18th NITROGEN WORKSHOP

THE NITROGEN CHALLENGE: BUILDING A BLUEPRINT FOR NITROGEN USE EFFICIENCY AND FOOD SECURITY

18th Nitrogen Workshop

PROCEEDINGS

Lisbon, Portugal, 30th June – 3rd July 2014

Editor: Cláudia M. d. S. Cordovil

INSTITUTO SUPERIOR DE AGRONOMIA
Universidade de Lisboa
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BULDING A BLUEPRINT FOR NITROGEN USE EFFICIENCY AND FOOD SECURITY

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NITROGEN WORKSHOP – A leading network on Nitrogen

On the behalf of the organizing Commission of the 18th Nitrogen Workshop, I wish to express a warm welcome to all the participants.

This book contains the proceedings of the 18th Nitrogen Workshop, held in Lisboa, Portugal, from 30th June to 3rd July 2014.

The Nitrogen Worlshop is a leading network on all issues related to Nitrogen, since 1982. From an European network, the Nitrogen workshop grew to an international network, integrating more and more countries and enlarging its focus to broader questions about Nitrogen.

There was a global participation of scientists and technicians, from 30 different countries from all the continents, who presented the results of their collaborative work. This is a clear sign of the importance of these workshops and stressed out the leading position of this network in the field of the challenging Nitrogen element. The doors are open to the growth of the network.

We are proud to present an outstanding scientific program with high quality speakers and innovative posters presented by more than 200 delegates, which will hopefully give rise to interesting and participated discussions. We have put our efforts in the selection of an innovative and comprehensive overview of the latest research developments to share with you and we hope you enjoy the workshop. More that 200 posters were displayed in large poster sessions, to allow the participants the exchange of ideas and information amongst them. It is worth highlighting a strong participation of students which ensure and keep alive the research in the subject of Nitrogen.

We hope that this Workshop will host great opportunities for the participants to meet and build future collaboration through friendship and professional interaction, and that this may be place and time for discussing the latest developments in science and politics for Nitrogen issues, and a means of moving forward to problem solving, innovation and knowledge transfer.

We wish to express sincere thanks to all the persons that made the 18th Nitrogen Worshop possible. To the colleagues who kindly accepted to revise the abstracts submitted to this Workshop, to the Chair of the 17th Nitrogen Workshop for his advice, availability and support at all times, to the Steering Committee of the International Nitrogen Initiative (INI) for scientific advice and to the European Centre of the INI for supporting this meeting.

Cláudia S. C. Marques dos Santos Cordovil

History of the Nitrogen Workshop

“The nitrogen challenge: building a blueprint for food and future”

- 17th International Nitrogen Workshop: Wexford, Ireland. **“**Innovations for sustainable use of N resources”.
- 16th International Nitrogen Workshop: Universita Degli Studi di Torino, Italy. 28 June - 1 July 2009. “Connecting different scales of nitrogen use in agriculture
- 15th International Nitrogen Workshop: University of Lleida, Spain. 28-30 May 2007. “Towards a better efficiency in N use”
- 14th International Nitrogen Workshop: PRI, Maastricht, The Netherlands. 24-26 October 2005. “N Management in Agrosystems in Relation to the Water Framework Directive”.
- 12thInternational Nitrogen Workshop: IGER, University of Exeter, UK.

  21-24 September 2003. “Controlling nitrogen flows and losses”.
- 11th International Nitrogen Workshop: INRA, Reims, France. 9-12 September 2001
- 10th International Nitrogen Workshop: The Royal Veterinary & Agricultural Univ Copenhagen, Denmark. 23-26 August 1999
- 9th International Nitrogen Workshop: Technische Universitat Braunschweig, Germany. 9-12 September 1996
- 8th International Nitrogen Workshop: University of Ghent, Belgium. 5-8 September 1994.
- 7th International Nitrogen Workshop: University of Edinburgh, UK. 3-26 June 1992
- 6th International Nitrogen Workshop: The Queen’s University Belfast, UK. 17-19 December 1990
- 5th International Nitrogen Workshop: Rothamsted Experimental Station, UK. 13-14 December 1988
- 4th International Nitrogen Workshop: University of Aberdeen, UK. 6-9 April 1987
- 3rd International Nitrogen Workshop: GRI, University of Reading, UK.

  16-17 December 1985
- 2nd International Nitrogen Workshop: Rothamsted Experimental Station, UK. 17 July 1984
- 1st International Nitrogen Workshop: Rothamsted Experimental Station, UK. 20 July 1982

18th Nitrogen Workshop

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Topic 1

Nitrogen use in agriculture

IMPACT OF NITROGEN FERTILIZER RATES ON INDUSTRIAL HEMP (CANNABIS SATIVA L.) BIOMASS PRODUCTION

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For many centuries, industrial hemp (*Cannabis sativa* L.) has been cultivated as a source of strong stem fibres and seed oil (Struiket et al., 2000).Industrial hemp's need for nitrogen is high, especially during the growing period (Amaducciet et al., 2002); moreover lack of nitrogen will result in lower yield, as growth phases (internodes' length, canopy area) will be missed and thus efficiency of radiation use will decline. In addition, in Latvia there are no recommendations for suitable nitrogen fertilizer rates for hemp cultivation. The aim of this study was clarification of nitrogen fertilizer rate impact on industrial hemp yield in Latvia and finding out of the optimal nitrogen fertilizer rate for better biomass production.

Materials and Methods

The field trial was carried out in 2011-2012 in Research and Study farm "Peterlauki" (56°53'N, 23°71'E) of the Latvia University of Agriculture, in the sod calcareous soils pH_{KCl} 6.7, containing available for plants P 52 mg kg $^{-1}$, K 128 mg kg $^{-1}$, organic matter content 21 to 25 g kg $^{-1}$ in the soil. Two factor experiments were conducted: factor A – cultivars (Futura 75, Tygra and Felina 32); factor B – various nitrogen fertilizer application rates (control – N0P0K0; background fertilizer (henceforth F) – N0P80K112; F+N30; F+N60; F+N90; F+N120; F+N150; F+N180 kg ha^{-1}). Statistical assessment of the data was made by ANOVA, variance analysis, and LSD test. Processing of data included also correlation and regression analysis methods.

Results and Discussion

Production of high quality biomass requires knowledge about optimal plant density for sowing. If plants are sown too dense or because of consequential interspecific competition, part of the plants die, others stop growing, and only the remainder contribute to the final production (Amaducciet et al., 2002). In 2012, the established plant density after full emergence varied between 217 and 258 plants per m^{-2}. The highest plant density (significant ($p< 0.05$)) was found in the plots where additional N fertilizer was not used (N0P0K0 – 256 plants per m^{-2}) and in the plots where fertilizer N60 was used (F+N60 – 258 plants m^{-2}). Whereas the lowest plant density (significant ($p< 0.05$)) was found if fertilizer N90 was used (F+N90 – 217 plants per m^{-2}).

During vegetation period, plant density decreases. At harvesting, the highest plant density was found for fertiliser N60 (233 plants per m^{-2}), but the lowest for N180 fertilizer (207 plants per m^{-2}). Some authors report that plant density decreases showed negligible plant lost at low density (about 30 – 90 plants per m^{-2}), while at high density (180-270 plants per m^{-2}) about 50-60% of the initial stand was lost. In other references it was stated that nitrogen caused high plant mortality, probably due to competitive effects in the initial phase of the cycle (Amaducciet et al., 2002; Jankauskiene and Gruzdeviene, 2012).

Hemp stalk length was significantly ($p< 0.05$) influenced by the nitrogen fertilizer rate applied and cultivar. Plant height gradually increases with higher N fertilizer dose, compared with the control (N0P0K0), however this growth varies among the cultivars tested. The highest stalk length was observed for the cultivar 'Futura 75' and if treated with any of the nitrogen fertilizer. The highest stalk length (3.18 m) was reached with nitrogen fertilizer F + N150 at 138 growing day. With application of the same fertiliser, stalk length of other cultivars ('Tygra' and 'Felina 32') was smaller, – 2.58 meters.

Analysis of the relationships between hemp stalk length and biomass yield indicates significant ($p < 0.05$) close linear positive correlation ($r=0.83$; $n=24$). Significant ($p < 0.05$) linear positive correlation ($r=0.53$; $n=24$) was found in 2012 and it shows that in 2011 in 69% of cases changes in yield might be explained by the stalk length, but in 2012 – only in 28%. Thus it may be concluded that hemp biomass yield depends upon not only nitrogen fertilizer rate, but also other factors, namely plant density, meteorological conditions and other factors currently not studied.

The effect left on dry matter yield by nitrogen fertilizer was significant ($p < 0.05$) in both years. The lowest amount of dry matter yield was harvested from plots where additional N fertilizer was not used (control plots – N0P0K0), respectively on average 4.34 t per ha^{-1} in 2011 and about 7.20 t per ha^{-1} in 2012.

Conclusions

Hemp biomass yield depends upon stalk length; moreover the phenomenon was approved by positive linear correlations.

The effect left by nitrogen fertilizer rate on the biomass yielded was significant ($p < 0.05$). The lowest green yield (and dry matter yield) was observed treating samples with N0P0K0 fertilizer, while the highest green biomass and dry matter yield was obtained by using F+N150 kg ha^{-1}. Increase in the N rate up to 180 kg ha^{-1} resulted in lower hemp green biomass and dry matter yield.

Acknowledgement

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NITROGEN USE EFFICIENCY IN INTENSIVE VEGETABLE GROWING: WHAT'S THE POTENTIAL OF ALTERNATIVE CROP ROTATIONS?

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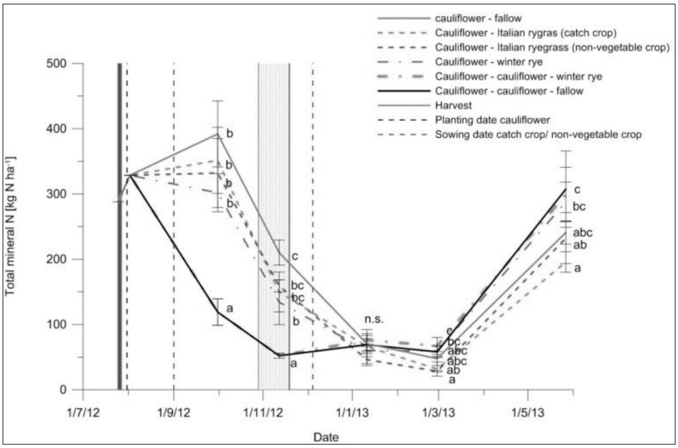
Vegetable crop residues take a particular position relative to arable crops as often very large amounts of biomass with a high N content are left on the field after harvest. Vegetable crop residues are characterized by small C:N ratios (De Neve & Hofman, 1996) and mineralize rapidly (Fox et al., 1990, Trinsoutrot et al., 2000). An important amount of vegetable crops are harvested during late autumn and despite decreasing soil temperatures, high rates of N mineralization and nitrification still occur (De Neve & Hofman, 1996). Furthermore vegetable crops are often the last crop before winter and several vegetable crops are harvested before a mature stage, leaving behind soils with considerable N content. These factors cause intensive vegetable rotations to be particularly prone to nitrate leaching during winter. In order to obtain water quality objectives set by the Nitrates Directive three 1,5-yr field experiments were set up to evaluate the inclusion of a catch crop or non-vegetable crop in vegetable rotations in autumn. N dynamics during the trial period were simulated by using the EU-Rotate_N model. In this study the impact of alternative crop rotations on N uptake, N losses during winter and N remineralization during the following spring are assessed.

Materials and methods

Alternative crop rotations. Three field experiments were established in the intensive vegetable growing region in Flanders, Belgium (autumn 2012- spring 2014). All field experiments were designed in fully randomized blocks with four replicates. The first alternative crop rotation examines the inclusion of Italian ryegrass (Lolium multiflorum) in cauliflower (Brassica oleracea var. botrytis) rotations. Per location two treatments, namely (i) cauliflower – Italian ryegrass (sown in August) and (ii) cauliflower – cauliflower – Italian ryegrass (sown in October) are compared to the conventional cauliflower – cauliflower combination. Following spring one or two cuttings of grass is harvested and removed. The remaining organic material is incorporated and a new cauliflower crop is planted. The second alternative crop rotation examines the use of two cover crops (Italian ryegrass or winter rye (Secale cereale) after a cauliflower crop. Similar as for the first alternative rotation two rotations, namely (i) cauliflower – cover crop (sown in August) and (ii) cauliflower – cauliflower – cover crop (sown in October) are compared to a conventional double cauliflower rotation. However in contrast to the first alternative rotation the cover crop will be incorporated during spring instead of harvested. Soil and plant sampling During the experiment soil samples were taken monthly with an auger in three layers: 0-30 cm, 30-60 cm, 60-90 cm and analysed for ammonium-N and nitrate-N after 1 M KCl extraction in order to determine soil mineral N profiles. Total crop yield and crop residue biomass (leaves and stalks) was determined at harvest. Plant samples (4 subsamples per treatment) were dried, ground and analysed for dry and organic matter, N and P content. Simulation of N dynamics Simulation of the N dynamics was performed with EU-Rotate_N. The model was calibrated through input of measured soil mineral N content, crop yield and crop N uptake.

Results and discussion

Evaluation and simulation of the results of the field experiments is ongoing and will be presented at the N Workshop, but some trends may already be identified. Soil mineral N content was significantly lower following a second cauliflower crop compared to catch crops or non-vegetable crops at two of the three field experiments because N uptake by the second cauliflower crop continued to deplete soil N content. The maximum soil nitrate content during autumn (90 kg NO3-N between 09/01 and 11/15) set by the Flemish legislation was only met for a double cauliflower crop (Figure 1). Total N uptake of Italian ryegrass (79 ± 1 kg N ha-1) was higher than winter rye (70 ± 1 kg N ha-1) despite a slower growth. Soil mineral N content was lower following incorporation of Italian ryegrass in spring compared to fallow soil (Figure 1). Hence Italian ryegrass may immobilize N at first which should be taken into account when planting the next crop. First simulation results indicate that gaseous losses may contribute significantly to total N losses (data to be shown at the conference). Figure 1 Mean soil mineral N content (0 – 90cm soil layer) measured at one location. Significant differences are indicated by different letters ($p < 0.05$, post hoc Tukey test).



Acknowledgments

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EVALUATION OF AMMONIUM NITRIFICATION IN THE PRESENCE OF DMPP NITRIFICATION INHIBITOR IN SOLUTION IN THE SOIL OF CITRUS TREES UNDER FIELD CONDITIONS

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About 85-90 million metric tonnes of nitrogenous fertilizers are added to the soil worldwide annually (Good et al., 2004). However, it has been estimated that 50-70% of this N is lost. Nitrate leaching is considered to be one of the most important mechanisms of N losses from soils (Shen et al., 2003), with consequence of the low fertilizer-N uptake efficiency (NUE), and also contributes to nitrate pollution of groundwater (Camargo & Alonso, 2006). Retardation of the biological oxidation of ammonium can reduce N losses due to leaching. Several chemical compounds are capable of retarding this oxidation by inhibiting the activity of *Nitrosomonas* bacteria, responsible for the first step in nitrification. Among these compounds, 3,4-dimethylpyrazole phosphate (DMPP), is a molecule, developed by BASF, very efficient in inhibiting the nitrification process in soil, is effective at low concentrations (0.5-1.0 kg DMPP ha^{-1}), allows the ammonium present in the soil to be blocked for a variable period of time, significantly reducing losses of N due to leaching and denitrification, allowing planning of mixed ammonium and nitrate nutrition of the crops, and no toxicological or ecotoxicological side-effects have been reported (Zerulla et al., 2001). Numerous studies have been carried out on small plants grown in pots or in assays in soil conditions with sampling at the end of the assay. However, there are no available studies to prove the efficiency of its daily use under field conditions and with numerous sampling during its application, showing its effectiveness throughout the fertilization period. Thus, the study focused on determining effect of daily DMPP applications on i) ammonium and nitrate concentration, both in the soil and soil solution, and ii) macro and micronutrient foliar.

Materials and Methods

Ten Uniform 11-year-old orange trees (*Citrus sinensis* (L.) Osbeck) c.v. Clemenules grafted on Carrizo citrange (*Citrus sinensis* x *Poncirus trifoliata*) rootstock were grown in field conditions with a plantation frame of 4 x 6 m (about 417 trees Ha^{-1}). The N fertilizer rate was 460 g N tree-year^{-1} based on tree canopy (Ø of 2.65 m), of which 20 % must be applied during the month of July (92 g of N·tree^{-1}), and, as the fertiliser was applied only for 10 days, a third of the necessary amount was applied during the assay (30.6 g of N·tree^{-1}). Nitrogen was supplied as ammonium sulphate either without or with the nitrification inhibitor DMPP (1 % DMPP relative to NH_4-N). The irrigation water used came from the irrigation well on the plot. Four treatments were carried out: I) Tree-Without: Citrus crop fertilised with AS; ii) Soil-Without: Bare soil fertilised with AS; iii) Tree+DMPP: Citrus crop fertilised with AS + DMPP; iv) Soil+DMPP: Bare soil fertilised with AS + DMPP. Each treatment was repeated five times in the experimental area with a tree per replica was used in the case of the citrus crop and bare ground in a zone equivalent to the area of a tree. The study was carried out during the month of July 2012 under the most unfavourable conditions for use of DMPP: high N needs for trees (July is the month with the greatest nutritional requirement of N) and with high temperatures. Three suction probes were used under each tree to gather the solutions from the soil at three depths (0-20, 20-40, and 40-60 cm). The soil solutions were subsequently collected in recipients (250 mL). To determine mineral fractions, both in soil KCl extracts and in soil solution were steam-distilled (Distillation Unit, Foss Tecator,); NH_4-N and NO_3-N were recovered in boric acid. To evaluate fertilizer N uptake, spring flush leaves were also sampled at the end of the assay (September).

Results and Discussion

Ten hours after N application, soil solution NH_4-N concentrations increased in all treatments. The highest values of NH_4-N concentrations in soil solution were found in Tree+DMPP treatments in the area of greatest root development (0 to 40 cm) mainly during fertilization period. At the end of the assay, the trial zone without trees had lower ammonium in the first two layers, without significant differences due to the effect of the DMPP input (Figure 1). The most remarkable differences among the AS (Tree-Without and Soil-Without) and AS+DMPP (Tree-DMPP and Soil-DMPP) treatments were observed in the first and last N application-end of sampling period interval, showing the residual effect of DMPP application.

With regard to nitrate concentration, at 40-60 cm depth, a higher concentration was noted in the treatments that did not receive DMPP (with and without tree). In this way, the risk of nitrate-N pollution in shallow groundwater would be minimized. In the second week, after continuous input of DMPP, higher nitrate concentration was observed in the treatments with tree than in those on bare ground. Highest nitrate concentration was found in bare soil zone that did not receive DMPP throughout the whole soil profile (Figure 2). Numerous authors found similar effect in soil samples (Quiñones et al. 2009).

Similar results were found in ammonium and nitrate concentration in soil samples at the end of the assay. DMPP application did not affect mineral foliar concentration, possibly due to the short duration of the assay (not shown).

Conclusions

The results of this study indicate that daily addition of the nitrification inhibitor DMPP to ammonium sulphate in drip irrigated adult citrus trees, reduced N loss through leaching as a consequence of the diminished nitrification rate, and brings on an ammonium nitrate mixed nutrition. Consequently, it can be concluded that high temperatures during the experiment did not depress the effect of DMPP.

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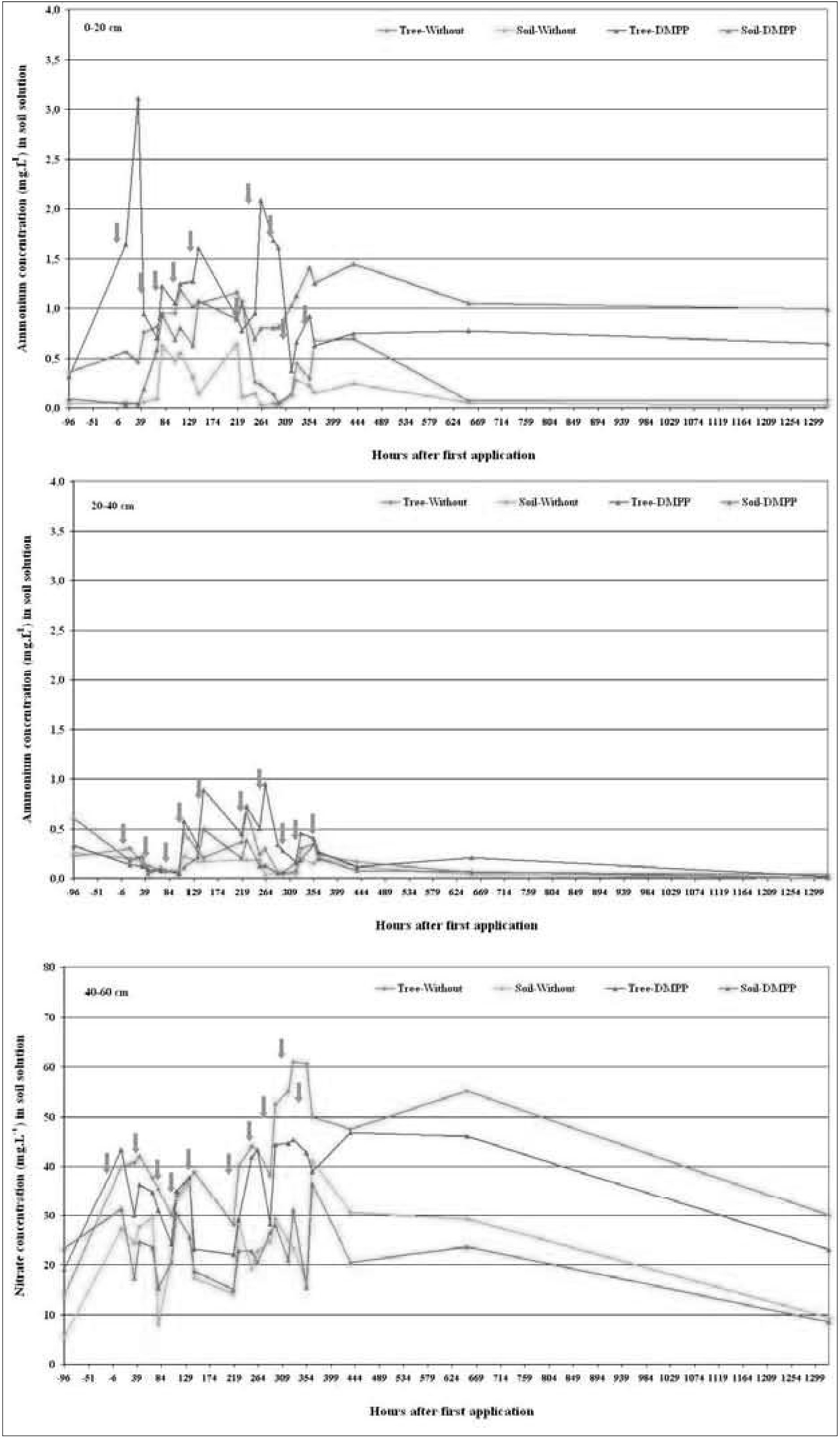

0-20 cm
Tree-Without
Soil-Without
Tree-DMPP
Soil-DMPP
Ammonium concentration (mg.L-1) in soil solution
Hours after first application
20-40 cm
Tree-Without
Soil-Without
Tree-DMPP
Soil-DMPP
Ammonium concentration (mg.L-1) in soil solution
Hours after first application
40-60 cm
Tree-Without
Soil-Without
Tree-DMPP
Soil-DMPP
Nitrate concentration (mg.L-1) in soil solution
Hours after first application

RESIDUAL INORGANIC NITROGEN CONTENT IN SOILS FROM INTENSIVE FARMS IN SOUTH BULGARIA

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Nitrogen in the soil is the most important element for plant development. It is required in large amounts and must be added to the soil to avoid a deficiency. When nitrogen is limited, crop growth is slow and yields are reduced. In case of nitrogen excess water pollution with nitrates is provoked. Agriculture is the strongest source of nitrates pollution. In general agricultural area with intensive farming coincide with nitrates polluted aquifers [2]. Optimization of nitrogen fertilizer rates is the aim of agronomists and farmers. The Canadian average residual soil nitrogen values (for the 0-20 cm soil layer) from 1981 to 1996 were fairly constant with a range of 12.9 to 13.9 kg N ha-1. However, residual soil nitrogen increased by 51% from 13.9 kg N ha-1 in 1996 to 21.0 kg N ha-1 in 2001. This increase is due to several factors including an increase in legume crop acreage and lower crop yields and reduced N uptake as a result of climatic constraints (droughts) [3].

Material and Methods

Main soils in studied areas are heavy and most frequently Vertisols and Chromic Luvisols are present [4]. Eighteen farms from administrative areas of Burgas, Stara Zagora and Yambol were studied (Table 1). Arable area of these farms varies from 15.7 to 903.22 hectares. Number of soils sampling is different and depends on the area of every crop rotation field – from 4 to 70. Survey is accomplished in the frames of National Agroecological Program – 2007-2013. Amount of residual N is calculated for 0-25 cm layer.

Inorganic nitrogen determination was according Bremner-Keeney, 1966 method for residual nitrogen assessment [1].

Statistics data treatment was accomplished with Statgraphics Centurion software.

Table 1. Arable area, number of fields and sample numbers of studied farms

	Yambol area			Stara Zagora area			Burgas area		
	Arable area	Fields	Sample numbers	Arable area	Fields	Sample numbers	Arable area	Fields	Sample numbers
Average	323.8	9	28	518	10	20	269.2	8	12
Max	849	13	70	903.22	16	35	644.59	18	21
Min	118.67	4	6	211.8	6	7	15.7	4	4
Total	2590.8	69	221	2072.13	39	82	1615.0	48	71

Results and Discussion

Survey of 18 intensive farms was accomplished in our study. Farmers in such farms aim to higher yields and nitrogen fertilizer rates are elevated. As consequence higher amounts of residual nitrogen could be expected.

All outliers from data ranges were removed before statistical treatment. That is why number of analyzed soil samples and soil samples statistically treated is different.

Average values for the residual inorganic nitrogen in soil is much higher than observed in other countries [3] – from 29 to 50 kilograms per hectares. It is calculated for 0-25 cm soil layer. This is explaining only 25% of obtained results. In our case high amounts of residual nitrogen are due, partly, to the dry period at the end of growing season.

Maximal amounts of residual nitrogen show that framers are applying non reasonable nitrogen fertilizer rates or some of them have applied fertilizers before soil sampling. Application of very high nitrogen fertilizers rates could explain the minimum amounts of residual nitrogen. Minimum of residual nitrogen in soil in Stara Zagora area is close to the average amounts found by Drury et al., 2007. Maximum amounts (up to 84 kg per hectare) and high coefficients of variation (up to 47.9%) are sign of large nitrogen rates of fertilization.

Table 2. Statistics for studied farms in 3 administrative areas

Statistical parameters	Yambol	Stara Zagora	Burgas
Count	183	56	64
Average	38.73	50.23	29.04
Standard deviation	15.01	20.49	13.91
Coeff. of variation	38.75%	40.78%	47.90%
Minimum	4.8	12.6	6.0
Maximum	73.5	84.0	60.0
Stnd. skewness	1.91	-0.23	1.61
Stnd. kurtosis	-0.83	-1.95	-1.29

Conclusion and perspectives

High variation of residual inorganic nitrogen in soils shows that more detailed studies in farms are needed. Best resolution of the problem is to apply spatial variability studies of fields. Use of variable fertilizing rates in one field is needed for optimization of applied amounts of fertilizers. It will decrease environmental pollution with nitrates.

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AGRONOMIC AND ENVIRONMENTAL PERFORMANCES OF ORGANIC FARMING IN THE SEINE WATERSHED: TURNING BACK OR MOVING FORWARD?

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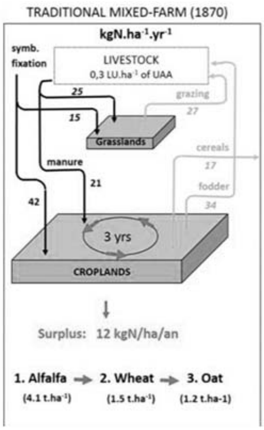
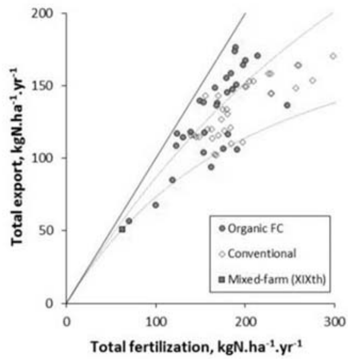
The Seine watershed (81 000 km), has long been the foodshed of Paris, meeting the demand for most animal and vegetal proteins consumed in the Capital city and supplying high quality water. Nowadays, in addition to providing most plant products consumed by the city, the traditional France's breadbasket exports 80 % of its huge cereal production on international markets (Billen et al. 2012). For the last 50 years, the Seine watershed has specialized into intensive crop farming (cereals, oilseed rape, sugar beet) entirely sustained by synthetic fertilizers and pesticides, while animal husbandry, has been concentrated within the Western and North adjacent regions. Those modern industrial agrosystems, are the main cause of severe surface and groundwater contaminations with pesticides and nitrates, thus endangering the drinking water resources of Paris agglomeration and leading to eutrophication problems. Organic farming appears as an alternative to the conventional input-intensive especially for pesticide contamination, but there are still lively debates on N leaching risks, soil fertility and productivity. In this study, we attempt to handle simultaneously those issues by analyzing and comparing long-term total N-use efficiency in: i) a 19th century system relying on the synergy between livestock breeding and crop production, ii) 28 organic farms specialized in field crops, and iii) conventional field crops rotations strictly following 'best' fertilization practices.

Material and Methods

We used the Soil Surface Balance, SSB (Oenema et al., 2003), extended over a whole crop rotation cycle to assess potential N losses to the environment. The inputs accounted for were synthetic fertilizers, manure, grazing excreta, symbiotic fixation (estimated with an empirical model based on exported biomass and taking into account rhizodeposition (Anglade et al., in prep)), and atmospheric depositions. Outputs were estimated as the N content of harvested crops and forage plus grazing. For organic field crop rotations, the SSB were established on the basis of farmer's interviews carried out in 2011-2012. Conventional crop rotations N balances were calculated using statistical data from the French Ministery of Agriculture for yields, and new regional references (2012) on soil nitrogen mineralization for estimating the appropriate rate of fertilizers according to the need of the crops, in compliance with the EU Nitrate Directive. Lastly, nitrogen fluxes on both grasslands and arable lands of a traditional mixed-farm were reconstituted combining historical statistical data (from 1870 to 1896) about land use and yields of cereals, legume fodder and permanent grasslands, with a careful reading of the famous realistic novel, The Earth, by Emile Zola which surprisingly contained details in abundance on daily farm routine at the end of the 19th century.





Results and Discussion

The self-sustaining mixed farming systems that were able to supply the fivefold increase in the Paris population during the 19^{th} century were based on close relationships between cereal cultivation, and livestock feeding (Fig.1). Apparently breaking with this past, and following industrial trends, many organic farms in the Seine watershed are specialized in the production of field crops without any breeding activity. Nonetheless, those organic systems differ significantly from intensive conventional practices by long and diversified crop rotations (7 to 14 years), including cereals and legume as crop (faba bean/lentil), fodder (alfalfa) and green manure (clover). Symbiotic fixation accounts on average across all farms for about 65% of total N inputs, the remaining part coming from atmospheric deposition (7 %) and exogenous sources (27%) like distillery residues and manure from distant husbandries. Legumes are also responsible for high protein yields over the whole crop rotation (N-export), that equalize or outperform conventional ones, at similar total fertilization rates. On average, organic N surpluses (35 $kgN.ha.^{-1}.yr^{-1}$) are also 40 % lower, although it exists a gradient of farms differing on N sources and crop rotations (Fig.2). It should be noted that among organic farms, huge N surplus > 50 $kgN.ha.^{-1}.yr^{-1}$ are linked to large amounts of exogenous inputs (> 50 $kgN.ha.^{-1}.yr^{-1}$) and/or the lack of outlets for fodder N-rich legumes. Considering that most of the cropland nitrogen surplus is leached, we have straightforward evaluated sub-root nitrate concentrations. The N fluxes of arable land in the traditional 19^{h} system were approximately in balance, thus sub-root water concentration (4,6 $mgN.l^{-1}$) was not surprisingly far below current drinking and ecological standards. By contrast, only 9 % of the intensive cereal rotations managed with official fertilization practices recommendations, and 50 % of the organic farms specialized in crop production, were found to deliver sub-root water meeting the drinking standards of 11mgN/l.

Conclusion and perspectives

The extension of organic agriculture, to meet water quality target while maintaining high protein productivity is substantially dependent upon local opportunities of valorizing legume fodder cereal by-products. Between evolutions and revolutions, are the futures contained in past agricultural systems?

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INTERCROPPING LEGUME AND NON-LEGUME, AN INNOVATIVE WAY TO VALORIZE N2 FIXATION AND SOIL MINERAL N SOURCES IN LOW INPUTS CROPPING SYSTEMS.

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Increasing concern about climate change and environmental impacts require transformation of actual cropping systems focusing on enhanced sustainability. Then, optimizing the use of natural N sources such as N2 fixation and soil mineral-N coming from organic matter mineralization is crucial. One solution could consist in increasing the diversification of crops across the European countries e.g. by implementing more legumes crops or by developping the use of intercropping (IC) such as cereal and legume mixtures. IC is the simultaneous growth of two or more species in the same field for a significant period and an application of ecological principles. IC is known to use available abiotic resources more efficiently than the corresponding sole crops particularly in low-input systems. Indeed, well chosen intercropped species do not compete for exactly the same resource niche but tend to use them in a complementary way leading to higher yield and grain protein compared to sole crops (e.g. Hauggaard-Nielsen et al. 2007; Bedoussac and Justes 2010). The main objective of our field studies was to analyse the functioning of cereal – grain legume intercrops as a first step to further optimize and finally propose optimised intercropped systems. Thus, we evaluated the potential advantage of intercrops for global yield and wheat grain protein concentration modified by N-fertilization, densities, species and cultivars.

Materials and Methods

58 organic field experiments were carried in France (Toulouse in the SW France and Angers in the NW) and Denmark from 2001 to 2010 with a large range of intercrops combinations (hard wheat, soft wheat or barley intercropped with pea or faba bean) with various cultivars, sowing densities and N treatments leading to a large range magnitude and dynamics of N availability. Grain yield was used to calculate the efficiency of IC based on the land equivalent ratio (LER) i.e. the relative land area under SC that is required to produce the yield achieved in IC. Cereal grain protein content was also analysed as a quality criteria and the percentage of N derived from N_2 fixation (%Ndfa) of legumes was evaluated in order to estimate all N sources.

Results and discussion

Total intercrop grain yield (cereal + legume) was almost higher than that of the mean SC grain yield (3,3 vs. 2,7 Mg ha-1 respectively; Figure 1) and the proportion of cereal yield in IC was higher than 50% and higher to that calculated from SC grain yields, indicating an higher competitivity of the cereal. LER values are almost higher than 1 (1,27 on average ranging from 0,93 to 2,41). High values (>1,5) correspond in general to situations in which at least one of the sole crop grain yield was low. IC was more efficient than SC without N fertilization or when N was applied late during cycle; this is mainly due to: i) a better light use (up to 10%) thanks to species dynamic complementarity for leaf area index and height, ii) growth complementarity over time (higher growth rate of the cereal and then of the legume), and iii) a dynamic complementary in N acquisition, i.e. N_2 fixation and soil mineral N. Disadvantages were observed with large available-N during early growth stages leading to higher cereal growth, increasing interspecies competition, reducing legume light absorption and then its biomass and yield. Cereal grain protein concentration was significantly higher in IC than in SC (11,1% vs. 9,8%; Figure 2) and the lower the SC grain protein concentration the higher the increase of N concentration in IC. The grain protein concentration increase in IC is due to: i) a lower cereal grain yield in IC than in SC (1,9 vs. 2,9 Mg ha-1 respectively) due to lower sown density density and so on fewer ears per square meter in IC and ii) a quite similar (ca. 90%) amount of available soil N for the cereal in IC compared to SC because of a high legume N_2 fixation rate in IC (75%) leading to a small amount of soil mineral-N uptake by the legume. The percentage of N derived from N_2 fixation of legumes was significantly higher in IC than in SC (75% vs. 62%) but the amount of N fixed was lower in IC (56 vs. 93 kg N.ha-1) due to a smaller biomass than in SC.

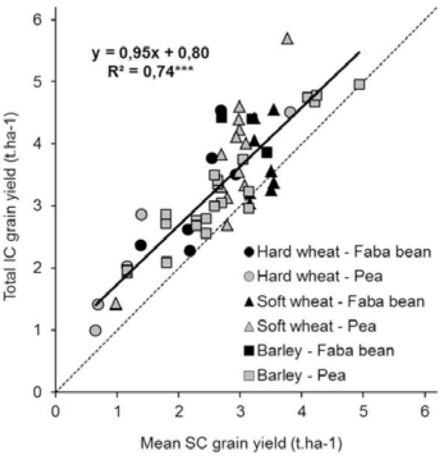

Figure 1: Comparison of the intercrop (IC) grain yield (cereal + legume) with the mean sole crop (SC) grain yield.

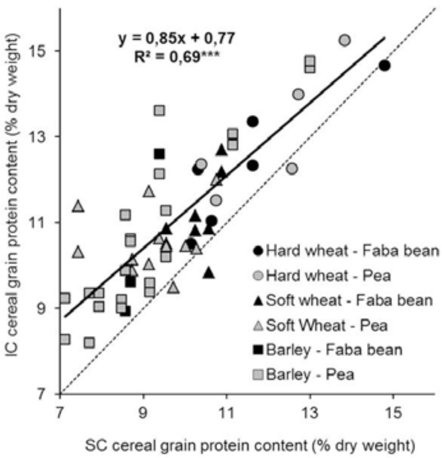

Figure 2: Comparison of the intercropped (IC) cereal grain protein concentration with that of the sole cropped (SC) cereal.

Conclusions

Our work confirms that IC is particularly suited to low N input systems due to the complementary use of N sources by species allowing a better wheat grain filling. However, a number of factors still need to be optimized in order to propose future cropping systems appropriately optimized, such as grain legume cultivars, sowing practices and design depending on specific goals (e.g the maximum total yield, global protein production, highest wheat grain protein content or multicriteria objectives).

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INCREASING SMALHOLDER CEREAL PRODUCTION IN THE HUMID HIGHLANDS OF ETHIOPIA VIA N-EFFICIENT FABA BEAN (VICIA FABA L.) – WHEAT ROTATIONS

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Most soils in Sub Saharan Africa (SSA) are deficient in nitrogen (N), which is the main constraint for cereal production. Traditional soil fertility restoring mechanisms like natural fallows are not longer an option due to high population pressure (Belane and Dakora, 2010; Vanlauwe et al., 2011). The use of mineral fertilizers could be a means to alleviate low nutrient levels and improve crop yields. However, at recommended application rates, mineral fertilizers are generally too expensive for resource-poor smallholder farmers in SSA. Hence, the exploitation of efficient N2-fixing legumes in cropping systems is a key strategy for sustainable agricultural intensification of low input cropping systems. Therefore, the objective of this study was to examine to which extent improved faba bean varieties enhanced soil N balances and grain yields of a subsequent wheat crop.

Materials and Methods

A two-phase field experiment was conducted on farmer's fields in Dedo (located at 7°28' N and 36°52' E at an elevation of 2160 m above sea level) in the tropical highlands of southwest Ethiopia involving faba bean and wheat. In the first phase, three improved faba bean varieties (Degaga, Moti, Obse) were grown at four levels of P fertilization (0, 10, 20 and 30 kg P ha–1) along with a local faba bean variety and wheat without N fertilization. N2-fixation was quantified by the 15N natural abundance technique (Peoples et al., 2009). The N balance was determined via two possible residue management scenarios: Scenario-I (i.e. common practice) assumed that all the aboveground biomass is exported from the fields; scenario-II assumed that all the above ground biomass except grains and empty pods is returned to the soil. In the second phase, agronomic efficiency of faba bean residues for a subsequent wheat crop was assessed. Again, no N fertilizer was applied to the succeeding wheat crop.

Results and Discussion

The amount of N2-fixed by faba bean varieties ranged from 258 ± 17 to 387 ± 15 kg N ha–1. Scenario-I gave a negative net N balance in the range of -86 ± 6 (variety, Degaga) to -9 ± 9 (variety, Moti) kg N ha-1 with significant differences between varieties. Scenario-II showed that all balances were significantly ($P < 0.01$) improved and the varieties were positively contributing N to the system in the range of 51 ± 13 (variety, Degaga) to 168 ± 14 (variety, Moti) kg N ha-1, which is equivalent to 110 – 365 kg N ha-1 in the form of urea (46% N). The superior haulm and total biomass production by Moti could be related to its high N2-fixation potential (Nebiyu et al., 2013). P addition did not bring any significant difference in N2-fixation, grain or total biomass yield of faba bean varieties, which suggested that P was not limiting. In the second crop phase, biomass and grain yield of wheat grown after faba beans increased significantly ($P < 0.05$) by 112 and 82%, respectively, compared to the yield of wheat following wheat (Table 1). Further, highest wheat grain N uptake (59 kg N ha-1) was obtained for the highest faba bean P application rate; with a significant linear relationship between grain N uptake and P application (kg N ha-1 = 7 x kg P ha-1 + 32). Incorporation of legume residues improved wheat yield through N addition via fixed N (Yusuf et al., 2009) and likely also via increased P availability due to the high P acquisition of faba beans. This study clearly demonstrates the prospects and importance of faba beans as a valuable component in sustainable agricultural intensification of cereal-based cropping systems in the humid tropical highlands of Ethiopia and sub-Saharan Africa.

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FERTILISER EFFECTS ON ORGANIC WHITE CABBAGE GROWTH AND NUTRIENT UPTAKE

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The economic importance of white cabbage results from its nutritious value and high yields which are related to fertilization practices and the season (Citak and Aonmez, 2010). To increase the yield of organic cabbage, research is needed to focus on how to provide nutrients (particularly N) to match crop requirements without the use of conventional fertilizers. The objective of this work was to assess the potential of three fertilizers to increase organic white cabbage yield and nutrient uptake, including an organic fertilizer (compost), Gafsa phosphate and limestone, and to evaluate the interactions between these fertilizers.

Materials and Methods

A randomized block designed pot experiment was set up with white cabbage (Brassica oleracea var. capitata) inside a greenhouse (unheated) located in NW Portugal, according to organic agriculture (OA) regulations (EC Reg. 834/2007). Each of 4 blocks included 12 treatments from the factorial structure of three factors: (i) organic fertilizer (source separated urban waste compost; 0, 15 and 30 t ha-1); (ii) Gafsa phosphate (Fertigafsa; 0 and 200 kg P2O5 ha-1) and (iii) limestone (Fertical; 0 and 8 t ha-1 calcium carbonate equivalent) based on 50,000 plants ha-1. Compost dry matter (DM) content, pH, electrical conductivity (EC) and organic matter (OM) content were determined by European standard procedures (CEN, 1999). Total N and P were measured by molecular spectrophotometry; K by flame photometry, and Ca, Mg and Fe by atomic spectrophotometry. NH4+N and NO3--N contents of fresh soil and composts were extracted with KCl 2M 1:5 and determined by molecular absorption spectrophotometry. Three-way ANOVAs were carried out with LSD post hoc procedures to assess differences on plant growth and nutrient contents between treatments. Statistical calculations were performed using SPSS 15.0 for Windows (SPSS Inc.).

Results and Discussion

The fresh weight of organic white cabbage increased significantly ($p < 0.05$) with the application of limestone, Gafsa phosphate or compost, without significant interactions between fertilizers (Fig.1). For the overall experimental treatments, cabbage yield increased 10% and 25% respectively with lime and phosphate, but the most significant increases were found with the application of compost: 44% at the rate of 15 t ha-1 and 64% for the highest compost rate (30 t ha-1). The positive effect of lime and phosphate on cabbage yield decreased with increasing rates of compost application, which can be explained by compost alkalinity and P content. N accumulation in leaves increased with the application of all fertilizers, but the accumulation of P and K improved significantly only with the application of phosphate and with the application of 15 t ha-1 of compost. The compost mineral N content (NH4−N NO3-−N) could explain the increased yield and N uptake by cabbages. Compost NH4−N content (1615 mg kg-1

DM) was above the content limit of 400 mg kg-1 DM suggested as an indicator of stabilization of composts (Zucconi and De Bertoldi, 1987). A ratio NH4−N/NO3-−N lower than 0.16 was established by Bernal et al. (2009) as a maturity index for composts of all origins. Here, this ratio was 15.6 indicating that the experimental compost was not matured. However, the growth of cabbage was not impaired by increasing rates of compost application, suggesting that there has been no toxicity effect caused by the application of this compost which, in addition to nutrient availability, may have contributed to the improvement of the physical and biological soil properties. Cabbage N content increased significantly with compost application and also with phosphate and lime. Nutrient contents in roots were always below nutrient contents in shoots except for Fe and Mg. The N and K contents in the leaves at least doubled those of the roots, but this increase was lower for P and Ca. Therefore, the distribution of N, P, K and Ca between leaves and roots was held for the benefit of the leaves, but to a lesser extent for P and Ca, compared to N and K.

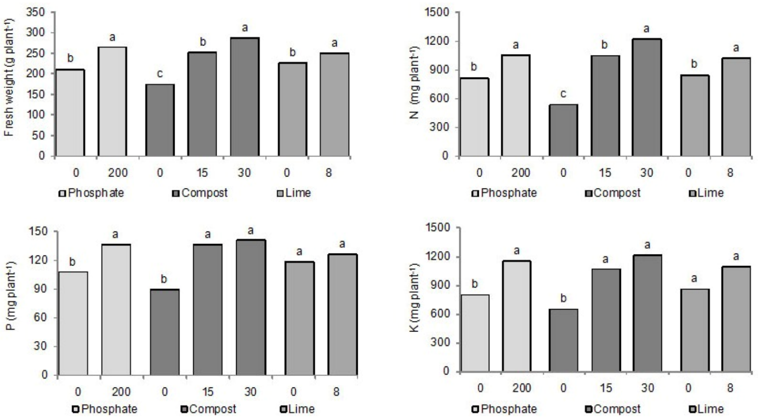

Figure 1. Mean fresh weight, N, P and K accumulation in white cabbage shoots with phosphate (kg P2O5 ha-1), compost (t ha-1) or lime (t ha-1) application, for the overall experimental treatments. Different letters above bars of the same fertilizer, mean significant differences between means ($p < 0.05$).

Conclusions

Organic white cabbage yield was significantly enhanced with the application of increasing rates of compost whereas lime and phosphate effects on cabbage yield were stronger when the compost was not applied. The response of cabbage to Gafsa phosphate was clear with or without lime or compost application, showing the importance of P fertilization to maximize organic cabbage yield. Partitioning of N, P, K and Ca between leaves and roots was held for the benefit of the leaves, but to a lesser extent for P and Ca, compared to N and K. These three fertilizers showed high potential to enhance organic cabbage nutrient uptake and can be recommended based on crop requirements and soil conditions to significantly enhance organic cabbage yields.

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EFFECT OF NUTRIENT ADDITION ON SOIL RESPIRATION IN MEDITERRANEAN GRASSLAND

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Nutrients such as nitrogen and phosphorous have been increasingly available in ecosystems largely due to human activities such as agriculture and combustion of fossil fuels. In spite of the global impacts of these human activities, few experiments have addressed how multi-nutrient availability controls plant biomass, species composition and functioning of grasslands. Also, soil respiration is the primary flux of carbon from soil to the atmosphere and is an important source of atmospheric CO_2. Knowledge of soil respiration dynamics and its controlling factors is therefore essential. In this experiment we will test how increased nutrient availability affects soil respiration in a Mediterranean grassland ecosystem.

Materials and Methods

To assess the effect of nutrient addition on soil respiration (Rs) the experimental set-up was established in 2012 in a Mediterranean grassland located in Companhia das Lezírias, Portugal. The experiment is a completely randomized block design: three blocks with 8 treatments per block and three additional bare soil control plots (N= 36 plots in total). Three nutrient addition treatments [Nitrogen (N), Phosphorus (P), Potassium plus other micronutrients (K+)], each with two levels (control, added), were crossed in a factorial design, for a total of 8 treatment combinations, and were established following the global Nutrient Network protocol. Nutrient additions were applied in autumn 2012 and 2013. Soil CO_2 fluxes (Rs) were measured every fortnight from February 2013 to February 2014. Soil water content (SWC) and temperature (Ts) were measured simultaneously. In addition to the above eight treatment combinations, “bare soil” control plots were established, where all vegetation was periodically removed. Leaf area index (LAI) was measured periodically during active plant growth using a ceptometer. Plant species composition and aboveground biomass were determined by clipping at the 2013 peak of biomass production. Soil (0-10cm depth) was sampled for determination of inorganic nitrogen pools (NH4 +-N and NO3 –N).

Results and discussion

Development of the herbaceous vegetation was confined to the period between February and May. During active plant growth, LAI increased significantly, reaching a peak in May. Effects of fertilization treatments on plant growth were noticeable from February on, as NK, NP and NPK+ plots responded rapidly to treatments. Peak aboveground biomass was highest in NPK+ and NP plots compared to unfertilized controls (950.63 and 715.80 gm^{-2} vs. 423.17, respectively).

Rs also displayed a distinct seasonal cycle, following changes in Ts and SWC, with Rs highest in spring and lowest during summer and winter. Ts did not differ considerably between fertilization treatments. Three months after the first fertilization, average Rs was highest in the NPK+ plots (2.97±0.42 $\mu mols^{-1}m^{-2}$), followed by similar rates in the NK and NP plots (2.52 $\mu mols^{-1}m^{-2}$ averaged across both treatments). These results were also reflected in the LAI dataset. Plots with only P or K fertilizations had lower and very similar effects on Rs (less than 1.60 $\mu mols^{-1}m^{-2}$). In spring, during peak growth, Rs rates increased in all treatments. Rs was highest in NK and NPK+ fertilization plots (3.22±0.52 and 3.10± 0.16 $\mu mols^{-1}m^{-2}$, respectively). In fact, soil NH4+-N and NO3 −-N concentrations measured in spring indicated a substantial available pool of N in NK and NPK plots. Although N plots presented higher available N and aboveground biomass than Control plots, Rs was lower in N plot, which might be related to differences in plant species composition or microbial community and activity. By June, as vegetation began to senesce, and until September, there was an accentuated decrease in Rs for all treatments, in accordance with the onset of the dry period. As first autumn rains occurred and while Ts was still mild, Rs rates increased with similar values for all treatments (averaging 1.49 $\mu mols^{-1}m^{-2}$). It is likely that this increase of Rs in autumn results more from a stimulation of the soil heterotrophic response and/or rainfall-derived pulses of carbon dioxide from soils than from an autotrophic response, as the rain at the end of the dry season occurs before annual grassland root re-growth

Conclusions

While nutrient addition seems to have an important effect on Rs rates, especially during spring growth, more data on root biomass, as well as data on soil chemistry, and microbial soil biomass and activity are needed to contextualize the observed responses.

STATISTICAL MODEL IMPACTS ON NITROGEN RECOMMENDATION, POTATO YIELD, NITROGEN UPTAKE EFFICIENCY AND RESIDUAL SOIL NITRATE

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Appropriate N fertilization is crucial to optimize potato yield and minimize environmental N losses (Zebarth and Rosen 2007). Many statistical models exist and can be used to establish the yield response to N and the optimal N rate (Bélanger et al. 2000) but the models will have different agronomic, environmental or agri-environmental implications. This study evaluated the effect of three statistical models of yield response to N and their implications with respect to tuber yield, nitrogen uptake efficiency (NUpE) and risk of nitrate leaching in five growing seasons.

Materials and Methods

The experiment was conducted during five growing seasons (2008-2012) on irrigated sandy soils in the area of Ste-Catherine-de-la-Jacques-Cartier (46°51'N, 71°37'W), near Quebec City, Canada. The experiment included 13 treatments replicated four times. Treatments were four N rates (60, 120, 200, and 280 kg N ha-1) for each of three N sources [ammonium nitrate (AN), ammonium sulphate (AS) and a polymer-coated urea controlled-release N (CRN)] plus an unfertilized control. The CRN was applied 100% at planting and the AN and AS were applied 40% at-planting and 60% at-hilling. At harvest, total yield (TY) and marketable yield (MY) were evaluated. The NUpE was calculated using N uptake in vines and tubers prior to vine desiccation. Risk of leaching was assessed using residual soil nitrate (RSN) in the 0-90 cm depth. Three models (quadratic, quadratic-plateau, and linear-plateau) were used to establish yield response to N and the optimal N rate.

Results and Discussion

The TY and MY were significantly influenced by N rate, but not by N source or year. The NUpE was always greater with AN and CRN than with AS (increased by 3%) and decreased with increasing N rate. The RSN increased with N rate but was also significantly influenced by N source and year. The CRN always produced greater RSN at harvest, particularly in a dry season and at high N rates.

With the quadratic model (r2= 48; MSE=34.6), the N rate to reach maximum TY (39.1 t ha-1) was 216 kg N ha-1 with a 95% confidence interval (CI) of 200 to 230 kg N ha-1 (Table 1). With the linear-plateau model (r2= 50; MSE=34.7), the N rate to reach maximum TY (36.9 t ha-1) was 84 kg N ha-1 with a CI of 74 to 94 kg N ha-1. With the quadratic-plateau model (r2= 50; MSE=32.2), the N rate to reach maximum TY (37.5 t ha-1) was 153 kg N ha-1 with a CI of 128 to 180 kg N ha-1. Similar results were obtained with the MY (Table 1).

The quadratic-plateau model produced slightly better statistical parameters (higher r2 and lower MSE). In comparison with the quadratic model, the quadratic-plateau model for TY predicted a 29% lower optimal N rate with a reduction of TY of less than 4%, and for MY predicted a 15% lower optimal N rate with a reduction of MY of less than 4%. In addition, use of the optimal N rate predicted by the quadratic-plateau model would increase the NUpE and reduce the RSN relative to other models examined.

Table 1. The maximum and marketable tuber yield, and the N rate to reach the maximum and marketable tuber yield, predicted using three statistical models: quadratic, linear-plateau and quadratic-plateau

N response for Total Yield						
-------------------------- Model --------------------- ---			---------- Tuber Yield --------- -		--------- N rate --------	
			Maximum	CI[z]	Nop[y]	CI
	r^2	MSE	------------ $t\ ha^{-1}$-----------		-------- $kg\ ha^{-1}$--------	
Quadratic	47.8	34.6	39.1	37.2-40.9	216	200-230
Linear-plateau	49.7	32.9	36.9	36.0-37.7	84	74-94
Quadratic-plateau	50.2	32.2	37.0	36.5-38.4	153	128-180
N response for Marketable Yield						
Quadratic	37.2	43.9	29.6	27.4-31.9	233	212-255
Linear-plateau	35.9	45.2	27.7	26.7-28.7	94	76-113
Quadratic- plateau	38.0	43.6	28.8	27.6-29.9	197	155-238

[z]: CI: 95% confidence interval.
[y]: Nop: the N rate to reach the maximum of total or marketable tuber yield.

Conclusion

The choice of the statistical model did not reduce significantly the TY and MY predicted but the optimal N rate calculated by the three models varied greatly. This suggests that when fertilizer N recommendations are based on field trials conducted across a range of soil types and environmental conditions, the recommended N rate may be strongly influenced by the choice of the statistical model used. In the long-term, the lower optimal N rate associated with the quadratic-plateau model is expected to be beneficial for the reduction of N losses in the environment.

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NITROGEN USE EFFICIENCY IN A SOIL AMENDED WITH DIFFERENT ORGANIC RESIDUES

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Nitrogen efficient use is important for economic and environmental sustainability of cropping systems. Three examples of fertilization practices considered to be able to contribute to the improvement of N use efficiency are: the use of a waste with higher C/N ratio, such as pulp mill sludge (Kirchmann and Bergström, 2003), specific application time and/or split application of nitrogen fertilizers (Roberts, 2008) and the use of nitrification inhibitors, such as the DCD (Zaman and Blennerhassett, 2010). In present study a field experiment was carried out for two years, in central Portugal, to evaluate agronomic nitrogen use efficiency (AE) and apparent N recovery efficiency (ARE), when different organic wastes (cattle slurry, sewage sludge and urban waste compost) were used as N sources in a double-cropping system producing maize and oats forage. The use of a nitrification inhibitor (DCD), the splitting application of residues and the use of an organic residue with high C/N ratio (pulp mill sludge) were evaluated as management practices that could improve N use efficiency.

Materials and Methods

The soil was a Cambisol, with 0.81% organic C, pH (H_2O) 6.2, and high P and K levels (>120 mg kg^{-1}). The 1st year autumn was rainy, and 2nd year one of the year most dried of last decade. The ten treatments tested consisted of: the splitting application at the establishment of the oats and maize crops of the organic residues sewage sludge (treatment SS), urban waste compost (UWC) and cattle slurry (CS); the yearly application of pulp mill sludge (PMS) to the oats crop, and SS and UWC to the maize crop only (SSm and UWCm); a mineral fertilizer treatment (MIN) and a Control were included, and the DCD effects were tested together with MIN (MIN+I) and CS (CS+I). PMS was applied in the first year only. Total N input was equal for all fertilization treatments (oats 80 kg N ha^{-1}; maize 170 kg N ha^{-1}), but amount of N applied by organic residues was variable (Table 1).

The field was divided in plots of 45m^2 and the experimental design was *randomized blocs*, with 3 replications. In order to measure yield, plants (at milky/farinaceous grain stage) of middle plots were harvested in the surface of 2.25 m^2 and 0.5 m^2 for maize and oats, respectively. AE was defined as the ratio of forage yield with N application minus forage yield without N application to N application. ARE was defined as the ratio that total plant N uptake with N application minus total plant N uptake without N application, then divided by N application and multiply by 100.

Results and Discussion

The forage production (Table 2) were different ($P < 0.001$) in the two years (mean production of 19.4 and 27.9 t DM ha^{-1} in 1st and 2nd year, respectively). Different climatic conditions between years contributed to this result. In any fertilization system was observed higher forage production than that obtained in MIN, and lower results were particularly observed in Control (59% of the forage yield in MIN) and with soil incorporation of urban waste compost (75 and 72% of the forage yield in MIN, in UWC and UWCm, respectively).

In general, the N use efficiency was lower when organic residues were used in the crops fertilization. For instance, with slurry application, forage yield per unit of N corresponded to 69 and 60% of that measured in MIN in 1st and 2nd year, respectively. However at the end of the trial, very similar results were observed between MIN and SSm. DCD didn't promote important changes in AE, namely when used with mineral fertilizers and when applied in spring fertilization (data not show). ARE values were around 75-80% with the utilization of mineral fertilizers and between 20 and 50% with organic residues incorporation. The lower value was obtained in UWCm and DCD didn't produce an evident effect on N recovery efficiency when added to the slurry or to the mineral fertilizer, namely in spring fertilization (data not show). Better results of ARE were obtained with maize than with oats (data not show), when N was incorporated through slurry or sewage sludge (37 and 43% N applied in CS and SSm in spring; 31 and 16% of N applied in CS and SS in autumn).

Table 1. Amounts (kg ha^{-1}) of N applied in each crop and treatment, through organic and mineral fertilizers.

Treatment	Oats			Maize		
	Organic fertilization	Mineral fertilization		Organic fertilization	Mineral fertilization	
		Sowing	Top-dressing		Sowing	Top-dressing
Control	0	0	0	0	0	0
MIN	0	30	50	0	90	80
MIN+I	0	80	0	0	170	0
PMS	10	20	50	0	90	80
SS	80	0	0	90	0	80
SSm	0	30	50	170	0	0
UWC	80	0	0	90	0	80
UWCm	0	30	50	170	0	0
CS	80	0	0	170	0	0
CS+I	80	0	0	170	0	0

Table 2. Forage production, agronomic N efficiency and apparent N recovery efficiency in different treatments and years.

Year	Control	MIN	MIN+I	PMS	SS	SSm	UWC	UWCm	CS	CS+I
Forage production (t DM ha^{-1})										
I	16.0cd	23.2ab	23.4ab	18.4bcd	19.5bc	25.9a	13.13d	17.6bcd	21.0abc	16.1cd
II	16.7e	32.6ab	33.2a	32.6abc	28.4abcd	28.3ab	28.5cde	22.4de	26.2abcd	30.3bcd
Agronomic N use efficiency (kg DM kg^{-1} N applied)										
I		29.1ab	29.6ab	9.8bcd	14.0bc	39.9a	-11.3d	6.6bcd	20.1abc	0.6cd
II		63.5	65.9	63.5	46.6	46.4	47.1	22.7	38.1	54.3
Apparent N recovery efficiency (% of N applied)										
I		60.8a	48.7ab	39.2bc	37.4bc	45.9ab	19.9d	13.8d	28.4cd	15.6d
II		85.6abc	110.7a	97.3ab	65.6bcd	48.6def	72.2bcd	28.0f	37.2ef	64.4cde

Conclusion

The amount of N removed from the soil through the vegetal material collected was mainly related with dry matter production, and was greater with more intensive use of mineral fertilizers. It was in UWCm treatment that was measured the lowest value of N removed by plants (21% of N applied). In order to increment N use efficiency with soil application of this residue, it is recommended simultaneous incorporation of mineral N. The same strategy should be considered when a waste with higher C/N ratio is used in crops fertilization. In similar cultural systems, is recommendable the soil application of slurry and sewage sludge in spring.

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EFFECTS OF TREES ON N2 FIXATION AND SOIL RHIZOBIUM ABUNDANCE IN AGROFORESTRY SYSTEMS

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The agroforestry system in Mediterranean region known by "montado" in Portugal consists of a mixed cropping system which features a widely spaced tree stand and a ground cover of arable crops or permanent pastures. Biodiverse pastures have been introduced in these systems. During the establishment, legume development is frequently restricted by competition for light and water by companion plants, often decreasing to less than 10% of initial vegetation after 2-3 years. The tree canopy in "montado" system may influence soil moisture and both the quality and quantity of light reaching the understory vegetation in a limited area of these systems. This study took place in four "montado" systems to evaluate the influence of a cork tree canopy on pasture composition, persistence, N2 fixation and soil Rhizobium abundance.

Material and Methods

In 2010 and 2012, microplots were arranged as split-plot in four "montado" systems located at two sites in south Portugal: Estremoz, a natural pasture >25-yrs-old and two improved pastures >5- and 12-yrs-old, and Vaiamonte with an improved pasture >30-yrs-old. Improved pastures were sown in the autumn with a mixture of Trifollium spp., Ornithopus sativus Brot., Lolium multiforum Lam. and Dactylis glomerata L. at a rate of 25-30 kg seeds ha-1. Seeds were inoculated with Rhizobium sp. Each site included a central cork oak tree (Quercus suber L.). Two sets of plots were installed in each site, one set consisting of three replicates under the tree canopy, and other three plots out of the canopy influence. Soils were classified as Dystric Cambisols at Vaiamonte and Eutric Luvisols at Estremoz. Soil samples were collected in December 2010 and 2012 to evaluate native rhizobial population by Most Probable Number. In both regions, average mean monthly temperature varied from 10 to 25 ºC, and annual rainfall was 800 and 350 mm, respectively at Vaiamonte and Estremoz. After plant emergency, 3 kg N ha-1 as 15NH415NO3 atom 5% 15N was applied to each plot to evaluate N2 derived from atmosphere (%Nda). In February 2011 and 2013, plant samples were harvested and new microplots were arranged in each site. In each new plot, aerial plant material was discarded and 15N fertilizer was added at the same rate and form as above. In April 2011 and May 2013, plants were harvested as before. Plant material was separated into legumes and non-leguminous plants, and roots and aboveground material to determine the dry weight, total N and %15N enrichment. N2 fixed by legumes was estimated by dilution method, using the adjacent non-fixing plants as controls.

Results and Discussion

The natural and improved pastures differed in vegetation growth (Table 1), the 12-yrs-old improved pasture showing the highest biomass, equivalent to 1574 kg DW ha-1 for the mean effects of cuts, years and canopy. Other pastures did not differ among each other (900 kg DW ha-1). Non-legumes dominated the swards (Table 1) only in the 1st cut under the canopy. After 5-, 12- and 30-yrs-old, composition of improved pastures was based on Lolium sp. (Vaiamonte) and Phalaris sp. (Estremoz) as non-legume species, whereas composite and Plantago plants were the dominant non-fixing plants in the natural sward at Estremoz (>25-yrs-old). Among the species introduced, only Lolium sp. survived after these years, in particular at Vaiamonte. The dominant legume in all swards was the subclover (>90%). %Nda by legumes was significantly higher in 2013 (74%Nda) compared with the 1st year (63%Nda) (Fig. 1). It was smaller in the oldest pasture (Vaiamonte) (54%Nda) compared with the others (73%Nda) which did not differ from each other. Overall, N2 fixation was relevant and oak tree canopy did not influence the fixation rate (67%Nda).

Table 1. Interaction of tree canopy on plant dry weight (g DW m-2) for the mean effect of years, pasture type and cuts, and ANOVA results for total herbage biomass

Source of variation	**Legume dry weight ($g\ DW\ m^{-2}$)**	**Non-legume dry weight ($g\ DW\ m^{-2}$)**
Tree canopy		
Under	53.2c	163.4a
Out	105.2b	105.6b
ANOVA	**F-value**	
Year (Y)	28.1***	
Tree canopy (T)	Ns	
Pasture type (P)	19.0***	
Cut (C)	150.9***	
Plant species (S)	49.2***	
Some interactions:		
YxC	17.1***	
PxC	3.7*	
TxC	15.6***	
TxP	6.8***	
CxS	48.5***	
TxCxS	3.9*	

ANOVA=Analysis of Variance; ns, *, *** = F-values not significant (p>0.05), and significant for p< 0.05 and p< 0.001, respectively; in each column, means with different letters are significantly different (p< 0.05) according to Bonferroni's test.

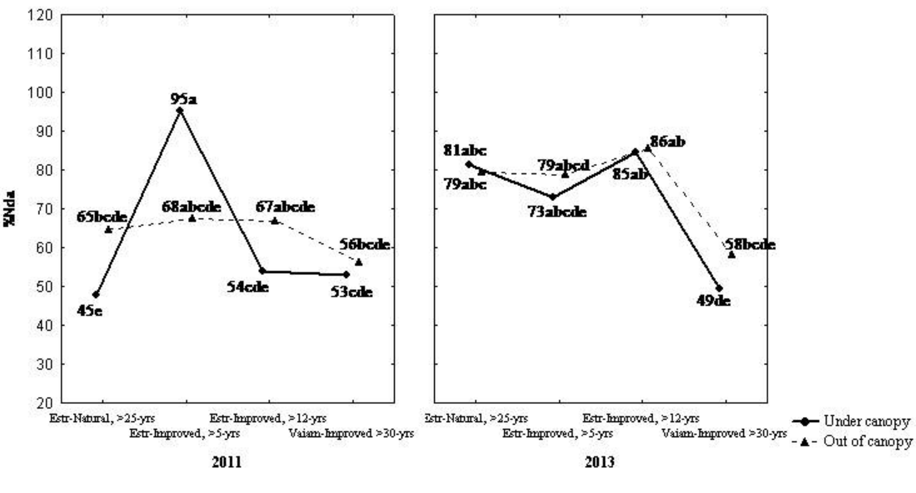

Fig. 1. Nitrogen derived from fixation (%Nda) measured in 2011 and 2013 in different pastures at south Portugal under the influence of a cork oak canopy, for the mean effect of cuts.

A decline of indigenous rhizobial population in the soil was observed under the cork oak canopy in improved pastures >30- and 12-yrs, which did not differ from each other (2.9x102 and 1.1x103 bacteria g-1 soil, respectively in the 1st and 2nd year), compared with outside the canopy influence (>104 bacteria g-1 soil in both years). This result was also observed in counts at Estremoz >5-yr, whereas in natural pasture 103 bacteria g-1 soil were found in all treatments.

Conclusions

Tree canopy did not influence symbiotic fixation in natural and improved mixed swards. Understory legume biomass was only significantly reduced during the winter. Older improved pastures (>12- and 30-yrs-old) contained a smaller rhizobium population under the tree canopy, but this effect was not confirmed in the younger and the natural swards.

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MAIZE NITROGEN RECOVERY AFTER APPLICATION OF A DIGESTED DAIRY COW SLURRY, ITS LIQUID AND SOLID FRACTIONS, AND OF A DAIRY COW SLURRY

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Knowing the crop recovery of N applied with livestock slurries is essential to formulate nutrient management plans that reduce economic costs and environmental pollution. After soil application, NH_4-N contained in digested and undigested slurries can be readily available for plant uptake or immobilised by the soil microbial biomass, and subsequently remineralised. Differently, slurry organic N is slowly mineralised. Incubation experiments suggest that in the short term most of the N fertilising value of slurries is due to their NH_4-N content, while the organic fraction is responsible of residual effects in subsequent years. In the three-year field experiment described here, we verified this possibility, comparing ammonium sulphate (AS), a digested slurry (DS), its solid (SF) and liquid (LF) fraction, and a raw cow slurry (SL).

Material and Methods

The field, located in Montanaso Lombardo (Italy), did not receive any organic fertilisers in the previous ten years. The 0-30 cm soil layer has: loam texture, pH(H_2O) 5.8, total N and organic C (% DM) 0.10 and 0.84. Six treatments were established with four replicates in a randomized block: the five fertilisers and a control without N fertilisation (CO).

On 31 May 2011, 17 May 2012 and 12 June 2014, fertilisers (Table 1) were applied immediately before ploughing at equal NH_4-N input. CO and AS were fertilised with 92 kg P_2O_5 ha^{-1} and 277 kg K_2O ha^{-1}. Maize (*Zea mays* L.) was sown shortly after fertilisation and harvested at milky-waxy maturity on 13 September 2011, 30 August 2012 and 3 October 2013; within 15 days, Italian ryegrass (*Lolium multiflorum* Lam.) was sown, without further fertilisation. Apparent nitrogen recovery of maize was expressed as a fraction of the total N (ANR) or of NH_4-N ($ANR_{NH4\text{-}N}$) applied. The mineral fertiliser equivalent (MFE) was calculated as the ratio of the ANR (or $ANR_{NH4\text{-}N}$) of each organic treatment to the ANR (or $ANR_{NH4\text{-}N}$) of AS. ANOVA was carried out separately for each year, using HSD Tukey test ($P < 0.05$) for mean separation. The treatment was considered a fixed factor. Homogeneity of variances was tested using the Levene test ($P < 0.05$).

Results and discussion

Maize N recovery in AS (Figure 1a-1b) was quite constant and high in all years ranging from 68% to 82%. Significantly differences in ANR and $ANR_{NH4\text{-}N}$ among the organic materials occurred only in 2011 (with the exception, for ANR, of LF and SF in 2013). In 2012 and 2013 the ANR of the organic materials was in the range 17-50%, while $ANR_{NH4\text{-}N}$ ranged from 44 to 98%. MFE and $MFE_{NH4\text{-}N}$ (Figure 1c-1d) increased from 2011 to 2013 in all treatments. In 2013, $MFE_{NH4\text{-}N}$ ranged from 82 to 133%, indicating that after three years the efficiency of applied NH_4-N was very close to that of mineral fertilisers. There were marked differences in the trend of $ANR_{NH4\text{-}N}$ among

Table 1. Characteristics of the fertilisers used in the field experiment (average values of the three years ± standard deviation).

Treatment	DM (%)	C (% DM)	Total N (% DM)	NH_4-N (% DM)	C/N org.	pH (H_2O)
DS	6.1 ± 0.4	38.5 ± 1.4	6.0 ± 0.4	2.9 ± 0.3	12.1 ± 0.8	8.1 ± 0.1
LF	4.7 ± 0.6	36.8 ± 1.4	6.6 ± 0.1	3.4 ± 0.1	11.3 ± 0.5	8.1 ± 0.2
SF	27.6 ± 2.0	43.6 ± 0.4	2.2 ± 0.1	0.6 ± 0.1	28.5 ± 4.2	9.4 ± 0.4
SL	6.8 ± 2.6	42.4 ± 1.5	4.7 ± 1.0	2.4 ± 0.5	19.5 ± 4.5	7.5 ± 0.3

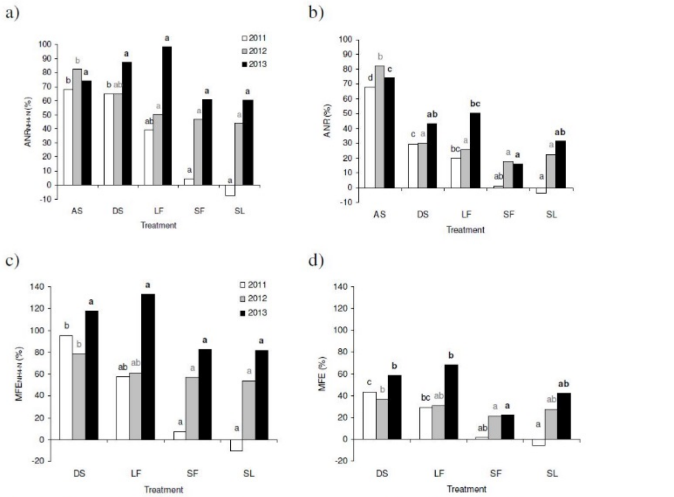

Figure 1. Apparent nitrogen recovery and mineral fertiliser equivalent of applied total N and NH_4-N. Within the same chart and the same year, data having different letters are significantly different ($P < 0.05$).

the organic treatments. In the three years $ANR_{NH4\text{-}N}$ was positive and relatively constant in DS (and less in LF), while in SF and SL it increased substantially. This could be ascribed to a different attitude of the materials to immobilise N and to produce residual fertilising effects. As DS and LF mainly contained stabilised organic matter with a low C/organic N ratio, they did not likely immobilise N and had a rather low residual effect on the following crops; most of the NH_4-N applied was available for maize during the year of application. Differently, SF and SL, because of a high content of more labile organic matter (SL) and a high C/organic N ratio, substantially immobilised N during several months after application and produced residual effects during the following crops.

Conclusions

The fertilising value of DS and LF was always positive, while it increased after repeated applications due to residual effects for SF and SL. To manage digested and undigested slurries it will be necessary to consider their residual effects in the years after application, which will become increasingly important when the number of repeated applications increases.

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RELEASE OF PLANT AVAILABLE NITROGEN FROM THE SOLID FRACTION OF TWO DIGESTATES

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Anaerobic digestion of animal manures and agroindustry wastes for energy production have been increasing, and digestates progressively substitute animal manures in crop fertilization. Separation of solid fraction from digestates is a common practice in digestion plants because of simplification in slurry storage and application. Sometimes separation and exportation of solid fraction are required by the law, to reduce nitrogen load for unit of land area. Plant available nitrogen released after application to soil of digestate solid fractions (DSF) is a basic information to calculate affordable fertilization plans. Recovery in solid fraction of most fibrous materials belonging to the raw digestate could results in a substancial nitrogen immobilization after application of solid fraction to soil. However, studies on DSFs mineralisation are still scarce. To contribute to fill this gap we studied the release of mineral nitrogen and its plant uptake in a soil amended with two different DSF. For this pourpose, a soil incubation experiment and a greenhouse pot experiment were conducted.

Material and Methods

In both experiments we used the same soil, DSFs, and doses of DSFs. The soil was sandy loam, pH(H_2O) 6.5, total N and organic C (g/kg DM) 0.97 and 7.81, C.E.C. (cmol(+)/kg) 5.39, Olsen-P and exchangeable K (mg/kg) 10 and 55. To increase soil fertility an equivalent amount of 23 kg/ha of P and 161 of K were added using potassium sulphate and superphosphate. Two DSFs (Tab 1) were obtained from a cattle livestock farm (CLF) and a slaughterhouse (SH). DSFs were applied to soil at a dose corresponding to 340 kg/ha of total N. In the incubation experiment 3 treatments were established with 4 replicates in a randomized design: the two DSFs and a control without N fertilisation (CO). To carry out the incubation with destructive samplings, the nursery experimental design described by Thuriès et al. (2000) was adopted. Each experimental unit consisted of a 250 mL large-neck plastic bottle containing an amount of wet soil (50% of WHC) corresponding to 120 g dry weight, and were incubated in the dark at 25°C and soil water content was adjusted weekly. On Day 0, 2, 4, 7, 14, 28, 42 and 63 after addition of DSFs, respired CO_2-C (data not shown) and soil nitrate and ammonium concentration were measured. In the pot experiment 4 treatments were established with 4 replicates in a randomized block design: the two DSFs, a control without N fertilisation (CO) and control fertilised with ammonium sulphate (AS). AS was applied at 100 kg N/ha. Pots contained 2.12 liters of soil at a bulk density of 1.3 kg/l. *Lolium perenne* (L.), cv. Pamir, was sown and after emergence 35 plant per pot were left. Greenhouse conditions were 16 h light (150-180 W/m^2) + 8 h dark per day, 22-30 and 16-22 °C during light and dark. Soil water content was maintained from 25 to 50% WHC by weighing and irrigating pots every one or few days depending on evapotranspiration. Aboveground biomass (AGB) was cut in November 12 2013, 61 days after plant emergence (66 after sown). Dry matter and total N concentration in AGB and soil mineral N concentration were determined. Apparent nitrogen recovery in plants (ANR) was calculated as the ratio of N plant uptake (net of CO) to N applied with fertilizers. Mineral fertilizer equivalent value (MFE) was calculated as the ratio of the ANR of DSFs to ANR of AS.

Table 1. Characteristic of the DSFs used in the incubation and pot experiments

Solid Fraction	DM (%)	C (% DM)	Total N (% DM)	NH_4-N (% DM)	C/N
CLF	31.95	45.25	6.36	0.95	20.82
SH	21.14	43.88	13.14	2.45	6.35

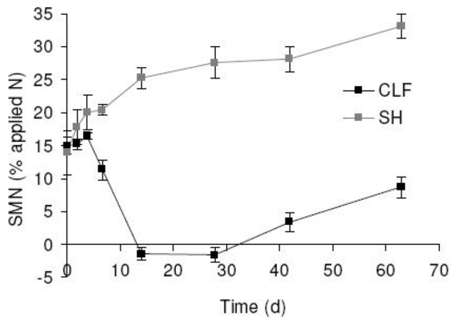

Figure 1. Dynamic of soil mineral nitrogen concentration after DSFs application in the incubation experiment.

Table2. Results of the pot experiment.

Treatment	AGB (g/pot)[a]	N in AGB (%)[a]	ANR (%)[a]	MFE (%)[b]
CO	2.5 a	1.14 a	--	--
AS	4.8 c	1.44 b	67 c	--
CLF	3.4 b	1.37 b	8 a	11
SH	6.1 d	1.75 c	33 b	48

[a]Data having different letters are significantly different ($P < 0.05$) according to the HSD Tukey test.
[b]Means are significantly different ($P < 0.05$) according to the T-test.

Results and discussion

Results of the incubation (Fig 1) showed a substantial N immobilisation in CLF until day 30 followed by an increase of SMN; at the end of the experiment, net organic N mineralization (NNM) was still negative. Differently, SMN concentration in SH increased during all the incubation period. The high C/N ratio of CLF can at list in part explain the strong N immobilization detected. In the pot experiment there was an increase in AGB and in the concentration of plant N in all fertilised treatments compared to CO (Tab 2). This increase was significantly higher in SH than in CLF. After 60 days, ANR of CLF was significantly lower compared to SH: 8% vs 33%. ANRs of the two DSFs strongly agreed with the values of SMN measured at the end of the incubation. According to ANRs, MFE of HS was also higher compared to CLF (Tab 2).

Conclusions

Results of the incubation and pot experiments were in strong agreement, confirming the importance of laboratory incubations to estimate potential plant available N. Nitrogen fertilising values of DSFs can be very variable and the determination of their ability to supply N for plants is reccomended to avoid N deficit or surplus in fertilisation plans.

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ROOT SOCIALIZATION: THE BASIS FOR IMPROVED PRODUCTIVITY AND NUTRIENT USE EFFICIENCY

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The world's population is expected to increase from 7 billion now to 8.3 billion by 2025. Humanity will need 70-100% more food by 2050, so the production of cereals, especially wheat, rice and maize, which account for half of human calorie intake, must be increased. Plant growth has already been enhanced by the input of chemicals, which act as plant growth regulators and as nutrients. The nutrients nitrogen and phosphorus are, together with potassium, applied to the soil as chemical fertilizers to improve grain yield. According to Roberts (2009) the present annual uses of chemical nitrogen, phosphorus and potassium fertilizers is 130, 40 and 35 million tonnes. The high input of chemicals to the soil raises a number of concerns such as water contamination leading to eutrophication and health risks for humans. Moreover, it results in soil degradation and loss of biodiversity. In this work we describe the benefits of exploring the relation between arbuscular mycorrhizal fungi (AMF) and plant growth promoting bacteria (PGPB), which in association with fertilizers may improve productivity and nutrient use efficiency.

Materials and Methods:

A four-block randomized field assay was established on May of 2012 in Beja (Escola Agrária de Beja, Portugal). Maize (Zea mays L. cv Sincero) was used to test the synergies between distinct microbial inoculants and fertilizers on plant growth. In each block there were 5 treatments: A) 100% of the dose of fertilizer recommended for this culture and soil type, 150 kg/ha of NPK fertilizer (18-46-0); B) 67% of the fertilizer applied in A; C) B + AMC inoculum; D) C + AMF inoculum; E) B+C+D. Each block had an area of 330 m2 and each treatment unit (within a block) had an area of 30 m2, the total assay area per treatment was 120 m2. The bacterial inoculant (AMC) consisted of *Bacillus megaterium*, *Saccharomyces ceravisae*, *Azospirillum brasiliense*, *Azotobacter crococcum*, *Rhizobium loti*, *Bacillus licheniformis* and *Bacillus pumilus*, which were incorporated in the fertilizer granules by the fertilizer company (1 L/t). The AMF inoculum was Glomus intraradices, which was directly applied in the field together with the seeds at a dosage of 5g/m2 (2 000 000 propagules /kg).

During the growing cycle, nutrients were added to the irrigation system daily: (205 $kg.ha^{-1}$ N; 3.5 $kg.ha^{-1}$ Ca; 1.77 $kg.ha^{-1}$ Mg; 1 $kg.ha^{-1}$ Zn). A mixture with 9% N, 37% organic matter, 23% organic carbon and 24% of free amino acids was applied once at 5.5 $kg.ha^{-1}$.

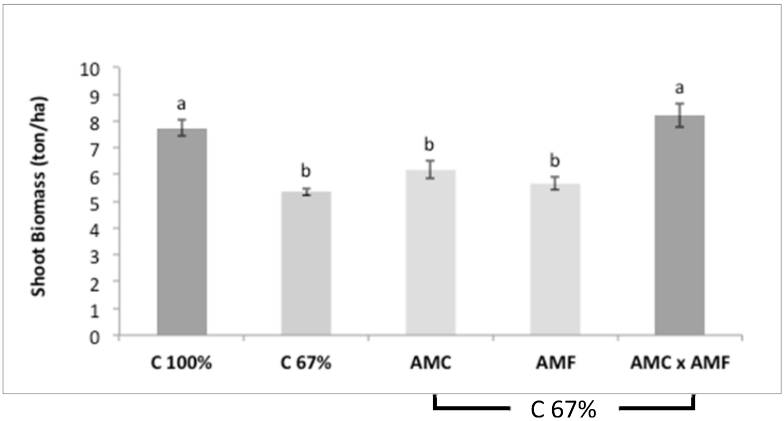
Seventy-five days after sowing, the aboveground parts of the plants were harvested, and analysed for biomass accumulation and nutrient content. Soil samples were also collected and analysed for nutrient content. The nutrient use efficiency was calculated for each treatment. Data were statistically analyzed through one-way ANOVA followed by Tukey post-hoc test (SPSS).

Results and Discussion:

Results show that at the flag leaf stage, maize plants fertilized with 100% of the recommended dose of fertilizer accumulated more biomass than those fertilized with only 67%, showing that the recommended dose was not overestimated. No significant differences in biomass accumulation were found among plants treated with 67% of the recommended dose of fertilizer, bacterium inoculum (AMC) or mycorrhizal inoculum (AMF). Therefore all these treatments resulted in smaller plants than 100% of the recommended dose of fertilizer. However, the simultaneous use of 67% of the recommended dose of fertilizer, the bacterial and the mycorrhizal inoculant originated plants with biomass similar to those treated with 100% of the recommended fertilizer dose. Although these results refer to the accumulation of vegetative biomass they show that there are ways of reducing the amount of the fertilizer applied to crops without reducing the productivity. The higher biomass accumulation in the plants treated with 67% of the recommended dose of fertilizer, the bacterial and mycorrhizal inoculant may be explained in the light of the biotic and abiotic interactions established between plants and microorganisms in the rhizosphere. Exploring the positive interaction between organisms and stimulating the formation of microbial consortia may be a very important strategy in the way to reduction the fertilizer application in 33% (PAC recommendation), which represents a big potential to decrease the environmental impacts of agriculture. Further studies are needed in order to assess: 1) the effects of the distinct treatments on grain productivity, and 2) how sustainable the process is.

Acknowledgements:

AMC Chemicals & Trichodex for PGPR and ADP-Fertilizantes for partnership and Fundação para a Ciência e a Tecnologia (FCT) of Portugal, project PTDC/AGR-PRO/115888/2009 for financial support.



Shoot biomass of maize plants **75 days** of sowing. Values (means±SE of 4 replicates) followed by different letter are significantly different at $P<0.05$ (Tukey *post-hoc* test).

VARIATION IN NITROGEN CYCLES ON DUTCH DAIRY FARMS

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In dairy farming systems, four major components can be distinguished, i.e, herd, manure, soil and crop (Fig 1). Nitrogen (N) cycles through these components: output from one component is input into another but losses are incurred in these transfers (Oenema, 2013).

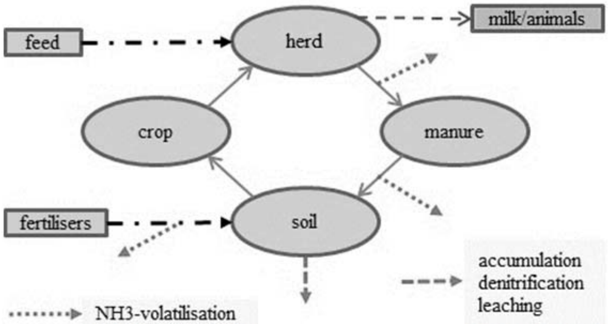

Figure 1 Nitrogen cycle for dairy farms

Dividing nitrogen output in milk and animals by nitrogen input in feed and fertilisers results in the nitrogen efficiency at farm level. Changes in stocks are also taken into account. More efficient use of feed and fertilisers results in a better cycling of nitrogen and thus in lower losses to the environment and less costs for purchases. Efficiency is partly governed by conditions that the dairy farmer can’t affect like soil type or weather conditions. However, management generally is the most dominant factor (Daatselaar et al, 2009).

To monitor and stimulate improvement in nitrogen efficiency the instrument Annual Nutrient Cycle Assessment (ANCA, in Dutch: Kringloopwijzer) is developed and introduced. Indicators, calculated by ANCA, enable dairy farmers to optimise the farm management as well as to justify this management towards authorities and the dairy processing industry (Oenema, 2013). This development underpins the need for the dairy farmer to know his position concerning the nitrogen efficiency. The objective of this study is to show the variation in nitrogen efficiency at farm level, even if soil type and intensity (ton milk/ha) are quite equal and to discuss the potential for further improvement in nitrogen efficiency.

Materials and methods

Since 1991, the Dutch Minerals Policy Monitoring Program (LMM) has collected data on farm practice and water quality for both a representative sample of farms in the Netherlands and some pilot farms like the participants in the Dutch project Cows and Opportunities. The analysis in this study concerned 277 dairy farms in the period 2009-2011. The farms were divided in the soil types clay, peat, dry sand and wet sand.

Within these soil types the farms were divided, based on the milk production per ha: < 13 ton, 13-16 ton and >16 ton. Due to the limited number of farms on peat soil only 2 classes were distinguished: < 13 ton and >=13 ton. Relevant data of these farms on farm practice were processed through the ANCA-instrument to obtain figures on nitrogen efficiency and other outcomes. Farmers can use the ANCA-instrument to calculate these outcomes for their own farm: then the average results of the corresponding group of LMM-farms are presented to the farmer as a benchmark. Hooijboer et al (2013) give a detailed description of the nitrogen input and output (for the calculation of the nitrogen efficiency) in the yearly derogation monitoring report which is one of the products of the LMM program.

Results

Table. 1. Averages for some indicators on Dutch dairy farms in the period 2009-2011. Farms divided by soil type and ton milk production per ha

Soil type	Clay			Peat		Dry sand			Wet sand		
Ton milk/ha	< 13	13-16	> 16	< 13	> 13	< 13	13-16	> 16	< 13	13-16	> 16
No of observations	36	21	16	29	21	22	12	27	46	25	20
Ton milk/farm	754	922	1358	533	931	534	811	1002	533	673	955
Ton milk/ha	9.8	13.8	17.9	10.3	15.0	9.9	14.1	20.8	9.7	14.1	19.8
Kg N soil surplus/ha	121	176	188	111	127	123	132	145	111	137	150
N-efficiency farm (%)	29.6	27.1	31.8	32.4	34.0	31.6	33.8	39.5	32.6	32.3	36.9

For every soil type it applies that the farms with a higher milk production per ha have a higher total milk production. Also, in most cases, both the nitrogen soil surplus per ha and the nitrogen efficiency are higher if the milk production per ha is higher. More intensive dairy farms rely more on purchased feed by which they avoid the nitrogen losses that would accompany the cultivation of forage crops on their own farms. However the bigger dependence on purchased feed can also lead to really better efficiency.

Besides the variation *between* the groups there is also variation *within* (figure 2). The boxes show the 25%-75% interval and the whiskers depict the 10%-90% interval. To avoid the visibility of individual observations the 10%-90% interval has been chosen instead of the more common minimum and maximum values for the lengths of the whiskers.In most groups the 10% farms with the highest nitrogen efficiency at farm level achieve a 1,5 fold or more higher nitrogen efficiency than the 10% lowest. In many groups the distance between the 25% highest and the 25% lowest in nitrogen efficiency is more than a factor 1.2. So a considerable number of farms, being in the same circumstances (soil type and intensity), have room to improve their nitrogen efficiency. No group differs significantly from another group at a confidence level of 90%: all groups have observations with a nitrogen efficiency of (or close to) 30%.

Conclusions

Milk production per ha and per farm are positively related on Dutch dairy farms. Both indicators also have a positive relation with the nitrogen surplus per ha as well as the nitrogen efficiency. When correcting for milk production per ha and soil type by division into groups considerable variation in nitrogen efficiency occurs in the different groups. These variations offer possibilities for many dairy farmers to improve the nitrogen efficiency, starting with benchmarking and followed by suitable improvement measures.

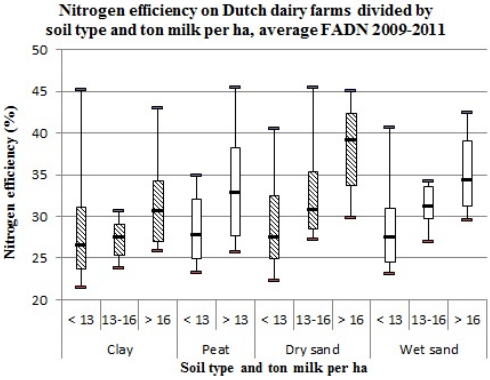

Figure 2 Nitrogen efficiency on Dutch dairy farms

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AZOFERT®: A DYNAMIC DECISION SUPPORT TOOL FOR FERTILIZER N ADVICE ADAPTED FOR ORGANIC PRODUCTS AND CATCH CROPS

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The increasing demand for high quality crops (protein content of cereals, technological quality of sugar beet, nitrate rate of vegetables) and protection of the environment (minimising nitrate leaching and gaseous losses) on the one hand, the evolution of agricultural practises with increasing and diversification of organic applications on the other hand, require an adaptation of reasoning and a rigorous management of the N fertilization, as well as an evaluation of environmental impacts.

Materials and Methods

AzoFert® is based on a complete inorganic N balance sheet (Meynard et al, 1997). The following equation is used to predict fertiliser-N rates, expressing that the variation of soil inorganic N between opening and close of balance sheet equals the difference between N inputs and outputs:

$$Rf - Ri = (Mn + X + Ap + Fns + Fs + Ir) - (Pf - Pi + Ix + Gx + Lx + Gs + Ls)$$
$$\text{With } Mn = Mh + Mr + Ma + Mci + Mp$$

Rf: soil inorganic N at close of balance sheet (at harvest), **Ri**: soil inorganic N at opening of balance sheet (end of winter for winter crops, sowing for spring crops), **Mn**: net mineralization from humus (**Mh**), crop residues (**Mr**), organic products (**Ma**), catch crops (**Mci**) and meadow (**Mp**) residues, **X**: amount of fertilizer N, **Ap**: N wet deposition, **Fns**: non symbiotic fixation, **Fs**: symbiotic fixation, **Ir**: N irrigation contribution, **Pf**: total N uptake by crop at close of balance sheet, **Pi**: N uptake by crop at opening of balance sheet, **Ix**: fertilizer N immobilised, **Gx**: fertilizer N lost as gas, **Lx**: fertilizer N lost by leaching, **Gs**: soil inorganic N lost as gas, **Ls**: soil inorganic N lost by leaching between opening and close of balance sheet.

AzoFert® integrates a dynamic simulation of soil N supplies (Machet et al., 2012). At the opening of the balance sheet (end of winter for winter crops, at sowing for spring crops), the soil inorganic N pool is measured at the rooting depth. In order to take into account the various contributions of crop residues, catch crops and organic products previously applied to the residual mineral N (varying with climate and characteristics of added organic matter), the decomposition of the different organic sources are simulated (using observed climatic data) from harvest of the previous crop, until the opening of the balance sheet. Decomposition is expressed over time using a "normalized time", based on temperature (T) and soil moisture (W) functions:

Normalized time $_{(\text{day i, day j})} = S_{ij}\ f(T) * g(W)$

Normalized time takes into account climatic variations and determines a potential rate of decomposition. From the opening of the balance sheet to the harvest of the crop, the subsequent net contribution of the organic residues or wastes and the net mineralisation of the humified organic matter are simulated using normalised days calculated from the past years mean climatic data of the area.

Results and Discussion

The decay of crop residues and organic amendments in the soil results in net mineralization or net immobilization of soil nitrogen. Each crop residue and organic product is characterised by a specific kinetic curve of decomposition according to N and C. The decay rate of these products depends on the nature of organic residues (chemical characteristics and C:N ratio) and temperature and moisture soil conditions (Nicolardot et al., 2001). AzoFert® estimates the direct effect of organic product or catch crop, using the curve of decomposition

Transposition in AzoFert® software is shown for vinasses and mustard catch crops residues, as indicated on figure 1.Thanks to this transposition in real time, it is possible to determine the mineralized part of products before the measure of soil mineral N pool at opening balance sheet, and the part which will be mineralized between soil mineral N measure and harvest (direct effect).

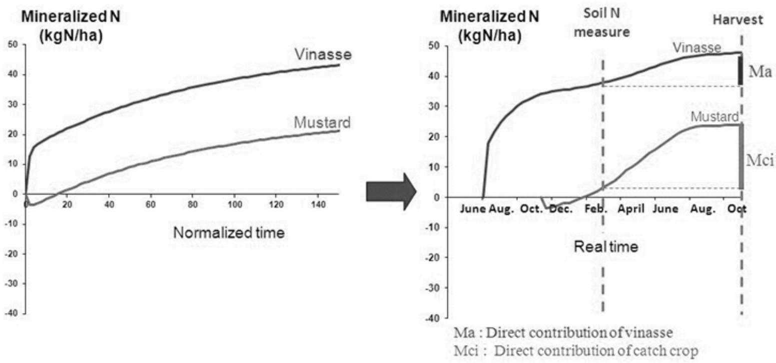

Figure 1: Curve of N decomposition in normalized and real time

A validation work was carried out by French Technical Institute for Sugar Beet (ITB) in order to test N recommendations. In the same farm, in deep loamy soils with frequent applications of organic products, there were annual (from 1992 to 1997) experiments on sugar beet, in order to know the optimal dose (table 1). Optimal dose varies a lot, from 0 to 180 kg N ha^{-1}, and AzoFert recommendation is close to optimal dose.

Table 1: Results of N annual experiments on sugar beet in the same farm

Experiment site	Soil inorganic N end of winter Kg N /ha	Humus mineralisation Kg N/ha	AzoFert dose Kg N/ha	Optimal dose Kg N/ha
Fieffes 1992	58	112	87	80
Fieffes 1993	79	131	0	0
Fieffes 1994	37	104	110	120
Fieffes 1995	33	84	165	180
Fieffes 1996	62	70	170	140
Fieffes 1997	99	139	40	0

Deep loamy soils with frequent applications of organic products

Conclusions

AzoFert® constitutes a decision support tool for fertilizer N advice based on a dynamic version of the predictive balance sheet method. The introduction of a dynamic simulation of soil N supplies allows its application to a larger range of cultural situations and pedoclimates. The integration of real data characterising the climate, soil type and cultivation practices leads to a significant improvement of N recommendations accuracy at field scale, especially with organic products and catch crops.

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PREDICTING YEAR-YEAR, FIELD LEVEL VARIATION IN MAIZE NITROGEN FERTILIZER REQUIREMENT

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Nitrogen fertilizer is a significant input for maize (*Zea mays* L) production, second in cost only to maize seed (Molenhuis, 2012). Considerable effort has been aimed at optimizing nitrogen fertilizer application rates so as to maximize profitability while limiting potential negative externalities associated with nitrogen fertilizer (Crews and Peoples, 2005). Maximum economic rate of nitrogen (MERN) have been shown to vary spatially within and across fields, but much less effort has been made to quantify temporal variation (Mamo et al., 2003). The objective of this research was to 1) assess using a long-term nitrogen trial established in 2009 at the Elora Research Station, University of Guelph, the year to year variation in MERN for a given field, and 2) assess whether existing nitrogen recommendation systems can effectively predict yearly variation in MERN for a given location.

Materials and Methods

A long-term nitrogen trial was established in 2009 at the University of Guelph, Elora Research Station ($43^0$39' N $80^0$25' W, 376 m elevation), Elora, Ontario, Canada. The trial consists of 5 nitrogen fertilizer rates (0, 28, 57, 115, and 188 kg-N ha^{-1}) injecting as urea-ammonium-nitrate (28%) solution at approximately the 6^{th} leaf stage mid-row to a depth of 5-10 cm. Each plot followed 115 kg-N ha^{-1} (Figure 1). All plots also received 30 kg N ha^{-1} starter fertilizer. Prior to UAN application soil nitrate (0-30cm) was assessed for each plot.

The experimental design is a factorial randomized complete block design with four replicates. The experimental unit consists of a six rows at 0.76m x 17m. The soil at the experimental site is a Guelph loam (fine loamy, mixed, mesic Glossoboric Hapludalf) with 32% sand, 48% silt, 20% clay, 4.5% soil organic matter, pH 7.7, and 1.36 ± 0.46 g cm^{-3} soil bulk density. The site's 30-yr total annual precipitation is 874 mm and mean annual air temperature is 6.7^{o}C. All plots are subjected to moldboard plowing in the fall of each year followed by spring disking just before planting.

Annual yield response to N rate was characterized by fitting a polynomial quadratic response model:

$Y=a+b(N)+c(N^2)$; (Equation 1)

Where: Y is maize yield (kg ha^{-1}), a is the fitted intercept, b is the fitted linear coefficient, c is the fitted quadratic coefficient, and N is the nitrogen rate (kg N ha^{-1}). The relative cost of nitrogen and value of grain maize influence MERN and is described by a price ratio coefficient (PR)

PR=\$N/\$Y; (Equation 2)

Where: \$N is the cost of nitrogen fertilizer expressed as \$ kg^{-1}, and \$Y is the value of grain maize expressed as \$ kg^{-1}. A maize price of \$220 Mg^{-1} and N cost of \$1.54 kg-N^{-1} was assumed. The maximum economic rate of nitrogen was determined by taking the first derivative of Equation 1 and rearranging to solve as follows:

MERN=(PR-b)/2c; (Equation 3)

Quadratic polynomials were fitted using Proc NLIN (Statistical Analysis System, version 9.1, SAS institute, NC, USA). Comparisons of N rate and profitability were made for MERN, Ontario General Maize N recommendations (OMAFRA, 2009) and the Pre-Sidedress Soil Nitrate test (OMAFRA, 2009).

Results and Discussion

Nitrogen response curves as well as MERN values varied considerably across years (Figure 2) even though management (N history, hybrid, tillage, and weed control) and soil type were constant across years. MERN values differed by 86 kg N ha^{-1} due to weather variability effects on soil nitrogen processes and crop growth and nitrogen demand. The relative consistency of the 0 kg N ha^{-1} rate suggests that nitrogen supply capacity was relatively constant across years. Neither the Ontario General Maize Nitrogen Recommendations, which accounts for soil type, previous crop, heat unit zone, maize:nitrogen price and expected maize yield, or the Pre-sidedress Soil Nitrate test were consistently effective in predicting the rate of nitrogen required (Table 1). Profit relative to MERN deviated by \$18-\$312 ha^{-1}. Interestingly, if actual yields could be predicted at the time of sidedress application, and the predicted value was incorporated into N rate calculation using the Ontario General Maize N Recommendations, then resulting estimates of nitrogen resulted in returns to nitrogen within \$10/ha of maximum (data not shown) Much of the variation in N requirement appeared to be determined by soil N supply and maize N demand requirements that were determined after decisions are made regarding sidedress nitrogen fertilizer rate, thus suggesting a need for a predictive modelling approach.

Figure 1: Nitrogen fertilizer treatments

Year	Nitrogen treatments (kg N ha^{-1})									
2009	0	28	57	115	188	115	115	115	115	115
2010	115	115	115	115	115	115	57	188	28	0
2011	188	115	28	0	57	115	115	115	115	115
2012	115	115	115	115	115	0	28	57	115	188
2013	115	188	0	28	57	115	115	115	115	115

Conclusions

Temporal factors are a significant cause of variation in maize N requirement. Existing nitrogen recommendation systems do not effectively account for this variation. Variation due to temporal variability may, in fact, be more difficult to predict than spatial variability, particularly since this variation appears to be caused by weather events that typically occur after nitrogen fertilizer decisions and applications are made. A predictive modelling approach must be evaluated as a means to improve nitrogen rate recommendations.

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Table 1: Comparison of MERN and rates derived from nitrogen recommendation systems

Year	MERN	NUE @MERN	Ontario General Recommendations: Recommended N rate	Ontario General Recommendations: Profit relative to MERN	Pre-Sidedress Soil Nitrate Test: Recommended N rate	Pre-Sidedress Soil Nitrate Test: Profit relative to MERN
	kg N ha^{-1}	kg kg N^{-1}	kg N ha^{-1}	\$ ha^{-1}	kg N ha^{-1}	\$ ha^{-1}
2009	155	21.8	145	-41.33	128	-18.41
2010	194	26.1	145	-150.49	138	-73.72
2011	184	24.6	145	-113.01	116	-107.45
2012	157	11.4	145	-18.54	101	-31.74
2013	241	27.9	145	-311.89	136	-215.18

Year	MERN	NUE @MERN	Ontario General Recommendations: Recommended N rate	Ontario General Recommendations: Profit relative to MERN	Pre-Sidedress Soil Nitrate Test: Recommended N rate	Pre-Sidedress Soil Nitrate Test: Profit relative to MERN
	kg N ha^{-1}	kg kg N^{-1}	kg N ha^{-1}	\$ ha^{-1}	kg N ha^{-1}	\$ ha^{-1}
2009	155	21.8	145	-41.33	128	-18.41
2010	194	26.1	145	-150.49	138	-73.72
2011	184	24.6	145	-113.01	116	-107.45
2012	157	11.4	145	-18.54	101	-31.74
2013	241	27.9	145	-311.89	136	-215.18

RESIDUAL SOIL MINERAL NITROGEN IN FUNCTION OF APPLIED EFFECTIVE AND CROP AVAILABLE NITROGEN

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Despite intense efforts eutrophication remains a major environmental concern across Europe, demanding a further reduction in nitrogen (N) losses from agricultural soils. Strict limitation of the N fertiliser application rates is considered as one of the best N management strategies to reduce residual soil mineral N (RSMN) at harvest and thus the potential risk of nitrate (NO_3^-) leaching during winter.

In the range from low to optimum fertilisation rates, most crops show a rather constant and low RSMN. This constant RSMN is considered to be the minimum mineral N buffer necessary for optimal growth (Hofman et al., 1981). When N fertiliser application rates are increased to rates above this optimum, RSMN shows a breakpoint and rises steeply.

The objective of this study is to optimise N fertiliser application rates for cut grassland, silage maize, potatoes, sugar beets and winter wheat in terms of a low RSMN whilst maintaining yield levels.

Materials and Methods

We used data from field trials with various N application rates carried out in Flanders and northern Wallonia (1991-2011). We analysed the dataset of RSMN (NO_3-N only) of the 0-90 cm layer (except for grassland and potatoes, where only the values up to 60 cm (rooting depth) were used). The RSMN in function of applied effective N and crop available N was calculated with the pooled data of the different trials. We tested the linear, segmented linear, exponential, quadratic and power model for the best fit of RSMN versus applied effective and crop available N. The breakpoint with the smallest confidence interval of segmented linear regression was calculated according to Oosterbaan et al. (1990).

Results and discussion

For grassland, segmented linear regression showed a breakpoint at 281±84 kg applied effective N ha^{-1} and at 514±109 kg crop available N ha^{-1} (Table 1), but even above these values RSMN remains below 50 kg N ha^{-1} up to 500 kg ha^{-1} applied effective N. The low RSMN can be explained by the ability of grassland to take up N efficiently even at high N application rates.

For silage maize, the RSMN increased exponentially in both scenarios. The maximum allowed effective N application rates for silage maize (in 2012 135-150 and 150-185 kg effective N ha^{-1} in Flanders and the Netherlands, respectively) (van Grinsven et al., 2013), correspond with a RSMN between 55 and 75 kg N ha^{-1}. These rather high RSMN lead to a high potential risk of NO_3^- leaching during winter.

For potatoes, a breakpoint was noticed for both scenarios. The breakpoint in function of the applied effective N was situated at 199 ± 77 kg N ha^{-1}. The RSMN at the breakpoint was about 70 kg N ha^{-1}. There is no ground for increasing the Flemish maximum allowed effective N application rates of 190 kg N ha^{-1} on sandy soils and 210 kg N ha^{-1}for non-sandy soils, given that the RSMN increases sharply after the breakpoint. A decrease of the N fertilisation limit is not recommended from an economic point of view, as the N fertilisation rate not only affects the total potato yield but also strongly influences the marketable yield.

For both scenarios, RSMN for sugar beets was low and constant over the entire range under study (0-160 kg applied effective N ha^{-1}). Despite low RSMN after sugar beets, NO_3^- leaching during winter remains possible. Only part of the N released from the sugar beet leaves can be found in the soil in the following spring. As harvest is often under wet soil conditions, part of the N of the sugar beet leaves is denitrified, but also part leaches.

For winter wheat a breakpoint in RSMN at 343±80 kg crop available N ha^{-1} and a continuous but small increase of RSMN versus applied effective N was found. The maximum allowed effective N fertilisation rates in 2012 (160-175 and 160-220 kg effective N ha^{-1} in Flanders and the Netherlands, respectively) (van Grinsven et al., 2013) result in low RSMN. However, without a catch crop, N mineralisation can lead to rather high mineral N in late autumn and a potential risk of NO_3^- leaching during winter.

Table 1 Residual soil mineral nitrogen (RSMN) at breakpoint as a function of crop available and applied effective nitrogen (N)

Crop	RSMN (+ st dev) for crop available N	RSMN (+ st dev) for applied effective N
Grassland	22 ± 12	18 ± 15
Silage maize	/	/
Potatoes	59 ± 26	72 ± 29
Sugar beets	12 ± 4*	13 ± 4*
Winter wheat	29 ± 14	/

*: RSMN is a constant value

Conclusions

RSMN is low for cut grassland, sugar beets and winter wheat due to their intensive rooting system. However, RSMN not only depends on the crop but also on crop available and applied effective N, except for sugar beets where RSMN is low independent of the amount of applied effective N. For winter wheat and silage maize, RSMN increases with a higher effective N application rate. The presence of a breakpoint between RSMN and applied effective N for grassland and potatoes gives the opportunity to deduce optimum N fertilisation rates limiting the potential risk of NO_3^- leaching during winter whilst maintaining yield levels.

Acknowledgement

We would like to thank Hooibeekhoeve, Inagro, Independent Centre for the Promotion of Forage (CIPF), Institute for Agricultural and Fisheries Research (ILVO), Provincial Institute for Biotechnical Education (PIBO),Royal Belgian Institute for Beet Improvement (KBIVB), Soil Service of Belgium (SSB), Test Centre for Potato Production (PCA) and Walloon Centre for Agricultural Research (CRA) for supplying the field data.

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ASSESSMENT OF SOIL-ATMOSPHERE N2O EXCHANGE IN PRE-ALPINE GRASSLANDS AND IMPORTANCE OF THE MONITORING TEMPORAL RESOLUTION

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Pre-alpine grasslands are ecosystems of high social, ecological and economical importance in Central Europe. Management of grasslands includes application of fertilizers to increase productivity. Besides positive effects in yields, use of fertilizers alters the nitrogen balance of the ecosystem and is responsible for nitrous oxide (N2O) emissions to the atmosphere, thus contributing to global warming. However, soil N2O emissions depend on a wide range of variables, including fertilizer type, application timing, soil characteristics and soil temperature and soil moisture conditions. Therefore it is essential to accurately quantify the contribution of different management practices Furthermore, in the frame of increasing temperature and rainfall patterns due to climate change, the soil N2O emissions are likely to change.

In this conference, we will present data from soil N2O emissions in grasslands as affected by application of fertilizers and climate change, as part a larger project. We will concentrate on crucial methodological aspects for accurate estimations of soil N2O emissions, with a special focus on temporal dynamics of N2O.

Materials and Methods

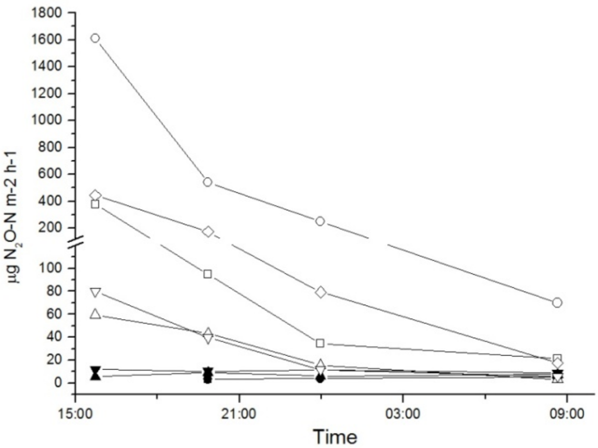
In the Ammer catchment -Southern Germany- a long term observatory has been established, within the TERENO project (Terrestrial Environmental Observatories, www.tereno.net). A translocation experiment has been conducted with big soil lysimeters (1 m2 section, 150 cm depth): they have been transplanted along an altitudinal gradient, acting as an *in situ* global change simulation experiment. In addition, two levels of “management intensity” were considered, varying in fertilization loads and cutting intensities. Specific information on the pre-alpine observatory of the TERENO project can be found at http://imk-ifu.fzk.de/tereno.php.

In order to monitor the greenhouse gas exchange processes in the atmosphere-pedosphere interface, an automatic measurement system has been implemented using a static chamber approach. Thus, a stainless steel chamber (1 m2 section, 80 cm height) is controlled by a robotized system. The chamber is hanging on a metal structure which can move both vertically and horizontally, so that the chamber is able to be set onto each of the lysimeters placed on the field. Furthermore, the headspace of the chamber is connected with a gas tube to an Aerodyne Quantum Cascade Laser, which continuously measures CO_2, CH_4, N_2O and H_2O mixing ratios. Such a system is implemented in two of the TERENO pre-alpine observation sites, with 12 and 18 lysimeters each, respectively. The chamber is set onto each of the lysimeters for 15 minutes and the increase in the concentration of the targeted gases in the chamber´s headspace is measured so that a soil-atmosphere exchange rate can be calculated. Each lysimeter is measured 5 to 8 times a day, depending on specific configuration.

Results and Discussion

The automatic system is capable to accurate calculate fluxes and the detection limit was estimated to be lower than 1 µg N_2O-N m^{-2} h^{-1}. N_2O emissions were largely affected by organic fertilizers –i.e. manure- applications. Therefore, high-intensively managed grasslands experienced higher N_2O emissions, due to higher fertilization loads. Freeze-thaw events also showed a significant contribution of the annual N_2O emission budget and such events occurred more often at lower altitudes in comparison to higher altitudes.

Interestingly soil pulse emissions of N_2O (as well as CH_4) happened in a very dynamic way after application of fertilization. For example, from a baseline emission rate of about 5-10 µg N_2O-N m^{-2} h^{-1} before application, soils turned into a very strong source of N_2O up to 1600 µg N_2O-N m^{-2} h^{-1} declining to about 500 in 4 hours, to 250 after 8 hours and to below 100 µg N_2O-N m^{-2} h^{-1} within a day (Figure 1). Weekly or even daily measurements may lead to a strong bias in the total N2O emissions: either underestimation if peak emissions are missed or overestimation if peak emissions are measured and wrongly interpolated in time.



Conclusions

As expected, fertilization application was closely related to the soil N_2O emissions. However, winter emissions due to freeze-thaw events were significant for estimation of annual budgets.

Sub-daily measurements are needed when aiming at an accurate estimation of greenhouse gas emissions from soils, especially after disturbances and management actions which may result in short-lasting pulses.

Acknowledgement

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IMPROVED PIG SLURRY AND CO-DIGESTATE MECHANICAL SEPARATION USING POLYMERS AND BIOCHAR

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Efficient separation of pig slurry and co-digestate into solid and liquid components is essential whenever removal of excess nutrients is required. Mechanical separation (typically achieved with screw presses and centrifugation) is quite effective in removing the dry matter and P from raw slurry and co-digestate. It fails, however, for N and K, where addition of flocculants may be required to enhance separator efficiency. Literature (Hjorth et al., 2010) indicates that organic, synthetic high polymer-polyacrylamide (PAM) is the most frequently chosen flocculant for manure treatment. Naturally occurring flocculants (e.g. chitosan) and biochar have received little systematic investigation to date on their potential to enhance solid—liquid manure separation. To our knowledge, there is also scarce information on the efficiency of co-digestate separation when flocculants are used prior to mechanical separation. Inclusion of pre-treatment based on biochar and chitosan should not increase separation costs markedly, given the fact that biochar and chitosan are by-products of larger processes (gasification and crabs or shrimp production, respectively), thus available at rather low costs. The principal hypothesis tested in our experiment was whether or not the addition of polymers and biochar (alone or in combination) improved the efficiency of screw press (SP) and centrifuge (CENT) separation. This was tested by observing the following indicators: dry matter, total N, ammoniacal N, and total P, K, Cu, and Zn.

Materials and methods

Fresh raw pig slurry (from fattening pigs) and co-digestate were sampled at two farms in Piedmont, Italy. The continuously stirred tank anaerobic reactor (CSTR) was run at a mesophilic thermal regime (at 40°C) with a retention time of 40 days and an average organic loading rate of 2.20 kg VS (volatile solids) m^{-3} d^{-1}. Approximately 750 l of raw pig slurry was collected from an agitated pre-tank over a period of 30 min while a total of 300 l of co-digestate was collected directly from the digester outlet into five 60 l barrels. Samples were transported to the laboratory and transferred to a tank in a fridge room at +5° C for the duration of the experiment. Screw press (SP) treatment was performed with a machine typically used for small-scale (household) tomato juice production, while the centrifugation treatment was performed in three main steps: (1) transfer of co-digestate (about 200 ml) to centrifuge tube, (2) centrifugation for 30 s at maximum speed (3500g) plus 3 min for acceleration and 8 min for deceleration (Becman J2-MC Centrifuge; rotor JA-10), and (3) careful suction collection of the supernatant. All separation tests (Table 1) were performed at the laboratory of the Department of Agriculture, Forest and Food Sciences at the University of Turin, Italy. All samples were analyzed in triplicate for DM, pH, total and ammoniacal N, total P, K, Cu and Zn. Separation efficiency of the treatments was evaluated using the Simple Separation Index (*Et*).

Table 1. Treatment ID and separation description

Treatment ID	Input material		Separation methods		
	Raw slurry	Co-digestate	Biochar	Polymer dosage (ml/0.5L manure)	Mechanical separation
SP	x	x			SP group
P+SP	x	x		175 of PAM	
C+SP	x			120 of Chitosan	
B+SP	x		0.5 g		
P+B+SP	x		0.5 g	150 of PAM	
C+B+SP	x		0.5 g	120 of Chitosan	
CENT	x	x			CENT group
P+CENT	x	x		175 of PAM	
C+CENT	x	x		120 of Chitosan	
B+CENT	x		0.5 g		
P+B+CENT	x		0.5 g	150 of PAM	
C+B+CENT	x		0.5 g	120 of Chitosan	

Results and discussion

Results showed that, when separating raw pig slurry, the pre-treatments markedly improved SP effectiveness, but failed to improve CENT separation efficiency to similar levels (Figure 1).

Added biochar increased the mass efficiency by 2 to 3 % for both separation methods, but had little effect on the chemicals tested. When added to raw slurry, chitosan loosened flocs sensitive to SP pressure, but was slightly affected by CENT. Liquids produced after chitosan pre-treatment reduced metal content only when centrifuged. Efficiencies were increased for total K, Cu, and Zn (75, 36, and 51%, respectively), but decreased for total and ammoniacal N and P (9, 19, and 51%, respectively) when chitosan and centrifugation were used in combination to separate co-digestate. Pre-treatment with PAM increased N, P, Cu, and Zn content in solids and decreased them in liquids when raw slurry was screw pressed. When centrifuged, PAM had little effect on solids, but significantly decreased the content of P, Cu, and Zn in liquids. These results suggest that screw press will generally outperform centrifugation as a mechanical separation method for farm applications. Addition of PAM to co-digestate before the screw press improved the P removal efficiency by 15%, with no other nutrient or trace metal retention effect. Alternatively, PAM added before the co-digestate centrifugation decreased the separation efficiency for total N, NH_4-N, and P by 16, 18, and 12%, respectively, without affecting other elements (Figure 2

Conclusions

The addition of biochar, chitosan, and their combinations to raw slurry failed to significantly improve the separation efficiency of mechanical separators, and did not significantly increase the elemental content of produced solid fractions. Application of chitosan to co-digestate increased the efficiency of centrifugation with respect to K, Cu, and Zn. PAM+SP effectively removed P, Cu, and Zn from raw slurry to solids, and produced the liquid fraction with the lowest dry matter and elemental contents. The addition of PAM prior to both mechanical separations of co-digestate showed no significant *Et* changes for all parameters, except for P (*Et* increased from 0.30 to 0.45) in the SP group.

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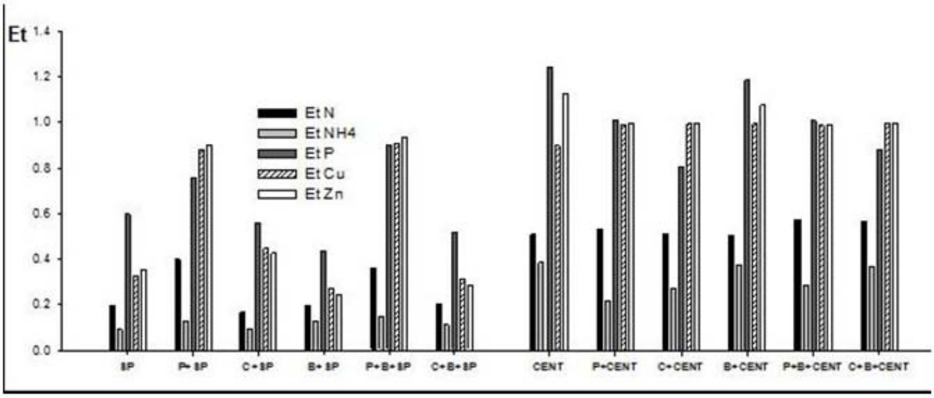

Figure 1. Simple separation index (Et) calculated for total nitrogen and NH_4-N, total P, Cu and total Zn for various pre-treatments on raw slurry denoted as P (PAM C-2260), C (chitosan), and two combinations (SP and CENT).

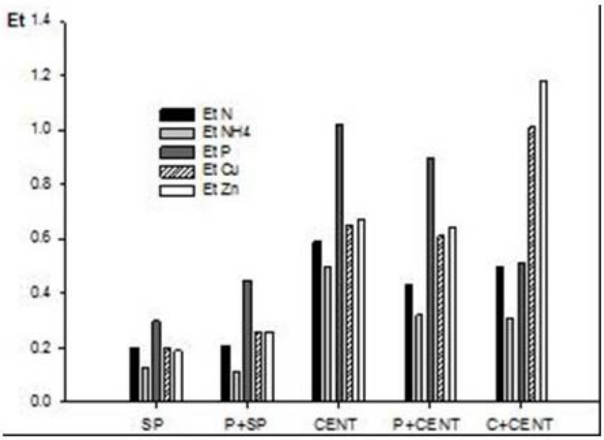

Figure 2. Simple separation index (Et) calculated for total nitrogen, NH_4-N, total P, Cu and total Zn for various pre-treatments on co-digestate as P (PAM C-2260), C (chitosan), and two combinations (SP and CENT).

NITROGEN LEACHING IN AUSTRIA – LONG-TERM LYSIMETER STUDIES FOR EVALUATION OF LEACHING FACTORS FOR TYPICAL MANAGEMENT PRACTICES ON GRASSLAND AND CROPLAND

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As part of the second commitment period of the Kyoto protocol 37 countries have agreed to legally binding reductions in their greenhouse gases emissions. The Intergovernmental Panel on Climate Change (IPCC) established guidelines which provide internationally agreed methodologies intended for use by countries to estimate greenhouse gas inventories to report to the UNFCCC (IPCC, 2006).

One of the listed greenhouse gases is nitrous oxide (N_2O), which has a global warming potential which is 296 times higher compared to carbon dioxide. Managed soils contribute to the emissions of N_2O directly and indirectly. The indirect N_2O emissions are caused by nitrogen leaching and runoff. The IPCC guidelines provide a calculation method to estimate N_2O emissions from leaching and runoff by using the $Frac_{Leach}$ factor, which is set to 0.3 as default value. This factor relates total nitrogen outputs to total nitrogen inputs. IPPC however encourages the use of country specific values in case a better basis for its estimation exists. As an example Ireland (0.1), Switzerland and Lichtenstein (0.2), Belgium and Netherlands (0.12) use lower values for $Frac_{Leach}$, based on scientific studies (Del Prado et al., 2006; Ryan et al., 2006; Neill, 1989; Prasuhn and Braun, 1994; Thomas et al., 2005). The aim of this work is to evaluate the validity of the factor $Frac_{Leach}$ in Austria using long-term lysimeter observations on both grassland and cropland under locally typical management.

Materials and Methods

In total we used data of 19 lysimeters, which were distributed all over the agricultural used area in Austria. Lysimeters were installed in both arable land and grassland. All lysimeters provided data on percolation and nitrogen contents of the percolating water. At all sites most of the required parameters of the IPCC equation (see Figure) were directly measured. However, belowground crop residues were only determined at specific sites. For those sites with missing information on belowground crop residues values were estimated using the default values of the IPCC guidelines. Land use was not changed and stable organic carbon content was assumed. This allows leaving F_{SOM} (mineralised N due to C-loss) unattended. The leaching factor $Frac_{Leach}$ finally was determined by relating nitrogen losses through leaching to the sum of all nitrogen inputs.

Results and Discussion

Results of the lysimeter exercise indicate a much lower $Frac_{Leach}$ as compared to the default value of 0.3. Especially on grassland in average only 2 % of available nitrogen are lost through leaching. $Frac_{Leach}$ for cropland is 0.19 in average, but also gives higher values at some sites, specifically when annual percolation is high or plant residues were not incorporated into the soil (Table1). Lowest values for cropland were gained in regions with low groundwater recharge.

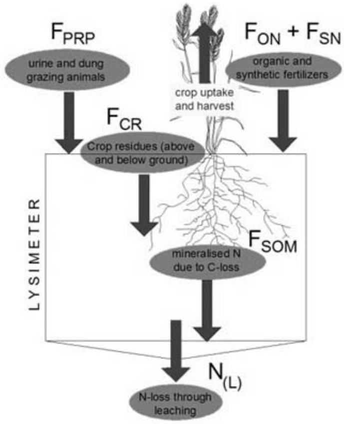

Figure 1: schematic display of FracLeach calculation leaching according to IPPC 2006 guidelines

Table1: Nitrogen inputs, leaching values and factors for selected Austrian lysimeter sites

	Petzenkirchen I	Petzenkirchen II	Petzenkirchen III	Petzenkirchen IV	Pettenbach L1	Pettenbach L2	Pettenbach L3	Pucking	Gumpenstein L1	Gumpenstein L2	Gumpenstein L3	Gumpenstein L4	Gumpenstein L5	Lobau L1	Lobau L2	Lobau L3	Hirschstetten S	Hirschstetten T	Wagna con I
land use	crop	crop	grass	grass	crop	grass	crop	crop	grass	grass	grass	grass	grass	crop	crop	crop	crop	crop	crop
annual rainfall (mm)	723	723	723	723	1030	1030	1030	753	1013	1013	1013	1013	1013	534	534	534	520	520	914
annual leaching (mm)	170	270	157	202	307	446	362	339	337	438	542	373	337	4	14	17	82	11	309
years of observation	5	5	6	4	17	6	14	7	3	3	3	3	3	14	14	14	3	1	5
N-Leaching (kg N ha-1)	187	252	33	17	475	121	250	476	3	4	21	4	12	30	148	104	114	14	487
N-Input organic fert. (FON) (kg N ha-1)	908	0	0	0	1520	955	116	597	120	238	214	311	409	0	0	572	0	0	885
N-Input syntethic fert. (FSN) (kg N ha-1)	366	506	0	0	652	597	1325	50	0	0	0	0	0	0	285		261	120	0
N-Pool Plant residues (FCR) (kg N ha-1)	357	398	2279	1753	1550	708	1171	390	201	327	319	377	331	1131	1013	1186	140	62	345
FracLeach	**0.11**	**0.28**	**0.01**	**0.01**	**0.13**	**0.05**	**0.10**	**0.46**	**0.01**	**0.01**	**0.04**	**0.01**	**0.02**	**0.03**	**0.11**	**0.06**	**0.28**	**0.08**	**0.40**

Although lysimeters can be used to estimate nitrogen leaching (Kroeze et al., 2003), they only represent point measurements. Furthermore nitrogen losses through runoff were not investigated. They might cause higher $Frac_{Leach}$ values if measurements and calculations are done at catchment scale.

Conclusions, Acknowledgments

The calculation of leaching factors with a lysimeter assessment delivers lower nitrogen leaching factors than suggested by the IPCC guidelines for both grassland and cropland. However, more detailed investigations have to be done at catchment scale to include also nitrogen losses through runoff.

We would like to thank our partners Joanneum Research Austria, Austrian Agency for Health and Food Safety, LFZ Raumberg Gumpenstein and BioScience Austria within the DAFNE100957-Project for providing data and expertise.

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INCORPORATION OF ORGANIC FERTILISERS IN SPRING TO WINTERWHEAT

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In both organic and conventional agriculture, high nitrogen fertiliser use efficiency is essential for the economics of cropping and for minimising the negative effects on the environment. The nitrogen mineralisation rate of various organic fertilisers has been examined in incubation studies and field trials (Delin & Engström, 2010). Low temperatures in spring delay nitrogen release from organic fertilisers, resulting in only small effects on yield, unless good contact between fertiliser and soil is achieved by incorporation. In Sweden, the efficiency of some organic fertilisers has been investigated in spring cereals (Gruvaeus, 2003) and organic winter oilseed rape (Stenberg et al., 2013), but not in winter wheat. This study therefore investigated the effect of organic fertilisers applied in spring to winter wheat and evaluated the effect of their incorporation into soil. It also compared application in late autumn with spring application.

Materials and Methods

Six winter wheat experiments were conducted at three locations in south Sweden (Halland, Västergötland and Västmanland) during two years, 2012-2013. The experiments had 12 treatments, comprising three commonly used organic fertilisers and control treatments with four different rates of mineral N fertiliser, randomised within four blocks. Organic and mineral fertilisers were applied 4 and 2 weeks, respectively, before crop growth stage 30 (GS30). Chicken manure was incorporated by harrowing after broadcasting, meat meal pellets were incorporated into the soil using a seed drill with fertiliser discs, while liquid bioresidues were injected by disc cutters to the recommended depth of 5-7 cm. By comparing grain yield in the organic amendments with mineral nitrogen fertiliser response, the nitrogen fertiliser replacement value (NFRV) was calculated.

Results and Discussion

In the three field trials performed in 2012, spring application of organic fertiliser gave yield increases averaging 1500 kg ha^{-1} (970-1900) and NFRV was on average 45% (23-61%). Incorporation of the organic fertilisers had no effect on yield in that year. On average for the three sites in 2013, the yield increase with spring fertiliser application was 1400 kg ha^{-1} (820-2200 kg ha^{-1}) and NFRV was 44% (21-70%). At two of the three sites in that year, incorporation of organic fertilisers affected yield (figure 1). Incorporation of bioresidues increased yield by 940 kg ha^{-1}and NFRV was doubled at one site with a heavy clay soil, whereas no effect was obtained for incorporation of the other fertilisers. Incorporation of meat meal pellets and chicken manure increased yield by 910 and 740 kg ha^{-1}, respectively, and NFRV was doubled at one site with a light-textured soil, but there was no effect of incorporation of bioresidues. Application of chicken manure in late autumn reduced yield by 44-60% and NFRV was halved compared with spring application. There were no differences in yield for meat meal pellets applied in late autumn or in spring.

The high and evenly distributed rainfall during the summer 2012 probably contributed to decreasing the impact of incorporation into soil. In addition, the fertilisers were not incorporated deeply enough at some sites. The opposing effects obtained for incorporation at the two sites in 2013 was probably due to differences in soil properties. On the light-textured soil, incorporation of the dry fertiliser was easier and therefore deeper than on the clay soil, which had a hard surface. This can explain why incorporation only had an effect on the light soil. On the clay soil, the hard surface blocked infiltration of the liquid bioresidues, which can explain why the effect was better after incorporation had broken the surface. On the light soil, the bioresidues probably did not need to be incorporated, since infiltration was good. The warm, dry weather in summer 2013 enhanced the effect of incorporation.

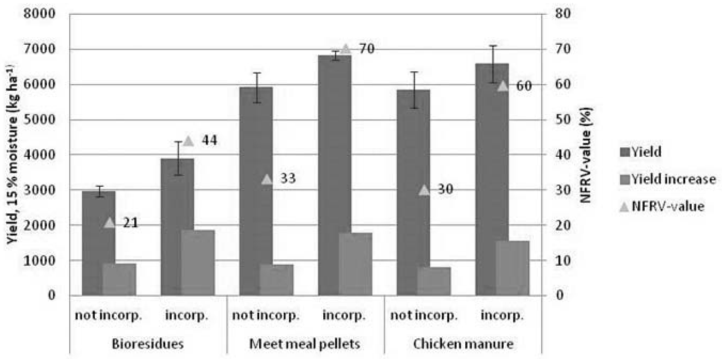

Figure 1. Effect of incorporation on winter wheat yield and nitrogen fertiliser replacement value (NFRV), for bioresidues (at Västergötland on clay soil), meet meal pellets and chicken manure (at Halland on sandy soil). Error bars indicate standard error.

Conclusions

Incorporation of dry fertilisers such as meat meal pellets and chicken manure can be recommended on light-textured soils and in dry weather conditions, such as in 2013. Incorporation of liquid fertilisers such as bioresidues is not needed on light-textured soils with good infiltration properties. On clay soils, where infiltration into soil may be blocked by surface hardening, incorporation of bioresidues can be recommended. Better techniques for incorporation of meat meal pellets and chicken manure on clay soils than used in this study are needed to improve the effect. Autumn application of chicken manure cannot be recommended.

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COUPLING MINERAL N AND SIMPLE MICROBIOLOGICAL INDICES IN SOIL INCUBATIONS TO INTERPRET N MINERALIZATION IN FIELD CONDITIONS

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Since several years, soil incubation has been used to get deeper information about the effect of fertilization on soil N kinetics, but the linkage between laboratory studies and processes at field scale still remains unclear. This work aims to assess the effect of compost fertilization on two soils adopting a lab to field approach, using mineral N and N-cycling bacteria dynamics from soil incubation to interpret results form a 3-year field trial.

Materials and Methods

A field experiment was carried out in Naples (NA) and Turin (TO) to compare the effects of compost and urea fertilization on soil fertility in two different Italian pedo-climates (Alluvione et al., 2013). Total maize dry matter (DM) production over 3 years showed a higher productivity at TO (21 *vs* 13 Mg ha^{-1} for TO e NA respectively). A 112-days incubation (T=28°C, soil moisture 40-60% of WHC) was carried out in 200 g soil pots to compare compost (COM), urea (UREA) fertilization with a not fertilized control (0N). Soils were collected during the 3rd year of experimentation at both sites, sieved at 5 mm and pre-incubated at 20°C for one week. Fertilizers were applied according to a field rate of 130 kg N ha^{-1} (as in the field), taking as reference a 0.20 m layer with bulk density of 1.2 and1.3 dm^3 kg^{-3}, for NA and TO soil respectively. NH_4-N and NO_3-N were measured according to the HACH method on samples collected at 0, 7, 14, 28, 42, 63, 84 and 112 days. Microbial enumerations of free-living N_2-fixing aerobic (N-fix) and nitrite-oxidizing (NO_2-ox) bacteria were performed at 28 and 112 days as reported by Pepe et al. (2013). Mineral N (min N) data were subjected to ANOVA considering each soil x fertilization x time combination as a single treatment, while a one-way three factors ANOVA (soil, fertilization and date) was carried out on microbiological data. Means were separated by LSD test when p-value was < 0.05.

Results and discussion

Min N values in NA soil were 50% to 80% of those in TO soil taking as reference 0N treatment, while fertilization did not produce any difference between soils with the exception of day 14 for COM (Fig.1). Fertilization effect as the difference of min N values recorded in fertilized soil and 0N is shown in Fig. 2. In NA soil, compost significantly increased min N (+36%) at day 42, and with a positive trend also at days 28, 63 and 112 (+26%, +21% and +8% respectively). The same treatment reduced min N content in TO soil on days 7 and 42 (-39 and -31% respectively) and also thereafter (although single data were not significantly different), thus showing N immobilization. In NA soil min N values recorded in UREA were significantly higher than in 0N

(+124% to +46%) during the whole incubation, while at TO, a significant increase was recorded only at day 63 (+44%). Min N values confirmed productive results of our field trial with reference to 0N soil (higher values for TO) and UREA (no difference between the soils), while COM showed an unexpected pattern. Although biomass production in COM was higher at TO than at NA (21 vs 8 Mg ha^{-1}), incubation showed the same min N values in both soils; furthermore a short-term N immobilization was found in TO soil even though field production was not different from that of 0N plots. It seems that the use of lab incubation mineral N as a key indicator could lead to a misleading interpretation of N availability of organic fertilizers in real field conditions. Dynamic of bacterial population allowed a better assessment of soil fertility and of its response to fertilization. N-fix bacteria (tab.1) were higher at NA and fertilization limited their growth in both soils. A significant increase at 112 days was found at NA, while TO soil showed an opposite trend. NO_2-ox bacteria were higher in NA soil at day 28 and values remained constant for 0N and COM, while increased in TO soil. Urea fertilization increased their number at day 28 only in TO soil, but a significant decline was found in both soils during the incubation. Abundance of N-fix bacteria was inversely correlated with N availability as indicated both by min N and field productivity, when fertilizers and soils were compared. It is possible that the increase of N-fix in NA soil is due to the reduction in the soil N supplying potential, while the decline in TO soil might be due to a log-lasting ability of this soil to supply min N. The initial peak of NO_2-ox bacteria in UREA was due to the input of easily available N and the decreasing pattern of this population could be related to a negative feedback of cumulated NO_3^--N. With the exception of UREA treatment, this population increased with increasing min N at TO, thus suggesting an increase of the nitrifying activity for this soil, in 0N and COM. On the opposite, constant values recorded at NA highlighted a lower attitude of this soil to sustain high min N flushes even in optimal conditions for mineralization. According to this results the growth pattern of N-cycling bacteria seems able to well discriminate between soils with different attitude to mineralize organic N, thus the combination of chemical and microbiological indicators is recommended to export results from soil incubation to an open field context.

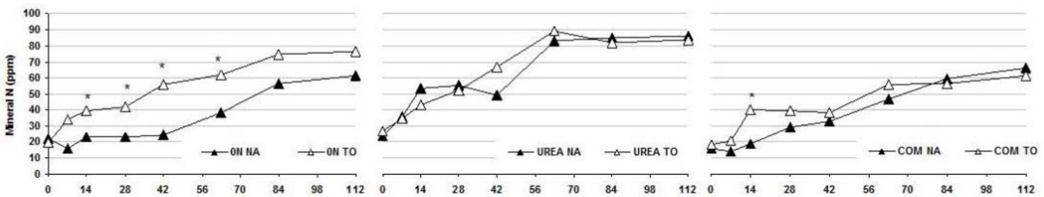

Figure 1 – Fertilization effect in NA and TO soil. *Indicates different values (p<0.05) within each sampling date

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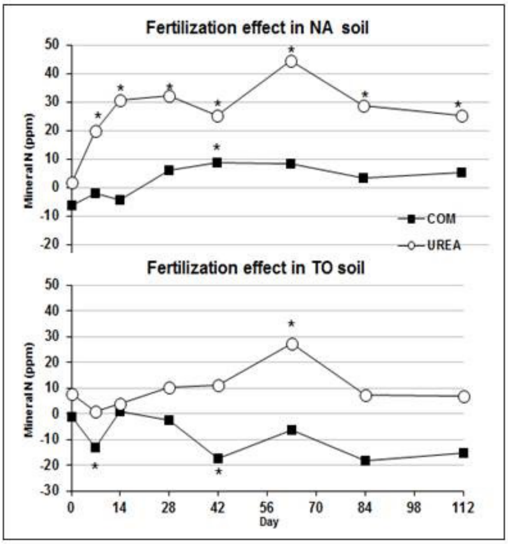

Figure 2 – Net fertilization effect (UREA-ON; COM-ON) in NA (a) and TO (b) soils. *Indicates values significantly different from 0 ($p<0.05$)

Table 1. Free-living (N_2-fixing aerobic and nitrite-oxidizing bacteria content in Naples (NA) and Turin (TO) soils under different fertilization at 28 and 112 days of incubation.

Soil	N_2-fixing bacteria ($CFU\ g^{-1}$)		NO_2-oxidizing bacteria ($CFU\ g^{-1}$)	
Treatments	28 d	112 d	28 d	112d
0N-NA	3.71^{b}	3.98^{a}	6.32^{ab}	6.48^{a}
COM-NA	2.89^{d}	3.29^{c}	6.18^{b}	6.48^{a}
UREA-NA	3.26^{c}	3.98^{a}	6.32^{ab}	5.17^{c}
0N-NA	2.26^{e}	1.98^{f}	3.26^{e}	5.15^{c}
COM-NA	2.30^{e}	1.71^{g}	3.26^{e}	5.15^{c}
UREA-NA	1.94^{fg}	1.32^{h}	5.15^{c}	4.22^{d}

Letters indicate significant differences among treatments ($p<0.05$)

TWENTY-FIVE YEARS OF LYSIMETER EXPERIMENTS IN THE CZECH REPUBLIC

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First field lysimeter experiments have been established by Central Institute for Supervising and Testing in Agriculture in 1984 on selected experimental stations in connection with the need of better understanding of movement of nutrients in the soil. The data were collected with the aim to evaluate the process of nutrient translocation in the soil with regard to plant nutrition, economics of fertilization and nowadays especially with regard to environmental compatibility. The construction of lysimeters was done according to representation of natural soil and water conditions. The collecting equipment was installed in undisturbed soil profile in the depths of 40, 60 and 80 cm (or in 40 and 60 cm only).

Materials and Methods

The basic long-term constant climatic and soil parameters for every standpoint are known (monthly and annual rain and temperature normal, soil type, parent soil substrate, bulk density of dry soil, maximum capillary water capacity). Continuously, every year observed parameters are meteorological data, growing crop, its yield, used fertilisers, percolation water retained in collecting equipment, used irrigation water, Nmin content in the soil – early in the spring, after crop harvest, before soil freezing, and basic agrochemical soil properties are determined– pH and available nutrient content (P, K, Mg, Ca) from spring sampling. In percolation, rain and irrigation water are determined pH value, nitrate and ammonia nitrogen, Cl, P, K, Mg, Ca, Na and SO4. Analysis of plant material (main and by-product) includes determination of contents of dry matter and essential nutrients (N, P, K, Ca, Mg). On the basis of results from lysimeter standpoints, we are able to observe inputs of nutrients from mineral and organic fertilizers, from rain and irrigation water to the soil (in complex). It is also possible to observe outputs of nutrients taken off by crops and nutrient losses determined in percolation water. From all this data, we are able to calculate nutrient balance. Based on Nmin determination in three terms, it is possible to observe dynamism of nitrate and ammonia nitrogen in soil and calculate the nitrogen losses during the winter. The primary aim of lysimeter measurements on the basis of analyses of percolation water is the monitoring of movement of nutrients, especially of nitrogen, in soil. Important contents of nutrients in percolation water are especially in the depth of 80 cm. These contents mostly represent losses for crops and at the same time a danger for quality of ground water.

Results

– From the point of soil characteristics the most important property is soil permeability. Light soils allow more rain water to penetrate to deeper horizons than heavy soils. Based on the intensity of infiltration (or water regime) we can separate our localities to percolative, periodically percolative and inpercolative regime. The intensity of infiltration is also influenced by total rainfall (and the distribution), soil type and crop.

– Most important nutrients from rainfall are nitrogen, calcium and sulphur.

– Nitrate form of nitrogen dominates in warmer sugar beet regions with chernozems and luvisols while slower nitrification leads to domination of ammonium nitrogen in colder regions with kambisols. Contents of both mineral forms of nitrogen decrease with depth of sampling.

– The highest environmental risk brings high content of nitrate nitrogen before winter, when there is very limited chance for its utilization by crops. In the spring we observed elevated amounts (circa 40 % more) of nitrate nitrogen in the middle and especially lower sampling depth, which indicates translocation of this nitrogen during winter.

– The most important factor of nutrient leaching is plant cover. The higher uptake by crops and shorter time without plants the lower risk of nutrient losses by leaching.

– Only smaller part of lost nutrients can be attributed to fertilizers used, majority comes from mineralization of soil organic matter.

AGRONOMIC EFFICIENCY, ENVIRONMENTAL BEHAVIOUR AND SOIL AMMONIA OXIDIZERS AS AFFECTED BY ORGANO-MINERAL FERTILIZATION

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The efficiency and sustainability of agriculture production is highly dependent on interactions within the soil–water–atmosphere–plant–animal ecosystem. While modern agricultural systems have provided considerable advantages to the growing world population, these activities have also produced some undesirable environmental consequences including impact of excessive losses of agricultural nitrogen (N) due to nitrification and subsequent denitrification, in high-imput farming systems. An alternative practice that could reduce N losses without necessarily reducing N inputs or crop yields is the use of organo-mineral fertilizers, which are mixtures of mineral components and organic fertilizer, with the organic components the minor amount. The aim of this study was to evaluate the agronomic and environmental efficiency of organo-mineral fertilization in pot experiment in a greenhouse using Lolium perenne, as well as the impact of different fertilization management on the abundance of soil ammonia-oxidizing bacteria and archaea.

Materials and Methods

This experiment was conducted in 30 cm diameter pots with soil in the greenhouse of the CRA-RPS research centre, Rome. Two kinds of fertilizer were applied, mineral as ammonium sulphate (M) and organo-mineral (OM) (21,21% N, 1% organic N from peat), at 160 kg/ha. A non-fertilized control treatment (C) was also included. Ammonia emissions, aboveground plant biomass, soil and water samples were collected during the 5-months experiment. Microbial biomass carbon Cmic was measured by the fumigation–extraction method. DNA was extracted from 0.25 g of each soil sample using a DNA PowerSoil® total DNA Isolation Kit (Mo Bio, Carlsbad, CA, USA), and quantified using QuantiTM dsDNA high sensitivity (HS) Assay Kit (Invitrogen NZ). Quantitative real-time PCR was performed on bacterial and archaeal amoA genes as described in Florio et al. (2014).

Results and Discussion

In our experiment, while no significant difference in ammonia emissions and between treatments could be detected, 15% decreased N losses from nitrates leached was observed in OM fertilization when compared to M fertilization. Furthermore, we found an increase in total soil N content as well as a slight increase in plant productivity at the end of the experiment in OM treatments when compared with M. The organic matter fraction of the fertilizer could have protected the N mineral fractions from leaching during the entire experiment; thus, this is in turn reflected on the limited environmental impact of organo-mineral fertilization. Furthermore, the increase in plant productivity and N concentration in plant suggest a better efficiency of organo-mineral fertilization with respect to mineral, as it's known that humic fractions provide physiological actions on plant growth due to different levels of intervention on the plant mechanisms, such as increasing nitrogen use efficiency, improving plant resistance to wilting by increasing osmotic pressure and affecting plant respiration by influencing the redox processes between dehydrogenase and oxidase Both microbiological and molecular approaches revealed that microbial community respond differentially to fertilizer treatments. In particular, Cmic increased both at T2 and T4 in OM when compared with both M and C, whereas at T3 no significant difference between treatments could be detected, suggesting that the organic matter fraction of the OM fertilizer can influence the dynamics of C immobilization in microbial biomass in the short-medium period. Molecular analysis revealed that AOB increase considerably following fertilization, as expected, at T2, corresponding to the highest nitrification activity in soil, and decrease after T3 until the end of the experiment with a similar trend both for M and OM, whereas they didn't shift in C during the entire incubation period. As opposed to AOB, AOA decreased during the experiment, starting from T1 in M and T2 in OM. However, a similar trend was found in C, suggesting that ammonia-oxidizing archaeal population was more sensitive to environmental changes than their bacterial counterpart, similarly with what found by Silva e Pereira et al. (2013) and Zinger et al. (2011). However, a more comprehensive understanding of the rules governing microbial communities will require additional field work to identify the key biotic and abiotic factors that drive the abundance and structure of these microbial groups.

Conclusions

Our results confirmed that the application of organo-mineral fertilizer to soil has the potential both to reduce the amount of fertiliser required, and to increase productivity; depending on soil conditions and plant species, these beneficial effects could be increasing N use efficiency, reducing N losses, fertilizer applications and consequently preventing environmental and health risk. Furthermore, the differential response of AOB and AOA to fertilizer treatments, probably due also to the different cellular biochemistry and metabolism existing between the domains Bacteria and Archaea, confirm that the actual role of AOA in N-cycling remains unclear.

Acknowledgements

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EFFECT OF NITRIFICATION INHIBITOR (DMPP) IN PEACH FRUIT QUALITY

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The new nitrogen fertilization practices to increase productivity and improve efficiency must guarantee fruit quality. It has been observed that excessive use of nitrogen in peach orchards delays ripening (Rufat, et al., 2011), and increases polyphenoloxidase (PPO) enzyme activity (Falguera et al., 2012, Pascual et al 2013). This enzyme largely determines postharvest fruit evolution. PPO is one of the main factors that determine postharvest fruit stability, which in turn may affect commercial issues of the fresh fruit market. The objective of this work was testing whether DMPP (3,4-dimethyl-pyrazole phosphate) combined with fertigation caused the same effects on fruit quality than conventional fertigation with nitrogen without DMPP.

Materials and Methods

The trial was conducted in 2013 in a commercial orchard of flat peach (*Prunus persica* (L.) Batsch. var. platycarpa (Decne.) LH Bailey cv. Planet Top) grafted on GF-677 rootstock and ridge planted at 2,5 m x 4,5 m in 2009 in the region of Segrià (Lleida, Spain). This area has a semi-arid Mediterranean climate. The soil is slightly saline (electrical conductivity 3,2 dS m^{-1}), with a silty loam texture, a pH of 7.8 and an organic matter content of 21 g kg^{-1}. A randomized complete block design with three replications was established. Four nitrogen fertilization level treatments, combined with DMPP were established (Table 1). Each elementary plot consisted of 6 trees and determinations were done on the 4 central trees. The harvest was on August 28th (commercial harvest). Mesocarp firmness (F) was determined with a manual penetrometer (Penefel, France). Total soluble solids (SS) concentration (◦Brix) was measured using a thermocompensated refractometer. Titratable acidity (AC) was obtained from 10 ml of juice of each sample, adding 10 ml of distilled water and titrated with NaOH 0,1 N. Maturity index (MI) was obtained as the ratio between SS and AC. Flesh color was measured with a Chroma Meter CR-400 tristimulus colorimeter in the CIELab color space. Parameters a*, b* and L* were determined. Fruit Nitrogen content was determined by Kjeldahl methodology. For polyphenol oxidase activity determination, 10 g of crushed peach flesh were mixed with 10 ml of McIlvaine buffer (pH 6,6) and 0,51 g (2,5%, w/v) of polyvinylpolypyrrolidone (PVPP) as phenolic scavenger. The mix was homogenized and centrifuged for 10 min at 5500 × g (RCF) and 5 °C. After centrifugation was complete, the pellet was discarded and the supernatant was used for PPO analysis, using 10 mM 4-methylcatechol as substrate, prepared in pH 6,6 McIlvaine buffer. The reaction was carried out in a 1 cm light path quartz cell, and the absorbance at 420 nm was recorded for 3 min with a spectrophotometer. One unit (U) of PPO was defined as the amount of enzyme that caused the increase of one absorbance unit (AU) at 420 nm in 1 min.

Results and Discussion

The lower activity of polyphenol oxidase (PPO) was found in the treatment T2 (50 kg N ha^{-1} with DMPP) (Table 2), followed by T1 (50 kg N ha^{-1} without DMPP), additionally lower PPO activity is observed in the other DMPP treatments, except for T8 (200 kg N ha^{-1} with DMPP) which registered the highest PPO activity. Treatments with no application of DMPP maintained the same tendency (Figure 1). The provided results indicated that PPO activity was influenced by high fertilization doses. Furthermore, it can be stated that the PPO activity is influenced by both the final amount of applied nitrogen (kg N ha^{-1} year^{-1}) and the concentration application of this nutrient in the irrigation water (ppm of N). No significant differences were found regarding the other studied parameters. Different doses of nitrogen, with or without DMPP had no effect on productivity or the standard parameters of fruit quality.

Table 1. Applied treatments characteristics.

Treaments	kg N ha^{-1}	DMPP	Days of aplication	Aplication dates		kg N ha^{-1} day^{-1}
				Starting	Ending	
T1	50	Without	62	8th June	8th August	0,81
T2	50	With	62	8th June	8th July	0,81
T3	100S	Without	31	8th June	8th July	3,23
T4	100S	With	31	8th June	8th August	3,23
T5	100	Without	62	8th June	8th August	1,61
T6	100	With	62	8th June	8th August	1,61
T7	200	Without	62	8th June	8th August	3,23
T8	200	With	62	8th June	8th August	3,23

Table 2: Effect of nitrification inhibitor (DMPP) in peach fruit quality, on N concentration fruit (% dry matter), yield (kg), mesocarp firmness (F), maturity index (MI), color parameters (CIELab L*, a*, b*) and polyphenol oxidase activity (PPO U mL^{-1}). Values followed by different letters indicate significant differences according to Tukey HSD test (P< 0,05).

Treaments	N kg ha^{-1} With and Without DMPP Days of aplication	N % Fruit	Yield kg	F N	M I	L	a*	b*	PPO (U mL^{-1}) Harvest
T1	50 (62 days)	0,97	58484	4,09	5,95	26	23	10	0,58bc
T2	50 DMPP (62 days)	1,07	60959	4,26	6,62	27	24	10	0,49c
T3	100S (31 days)	1,03	60212	4,31	6,3	23	20	9	0,79a
T4	100S DMPP (31 days)	1,03	60261	4,74	6,22	20	18	8	0,77a
T5	100 (62 days)	1,12	56328	4,11	6,46	28	28	11	0,76a
T6	100 DMMP (62 days)	1,06	52719	3,89	6,96	24	21	9	0,63abc
T7	200 (62 days)	1,13	59616	4,81	5,99	24	23	10	0,71ab
T8	200 DMPP (62 days)	1,23	60701	4,73	5,76	29	28	11	0,81a
Model (Prob>F)		ns	ns	ns	ns	ns	ns	ns	0,0004
Treatment (Prob>F)									0,0002

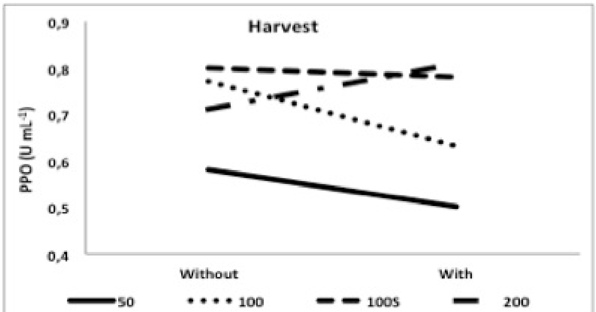

Figure 1. PPO activity on harvest date for DMPP and no-DMPP treatments.

Conclusions

Previous studies have shown the effects of nitrogen fertilization on the PPO activity in harvest and post-harvest stage, and effect of this on the fruit conservation. Preliminary results of this study show that the use of DMPP applied via fertigation could improve the potential conservation of the fruit peach by reducing the PPO activity at harvest.

Acknowledgements.

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ASSESSMENT OF POTENTIAL ENVIRONMENTAL BENEFITS OF LOW N-INPUT GIANT REED LIGNOCELLULOSIC FEEDSTOCK PRODUCTION THROUGH THE LCA APPROACH.

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The European Union directive (RED) encourages the production and use of biofuels in transport sector (at least 10% in 2020), by using lignocellulosic materials, wastes and residues (second or third generation biofuels), entailing more benefits in terms of land use, food security, GHG emission reductions and other environmental aspect. Agricultural crops for energy production has however been shown to cause relevant impacts, such as acidification and eutrophication, also higher than conventional fossil fuels [1]. Large emissions of reactive N and P are linked to fertilizer application [2], which therefore should be restrained to achieve a real sustainability of biofuel chains. The aim of this study is to compare the environmental constraints of lignocelluloses feedstock cultivation under high and low N-fertilization management.

Materials and Methods

A Life Cycle Assessment (LCA) was applied according to the ISO 14040-44/2006, to a giant reed (*Arundo donax* L.) cultivation grown from 2009 to 2012 in an experimental farm located in the Sele River Plain (Southern Italy). The LCA was carried out by means of SimaPro 7.3.3 software coupled with the ReCiPe (v., 2.0-2008) midpoint hierarchic impact assessment method. It was based on collected primary data for high and low N-fertilization management (Table 1), computation of Direct Field Emissions (DFE) [3] and the inclusion of non productive phases of cultivation, opportunely sharing the impacts of the latter for the whole crop lifetime (15-years). System boundary and functional unit were respectively set as 1 ha of cropped land and 1 kg of dry biomass. Impact categories analyzed were: Climate change (**CC**, Kg CO_2eq), Ozone depletion (**OD**, kg CFC-11eq); Terrestrial acidification (**TA**, kg SO_2eq), Freshwater eutrophication (**FE**, kg Peq), Marine eutrophication (kg **N**eq), Photochemical oxidant formation (**POF**, kg NMVOCeq), Particulate matter formation (**PMF**, kg PM_{10} eq), Fossil depletion (**FD** kg oileq).

Results and discussion

In the first year of cultivation (Fig.1A), with similar production (Table 1), **N50** gained a lower impact, markedly for the categories strictly linked to **DFE** of GHGs (**CC**), ammonia (**TA**, **ME** and **PMF**) and nitrate (**ME**) (Table 2). This result was more evident for **ME**, deeply affected in **N100** by a significant risk of nitrate leaching in the first year of cultivation (Fig.1 A, Table 2). From the second year, impacts decreased for both crops, due to the raising yields (Fig.1 B, C, D). Moreover, in consequence of diverging crop yields (Table 1), the lower upstream and downstream emissions entailed by **N50** were counteracted by the lower harvested biomass (on average about 22% lower in **N50** as compared to **N100**). In detail, in the second and fourth years (Fig.1 B, D), **N50** showed higher impacts associated to the harvest operations, due to fuel and exhaust emissions of bromotrifluoro-methane (**OD**) and NOx (**POF**), upstream phosphate emissions (**FE**) and oil consumption (**FD**). Anyway a net benefit could be always detected for the impact categories sensitive to **DFE** of reactive nitrogen (**CC**, **TA**, **ME** and **PMF**) and CO_2 (**CC**) (Table 2). In the third year, in absence of N fertilization at shoots re-sprouting, the final outcome was entirely ascribable to the percentage difference in crop yield among treatments (Fig.1 C). The impact of **N50** as **CC** might be further reduced including the soil carbon storage (**SCS)** in **DFE** computation. According to the experimental estimates of short-term **SCS** in top soil at the end of the fourth year (**N100**: 1.0 ton C ha^{-1}; **N50**: 4.6 ton C ha^{-1}), it might be assumed a total CO_2 sequestration of about 2.8 Ton ha^{-1} and 12.5 Ton ha^{-1}in **N100** and **N50**, respectively. Likely in **N50**, the low urea applied caused a reduction of soil organic matter mineralization, leading to a net sink of atmospheric CO_2.

Conclusions

A potential environmental benefit was highlighted for the low N-input giant reed cultivation, due to the reduced DFE entailed. Anyway the final outcome appeared to be very sensitive to the agronomic output, pointing out the relevance to better investigate crop yield patterns in the long-term, also to verify the feasibility of lignocellulosic feedstock large-scale cultivations to supply a regional biorefinery system.

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Table 1: Input and output flows. Gray background for practices with impacts properly shared for the whole lifetime.

Data input	**High fertilization (N100)**	**Low fertilization (N50)**
Seedbed preparation:		
Ripping and hoeing (combined rotary harrow) - diesel	12 L ha^{-1}	12 L ha^{-1}
Crop estabilishment:		
Seeding (rhizomes planting) - diesel	12 L ha^{-1}	12 L ha^{-1}
Field mantainance:		
Late N fertilization each year, except III year, (as urea 46%)	100 Kg ha^{-1} of N	50 Kg ha^{-1} of N
Combined fertilizer spreader - diesel	5 L ha^{-1}	5 L ha^{-1}
Harvest:		
Mowing by rotary mower - diesel	66 L ha^{-1}	66 L ha^{-1}
Combined harvester - diesel	26 L ha^{-1}	26 L ha^{-1}
Final eradication:		
Mower+Sprayer +Potato digger+Combined rotary harrow - diesel	149 L ha^{-1}	149 L ha^{-1}
Glyphosate (as Roundup)	2.72 kg ha^{-1}	2.72 kg ha^{-1}
Output		
Harvested biomass (d.w.)	10.3 t ha^{-1} (I) 22.6 t ha^{-1} (II) 18.7 t ha^{-1} (III) 23.8 t ha-1 (IV)	10.3 t ha^{-1} (I) 16.6 t ha^{-1} (II) 16.4 t ha^{-1} (III) 17.5 t ha-1 (IV)

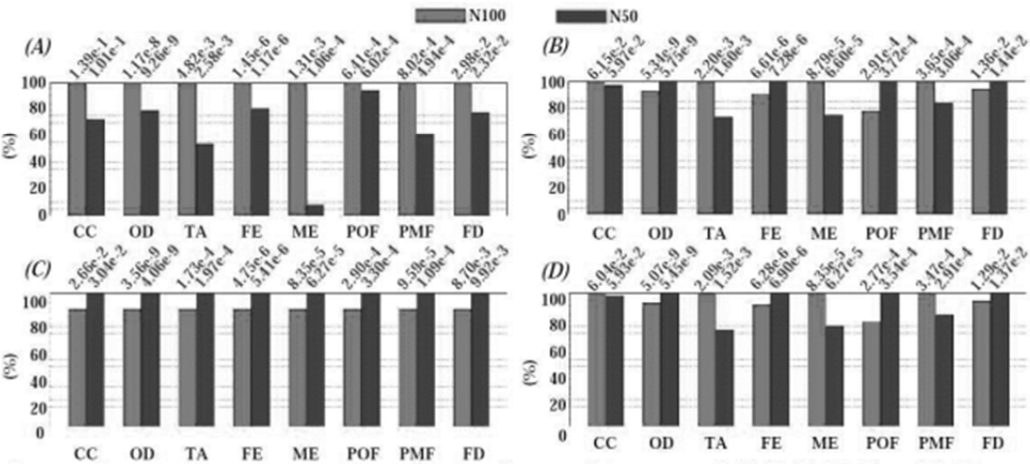

Figure 1: Percentage differences of total burdens between **N100** and **N50**, during the I *(A)*, II *(B)*, III *(C)* and IV *(D)* year of cultivation. For both crops, absolute value of total impacts in each category are reported at the top of columns.

Table 2: Contribution of DFE to total burdens in target impact categories for each year.

N50							N100						
CC		TA, ME, PMF and POF					CC		TA, ME, PMF and POF				
DFE	% CC	DFE	% TA	% ME	% PMF	% POF	DFE	% CC	DFE	% TA	% ME	% PMF	% POF
N_2O	26.12 (I)	NH_3	83.89 (I)	76.42 (I)	57.34 (I)		N_2O	29.66 (I)	NH_3	89.81 (I)	12.42 (I)	70.51 (I)	
	22.57 (II)		84.03 (II)	76.50 (II)	57.42 (II)			31.75 (II)		84.30 (II)	56,41 (II)	70.47 (II)	
	26.12(IV)		83.98 (IV)	76.42 (IV)	57.34 (IV)			26.34 (IV)		84.29 (IV)	47,44 (IV)	70.51 (IV)	
CO_2	7.56 (I)	NO_x	0.30 (I)	<0,01 (I)	0.80 (I)	3.06 (I)	CO_2	10.92 (I)	NO_x	0.60 (I)	0.67 (I)	0.82 (I)	4.84 (I)
	7.93 (II)		0.34 (II)	0.60 (II)	0.82 (II)	3.08 (II)		11.30 (II)		0.40 (II)	0.67(II)	0.83 (II)	3.57 (II)
	7.56 (IV)		0.30 (IV)	0.53(IV)	0.69 (IV)	2.58 (IV)		10.93 (IV)		0.33 (IV)	0.57 (IV)	0.72 (IV)	3.20 (IV)
		NO_3^-							NO_3^-		85.26 (I)		

NITROGEN TRANSFER BETWEEN COMPANION CROPS IN ANNUAL LEGUME-BASED INTERCROPS

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Legume-based intercrops have the potential to combine high economic performance and low environmental impact in systems by reducing the amount of fertiliser supplied (Pelzer et al. 2012). Indeed, growing a legume (Fabaceae) with a non-N fixing crop in the same field leads to a more efficient use of light and soil resources, and has a positive effect on plant productivity (Malézieux et al. 2009). In annual cereal-legume intercrops, niche separation contributes to increase yields and/or total N of the cereal (Corre-Hellou et al. 2006). In perennial legume-grass communities, it has also been shown that grasses can benefit from N provided by the neighboring legumes (Gylfadóttir et al. 2007). In annual crops, N transfer between the legume and the companion crop remains poorly documented. Our aim was to test whether N from the legume can benefit to the non N-fixing companion crop.

Material and Methods

Four experiments were undertaken in a greenhouse within 3 successive years. In three of them, durum wheat was intercropped with field pea in pots filled with soil. A different soil was used in each experiment. In one of these assays, we compared the amounts of nitrogen transferred between species either with intermingled roots, or root separated by a fine mesh (30 μm). In the fourth experiment, rapeseed was intercropped with faba bean in rectangular rhizotrons made of transparent Altuglass® protected from light by a black cover and inclined at 45° for allowing root survey. In all the experiments, using 15N-urea, we labelled either the legume or the non-fixing crop for assessing N transfer from the legume to the non-fixing crop and conversely. We applied a stem labelling method to field pea, faba bean and rapeseed (Mahieu et al. 2009, Jamont et al. 2013) and a leaf labelling method to durum wheat (n=5). Experimental designs also included unlabeled controls (n = 5). Calculations were performed as described by Gylfadóttir et al. (2007).

Results and Discussion

In all experiments, the amount of N transferred from the legume to the non-fixing plant was small (less than 1200 μg N.plant-1) and did not differ significantly from that transferred from the non-fixing crop to the legume. With the experiments undertaken on field pea-durum wheat, we have shown that the amount of N transferred from a plant to its companion crop increased significantly up to pea flowering, before stabilization until seed maturity (Fig. 1). N transfer between species was significantly higher when roots were intermingled than when they were separated by mesh (less than 50 μg N.plant-1; Fig. 2). In pea-durum wheat intercrops, the amount of N transferred from the legume to the companion plant was positively correlated to the legume root N ($p < 0.01$), and to the legume above ground N ($p < 0.01$; Fig 3). Such relationship was not found between N transfer and the %N of the plant parts. Although N rapeseed was higher in intercrops than in the pure controls, N transferred from the faba bean to the rapeseed was also similar to that transferred from the rapeseed to the faba bean (data not shown). Up to 32 days after sowing, root distribution in the rhizotrons was favourable to physical sharing of the soil N: 64 % of faba bean root length was located in the upper part, as 70 % was in the lower part for rapeseed. The biological nitrogen fixation of the intercropped faba bean reached around 87%, increasing niche separation between plants.

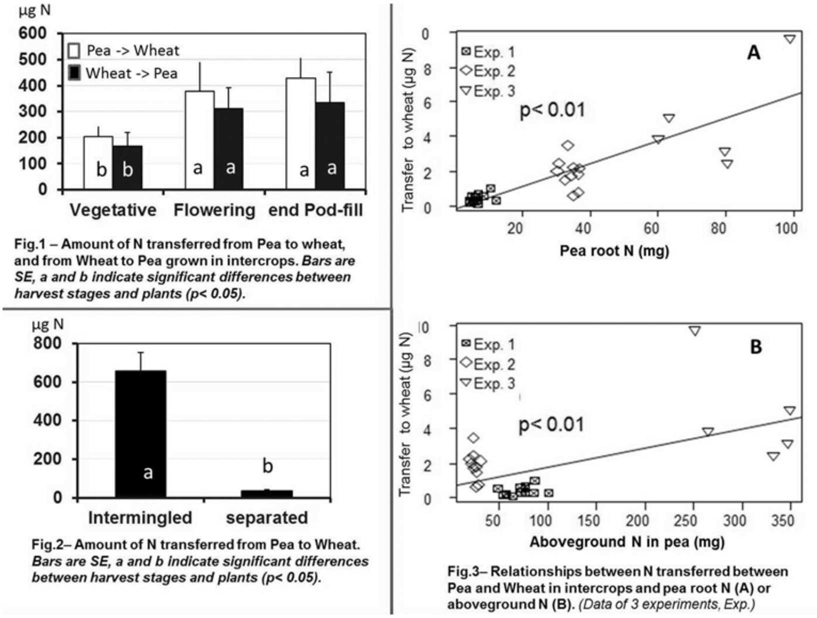

Fig.1 – Amount of N transferred from Pea to wheat, and from Wheat to Pea grown in intercrops. *Bars are SE, a and b indicate significant differences between harvest stages and plants (p< 0.05).*

Fig.2– Amount of N transferred from Pea to Wheat. *Bars are SE, a and b indicate significant differences between harvest stages and plants (p< 0.05).*

Fig.3– Relationships between N transferred between Pea and Wheat in intercrops and pea root N (A) or aboveground N (B). *(Data of 3 experiments, Exp.)*

Conclusions

During the legume crop cycle, net N transfer from the legume to the non-legume crop was negligible. Our results suggest that net N transfer can be increased by some particular legume traits: well developed hairy roots able to intermingle with that of the companion plant, and high plant N (roots and aboveground parts). However, such legume traits may compete for other abiotic factors such as light and/or water with the companion plant. In annual intercrops, niche complementarity seems to be a better lever to increase intercropping performances than N transfer. We can suppose that the fixed nitrogen benefits either in the companion species after maturity of the legume or in the succeeding crop, but not in the companion crop in the time of the legume cycle.

Aknowledgements

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GROUND AND AIRBORNE LEVEL OPTICAL SENSORS: IS IT POSSIBLE TO ESTIMATE MAIZE CROP N STATUS?

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Adjusting N fertilizer application to crop requirements is a key issue to improve fertilizer efficiency, reducing unnecessary input costs to farmers and N environmental impact. Among the multiple soil and crop tests developed, optical sensors that detect crop N nutritional status may have a large potential to adjust N fertilizer recommendation (Samborski et al. 2009). Optical readings are rapid to take and non-destructive, they can be efficiently processed and combined to obtain indexes or indicators of crop status. However, other physiological stress conditions may interfere with the readings and detection of the best crop nutritional status indicators is not always and easy task. Comparison of different equipments and technologies might help to identify strengths and weakness of the application of optical sensors for N fertilizer recommendation. The aim of this study was to evaluate the potential of various ground-level optical sensors and narrow-band indices obtained from airborne hyperspectral images as tools for maize N fertilizer recommendations. Specific objectives were i) to determine which indices could detect differences in maize plants treated with different N fertilizer rates, and ii) to evaluate its ability to identify N-responsive from non-responsive sites.

Materials and Methods

The study was conducted in the central Tajo river basin near Aranjuez (Spain) in 2012. The soil is a silty clay loam (Typic Calcixerept) and the climate Mediterranean semiarid. The experiment was design as a randomized complete block with six treatments per block and four replications. Plot size was 6 by 12 m. Treatments consisted in N fertilizer rates, and ranged from 0 to 200 kg N ha^{-1}, with 40 kg N ha^{-1} increases. The experiment was sown with maize (*Zea mays* L.) in early spring (20/04/12) with a plant population density of 80000 plants ha^{-1}. N fertilizer was hand broadcast to plots as ammonium nitrate in two applications: ½ when maize had 4 leaves (23/05/12), and ½ when had 8 leaves (26/06/12). Irrigation was uniformly applied by a sprinkle according to the crop evapotranspiration. Sensors Readings were conducted in two different dates: at stem elongation just before the second fertilizer application (21/06/12), and at flowering when treatment differences were expected to be more evident (23/07/12). Ground level measurements were taken with three optical sensors (SPAD® (Konica Minolta Inc., Japan), Dualex® and Multiplex® (Force-A, Orsay, France)). Airborne data acquisition was conducted by flying a hyperspectral (Micro Hyperspec VNIR model, Headwall Photonics, MA, USA) and an incoming irradiance (Ocean Optics HR2000 fiber-optic spectrometer, FL, USA) sensors 300 m over the experimental plots. In both sampling dates, fifteen measurements were taken with the hand held optical sensors in the uppermost fully developed leaf of 15 representative plants in the central plot rows.

Results and Discussion

Maize yield was highly correlated with N uptake and fertilizer application (Figure 1). During both dates, and at ground level, chlorophyll (Chl; SPAD, Chl Dualex, SFR) and N balance (NBI, NBI-G) indices tended to increase with N rate application and showed differences between lower and higher N application treatments, whereas flavonoids (Flav) and anthocyanins (Anth) tended to decrease. These results are in agreement with Cerovic et al. (2005) and Tremblay et al. (2007) who observed that N deficiency reduced Chl content and increased polyphenols. Good correlation between sensors indices was observed (~0.9) except for the Multiplex at steam elongation, due to the low signal intensity when leaves were too narrow. The structural index presenting a better relationship with LAI and ground level Chl measurements was NDVI (Table 1). However, other airborne indices calculated from narrow-band reflectance measurements (i.e. R750/710 or TCARI/OSAVI) and Chl solar-induced fluorescence (FSIF) were higher correlated with Chl than NDVI and presented a larger potential for future application to estimate crop N status (Fig 1). Table 1. Pearson correlation coefficients for the linear model between indexes (shadowed for those based on ground level sensors) and crop parameters at steam elongation (SE) and maize flowering (Fl). Indexes are frequently used in the remote sensing literature. A detailed description can be found in Quemada et al. (2014). Maize yield versus crop N uptake (A), N applied as fertilizer (B) and R750/R710 index (C).

Conclusion

The results suggest that despite numerous sources of variation, indices based on airborne measurements were as reliable as ground level equipment at assessing crop N status and predicting yield at stem elongation and flowering. Ground level indices more reliable to differentiate between maize plants treated with different N fertilizer rates were SPAD readings, Chl Dualex and SFR Multiplex. The airborne chlorophyll indices (i.e. R570/R710) and SIF were more accurate in detecting N stress in maize that structural indices (i.e. NDVI). More research is needed to account for other sources of variability that may interfere in the identification of the N nutritional status.

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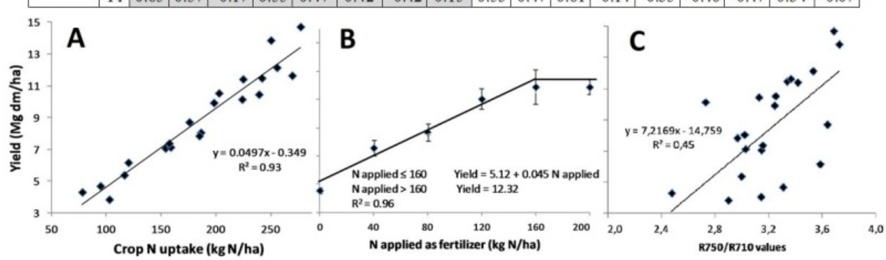
		SPAD	Chl	Flav	NBI	SFR	NBI-G	Anth	FERARI	NDVI	RDVI	R750 / R710	R700 / R760	TCARI	TCARI / OSAVI	PRI	BGI1	FSIF
Yield	SE	0.48	0.44	-0.10	0.22	-0.07	-0.10	0.04	0.12	0.46	0.47	0.55	0.33	0.14	-0.17	-0.48	0.23	0.59
	Fl	0.67	0.62	-0.06	0.54	0.55	0.38	-0.45	0.26	0.61	0.57	0.67	-0.04	-0.29	-0.42	-0.55	0.33	0.52
Biomass	SE	0.43	0.38	-0.18	0.26	-0.09	-0.04	-0.08	0.07	0.43	0.44	0.51	0.28	0.10	-0.20	-0.43	0.27	0.49
	Fl	0.65	0.56	-0.05	0.49	0.52	0.34	-0.42	0.26	0.60	0.56	0.63	-0.04	-0.27	-0.40	-0.53	0.30	0.49
Grain N	SE	0.49	0.42	-0.02	0.15	-0.04	-0.12	0.04	0.19	0.50	0.52	0.57	0.42	0.24	-0.05	-0.54	-0.30	0.57
	Fl	0.64	0.59	0.00	0.49	0.55	0.30	-0.43	0.31	0.64	0.61	0.66	0.03	-0.23	-0.37	-0.59	-0.18	0.57
N uptake	SE	0.50	0.47	-0.18	0.30	-0.09	-0.03	-0.01	-0.01	0.37	0.38	0.48	0.20	0.00	-0.29	-0.38	0.30	0.15
	Fl	0.65	0.57	-0.17	0.55	0.47	0.42	-0.42	0.15	0.53	0.47	0.61	-0.14	-0.35	-0.46	-0.47	0.34	-0.07



MODIFICATION OF THE NITROGEN MINERALIZATION CAPACITY OF SURFACE AND SUBSURFACE LAYERS OF AFFORESTED SOILS

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Since the beginning of the 1990s, the European Union has promoted the transformation of marginal agricultural land into forest land (EC, 1992), with the aim of enhancing the accumulation of carbon and nitrogen in both the plant biomass and the soil. However, questions have been raised about the suitability of this approach, which has not always been correctly applied: in many instances, prime agricultural land has been afforested. Moreover, the practice has led to notable changes in the European agricultural landscape, as the afforestation has often been carried out with exotic species. Nonetheless, very little is known about how the practice affects the soil organic matter. This lack of information is especially important in relation to nitrogen, as the transformation of agricultural land to forest land will greatly modify the N cycling, mainly due to cessation of nitrogenous fertilizer application. In the present study, we investigated the depth-related variation in the nitrogen mineralization capacity of the different soil layers (to a depth of 100 cm) in afforested plots and in reference cropped plotsn (representing pre-afforestation conditions).

Materials and methods

Three study plots (AF) were established in land previously dedicated to maize culture (for more than 100 years) and that was afforested 11 years ago. One of the plots was afforested with *Quercus rubra* L. (Baloira) and the other two with *Populus x euroamericana* (Dode) Guinier (Laraño and Pontevea). Control, cropped, plots (CT) were established in land still dedicated to maize culture, next to each of the afforested plots. Soil samples were taken to a depth of 100 cm in all plots (afforested and cropped) with the aid of a soil probe (Eijkelkamp®, model 04.15). The cylindrical samples thus obtained were divided into 10 cm segments, which were each analyzed separately. Soil samples were collected at three points in each each plot. The soil samples were sieved (< 4 mm) and stored at 4° C until analysis. Soil samples at field moisture were incubated in the dark for 10 days at 25 °C. The total inorganic nitrogen was extracted before and after incubation with 2 M KCl (1:50 w:v) and was determined following the method described by Bremner and Kenney (1965). Potential nitrogen mineralization was evaluated as the difference between the N contents at the beginning and at the end of the incubation period. The main characteristics of all soil samples were determined following the methods described by Guitián and Carballas (1976).

Results and discussion

In general, the total C and N increased in the upper soil layer (0-10 cm) after afforestation, except in the Laraño plot, in which the total C and N contents were almost the same as in the cropped plot (Tab 1). Moreover, although the total C and N contents were very similar in the upper 40 cm of the cropped soils (due to mixing of the soil during ploughing), clear differences between the upper (0-10 cm) and lower layers were observed in the afforested soils. The soils in the afforested plots contain less total inorganic nitrogen than the cropped soils, probably as a result of the cessation of nitrogenous fertilizer application. However, in all three afforested plots, the potential nitrogen mineralization capacity of the upper soil layer was higher than in the same layer in the corresponding cropped plots (Fig. 1). In the subsurface layers, the change in the potential nitrogen mineralization capacity was similar to that observed for the organic matter distribution: in the cropped plots, the potential nitrogen mineralization capacity was similar in all layers between 0 and 40 cm, whereas it was clearly stratified in the afforested plots, with a strong increase in this parameter between 0 and 10 cm.

Table 1 Mean values of the main properties and the total inorganic N content of soils in the control (cropped) plots (CT) and variation in percentage of these properties in the soils from the afforested plots (AF).

	Total C (%)						Total N (%)						pH KCl						Total inorganic N (mg kg^{-1})					
	Baloira		Laraño		Pontevea		Baloira		Laraño		Pontevea		Baloira		Laraño		Pontevea		Baloira		Laraño		Pontevea	
Depth (cm)	**CT**	**AF**	**CT**	**AF**	**CT**	**AF**	**CT**	**AF**	**CT**	**AF**	**CT**	**AF**	**CT**	**AF**	**CT**	**AF**	**CT**	**AF**	**CT**	**AF**	**CT**	**AF**	**CT**	**AF**
0-10	1.89	65	2.50	-1	1.85	101	0.17	72	0.19	-7	0.18	50	3.76	5	4.19	8	4.21	5	10.15	-10	9.10	-8	4.55	92
10-20	1.84	-24	2.46	-24	1.94	0	0.16	-4	0.19	-22	0.17	-13	3.84	3	4.32	-5	4.25	-6	9.10	-42	10.15	-24	7.35	-33
20-30	1.97	-39	2.31	-34	1.75	10	0.17	-29	0.16	-20	0.16	-15	3.81	5	4.26	-5	4.35	-7	9.10	-54	14.70	-48	6.65	-53
30-40	1.87	-57	1.92	-35	1.84	-5	0.16	-44	0.12	-20	0.17	-22	3.90	3	4.22	-2	4.32	-4	9.10	-31	9.10	-27	6.65	-53
40-50	1.01	-25	1.91	-41	1.01	-16	0.10	-38	0.14	-18	0.10	-70	4.05	-1	4.19	2	4.67	-5	8.75	-48	4.90	-43	9.80	-75
50-60	1.07	-43	1.96	-29	0.47	60	0.12	-45	0.14	-21	0.05	-11	4.23	-6	4.35	-3	4.85	-5	9.80	-54	2.80	25	3.85	-73
60-70	1.58	-66	1.84	-14	0.43	12	0.13	-62	0.13	-16	0.05	-27	4.22	-3	4.40	-3	4.87	-7	7.70	-50	2.80	38	2.10	-67
70-80	1.65	-69	1.44	4	0.35	60	0.11	-56	0.12	-21	0.03	-18	4.22	-2	4.43	-4	4.93	-10	3.85	0	1.75	180	1.75	0
80-90	1.23	-61	1.41	-4	0.34	38	0.10	-57	0.10	-20	0.03	-31	4.28	-5	4.50	-5	5.00	-11	3.15	-44	2.10	150	2.45	-71
90-100	0.97	-50			0.24	104	0.09	-46			0.03	-1	4.30	-7			4.93	-10	3.15	-11			3.15	-44

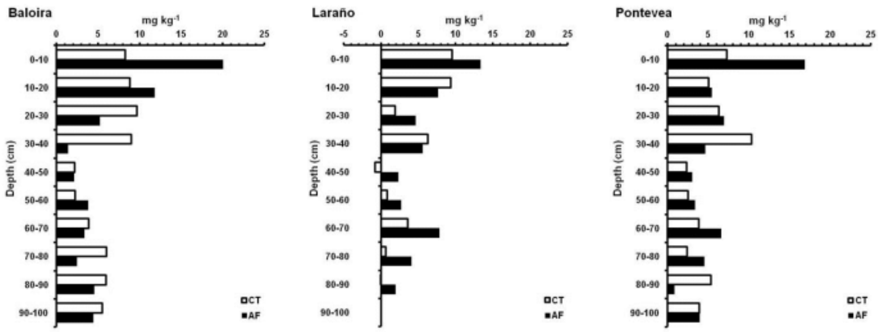

Fig.1 Potential nitrogen mineralization capacity in all the soil layers in each of the three pairs of cropped (CT)-afforested (AF) plots.

Conclusions

The differences between the afforested and cropped plots were due to changes in management practices, such as cessation of the nitrogenous fertilizer application, as well as to the change in vegetation resulting from the afforestation. In the afforested soils, the cessation of nitrogenous fertilization application was accompanied by an increase in the organic nitrogen mineralization capacity, to yield bioavailable nitrogen.

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ESTIMATION OF NUTRIENT VALUE OF SLURRY IN DAIRY FARMS IN GALICIA FROM THE DENSITY AND ELECTRICAL CONDUCTIVITY

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In Galicia, the primary milk production region of Spain, feeding dairy cows is based on concentrates and own-grown crops (maize and grass silage). The own slurry produced in the farms is used in the fertilization of the crops and generally the farmer doesn´t know the nutrient value of this slurry. Quick methods for estimating the composition of slurry from physical-chemical parameters such as electrical conductivity and density allow in situ estimation of nutrients by the technicians who advise these farms and therefore immediate adjustment of slurry doses, with subsequent environmental and economic benefits. Good correlations between electrical conductivity and total and ammoniacal nitrogen were found in cattle slurries (Provolo and Martínez Suller, 2006; Martínez Suller et al., 2010) although some authors only found good correlations with ammoniacal nitrogen (Mangado et al., 2006; Parera i Pous et al., 2010), reasonable correlations with potassium (Provolo and Martínez Suller, 2006; Parera i Pous et al., 2010) and low correlations with phosphorus (Provolo and Martínez Suller, 2006; Parera i Pous et al., 2010). Good correlations between dry matter and nitrogen total and phosphorus and low correlations with potassium were found (Martínez Suller et al., 2010). Dry matter is highly correlated with density that is a direct and quick measurement. So Mangado et al. (2006) found a good correlation between phosphorus and density. The aim of this study was to obtain equations linking direct readings of physicochemical parameters such as density and electrical conductivity with the fertilizer value of slurries to estimate reliably the nutrient value of slurry generated in Galician intensive dairy farms.

Materials and Methods

Thirty-eight samples of cattle slurry generated in different dairy farms were analyzed. Slurry dry matter (DM) was determined after 24 h of oven-drying samples at 105ºC. Analysis of N, P and K was determinated in fresh samples. Fresh samples were previously digested with sulphuric acid and hydrogen peroxide. Total N was analyzed by a colorimetric method using a continuous flow analyzer. Slurry K content was determined by atomic absorption spectroscopy. Total P was determined by a UV-vis spectrophotometer. Density was measured directly in the slurry, applying it to a measuring cylinder, stirring, introducins a hydrometer and taking measurement at 5 minutes when stabilized. Electrical conductivity was measured with a conductivity meter (Crison) equipped with a titanium electrode that was inserted directly into the slurry or into diluted slurry with distilled water in the ratio of 1:9 to prevent electrical and ionic interactions between the ions. Correlations between nutrient value of slurry: Kg of N, Kg of P2O5 and Kg of K2O per cubic meter with density (D) and electrical conductivity (EC) were calculated. After these correlations, simple and multiple regressions were done with these variables. The data were analyzed using the SPSS statistical package.

Results and Discussion

Analyzed slurries had an average chemical compositon of 7.67% of dry matter, 35.97 g•kg-1 of N, 6.52 g•kg-1 of P y 32.46 g•kg-1 of K. Correlations between nutrient value of slurry and electrical conductivity (Table 1) are reasonable for potassium and low for phosphorus, in agreement with other authors (Provolo and Martínez Suller, 2006; Parera i Pous et al., 2010). Correlations are somewhat low for nitrogen but higher that those found by Parera i Pous et al. (2010). Correlations were better for electrical conductivity measured in diluted slurry with distilled water in the ratio of 1:9 (ECdil) than for electrical conductivity measured directly (EC), so that the first was taken to obtain the regression equations. Correlations between nutrient value of slurry and density (Table 1) are good for phosphorus, in agreement with other authors (Mangado et al., 2006) and somewhat low for nitrogen and potassium. The best regression equations obtained are displayed in Table 2. Phosphorus value of slurry showed a good simple regression (R2 0.82, P< 0.001) in relation to the density. Simple regression equations obtained for nitrogen and potassium value with electrical conductivity were not so good. Multiple regression equations that considered as variables D and ECdil significantly improved prediction of nitrogen and potassium and lightly prediction of phosphorus.

Table 1. Pearson correlations between parameters.

	EC $mS{\cdot}cm^{-1}$	$EC_{dil}(1{:}9)$ $mS{\cdot}cm^{-1}$	D $(kg{\bullet}m^{-3})$	N $(kg{\bullet}m^{-3})$	P_2O_5 $(kg{\bullet}m^{-3})$
EC(1:9)	0.93***				
D	0.26	0.35*			
N	0.65***	0.75***	0.70***		
P_2O_5	0.42**	0.56**	0.90***	0.89***	
K_2O	0.69***	0.79***	0.53**	0.81***	0.63***

*** P<0.001; ** P<0.01; *** P<0.05

Table 2. Simple and multiple regression equations.

$Kg{\cdot}m^{-3}$	Equation	Coefficient of determination
	Simple regression equations	
N	$1.31EC_{dil}+0.63$	0.57***
P_2O_5	1.97D-1.03	0.82***
K_2O	$1.73EC_{dil}-0.11$	0.63***
	Multiple regresión equations	
N	$1.03EC_{dil}+4.12D-3.38$	0.81***
P_2O_5	$1.75D+0.25EC_{dil}-1.23$	0.88***
K_2O	$1.51EC_{dil}+6.00D-6.13$	0.73***

*** P<0.001

Conclusions

In this work was proved that in Galician Dairy farms is appropriated to use physicochemical parameters such as density and electrical conductivity to estimate the fertilizer value of slurries. Good regression equations between nitrogen, phosphorus and potassium value with the combined measure of both parameters were obtained. The implementation of these quick measurements in situ and the application of obtained equations will allow farmers immediate adjustment of the dose of slurry applied to silage crops.

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FIRST 20 YEARS OF DNDC: MODEL EVOLUTION AND GRAMP

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The DNDC (DeNitrification and DeComposition) model was first developed by Li et al. (1992) as a rain event-driven process-orientated simulation model for nitrous oxide, carbon dioxide and nitrogen gas emissions from the agricultural soils in the U.S. Over the last 20 years, the model has been modified and adapted by various research groups around the world to suit specific purposes and circumstances.

The Global Research Alliance Modelling Platform (GRAMP) is a UK-led initiative for the establishment of a purposeful and credible web-based platform initially aimed at users of the DNDC model. With the aim of improving the predictions of soil C and N cycling in the context of climate change the objectives of GRAMP are to: 1) to document the existing versions of the DNDC model; 2) to create a family tree of the individual DNDC versions; 3) to provide information on model use and development; and 4) to identify strengths, weaknesses and potential improvements for the model.

Materials and Methods

At present limited documentation exists on the differences between successive updates for each of the DNDC model versions. Consequently, users are often unaware of more appropriate versions of the model for their purposes. To rectify this GRAMP has created a database of DNDC model versions and constructed a “family tree”. Versions of DNDC were found through a combination of literature searches, web searches and input from the DNDC user-community.

The published literature was reviewed to identify how different modellers applied the DNDC model and the techniques used for model calibration. A range of statistical indicators were also used to compare the performance of different versions of DNDC. Further information on important changes to the model was also obtained as part of a survey distributed to *c.* 1500 model users around the globe. Information gathered included data on validation practices and datasets and records of changes made to individual versions of the model

Results and discussion

Through GRAMP, 17 different versions of the DNDC model have been identified and their history documented. Figure 1 is a schematic diagram of the model versions and how they relate to each other and the early versions of DNDC.

As part of GRAMP over 250 publications involving modelling with the 17 DNDC model versions were identified (Figure 2). In addition to the GRAMP team, the 98 survey respondents identified many strengths and weaknesses of the DNDC model versions which in addition to obstacles to process model uptake and recommendations for addressing the issues arising form the basis of a discussion document.

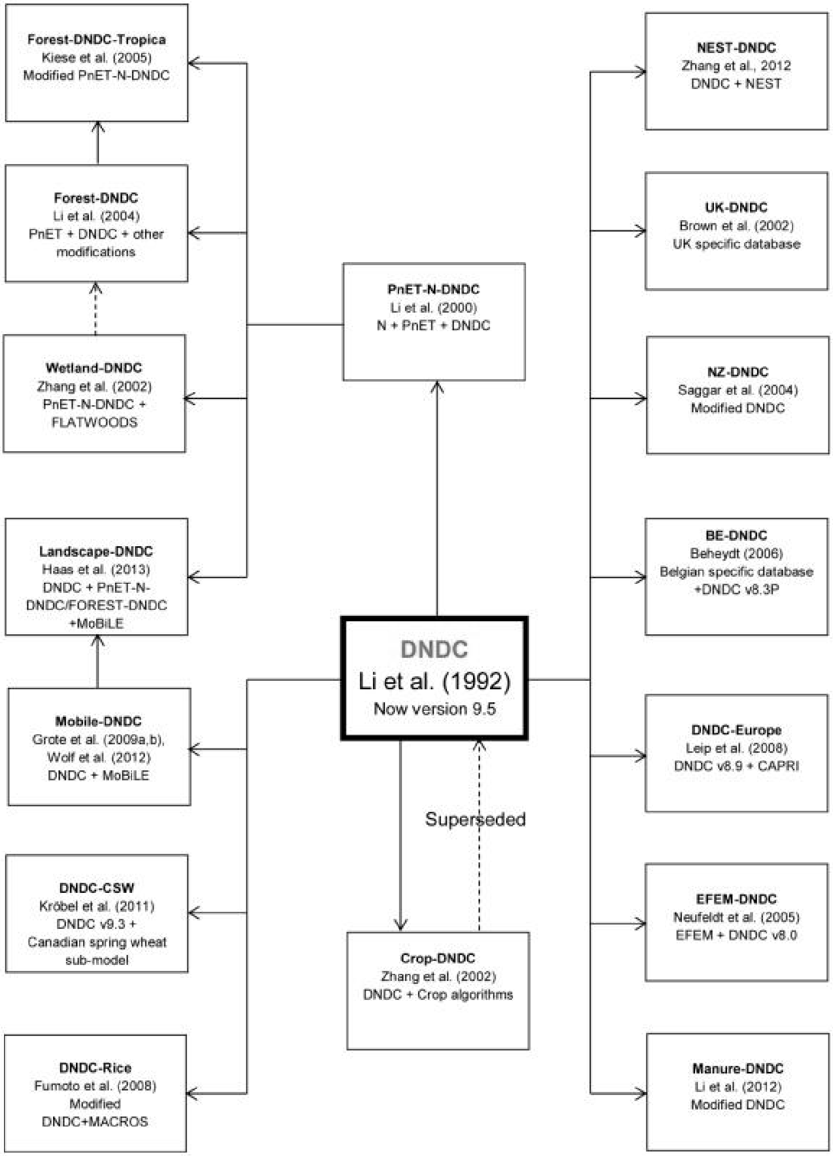
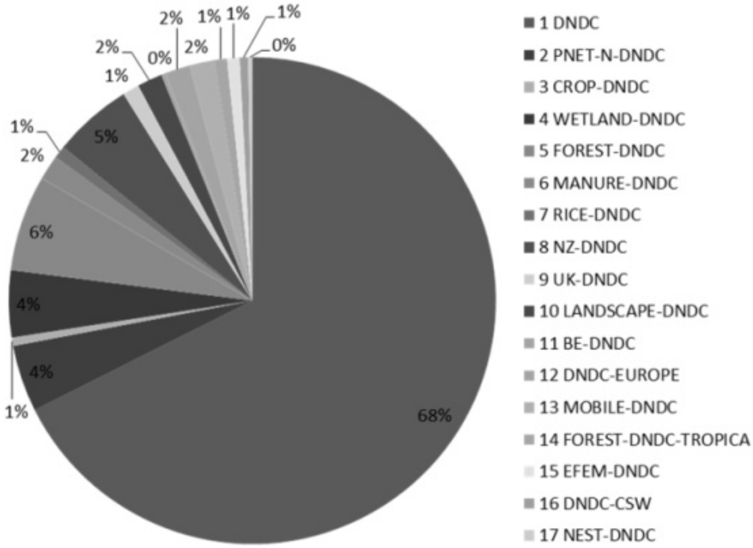


Figure 1. Schematic diagram of the DNDC extended family.



Conclusions

Throughout its 20 year history, the DNDC model has undergone many changes and its on-going value to the scientific community is reflected in the range of versions in current use, the number of current users, and an extensive published literature. However, in common with all biogeochemical process models, the DNDC model has both strengths and weaknesses. The GRAMP project has much to offer to the DNDC user community in terms of promoting the use of DNDC and addressing the deficiencies in the present arrangements for the model's stewardship.

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DEVELOPING A TOOL TO COMPARE NITROGEN USE EFFICIENCY OF DIVERSIFIED FARMING SYSTEMS

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The improvement of nitrogen (N) efficiency is a major way to enhance the productivity and reduce the environmental impacts of farming systems. Nitrogen Use Efficiency (NUE) is the most widely used indicator to assess N efficiency in farming systems, but presents several limitations. Importantly, it does not allow comparing farms with different crop and livestock productions, as N efficiency in livestock systems is usually lower than in cropping systems. Our goal was to propose an efficiency indicator that enabled comparisons between diversified farming systems.

Materials and methods

The first step in the calculation of this indicator was the estimation of N losses due to inputs production with life cycle inventory data. This allowed comparing farms relying on external inputs and more autonomous farms. The second step was the inclusion of soil N changes in the calculation of efficiency with a simple soil model. This helped comparing farms that did not manage their soil N stock in similar ways. These two modifications to NUE are discussed in more detail in Godinot et al. (submitted). The third step was the calculation of theoretically attainable N efficiency for all farm products. Estimates of attainable N efficiency for the main N fluxes in the farming system were found in the literature (Table 1). Based on these values, it was possible to calculate a theoretical attainable N efficiency for specialized and mixed farming systems given their outputs. The last step was the calculation of Relative Nitrogen Efficiency (RNE) as the ratio between actual efficiency and theoretically attainable efficiency. The closer to 1 this ratio was, the closer the farming system was from its efficiency potential, irrespective of the nature of its outputs. NUE (including input production and soil N change) and RNE were calculated for 38 mixed dairy and crop farming systems (MFS) in Brittany, and for various farm types from the literature.

Results and discussion

Interests of RNE

As shown in Figure 1, NUE was highly influenced by the nature of outputs: crop farms were always more efficient than animal farms. Within animal productions, pigs were more efficient than dairy cows, and dairy cows than beef cattle. RNE calculation allowed comparing different farming systems. In all types of production, some farms were over 80% of their efficiency potential, while some others were far from it. From these results, it was clear that some farms had a large margin of efficiency improvement. An important interest of RNE was to compare mixed dairy and crop farming systems producing variable quantities of animal and crop products. In our 38 MFS sample, there was a strong correlation between NUE and crop output. RNE gave supplementary information, as some farms presenting similar efficiency had very different RNE due to the different share of animal and crop products.

Limits of the method

To calculate theoretically attainable efficiency, we used the highest attainable efficiency value that we found for each N flux. Therefore, we did not take into account that forages usually present a higher N efficiency than grain crops; that animals have different efficiency depending on their feed; or that manure losses are depending on its nature. It therefore gives an idea of the highest attainable efficiency, not considering the particularities of each farm.

In order to calculate attainable efficiency, the maximal values of all fluxes were assumed to be independent. This assumption is questionable due to pollution swapping: low losses in one compartment can result in higher losses in the following compartments. This might lead to an overestimation of potentially attainable efficiency. However, as we use the same calculation method for all farms, this bias does not prevent the comparison of farming systems

Table 1: Attainable efficiency for the main nitrogen fluxes in farming systems.

Flux	Attainable efficiency	Source
Feed to cattle milk	0.36	Chase, 2004
Feed to beef cattle	0.21	Watson and Atkinson, 1999
Feed to pig	0.41	Cederberg and Flysjö, 2004
Crop to feed	0.86	Jarvis and Aarts, 2000
Harvestable to harvested crop	0.95	Rotz, 2004
Soil to harvestable crop	0.80	Powell et al., 2010
Excretion to soil	0.93	Jarvis and Aarts, 2000
N fertilizer production	0.99	IPPC Bureau, 2007

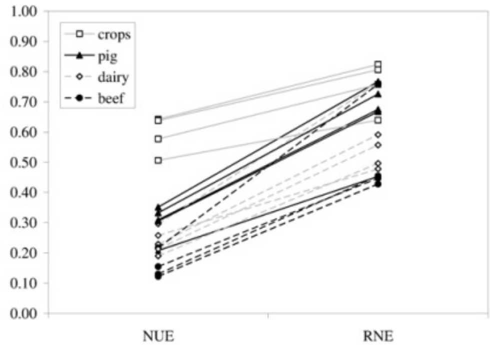

Figure 1: graphical comparison between Nitrogen Use Efficiency (NUE) and Relative Nitrogen Efficiency (RNE) for various farming systems

Conclusions

Relative N efficiency proved to be a useful tool to evaluate the gap between actual and potential N use efficiency of farming systems. It also allowed comparisons of different farming systems, which is especially interesting when comparing farming systems that are close but not comparable, such as specialized and mixed dairy farms. However, it is important not to forget that the choice of productions systems remains a major way to improve N efficiency at the global scale.

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MULTIVARIATE ADAPTIVE REGRESSION SPLINES TO SIMULATE SOIL NO3 CONTENT UNDER POTATO (SOLANUM TUBEROSUM L.) CROP

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Nitrate (NO3) leaching is a major issue in sandy soils intensively cropped to potato. Due to their shallow root systems, potatoes are generally cropped in light textured soils. Studies conducted in the North eastern part of the U.S. (Maine, New York) and Canada (Ontario) showed that only 45 to 60% of the N applied at planting at rates of 160 to 225 kg N ha–1 was recovered by tubers (Bouldin and Selleck 1977; Cameron et al., 1978). Modelling could help optimizing N fertilization strategies in time and space and reduce nitrate losses to the environment. Lack of input data is an important barrier for the application of classical process-based models to simulate soil nitrate content (NC). Alternative approaches using empirical relationships between NC and surrogate variables could be considered for operational purposes. This study evaluates a multivariate adaptive regression splines (MARS) model to simulate the NC daily seasonal dynamic in the 0-40 cm soil layer of potato fields. Input candidates were chosen for known relationships with NC: temperature, rainfall, leaf area index (LAI), day of year, and day after planting. Results were compared to field data collected between 2004 and 2012 in experimental plots under potato cropping systems on two farms in the Province of Québec, Canada. Training and validation of the models were operated on independent plots.

Materials and methods

The NC was measured from 26 rainfed agricultural plots in 2004, 2005, 2010, 2011, and 2012 located on two farms in eastern Canada. Composite soil samples were collected in each plot down to a 40-cm depth (2 layers of 20 cm each) in early spring before planting, every 2 weeks during the cropping season and after harvest. The meteorological data were aggregated at a daily time-step from in-situ micro-meteorological stations (Parent and Anctil, 2012) from planting to harvest. An averaged LAI value was obtained weekly from 12 plants in each plot during the growing season and biweekly during senescence, using a LICOR LAI-2000 instrument. MARS models were built using the ARESlab toolbox ver.1.5.1 (Jekabson, 2013) for Matlab. MARS builds a model in two stages: a forward and a backward pass (Friedman, 1991; García Nieto et al., 2012). The available 26 field plots were split into training and validation datasets using a self-organizing feature map (Kohonen, 1982) to ensure statistical homogeneity. 65% of them (17) were selected to train the model while the remaining 9 were used for validation.

Results and discussion

The most performing model was a 5-input MARS model built with cumulative LAI, cumulative rainfall, cumulative temperature, the day of year and the day after planting. The mean absolute error after final optimisation was 49.3 mg NO3 kg-1 on validation (i.e. 26.9% of the mean NC value of validation samples), which is satisfactory considering the intrinsically complex nature of NC (Fig.1). The variable explaining largest variance was the ∫LAI, suggesting the importance of local heterogeneity of NC. It also suggests that, as a growth indicator, LAI integrates many factors responsible for local heterogeneity of NC such as cultivar induced variability. As compare to a multiple linear regression, the proposed approach allowed to explain 54% more variance. The inclusion of the LAI as input variable adds a spatial dimension to the model. Map of simulated NC, coupled with N fertilization grids based on NC, would allow the mapping of N recommendation rate. This mapping is required for an effective variable rate N application.

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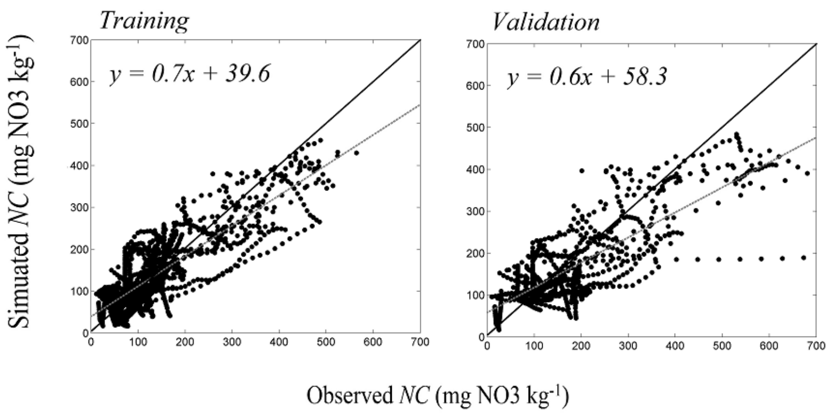

GOOD MANAGEMENT PRACTICES EFFECT ON N UPTAKE AND SURPLUS: AN OVERVIEW OF EUROPEAN LONG TERM TRIALS

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Agricultural practices can be considered as good if they positively affect the efficiency of resource use, soil fertility, C sequestration, or farm economic profitability (Costantin et al, 2010). Not all effects go in the same direction, and some drawbacks can be acceptable (e.g. a slight decrease in yield) if other advantages are met (e.g. a reduction in costs, or an increase in SOM). Farmers need a tool which helps them to quantify expected changes on target indicators, tailored on specific farm characteristics and pedo-climatic conditions, so that they can choose on the basis of their own specific objectives (Easterling, 2003). An EU-funded project, Catch-C (www.catch.eu) is analyzing the effects of different soil management options on productivity, climate change mitigation and soil quality using long-term field experiments (LTEs) across Europe, partly run by the project partners, and partly collected from international, national and technical literature. Data from over 350 LTE studies were collected and analyzed. This work presents preliminary results on the effects of a series of good management practices on crop yield, N uptake and surplus.

Materials and Methods

Multi-years averages of crop yields, N uptake and field N surplus (supply-removal) from more than 100 LTEs across Europe were analyzed. Agricultural practices considered were the use of crop rotations, harvested catch crops, crop used as green manures, no- and minimum tillage, organic fertilization with farmyard manure (FYM), bovine slurry or compost, and crop residues management. Yield obtained using each practice were divided by those obtained when the practice was not adopted, in the same conditions. The indicator obtained, a relative ratio (RR), is greater than 1 when the practice increased yield. N uptake was also analyzed using RR, while N surplus was examined using the difference between the practice and reference treatment (DIFF). A negative value indicates a reduction in surplus.

A multiple linear model using climate, soil type and duration of practice (4 levels each), and crop (12 types) as nominal factors was performed to evaluate which conditions mostly affected the performance of a practice.

Results and Discussion

The table reports main results obtained on yield. Data on N uptake and N surplus are here discussed but not shown.

In more than 80% of the cases, a crop grown in a rotation outyielded the crop grown in a monoculture, and the average increase in yield was 5%. Best performances were obtained in Western Europe climate, sand or loam soils, wheat or grain maize, and in long-lasting experiments (10-20 years). N uptake was also increased and N surplus was reduced. In 60% of the cases, the use of a harvested catch crop (both leguminous and non-leguminous) resulted in a yield increase of the main crop and best results were obtained in Eastern Europe, soils other than silt, barley, maize or minor cereals, and in long-lasting experiments. N uptake was also increased in 80% of cases, and consequently N surplus was reduced. Little or no effect of green manure on yield and N uptake was observed, in all pedo-climatic conditions explored. This means that the performance of green manuring could not be predicted on the basis of the considered factors.

A reduction in yield and N uptake is to be expected when no tillage is applied, but on average this reduction was limited to 4%. Silt soils performed best. Similar but less encouraging results on yield and N indicators were obtained using minimum tillage (defined as non-inversion tillage at a shallower depth than ploughing). Furthermore, the performance of this technique on yield was not influenced by the factors here considered, whereas N uptake was increased and N surplus was reduced in Western Europe. When organic and mineral fertilizers were applied at the same N rate, similar crop yield and N uptake were observed. However, the performance of organic fertilizers depended on the soil type (best results in coarse-textured soils), climate (the colder, the better) and duration of practice (more than 5-10 years). Incorporation of crop residues caused a reduction in yield (probably because of N immobilization) especially in badly-structured soils, in all crops. Quite surprisingly, burning cereals straw positively affected yield and N uptake, while reducing N surplus, although observations were mostly located in Western Europe and sandy soils.

Table. Statistics of RR of yield and factors that affected RR

Good practice	n.	mean	min	max	% of cases >1	factors
crop rotation	27	1.05	0.81	1.34	81.5	crop, climate, soil, duration
catch crops	41	1.05	0.75	1.58	63.3	crop, climate, soil, duration
green manure	8	1.00	0.64	1.30	50.0	-
no tillage	36	0.96	0.68	1.31	40.0	soil
minimum tillage	97	0.97	0.54	1.52	35.1	-
FYM	60	0.94	0.43	1.45	48.3	climate, soil
slurry	37	0.98	0.52	1.58	48.6	soil
compost	21	0.95	0.69	1.67	42.9	climate, crop, duration
incorporating crop residues	35	0.93	0.40	1.16	48.6	soil, duration
burning crop residues	9	1.03	1.01	1.06	100.0	-

Conclusions

When assessing farm-compatibility of good management practices several aspects have to be considered. Promoting productivity, climate change mitigation, and soil quality at the same time can sometimes be difficult to achieve.

LTEs provide plenty of information that rarely is aggregated, compared and well exploited (Olesen et al, 2006; Merbach and Deubel, 2008). Nitrogen management interacts with most other farm practices. In order to reach a higher N use efficiency, a simultaneous and complete analysis of many interactions must be considered. Only when this is done it is possible to identify bottlenecks and potentially overcome the social and economic barriers that slow the adoption of good practices for an efficient N use.

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CROP ROTATION AND RESIDUES INFLUENCE N2O EMISSION AND N EFFICIENCY RATHER THAN TILLAGE UNDER A RAINFED MEDITERRANEAN AGRO-ECOSYSTEM

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Conservation tillage and crop rotation have spread during the last decades because promotes several positive effects (increase of soil organic content, reduction of soil erosion, and enhancement of carbon sequestration) (Six et al., 2004). However, these benefits could be partly counterbalanced by negative effects on the release of nitrous oxide (N2O) (Linn and Doran, 1984). There is a lack of data on long-term tillage system study, particularly in Mediterranean agro-ecosystems. The aim of this study was to evaluate the effects of long-term (>17 year) tillage systems (no tillage (NT), minimum tillage (MT) and conventional tillage (CT)); and crop rotation (wheat (W)-vetch (V)-barley (B)) versus wheat monoculture (M) on N2O emissions. Additionally, Yield-scaled N2O emissions (YSNE) and N uptake efficiency (NUpE) were assessed for each treatment.

Materials and Methods

The experiment was located in “Canaleja” field station (Madrid, Spain) on a sandy clay loam soil (Calcic Haploxeralf) under rainfed conditions. The experimental design was a three-replicated split-plot. Tillage was the main factor (plot) in a randomized complete block design and subplots (as secondary factor) were W, V, B and M, in a complete randomized design. Crops were sown at the beginning of November and harvested on 18th June. All subplots were fertilized with 70 kg N ha-1year-1, except vetch ones, split in two applications (at seeding and spring). Nitrous oxide emissions were sampled periodically by Static Closed Chamber method (Ábalos et al., 2012) and quantified by Gas Chromatography. Cereal N content was quantified by Elemental Analysis.

Results and Discussion

Nitrous oxide cumulative fluxes were similar between tillage treatments. Conversely, significant differences ($P < 0.05$) were found among crops, i.e. M showed higher N2O fluxes than cereals in rotation (Table 1). These results highlight the importance of the type of residue from the previous crop (wheat for M, vetch for B and fallow for W), that seems to be responsible for the differences observed among crops fertilized with the same N rates (Malhi and Lemke, 2007). Fluxes of N2O for legume non-fertilized crop were not significantly different to low N input fertilized cereals (Jensen et al., 2012). Yield-scaled N2O emissions were greater in M than in W (Table 1), regardless tillage system. However, B and V showed similar YSNE values for all tillage treatments.

According to NUpE, there were not significant differences for tillage systems and crops. Nevertheless, cereals in rotation showed higher values than M in all tillage treatments, whereas lower values in conservation tillage (MT and NT) were obtained in comparison with CT (Table 1). Mean Emission Factor (kg N2O ha-1 kg N min applied-1) in all treatments was around 0.1%, lower than IPCC default value (1%) because of climate, soil conditions and land management (rainfed and low N input) (Aguilera et al, 2013). Conclusion Taking into account N2O emissions, yields and N efficiency, rotation is a good alternative to improve sustainability of crops. Nevertheless, tillage treatments showed high variability, and is necessary to consider the best combination of both factors.

Table 1 Effect of tillage treatments and crop on N2O cumulative emission, NUpE and YSNE.

Effect	N_2O cumulative emission (mg N-N_2O m^{-2})	NUpE (kg N_{plant} kg Nmin $_{applied}{}^{-1}$ x 100)	YSNE (mg N-N_2O kg $grain^{-1}$)
Tillage x crop	ns	ns	ns
Tillage	ns	ns	ns
CT	18.4	125	51.9
MT	18.5	102	61.2
NT	17.4	102	71.1
S.E.	1.8	10	8.5
Crop	**	ns	**
M	23.1 a	92	81.0 a
W	15.4 b	116	41.8 b
B	14.8 b	120	–
V	19.0 ab	–	–
S.E.	1.6	11	7.0

Within a column, means followed by the same letter are not significantly different according to Fisher's LSD at a 0.05 probability level. *P< 0.05; ** P< 0.01; ns=not significant

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NITROGEN AND OZONE INTERACTIVE EFFECTS ON THE NUTRITIVE QUALITY OF ANNUAL PASTURES

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Pastures represent one of the most important ecosystems covering a major fraction of European landscapes. They are important for agricultural production and biodiversity conservation. However, their response to increasing tropospheric ozone (O_3) and increasing nitrogen (N) availability, two of the main drivers of global change, is still uncertain. Annual species predominate in the dehesa traditional agro-forestry systems. The dehesas currently cover 3.5-4 million hectares in the Iberian Peninsula sustaining livestock farming, agriculture and timber production. Previous studies with individual species have shown that N fertilization counterbalances O_3 effects on plant senescence and on flower biomass production when plants are exposed to moderate O_3 concentrations (Sanz et al., 2007, 2011). The response of the whole community is expected to be more than the sum of responses of individual plants, thus experiments with annual communities are needed to understand changes that could be occurring at field level. An experiment was carried out to study the interactive effects of O_3 and N fertilization on a simplified annual community composed of six representative species. The experiment was performed in an Open Top Chamber (OTC) facility located in the Spanish central plateau (Santa Olalla; 450 m.a.s.l.; 40°3'N, 4°26'W). Plants were exposed to four O_3 treatments: charcoal-filtered air (CFA), non-filtered air (NFA), non-filtered air supplemented with 20 ppb O_3 (NFA+) and non-filtered air supplemented with 40 ppb O_3 (NFA++). Additionally, three nitrogen fertilization treatments were established aiming to reach N integrated doses of "background", +20 or +40 Kg N ha^{-1}. Ambient air chamberless plots (AA) were considered to evaluate the chamber effect. A mixture of six species of three representative families, were sowed. The study was carried out during two growing seasons in 2011 and 2012. Ozone significantly induced visible injury and reduced pasture total green biomass by 14% and 25% in NFA+ and NFA++ treatments respectively compared with CFA (Figure 1). The total senescent biomass increased 40% in NFA+ and NFA++. Nitrogen fertilization partially was counterbalance O_3-induced effects on green biomass only under moderate O_3 concentrations but not with high O_3 levels. On the other hand, O_3 exposure reduced the fertilization effects of N additions. Ozone also affected canopy gas exchange decreasing gross primary production through reducing net ecosystem CO_2 exchange and increasing soil respiration. Besides, soil N_2O emissions increased with O_3 exposure while N fertilization significantly enhanced NO emissions. Interestingly, increasing N availability was not enough to significantly affect the yield or gas exchange rates of the annual pasture. The background soil N content of the soil (30 kg Nmin ha^{-1}) seemed to be enough to cover the N nutritional demand of the pasture. In fact, natural annual pastures usually grow in low fertile soils (Vázquez de-Aldana, 2008). Other nutrients, such as phosphorus, could be limiting plant development in this type of ecosystems. Despite the low responsiveness to N, a significant interactive response between N and O3 was detected in green and total aboveground biomass production. Also increasing N availability affected quality parameters such as C/N ratio, crude protein and lignin, neutral or acid detergent fiber

(NDF or ADF), real food values (RFV) of some annual pasture species individually. NDF is inversely related to voluntary forage intake, and ADF is inversely related to forage digestibility of ruminants traditionally raised in this ecosystem. O_3 exposure also altered some quality characteristics of these annual community (Table 1). Therefore, both O_3 and increasing N availability seem to be affecting pasture growth and quality affecting livestock production on these extensive breeding areas.

Table 1. Nutritive quality parameters (means±se) depending on O_3 and N treatments in a simplified annual community in 2012. N0=N background, N20=N background+20 Kg N ha^{-1} and N40=N background+40 Kg N ha^{-1}. Statistical results of differences among treatments using the Univariate Tests of Significance ANOVA (p) considering a split-plot design and the Tukey test (HSD); comparisons between each treatment with the control were analysed with the Dunnett test (D). Different letters indicate significant differences among O_3 treatments obtained with the HSD test.

	Treatment	C/N			NDF			ADF			LIGNIN			RFV		
		p	HSD	D	p	HSD	D	p	HSD	D	p	HSD	D	p	HSD	D
	Nitrogen	ns	<0,1		ns			ns			ns			ns		
	Ozone	<0,1	<0,05	<0,1	ns			ns	<0,05	<0,005	ns			ns	<0,05	<0,05
	Nitrogen x Ozone	ns			ns			ns			ns			ns		
O_3	FA	16,64±0,48^{ab}			48,19±1,27			46,81±2,01^{a}			4,69±0,80			101,44±2,72^{ab}		
	NFA	16,38±0,69^{ab}			49,13±1,59			46,98±2,58^{a}			5,01±1,11			100,19±5,47^{a}		
	NFA+	16,09±0,43^{a}			50,30±0,60			41,28±2,70^{b}			3,09±0,77			105,27±4,24^{ab}		
	NFA++	18,89±0,88^{b}			48,03±0,94			39,54±2,77^{b}			3,17±0,86			112,99±4,94^{b}		
N	N0	18,14±0,61			49,41±0,80			44,27±1,90			3,91±0,66			103,05±3,52		
	N20	17,18±0,55			48,33±0,98			43,75±2,05			3,99±0,72			106,09±3,57		
	N40	16,30±0,41			48,85±0,79			44,27±2,20			4,21±0,78			103,96±3,42		

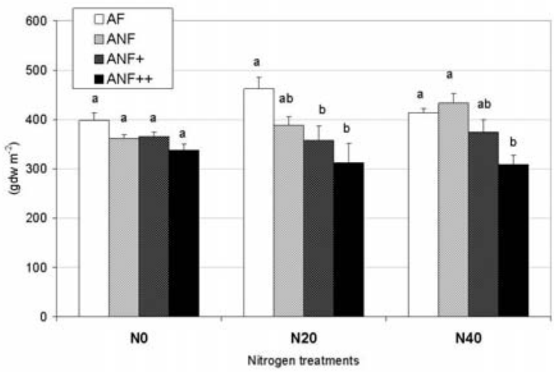

Fig 1. Green biomass for the different O3 and N treatments at the second harvest. FA= charcoal filtered air, NFA= non filtered air, NFA+= non filtered air supplemented with 20 nl l^{-1} of O_3, NFA++= non filtered air supplemented with 40 nl l^{-1} of O_3. N0= soil N background, N20=20 Kg N ha^{-1}; N40=40 Kg N ha^{-1}. Different letters indicate significant differences among means (mean ± se).

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THE SIMULATED IMPACT OF DIFFERENT FERTILIZATION SYSTEM ON YIELD AND THE UTILIZATION OF NITROGEN FROM WHEAT ROOT ZONE LAYERS UNDER SHORTAGE OF WATER

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The water shortage reduces the uptake and utilization of nutrients by crops due to less demand and availability for root uptake. Due to drought N from fertilizers broadcast applied during growth is often not effectively utilized. When top soil resources are exhausted nitrate shifted during winter to deeper layers is effectively depleted from moist soil by relatively sparse roots. It suggests possible strategy to manipulate distribution of nitrate in a soil profile for better utilization by a crop. However, the accumulation of nitrate in deep layers also increases the risk of leaching. The aim of the study was to examine the impact of nitrate content manipulation in subsoil layers under differentiated water supply, using crop model.

Materials and Methods

Model CERES-Wheat (Jones et al. 2003) was calibrated (Haberle 2007) and validated with data from field experiments with winter wheat (*Triticum aestivum* L.) in Prague-Ruzyne. In the experiment, the wheat at low (N1) and high N levels (N2) was grown under three water regimes differentiated from anthesis: Stress (S), ample water (Ir) and rain-fed treatments (R) (Haberle and Svoboda 2007, Haberle et al. 2008, Raimanová and Haberle 2010). In the experiments root, soil moisture and N_{min} distributions, N uptake and grain yield were observed. Daily meteorological data from six wheat season (2003-2009) modified according to S and Ir water supplies were used. Two levels of N_{min} in 0-150 cm soil zone (mostly in 0-45 cm) at autumn, high (195 kg N/ha) and medium (110 kg N/ha), were simulated. Three systems of fertilization were simulated: 1) the standard spring application of 200 kg N/ha in calcium nitrate (CAN) divided to three doses (30-100-70) (treatment SPR), 2) the application of 200 kg N/ha at autumn, before sowing (AUT), and 3) 30 kg N/ha at early spring and the (hypothetical) injection of 170 kg N/ha in CAN at the start of April to depth of 60 cm (INJ) were simulated. The yield, the content of grain N (as the indicator of grain quality), root depth and distribution in soil, water and nitrate contents and distributions, and indicators of N and water availability for the wheat crop were simulated.

Results and Discussion

Water stress reduced yields on average of years to 72-79 % (medium N_{min}) and 82-84 % (high N_{min}) of control (R). Ir increased yields by 4-9% in comparison with R, similarly to impact of the water regimes observed in the field experiments. The SPR reduced grain yield to 63-73 % of AUT and INJ yields under medium N_{min}. Under high N_{min} reduction reached only 4-5%. S treatment increased grains % N in all fertilization treatments (6-18 %), ample water supply reduced it by 1-2 %. SPR fertilization reduced grain N significantly under medium N_{min} in all water regimes, average values were 2.62 %, 2.47 % and 2,18 % in AUT, INJ and SPR systems. At high N_{min} level the effect was weaker, average grain N contents were 2.75 %, 2.74 % and 2.58 % in AUT, INJ and SPR systems. Autumn fertilization increased nitrate content down to 60 cm at spring but the downward shift was (somewhat unrealistic) low, probably due to high water capacity of the soil and low precipitation. Stress and irrigation did not modify nitrate content in subsoil (under 60 cm) at harvest, the differences were evident in shallow layers. The simulation of leaching needs to be better calibrated. Without fertilization nitrate was depleted to low contents, 39-40 kg N/ha in the whole profile, under all water regimes. The model simulated that with nitrate fertilization at autumn at medium and high N_{min} 78-91 kg and 143-156 kg of nitrate N per ha were left at harvest. INJ fertilization systems increased (14-18 kg N/ha) and reduced (26-28 kg N/ha) residual nitrate content under medium and high N_{min}, resp., in comparison with AUT. SPR strongly increased residual nitrate, on average by 82 kg N/ha under medium N_{min}, in agreement with the impact on yield, and slightly reduced the content under high N_{min}. Rooting depth was slightly reduced by post-anthesis stress (by 13 cm) in comparison with ample water supply (Ir) while we observed the same or slightly higher depth under stress. Simulated effect of N_{min} and N rate was negligible, in agreement with results of our previous field experiments at the site.

Conclusions:

The simulation study and the results of field experiments with induced water stress suggest that manipulation of nitrate distribution in root zone is worth of further study as the strategy for effective nitrogen utilization under post-anthesis drought and reduction of residual nitrate content.

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EFFECTS OF LEGUMINOUS COVER CROPS ON SPRING BARLEY PRODUCTION ON A LIGHT TEXTURED SOIL IN IRELAND

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Vegetative cover established after harvest of a spring cash crop and allowed grow over the fallow period can significantly reduce nitrate leaching by accumulating nitrogen in its biomass (Hooker at al., 2008). In order to replace fertiliser N inputs to subsequent crops, the N accumulated must be mineralised in the soil sufficiently early in the growing season of the subsequent crop to be of benefit to that crop. In many instances the effect of non-leguminous covers, particularly natural regeneration, on the N nutrition of the subsequent crop can be small (Hackett, 2012). In addition to accumulating residual N from the soil, use of leguminous cover crops can lead to accumulation of N through biological nitrogen fixation. Little work has been carried out examining the potential of leguminous cover crops under Irish conditions. The objective of this work was to determine the effect of leguminous cover crops on yield and N nutrition of succeeding spring barley under typical Irish conditions.

Materials and Methods

A split-plot spring barley experiment with four replications was carried out in two seasons, 2012 and 2013 on a light textured soil at the Crops Research Centre, Oak Park, Carlow, Ireland. Main plot factor was vegetative cover type, sub-plot factor was N rate (0 kg N/ha, 120 kg N/ha). Six vegetative covers were included four of which were legumes; hairy vetch (*Vicia villosa* Roth., cv. Ostaat Dr. Baumanns), peas (*Pisum sativum L.*, cv. Arkta), white mustard (*Sinapsis alba* L, cv.), lentil (*Lens culinaris*L. cv. Lentifix), grass pea (*Lathyrus sativus* L., cv. Nfix) and natural regeneration (NR weeds and volunteer cereals from the previous crop). Vegetative covers were sown on 25 August 2011 and 28 August 2013. The covers were subsequently incorporated by ploughing and spring barley sown on 20 February 2012 and 5 March 2013. Of the 120 kg N /ha applied to the N treatments, 30 kg N/ha was applied at sowing and the remainder at the mid tillering stage of crop growth. All other inputs were applied according to standard farm practice uniformly across the trial areas. At crop maturity grain yield, grain protein and grain N accumulation were determined. Results were analysed using ANOVA.

Results and Discussion

Visual inspection of the growth of the cover crops indicated that growth in the first season was much greater than in the second season. In the second season extensive damage due to slug and bird grazing meant that growth of all covers was very low. Grain yield, protein content and grain N accumulation results are presented in Table 1. N application significantly increased barley grain yield, protein content of the grain and grain N accumulation in both seasons. There was a significant effect of cover type on grain yield, protein and grain N in 2012 but not in 2013. However the effect of cover type in 2012 on yield and protein content was influenced by N application. Where no fertiliser N was applied hairy vetch, peas, lentil and grass pea significantly increased yield by 3.3 t/ha, 1.9 t/ha and 1 t/ha respectively compared to the NR treatment, while mustard had no significant effect on yield compared to the NR treatment. Where fertiliser N was applied hairy vetch and peas increased yield significantly compared to the NR but yield increases were lower than where no fertiliser was applied (0.49 t/ha and 0.32 t/ha respectively). A response to N inputs greater than 120 kg N/ha would be expected on this site. These results suggest that leguminous cover crops can supply N at a time where it can be used for yield formation in spring barley to crops that have received sub-optimal amounts of fertiliser N. They also suggest that leguminous cover crops can give yield benefits greater than the current practice of using NR on land during the fallow period between spring crops. For protein in 2012 there was no significant difference between cover types where no fertiliser was applied with the exception of grass pea, which gave a significantly lower protein content than the other covers. Where fertiliser was applied, hairy vetch gave a significantly greater protein content than peas which in turn gave a significantly greater protein content than lentil, with all three being significantly greater than the NR treatment. Grass pea and mustard had similar proteins to the NR treatment where no fertiliser was applied. There was no significant interaction between cover type and N application for grain N accumulation in 2012. Hairy vetch, peas and lentil significantly increased grain N uptake compared to the NR treatment by 36.8, 22.8 and 11.1 kg N/ha respectively, averaged over N rates. There was no significant difference between grass pea, mustard and the NR respectively.

Table 1. Effect of cover type and N application on grain yield, protein content and grain N accumulation of spring barley over two seasons.

Cover type		Yield (t/ha @85% DM)		Protein (% @100%DM)		Grain N (kg N/ha)	
2012		-N	+N	-N	+N	-N	+N
Hairy vetch		6.91	8.70	8.75	11.17	82.52	132.08
Peas		5.56	8.51	8.79	10.24	66.63	118.61
Lentifix		4.62	8.34	8.60	9.54	53.85	108.26
Nfix		4.06	8.12	8.16	9.20	45.09	101.51
Mustard		3.58	7.86	8.76	9.05	42.70	96.95
NR		3.62	8.02	8.63	9.07	42.47	98.96
LSD	Cover type	0.285		0.408		4.534	
(5%)	N application	0.155		0.128		1.950	
	C x N	0.411		0.338		ns	
2013							
Hairy vetch		3.71	7.96	7.89	11.16	39.82	120.89
Peas		4.13	7.97	8.09	11.41	45.45	123.38
Lentil		3.95	8.04	8.34	11.32	44.81	123.78
Grass pea		3.70	8.02	8.27	10.95	41.62	119.44
Mustard		3.64	8.20	8.04	10.99	39.88	122.67
NR		3.60	7.76	7.76	11.09	37.97	116.93
LSD	Cover type	ns		ns		ns	
(5%)	N application	0.165		0.168		2.020	
	C x N	ns		ns		ns	

Conclusion

Leguminous winter covers have the potential to supply significant amounts of N to spring barley on sandy soils under Irish growing conditions. However the effect varies between seasons, which appears related to the amount of growth achieved by the cover crop. Further work is required to determine if the supply of N from a leguminous cover can be predicted with sufficient confidence to allow reductions in fertiliser N.

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STABILISATION OF RED CLOVER NITROGEN RHIZODEPOSITION IN DENSITY FRACTIONS OF SOILS WITH DIFFERENT FERTILIZATION HISTORIES

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Clover-grass leys provide essential input of external nitrogen (N) by biological di-nitrogen fixation to crop rotations in low input agro-ecosystems such as organic farming. While aboveground biomass is generally removed as fodder and only a proportion of its N will return to the field as manure, N in stubble and belowground N (BGN) provide the major benefit to subsequent non-legumes. BGN is defined as root N plus the N derived from rhizodeposition (NdfR) and comprises a large variety of N compounds lost from living plant roots and root turnover (Wichern et al. 2008). Those compounds can directly be used by soil microorganisms and are rapidly turned over. Generally it is assumed that at the end of a vegetation period 45-60% of red clover NdfR is stabilized in soil organic matter (SOM), whereas the remainder will be recovered as dissolved N, microbial N and N transferred to associated grasses. The objectives of our study were i) to determine the stabilization of NdfR by red clover in different soil density fractions and ii) to evaluate the influence of different fertilisation histories on the stabilization of NdfR in a long term field experiment using ^{15}N labelling methods.

Material and methods

We studied during 2011 and 2012 the NdfR inputs by red clover as well as the stabilization in SOM fractions in a two component grass (*Lolium perenne* L.)-clover (*Trifolium pratense* L.) mixture using ^{15}N leaf labelling- and density fractionation-methods (based on Sollins et al. 2009). In spring 2011 the red clover-ryegrass mixture was planted in microplots (PE tubes with 39 cm diameter, 0-25 cm soil depth), situated in the clover-grass ley of the DOK long term field experiment (Basle, Switzerland) comparing organic and conventional cropping systems since 1978 (DOK experiment: bio-Dynamic, bio-Organic, Konventionell) (Mäder et al. 2002). Four DOK treatments were selected: Control with zero fertilization (NON), bio-organic with regular (ORG2) and half (ORG1) dose manure fertilisation and conventional with regular mineral fertilization (MIN2). To determine NdfR 11 clover plants were ^{15}N leaf-labelled after each cut with ^{15}N enriched urea. In October 2011 and 2012 the soils of the microplots were excavated, physically recoverable roots were removed by hand and separated into clover and grass roots. The soil was extracted two times with 0.05 M K_2SO_4 to remove roots and dissolved N in the first step and microbial N in the second step. The remaining soil was separated into four fractions comprising two particulate- and two mineral associated-organic matter fractions by density fractionation with sodiumpolytungstate. Roots and soil fractions were analysed for N content and ^{15}N and the amount of NdfR stabilized in the different soil fractions was calculated according to Janzen & Bruinsma (1989), assuming that the NdfR had the same isotopic composition as the roots. For NdfR inputs see Mayer et al. 2014 in the same proceedings.

Results and discussion

After 6 (2011) and 18 (2012) months of meadow use the percentage of red clover NdfR in the respective fractions (%NdfR) decreased from the lightest [< 1.65 g cm^{-3}] to the heaviest fractions [2.25-2.55 g cm^{-3}] and [> 2.55 g m^{-3}] (fractions [< 1.65] > [1.65-2.25] > [2.25-2.55] = [>2.55] g cm^{-3}, table 1a). From 2011 to 2012 %NdfR increased in all fractions and in the bulk soil due to an accumulation of NdfR inputs. In 2011 no differences could be found between the treatments whereas in 2012 %NdfR in fraction [2.25-2.55] and [>2.55] g cm^{-3} and in the bulk soil was significantly lower in NON compared to the remaining treatments. In contrast to %NdfR the largest proportion of NdfR (g m^{-2}) in the respective fraction was found in the clay-rich fraction [2.25-2.55] g cm^{-3} (fractions [2.25-2.55] > [1.65-2.25] > [< 1.65] > [< 2.55] g cm^{-3}, table 1b) which was the fraction with the largest absolute soil N amount (N amount: fractions [2.25-2.55] > [1.65-2.25] = [< 2.55] > [< 1.65] g cm^{-3}). The stabilisation in the clay-rich fraction, [2.25-2.55] g cm^{-3} was already observed after 6 months in 2011. This indicates a fast stabilization of major amounts of NdfR via microbial incorporation and subsequent sorption of amino acids and proteins from microbial residues to the active sites of clay minerals (Miltner et al. 2009). From 2011 to 2012 amounts of NdfR increased in all fractions except the heaviest (data not shown). Also the relative proportion of fraction [2.25-2.55] g cm^{-3} decreased whereas the relative proportion of fraction [1.65-2.25] g cm^{-3} increased significantly.

Table 1a) Percentage of red clover N derived from rhizodeposition in soil density fractions after six (2011) and 18 months (2012) of clover-grass establishment in different DOK cropping systems and b) proportion of amounts of NdfR in the respective fraction

	a) percentage of N derived from rhizodeposition					b) proportion of NdfR in fraction (total = 100%)			
fraction	<1.65	1.65?2.25	2.25?2.55	>2.55	bulk soil	<1.65	1.65?2.25	2.25?2.55	>2.55
treatment	2011					2011			
NON	6% $^{n.s.}$	1.4% $^{n.s.}$	0.7% $^{n.s.}$	0.5% $^{n.s.}$	0.9% $^{n.s.}$	25% $^{n.s.}$	18% $^{n.s.}$	48% $^{n.s.}$	8% $^{n.s.}$
MIN2	4% $^{n.s.}$	1.8% $^{n.s.}$	1.2% $^{n.s.}$	0.8% $^{n.s.}$	1.4% $^{n.s.}$	16% $^{n.s.}$	24% $^{n.s.}$	51% $^{n.s.}$	10% $^{n.s.}$
ORG1	5% $^{n.s.}$	1.8% $^{n.s.}$	1.0% $^{n.s.}$	0.6% $^{n.s.}$	1.2% $^{n.s.}$	20% $^{n.s.}$	23% $^{n.s.}$	48% $^{n.s.}$	10% $^{n.s.}$
ORG2	4% $^{n.s.}$	2.0% $^{n.s.}$	1.2% $^{n.s.}$	0.9% $^{n.s.}$	1.5% $^{n.s.}$	16% $^{n.s.}$	27% $^{n.s.}$	46% $^{n.s.}$	11% $^{n.s.}$
	2012					2012			
NON	36% $^{n.s.}$	12% $^{n.s.}$	3% ^{b}	2% ^{b}	5% ^{b}	18% $^{n.s.}$	30% $^{n.s.}$	46% $^{n.s.}$	6% $^{n.s.}$
MIN2	46% $^{n.s.}$	15% $^{n.s.}$	5% ^{a}	3% ^{a}	8% ^{a}	26% $^{n.s.}$	30% $^{n.s.}$	40% $^{n.s.}$	5% $^{n.s.}$
ORG1	37% $^{n.s.}$	16% $^{n.s.}$	5% ^{a}	3% ^{a}	8% ^{a}	17% $^{n.s.}$	34% $^{n.s.}$	43% $^{n.s.}$	6% $^{n.s.}$
ORG2	36% $^{n.s.}$	14% $^{n.s.}$	5% ^{a}	3% ^{a}	8% ^{a}	23% $^{n.s.}$	33% $^{n.s.}$	38% $^{n.s.}$	6% $^{n.s.}$

Levels not connected by same letter are significantly different (least significant difference $p < 0.05$)

Conclusions

Large amounts of NdfR were rapidly stabilized in the clay-rich fraction after 6 months. With increasing duration of the clover-grass ley amounts of NdfR increased in density fractions whereas the relative proportion of clay-rich fraction decreased, the relative proportion of the nearest lighter fraction increased. Effects of fertilisation history on stabilisation of NdfR were small and stabilisation was mainly driven by input.

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EXPLORING OPTIMAL FERTIGATION STRATEGIES FOR ORANGE PRODUCTION, USING COMBINED SOIL-CROP MODELLING

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From a literature review (Qin et al., this issue) it became clear that in fruit tree cropping systems the efficiencies of water use and nitrogen use in drip-fertigated situations are still low. This is due to, for example, erratic rainfall events, and lack of knowledge in water and N demands. Conducting a large number of field trials for perennial crops is time-consuming and costly. Therefore, the objective of this study was to use a combined soil-crop model in testing several pre-defined fertigation strategies to determine which strategies are good and which should be avoided.

Materials and Methods

A 3D model describing water movement and N transport in the soil including uptake by roots (FUSSIM3D; Heinen and de Willigen, 1998) was sequentially coupled to the FAO crop model AquaCrop (Steduto et al., 2008; 2012). Average climatic conditions for the south of Spain were imposed. Different fertigation scenarios were considered including the following factors: 1) three levels of water supply, 2) three levels of N-supply, 3) five partitionings (see below) of N-supply during the growing season, 4) three soil types, and 5) two time-windows of application (continuous or in pulses). A total yearly N-demand of 200 kg/ha was assumed which was distributed over the growing season according to a bell-shaped pattern. The partitionings considered were 3-2-1, 2-2-2, 1-2-3, 2-3-1, 1-3-2, where, e.g., 3-2-1 means that in the first period 3/6th of the total supply is delivered, in the second period 2/6th, and in the third period 1/6th. The five partitionings did not exactly match this bell-shape as it was assumed that in most cases the exact N-demand distribution over time is unknown. Fertigation scenarios were judged based on predicted yields, and predicted N-losses from the soil as denitrification and leaching from the bottom of the root zone.

Results and Discussion

Modelled yields ranged between 33 to 43.5 ton/ha/yr where the highest value was the assumed maximum yield possible for oranges under the considered circumstances. N-uptake, N-leaching from the root zone and denitrification increased with increasing N-supply. More irrigation led to wetter conditions in the soil and consequently to more leaching and denitrification. Not surprisingly the worst scenarios were those with too low or too high water and/or N-supply with respect to the total demand. Good results were obtained for strategies where the N-supply matches the N-demand curve most closely. Table 1 presents the ranking of several scenarios based on either yield or losses to the environment, or to a combination of both.

Table 1. All scenarios with a N-supply of 200 kg/ha, put in order of the highest yield, the lowest N-leaching from the root zone, the lowest denitrification, the lowest total N-loss and two overall ranks, respectively. In the first overall rank the same weight is given to yield (Y), N-leaching (L) and denitrification (D), in the second overall rank Y, L, D are multiplied with a weight of 6, 2 and 1 respectively. The best three scenarios per category are green and the worst three are red.

			Individual ranks				Summed ranks	
#	F_irr	Portion	Yield (Y)	Leach (L)	Denit (D)	L+D	Y+L+D	6Y+2L+D
1	100	3-2-1	1	8	3	6	4	2
2	100	1-2-3	12	10	12	10	12	12
3	100	2-2-2	8	3	6	3	6	8
4	80	3-2-1	6	4	1	3	2	5
5	80	1-2-3	11	6	6	6	9	10
6	80	2-2-2	6	1	1	1	1	4
7	120	3-2-1	5	9	3	9	6	7
8	120	1-2-3	13	11	14	12	13	13
9	120	2-2-2	9	6	11	8	10	9
10	100	3-2-1 P	3	12	3	11	8	5
11	100	1-2-3 P	14	14	12	14	14	14
12	100	2-2-2 P	10	13	9	13	11	11
13	100	1-3-2	3	2	9	2	5	3
14	100	2-3-1	1	4	6	5	2	1

Conclusions

The best fertigation strategy ideally results in: 1) high crop yields, 2) low N-leaching losses, 3) and low denitrification losses. The most dominant factor in the scenario simulations was the supplied N-amount. Judged by yield alone, it was smart to supply 100% irrigation and divide the N-supply according to the 2-3-1 or the 3-2-1 fractionation. Judged by total N-loss, the worst fertigation strategies that can be chosen were to either apply the N in short pulses or to supply most of the N in the last period. Lowest total N-losses were obtained when N was applied through fertigation evenly, according to the N-demand pattern, or applying most of the N in the first period in combination with a low irrigation amount.

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WATER AND NITROGEN INTERACTIONS IN ORANGE PRODUCTION: A META-ANALYSIS

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Sustainable orange production requires quantitative insights in the interactions between water and nitrogen use in crop yield, and in water and nitrogen use efficiencies. Here, we report on a review and meta-analysis of studies about water and nitrogen use in orange production.

Materials and Methods

We used the Web of Science database for literature search. Publications with reports on orange yield, water and nitrogen use were selected for the meta-analysis. In order to compare water and N saving potentials between regions, we used maximum yield, median water use efficiency (WUE) and median nitrogen use efficiency (NUE) as the reference targets. For statistical analysis, we used quantile regression via R (version 3.02).

Results and Discussion

There were large variations between regions. Orange yields ranged between 2 and 93 ton/ha. Water inputs ranged between 500 and 4000 mm per year, and N inputs from 20 to 350 kg/ha. As a result, WUE and NUE varied also widely, from 0.1 to 7.9 kg/m^3 and from 10 to 1950 kg/kg N, respectively. On average, water and N inputs could be reduced by 16% to 30% and by 26% to 47%, respectively, without severe reduction in yield, while maintaining median level of WUE and NUE (Table 1). Overall, yield responded positively to both water and N inputs, but there was a negative water times N input interaction. WUE responded negatively to water input but positively to N input; NUE responded positively to water input but negatively to N input. Water and N interactions were significant in yield, but not in WUE and NUE. Optimizing water and N inputs in orange production is complicated by erratic rainfall events, and by lack of knowledge on water and N demands by orange trees.

Conclusions

Current water and N use in orange production are not yet fully optimized because of complexities in synchronizing water and N supply to the plant demand simultaneously. There is still a great potential for improving WUE and NUE in orange production. Reducing water and N inputs to the optimal levels is the first step towards increased WUE and NUE. The impacts of different water and N management (e.g. fertigation) strategies need to be tested further.

Table 1. Water and N saving potentials in targeting maximum yield, median WUE and median NUE in different regions. Maximum yield, median WUE and median NUE were listed in green columns and set as the production targets (i.e. T1, T2 and T3). Maximum input for water and N were listed in red columns. Accordingly, water and N saving potentials for each specific targets were listed in blue columns, as relative of maximum water or N input. For example, water saving potential for maximum yield is calculated as: saving potential = (water input for maximum yield – maximum water input) / maximum water input.

Region	T1	T2	T3	Maximum input		T1	T2	T3	T1	T2	T3
	Yield (ton/ha)	WUE (kg/m3)	NUE (kg/kg N)	Water (mm)	N (kg/ha)	Water saving potential (%)			N saving potential (%)		
Spain	62	5.6	223	1276	260	-38	-51	-44	-42	-42	-8
Iran	27	2.1	137	1188	150	-6	-37	-12	0	0	0
FL,USA	93	3.5	318	2297	336	-20	-29	-20	-17	-58	-17
Uruguay	83	3.8	313	1877	192	-22	-15	-42	0	0	0
Brazil	57	4	320	1153	260	0	-14	-14	-62	-62	-62
Cook Islands	72	1.9	168	3924	351	-45	-53	-36	0	-52	0
Australia	62	3.2	345	1348	255	-12	-31	-31	-61	-61	-61
AZ,USA	20	0.7	108	1970	204	0	-12	-12	-33	-67	-67
Israel	77	3.4	255	1946	320	0	-31	-4	-47	-80	-23
Mean	**61**	**3.1**	**243**	**1887**	**259**	**-16**	**-30**	**-24**	**-29**	**-47**	**-26**

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EFFECTS OF DROUGHT AND LEGUME CONTENT ON GRASSLAND NITROGEN YIELD AND SYMBIOTIC N2 FIXATION

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Increased incidence of drought, as predicted under climate change, highlights the need to design grassland management systems for forage production that are adapted to future climate scenarios. Compared to monocultures, simple grassland mixtures can result in increased yields, greater stability in response to disturbance (i.e. drought), reduced invasion by weeds and improved nutrient retention (Hooper *et al*., 2005; Finn *et al*., 2013). In particular, the interaction between species with and without the ability for symbiotic N_2 fixation (SNF) increases yield benefits (Nyfeler *et al*., 2011). This study assessed the effect of summer drought on the N yield of monocultures and mixtures with experimentally varied legume proportions. We used measurements of SNF to reveal how legumes and non-legumes adjust their N nutrition in response to these factors.

Materials and Methods

In August 2010, monocultures and mixtures were sown in 66 plots at Tänikon research station, Switzerland in a completely randomised design with 3 replicates. Four species were selected based on their ability for symbiotic N_2 fixation, two legumes (*Trifolium repens* L and *Trifolium pratense* L) and two non-legumes (*Lolium perenne* L and *C intybus* L). Plots were sown to consist of four monocultures, six binary communities (50% of two species), and one equi-proportional community (25% of each of four species). Using rainout shelters, half of the plots were subjected to a drought treatment of 10 weeks of rain exclusion in 2011. Plots received 145 kg N ha^{-1} yr^{-1} split over five applications. We used the isotope dilution method to quantify SNF: A solution of double ^{15}N labelled (98 Atom%) ammonium nitrate was injected at 5 cm depth in a 50 × 50 cm sub plot within each 3×5 m plot. At the end of the drought treatment, the sub-plots were harvested at 6 cm height and separated into component species, and samples were dried and subjected to ^{15}N and total N analysis. The remainder of the plot was harvested with a plot harvester and a subsample taken for DM and total N analysis. We used the following model (based on Nyfeller *et al*., 2011) to assess the effect of drought and legume proportion (Legume):

$$y = \alpha + \beta_1 \cdot \text{Drought} + \beta_2 \cdot \text{Legume} + \beta_3 \cdot \text{Legume}^2 + \beta_4 \cdot \text{Legume} \cdot \text{Drought} + \beta_5 \cdot \text{Legume}^2 \cdot \text{Drought}$$

Results and Discussion

Total N yield was reduced under drought conditions ($p < 0.001$) by on average 17 kg N ha^{-1} cut^{-1} (Fig. 1a). The reduction was highest in non-legume stands (61%) and lowest for the mixed and pure legume stands (23% and 18%, respectively). The N yield from SNF was not affected by drought (Fig. 1d), which could be attributed to the higher percentage of legume N derived from SNF (%SNF) under drought compared to control conditions. Total N yield of the sward was strongly linked to the sown legume proportion ($p < 0.001$), and the N yield ranged from 19.5 kg N ha^{-1} to 82 kg N ha^{-1} for swards with legume proportion of 0 and 1, respectively (Fig. 1a). The N yield in mixed swards (legume proportion 0.5) was higher than expected from the proportional contributions of the non-legumes and legumes, indicating that positive mixing effects (overyielding) were occurring, as evident from the highly significant non-linear legume effect (Fig. 1a). For mixed swards containing 50% legumes, the overyielding was on average 26%. The overyielding could be mainly attributed to the higher than expected N uptake by non-legumes in mixed swards (Fig. 1b), indicated by a highly significant ($p< 0.001$) non-linear relationship between legume proportion in the sward and N yield of non-legumes. This may be the result of the competitive advantage of non-legumes to take up N, which can lead to 'nitrate sparing' (Nyfeler *et al*., 2011). At the same time, legumes maintained their N yield by increasing the %SNF. This fits well with other evidence that legumes regulate their activity of SNF to close the gap between their N-demand for growth and the availability of N from non-symbiotic sources. In conditions with low availability of soil N (i.e. drought conditions and swards with a high proportion of non-legumes), legumes were mainly depending (90-98%) on SNF, whereas in condition with higher soil N availability (i.e. non-drought conditions and swards with a high proportion of legumes) the %SNF was down to 60%.

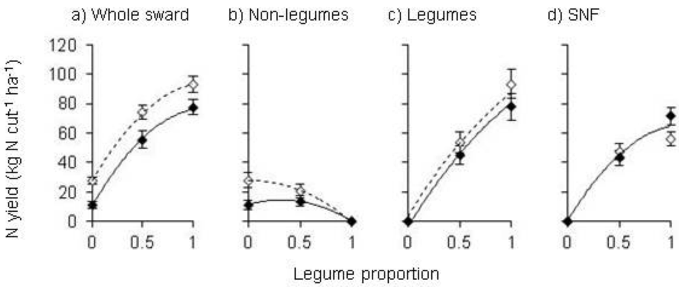

Fig. 1. **Total N yield (kg N cut^{-1} ha^{-1}) (a), non-legume N yield (b), legume N yield (c) and N yield from SNF (d) as affected by the legume proportion under control (open diamonds and dashed lines) and drought (solid diamonds and lines) conditions during the second harvest of the drought period. Data points represent the treatment mean (± SE). Regression lines are based on the model indicated above (Materials and Methods), with Leg = legume proportion.**

Conclusions

The combination of legumes and non-legumes in simple grassland mixtures maintained their positive effect on N yield under drought conditions. Measurement of SNF provided valuable insights into the processes behind the observed resistance to drought.

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RESIDUAL SOIL MINERAL NITROGEN AS A READILY AVAILABLE SOURCE TO CROPS IN AN IRRIGATED MEDITERRANEAN AREA

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Nitrogen (N) is one of the major plant nutrient affecting the crop yield and quality. Intensified agricultural production can not be thought without fertilization and irrigation. On the other hand, N management and efficient use of N fertilizers are also vital from the economical and environmental standpoints. Considering pre and post harvest soil profile mineral nitrogen (Nmin) as nitrate-nitrogen plus ammonium-nitrogen (NO3-N + NH4-N) in fertilizer recommendations can be an approach in N management. Akarsu Irrigation District (AID, 9,495 hectares) in the Seyhan Plain of southern Turkey is under intensive irrigated agriculture with excess use of N fertilizers (Qualiwater, 2010). Based on the results of limited soil and plant analysis, soil Nmin level in the region's soils is hardly covered in the fertilizer recommendations. Therefore, the objective of this research was to determine the pre and post harvest Nmin values in the rooting depth between 2007-2009 to be used in fertilizer recommendations as a readily available N source to main crop corn.

Materials and Methods

The study area (9,495 ha) is located in the Mediterranean coastal region comprising the most intensively cropped and irrigated area of Turkey. A Mediterranean-type climate prevails in the region. The soils of the location are mainly alluvial, relatively uniform, deep and high in clay, mostly smectite, calcium carbonate (20-35%), and pH (7.5-8.5), but low organic matter (0.8-1.2%), and deep cracks are common during the dry summer season. The cropping pattern of the district was dominated by wheat (Triticum aestivum), corn (Zea mays L.), citrus (Citrus sinensis L.), cotton (Gossypium hirsutum L.) and vegetables during the study years. Double cropping (wheat-corn or –cotton) is a common practice in the area. Based on the digitalized cropping-pattern maps, over 70% of the total irrigated land was covered by wheat and corn in the years under the study. Fertilization practices in the AID were obtained through farmer's interviews. Nitrogen fertilizer application rates to the crops varied between 70 to 340 kg N ha-1, with an average of 245 kg N in these 3 years. Based on the cropping pattern, N fertilizer application continues year-around except a few months in early fall when no crops are fertilized in the field. Preplant and post-harvest soil samples from the depths (0-30, 30-60 and 60-90 cm) of 54, 56 and 23 main crop corn fields in these respective years were processed, and analyzed for NO3 and NH4 to calculate Nmin in the profile. Soil NO3 and NH4 concentration values in each depth were converted to kg NO3-N ha-1 and kg NH4-N ha-1, and added up (NO3-N + NH4-N) for the 0-90 cm rooting depth.

Results and Discussion

Based on the local cropping pattern, wheat, corn and citrus are the major crops of the Akarsu ID. Distribution and growth of the other crops, i.e. cotton, vegetables, melons, legumes and fruit trees vary year to year. The survey data, specifically collected for this research, indicated that application rates of N fertilizers to these crops are exceedingly high compared to the recommended rates; for example, 195 kg N for wheat, 340 and 320 kg N for main and second crop corn and 180 kg N h-1 for citrus. The most N fertilizers were applied during fall, winter and spring as preplant treatment and side dressing without considering pre existing soil Nmin. Since the wheat and corn growth is slow during late fall and early spring, vital amount of soil and fertilizer N accumulate in the profile as more than plant use. Post harvest soil Nmin values after the main crop corn showed that approximately ¼ of the fertilizer applied remained in the 0-90 cm depth as residual N or readily plant available (Table 1). These post harvest Nmin values closely agree with the preplant values. Therefore, up to 71 to 94 kg Nmin ha-1 were hold or equilibrated in the rooting depth as readily available residual N. Similar pre plant values in the area were also reported earlier for wheat (Ibrikci et al., 2001).

Table 1. Preplant and post harvest Nmin values (kg Nmin ha^{-1}) in the main crop corn fields in 2007, 2008 and 2009.

Depth	2007[a]		2008		2009	
(cm)	Preplant	Post Harvest	Preplant	Post Harvest	Preplant	Post Harvest
0-30	27.8	34.6	33.8	37.9	27.8	25.1
30-60	42.2	30.4	42.8	39.7	34.0	27.7
60-90	6.2	5.8	6.8	6.9	32.0	25.5
Total	76.2	70.8	83.3	84.5	93.8	78.3

[a] The numbers are average of 54, 56 and 23 corn fields in 2007, 2008 and 2009 respectively

Residual soil profile Nmin after corn is an excellent available source of N to wheat. This amount is not only economically important but also important for being a safe source to seeds at germination. If the rotation continues as corn-corn, there is average of about 13 and 9 kg N ha-1 increase in the profile Nmin from post harvest until next year's corm planting (september to march). Most probably, the increment is a result of N mineralization either from the soil organic matter or from the plant residues incorporated into the soil. However, when each horizon's data was evaluated, it is not easy to withdraw a conclusion about consistent increments or declines between the post harvest and preplant periods.

Conclusions

Considerable amounts of preplant and post harvest Nmin values (71 to 94 kg Nmin ha-1) exist in the rooting the depths of irrigated corn fields. Readily plant available this N source needs to be used in fertilizers recommendations not only for crop production but also for environmental protection and the farmers' economy.

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FODDER VALUE IN WINTER TRITICALE VARIETIES FEPENDING ON PRODUCTION TECHNOLOGY

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In Poland, triticale grain is mainly used for fodder purposes. For this reason, the nutritional and digestibility value of the triticale grain is important. The aim of the research is to determine the effects of cultivation and variety on the content of essential amino acids in winter triticale grain under monoculture conditions.

Materials and methods

The effect of two technologies – integrated and intensive for nutritional value of winter triticale Pizarro and Pigmej varieties was studied in the IUNG-PIB Experimental Station in Pulawy. As a forecrop, winter wheat was cultivated. The study was conducted on soils classified with a good wheat complex valuation class IIIa and IIIb. The technologies differed inter alia with the level of mineral fertilisers and chemical plant protection against weeds, diseases and pests. The integrated technology dose of potassium fertilisers and phosphorus were determined based on the content of these components in the soil. The total dose of nitrogen was determined on the basis of the expected yield, soil conditions and knowledge of the field, also taking into account the type of cropping and fertilisation. Clarification into the individual doses of nitrogen was made on the basis of soil and plant tests. The amount of the first dose was specified on the basis of mineral nitrogen (N min) test. The size of the second dose was corrected and based on the assessment of nutritional status of plants through plant tests. Spraying in the integrated production technology was used after the pathogens threshold. At the stage of full grain maturity, the yield per hectare was specified. The grain samples were collected to determine the content of protein, fibre, fat, ash and alkylresorcinols and the content of essential amino acids in the triticale grain.

Results and Discussion

Under intensive technology, winter triticale grain yield increased by 4% compared to the grain yield obtained in integrated technology (Table 1). Researchers Zawiślak and Adamiak (1997) demonstrated a lack of significant effect on intensifying the production of an increased yield of triticale in terms of cereal monoculture. In studies of Nieróbca et al. (2008), triticale yielded best in medium intensive and intensive technology. The used technologies of cultivation have an impact on the content of essential amino acids in the studied grain triticale cultivars. It was at a higher percentage of carbohydrates and fat in the grain of triticale that are cultivated under intensive technology. Triticale grain of integrated technology contained more fat than grain from the integrated technology. In terms of the integrated technology, they had a significantly higher proportion of valine, isoleucine, leucine and phenylalanine in the protein. Research done by Stankiewicz (2005) indicated that the use of herbicides in the cultivation of triticale did not cause a significant change in the composition of amino acids in the grain. The Pigmej variety yielded about 15% higher and had a higher content of carbohydrates, fat and alkylresorcinols than the Pizarro variety. In contrast, amino acid content in the grain of the Pigmej variety was lower than the Pizarro variety, except for arginine. Significant differences in the content of lysine and methionine were found between genotypes in the research done by Fernandez-Figares et al. (2000). In the author's own research, the Pizarro variety had a higher lysine content, while the methionine content was similar in both cultivars.

Table1. The influence of technology production and varieties on fodder value of winter triticale.

Trais	Technology production			Varieties		
	integrated	intensive	$LSD_{0.05}$	Pizarro	Pigmej	$LSD_{0.05}$
Grain yield ($t ha^{-1}$)	6.11	6.34	0.21	5.78	6.67	0.87
Content in grain						
Protein (% d.m.)	1.59	1.62	ns	1.64	1.57	ns
Fibre (% d.m.)	1.43	1.27	0.11	1.38	1.32	ns
Carbohydrates (% d.m.)	14.3	19.0	3.91	14.0	19.2	4.81
Fat (% d.m.)	1.91	2.10	0.12	1.92	2.08	0.13
Ash (% d.m.)	1.72	1.76	ns	1.79	1.69	0.98
alkylresorcinols ($mg\cdot kg^{-1}$)	307	295	ns	274	328	36.9
Amino acid ($g\cdot kg^{-1}$)						
Threonone	3.58	3.51	r.n	3.60	3.49	0.11
Valine	4.65	4.54	0.10	4.66	4.52	0.12
Isoleucine	3.86	3.72	0.11	3.81	3.77	ns
Leucine	7.26	7.14	0.12	7.23	7.17	ns
Phenylalanine	5.11	4.92	0.17	4.97	5.06	ns
Histidine	2.67	2.58	ns	2.61	2.64	ns
Lysine	3.79	3.78	ns	3.90	3.67	0.21
Arginine	4.99	4.94	ns	4.84	5.09	0.22
Methionine	1.97	2.16	0.15	2.07	2.06	ns
Cysteine	1.95	2.13	0.17	2.10	1.99	0.10

ns – non-significant differences

Conclusions

1. The intensive technology of cereal cultivation in monoculture conditions contributed to an increase in the content of certain amino acids such as methionine, cysteine and decreased content of valine, isoleucine, leucine and phenylalanine, compared to the integrated technology.
2. Grain from intensive technology is characterised by a higher content of carbohydrates and fat as well as a lower fibre content.
3. Variety Pizarro contained a higher amount of alkylresorcinols content threonone, valine, lysine and cysteine, but had a lower content of arginine in the protein than grain from the Pigmej variety.

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NITROGEN USE EFFICIENCY IN NATURAL ECOSYSTEMS AND LOW INPUT AGRICULTURAL SYSTEMS

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World livestock production systems can be classified in mixed and landless production systems and in pastoral systems. Mixed and landless systems are high input systems relying on a mix of concentrates (food crops) and roughage, consisting of grass, fodder crops, crop residues, and other sources of feedstuffs. Pastoral systems are low input systems depending almost exclusively on grazing and with low external inputs like synthetic fertilizers. Pastoral systems have a share of circa 18% in the global production of meat from beef as well as from sheep and goat (Bouwman et al., 2005).

This paper deals with biological nitrogen fixation (BNF) in grasslands as source of nitrogen instead of synthetic nitrogen fertilizers and with sustainable use of low input agricultural systems.

Materials and Methods

Published nitrogen balance sheets for dairy and sheep farms are used in this study. Balance sheets are only useful and interpretable when the system boundaries are given. For this study only external inputs and outputs like synthetic fertilizer, purchased animal feed, produced milk and meat and so on are taken into account. Farm grown forages internally used as feed and animal manure internally used as organic fertilizer are considered as internal entries which do not enter the balance sheet. Biological fixed nitrogen by forage legumes is considered as an entry on the balance sheet because when the fixed nitrogen becomes part of the farm soil system it is not distinguishable from synthetic nitrogen fertilizer. In this way it contributes in the same way as synthetic nitrogen fertilizer to nitrogen losses to the air and groundwater.

Results and Discussion

Nitrogen balance sheets for 6 dairy farms with varying nitrogen inputs of fertilizer and BNF are presented in Table 1. Farms in Ireland (1 and 2) and The Netherlands (3 and 4) show that use of BNF can replace fertilizer consumption and the table reveals that the nitrogen use efficiency (NUE) per hectare is equal for farms 1 and 2 respectively farms 3 and 4. Likely due to the higher stocking rates the farms with fertilizer (2 and 4) have 16-18% higher output of milk and meat per hectare in comparison with the BNF farms (1 and 3). The low input farm in New Zealand (5) shows a high NUE whereas the high input farm (6) has a moderate NUE. BNF by forage legumes offers good opportunities for reducing fertilizer consumption and as a side effect reduces the environmental costs of fertilizer production. Nevertheless forage legumes have some limitations in the management of production, conservation and utilization (Rochon et al., 2004; Peyraud et al., 2009).

Table 1. Nitrogen balances for different farming systems. Nitrogen input, product (meat and milk) and surplus in kg N per hectare, stocking rates per hectare and NUE calculated as product/input.

No		Stocking rate per ha	N fertilizer	BNF	N-feed	Total N input	N-product	N-surplus	NUE
1	Ireland	1.75 cow	80	88	27	205	67	138	0.33
2	Ireland	2.1 cow	180	9	32	232	79	153	0.34
3	The Netherlands	1.9 cow	16	176	47	279	69	210	0.25
4	The Netherlands	2.2 cow	208	0	85	333	80	253	0.24
5	New Zealand	3.3 cow	0	160	0	165	78	87	0.47
6	New Zealand	4.4 cow	410	40	41	496	114	382	0.23
7	New Zealand, 980 mm	20 sheep	0	130	0	140	18	122	0.13
8	Australia, 200 mm	1 sheep	0	5	0	7	1	6	0.14
9	Australia, 700 mm	10 sheep	0	130	0	136	11	125	0.08

Sources: 1+2 = Humphreys et al., 2008; 3+4 = Schils et al., 2000; 5-9 = Ledgard, 2001

Nitrogen balance sheets for 3 sheep farms in New Zealand (7) and Australia (8 and 9) are characterized by the absence of fertilizer and external feed. This means that variability in rainfall has a direct impact on the amount of BNF per hectare and subsequent on the stocking rates of sheep. The NUE of the sheep farms is lower compared to the dairy farms because the conversion of feed into meat is less efficient compared to the conversion of feed into milk. In pastoral farming systems, pasture production normally exceeds animal demand in the spring–summer period. Animal output and NUE of these farming systems will increase by conserving forage in spring-summer time for use during the following winter (Hou et al., 2008). In rangelands, plant and animal output together with NUE will increase when stocking rates are adapted to carrying capacities (Oliva et al., 2012).

Conclusions

Forage legumes offer good opportunities for reducing nitrogen fertilizer consumption but have some limitations in the management of production, conservation and utilization. Animal output and nitrogen use efficiency of rangelands will increase when stocking rates are adapted to carrying capacities and when the surplus of summer forage is conserved for the winter.

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INFLUENCE OF NITROGEN FERTILISATION ON CARBON AND NITROGEN MINERALISATION, MICROBIAL BIOMASS AND SOIL RESPIRATION IN GRASSLAND SOILS

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Nitrogen (N) released from soil organic matter (SOM) and plant residues is an important source of available N in permanent grasslands. Inorganic fertiliser application can alter root biomass and quality and therefore change the input and turnover of SOM. It has been shown, that low inorganic N fertilisation increased litter decomposition and high inorganic N fertilisation inhibited litter decay, but that also the litter quality plays an important role in influencing these processes. However, interactions of inorganic fertilisation and mineralisation processes in grassland soils are not well understood. Objectives of the present study were to (i) determine the potential carbon (C) and N mineralisation and the soil microbial biomass (MB) of grassland soils which have received different levels of inorganic and organic fertilisation, to (ii) evaluate the contribution of roots to C and N mineralisation and to (iii) determine the evolution of CO_2 and N_2O in relation to inorganic fertilisation status.

Materials and Methods

Investigations were done in a fertilisation field trial on permanent grassland (*Lolium perenne* L.) in Kleve (51.8°N, 6.2°E, 20 m a.s.l.). Design consists of various inorganic (calcium ammonium nitrate – CAN) and organic fertiliser (cow slurry) treatments from 85 to 340 kg N ha^{-1} in 4 replicates, and a plot without fertiliser addition. Total amount of fertiliser was applied in 4 steps throughout the growing season. Gas fluxes of the different treatments were measured *in situ* before each of 4 cuts using a portable photo-acoustic measuring device (INNOVA 1412i). Soil samples were taken at the same time for extractable organic and inorganic C and N fractions and the MBC and N. Microbial biomass estimated by chloroform-fumigation extraction (Brookes et al., 1985) including a pre-extraction step (Mueller et al., 1992). In a short term incubation, we investigated the effect of contrasting inorganic fertilisation rates and the presence of coarse roots on C and N mineralisation and MB under optimal conditions. Soil samples were collected and roots were separated manually. Soil samples were incubated either with or without roots. Evolution of CO_2 was measured every 2 to 7 days.

Results and Discussion

The evaluation of the short term incubation experiment provides a net N immobilisation in all treatments. The lowest amounts of mineralised N were detected at low external N levels. However, the amount of the added fertiliser and the immobilisation showed no linear correlation. In particular the mineralised N stagnated after 170 kg N ha^{-1} (CAN) at a level of 25 mg N kg^{-1} soil. A small effect of roots was detected, as we found a lower immobilisation of N in the presence of roots. Increasing inorganic fertiliser application reduced the MBC. An addition of roots entails a lower microbial biomass. Soil respiration within the incubation (fig 1) shows the relation of inorganic N-fertiliser amount and soil respiration is not linear. Highest cumulative CO_2-C fluxes were detected at lower external N. At increasing fertiliser amendment, cumulative flux decreased, which corresponds to the decrease in MBC. In contrast to the incubation experiment, gas fluxes in the field showed an increased soil respiration also at higher external N input (fig 2). This was due to the fact, that in the field CO_2 emissions are composed of root respiration and rhizodeposit influenced microbial respiration. Rhizorespiration was affected by increasing fertiliser N input, as reflected by an increased above-ground biomass and N yield, which may result in an increased root respiration and root exudation.

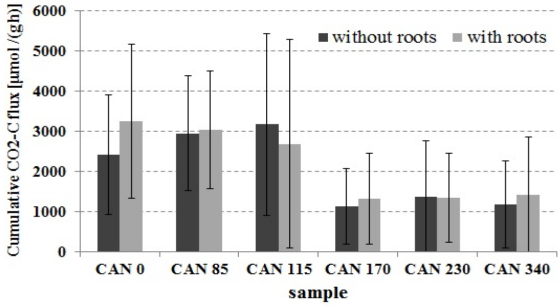

Fig.1: Cumulative carbon dioxide fluxes of fertiliser treatments with and without roots at the end of the imcubation experiment (Error bars indicate SD, n=5).

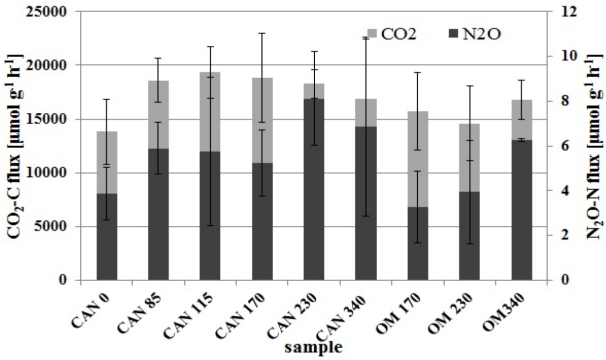

Fig. 2: Soil respiration and N_2O release in the field october 2013 (Error bars indicate STD; n=4).

In the field, N_2O emissions increased with increasing amount of slurry addition, even though soil respiration did not increase. In contrast, increasing inorganic N addition did not increase N_2O emissions significantly. This indicates that not only available N influences N_2O release, but also available C has to be present in sufficient amounts as it is in slurry.

Conclusions

At low inorganic N availability microbes tend to mineralise more organic matter, whereas at high inorganic N availability microbes tend to use more of the available inorganic nitrogen for their metabolism and mineralise less organic matter. This behaviour is also reflected in different gas emissions within the incubation experiment. Furthermore, *in situ* measurements deliver differences in the gas emissions which can be reduced to existing rhizosphere processes in the field.

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NITROGEN MINERALIZATION OF SUGARBEET VINASSES IN INTERACTION WITH CATCH CROPS

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Adequate nitrogen (N) fertilization is essential for sugar beet crop management, because excess N decreases sugar rate and extractable sugar. In France, the N balance sheet method is widely used to forecast N fertilizer requirements of sugar beet crop (Machet et al 2007). The balance is made on the inorganic N pool in the soil rooting zone, during the growth cycle of crops; it enables to determine the amount of N-fertilizer necessary to fit soil N supplies to crop requirements. Soil N supplies take into account the N contribution of organic products and catch crops. In many cases, before establishment of sugar beet crop, various organic products are spread, particularly vinasses and mandatory introduction of catch crops bounded to the regulation. Vinasses are coproducts from sugar refinery. They are used as organic fertilizer for the conventional and biological cultures and meet the criteria of the NF standard U42-001-2. The objective of this study was to estimate the N effect of vinasse in interaction with catch crops, in field conditions on annual experiments. The results allowed to test the decision-making tool AzoFert® in conditions of vinasses application and presence of catch crops in winter.

Materials and Methods

Field experiments were set up each year between 2005 and 2013 in northern France. Soils are deep loamy soils with high water content. The climatic contexts and cultural systems are similar. In all situations, the preceding crop is winter wheat with buried straw. The five different experimental treatments (with four randomized replicates) are: Control treatment without vinasse and with or without a catch crop; Spreading of vinasse in August after winter wheat harvest, followed or not by a catch crop; Spreading of vinasse in spring before sugar beet sowing.

Vinasses are spread at a mean rate of 3.3 t.ha^{-1}. Total N content is 22 kg N ton^{-1} vinasse (equivalent to 73 kg N ha^{-1}). Catch crop is a white mustard, *Sinapsis alba,* sowed just after wheat harvest. Sugar beet is sowed in March. For each treatment, N response curve is set up, with 0 (control), 40, 80, 120 and 160 kg N ha^{-1} as ammonium nitrate. Optimal N rate is determined as the lower rate which allows to reach the statistically highest sugar yield. The changes in soil nitrate and ammonium are followed at different dates (August, November and February) until 120 cm depth (four layers of 30 cm). Catch crops are sampled each year, in November, before their burying in the soil by ploughing. Dry matter and nitrogen (N) and carbon (C) contents are determined on above and below-ground biomasses. In October, sugar beet crop is sampled on control treatments, to determine N and C contents in the tops and roots. Roots yield, sugar content, sugar yield and qualitative criteria on roots as glucose and alpha-amino N are determined after harvesting on every treatment.

Results and Discussion

Residual mineral N in the soil after wheat harvest is on average 34 kg N ha^{-1}. Between August and November, evolution of the mineral N pool is similar in the plots with catch crops, with or without spreading of vinasse. The amount of mineral N decreases to reach a minimal value, on average 17 kg N ha^{-1}, at time the catch crop is destroyed. In the control treatments without catch crop, mineral N pool is higher than in the control treatments with catch crops. During autumn, the extra mineral N due to vinasse addition is on average 11 % of vinasse-N applied with a high variability between the years (0 to 27 %). At the end of winter, mineral amounts increase because N supplies (mineralization of soil OM, wheat residues, catch crop residues and vinasse) are higher than N leaching losses. This increase is more important in treatments with catch crops. In bare soils treatments (without catch crop in autumn), leaching losses are similar with or without vinasse addition, about 14 kg N ha^{-1} and a concentration of water drainage at 90 cm depth of 60 mg N l^{-1}. With a catch crop, N leaching losses reach 3 kg N ha^{-1} and the N concentration in water is calculated with Burns model at 14 mg N l^{-1} (Justes et al 2012).

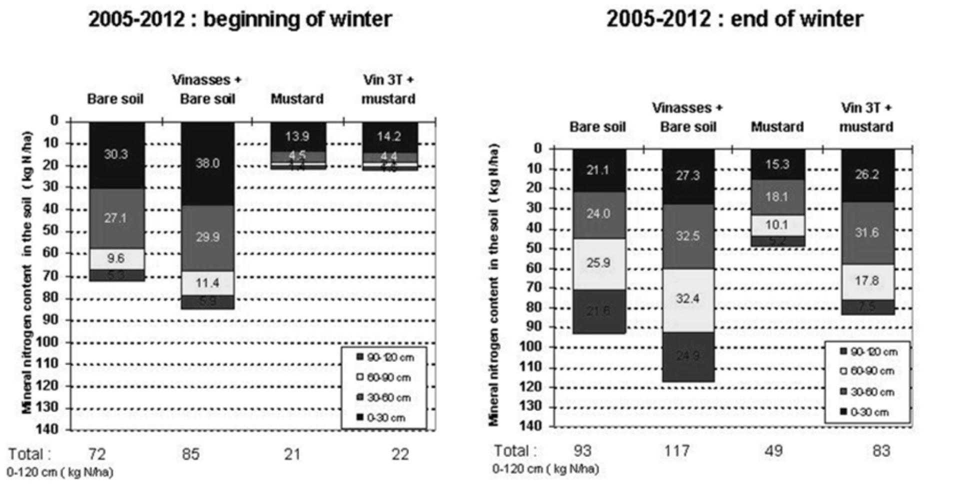

Figure 1 : Evolution of Nitrogen soil contents for different treatments between the beginning and the end of winter

The mean aerial biomass of the catch crops is 1.7 t DM ha^{-1} in treatments without vinasse compared to 2.3 t DM ha-1 with vinasse. Catch crop N is 40 and 59 kg N ha^{-1} without or with vinasse, respectively, representing an apparent recovery of vinasse-N by catch crop of, on average, 20 % added N (3 % to 28 % depending of the year). The apparent recovery of vinasse-N by sugar beet crop, measured on the control treatment (withouth mineral fertilization), reach 56 % added N (SD = 20) and 49 % added N (SD = 30) for August and spring vinasse spreadings, respectively. N response curves show that the N optimal doses measured on the experiments agree with the rate of fertilization proposed by the decision-making tool AzoFert®.

Conclusions

The experiments set up during the period 2005-2012 allow to build up a significant database on N fertilization management of sugar beet crops. It contributes to a better knowledge of the dynamics of N in response to different scenario of vinasse spreading and intercrop management, particularly the establishment of catch crops. These results contribute to improve the relevance of AzoFert® tool for the N fertilization of sugar beet crops.

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EVALUATION OF LABORATORY AND FIELD INCUBATION TO PREDICT NITROGEN FERTILIZATION IN AN ORGANIC HORTICULTURAL ROTATION

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Nitrogen (N) is usually the limiting nutrient in organic agriculture, but some of the problems with N availability may be solved through the judicious use of green manure and composted farmyard manure. Therefore, there is a need to determine the N mineralization of organic materials incorporated into the soil along the crop rotation in order to synchronize the release of N with crop requirements. The laboratory incubations will probably not reflect the rates of mineralization which occur in the field due to soil perturbations (storing, mixing and sieving) and the lack of temperature and soil moisture fluctuations (Lomander et al., 1998). Incubation of soil in buried soil cores are intended to closely resemble the fluctuating environmental conditions in the field. Here, the N mineralization determined by field and laboratory incubation was assessed for fertilizers incorporated into the soil to improve fertilizer recommendations.

Materials and methods

A field crop rotation with a cover crop of rye (S. cereal L.) consociated with hairy vetch (V. sativa L.) over the autumn and winter, followed by potato (Solanum tuberosum L.) and lettuce (Lactuca sativa L.) was set up on a sandy loam soil, as a randomized block design with four replicate plots per treatment. Treatments incorporated before planting potato included: (i) green manure (GM); (ii) and (iii) GM with 20 and 40 t ha-1 of compost (C20 and C40); (iv) and (v) GM with 1 and 2 t ha-1 of a commercial organic fertilizer (OF1 and OF2), and (vi) a reference treatment (T0) without fertilizers. The compost was collected from a pile of cow manure aged for 7 months with an N content of 19.8 g kg-1, and an ammonium and nitrate content of 69 and 134 mg kg-1 respectively. The organic fertilizer was based on poultry feathers, fermented and granulated, and had a total N content of 118.7 g kg-1, and an ammonium and nitrate content of 7272 and 2.1 mg kg-1 respectively. The field incubation was carried out during the potato and lettuce crops (196 days). Five core samples of soil were buried for periods of 14 days in micro-perforated polyethylene bags and other five were frozen. The difference between the amount of inorganic N in frozen and incubated samples was used to calculate N mineralization in each plot. In the laboratory incubation the fertilizers were freeze-dried and grounded using a 2 mm sieve. The amounts of compost, organic fertilizer and cover crop used in the incubation were equivalent to 25, 1.2 and 40 t ha-1 respectively. The soil moisture was adjusted to 60% of soil water–holding capacity and subsamples of 50 g of dry soil equivalent were incubated for 196 days at 25ºC. The evolved CO_2 trapped in a NaOH solution and N mineralization were calculated. Two models of N mineralization were fitted to the results.

Results and discussion

Potato yield was not statistically different between the treatments OF1 and OF2 (36 and 42 t ha-1 respectively) and C40 (36 t ha-1). However, lettuce yield increased significantly ($P< 0.05$) for treatments C20 and C40 (18 and 19 t ha-1 respectively) compared to all other treatments. Lettuce yield increased with compost application compared to the organic fertilizer because the organic N in the organic fertilizer was fast mineralized during the previous potato crop, probably due to its low C/N ratio (3.9) and high content of soluble C and N (42704 and 36585 mg kg-1 respectively), which increased the microbial activity releasing a high amount of mineral N in a short period of time. On the other hand, the application of 40 t ha-1 of compost caused N immobilization for 72 days, probably due to the increase in available C for microbial biomass, promoting the N remineralization during the period of potato as well as lettuce growth. A positive correlation was found between the sum of mineralized N and potato yield ($R2= 0.90$; $P< 0.05$), so field incubation is probably a good indicator to predict fertilization (Yan et al., 2006). In laboratory incubation, the applied organic N mineralized in compost (12 %) was much lower compared to the applied organic N mineralized with 20 and 40 t ha-1 compost in the field incubation (73 and 50% respectively) (Table 1). Probably the increasing soil respiration by incorporation of rye and vetch (4049 mg C-CO2 kg-1) indicating high microbial activity in the field increased N mineralization in compost. The storage conditions of compost before laboratory incubation may also lead to a decrease in N mineralization. Jost et al. (2013) reported that cow faeces microbial characteristics revealed higher impacts on plant N uptake than soil microbial properties, thus the changes of microbiology in frozen samples of fresh compost can decrease N mineralization (Wu et al., 2001).

Conclusions

The proper management of composted farmyard manure combined with rye and vetch can increase organic vegetable crop yields in a horticultural rotation. The field incubation was more effective to predict fertilization according to the potato and lettuce yields than laboratory incubation.

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Table 1. Model equation of mineralized N (mg kg^{-1} DM) and applied organic N mineralized (%) ($Org_{min}N$) in compost and organic fertilizer.

Fertilizers	$*N_0$	$**N_0$	k_1	k_2	Org_{min} N (%)
Field incubation					
* 20 t ha^{-1} Compost	58		0.0001	0.0001	73
* 40 t ha^{-1} Compost	80		-0.01	0.0014	50
* 1 t ha^{-1} Organic fertilizer	57		0.006	0.00005	72
** 2 t ha^{-1} Organic fertilizer		104	0.009	0.6	61
Laboratory incubation					
* Compost	12		0.3	0.001	12
** Organic fertilizer		61	0.057	0.9	61

$*N_m = N_0 [1-\exp(-k_1t-k_2t^2)]$ and $**N_m = N_1 [1-\exp(-k_1t)] + N_2 [1-\exp(-k_2t)]$: N_m represents accumulated mineralized N; k_1 and k_2 are mineralization constant rates; $*N_0$ and $**N_0 - N_1+ N_2$ represents the amount of potentially mineralizable N.

ENVIRONMENTAL IMPACTS OF MAIZE CULTIVATION IN DENMARK AND IN NORTH CHINA PLAIN

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In Denmark silage maize substitutes the labor-intensive forage crops such as grass and grass-clover. The period after harvest is often with bare soil and coincides with high drainflow season, increasing the amount of nitrogen (N) leaching. Catch crop extends the growth season to avoid bare soil during autumn and winter, thus to minimize N leaching. Spring-sown catch crop (intercropping) may establish better than autumn-sown, but may also compromise yield due to competition. In the North China Plain (NCP) no bare soil period exists after maize harvest because winter wheat is sown. This traditional annual crop rotation receives more than 600 kg N ha^{-1} urea fertilizer to ensure high yields. Studies show deteriorating N effect on the soil and water environment in the NCP (Ju et al., 2009). The aim was to use field measurements and model simulation to quantify N losses from maize fields under current and alternate management in Denmark and in the NCP.

Materials and Methods

In Denmark, maize was sown after grass-clover each year from 2009 to 2011 on sandy loam and coarse sand soils. Maize was grown as monocrop or intercropped with red fescue catch crop, at 20, 80 and 140 kg N ha^{-1}(N20, N80 and N140 respectively). In the NCP, maize-winter wheat crop rotation was studied in 2012-2013 on silty loam soil at 0, 200, 400 and 600 kg N ha^{-1} fertilizer rates (N0, N200, N400 and N600). At both sites, soil water content and crop growth were measured throughout growing season; soil NO_3-N was also periodically measured. For the Danish site, daily NO_3-N concentrations were calculated by interpolation between measuring dates according to Lord and Shepherd (1993). These were multiplied with DAISY-simulated daily water percolation to obtain accumulated (annual) N leaching. Similarly, N leaching for NCP site was obtained after calculation of water percolation with the water balance method (Moreno et al., 1996). Field measurements were used to calibrate the DAISY model for soil water, crop growth and N leaching.

Results and Discussion

In Denmark in 2009, N leaching was affected by both catch crop and fertilizer rate and was strongly related to soil type. On sandy loam, intercropped maize resulted in about 23% lower N leaching compared to monocrop (Fig. 1a). N leaching reduction was largest at N20, which had similar yield and N content as N80 and N140, implying that mineralized organic N from the previous grass-clover sufficed for both maize and catch crop. In a similar study in North Germany, Kayser et al. (2008) also found organic N mineralization after grass-clover sufficed for high maize monocrop yields without N fertilizer even for two years, and adding fertilizer N only contributed to N leaching. On coarse sand, N leaching was overall high and the catch crop was able to diminish it to a large extent. There was about 50% reduction in N leaching from intercropped maize compared to monocrop. However, yields were reduced by about 15% (Fig. 1b) due to competition and lack of soil N. Adding 60 kg N ha^{-1} (N140) to the recommended fertilizer rate (N80) diminished yield loss, while the catch crop offset N leaching. The presented offset in N leaching on coarse sand for N140 (Fig. 1a) was in 2009, although in 2010-2011 it was to a lesser degree.

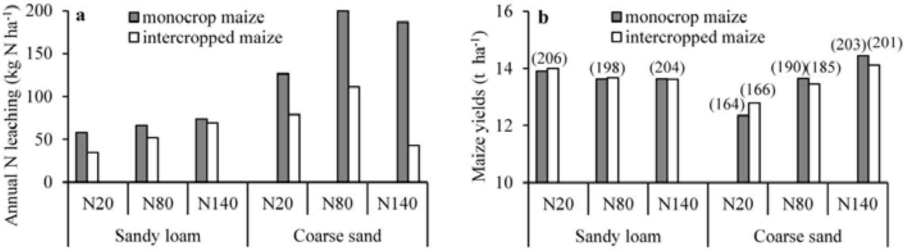

Fig. 1. Field-measured nitrogen leaching (a) and maize yields (b) from different management treatments in Denmark. Annual values refer to April 2009-April 2010.Values in brackets are harvested N (kg N ha-1). N80 is the recommended fertilizer rate (80 kg N ha-1) for maize after grass-clover on sandy soils.

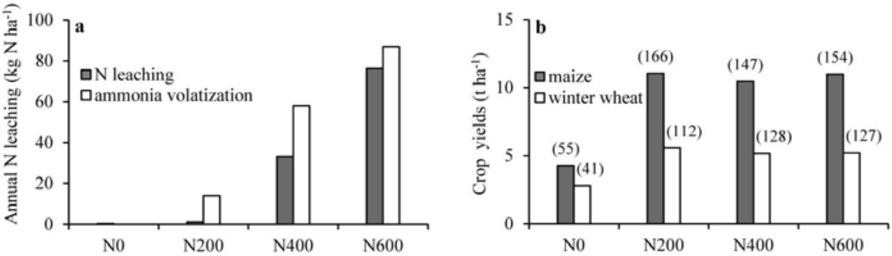

Fig. 2. Field-measured nitrogen leaching and simulated ammonia volatization (a) and measured crop yields (b) from different management treatments in the NCP. Annual values refer to June 2012-June 2013.Values in brackets are harvested N (kg N ha-1). N600 is the annual fertilizer rate (600 kg N ha-1) used by most farmers in the NCP.

In the NCP, N leaching and NH_3 volatization due to use of urea were the main N losses from the system. Both responded dramatically to fertilizer rates (Fig. 2a). Respective N leaching for N0, N200, N400 and N600 treatment was 1, 2, 33 and 76 kg N ha^{-1} and volatization 0, 14, 58 and 87 kg N ha^{-1}. NH_3 volatization was simulated assuming that increased amount of urea increases soil pH and thus leads to higher percentage-wise losses. There were no significant differences for maize and winter wheat yields between N200 to N600 fertilizer rates (Fig. 2b). The N0 treatment showed relatively high yields, implying high organic N mineralization, especially during maize season when both temperatures and precipitation are high. There is also additional N source to the system from atmospheric deposition (100 kg N ha^{-1}) that should be considered (Ju et al 2009). DAISY simulations for the NCP showed that not only the rainy maize season affects N leaching, but also the dry wheat season during which water transports significant amounts of accumulated NO_3-N downwards to the deep soil layers.

Conclusions

Intercropping is a promising measure to reduce N leaching from maize fields in Denmark. On sandy loam previously cropped with grass-clover, lower N leaching was achieved with N20 fertilizer rate without yield decrease. On coarse sand, higher fertilizer rate (N140) was required to compensate for competition, while catch crop offset the N leaching. In the NCP, annual fertilizer rate of 200 kg N ha^{-1} drastically reduced the N losses from the system without significant yield loss, compared to higher rates 400-600 kg N ha^{-1} commonly used. The optimum rate is somewhere between 200 and 400 kg N ha^{-1} and should be investigated in details. Also, long-term effect of decreased N input to the maize-winter wheat system should be studied to assess if yields are compromised.

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INFLUENCE OF NITRIFICATION INHIBITOR DMPP ON YIELD, FRUIT QUALITY AND SPAD VALUES OF STRAWBERRY PLANTS

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3,4-dimethylpyrazole phosphate (DMPP) is a nitrification inhibitor with highly favourable properties. It has undergone thorough toxicology and ecotoxicology tests and application-technology experiments, and has been shown to have several distinct advantages compared to the currently used nitrification inhibitors. DMPP has high efficiency in inhibiting soil nitrification and increasing N fertilizer efficiency, crop yield and fruit quality and it has also no toxicological effects. The aim of this research was to evaluate the effect of 3,4-dimethylpyrazole phosphate (DMPP) on yield, fruit quality and SPAD values in young leaves of strawberry plants in soil growing system.

Materials and methods

The experiment was carried out in Huelva (Spain) in a greenhouse from October 2012 to June 2013 on one strawberry (*Fragaria* x *ananassa* Duch.) cultivar 'Candonga'. A completely randomized block design (4 treatments x 1 cultivar) with 3 replications was established. The treatment were: T1: 100% NOVATEC® Solub 21 (21% N-NH_4^+, 0.8 % DMPP, 60% SO_3), T2: 75% NOVATEC® Solub 21 (21% N-NH_4^+, 0.8 % DMPP, 60% SO_3) + 25% Ammonium Nitrate 34% N-(NH_4NO_3), T3: 25% NOVATEC® Solub 21 (21% N-NH_4^+, 0.8 % DMPP, 60% SO_3) + 75% Ammonium Nitrate 34% N-(NH_4NO_3), T4: 100% Ammonium Nitrate 34% N-(NH_4NO_3). Ripe fruits from each treatment were harvested through the experimental period (early and late crop cycle). The total marketable fruit yield (sum of early and late production) from 1st of January to the 31st of May in g per pots (10 plants per plot) was recorded weekly during the harvest. The early marketable fruit yield was recorded from 1st of January to 31st of March in g per pots. The main effects of DMPP on yield (fruit weight), fruit quality and SPAD values in young leaves of strawberry plants were evaluated.

Results and Discussion

The highest values of fruit weight (20.15 g fruit^{-1}) and (19.56 g fruit^{-1}) were observed in treatment T2 and T1 respectively (Table 1). There was fruit weight increment resulted from treatment with DMPP. Several studies have demonstrated that DMPP increases crop yield in other crops such as barley, maize and wheat (Linzmeier et al., 2001; Pasda et al., 2001). During early crop cycle significant differences in fruit size between treatments were recorded. The highest values of fruit size (29.51 mm) and (29.71 mm) were observed in treatment T1 and T2 respectively (Table 1). Fruit weight increment resulted from treatment with DMPP might be due to the fruit size increment. There were significant differences between treatments (T1, T2) and T3 and T4 (control). The values of fruit size in late crop cycle were higher than in early crop cycle. However, there were not significant differences between treatments (Table 1). Respect to TSS and TA registered during the early and late crop cycle there were not significant differences between treatments (Table 1). The values of TSS and TA in late crop cycle were higher than in early crop cycle. The highest values of TA (3.23 %) were observed in treatment T1 (Table 1). The lowest values of firmness were registered in treatment T1 (282.42 g cm^{-2}), T2 (295.51 g cm^{-2}) and T3 (292.86 g cm^{-2})

respectively (Table 1). Respect to AsA and during late crop cycle there were not significant differences between treatments. The highest value (58.68 mg AsA 100 g^{-1}) was observed in treatment T1. AsA increment resulted from treatment with DMPP might be due to the fruit size increment. AsA increment could be influenced by DMPP. The lowest values of AsA were registered at the end of late crop cycle (week 26) (45.02 mg AsA 100 g^{-1}). AsA contents of strawberry fruit can be variable among cultivars. SPAD values followed different tendencies during the total crop cycle. There was a change in SPAD values with time. An increase was observed from week 13 to week 19 after planting and there was a decrease in SPAD values from week 22 to week 27 due to the end of crop cycle (data not shown). Results suggested that 'Candonga' cultivar yield was affected by treatment and crop cycle (weeks). There were significant differences between T1 treatment and others treatments. SPAD values were higher in tretament T1 (56.31) (Figure 1). The highest values were registered in weeks 13, 15, 19 and 22 (early crop cycle) (data not shown). In 'Candonga', it seems there is some beneficial effect of treatment with DMPP during early crop cycle on the increment of SPAD, in particular after week 14. In this same, more intensive green colour of leaves as a result of enhanced chlorophyll content was observed with DMPP. During early crop cycle significant differences in fruit size between treatments were recorded. Therefore, fruit size increment resulted from treatment with DMPP might be due to the SPAD values increment.

Conclusions

Cultivar type is the most important factor that is involved on the determination of the post-harvest quality and shelf-life. Therefore, more research is required with different strawberry cultivars in order to confirm the results obtained so far. In early crop cycle fruit weight increment resulted from treatment with DMPP might be due to the fruit size and SPAD values increment. In Candonga, it seems there is some beneficial effect of treatment with DMPP during early crop cycle. In this same, the possible effects on fruit quality of DMPP that have not been evaluated need to be elucidated. In conclusion, the study of fruit size, firmness and SPAD values recorded during early and late crop cycle revealed effect of treatment in strawberry soil growing system.

Table 1. Fruit weight and fruit quality recorded during crop cycle.

Treatments	Fruit weight² (g $fruit^{-1}$)		Fruit Size³ (mm)		Total soluble solids (mg kg^{-1})		Titratable acidity (TA)		Firmness (g cm^{-2})		Vit C (mg AsA 100 g^{-1})
	Early production	Late production	Early production	Late production	Early production	Late production	Early production	Late production	Early production	Late production	Late production
T1	19.56 ± 4.28 a	21.64 ± 4.62 a	29.51 ± 4.75 a	31.65 ± 3.19 a	6.02 ± 0.57 a	7.34 ± 1.59 a	2.74 ± 0.41 a	3.23 ± 0.69 a	282.42 ± 53.56 b	231.08 ± 45.19 b	58.68 ± 10.53 a
T2	20.15 ± 3.96 a	21.39 ± 4.62 a	29.71 ± 3.45 a	30.79 ± 3.18 a	6.32 ± 0.72 a	7.24 ± 1.54 a	2.76 ± 0.40 a	2.85 ± 0.56 a	295.51 ± 31.47 b	233.28 ± 34.10 ab	53.98 ± 7.19 a
T3	18.46 ± 3.04 a	22.25 ± 5.11 a	26.55 ± 2.22 b	31.81 ± 3.02 a	6.53 ± 1.19 a	7.46 ± 1.46 a	2.95 ± 0.71 a	3.10 ± 0.81 a	292.86 ± 28.94 b	253.75 ± 34.32 ab	56.03 ± 12.05 a
T4 (Control)	18.31 ± 3.45 a	22.05 ± 4.68 a	26.37 ± 2.10 b	31.77 ± 4.15 a	6.12 ± 1.22 a	6.95 ± 1.28 a	2.72 ± 0.29 a	3.11 ± 0.62 a	310.03 ± 30.60 a	257.56 ± 33.53 a	53.83 ± 13.93 a
Significance	NS	NS	*	NS	NS	NS	NS	NS	*	*	NS

NS: effect of treatment not significant. Values followed by the same letter are not statistically different according to Duncan test, at 0.05.

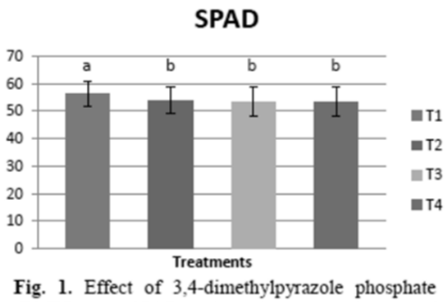

Fig. 1. Effect of 3,4-dimethylpyrazole phosphate (DMPP) on SPAD values in young leaves of strawberry plants in soil growing system. Values are mean ± SD.

Acknowledgement

Supported by the agreement by COMPO EXPERT Spain S.L. "Effect of nitrification inhibitor (DMPP) on productivity and quality of strawberry plant". Pedro Díaz for technical assistance.

ARE REDUCTION OF NITROGEN POLLUTION AND QUALITY REQUIREMENTS IN AGRO-FOOD CHAIN COMPATIBLE?

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Hard wheat (BAF in the French classification) is a key crop for bread wheat chain because its good technological qualities partially correlated to its high grain protein content (GPC). GPC is the main contractual criteria for quality grading and results from a tradeoff between carbon and nitrogen nutrition. Grain Yield (GY) is known to be negatively correlated with GPC among genotypes although nitrogen and water supply strongly influence this relationship (Oury et al., 2003). In the center of France, pedoclimatic conditions allow growers to reach high GPC. However to maintain production in this territory, grain collecting firms (GCF) have promoted more intensive cultural practices, especially higher fertilization rates, leading to high nitrate levels in ground waters and in water catchments. We analyzed the management practices of the BAF supply chain from growers to the industries in order to identify the main challenges for N reductions in cropping systems and leverage points in the wheat chain.

Materials and Methods

Surveys were carried out with various stakeholders in the BAF supply areas of 3 GCF (fig1) in order to analyze (i) determinants of quality criteria and the supply management strategy for GCF, (ii) the management of wheat production in 69 farms.

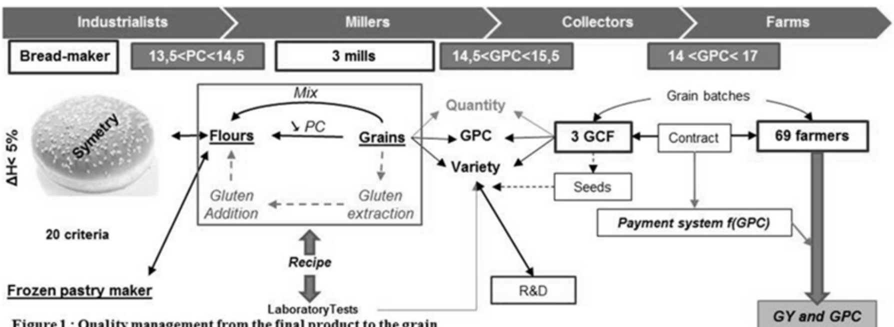

Figure 1 : Quality management from the final product to the grain
surveys; → common characteristics; - -> according to the structure; GPC: Grain Protein Content; PC: Protein Content; GY: Grain Yield; GCF: Grain Collector Firms

Results and discussion

BAF grown in these supply areas is mainly for industrial use (frozen pastry-making, bread-making for typical special bun or sandwich breads). Standard making-processes involve very strict requirements so as to have both homogeneous quantity and quality. To supply flour of standard and constant quality, millers impose specifications to GCF with respect to the variety and GPC. GCF implement coordination tools in order to respond to market demands while fulfilling both agricultural and industrial capacities.

These tools include both the terms of commitments and the instruments of technical and commercial management. The payment system considers both GY and GPC. In this area, soils are heterogeneous and useful water reserves vary from 60 mm to 210 mm. Therefore drought stress can begin during May for the lowest water reserves whereas wheat flowering generally occurs at the end of the month: this leads to high GPC but low GY (5-6 t/ha). However, to meet bread chain requirements, growers try to reach both high GPC (14 – 17%) and high GY (6.5-8t/ha). To reach such yields, BAF is grown either on deep soil or on low water reserve soils with irrigation during drought conditions. To compensate the dilution of proteins resulting from GY increase, growers use a lot of inputs, notably nitrogen fertilizers: 220 to 260 kg/ha. The level of N fertilization is calculated by the balance method: total N requirement is the product of the target yield by N need per unit yield (b). b depends on the variety and is correlated with its potential GY, it varies between 2.8 and 3.2. However, for BAF, a higher b value is used (bq) to reach the GPC targets (3.9-4.1 for Courtot and Galibier). This leads to use more nitrogen than needed to achieve the target yield and can potentially result in a strong increase of soil N-NO3 content at harvest (Comifer, 2013). Current regulations are implemented under Nitrate Directive and aim to modify and control growers' practices, but our results show that these current regulations do not achieve their objectives because of the bq value. Four leverage points could be considered but face a lot of resistance in the wheat chain. Firstly, reducing yield to reach GPC with less N is difficult because it would lead to a decrease in growers' income and a reduction of the quantity of grain available to collectors. Secondly, reducing quality targets has to be examined: (i) as GPC are often higher than the 14.5% target, could GCF modify their bonus for the highest GPC? (ii) Could industries further adapt their process to lower and more variable protein content? The third leverage point is upstream in the wheat chain. As BAF is a small market only a few varieties are available and often old. Some work has already shown that bread making qualities depend not only on GPC but also on gluten composition. New varieties with lower GPC but good qualities are now available but are not well known by the stakeholders of the supply chain. Finally, the last leverage point could be in the way grain quality is tested for: GPC is easy to measure at all the chain stages but is far to be enough to predict bread making qualities as shown by the multiple other controls at the mill and fabrication stages. Finding quick but more predictive tests appears as a major issue.

Conclusion

To address environmental issues, the management tools currently used are mostly technical, aiming at a modification of farmer practices on a given crop and have showed their limits. Alternative agroecological practices exist but can lead to less homogeneous products in quantity and quality. The determinants of cultural practices overpass the farm scale and also depend of the whole supply chain management. These determinants construct a technological regime and a lock-in situation that hinders the development of agroecological innovations. Breaking out of these lock-in situations requires acting simultaneously on different leverage point as shown for other supply chains (Meynard et al., 2013).

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IMPACT OF FERTILIZATION HISTORY ON RED CLOVER BELOW GROUND NITROGEN INPUTS IN A CLOVER-GRASS LEY AND N-TRANSFER TO GRASS

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Clover-grass leys provide essential input of external nitrogen (N) by biological di-nitrogen fixation to crop rotations in low input agro-ecosystems as organic farming. While above ground plant parts are generally removed as fodder and only a proportion of its N will return by manures, N in stubble and below ground N (BGN) provide the major benefit to subsequent non-legumes. BGN is defined as root N plus N derived from rhizodeposition (NdfR), the latter comprising the release of all kinds of N compounds from living plant roots and its turnover (Wichern et al. 2008) and therefore can partly be taken up by associated grass in clover-grass mixtures (Oberson et al. 2013). The objectives of our study were i) to determine the BGN inputs by red clover including NdfR and the N transfer to associated grasses and ii) to evaluate the effect of fertilization history in a long term field experiment using ^{15}N labelling methods. The effects of agricultural management on the ratio of BGN to total N (Nt) and determining factors will be presented.

Material and methods

We studied the BGN inputs by red clover *(Trifolium pratense* L.*)* and the transfer of clover N to the associated ryegrass *(Lolium perenne* L.*)* in a two component clover-grass mixture using ^{15}N leaf labelling methods during 2011 and 2012. The red clover-ryegrass mixture was established in micro-plots (PE tubes with 39 cm diameter, 0-25 cm soil depth), situated in the clover-grass ley of the DOK long term field experiment (Basle, Switzerland) comparing organic and conventional cropping systems since 1978 (DOK experiment: bio-Dynamic, bio-Organic, Konventionell) (Mäder et al. 2002). Four DOK treatments were selected: Control with zero fertilization (NON), bio-organic with regular (ORG2) and half (ORG1) dose of manure fertilization and conventional with regular mineral fertilization (MIN2). To determine BGN inputs 11 clover plants were leaf labelled with ^{15}N enriched urea after each cut. In October 2011and 2012 the soils of the micro-plots were excavated, physically recoverable roots were removed by hand and separated into clover and grass roots. The plant material and the remaining soil were analysed for N content and ^{15}N. The amount of NdfR as well as the transfer to associated grass was calculated according to Jansen and Bruinsma (1989) assuming that the NdfR had the same isotopic N composition as the roots. The proportion of clover N derived from symbiotic di-nitrogen fixation (%Ndfa) was determined using the ^{15}N natural abundance approach.

Results and discussion

Over the whole growing period the total amount of assimilated N of the clover-grass ley increased from the unfertilized treatment NON to MIN2 and ORG1 to ORG2 ranging from 21 to 48 g m^{-2} after 3 months, 30 to 67 g m^{-2} after 6 months and 83 to 164 g m^{-2}

after 18 months of clover-grass ley cultivation (Table 1 a-c). Clover N contributed predominantly to N_t throughout the whole growing period. The relative contribution was lowest in MIN2 and largest in ORG2 and ORG1. The amount of BGN (g m^{-2}) was lowest in NON and highest in ORG2 during the whole ley, whereas the proportion of BGN in % of N_t was highest in NON after 3 months, nearly 50%, and tended to be higher after 6 and 18 months. The highest average proportion was found at the beginning of the clover-grass ley decreasing to a minimum after 6 months. After 18 months BGN constituted between 24% and 30% of N_t. However, that was only 50% to 60% of the proportion found by Hoegh-Jensen et al. (2001). Clover N contributed distinctly to the N uptake of associated grass. The benefit for grass was lowest in the beginning and constituted 13% to 41% of grass N after 3 months. Significant differences were found between ORG2 with a maximum of 41% and NON and ORG1with a minimum of 13% and 18% respectively. Towards the end of the growing period the proportion of N derived from clover (%Ndfc) increased to a level of 41% to 51%, but differences between treatments decreased. Oberson et al. (2013) found similar %Ndfc after 2 years in the DOK clover-grass ley, but not at the beginning where we found relevant %Ndfc after 3 months. At the beginning of the clover-grass ley the %Ndfa showed in tendency a differentiation with lower %Ndfa for MIN2 and NON and higher %Ndfa for ORG2 and ORG1l, whereas significant differences were found only between MIN2 and ORG1. After 6 months the differences were almost balanced and increased after 18 months to more than 95% in all treatments.

Conclusions

BGN inputs constituted one fourth to one third of clover-grass ley-N after two vegetation periods. The absolute BGN inputs doubled from the beginning to the end of the growing period. Malnourished plants invested absolutely less N below ground but relatively more. Corresponding to increasing BGN inputs, associated grass benefited from clover N over time and received up to 50% of its N from clover. Although the percentage of clover was lowest in the mineral fertilised treatment the proportion of clover N found in grass was comparable to the other treatments, indicating that clover plants in this treatment transfer absolutely more N to the associated grass.

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Duration clover-grass ley	treatment	N_t [g m^{-2}]	BGN [g m^{-2}]	BGN in % N_t	Ratio clover N : grass N	%Ndfa	% Ndfc
a) 3 months (soil sampling after 2. cut 2011)	NON	21 c	10 b	49% a	6.1 bc	75% ab	13% b
	MIN2	37 b	14 ab	37% ab	3.2 c	69% b	24% ab
	ORG1	35 b	12 ab	34% b	8.5 ab	98% a	18% b
	ORG2	48 a	19 a	39% ab	10.5 a	92% ab	41% a
b) 6 months (soil sampling after 4. cut 2011)	NON	30 c	7 b	22% n.s.	7.0 b	88% n.s.	36% n.s.
	MIN2	50 b	11 ab	21% n.s.	4.3 b	83% n.s.	45% n.s.
	ORG1	53 b	8 ab	16% n.s.	11.1 a	97% n.s.	37% n.s.
	ORG2	67 a	11 a	17% n.s.	11.7 a	94% n.s.	48% n.s.
c) 18 months (soil sampling after 5. cut 2012)	NON	83 c	26 b	30% n.s.	9.2 a	98% n.s.	41% n.s.
	MIN2	151 ab	37 a	25% n.s.	5.0 b	97% n.s.	51% n.s.
	ORG1	143 b	37 a	26% n.s.	7.9 a	95% n.s.	44% n.s.
	ORG2	164 a	40 a	24% n.s.	7.9 a	96% n.s.	50% n.s.

Means not connected by same letter are significantly different (Least significant difference, $p < 0.05$)

Table 1: Clover-grass ley above- + below ground N (Nt) and below ground N (BGN), proportion of BGN of Nt (BGN in % Nt), proportion of N derived from symbiotic N_2 fixation (Ndfa) and proportion of grass N derived from clover (Ndfc) after a) 3 months, b) 6 months and c) 18 months of clover-grass ley

GRASS-FUNGAL ENDOPHYTE SYMBIOSES EFFECTS ON NITROGEN FIXATION AND DYNAMICS IN A KENTUCKY PASTURE

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Tall fescue, an important forage species of pastures in the eastern U.S., can associate with an endophytic fungus, *Neotyphodium coenophialum*. However, endophyte presence in tall fescue can negatively impact animal production through production of toxic alkaloids; therefore, non-toxic 'novel' strains of the endophyte have been identified and are being adopted by forage producers. Because fungal endophyte presence and strain have been shown to alter plant community dynamics in tall fescue dominated grasslands, it is important to also investigate whether these fungal endophyte associations influence belowground processes such as nutrient cycling. The objective of this study was to assess the impact of grass-endophyte symbiosis on nitrogen fixation and dynamics utilizing ^{15}N natural abundance techniques.

Materials and Methods

The study site, established in 2008, consisted of tall fescue that was either (1) infected with the common toxic strain of the endophyte (CTE), (2) infected with the non-toxic AR 542 or (3) AR 584 strains, (4) not infected with *Neotyphodium* (endophyte-free), or (5) contained an equal mixture of treatments 1—4. To assess the effect of endophyte presence and strain on the amount of nitrogen derived from biological fixation via legume symbiosis and nitrogen use in co-occurring tall fescue, $\delta^{15}N$ natural abundance was measured in red clover (RC), tall fescue associated with red clover (TF+RC), and tall fescue not associated with clover (TF-RC) collected from plots of each of the tall fescue – endophyte treatments.

Results and Discussion

$\delta^{15}N$ in RC samples, while unaffected by tall fescue endophyte status, were significantly depleted ($\delta^{15}N$= -1.56, on average), suggesting reliance on N-fixation. However, the %N in red clover samples was affected by endophyte treatment, with CTE+ tall fescue plots containing significantly lower values than E-, AR542, or mixed plots. In TF samples, $\delta^{15}N$ was significantly affected by endophyte treatment. For TF(+RC), tall fescue infected with AR 542 was significantly more enriched with ^{15}N than either AR584 or CTE. However, for TF(-RC), E- samples were significantly more ^{15}N-depleted than other treatments. In addition, $\delta^{15}N$ natural abundance was measured in bulk soil samples from each plot over a period of 3 years in order to gain insight into long-term changes in N-cycling in this system. No significant endophyte effect on was found on ^{15}N of bulk soil samples from each plot, though the entire soil system became increasingly ^{15}N-depleted over time, which indicates that the site is retaining ^{15}N -depleted forms of N, presumably products of N-fixation.

Conclusions

The results of this study indicate that endophyte infection and strain have differing effects on nitrogen dynamics in both tall fescue and neighboring red clover. Deployment of novel endophyte associations in pastures should take into consideration grass-fungal endophyte effects on belowground processes and nutrient

NITROGEN DINAMICS IN SOILS: THE EFFECT OF DIFFERENT PRACTICES AND THE ROLE OF MICROBIOTA ON N OXIDATION AND REDUCTION

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The development of intensive farming-systems contributes to the degradation of the environment, particularly with respect to water quality (Oheler at al., 2006). Incorporation of animal manures in soil may impact NO_3 leaching and N_2O emissions. Upon decomposition, these organic materials are fragmented and processed by the soil fauna and microbial communities to produce more or less stabilized N and C molecules and dissolved compounds (Tatti et al., 2013). Alluvial ground-water is particularly vulnerable to nitrate (NO_{3-}) leaching due to nitrogen (N) losses from agricultural soils. However, the relationship between groundwater NO_{3-} concentration and N sources used at the soil surface is complex. The microbial process of denitrification results in the permanent removal of nitrate through the conversion of nitrate to nitrous oxide (N_2O) or dinitrogen (N_2) gases, which can then diffuse out of the ecosystem. Denitrification requires organic carbon and nitrate, low dissolved oxygen concentrations and the presence of denitrifying bacteria (Groffman, 1994). These conditions are likely to occur in agricultural soils. The activity of microorganisms is regulated by many factors, such as soil pH, organic matter content, and the form and availability of N sources (Cela and Sumner, 2002). Agricultural management practices can strongly influence the size, composition, and activity of the microbial communities in soils. Agricultural practices significantly alter ammonia-oxidizing bacteria (AOB) populations, which subsequently impact nitrification and production rates of nitrous oxide (N2O) (Kong et. Al 2010). Cropping systems that receive high organic matter inputs have been characterized by greater microbial activity as well as greater N mineralization compared to systems receiving only mineral fertilizer-N (Kramer et al., 2002). The aim of the project is to better understand where and in which conditions denitrification processes occur.

Materials and Methods

Between May-June 2013, soil cores at 4 depths (0-15; 25-35; 45-55; 65-75 cm) were collected at 5 different sites in Po Valley, in maize-based cropping systems: Landriano (LA, province of Lodi), Bigarello (MNA and MNB, province of Mantova), Calvenzano (CA, province of Bergamo), and Pizzighettone (PIZD and PIZ, province of Cremona). The soils of these sites were respectively *cambisols, calcisols, luvisols, cambisols* according to the WRB soil classification and the annual rainfall ranges from 635 to 1075 mm. Organic N fertilization had an annual mean amount respectively of 154, 224, 0, 621, 250, 320 ha^{-1} year and mineral N mean annual amount was 83, 0, 240, 0, 110, 138 ha^{-1} year. The DNA extracted has been utilised for 5 reactions of real time PCR. The targets (amoA bacterial, amoA archea, nirK, nifH e nosZ), the protocols of real time were applied on the samples following an automatic procedure, using the Tecan Freedom Evo 100 robot. The reactions of real time PCR have been processed using grids of 384 wells on the real time 7900HT Applied Biosystem.

Results and Discussion

All the soils sites were investigate in terms of dry matter (DM), nitrate nitrogen (N-NO_3), ammonia nitrogen (N-NH_4), total nitrogen (Ntot) and phosphorus (P_2O_5) content. Table 1 reports the mean values for each site. The concentration of nitrate in the soils related to the depth of sampling is shown in Figure 2. It is possible to notice that for all sites, except for PIZ, with the deepening of the soil, nitrate concentrations decrease significantly. Ammonium oxidation was present in the superficial layer (0-45 cm) rather than in deep (55-65 cm). No distinction was evident for archea and eubacteria with some site-dependent exceptions. The nif genes are genes encoding enzymes involved in the fixation of atmospheric nitrogen into a form of nitrogen available to living organisms. Analysis of the presence of nifH indicated, as average, the absence or very low presence of N-fixation at highest depth (65 cm). On the contrary and according to soil type and the fertilizers used, highest activity varied between 5-15 to 15-55 cm of depth. Nitrite reductase genes (nirK) was detected in all soil profiles but it was more present in the superficial layers (5-45 cm) and much less at highest dephts.

Site	DM %	N-NO_3 mg kg^{-1} DM	N-NH_4 mg kg^{-1} DM	Ntot g kg^{-1} DM	P_2O_5 g kg^{-1} DM
CA	84.29	6.093	4.069	1.063	38.497
LA	83.56	6.674	7.818	1.143	67.748
MNA	79.51	5.141	0.952	0.940	5.488
MNB	79.33	6.992	1.292	0.508	4.790
PIZ	78.24	16.135	2.577	2.012	82.016
PIZ_D	82.95	7.365	1.566	0.973	44.196

Table 1 – Mean values of dry matter, nitrate nitrogen, ammonia nitrogen, total nitrogen and phosphorus in the

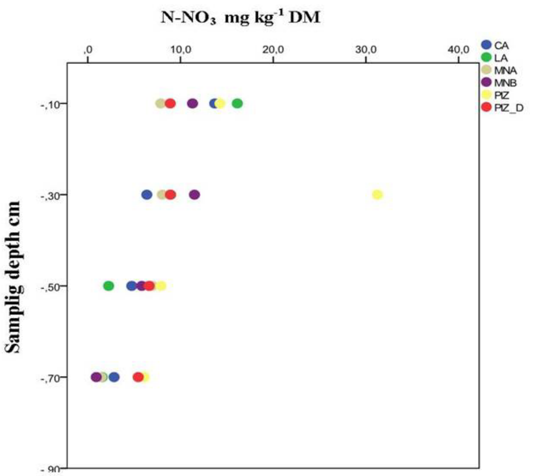

Figure 1 – N-NO_3 soil content mg kg^{-1} DM at different sampling depth (cm) in the different sites.

At the last nitrous oxide reductase (nosZ) gene followed the nirK genes being more present in higher soil layer 5-35 cm and absent or much less represented in deep soil layer (65-75 cm) (questa non l'ho capita). Nitrate content depended by total N content ($r = 0.84$; $p < 0.01$) and less by ammonia content. In this context as average, genes Nir and Nos were detected more frequently when the presence of nitrate was higher within each soil profile for Nos and with a very good correlation with Nir genes ($r = 0.65$, $p < 0.05$). This preliminary test (elaboration of data is in progress) seems to indicate that N both N oxidation and N reduction were present in all soil studied and differently fertilized. The highest activity was detected in superficial and medium depth and generally was absent or very low in deepest soil horizon. Microbial activity seems connected with the presence of nutrient and organic matter.

Conclusions

This work sought to relate the abundances of AOB, denitrifiers, and total bacterial communities within different soil microenvironments regimes. We found that AOB and denitrifier community abundance did relate to variable NO_3 concentrations in soils.

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REDUCING SOIL N TRANSFORMATION OF UREA BY A BIOLOGICAL INHIBITORS OF UREASE ACTIVITY

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Nitrogen (N) transformation is essential to natural and agricultural ecosystems, but the loss of this nutrient causes deleterious effects on the environment to both water reservoirs and atmosphere via volatilization of nitrogenous gases. N use efficiency in agriculture is only 30 to 50%. N mineralization makes N available for plants by converting organic N to inorganic forms through soil organisms and catalyzing the urea hydrolysis to ammonium by enzymes. One of the most important enzymes is urease, which can be associated to cells of microorganisms or immobilized in clays (Lai and Tabatabai, 1992). Inhibition of the urease activity decreases the rate of N mineralization, especially when N fertilization is based on urea or organic materials. Urease inhibitors, which are chemical compounds as heavy metals, thiols and phosphoramide, can be used to reduce N mineralization (Upadhyay, 2012). Another alternative is the use of organic compounds produced from plants that inhibit N mineralization, such as Nimin ®, extracted from Neem tree, which retards urea hydrolysis; or root exudates of Brachiaria humidicola (Rendle) Schweick, which inhibits nitrifying bacteria reducing the nitrification (Gopalakrishnan et al. 2009). The aim of this study is to evaluate the effect of natural inhibitors obtained from plants on urease activity, as a mechanism to reduce N losses in agricultural systems.

Materials and Methods

Vegetal material was obtained from the Mediterranean zone of Central-South of Chile, collecting samples of leaves and bark of Cryptocarya alba (Mol.) Looser and Eucalyptus globulus (Labill). The material was washed, oven-dryied at 60°C and reduced to particles smaller than 2 mm in an electric grinder (Moulinex, Spain). Plant extracts were obtained by boiling the plant material with water as solvent (plant material: solvent in a 1:10 ratio) for 45 minutes. The solution was then filtered and kept in dark at 4°C. Total solids were determined gravimetrically, using total solid contained in each extract to calculate the doses of application.

Urease activity in a model system. A 90 µL urease solution (1,000 IU, Sigma Aldrich) and 8 mg total solids of each plant extract were mixed with phosphate buffer and EDTA, and incubated at 10°C for 5 h. A 20 µL aliquot was obtained every 1 h adding urea (150 mM), phenol nitroprusside solution and sodium hydroxide solution with sodium hypochlorite. Samples were placed in a temperature-controlled bath at 50°C for 3 minutes, then cooled in an ice bath and measured in a spectrophotometer ultra violet/visible at 625 ηm against blank reagent. Tannic acid (8 mg) was used, determining the urease inhibition in terms of percentage (%) respect to tannic acid..

Urease activity in soil. Measurements were conducted using an Alfisol soil (Haploxeralfs) with applications of plant extracts (doses equivalent to 9.5 kg ha^{-1}). Soil was fertilized with urea in solution (equivalent to 150 kg ha^{-1}) and urease activity was determined after 3 days of incubation (at 22°C and 60% water filled pore space, WFPS). The colorimetric determination of ammonium produced was determined by UV/Vis spectrophotometer at 636 ηm..

Statistical analysis. distribution was verified performing a modified Shapiro-Wilks test. Significant effects at $p \leq 0.05$ were evaluated using Tukey test ($p \leq 0.05$).

Results and discussion.

The results of urease activity in a model system showed differences in the ability to inhibit the enzyme activity of treatments. Leaves from *E. globulus* and *C. alba* had low effect on this enzyme. However, the plant extract from bark of both plant species showed an inhibition of 69% and 65% respectively, higher than the inhibition obtained from tannic acid (control). Plant extracts produced a decrease in soil urease activity in an average of 46% respect to control soil (Fig. 1; $p \leq 0.05$). Both assays indicate that these plant species could inhibit N mineralization. The mechanism of the inhibition is attributed to phenols compounds present in the plant extracts. A previous analysis (results not published) and diverse authors (Kiran and Patra 2003; Mohanty *et al*. 2008) indicate that compounds such as tannin and phenol derivatives reduce the nitrification in the soil.

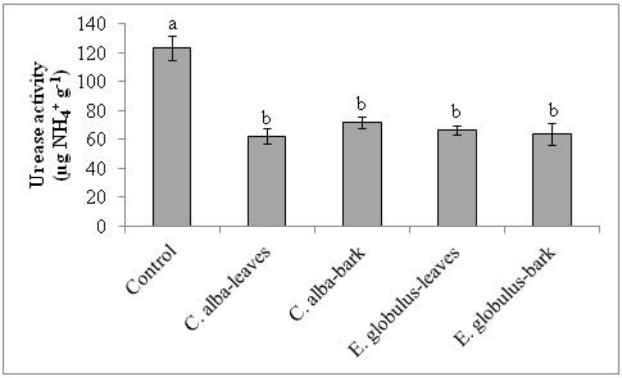

Fig. 1. Urease activity in an Alfisol with applications of plant extracts

Conclusion

Bark of *C. alba* and *E. globulus* are promissory alternatives to decrease the rapid N mineralization from urea fertilizer in agricultural soils.

Acknowledgements

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TRANSFER PROCESSES OF C AND N FROM PEAS TO CEREALS

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Living plant roots continuously release organic and inorganic compounds to the surrounding soil. This process is defined as rhizodeposition, consisting of root exudates, mucilage, sloughed cells and tissue, cell lysates and root debris (Uren et al. 2001; Wichern et al. 2008) and is a major source of C and N inputs into the soil (Jones et al., 2009; Wichern et al., 2008). Additionally, N rhizodeposition of legumes is expected to contribute to the nutrition of cereals in intercropping systems. However, the importance of mycorrhizal connections and direct root contact is not well understood. Thus the aim of this project was to estimate the temporal distribution of N rhizodeposition of peas (*Pisum sativum* L.) and the quantitative contribution of mycorrhiza and root contact to the N transfer to the intercropping partner.

Materials and Methods

To examine the impact of different transfer pathways (root contact, mycorrhiza, diffusion), a pot experiment was set up under controlled conditions to quantify the transfer of the rhizodeposits into different soil compartments. In this pot trial two pea varieties were used as donor plants: *Pisum sativum* L. cv. Frisson, which is able to establish the symbiotic association with arbuscular mycorrhizal fungi and its isogenetic mutant P2, which is not capable of the symbiotic association (Duc et al., 1989). In every pot there were one pea und two cereal plants. As intercropping cereal and receiver plant Triticale (×Triticosecale Wittm. ex. A Camus) variety ‚Benetto' was used. Plants were labeled with a solution of ^{15}N urea (0.5% solution, 95 atom%) and ^{13}C glucose (2% solution, 99 atom%) using the wick method (Russell and Fillery, 1996; Wichern et al., 2007). Labeling was conducted four times with 1 ml applied every two weeks and began in the three-leaf-state (EC 13). Plants were harvested at full ripeness (EC 89). To determine the different transfer pathways, root separations were installed in some of the pots. Without the root separation (W), transfer of C and N is possible by root contact, mycorrhiza and diffusion. With a single walled root separation (S) by a mesh-size of 30 µm root contact is prohibited. The third treatment, a double-walled root separation (D), avoided diffusion as well but allowed mycorrhizal transfer.

Results and Discussion

It was found that mycorrhization of the pea variety Frisson was around 18 %, whereas that of P2 was less than 1% (Fig. 1). As expected, the above-ground biomass was higher in Frisson than in P2 and decreased for the cereal in the order W>S>D (Table 1). The same pattern was observed for total plant N (Table 1). Moreover, the enrichment of the triticale roots with ^{15}N [atom%excess] decreased with increasing root separation W>S>D, indicating reduced transfer from the pea plant to the cereal through mycorrhizal hyphae alone (Table 2). However, ^{15}N transfer from pea to triticale was substantially lower in P2 compared to Frisson, indicating that mycorrhiza hyphael networks, which were present in Frisson but not in P2, might be an important N pathway between intercropping partners (Table 2). One reason for this contradicting result might be a more intensive mycorrhizal connection between peas and triticale without a barrier, compared to the double-walled separation, where mycorrhizal contact area between peas and triticale will be lower. This is also indicated by the relative distribution of N [%] in P2, which showed an increase of N in rhizodeposition [% of total N] with increasing root separation (Fig. 2, Fig. 3). As a consequence, more of the N rhizodeposits has been transferred to the triticale plant in association with Frisson, when only mycorrhiza (D) was established compared to the same treatment (D) with the P2 mutant. However, this phenomenon can also be explained by the lower total amount of plant N with a relative stronger transfer below-ground.

Conclusion

Transfer of N from legumes to cereals follows different pathways, such as root contact, diffusion and mycorrhizal connections and quantification of their contribution remains difficult. From the results of our study, we suggest mycorrhiza being responsible for one third to half of the N derived from peas found in triticale. But for all that, some methodological concerns, such as different growing patterns of Frisson and P2 and the difficulty to quantify total N transfer from a reduced contact area remain, and have to be taken into consideration in interpreting the data. Future research should further elaborate on the relevance of the different transfer pathways, by combined use of plant varieties with their mutant isolines and isotopic labeling. We expect a more in depth view and quantitative verification from the results of a field trial.

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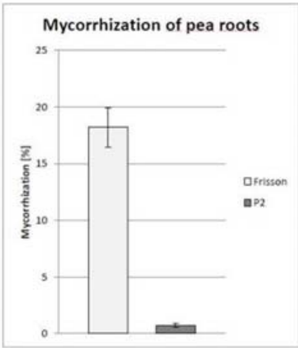

Figure 1: Mycorrhization of two pea varieties (Frisson and P2). Mean ± SE; n=36

Table 1: Above ground biomass and total plant N of two pea varieties (Frisson and P2) and intercropping partner Triticale. Mean ± SE; n=12

Above-ground biomass [g]				
Root separation	Frisson	Triticale	P2	Triticale
Without	11.44 ± 1.16	3.63 ± 0.50	6.66 ± 0.51	3.65 ± 0.38
Single	12.54 ± 0.69	3.04 ± 0.21	6.40 ± 0.56	3.03 ± 0.29
Double	11.65 ± 1.03	2.92 ± 0.23	5.42 ± 0.38	2.94 ± 0.17
Total N / plant [mg]				
Root separation	Frisson	Triticale	P2	Triticale
Without	400.78 ± 43.86	163.43 ± 21.89	180.55 ± 13.85	157.50 ± 18.22
Single	460.86 ± 19.24	137.74 ± 8.37	193.51 ± 11.36	136.40 ± 11.46
Double	444.88 ± 33.54	125.63 ± 8.10	156.87 ± 10.80	136.20 ± 7.38

Table 2: Enrichment with ^{15}N in roots of two pea varieties (Frisson and P2) and intercropping partner Triticale. Mean ± SE; n=6

Enrichment with ^{15}N [atom%excess]				
Root separation	Frisson	Triticale	P2	Triticale
Without	1.99 ± 0.24	0.1183 ± 0.0249	2.12 ± 0.22	0.0427 ± 0.0060
Single	1.63 ± 0.21	0.0846 ± 0.0301	2.41 ± 0.20	0.0164 ± 0.0018
Double	2.28 ± 0.34	0.0346 ± 0.0219	2.02 ± 0.23	0.0079 ± 0.0014

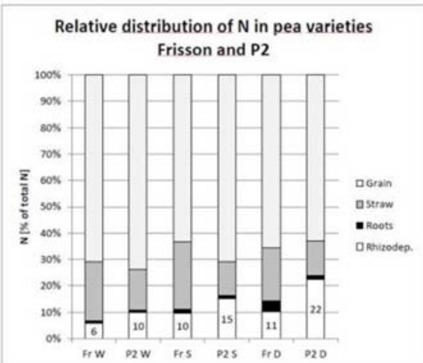

Figure 2: Relative distribution of N [%] in two pea varieties. Fr=Frisson, W=without root separation., S = single root separation, D=double root separation. Mean; n=6

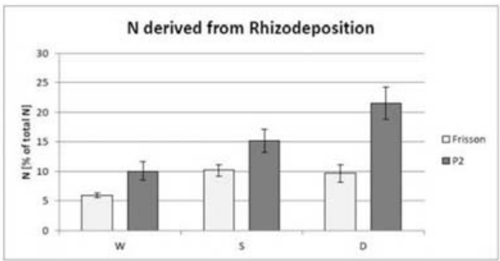

Figure 3: Proportion of total N derived from rhizodeposition of two pea varieties (Frisson and P2). W=Without root separation, S=single root separation. D=double root separation. Mean ± SE; n=6

ENZYME ACTIVITY AND MICROBIAL DIVERSITY IN THE RHIZOSPHERE OF TWO MAIZE LINES DIFFERING IN N USE EFFICIENCY

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Microbial biomass, B-glucosidase, acid and alkaline phosphomonoesterase, phosphodiesterase, urease and arylsulphatase activities and the microbial community structure were determined in the rhizosphere of two contrasting maize lines (Lo5 and T250) differing in the nitrogen use efficiency (NUE). The Lo5 and T250 inbred maize characterized by high and low NUE, respectively, were grown in rhizoboxes allowing precise sampling of rhizosphere and bulk soil and solution. Microbial community structure was determined by using a phylogenetic group specific PCR-DGGE approach in the rhizosphere and bulk soil of both Lo5 and T250 maize lines. High NUE Lo5 maize induced faster in inorganic N depletion in the rhizosphere than the low NUE T250 maize, and induced the larger changes in microbial biomass and enzyme activities. The two maize lines induced differences in the studied microbial groups in the rhizosphere, with the larger modifications induced by the high NUE Lo5 maize line.

The Lo5 maize line with higher NUE induced larger changes in the chemical properties and in the enzyme activity, microbial biomass and community structure than the low NUE T250 maize line, probably due to differences in the root exudates of the two maize lines.

INHIBITION AND RECOVERY OF SYMBIOTIC N2 FIXATION BY PEAS (PISUM SATIVUM L.) IN RESPONSE TO SHORT-TERM NITRATE EXPOSURE

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Cereal-legume intercrops are a promising way to combine high productivity and several ecological benefits in temperate agro-ecosystems. However the proportion of each species in the mixture at harvest is highly variable. N-fertilisation may be a mean for controlling these proportions (Naudin *et al.*, 2010). Symbiotic N_2 fixation (SNF) can be inhibited by nitrate exposure. Although the effect of nitrate on SNF has been extensively investigated different scales, few studies have highlighted the impact of different timings of nitrate exposure on the SNF apparatus (N_2 fixation rate and nodule biomass) and on its ability to recover after nitrates removal. The aim of this study was to test the consequences on SNF of short-term exposure to nitrate at different growth stage of pea.

Materials and Methods

A field experiment was carried out in western France in 2007-2008. Winter field pea (*Pisum sativum* L.) and winter wheat (*Triticum aestivum* L.) were grown as intercrops (IC) in a substitutive design (at 40 pl m^{-2} and 130 pl m^{-2} for pea and wheat, respectively) with or without N fertilisation. Experiments were arranged in randomised complete block designs with three replicates. Weeds, pests and diseases were controlled with pesticides when required. No irrigation was provided. N was applied as NH_4NO_3 as liquid fertilizer at several single dates from wheat tillering to the end of wheat stem elongation (Table 1). A greenhouse experiment was carried out in Angers (France) in 2008. Pea plants were grown in hydroponic conditions in a N-free nutrient solution and exposed to nitrate (5 mM NO_3^- L^{-1}) for one week during either early vegetative growth, flowering or seed filling. After nitrate removal, the plants were grown either under natural light or shade (Table 2). In all experiments, N-fertilizers were enriched with ^{15}N in order to estimate the dynamics of the contribution of nitrogen derived from air (%Ndfa) to aboveground N in peas.

Results and discussion

In the field experiment, %Ndfa of intercropped pea in unfertilized intercrops increased gradually with thermal time up to 90% at the end of the crop cycle (Fig 1a). The earliest N-supply (IC Tillering) entailed a transitory high decrease in %Ndfa. After %Ndfa increased again but remained below the unfertilized intercrop during a main part of the crop cycle. It reached the same value as %Ndfa observed in unfertilized intercropped peas at maturity. The latest N-supply entailed a higher decrease in %Ndfa without a recovery. Soil N-mineral content in 0-30 cm soil layer tended to increase during the few days after N-applications (Fig 1b). Then, it remained close to soil N-mineral content observed in no-fertilized conditions. The intensity of the transitory inhibition of N_2 fixation during the two weeks succeeding N-applications is mainly dependent on the nitrates availability. %Ndfa of N-fertilized intercropped peas decreased linearly with soil nitrate content in 0-30 cm soil layer ($y = -1.9183x+103.6441$; $r^2 = 0.9587$**). Total inhibition threshold of nitrate content on symbiotic N fixation could be observed between 35 and 50 kg NO_3^- ha^{-1}, as previously shown in sole crop by Voisin *et al.* (2002). In the greenhouse experiment, the period of root exposure to nitrate had a massive effect on nodule dry weight after nitrate removal (Fig 2). Considering nitrate exposure at the vegetative stage, the first plant response during the two weeks following the nitrate removal was an increase in nodule growth that was not observed in control plants. Nodule growth was prolonged during the growth cycle, whereas it stopped at mid-flowering in control plants (Fig 2a). The second plant response involved a prolonged period of new nodule generation until the end of the growth cycle, whereas new nodule generation in control plants stopped earlier at mid-flowering (Fig 2b). At the end of the growth cycle, nodule biomass was 36% higher in nitrate-treated plants than in the control plants (Fig 2a). Similarly, nitrate removal after an exposure during flowering stage resulted in the simultaneous initiation of new periods of nodule generation and nodule growth, whereas in control plants, nodule biomass and nodule number had stopped increasing. Finally, in nitrate-treated plants, at the end of the growth cycle, nodule biomass was 26% higher than in the controls (Fig 2b). By contrast, considering nitrate exposure at the seed filling stage, the plant response after nitrate removal was null as nodule biomass remained stable until the end of the growth cycle, as it did in control plants. However, due to a decrease during nitrate exposure, nodule biomass was 36% lower than in the control plants at physiological maturity (Fig 2c).

Table 1
Growth stages of wheat were described based on Zadoks'scale (ZGS). "GS": Growth stage. Vegetative stages of pea were noted "Veg".

Treatments	Crops conditions at the date of N-fertilization: Wheat ZGS	Pea GS	N-fertilization: Time	Rate (kg N ha^{-1})
IC N0	—	—	—	0
IC Tillering	ZGS23	Veg (8-leaf)	07/02	45
IC E1	ZGS30	Veg (10-leaf)	07/03	45
IC E1+1	ZGS32	Veg (14-leaf)	10/04	45
IC Flo	ZGS65	Beginning of Seed Filling	15/05	45

Table 2
DAG: days after seedling germination; Veg: vegetative stage; Flo: flowering; SF: seed filling; L+ and L-: growth under natural light and under shade, respectively.

Treatments	Date of nitrate exposition (DAG)	Dose of nitrate exposition (mM NO_3^- L^{-1})	Growth conditions after nitrate exposition
Control	—	0	natural light
N_{Veg} L+	from 14 to 22	5	natural light
N_{Veg} L-		5	shaded
N_{Flo} L+	from 49 to 57	5	natural light
N_{Flo} L-		5	shaded
N_{SF} L+	from 56 to 64	5	natural light
N_{SF} L-		5	shaded

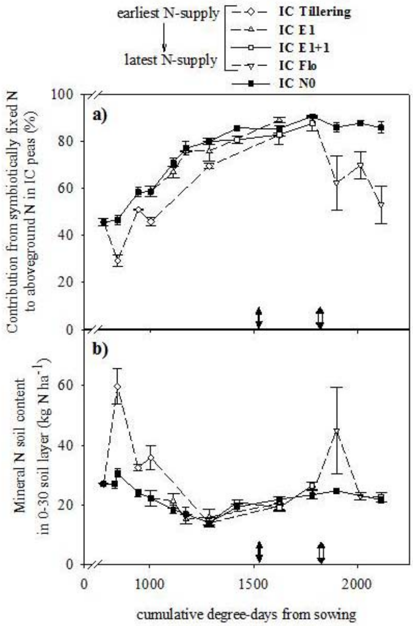

Figure 1 Contribution from symbiotically fixed N to aboveground N in peas and mineral N soil content in 0-30 cm soil layer after N-fertilization on intercrop.
Values are means (n=3) and standard error calculated for respective treatments. ↥: beginning of pea flowering. ⇳: beginning of pea seed filling.

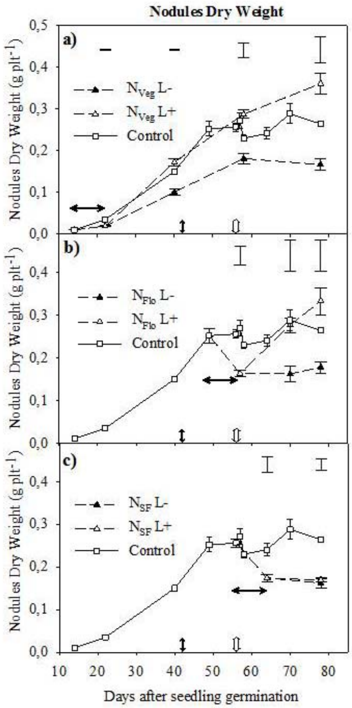

Figure 2 Dynamics of nodule dry weight accumulation
Control: pea plants never exposed to nitrate and grown under natural light. N_{Veg}, N_{Flo}, and N_{SF}: pea plants exposed to nitrate (5 mM NO_3^- L^{-1}) during vegetative growth (a), flowering (b) or seed filling (c), respectively. L+ and L-: under natural light or shade, respectively, from after nitrate exposure until maturity. Values are means (n=8) ± SE (standard error bars in the plots). Analysis of variance (type III sum of squares, α= 5%) was carried out to compare treatments at the respective dates of observation: vertical bars represent Tukey's HSD (α= 5%). ↔: exposure to nitrate ↥: beginning of pea flowering. ⇳: beginning of pea seed filling.

Conclusion

Our results demonstrated the sensibility and reactivity of SNF of pea to different dynamics of nitrate exposure (Naudin *et al.*, 2010; 2011). It illustrated the significance of SNF of legumes to act as a buffer when soil mineral N varied by adapting its N acquisition pathways.

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CANOLA RESPONSE TO N FERTILIZER AND ROTATION SYSTEMS IN PRINCE EDWARD ISLAND

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Prince Edward Island (PEI) producers are consistently growing more canola than ever before (Statistics Canada, 2011). Potatoes are still the main crop in PEI representing around 25% of Canadian potato production. Currently, potatoes are rotated with barley under seeded with red clover with the latter being incorporated in the third year. Research is needed to assess how canola would fit into the potato production system. Prince Edward Island soils are loamy to sandy and have low organic matter content. The objective of this study was to evaluate potential canola yield and N dynamics during the growing season following different potato rotation systems and N sources.

Materials and Methods

The experimental design situated at Harrington Research Farm (PE, Canada) was a split- plot with the main factors being 4 rotation systems installed in 2006: i) potatoes-barley (PB); ii) potato-sudan grass/winter rape (PSW); iii) potato-canola-winter rape (PCW); iv) potatoes-sudan grass/winter rape-canola/winter rape (PSWCW). The second factor was composed of 3 N sources: mineral N fertilizer, liquid manure and lobster flake applied during potato phase from 2006 to 2012. From 2006 to 2011, when canola was part of the rotation, it was plowed under as green manure. In 2013, all plots were seeded to canola and liquid manure and lobster flake were not applied in 2013 to assess their residual effects. Nitrate dynamics during the growing season was assessed using anion exchange membranes. Canola yield, grain protein concentration and grain N uptake as well as one thousand kernel weight (TKW) were determined.

Results and Discussion

Data from anion exchange membranes showed that PSW and PSWCW were associated with higher NO_3 released on July 23th, but PSW was shown to mineralize more N later in the growing season than other rotation systems (Figure 1A). Among N sources, lobster flake and mineral fertilizer were associated with higher nitrate adsorbed onto anion exchange membranes (Figure 1B). The N release pattern of liquid hog manure appeared to be more constant than lobster flake and mineral N (Figure 1B). Canola yield, grain protein concentration (GPC), grain N uptake and thousand kernel weight (TKW) followed the ranking: lobster flake>liquid manure> mineral N (Table 1). Among the rotation systems, sudan grass (PSW> PSWCW) was associated with higher yield, GPC, N uptake (Table 1). Mineral N fertilizer was associated with lower grain yield, grain N uptake and TKW compared to organic sources probably due to the asynchrony between N released and canola N demand. Values of canola protein contents observed in this study are lower than the Western Canadian canola five year average of 20.4% (Canadian Grain Commission board, 2013). It was reported an inverse relationship between GPC and canola oil contents (Brennan et al., 2000).

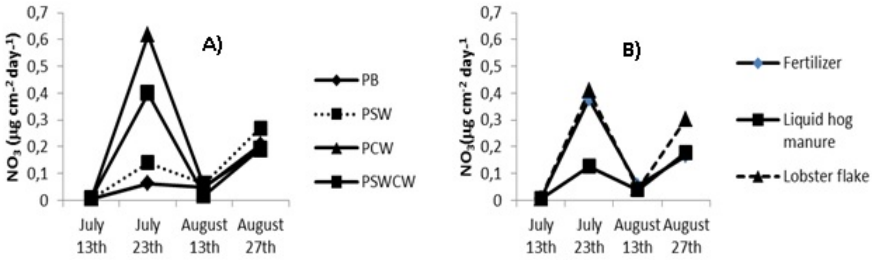

Figure1. Nitrate dynamics during 2013 canola growing season by using anion exchange membranes. BP, potato-barley; PSW, potato-sudan grass/winter rape; PCW, potato-canola winter rape; PSWCW, potato-sudan grass/winter rape-canola/winter rape. From 2006 to 2011, when canola was part of the rotation, it was plowed under as green manure.

Table 1. Residual effects of N sources and crop rotations on canola grain yield, grain N uptake, grain protein concentration (GPC) and on thousand kernel weight (TKW)

	Grain yield (Mg ha^{-1})	Grain N uptake (kg N ha^{-1})	GPC^{Y} (%)	TKW (g)
N sources				
Mineral N	2.1b	57.3c	17.4a	2.95b
Liquid manure	2.2b	61.8b	17.4a	3.07ab
Lobster flake	2.6a	76.5a	18.2a	3.17°
Rotation systems				
PB	2.2b	61.5bc	17.0b	2.93b
PSW	2.6a	74.3a	17.6ab	3.14°
PCW	1.9c	56.6c	17.9a	3.09°
PSWCW	2.3b	68.3ab	18.28a	3.08°
Analysis of variance(p values)				
Rotation effects	< 0.01	< 0.01	< 0.01	< 0.01
N sources effects	< 0.001	< 0.01	0.06	0.018
Rotationsx N source	0.181	0.303	0.918	0.708

^{Y}= N x 6.25 (Canadian Grain Commission Board, 2012). BP, potato-barley; PSW, potato-sudan grass/winter rape; PCW, potato-canola winter rape; PSWCW, potato-sudan grass/winter rape-canola/winter rape. From 2006 to 2011, when canola was part of the rotation, it was plowed under as green manure.

Conclusion

This study demonstrated that liquid manure and industry by-products, such as those from fisheries, can constitute an excellent canola nutrient source one year after their last application. Nitrogen source and cropping histories impact canola yield and quality and synchronizing N mineralization from different sources with canola N peak demand remains challenging.

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THE EFFECTS OF PRECEDING CROPS AND THEIR MANAGEMENTS ON SUBSEQUENT POTATO PRODUCTION

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Efficient nitrogen management is linked to environmental and economic benefits. Prince Edward Island contributes to about 25% of Canadian potato production (Statistics Canada, 2009). Previous studies have reported increasing nitrate losses under potato production (Jiang et al., 2011). Identifying best management practices (BMPs) to sustain potato productivity while minimizing water contamination by nutrient loading is still challenging. The objectives of this study were to evaluate the management impacts of annual rye, white and red clover on subsequent potato production. Two seeding times (early and late) and weed management regimes (spray with herbicide, no spray) were compared.

Materials and Methods

An experimental was established at Harrington Research Farm (PE, Canada) in summer 2012 which compared 4 potato preceding crops (bare soil, red clover, white clover and annual rye). Red clover, white clover and annual rye were seeded at two different dates (June 13 and August 27th). Potato preceding crops were either sprayed (liberty at 2.5L ha^{-1}) or not sprayed to control weed pressure in 2012 and were incorporated in spring 2013 prior potato seeding. For the potato phase, the experimental unit was split (no fertilizer versus recommended fertilizer rate). Only results associated with unfertilized plots will be presented here to elucidate how management of different crops affected the capacity of a soil to supply nutrient without the confounding effects of fertilizer. Parameters measured include: chlorophyll meter readings (3 dates), vine and tuber dry matter and total N accumulation prior vine desiccation (top kill). At harvest, potato marketable yield was determined. A two weak aerobic incubation was carried out on soil (0-15 cm) taken in spring before potato seeding to analyze the soil N supply capacity (SNS).

Results and Discussion

Spray effect was statistically significant with higher values recorded in sprayed treatments (Figure 1A) for samples taken on July 2 and 17th and the reverse effect was observed for readings taken on August 1st but there was no effects of seeding time or preceding crops on chlorophyll meter readings (Figure 1B and 1C). Similar trends were also observed with vine and tuber dry matters, N uptake and total marketable yield with higher values associated with sprayed plots (Table 1).

Soil N supply capacity was higher with rye or clover compared to a bare soil (Table 1). Results from chlorophyll meter readings suggest that biomass from sprayed plots tended to mineralize early in the growing season following their incorporation but the reverse was observed later in the growing season for unsprayed plots, implying that weed management regimes affect quantity and quality residues and thus their mineralization rate.

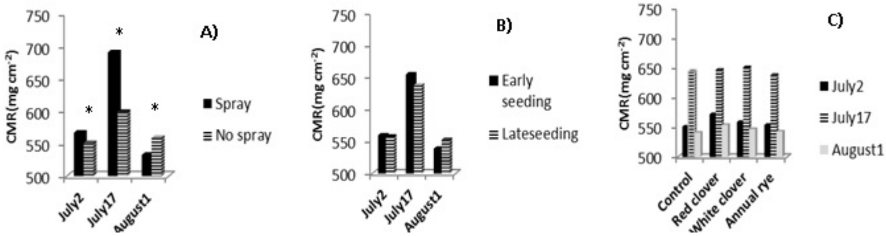

Figure 1. Chlorophyll meter readings (CMR) measured in unfertilized plots during potato phase in 2013. * indicates that the means within a group are statistically different at 0.05 probability level.

Table 1. Effect of cover crop and green manure management on soil nitrogen supply capacity (SNS) before seeding potato, potato dry matter accumulated before vine desiccation, N uptake, and marketable potato yield in unfertilized plots

Main effects		**SNS**	**Dry matter before vine dessication (Mg ha^{-1})**		**N uptake (kg N ha^{-1})**		**Marketable yield**
		mg kg^{-1}	Vines	Tubers	Vines	Tubers	Mg ha^{-1}
Seeding time (ST)	Early	38	1.96	8.6	44.5	68.3	36
	Late	30	1.40	7.4	29.8	59.3	28
Spray (S)	No	34	1.38b	7.3b	26.8	58.7	27b
	Yes	34	1.98a	8.7a	47.2	68.8	37a
Type of crops (T)	Control	28b	1.68	7.8	38.4	68.7	31
	Red clover	36a	1.79	8.4	38.7	64.1	32
	White clover	37a	1.84	8.2	39.3	65.4	34
	Annual rye	36a	1.40	7.7	31.9	56.9	31
Analysis of variance (p values)							
S		0.873	0.026	< 0.01	0.020	0.279	< 0.01
ST		0.057	0.164	0.121	0.220	0.429	0.126
SxST		0.412	0.548	0.855	0.495	0.897	0.472
T		0.011	0.629	0.637	0.912	0.827	0.985
SxT		0.984	0.755	0.136	0.928	0.870	0.704
ST xT		0.476	0.469	< 0.01	0.446	0.225	0.883
SxSTxT		0.335	0.669	0.804	0.827	0.830	0.934

SNS, sum of NO_3-N extracted at time zero and NO_3-N extracted after 14 days of soil aerobic incubation. Values followed by different letters within a group are statistically different at 0.05 probability level.

Conclusion

Overall, compared to a bare soil fallowed during one growing season, growing a grass or a legume showed an increase in the soil N supply capacity but any significant effect of crop type was observed on chlorophyll meter readings, dry matter, total N uptake or total marketable yield. Crop management was shown to affect subsequent potato production with sprayed plots showing higher dry matter, N uptake, and marketable yield. There were also trends towards higher values with early seeding than late seeding. Further studies are needed to validate these findings and to take into account differences in annual variations and soil types.

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LEGUME NITROGEN TRANSFER IN GRASS-CLOVER LEYS – A 15-NITROGEN NATURAL ABUNDANCE STUDY IN ORGANIC AND CONVENTIONAL CROPPING SYSTEMS

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The di-nitrogen (N_2) fixation by the legume-rhizobia symbiosis provides an essential nitrogen (N) input into agro-ecosystems and is the most important external N source in organic farming. To cope with increasing N demand for greater food and forage production, ways to increase this input and to provide symbiotically fixed N to other crops than the legume itself need to be further explored. An option providing this benefit is the transfer of legume N to associated grasses in grass-legume mixtures. While this N transfer has been demonstrated using ^{15}N enrichment methods (Nyfeler et al. 2011), the potential of the ^{15}N natural abundance has been explored less. Furthermore, N transfer has not yet specifically been studied under organic vs. conventional cropping. The objective of the overall study was to determine the input of symbiotically fixed N in grass-clover leys under conventional and organic cropping. In our presentation we will focus on the N transfer.

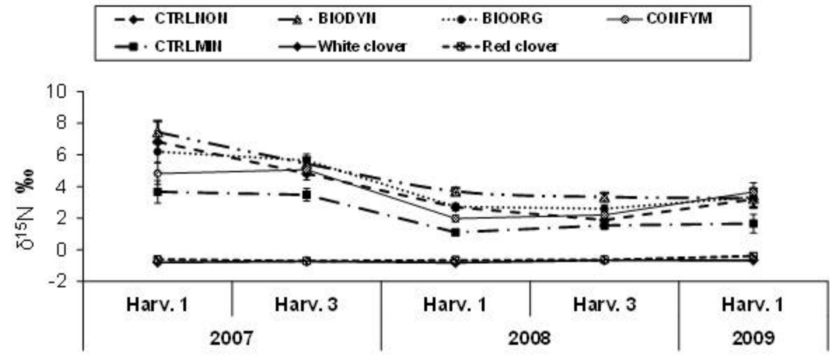
Material and methods

We studied the N_2 fixation by clover and the transfer of clover N to the associated grasses in grass-clover mixtures using ^{15}N natural abundance methods (Oberson et al. 2013). The mixtures were included in the crop-rotation of a long term field experiment started in 1978 to compare organic and conventional cropping systems (DOK experiment: bio-Dynamic, bio-Organic, Konventionell) (Mäder et al. 2002). Mixtures comprised white clover (*Trifolium repens* L.*)*, red clover *(Trifolium pratense* L.*)* and several grass species, with perennial ryegrass *(Lolium perenne* L.*)* being the dominating species. We followed over a two year lasting ley phase (2007-2009) the yields, N concentrations and the ^{15}N natural abundance of red clover, white clover and the grasses. We estimated the proportion of N in clover derived from the atmosphere (PNdfa, in %) using the method from Shearer and Kohl (1986). The proportion of N in the grasses derived from clover (PNdfc, in %) was calculated from the changes of δ^{15}N over time in ryegrass by adapting the formula of Daudin and Sierra (2008) and by carefully checking the assumptions underlying its application (Oberson et al. 2013).

Results and discussion

The monitoring of δ^{15}N from May 2007 until April 2009 showed that δ^{15}N in red and white clovers was already below 0 in the first year of utilization of the grass-clover ley and remained low over the duration of this study (Fig. 1). The δ^{15}N of both clovers were not significantly affected by the treatments. In contrast, the δ^{15}N of ryegrass was significantly affected by treatments ($p < 0.05$) and significantly decreased with time, particularly from year 1 to year 2 ($p < 0.001$) (Fig. 1). Throughout the study, the δ^{15}N values of the ryegrass were significantly higher than the $\delta^{15}N$ values of both clovers (*p*< 0.001). The clear and significant differences in $\delta^{15}N$ between clover and ryegrass translated into high PNdfa (on average 90%). The PNdfa was not significantly affected by the treatment or clover species and remained at high level. The decrease in $\delta^{15}N$ in ryegrass over time suggested that the soil N pool which it was exploiting became increasingly affected by clover derived N (Fig. 1). Other factors causing changes in $\delta^{15}N$, such as modifications in form and amount of fertilizers, could be ruled out. Furthermore, the decrease in $\delta^{15}N$ occurred in all fertilized treatments and in the unfertilized control, and the $\delta^{15}N$ signature of the grass reached values that were below the soil and fertilizer N sources of the organic treatments. The resulting proportion of N in the ryegrass growing in the two-year-old leys derived from clover ranged from 46 to 60%. Because the PNdfc was similar in all treatments, the amount of clover N in grasses was higher with greater grass N yields. As clover N in each system was largely derived from the atmosphere, belowground N transfer to the grass constituted an additional input of atmospheric N (35 to 100 kg ha^{-1} yr^{-1}).

Fig. 1 Evolution of $\delta^{15}N$ (‰) over time in ryegrass and clover sampled in grass-clover leys under organic (BIODYN, BIOORG) and conventional (CONFYM) cropping systems, in the unfertilized control (CTRLNON), and in the mineral fertilized control (CTRLMIN). Because the $\delta^{15}N$ for clover was not significantly affected by the treatments, average and standard error of mean over all five treatments is shown for white and red clover, with n=4 per treatment (reprinted from Oberson et al., 2013).



Conclusions

The natural abundance method proved useful to assess the N transfer from clover to grasses. The N transfer provides an important N source to grasses growing in association with clover. This transfer needs to be included in N budget studies as it increases the input of symbiotically fixed N beyond the N contained in legume biomass. Organically and conventionally cropped grass-clover leys fully profited from symbiotic N_2 fixation.

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IMPROVING THE UNDERSTANDING OF NITROGEN PROCESSES BY COMBINING FIELD OBSERVATIONS AND DYNAMIC ECOSYSTEM MODELLING

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Nitrogen and carbon processes in forest ecosystems are important for several environmental issues. However, scientific knowledge of these highly complex interactions is still limited. By combining empirical data with dynamic models, an increased understanding of these processes could be achieved which would reduce uncertainties in projections of future effects caused by e.g. changes in climate, atmospheric nitrogen deposition and forest management strategies. In this project, we have combined dynamic ecosystem modelling with empirical data from field sites in Southern Sweden, with the overall objective to increase the understanding of nitrogen processes and cycling in forest ecosystems. One detailed focus has been to investigate the risk of nitrogen leaching from planted forests, which is a risk that needs to be better understood, e.g. when considering an increased use of artificial fertilization in forests for increasing carbon uptake and biomass productivity (wood and biofuel).

Materials and Methods

Empirical field data was provided by the Swedish Throughfall Monitoring Network (SWETHRO) (Pihl Karlsson et al., 2011). These data from forest sites includes monthly open field deposition and throughfall measurements, and soil solution data measured three times per year (representing before, during and after the growing season). The modelling was performed with ForSAFE (Wallman et al., 2005), a dynamic ecosystem model handling dynamics and feedbacks between different processes. The ForSAFE model uses algorithms integrating processes of tree growth, decomposition, weathering, hydrology and ground vegetation. For simulations for the 21st century, we utilized atmospheric deposition scenarios provided from the MATCH model (see e.g. Engardt and Langner, 2013), with climate data from global climate models. The ForSAFE model was applied with site-specific setups (based on empirical observational data from SWETHRO), and simulations were performed with different MATCH deposition scenarios.

Results and Discussion

Two sites in southern Sweden situated only a few kilometres apart, show highly different characteristics regarding observed soil water chemistry. One site shows very low N concentrations in soil water, while the other shows rather high concentrations. Applying the ForSAFE model, preliminary results indicate that differences in the soil chemistry between the two investigated sites might explain at least part of the observed difference in nitrogen concentrations. Further detailed investigations are ongoing, including applying different scenarios of forest management, climate and atmospheric deposition. Additionally, initial analyses suggest that the modelled results are affected by uncertainties related to past land management as well as uncertainties in climate and deposition input data.

Conclusions

We show that a combination of field site observations and dynamic ecosystem modelling can enhance the understanding of the processes regulating the nitrogen processes and cycling in forest ecosystems, and by so also result in improving dynamic models. This could reduce uncertainties in future projections, and be important when evaluating positive and negative effects of nitrogen fertilization or other forest management strategies. Furthermore, these results could be a useful contribution for the environmental protection work related to key environmental issues such as climate change, carbon sequestration, eutrophication and acidification.

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APPLICATION OF SUGAR BY-PRODUCTS AND NUTRIENTS BY FERTIGATION ENHANCES FRUIT TREE GROWTH AND YIELD PRECOCITY

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Sugars application has been shown as effective method for increase rooting on urban and nursery arboriculture. Sugars (commonly from molasses) application promotes the growth of fine roots and, consequently, improves root and shoot growth and photosynthetic rate on trees (Percival et al., 2004; Percival and Fraser, 2005; Martínez-Trinidad, 2009). However, this subject is rare on fruit tree research (van Schoor and Stassen, 2008). The aim of this study is to demonstrate the effectiveness of the sugar compounds applied by fertigation for on peach tree growth and yield.

Materials and methods

A two-year field experiment on the region of Segrià (Lleida) was conducted in a commercial orchard of flat peach (*Prunus persica* (L.) Batsch. var. platycarpa (Decne.) LH Bailey cv. Planet Top®) grafted on GF677® rootstock. Orchard was planted in 2009 in ridge, spaced 2.5 x 4.5 m. Three nutrient treatments based on modified Hoagland's solution (N, 145 Ca, 1.4 Mg, 17.9 P, 9.9 K, 172.2 S, 25.1 Cu, 0.23 B, 0.08 Fe, 5.60 Mn, 0.60 Mo, 0.02 Zn, 0.23 -all in ppm- pH, 2.3 CE:2 dSm) were applied: NUT100 (solution application along growth season), NUT50 (half solution concentration) and NUT25 (one quarter solution concentration). NUT50 and NUT100 were combined with application of a commercial sugars compound made from molasses (RootTip®, AltincoAgro, S.L.) at doses of 20 (S50) and 40 kg ha^{-1} (S100) by fertigation and no sugar application (S0). Plots of 8 trees each one was arranged as randomly block design with three replications. All treatments were irrigated with ETc calculated from data of field weather station. Irrigation water and nutrient solution was applied by high frequency pulses (daily, as minimum). Capacitance probes were installed for soil moisture monitoring to avoid water stress. Statistical analysis of data was carried out using the SAS-STAT package (SAS®, Version 9.2. SAS Institute Inc., Cary, NC, 1989-2009).

Results and Discussion

Rich-nutrient solutions on first years of peach tree growth enhance canopy development and promote rapid tree fruiting and improve fruit weight (Table 1). In front, nutrient solutions at low or moderate concentration diminish fruit N/Ca ratio because diminution of nitrogen concentration and calcium increase (Figure 1). Application of sugar compounds at high dose (S100) exhibit an interesting and positive effect on the vegetative growth and yield. Nutrient contents in fruits were not affected by sugars, regardless of nutrient treatments.

Conclusions

Use of rich-nutrient solutions on peach trees improve canopy development and promote rapid tree fruiting. The effect of nutrient solutions combined with sugar compounds application had a positive additive effect on vegetative growth and fruit yield, improving fruit N- Ca balance.

Table1. Effects of Nutrient and sugars treatments on yield, fruit weight, Fruit firmness (FF), Soluble Solids (SS), fruits by tree, contents on fruit and leaf (prefix fr or le respectively) Nitrogen, Calcium, Magnesium, Potassium, fruit Nitrogen/Calcium ratio (frN/Ca) Phosphorous and Sulfur, Carbon/Nitrogen relation in fruit (C/N), fruit skin L and a color (Cie-Lab system) and trunk diameter (TD). Values followed by different letters indicate significant differences according to Tukey HSD test (P <0.05). Significance of effects and interactions is noted by (*) (**) (***) at P level < 0.05; <0.01, < 0.001; and ns non-significant respectively.

	NUT				S				N x S
	NUT25	NUT50	NUT100		S0	S50	S100		
yield (t ha-1)	38.1 b	46.3 ab	50.7a	*	45.2 b	43.9 b	55.0 a	**	ns
FW (g)	131.6	141.9	138.8	ns	141.4	144.6	142.9	ns	ns
FF (Nw)	4.2	5.1	5.1	ns	5.2	5.2	5.0	ns	ns
SS (Brix)	13.9	13.8	13.5	ns	13.9	13.7	13.3	ns	ns
fruits tree-1	312.2	332.1	364.7		329.1 b	310.9 b	409.0 a	**	ns
frN (ppm)	6729.9 b	9827.5 a	11061.8 a	**	9763.0	10106.4	9971.7	ns	ns
frCa (ppm)	635.0 a	511.3 b	485.4 b	**	516.0	498.2	468.1	ns	ns
frMg (ppm)	584.2	570.6	589.9	ns	594.0	569.3	578.9	ns	ns
frK (ppm)	13186.2	12785.9	13785.0	ns	13522.4	13076.4	13351.8	ns	ns
frP (ppm)	1226.5	1191.5	1234.5	ns	1259.7	1224.4	1147.6	ns	ns
frS (ppm)	352.4	412.0	416.1	ns	435.7	411.6	391.4	ns	ns
leN (%)	2.70 b	3.35 a	3.37 a	**	3.35 ab	3.56 a	3.12 b	**	ns
leCa (%)	3.6 a	2.9 ab	2.7 b	*	3.00	2.96	3.11	ns	ns
leMg (%)	0.43	0.34	0.32	ns	0.34	0.31	0.35	ns	ns
leK (%)	2.25	2.27	2.41	ns	2.42	2.33	2.28	ns	ns
leP (%)	0.17	0.17	0.17	ns	0.16	0.18	0.15	ns	ns
leS (%)	0.06	0.07	0.07	ns	0.07	0.08	0.06	ns	ns
C/N	20.9	14.3	12.3	ns	12.6	13.8	13.4	ns	ns
frN/Ca	11.0 c	19.2 b	23.2 a	**	19.3 b	18.6 b	22.5 a	ns	ns
skL*	56.0	52.3	54.0	ns	53.6	53.0	54.3	ns	ns
ska*	20.2	19.3	17.1	ns	18.0	18.5	17.8	ns	ns
TD (mm)	67.0 b	68.5 b	73.8 a	**	71.1 b	67.4 b	76.3 a	**	ns

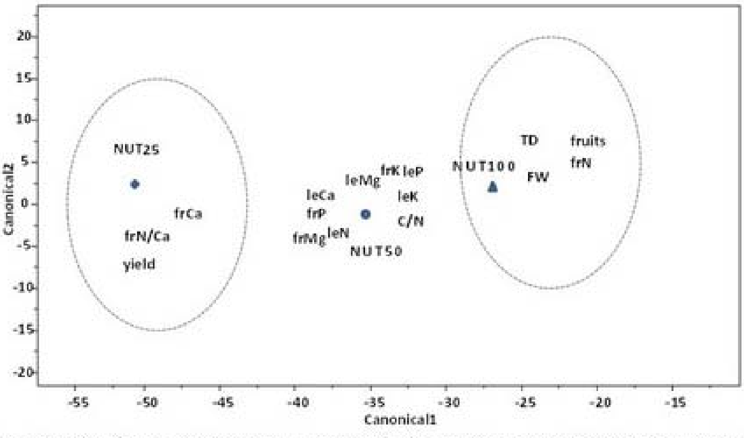

Figure1. Results of canonical discriminant analysis for Nutrient treatments. Canonical component 1 explains 79.2% of total variability.

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CANOPY MANAGEMENT ON RAINFED VINEYARDS FOR OPTIMIZATION OF NITROGEN AND WATER USE AND WINE QUALITY IMPROVEMENT

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Rainfed systems on viticulture dominate world's production of grapes for wine. In these conditions, several gaps limit grape yield and wine quality. Soil characteristics, mainly the interaction between water holding capacity and nutrients availability, constitute the keystone to define technological roadmap adapted to site-specific response. Grapevine canopy management strategies affect to grapevine water availability and nutrients uptake during the growing season, contributing to wine quality by the enhancement of microclimate canopy characteristics and physiological responses to water status, mainly during grapes maturation (Hunter and Archer, 2001; Choné et al. 2001). The objective of this study was to evaluate the effects of canopy management strategies on plant organic and inorganic nitrogen and wine composition and quality.

Materials and methods

A two-year field experiment (2009-2010) on grapevine was conducted on an adult vineyard cv. Tempranillo in Somontano region (Northeast Spain). Vineyard was selected according to the homogeneity on culture practices and cellar performance classification. Two sites in vineyard were selected according to soil deep and water holding properties (first point with no limiting rooting deep and second point with shallow soil and limited rooting conditions). Three canopy management treatments were randomly arranged (4 replicates of 10 plants) on each site: canopy management according common practices as a vertical shoot positioning and topping after veraison (C), leaf removal of fruiting zone at veraison (LR), and repeated shoot topping -3 times- after berry set (ST). Macro and micro elements were analysed in leaf petioles at veraison time. Plant water status was measured as stem water potential (y_s) at veraison and at harvest time. Exposed canopy area was measured by image analysis. Yield, crop load, weight of clusters and berries, sugars, pH, acids, phenolic compounds, yeast available nitrogen (YAN) and amino acids were analysed in must at harvest time. Wine analysis and tasting was performed after microvinification in controlled fermentative conditions to make quality wine (50L). Statistical analysis was carried out using Principal components Analysis.

Results and Discussion

First two PCA components account 74.7% of experimental variance (Fig 1). Analysis shows complexity of the interaction between canopy management practices and water availability (Tab 1). Analysis reveals a well- known effect: soils with more water availability (Point 1) promote higher berry weight, a negative factor for wine quality. Independently, ST and LR promote a conservative water behavior and a correlative plant water status improvement. In such conditions greater absorption of nitrogen in the plant occurs when water deficit is not produced on veraison. Low y_s on veraison and harvest does not promote nitrogen accumulation and YAN in must, but, in general, improves nutrient content in petioles. Canopy control (ST and LR) is shown as positive as measure for water saving and shows an effect on wine style. Wine obtained from ST is more complex and better acid balanced (Acid E). Meanwhile, LR exhibits a positive effect on quality when water is available. Control treatment shows a higher concentration of amino acidsand aroma potential (PAI), although the wine not exhibit better characteristics than the LR and ST.

Table1. Load matrix for two first PCA factors.
Relevant variables are marked in bold.

	Factor 1	Factor 2
C	**0,79**	0,26
LR	-0,30	**0,56**
ST	**-0,62**	**-0,77**
POINT 1	**-0,92**	0,29
POINT 2	**0,92**	-0,29
Aspartic Ac.	**-0,67**	0,06
Serine	**0,74**	-0,06
Glutamine	**0,88**	-0,10
Histidine	**0,92**	0,10
Glycine	**0,63**	**0,62**
Alanine	0,05	**0,78**
GABA	0,55	0,30
Tyrosine	**0,93**	-0,20
Valine	**0,88**	0,24
Methionine	**0,92**	-0,29
Triptophan	**0,69**	0,12
Phenylalanine	**0,99**	-0,06
Isoleucine	**0,90**	-0,21
Leucine	-0,34	0,47
Lysine	0,15	**0,92**
Proline	0,15	**0,92**
T AMIN	**0,65**	0,49
YAN	-0,36	**-0,63**
Y Veraison	-0,50	**-0,86**
Y Harvest	**-0,90**	0,37
Berry wg	**-0,79**	0,54
N-nitrate	-0,18	**0,98**
Phosporous	-0,97	-0,18
Magnesium	**-0,83**	-0,44
Potassium	0,54	**0,83**
Calcium	0,20	**-0,87**
Color Int	-0,30	0,56
Complexity	0,27	**-0,95**
Body	**0,60**	0,17
Sweetness	**0,60**	0,17
Unctuosity	-0,30	**0,56**
Alcohol E	-0,30	**0,56**
Acid E	-0,21	**-0,95**
Equilibrium	**0,62**	**0,77**
PAI	**0,92**	-0,29

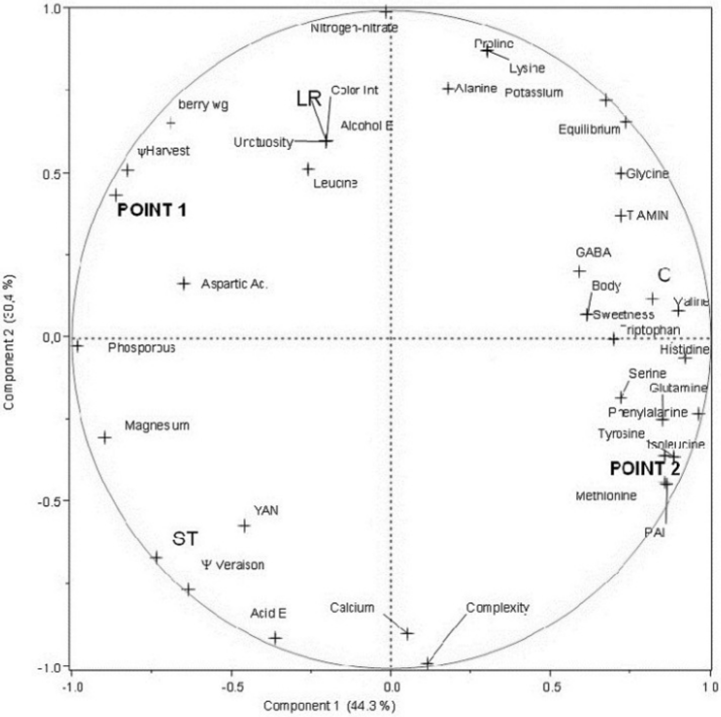

Figure 1. Biplot presentation of principal component analysisof variables selected from must analysis, wine taste, petiole nutrient contents and stem water potential at veraison and hervest. PCA includes Point and canopy management effects.

Conclusions

Under rainfed Somontano conditions nitrogen does not play a prominent role in yield and wine quality of Tempranillo grapes. Canopy management treatments has a strong effect on plant water status, plant nitrogen content, must and wine organic nitrogen composition and wine profile. Canopy treatments could be clearly distinguished by sensory evaluation. The yeasts synthesize fusel alcohols from amino acids and these alcohols produce acetate esters. Fermentative aromas derived from amino acids presents in must could be manipulated by canopy treatments. So, canopy management strategies should be considered a priority on technical research for improving wine quality and to promote terroir wine style.

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USE OF IMAGE ANALYSIS FOR EVALUATION OF FRUIT TREES GROWTH UNDER DIFFERENT NUTRIENT STRATEGIES: PRELIMINAR RESULTS

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Evaluation of growth in fruit trees is essential for the study of biomass production, particularly in research on nutrient and water response. Usually, on multiannual assays, several measures are made to determinate canopy growth, as trunk diameter or section, leaf and pruning weight, light interception by canopy or growth of tagged shoots, all of them with the aim to avoid plant destructive techniques. Several authors have shown the feasibility of image analysis for biomass quantification for different purposes (Campillo et al., 2011; Lu et al., 2005; Sedar and Demirsoy, 2006). The objective of this piece of work is to demonstrate the feasibility of image from digital camera for tree growth evaluation under different nutrient treatments.

Materials and methods

A two year field experiment (2011-2012) was conducted on peach (*Prunus persica* (L.) Batsch. cv. Fortune®) grafted on Cadaman® planted in 1 m^{-3} containers in Lleida region (Northeast Spain). Three nutrient treatments were applied along growth season: NUT100 (complete Hoagland's solution, Hoagland and Arnon, 1950), NUT50 (half Hoagland's solution), NUT25 (one quarter Hoagland's solution). All treatments were irrigated with ETc calculated from data of field weather station, according to Penman-Monteith method (Allen et al 1993). Capacitance probes (ECH2O-10HS, Decagon Devices Inc., Pullman, Washington, USA) were installed in containers for soil moisture monitoring near field capacity. Plots of 3 control trees for each nutrition treatment were arranged on a complete 3 replications block design. Tree whole growth was measured in one tree per plot each year using trunk sectional area at the beginning and at the end of growth. At the end of vegetative growth period (midsummer) canopy lateral projection area was measured using digital camera images, same trees were defoliated on autumn and also an image was taken for whole tree measure of shoots length. For these purposes it was used a reflex camera (Nikon 300s with Zoom lens Nikkor 18-105 mm f/3.5-5.6G, Nikon, Japan) and specific software (Photoshop CS5, Adobe Systems, USA; ImageJ, NIH, USA). Trees were excavated and fractioned in aboveground and root system parts. Leaves, wood and roots were dried to constant weight at 65ºC temperature. Statistical analysis of data was carried out using the SAS-STAT package (SAS®, Version 9.2. SAS Institute Inc., Cary, NC, 1989-2009).

Results and Discussion

Full nutrient solutions, as Hoagland solution, improved vegetative growth on whole tree. In fact, results suggest that the half concentration solution was sufficient to achieve high tree growth rates on young peach trees (Table 1). The application of non-limiting nutrient solutions stimulated aboveground growth versus root growth. This phenomenon might be due to a minor development of the root system in a nutrient-rich environment. Assessment of tree growth by using image techniques was feasible and, in addition, presented many advantages compared to common techniques used in fruit tree research (Table 2).

Table1. Effects of Nutrient treatments on wood biomass (AB), Root biomass (RB), Leaf biomass (LB), Total biomass (TB) – all as dry biomass- , Area of lateral image of canopy (CA), Trunk cross sectional area (TCS), trunk diameter increase (DI), total tree framework length (LS), ratio canopy /root (A/R) and annual shoot growth (LSY) . Values followed by different letters indicate significant differences according to Tukey HSD test (P <0.05). The ANOVA model significance is shown as p>F, ns indicates no model significance.

	NUT-100	NUT-50	NUT-25	Significance p>F
AB (kg)	1.32 a	1.26 ab	1.12 b	0.02
RB (kg)	1.26	1.30	1.13	ns
LB (g)	375.2 a	351.6 ab	313.5 b	0.005
TB (kg)	2.96 a	2.93 a	2.55 b	0.03
CA (m^2)	0.90 a	0.88 a	0.78 b	0.01
TCS (mm^2)	1048.9 a	964.2 ab	869.1 b	0.001
DI (cm)	14.80	16.09	13.14	ns
LS (m)	30.04 a	28.23 b	27.58 b	0.03
A/R	1.21 a	0.97 b	1.05 b	0.04
LSY (m)	23.5 a	20.12 b	18.6 c	0.001

Table2. Pairwise correlations between all growth variables. Results in bold indicate significance at probability level p< 0.05.

	AB	RB	LB	TB	CA	DI	TCS	LS
RB	0.25							
LB	**0.77**	0.18						
TB	**0.67**	**0.87**	**0.57**					
CA	**0.68**	0.08	**0.61**	0.39				
DI	**0.54**	0.39	**0.48**	**0.62**	0.27			
TCS	**0.55**	0.37	**0.50**	**0.61**	0.28	**1.00**		
LS	0.03	0.26	0.16	0.20	0.33	0.16	0.18	
LSY	**0.61**	0.32	**0.71**	0.49	**0.68**	0.32	0.43	**0.83**

Conclusions

The use of full nutrient fertigation solutions improves tree growth and offers the opportunity to increase orchard yield precocity. However, full nutrition solution increases the relationship between aboveground biomass and root biomass. The results of tree images analysis shows that this method provides higher accuracy compared with common methods and could provide higher interpretation capacity for interpretation of experimental results.

Acknowledgements

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NITROGEN TRANSFORMATION DURING ANIMAL WASTE COMPOSTING

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One of the characteristic of cattle and lamb feedlot is the generation of waste which, if not treated and managed correctly, may endanger the environment. Therefore, before reaching its final destination, this waste must pass through a stabilizing process. Among the various methods for treating it is the composting. During the process of composting there may be losses of nitrogen. This losses may have a negative impact by decreasing the amount of nutrients and consequently reducing the quality of the compost besides increasing environmental issues. Losses of nitrogen during the process of composting may happen due to the volatilization of NH_3, leaching and denitrification (Bernal, 2009). The aim of this study was to evaluate the dynamics of the nitrogen during the process of beef cattle and lamb ranching waste composting.

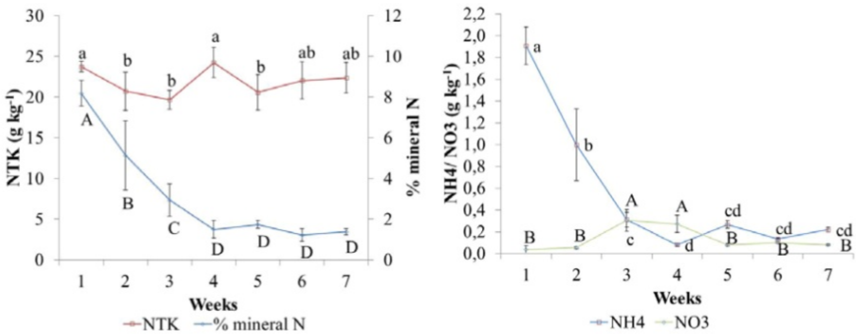
Materials and Methods

This study consisted in the use of waste taken from cattle and sheep feedlots. The breeding stock was subjected to a diet consisting of 60% of forage and 40% of concentrated. The wastes were submitted to a composting experiment in a covered shed with impermeable floor in proportions of 1:1, considering the dry mass weight of the material. Four piles of natural matter, 487 kg of each, were installed. In this way, the piles consisted of 307 kg of cattle manure and 180 kg of sheep litter. The irrigation, turning, and collecting of samples for the analysis of the piles were performed in a weekly base. The moisture value of the piles was maintained in 60%.Through the samples values of total Kjedahl nitrogen (NKT), ammonium (N-NH_4^+) and nitrate (N-NO_3^-) were determined. For the NKT analysis the methodology proposed by Malavolta et al. (1989) was used. In order to determine the ammonium and nitrate the samples were subjected to extraction with KCl 2 mol L^{-1} and subsequent distillation with the addition of MgO in order to obtain N-$NH_4^{+.}$ Devarda alloy was used in order to obtain the N-NO_3^-, according to the adapted methodology from BREMNER; KEENEY (1965). For the evaluation of the results the analysis of comparison of means through LSD in 5% significance was performed.

Results and Discussion

There was significant reduction in the percentage of N mineral during the first six weeks of the composting process (Figure 1A), mainly due to the decrease of the values of N-NH_4 (Figure 1B).

The small reduction of NKT during the process of composting (from 23.72 g.kg^{-1} to 22.34 g.kg^{-1}) can be explained by the losses of carbon in the form of CO_2, which caused the concentration of the element (BRITO et al., 2008). However, despite the small variation on the values of NKT, it is possible to note that the N-NH_4 in the beginning of the process was not converted into N-NO_3^- by nitrification (SZANTO et al., 2007), but it was probably immobilized or transformed into organic matter (LU et al.,2013), and that explains the small variation on the values of NKT. Still regarding the stability of the concentration of NKT, the possible losses of N in the form of of N-NH_3 cannot be discarded. Lu et al. (2013), while evaluating swine manure and rice straws composting, noted an increase on the values of N-NH_4, specially during the thermophilic phase due to the high temperature, the high concentration of N-NH_4 and the increase on the value of pH. Temperatures higher than 50°C were registered from the second to the fourth week and the increase of pH was also noted during the process, thus explaining the reduction on the concentration of N-NH_4. These losses of nitrogen occur mainly through the volatilization of N-NH_3 (BUSTAMANTE et al., 2008) and through the aeration on the process of tilling the piles (GÓMEZ-BRANDÓN et al., 2008; GUARDIA et al., 2008). The generation of nitrate lasted until the sixth week (Figure 1B). Szanto et al. (2007) verified when composting straw-rich swine manure that the NO_2- and NO_3- did not accumulate but probably were converted into N_2 and N_2O by denitrifying bacteria. It appears, therefore, there was a reduction of 83% of the N mineral during the process of composting, which may have been converted into organic matter (remaining in the final product) or lost though volatilization.



The use of equal low case letters represent equal means for the NKT and NH, while equal capital letters represent equal means for the % of N mineral and NO_3 on 5% of significance by the LSD test. Figure 1 – (A) Concentration of NKT and N mineral, (B) concentration of ammonium and nitrate during the period of 12 weeks of composting.

Conclusions

There is a decrease in the % of N mineral during the process of composting.

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THE AMOUNT OF MINERAL NITROGEN IN ORGANIC SOILS OF GRASSLANDS IN POLAND

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As a result of organic matter mineralisation, soils are enriched with nitrogen available for plants in a form of ammonium ions (N-NH4) and nitrates (N-NO3). The process is favourable for the growth and yielding of crop plants; it may, however, pose environmental risk associated particularly with water pollution by nitrogen (in case mineral nitrogen is accumulated in soils in amounts exceeding the uptake by plants). The risk is especially high in grassland soils of peat origin since they are very rich in organic matter. Mineralisation of such soils has already been elucidated in agri-environmental aspects, there is still, however, a need of more advanced studies (e.g. with respect to conditions, parameters and effects of mineral nitrogen release from soil organic matter). This paper is an attempt to meet these needs. Its aim was to make a quantitative assessment of the content of mineral nitrogen in organic soils of grasslands in Poland and its seasonal dynamics.

Materials and Methods

Studies were carried out in the years 2008-2012. Soil samples were collected from over 160 permanent monitoring sites selected in organic soils covered by permanent grasslands in Poland. This monitoring was established by the National Agro-Chemical Station (KSCh-R) and regional agro-chemical stations in cooperation with the Institute of Technology and Life Sciences (ITP) in Falenty. In the beginning in 2008, soil characteristics were analysed in particular sampling sites and the content of soil organic matter was determined as a loss on ignition at a temperature of 550°C for 7 hours.

Soil samples for the determination of inorganic nitrogen were collected twice a year: in early spring before application of nitrogen fertilisers (i.e. before or just after start of vegetation) and in autumn after plant harvest from a layer of 0 – 30 cm. The content of nitrate-nitrogen (N-NO3) and ammonium-nitrogen (N-NH4) was determined in soil samples according to PN-R-04028:1997: Chemical and agricultural soil analysis. Methods of soil sampling and determination of nitrate and ammonium ions in mineral soils.

Based on obtained results, the resources of inorganic nitrogen (N-NO3 +N-NH4) in 30-cm thick soil layer i.e. the amount of inorganic nitrogen accumulated in that layer per 1 ha of grasslands (in kg N ha-1) were estimated for the period 2008-2012. Resources of inorganic nitrogen in mineral soils were calculated as a product of inorganic nitrogen (NO3-N + NH4-N) content in soil and its bulk density.

Bulk density of organic soils was determined using the equation:

$y = -0.000006x3 + 0.001x2 – 0.0623x + 1.6346$; R2 = 0.9489

where:

y – bulk density, kg dry mass·dm-3

x – organic matter content, %.

The relationship was derived from statistical analysis of 186 pairs of data (bulk density and organic matter content in soil) obtained by various authors.

Results and Discussion

Mean concentration of mineral nitrogen (Nmin) in organic meadow soils was 51 mg N·kg-1 in spring and 55 mg N·kg-1 in autumn of the years 2008-2012 – Table 1. The concentration of nitrate-nitrogen was higher in autumn than in spring while the reverse was true for ammonium-nitrogen concentrations. The resources of inorganic nitrogen in 30-cm soil layer were from ca. 117 kg N·ha-1 in spring to more than 131 kg N·ha-1 in autumn during the study period.

In particular study years, the content of inorganic nitrogen in organic soils increased between spring and summer and decreased between autumn and spring of the next year. In the summer periods, the resources of Nmin increased by 7.9-17.9 kg N·ha-1 and in the winter time they decreased by 8.9 -16.2 kg N·ha-1. Meteorological conditions largely affected the range of changes of inorganic N resources. There were significant correlations between soil richness in nitrogen in spring and autumn and mean precipitation and air temperature ($p< 0.05$, $n=10$, $R=0.87$ and 0.85, respectively). Recorded decrease in the amount of inorganic nitrogen in organic soils during winter half-year (after vegetation period) was undoubtedly associated with nitrate leaching to ground waters.

Conclusions

As a result of organic matter mineralisation in organic grassland soils in Poland, the resources of inorganic nitrogen are released in amounts exceeding plant capability of their uptake in the vegetation period. Nitrogen not absorbed by plants is a potential source of water pollution. To ensure sustainable management of grasslands situated on organic soils, there is a need of elaborating methods to control the efficiency of mineralisation of soil organic matter.

Table 1. Mean content of nitrate-nitrogen ($N\text{-}NO_3$), ammonium-nitrogen ($N\text{-}NH_4$) and the resources of inorganic nitrogen N_{min} ($N\text{-}NO_3 + N\text{-}NH_4$) in 0-30 cm layer of organic meadow soils in various years

Year	Spring				Autumn			
	n	$N\text{-}NO_3$, mg N•kg^{-1}	$N\text{-}NH_4$ mg N•kg^{-1}	N_{min}, kg N•ha^{-1}	n	$N\text{-}NO_3$, mg N•kg^{-1}	$N\text{-}NH_4$ mg N•kg^{-1}	N_{min}, kg N•ha^{-1}
2008	162	13.4	34.1	113.7	162	22.6	31.9	129.6
2009	161	13.8	35.0	113.4	162	21.1	36.3	131.3
2010	164	13.1	38.6	122.3	165	18.6	37.0	131.4
2011	162	8.6	45.4	122.5	163	12.3	39.9	130.4
2012	162	9.6	42.2	116.9	162	18.9	35.2	126.6
Mean from the years 2008-2012		11.7	39.1	117.8		18.7	36.1	129.9

TECHNOLOGICAL QUALITY OF WINTER WHEAT DEPENDING ON NITROGEN APLICATIONS

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Nitrogen is one of the most important agronomic factors affecting the yield and technological quality, especially protein content and gluten amount as well as gluten quality. The most important from an economical and environmental point of view is nitrogen use efficiency, defined as the grain dry matter, protein and gluten content of supply of available nitrogen from the soil and fertilizer. The effectiveness of nitrogen depends on many factors mainly the term of N applications and the variety. The aim of this study was to determine the effects of winter wheat cultivars on effectiveness of nitrogen fertilization on the grain yield and content of protein and gluten content in winter wheat grains.

Materias and Methods

Field experiments were carried out at the experimental station Radostowo ($\varphi = 53^o59'$, $\lambda = 18^o45'$) in Poland in 2008/9, 2009/10 and 2010/11. Ten Polish cultivars were grown at two rates of N fertilization. Each experiment used a split-plot design in which N treatment was randomized on main plots, cultivar was randomized on the sub-plots and each treatment was replicated four times. The first N (N1) fertilization treatment was intended to replicate commercial practice and equalled 160 kg N ha^{-1}. The second fertilization (N2) was calculated using measurements of the amount of mineral N in the soil in the beginning on vegetation period. The first N treatment was applied in two terms (tillering and shooting) the second in three terms (in tillering, shooting and heading) during vegetation period of winter wheat. All N fertilization was applied as granules of ammonium nitrate and each split was applied on the same calendar date for the 10 cultivars. The level of N fertilizer applied was kg N ha−1for 2008/9 and 2007/8, respectively. The complex agricultural suitability of soil was wheat very good. The cultivars within an experiment were sown at the same seed rate 450 seeds per m^2. All other crop inputs including pest, weed and disease control, potassium, phosphate and sulphur fertilizers were applied at levels to prevent non-N nutrients or pests, weed sand diseases from limiting the yield.

Results and Discussion

No significant differences were found between two ways of N applications in the grain yield. The highest grain yield was found in 2009 and 2011, the lowest in 2010. Difference in yield levels between the 2009 and 2010, and 2011 equaled 1,01 t ha^{-1}.and 0,49 t ha^{-1}. 2009 was the year that in significant differences in grain level between the cultivars. The highest yield was found for the Satyna, Ostroga and Nateja cultivars the lowest for Izyda and Muszelka (tab. 1).In the 2010 was found the interaction between cultivar and nitrogen application. On the N1 the highest yield gave Kohelia and Markiza, the lowest Izyda, Batuta, Ostroga, Satyna. The N2 method of nitrogen application did not cause differences in the level of yielding varieties (tab.1). The highest gluten content was found in Tonacja, Ostroga and Nateja and the lowest in Izyda, Markiza and Satyna cultivars. The differences between Tonacja and Markiza equal 3,6%. In was also found interaction. On the first N applications the highest amount of gluten was found in Tonacja, Satyna and Kohelia cultivar however, second N2 resulted in Ostroga, Tonacja, Batuta, Nateja giving the highest gluten content (tab.2). The highest protein content was found in Fregata, Izyda, the lowest in Tonacja and Kohelia cultivar. On the first N applications the is no differences in protein content between the cultivars but on the second N2 the highest amount of protein was found in Fregata and Izyda cultivar (tab.3).

Table 1. Grain yields of winter wheat cultivars depending on N fertilization. No - -not significant

Year	Nitrogen	Cultivar										Average
		Tonacja	Muszelka	Satyna	Markiza	Nateja	Ostroga	Batuta	Izyda	Fregata	Kohelia	
2009	N1	10.1	9.6	10.3	9.8	10.1	10.0	9.6	9.6	9.8	10.0	9.9
	N2	9.6	9.2	10.1	9.5	10.0	10.3	9.9	9.7	9.6	9.6	9.7
	average	9.8	9.4	10.2	9.7	10.0	10.1	9.8	9.6	9.7	9.8	
	Nitrogen (a) no; Cultivar (b) .0.481; a x b = n.o											
2010	N1	8.80	8.98	8.15	9.03	8.98	8.25	8.40	8.38	8.93	9.15	8.71
	N2	8.75	8.62	9.07	8.97	8.87	9.08	8.83	9.00	8.80	8.73	8.87
	average	8.78	8.80	8.61	9.00	8.93	8.67	8.62	8.69	8.87	8.94	
	Nitrogen (a) no; Cultivar (b) .n.o; a x b = 0.497											
2011	N1	9.46	9.25	9.53	9.35	9.43	9.45	9.13	9.00	9.20	9.07	9.28
	N2	9.42	9.53	9.38	9.22	9.10	8.87	9.65	9.53	9.37	9.30	9.34
	average	9.44	9.39	9.46	9.28	9.27	9.16	9.39	9.27	9.28	9.31	
	Nitrogen (a) no; Cultivar (b) .0.781; a x b = n.o											
2009-2011	N1	9.45	9.28	9.33	9.39	9.50	9.23	9.04	8.99	9.31	9,41	9.29
	N2	9.26	9.12	9.52	9.23	9.32	9.42	9.46	9.41	9.26	9.21	9.32
	average	9.36	9.20	9.42	9.31	9.41	9.33	9.25	9.20	9.28	9.31	
	Nitrogen (a) no; Cultivar (b) .n.o; a x b = n.o											

Table 2. The effect of nitrogen application on gluten content in winter wheat cultivars (%)

Fertilizer	Cultivars										Average
	Tonacja	Muszelka	Satyna	Markiza	Nateja	Ostroga	Batuta	Izyda	Fregata	Kohelia	
N1	31.2	26.7	31.1	26.4	29.7	28.8	25.5	28.7	29.4	31.1	28.7
N2	31.9	30.9	26.1	29.7	31.3	33.1	31.7	27.3	29.1	26.1	29.7
Average for cultivars	31.6	28.8	28.1	28.0	30.5	31.0	28.6	28.0	29.2	28.6	
LSD for; Nitrogen (a) Cultivar (b)	r.n. 1.958										
interaction; I x II	0.412										

Table 3. The effect of nitrogen application on protein content in winter wheat cultivars (%)

Fertilizer	Cultivars										Average
	Tonacja	Muszelka	Satyna	Markiza	Nateja	Ostroga	Batuta	Izyda	Fregata	Kohelia	
N1	11.5	11.2	11.4	11.6	11.1	10.8	10.9	11.6	11.7	10.7	11.3
N2	11.9	12.7	12.5	12.1	12.5	13.0	12.0	13.3	13.6	12.5	12.6
Average for cultivars	11.7	12.0	12.0	11.9	11.8	11.9	11.8	12.5	12.7	11.6	
LSD for; Nitrogen (a) Cultivar (b)	r.n. 0.770										
interaction; I x II	1.547										

Conclusion

Presented studies indicate that there is a differentiation in the use of nitrogen by variations in both the size of grain yield and protein content and quantity of gluten.

CONTRIBUTION OF DAIRY RATION COMPONENTS TO NITROGEN IN MILK, MANURE, CROPS AND ENVIRONMENTAL NITROGEN LOSS

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Of the total N consumed by dairy cows, a general range of 20 to 35% is secreted in milk, and the remaining N is excreted in manure. Feed N use efficiency (FNUE, the percent of total N consumed that is secreted as milk N) and the relative amount of N excreted in urine and feces can be highly influenced by feed management, such as the type and amount of N consumed. Dairy rations containing approximately 26.2 g N/kg of total ration dry matter (DM) maximize milk production and FNUE (Broderick, 2003 and 2009). As ration N increases above requirement, FNUE declines, excess N consumption is excreted as urea in urine, and this urea hydrolyzes and is lost as ammonia (NH_3). Broderick (2003) found that an increase in ration N from 24.2 to 29.4 g/kg decreased FNUE from 31 to 25%, and increased the excretion of urinary N from 23 to 35% of N consumed. Misselbrook et al. (2005) determined that dairy slurry from a high N (31.0 g/kg DM) ration emitted 2 to 5 times more NH_3 after its application to soil compared to slurry from a low N (21.8 g/kg DM) ration. For many dairy herds, improved feed management including feeding rations balanced in energy and crude protein, can enhance milk production and FNUE, and reduce manure N excretion and N loss. Yet these practices may also reduce manure N availability to crops and decrease yield. For example, Powell et al. (2006) determined that feces from cows fed a high N (29.4 g/kg DM) ration applied to soil resulted in 35 to 40% more crop N uptake and yield than feces from cows fed a low N (24.2 g/kg DM) ration. To better understand the integrated nature of N use and loss in dairy production systems, a series of experiments was designed to quantify the relative amounts of N from individual ration components secreted in milk, excreted in urine and feces, available to crops after manure application to soil, and lost as NH_3, nitrate (NO_3^-) and nitrous oxide (N_2O).

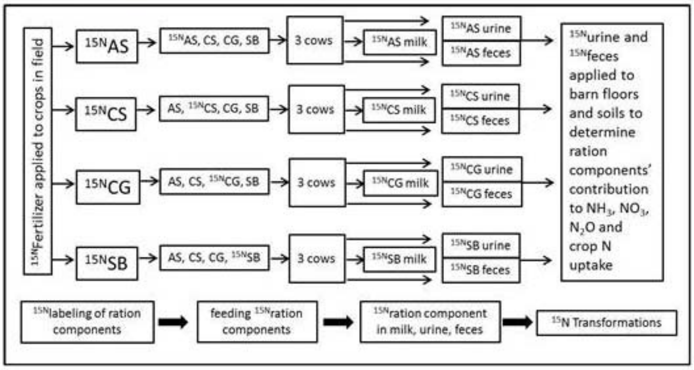
Materials and Methods

Alfalfa silage (AS), corn silage (CS), corn grain (CG) and soybeans (SB) were grown in the field with applications of 15N-enriched fertilizer (Fig 1). Each 15N-enriched feed was then fed individually (as components of total mixed rations) to 12 mid-lactation cows (3 cows per ration component). The mass of milk, urine, and feces produced by each cow were recorded and sampled the day before and 3 days after the 4-day 15N feeding period. Collected feces and urine were composited within each 15N enriched ration and frozen for later use in laboratory, greenhouse and field trials to assess each component manure N availability to crops and environmental N loss.

Results and Discussion

Concentrations of fiber and N in the ration components fed to the cows are provided in Tab. 1. Highest 15N incorporation in the field was achieved with CS and CG, and lowest with AS and SB (due to 15N dilution by the atmospherically-fixed N by these legumes). The experimental methods used to ensile the 15N-enriched corn and alfalfa, the milling of corn grain and the extraction of soybean grain to produce soybean meal did not appear to impact ration intake, milk production or N excretion by dairy cows, as indicated by the narrow range (and non-significant differences among rations containing the 15N-enriched components) in dry matter intake, N intake, milk production, FNUE and N excretion in urine, urea and feces (Tab. 2)

Fig. 1. ^{15}N labeling of dairy ration components, milk, urine and feces, and use of 15N-labeled manure to study N transformations



Tab. 1. Concentrations of neutral detergent fiber (NDF), total N (TN) and ^{15}N in ration components fed to dairy cows

Ration component	NDF (g/kg DM)	TN (g/kg DM)	^{15}N (atom %)
Alfalfa silage	443	29.8	0.730
Corn silage	448	11.3	1.857
Corn grain	88	11.8	2.040
Soybean meal	92	57.4	1.385

Tab. 2. Range dry matter intake (DMI), N intake (NI), milk production, feed N use efficiency (FNUE) and N excretion by 12 cows fed rations containing 15N-enriched components

Parameter	Range (min. – max.)
DMI (kg/cow/d)	22.8 – 23.6
NI (g/cow/d)	591 – 613
Milk (kg/cow/d)	24.8 – 28.5
FNUE (%)	22 – 25
Urinary N (g/cow/d)	231 – 233
Urea-N (g/cow/d)	175 – 193
Fecal N (g/cow/d)	154 - 174

Conclusions

The synergistic nature of feed N and manure N management on dairy farms necessitates integrated feed-milk/manure-soil/crop-environmental N loss experiments. Ration components were enriched in the field with the stable isotope ^{15}N and fed to lactating cows. Cows responded similarly to feeding ^{15}N-enriched and non-enriched ration components. Feces and urine from each ^{15}N enriched ration component will be applied to emission chambers that simulate barn floors and field soil surfaces, and ^{15}N concentrations in NH_3, NH_4, NO_3 and N_2O will be measured. Manure-soil incubations, greenhouse and field trials will be conducted to determine each ration component's manure N contribution to crop N uptake.

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NITROGEN DYNAMICS IN THE RHIZOSPHERE

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The fate and dynamics of organic N in the rhizosphere has been the focus of a large number of studies (Kuzyakov and Xu, 2013). Plants have been shown to both, exude amino acids from roots, and have the capacity for amino acid uptake (Jones and Darrah, 1994; Jones et al., 2009). However, determining the fate of amino acids exudates, and N dynamics, in the rhizosphere, has been limited by a lack of non-destructive sampling methods (Oburger et al., 2013). In a novel, *in situ* approach, microdialysis probes were used to monitor N dynamics in the rhizosphere of maize seedlings.

Materials and Methods

Microdialysis probes were inserted into a rhizotube filled with an agricultural Eutric cambisol. Maize seedlings were grown, over a 68 hour period, in the rhizotubes, so that the growing root passed directly over the membrane of the microdialysis probe. The probes were perfused with de-ionized water, at a rate of 5 µl min^{-1}, and the dialysate was sampled over a four hour time period. Samples were then chemically analysed for total amino acids, ammonium and nitrate. In addition, the fate of amino acid inputs in the same soil was investigated using an isotopically labelled amino acid mixture, at a range of concentrations representing that which could be found in the rhizosphere (1, 10, 100, 1000 µM). A 300 µl aliquot of amino acid mixture of each concentration was applied to 1 g (DWE) of field moist soil and incubated, at 20°C, for 1, 5, 10, 30 and 60 min. The same treatments were applied to sterilised soils and the results used as a control to determine the significance of abiotic mechanisms. Carbon dioxide traps were used to capture any $^{14}CO_2$ produced during mineralization and ^{14}C-labelled amino acids remaining in soil solution was collected using a centrifugation drainage technique (Hill et al., 2008). Measurements of ^{14}C, in the CO_2 traps and soil solutions, were performed with liquid scintillation counting. Net amino acid efflux from the roots of maize seedlings, grown in a sterile nutrient solution was also investigated using a nano sampling technique coupled with spectrofluorometric analysis.

Results and discussion

A significant concentration of amino acid exudation, which peaked 1-2 cm from the root tip, was determined using the nano sampling method. However, results from the microdialysis sampling showed the concentration of amino acids in the soil solution remained relatively constant throughout the experiment. The lack of amino acid spike in the soil solution, due to root exudates, may be explained by the fact that the depletion of the isotopically labelled amino acid mixture from the soil solution was extremely rapid. Figure 1 shows that only 10% of the 10 µM treatment remaining in the soil solution after 1 minute. Nitrate concentration decreased following the growth of the root past the probe, which is likely due to plant uptake.

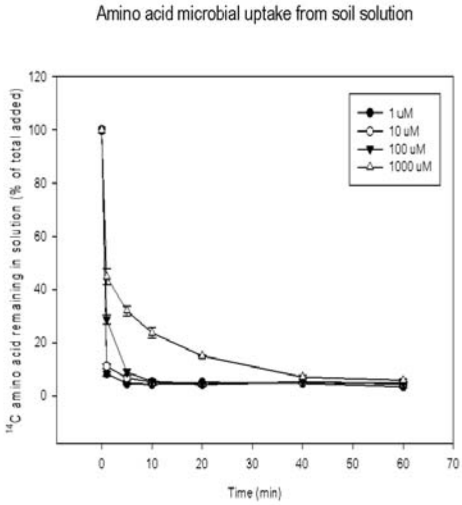

Fig. 1. Amount of 14C-label remaining in soil solution after the addition of a 14C-labelled amino acid pulse (1µM – 1000µM) to an agricultural soil. Values represent means ± SEM (n = 3).

Conclusions

This study demonstrates the feasibility of microdialysis sampling within the rhizosphere and suggests that LMW DON exuded by roots, is rapidly cycled by carbon limited microorganisms in the rhizosphere.

Acknowledgements

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GROWTH AND NITROGEN RECOVERY IN THE ABOVE-GROUND BIOMASS OF ELEVEN SELF-RESEEDING ANNUAL LEGUMES GROWN IN A RAINFED OLIVE ORCHARD

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Most of the traditional dry-farmed olive orchards of the NE of Portugal are planted in shallow soils on sloping terrain. The olive yields obtained are usually low, due to the severe environmental constraints under which these orchards are grown. Growing olive trees in such hard conditions, may recommend the management of the orchards as organic, a low-input farming system from which the farmer profit can arise from the appreciation of the price of the olive oil. The natural soil fertility of these orchards is usually very low, being nitrogen the most limiting nutrient to the growth of the trees (Rodrigues et al., 2011). Objectively, to manage these orchards as organic and to maintain the soil fertility and the tree nutritional status in an acceptable level, there is a single option: the introduction of legume species as cover crops. Legumes can access atmospheric N_2, due to the symbiotic relationship that they can establish with nitrogen-fixing bacteria (Russelle, 2008). In this work eleven self-reseeding annual legumes were introduced in a rainfed olive orchard in order to test their suitability to be used as cover crops. The legume species/varieties were grown as pure stand and managed without grazing, since currently the farmers of the region are not raising animals. Data on dry matter yield, nitrogen recovery and persistence of the sown species are presented.

Materials and Methods

The field trial took place in Mirandela (NE Portugal) in an olive orchard of ~20 years old. The olive orchard is installed in a Leptosol loamy textured, pH acid and low organic matter content. The climate is of Mediterranean type. In the autumn of 2009 were sown the following eleven species/varieties: *Ornithophus compressus* L. cv. Charano, *O. sativus* Brot. cvs. Erica and Margurita, *Trifolium subterraneum* L. ssp *subterraneum* Katzn. and Morly cvs. Dalkeith, Seaton Park, Denmark and Nungarin, *T. resupinatum* L. ssp *resupinatum* Gib and Belli cv. Prolific, *T. incarnatum* L. cv. Contea, *T. michelianum* Savi cv. Frontier and *Biserrula pelecinus* L. cv. Mauro. Seed rates varied according to that recommended for each species/varieties. The area of the individual plots was 49 m^2 (3 replications per treatment). Dry matter yield was determined in May, by cutting the biomass of a grid of a 0.25 m^2. The samples were oven-dried at 70 ºC and ground. Nitrogen concentration in the dried samples was determined by a Kjeldahl procedure. Nitrogen recoveries were estimated from tissue nitrogen concentrations and dry matter yields.

Results and Discussion

B. pelecinus cv. Mauro showed very low nitrogen recoveries since the first year due to a very low rate of seed germination. Nitrogen recovery by *T. incarnatum* cv. Contea exceeded 450 kg N ha^{-1} in the four growing seasons. The cvs. of *T. subterraneum* yielded nitrogen recoveries close to 200 kg N ha^{-1} (cvs. of short growing cycle, Nungarin and Dalkeith) and close to 300 kg N ha^{-1} (the longer growing cycle cultivars, Denmark and Seaton Park). The cvs. of *O. sativus* produced higher nitrogen recovery values (~300 kg N ha-1) than *O. compressus* cv. Charano (264 kg ha^{-1}). *T. michelianum* exceeded 300 kg N ha^{-1}, whereas *T. resupinatum* just surpassed 200 kg N ha^{-1}. Most species/varieties reached the higher nitrogen recoveries in the first growing season (spring of 2010). The fourth growing season was also good for several species/varieties whereas the third was the worst for all of them.

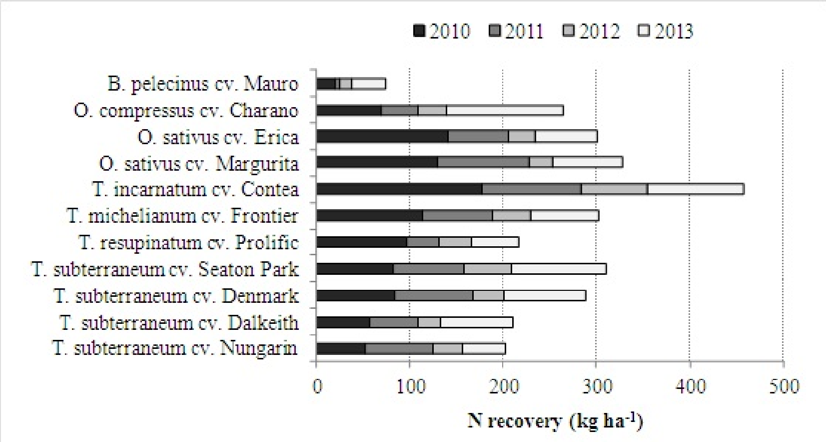

Figure 1. Nitrogen recovery in the above-ground biomass of eleven legume species/varieties grown in pure stands in a rainfed olive orchard during four consecutive growing seasons.

The first growing season was highly favorable to the establishment and growth of the sowed legumes species. Also the absence of grazing allowed the formation of a great seed bank from the first year, leading to a good establishment of the swards in the second growing season. The climate conditions in the second year were not so favorable, with less rain during spring. In the third year, the first big problem occurred. It was observed a false break (germination-inducing a rainfall event late in August followed by death from severe drought during September). The false break associated to the increase in soil fertility, as a result of two consecutive seasons of the growth of the legume species, has created an opportunity for weeds. In the third year the sward appeared dominated by Rattail Fescue (*Vulpia myuros*). At the end of the third year it was feared the end of the sown legumes. The autumn of the fourth year was again very favorable for legume seeds germination and plant growth. In spring 2013, the dominance of most of the sown legume species/varieties was restored. In summary, most species/varieties are able to persist for long-term in this environment. Farmers should chose the most appropriate for rainfed or irrigated orchards taken into account their N fixing capability and the length of their growing cycles.

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MONITORING NITROGEN NUTRITIONAL STATUS OF VEGETABLES IN THE SOCIAL GARDEN OF POLYTECHNIC INSTITUTE OF BRAGANÇA, NE PORTUGAL

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Urban agriculture is a global phenomenon. In developing countries urban agriculture has had crucial importance in alleviating problems of extreme poverty of the populations of the larger cities. In developed countries has been particularly important during periods of economic depression. Nowadays, the urban agriculture in developed countries is increasing, aimed more at leisure, sports and recreation of the urban dwellers. These spaces are also emphasizing environmental education, by implementing environmentally friendly agricultural practices. In the case of Portugal, urban agriculture is frequently regulated by norms, implicitly or explicitly, similar to those established for organic farming. In the urban agriculture project of the Polytechnic Institute of Bragança (IPB) the participants are also encouraged to use sustainable farming practices. The fertilizers used are usually farmyard manures and other organic wastes. This work takes part of a larger project that aims to monitor the nutritional status of the plants, the residual mineral nitrogen (N) content in the soil and the contamination of plants and soils with heavy metals. In this report, it will be present results only for the first two goals.

Materials and methods

During 2013 several vegetable species were sampled in the social garden of IPB for analysis. Data from nine species sampled from six gardens each and involving a total of thirteen different gardens were considered. The plants were sampled during the growing season of 2013 following the norms established for each vegetable, regarding the sampling date and the proper tissue to be analyzed (Mills and Jones, 1996; LQARS, 2006). In the autumn, soil was monthly sampled at two depths (0-20 cm and 20-40 cm) in each of the selected gardens. Plant samples were oven-dried at 70ºC, ground and analysed for tissue nutrient concentration. N concentration in plant tissues was determined by a Kjeldahl procedure. The soil samples were frozen after collected. Soil extracts were thereafter prepared by using a concentrated (2*M*) KCl solution and the extracts analysed for NO_3^- and NH_4^+ concentration by UV and visible spectrophotometry.

Results and discussion

N concentration in plant tissues was always close to the lower limit of the sufficiency range for all vegetables (Table 1). The average soil mineral N levels, in the gardens where plant samples were taken, were quite low when compared with values previously recorded for other agro-ecosystems (Magdoff et al., 1984; Rodrigues, 2004a). The concentration of mineral N in soil in the sampled gardens varied between 2.9 and 8.9 mg kg^{-1} (table 2). The gardeners fertilize their crops with farmyard manure that has been freely provided by IPB, and in lesser extent with other organic wastes.

This kind of manure usually presents N concentrations ranging between 11 to 22 mg kg^{-1} and C/N ratios between 10 and 20 (Rodrigues, 2004b; Rodrigues et al., 2006). These manures release N very slowly, being the effect on vegetation modest (Rodrigues et al., 2006), which may explain the low N concentrations in plant tissues and the low residual mineral N in the soil. The small differences that were observed in residual soil mineral N are certainly the result of the greater dedication of some gardeners. It was observed a close relationship between the gardens with great variability in species and the higher soil mineral N levels (the diversity of species grown can be seen as an index of the dedication of the gardeners).

Table 1. Leaf N concentration (mean±mean confidence intervals), sufficiency N ranges for the most commonly grown vegetables in IPB gardens and soil mineral-N (0-40 cm soil layer) in the gardens where each of the vegetables was grown.

	Leaf N concentration g kg^{-1}	Sufficiency range* g kg^{-1}	Soil mineral-N mg kg^{-1}
Strawberry	19.9±2.6	21-40	6.1±0.3
Carrot	21.3±1.0	21-35	6.0±0.4
Lettuce	23.7±1.3	25-50	6.3±0.7
Tall cabbage	29.5±1.7	31-55	6.3±0.6
NZ spinach	28.9±1.7	---	4.2±0.9
Onion	22.8±0.8	45-55	6.0±0.7
Bean	29.9±2.3	30-60	6.3±0.8
Pepper	32.0±2.0	35-50	5.8±0.6
Tomato	31.5±1.3	30-50	6.0±0.7

*Adapted from Mills and Jones (1996) and LQARS (2006).

Table 2. Soil mineral N (0-40 cm soil layer) and vegetables sampled in the corresponding garden.

Garden number	Vegetables	Soil min-N mg kg^{-1}
5	Carrot, lettuce	5.0
6	Strawberry, lettuce, cabbage, NZ spinach	5.6
8	Carrot, lettuce, cabbage, onion, bean, pepper, tomato	5.6
9	Cabbage, onion, pepper, tomato	6.9
11	Lettuce, cabbage, onion, bean, tomato	8.9
15	Strawberry, NZ spinach	5.9
19	Strawberry, carrot	7.1
20	NZ spinach	2.9
22	Carrot, lettuce, bean, pepper	7.6
32	NZ spinach	3.6
40	Onion, bean, pepper, tomato	3.7
62	Carrot, lettuce, cabbage, onion, bean, pepper, tomato	4.8
79	Strawberry, carrot, cabbage, onion, pepper, tomato	6.0

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NITROGEN CONTENT IN ABOVE-GROUND PLANT PARTS AS AN AID TO ESTABLISH MORE ACCURATE FERTILIZER-NITROGEN RECOMMENDATIONS FOR GRAPEVINE

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Nitrogen is the nutrient applied most frequently as fertilizer in annual and perennial crops. In grapevine, nitrogen determines the vigor and yield of vine and several attributes of must quality (Akin et al., 2012; Pérez-Alvaréz et al., 2013). The close relationship between nitrogen application, vine performance and the quality of wine requires the rational use of fertilizer-N. In addition, the excessive use of fertilizer-N may cause environmental damage (Powlson, 1993). The current recommendation systems for vine are usually based on plant analysis complemented by soil testing. These tools are important but insufficient to provide quantified rates of nutrients to apply. To increase the accuracy of the fertilization programs, data on nutrient content and dynamic in plant tissues may be helpful. In this work, nutrient content in different vine parts (leaves, canes, cordons and trunk) and at different dates (from September 14th to November 28th) was determined to understand the fate of the nutrients at the end of the growing season as a mean of increasing precision of the fertilizer recommendation system. In this extended abstract, it will be presented data on nitrogen content and dynamic in those plant tissues and dates.

Materials and Methods

At harvest, on September 14th, the clusters of three vines were cut and separated into rachis, pulp and seeds. Dry matter yield and nitrogen concentration of the different tissues were recorded. The canes were divided into leaves and wood, weighed dry and analyzed for nitrogen concentration. Trunk and cordons were also weighed after dried, and analyzed for nitrogen concentration from sawdust samples and from the outer layer (phloem vessels) of the trunk. The procedure was repeated on October 16th, excluding the clusters that were not present at that time. On November 02nd and 28th the analyses were performed in the woody parts (the leaves had begun to fall). On October 16th, samples of normal (green) and chlorotic leaves taken from a similar position in the canopy were analyzed for nitrogen concentration to infer on nitrogen lost during the senescing process.

Results and Discussion

On September 14th, the leaves were the most nitrogen concentrated tissue (~18 g kg^{-1}), followed by the seeds (~15 g kg^{-1}). The woody vine parts (trunk, cordons and canes) presented low nitrogen concentrations (< 4 g kg^{-1}). Nitrogen concentration in leaves decreased from 18.3 g kg^{-1} on September 16th to 13.3 kg kg^{-1} on October 14th. Nitrogen concentrations in green and chlorotic leaves on October 14th were, respectively, 11.9 and 6.2 g kg^{-1}. Cane nitrogen concentrations increased from September 16th to November 28th. Nitrogen concentration in trunk increased from September until November 02nd. The above-ground parts of a vine contained 84 kg N ha^{-1}, distributed by trunk and cordons, canes, leaves and clusters, respectively in the approx. amounts of 14, 8, 42 and 20 kg N ha^{-1}.

Nitrogen present in the vine at harvest may be lost from the soil/plant system or recycled through remobilization to the perennial structures and taken up from soil in the next season. The schematic view of the process is presented in table 1.

The clusters contained ~20 kg N ha^{-1}, representing 1.4 kg N per ton of fresh fruit. This nitrogen represents an entirely lost from the soil/plant system which should be taken into account in the fertilizer recommendation program. During senescence, nitrogen from leaves may be remobilized to perennial structures or volatilized as NH_3 to the atmosphere. The results here reported indicate that a significant portion is remobilized to woody parts. Nitrogen in fallen leaves undergo mineralization and thereafter nitrogen can be taken up by roots or lost from soil by NH_3 volatilization or NO_3^- leaching and denitrification. The importance of each component may depend on environmental conditions and soil management techniques influencing nitrogen use efficiency. Nitrogen in prunings may be lost from the system or recycled in it depending if prunings are removed and used as firewood or left on the ground as an organic residue. The advisory system should take all those aspects into account in preparing nitrogen recommendations for vineyards.

Table 1. Fate of nitrogen from clusters, leaves and canes at/and thereafter harvest.

Clusters	20 kg N ha^{-1}	Lost from soil/plant system		
Leaves	42 kg N ha^{-1}	Lost or recycled in soil/plant system		
			Remobilized to perennial structure	
			Volatilized from canopy	
			Mineralized in soil	
				Volatilized as NH_3 from soil surface
				Leaching with autumn/winter rains
				Denitrified in waterlogged conditions
				Taken up by crop and weeds
Canes/prunings	8 kg N ha^{-1}	Lost or recycled in soil/plant system		
			Removed and burned	
			(or)	
			Mineralized in soil	
				Volatilized as NH_3 from soil surface
				Leaching with autumn/winter rains
				Denitrified in waterlogged conditions
				Taken up by crop and weeds

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PRELIMINARY CRITICAL NITROGEN NUTRITIONAL INDICES FOR LEMON BALM (MELISSA OFFICINALIS) GROWN IN A MEDITERRANEAN ENVIRONMENT IN NORTH-EASTERN PORTUGAL

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The management of crop fertilization presupposes previous diagnoses of soil fertility and plant nutritional status. In Europe, regular soil testing and plant analysis is mandatory for farmers receiving EU subsidies. Soil testing provides information on the potential availability of nutrients in the soil, a key component in the fertilization recommendation systems. By analyzing plant tissues we can access the nutritional status of a crop, which allows understanding if supplemental fertilization is needed. Without these means of diagnostic, fertilization is an entirely empirical practice. The sector of Medicinal and Aromatic Plants (MAP) seems to be out of that logical. For most MAP it were not yet established sufficiency ranges or critical levels that allows the interpretation of plant analysis results (see Mills and Jones, 1996; INIAP-LQARS, 2006). Taken into account that most of MAP production in Europe is organic, and that this may requires the use of expensive commercial organic manures, the situation seems to be unsustainable. There is a real need for sufficiency ranges or critical levels allowing the use of plant analysis as a diagnostic criterion on its nutritional status for a judicious decision on the rate of nutrients to apply. This work is part of a wider project that is still beginning, which main goal is to establish sufficiency ranges or critical levels for some MAP that are having great economic importance in Portugal. In this paper, preliminary results for lemon balm (*Melissa officinalis* L.) are presented.

Materials and Methods

Data were collected in a field fertilization trial of lemon balm where different rates of the nutrients were applied in the form of liquid fertilizers allowed for organic farming. The experiments were located in NE Portugal. The region benefits from a Mediterranean climate. The soil is a Leptosol derived from schist. The texture is sandy-loam, pH 6.5 and an organic matter content of 14 g kg^{-1}. Plants are spaced at 40 x 30 cm. They were planted in holes made in a black plastic mulch used to protect the crop against weeds. The results here presented were obtained from a cut made at the beginning of flowering on September 2, 2013. The dry matter yield and nitrogen nutritional status indices were recorded from experimental plots of 18 useful plants. In the field, SPAD readings were taken by using the portable SPAD 502 Plus chlorophyll meter, which provides an indication of the relative amount of chlorophyll present in plant leaves. The values are calculated based on the amount of light transmitted by the leaf in two wavelength regions in which the absorbance of chlorophyll is different (650 and 940 nm). The Normalized Difference Vegetation Index (NDVI) was also determined by using the portable FieldScout CM 1000 NDVI meter. The Meter senses the light at wavelengths of 660 nm (chlorophyll absorption) and 840 nm (entirely reflected by chlorophyll) to estimate plant health (the relative greenness of the leaf). A NDVI value (-1 to 1) is calculated from the measured ambient and reflected light data [(%Near Infrared – %Red) / (%Near Infrared + %Red)]. From each individual plot five plants were cut and carried out to the laboratory. A subsample was oven dried at 70 ºC allowing estimate the dry matter percentage of the biomass. The other subsample was used to prepare tissue samples for elemental determination. Samples of young and fully expanded leaves (between the 4th and the 10th leaves from the tip) were prepared as well as samples of shoot tips (including stems and leaves) with approximately 8 cm in length. Tissue samples were thereafter dried at 70 ºC, ground and analyzed for nitrogen concentration by a Kjeldahl method. Thereafter, critical levels were estimated for the four indices of plant nutritional status by using the original Cate-Nelson graphical method which is based upon dividing the *Y-X* scatter diagram into four quadrants and maximizing the number of points in the positive quadrants.

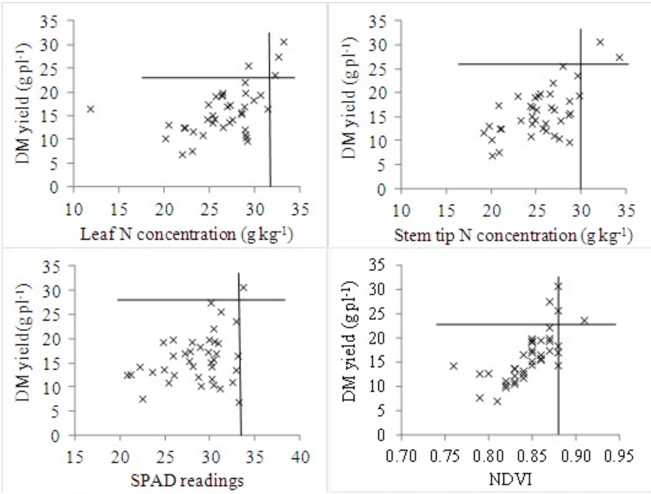

Figure 1. Scatter diagrams of dry mater (DM) yield of lemon balm *versus* plant nitrogen nutritional status indices (leaf and stem tip nitrogen concentration, SPAD readings and Normalized Difference Vegetation Index, NDVI). The vertical lines define the critical value for each nitrogen nutritional index.

Results and Discussion

It was found a linear relationship between dry matter (DM) yield and leaf nitrogen concentration (Figure 1), suggesting that nitrogen was a limiting factor of crop growth in many experimental plots. Significant linear relationships were also found between DM yield and stem tip nitrogen concentration and NDVI. SPAD readings did not correlate so well with DM yield. The results point to critical levels for leaf nitrogen concentration close to 3.2 g kg^{-1} (Figure 1). If the tip of stems were used as sampling tissue, critical nitrogen concentration was found close to 3.0 g kg^{-1}. Critical SPAD reading was found close to 34 units and the critical NDVI was found close to the value 0.88.

Acknowledgment

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THE EFFECT OF DIFFERENT WINTER COVER CROPS ON SORGHUM NUTRITIONAL STATUS AND DRY MATTER YIELD

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Growing winter cover crops has a great agroecological meaning, since it allow maintaining the residual inorganic N in the soil/plant system, thus avoiding leaching of N (Rodrigues et al., 2002). As winter cover crops, it can be grown diverse species (Jensen, 1992). If legume species were used, they can access atmospheric N through the establishment of a symbiotic relationship with N-fixing bacteria (Russelle, 2008). Thus, the winter leguminous cover crops can have a dual role: to uptake residual inorganic N; and promoting the growth of the following crop through a green manuring effect, which may reduce the need for expensive N fertilizers. Lupine (*Lupinus albus*) appears as a suitable legume species to be grown in this region, since it presents high growth rates in winter and a great ability to fix N (Rodrigues et al., 2013). In this work, the use of lupine as a winter cover crop was compared to small grains and natural vegetation (weeds) by measuring their effect on irrigated sorghum grown as a summer crop. The effect of the different winter cover crops was evaluated by comparing sorghum dry matter yield, plant N nutritional status and N recovery by sorghum plants.

Materials and methods

In spring 2012 the biomass of the winter cover crops (lupine, cereal and weeds) was cut. Lupine was managed of two different ways, incorporating all the above-ground biomass in the soil and removing the above-ground biomass. Thus, four treatments were taken into account: weeds (natural vegetation); cereal (a mixture of small grains); lupine total (integral incorporation of plant residues in soil); and lupine root (aerial biomass removed). Sorghum [*Sorghum bicolor* (L). Moench] was sown after the incorporation of the residues of the winter cover crops in the soil at a rate of 10 kg seed ha^{-1}. No fertilizers were applied. During the growing season two cuts of biomass were performed. Dry matter (DM) yields were determined from samples of 0.25 m^2 after over-drying the samples at 70 ºC. The dried samples were ground and analyzed for total N by a Kjeldahl procedure. Plant N recoveries were determined from DM yields and tissue N concentrations. Plant nutritional status was also accessed by determining N concentration in leaves and stems and performing SPAD readings (SPAD-502 chlorophyll meter).

Results and discussion

Sorghum DM yields varied significantly among the different cover cropping treatments (Table 1). Total DM accumulated in the two cuts reached 11.9 Mg ha^{-1} in the Lupine-total treatment and was only 5.7 Mg ha^{-1} in the Cereal treatment. N concentration in sorghum tissues was not significantly affected by the different cover crops. However, N recovery varied significantly among winter cover crop treatments due the differences found in DM yield. In the Lupine-total treatment, sorghum recovered 142.6 kg N ha^{-1} while in the Cereal treatment sorghum recovered only 60.3

kg N ha^{-1}. Leaf N concentration and SPAD readings showed that Cereal was the winter cover crop that decreased more the N nutritional status of sorghum (Table 2). The results also showed that the lupine promoted the sorghum DM yield, likely due to their N-rich residues. The N concentration in sorghum tissues and SPAD readings were not different in lupine plots in comparison with the uncultivated plots (weeds) probably due to a dilution effect caused by the increase in DM yield, since N recovery was higher in the plots where lupine was grown as winter cover crops. In the lupine plots, the removal of shoots did not reduce the beneficial effect on sorghum yield, having been obtained similar values when the above-ground biomass was buried or it was removed.

Table 1. Dry matter yield, tissue N concentration and N recovery by sorghum grown after three winter cover crops, weeds, cereal and lupine, determined in two cuts (August 8th and October 9th) in the summer season.

	DM yield (Mg ha^{-1})			Tissue N conc. (g kg^{-1})		N recovery (kg ha^{-1})		
Treatment	1st cut	2nd cut	total	1st cut	2nd cut	1st cut	2nd cut	total
Weeds	3.8 b	4.1 ab	7.9 b	12.2 a	11.4 a	46.3 b	46.9 ab	93.1 b
Cereal	2.4 b	3.3 b	5.7 b	11.5 a	10.0 a	26.9 b	33.4 b	60.3 b
Lupine-root	7.0 a	4.8 ab	11.7 a	12.6 a	11.1 a	86.5 a	52.2 ab	138.7 a
Lupine-total	6.5 a	5.4 a	11.9 a	11.9 a	12.0 a	77.1a	65.5 a	142.6 a

Means followed by the same latter in columns are not statistically different (Tukey HSD, $\alpha < 0.05$).

Table 2. Sorghum leaf and stem N concentrations and SPAD readings as a function of the previous winter cover crop treatment.

	Leaf N conc. (g kg^{-1})		Stem N conc. (g kg^{-1})	SPAD readings
Treatment	July 19th	August 8th	August 8th	August 9th
Weeds	30.5 a	16.0 a	2.8 a	32.2 ab
Cereal	22.2 b	13.9 b	3.2 a	27.8 c
Lupine-root	28.1 a	16.8 a	2.6 a	31.6 b
Lupine-total	33.2 a	17.7 a	2.4 a	34.5 a

Means followed by the same latter in columns are not statistically different (Tukey HSD, $\alpha < 0.05$).

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TESTING OF A STABILIZED NITROGEN FERTILIZER IN A RICE-WHEAT DOUBLE-CROP ROTATION IN SOUTHEASTERN CHINA

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China's intensive agriculture is characterized by very high nitrogen (N) balance surpluses and low N use efficiencies (NUEs) (Roelcke et al., 2004; Ju et al., 2009). The soils in southeastern China show anaerobic conditions during the irrigated lowland summer rice (*Oryza sativa* L.) vegetation period and aerobic conditions during the upland winter wheat (*Triticum aestivum* L.) cropping season. The alternating water regime leads to high mineral N transformation losses (Roelcke et al., 2002). Besides an overall reduction of the excessive mineral N application rates, strategies for improving fertilizer NUEs may include the use of more efficient N fertilizers, such as urea- or ammonium-(NH_4^+)-based fertilizers with nitrification inhibitors whose effect is based on a stabilization of the NH_4^+-N. The aim of our research was to test the stabilized, urea-based fertilizer with nitrification inhibitor ALZON® 46 by the company SKWP under field conditions of Chinese agriculture. Specific objectives were to investigate whether higher grain yields could be achieved using ALZON compared to conventional urea under equal N application rates, whether similar grain yields could be achieved with ALZON under reduction of N application rates by 33% (rice) and up to 40% (wheat), whether fertilizer NUEs could be increased and N losses reduced, and whether labour could be saved by reducing the number of split applications compared to farmers' practice.

Materials and Methods

A two-year exact field experiment in the county of Huai'an in northern Jiangsu Province (33°35'N 118°53'E). It comprised a total of four growing seasons (2009 summer rice to 2010/11 winter wheat). The experimental design consisted of four treatments with four replicates each in a latin square design. Each plot was approx. 80 m^2 in size and was surrounded by 40 cm high ridges in order to prevent an exchange of water and dissolved fertilizer during the summer rice vegetation period (Fig. 1).

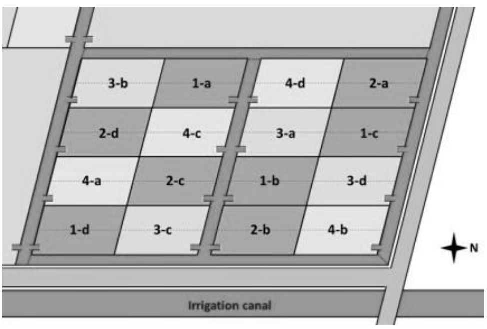


Figure 1: Experimental design of the exact field experiment in Huai'an using a latin square design with four N fertilization treatments and four replicates.

Table 1: N application rates in the exact field experiment in 2009-2010/2010-2011. SA: no. of split applications.

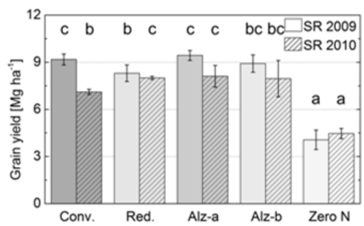
Treatment	Rice		Wheat	
	[kg N ha^{-1}]	SA	[kg N ha^{-1}]	SA
1) Conv. urea	225/300	4/4	250	3
2) ALZON® 46-a	225	3/3	180	2
3) ALZON® 46-b	200	3/3	150	2
4) Zero N	-	-	-	-
5) Reduced urea	225	4/3	180	3

The four N treatments (Table 1) included 1) 'Conventional' urea N fertilization (farmers' practice), 2) ALZON® 46-a: ALZON fertilization with 25% (summer rice) and 30% (winter wheat) less N compared to treatment 1), ALZON® 46-b: ALZON fertilization with 33% (summer rice) and 40% (winter wheat) less N compared to 1), 4) Zero N treatment (control). Additionally, 5), a 'Reduced urea' treatment from a directly adjacent on-farm demonstration field experiment with an N application rate equal to 2) was included in the evaluation. Field and crop management in the exact and the demonstration experiments were identical.

Results and Discussion

Mean grain yields of the summer rice and winter wheat crops during the two growing seasons are shown in Figs. 2a and b. For summer rice in 2010, a significant yield decrease occurred in the 'Conventional' urea treatment compared to 'Reduced' urea and 'Alz-a', because most of the area of the 'Conventional' urea plots was affected by lodging. For the second winter wheat crop (2010/11), there was no significant difference in grain yields between the two ALZON treatments and the 'Conventional' urea treatment, although 28% and 40% less N was applied in 'Alz-a' and 'Alz-b', respectively. However, significantly higher yields could be achieved in the 'Alz-a' treatment compared to the 'Reduced' urea treatment. Significant increases (P<0.05) in mean agronomical N efficiencies (AE_N) were observed between 'Conventional' urea and the two ALZON treatments (Figs. 3a and b). Residual mineral N (N_{min}) contents at wheat harvest in June 2011 showed a clear differentiation between treatments (Fig. 4).

a)



b)

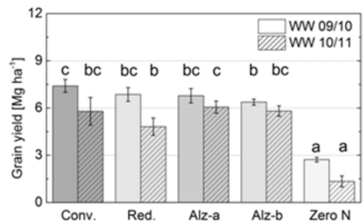


Figure 2a-b: Mean grain yields of summer rice in 2009 and 2010 (n=4) (a) and wheat 2009/10 and 2010/11 (n=4) (b). Error bars: ± s.d. Different letters: Significant difference by Fisher's LSD test between treatments of one year (P<0.05).

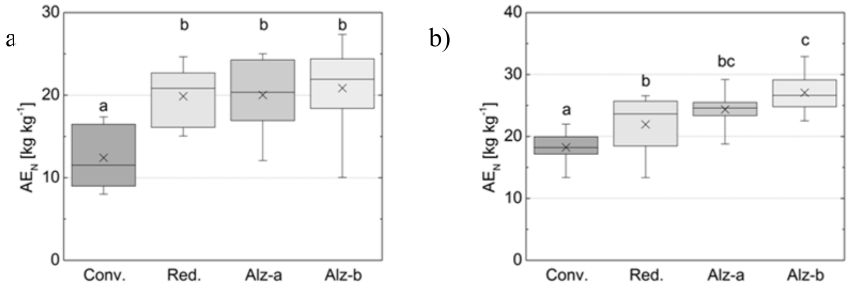


Figure 3a-b: Box-and-whisker plots of mean AEN of summer rice crops 2009-2010 (n=8) (a) and mean AEN of winter wheat crops 2009/10-2010/11 (n=8) (b). The median is depicted with a solid line and the mean with a cross, box plots with the same letter do not differ significantly by Fisher's LSD test (P <0.05).

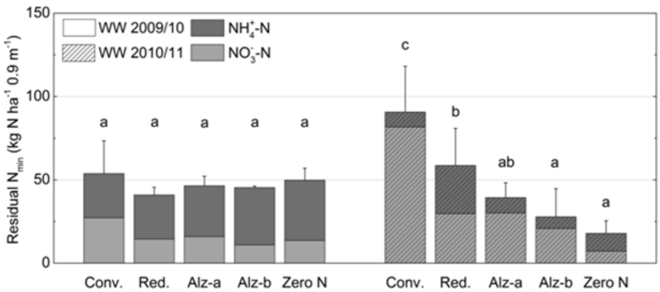


Figure 4: Mean residual mineral N (N_{min}) contents (n=4) in 0-0.9 m depth after harvest of winter wheat in June 2010 (left) and June 2011 (right); error bars: ± s.d.; same letters do not differ significantly by Fisher's LSD test ($P < 0.05$).

Conclusions

Use of N fertilizers with nitrification inhibitors can be recommended for winter wheat crops in SE China. It was possible to increase NUEs and to reduce the risk of N losses for winter wheat, particularly in dry years. In contrast, less marked effects of ALZON on grain yields and NUEs were observed for rice.

Acknowledgements

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IMAGE TECHNIQUES TO EVALUATE THE VEGETATIVE GROWTH OF AN OLIVE ORCHARD

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Digital photography has evolved considerably in the last years, making it affordable for everyone today. The digital image provides a lot of information that can be used for different purposes and allows to incorporate photography as a new powerful, economical and available tool. This work evaluates and compares the vegetative growth response to different irrigation and fertilization strategies of an Arbequina olive orchard grown under super-intensive conditions using this new tool.

Materials and Methods

The trial was conducted on a commercial adult olive plot (cv. Arbequina) in Torres de Segre (Lleida) during the year 2013. Trees were spaced 4,5 x 2,2 m (1010 trees.ha^{-1}). The trial consisted of 48 plots as a result of crossing four irrigation treatments with three fertilizing treatments, randomly distributed in four blocks. The irrigation treatments were: drip irrigation at 100% of the calculated irrigation dose (R-100), and drip irrigation at 40% dose (RDC-0.4), at 60% dose (RDC-0.6) and at 80% dose (RDC-0.8) during pit hardening. The fertilization treatments were three doses of nitrogen (N) of 0, 50 and 100 kg of N/ha. Tree growth was assessed in 4 control trees per plot using four methods: 1) tape measured trunk perimeter (TP), 2) visual canopy volume measurement (CV), 3) lateral canopy area (LCA), and 4) zenith canopy area measurements (ZCA), all done at the end of the season. For the last two methods a single-lens reflex digital camera (Nikon D5100, Nikon, Japan; with Tamron SP AF 10-24 mm F/3,5-4,5 Di II LD ASL IF, Tamron, Japan) and specific software (Photoshop CS6, Adobe Systems, USA; ImageJ, NIH, USA) were used, following the methodology proposed in Lordan et al., 2013. This technique consists on taking a digital photo of all the trees of the plot, selecting the part of the photo that will be analyzed.

Results and Discussion

The different methodologies used in this work to measure vegetative growth allowed us to observe that the trees under RDC-0.4 irrigation strategy had the lowest growth data. For the fertilization doses, plots without N had lowest ones (Table 1). In the other hand, in the case of irrigation, most of times higher values corresponded to R-100 treatments, without statistical significant differences between treatments, except for RDC-0.4 (Table 1). With regard to fertilization, higher values corresponded to N-100 treatments. However it should be noted that in the case of trunk perimeter it does not exist statistical significant differences. Both ZCA and lateral canopy area (LCA) provided fast and accurate useful information to evaluate tree response to different treatments of irrigation and fertilization being ZCA the methodology that gave most clear differences, mainly for fertilization treatments (Table 1). Moreover, the entire plot digital images can be studied with a single photo (common methods referred to one tree), evaluating a larger area in a quicker, cheaper and more accurate way.

Table 1. Effects of irrigation and fertilization treatments on trunk perimeter (TP), canopy volume (CV), zenith canopy area (ZCA) and lateral canopy area (LCA). Values followed by different letters indicate significant differences according to Tukey HSD test (P< 0.05).

Concept	Units	R-100	RDC-0.8	RDC-0.6	RDC-0.4	N-100	N-50	N-0
TP	cm	36,40a	32,34ab	35,24ab	32,34b	36,91a	34,49a	34,19a
CV	m^3	4,71a	4,95ab	3,83ab	3,90ab	5,04a	4,46ab	4,01b
ZCA	m^2	11,18a	10,48ab	10,47ab	8,74b	12,44a	10,49b	9,17c
LCA	m^2	5,16a	5,12a	4,93a	4,07b	5,32a	4,93ab	4,39b

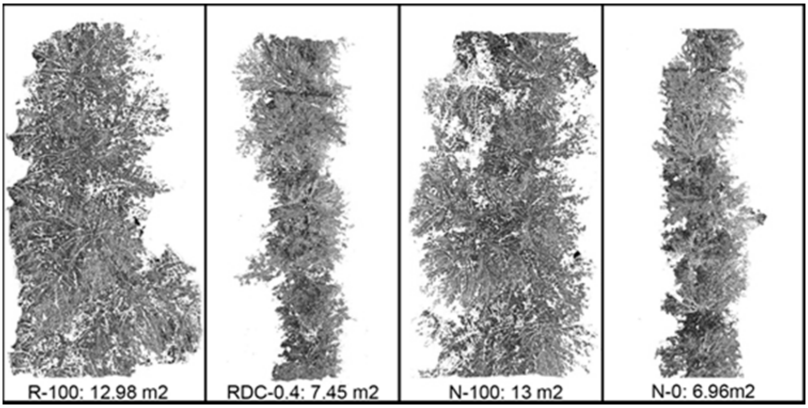

Figure 1. Images of four different plots corresponding to irrigation and fertilization treatments using zenith canopy area methodology.

Conclusions

Digital image is presented as a useful, effective and worthy analytical tool with a high future potential for research. This new method provides complementary information in a quick and easy way to any user.

Acknowledgements

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THE IMPACT OF NITROGEN FERTILIZER NORM ON INDICATORS OF NUTRIENT USE FOR SPRING BARLEY

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High yields of barley grain are produced when the soil nutrient stock is adequate at all growth stages of plants. Nitrogen fertilization is the most effective yield increasing factor in barley, however soil fertility, amounts of previously applied fertilizer, variety potential, meteorological situation during vegetative growth and other conditions (Moreno et al., 2003), including soil properties, should be considered. Increased N fertilizer rates result in increased grain yields only up to a certain yield limit, at the same time return from 1 unit nitrogen use with grain yield gradually decrease (Anbessa, Juskiw, 2012, Shafi et al., 2011). Excessive nitrogen is, possibly, a hazard to environment because N-compounds unused by plants in the soil is a risk of surface and, particularly, groundwater pollution, therefore N-containing fertilizer input in the soil is limited based on measures developed by the European Union (Council Directive of 1991).Wherewith, the research goal is to determine allowed and economically grounded mineral fertilizer, and mostly N fertilizer, rates and indicators of nutrient utilization based on the results obtained in several years long research conducted in different soil and agro-climatic conditions.

Materials and Methods

Field trials with spring barley (*Hordeum vulgare*) cultivar 'Tocada' were conducted in the Research and Study farm 'Peterlauki' of the Latvia University of Agriculture, State Stende Cereals Breeding Institute and State Priekuli Field Crops Breeding Institute over the period 2008 – 2012. Nitrogen fertilizer rates were used: 1. $N_0P_0K_0$ – check, 2. PK background, the following with a step N_{30} to N_{210} kg ha^{-1}. Outcome, plant nutrient balance and utilization coefficients were calculated. Total N was determined by the Kjeldahl method. Nitrogen outcome was calculated based on total nitrogen content in seeds and straw. Apparent recovery fraction (*ARF*) for nitrogen was calculated according formula, suggested by Montemurro (Montemurro et al., 2007).

Results and Discussion

Increasing of yield was obtained by nitrogen fertilizer N_{90-120} following increase of nitrogen was not effective (Figure 1). Plant nutrient removal with yield is dependent on crop yield level and nutrient content in basic products (grain) and by-products (straw). Nitrogen removal is associated with yield increase as affected by N fertilizer and with the increase of N content in grain and straw. The increase in P_2O_5 associated almost only with the increase in grain yield. Difference between minimum and maximum value of K_2O removed was more than two times greater. Utilization coefficients of plant nutrients are, to a great extent, dependent on meteorological situation in the growing season. On the average mineral N utilization coefficient was the highest with nitrogen fertilizer rate N_{90} – N_{120} – 0.50-0.51, keeping constant relationship that utilization coefficient gradually decreases with each succeeding N fertilizer rate. Utilization indices of P_2O_5 are comparatively small however the increase in N fertilizer rate resulted in the increase of P_2O_5 removal.

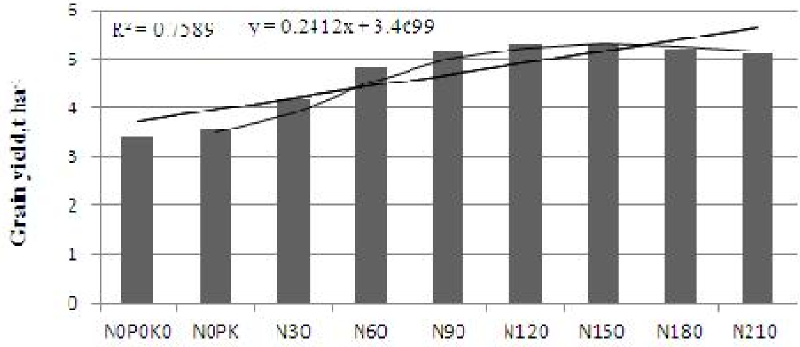

Figure 1. Mean grain yield as finction of N fertilizer rates, t ha^{-1}

Conclusion

The increase of barley grain yield under conditions of Latvia is comparatively stable with nitrogen fertilizer rates up to N90. Further need for nitrogen is determined depending on the character of the growing season and plant status. In case of need, N30 as N-topdressing is added. Increasing nitrogen fertilizer rates to N90 – N120 in most cases result in increased agronomic effectiveness of nitrogen, however succeeding increase in N fertilizer rates have a negative effect. The increase of N fertilizer rates rapidly increase K_2O content in straw and wherewith potassium removal with yield.

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MAXIMUM NITROGEN RATES APPLICATION AND POSITIVE EFFECT OF THE ROTATION WITH ALFALFA ON THE WHEAT FIELD IN THE NORTHERN ITALY

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It is of vital importance for the maintenance of the agricultural activities and for the protection of the environment to know and to be able to apply the right amount of fertilizer to the soil. This allows to obtain the best crop yield, to limit loss of fertilizers in environment. Also it is important to control cost management and to limit surface and underground waters pollution. The purpose of this study was to compare wheat (*Triticum aestivum aubusson*) in monocropping and in rotation with alfalfa and identify the best nitrogen rate for maximize wheat yield.

Materials and methods

The experiment was carried out in Montanaso Lombardo (Italy), geographical coordinates 45°20’32’’N; 9°26’46’’E. The soil is classified as a Gleyi-Luvic Calcisols, in according with FAO classification. The climate is temperate sub continental, with about 800 mm of rainfall during the year, with a maximum in October. The annual average temperature is 12.9°C, while coldest and hottest months averages are 1.5°C and 23.7°C. There was a large wheat field, cultivated as monocropping. We had sown in different years three small experimental areas of 2000 m^2 with alfalfa. The alfalfa was grown in those areas for three years; after this period each small area was broken by plowing and the wheat was subsequently replanted. We have made a comparison between wheat production in the restored area, and a nearby area of same size, where wheat was kept in monocropping. In both these areas, a randomized block trial was established to test 4 different levels of N supply, each in 4 replicates. These comparisons were made in years 2008/2009, 2009/2010 and 2012/2013. The N supply rates were respectively 0, 50, 100, 150 kg/ha. Maximum yield N rate (MNR) was assessed using the quadratic equation for estimating grain yield response curve, as a function of N rate: $y = a + bx - cx^2$ (proposed by Kwaw-Mensah and Al-Kaisi, 2006), where y is yield and x is N rate. Placing the first derivative equal to zero and solving for x, the value of the MNR was found: $x = b/2c$. In addition two factorial variance analysis, coupled with Tuckey test, was used in order to assess the significance of the differences among the theses. Data yields, usually expressed in t/ha, were normalized respect to the less productive thesis of each year, corresponding to monocropping 0 N supply, which has been equated to 100. In 2008/2009 average yield of 4 replicates of less productive thesis was 2,1 t/ha, in 2009/2010 was 1,9 t/ha and in 2012/2013 was 2,4 t/ha.

Results and Discussion

The MNRs for each year, calculated on the basis of quadratic equations fitted to data, are summarized in table 1; the resulting MNR values were all inside or nearby the tested N rates, only a value for 2012/2013 fell beyond these limits. There was close to normal amount of rainfall in 2008/2009 and 2009/2010 years. Need to be underlined, that in 2012/2013 there were particular rainfall conditions, 1075 mm, a double amount compared to normal conditions, 558 mm (30 years average), during the growing season of wheat. Comparing the results, the effect of management (monocropping/rotation) was significant (p 0,05) for yields among the different theses, for all the N supplies applied, for the years 2008/2009 and 2009/2010 (Fig. 1). In 2012/2013 there were no significant differences for management. For the monocropping in 2008/2009 all the N rates gave significant differences in yield, while for rotation the significance was achieved only between 0 and others N rates. In 2009/2010 for monocropping the results were similar to 2008/2009 except for an anomalous yield on 100 kg/ha N rate. In the same year rotation showed again a significant difference only among 0 kg/ha and all others N supplies. In 2012/2013, the only significant difference was between the supply of 0 kg/ha and all the others, for the cultivation in rotation.

MNR kg/ha N	monocropping	rotation
2008/2009	108	100
2009/2010	163	145
2012/2013	262	100

Tab. 1: Calculated MNR.

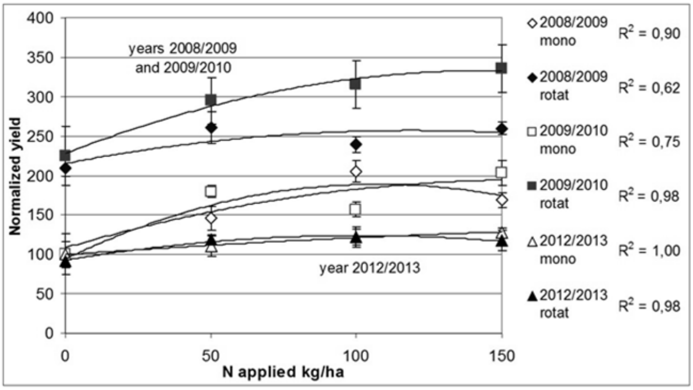

Fig. 1: Quadratic functions interpolating wheat normalized yield for rates of N supply.

Conclusions

Rotation wheat with alfalfa showed a clear positive effect, which was more significant than effect caused by any of N supply. This effect could be explained by soil Nitrogen enrichment caused by alfalfa residues. On the other hand, culture rotation can help to suppress allelopaty. In 2012/2013, beneficial effect caused by the rotation, observed in previous years for all N supplies, has disappeared. Probably it could be explained by the high rainfall, which prevented much of the Nitrates accumulation in the soil during wheat growing season. Overall, wheat yields of land in rotation with alfalfa were higher than those in monocropping. Analysis of variance showed that for wheat in rotation with alfalfa only the 50 kg/ha N rate gave a significant yield increase, in years with regular rainfall. The same conclusion can be drawn by MNR analysis although with this approach the critical rate was estimated to be among 100 kg/ha.

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TEMPORAL DYNAMICS OF NITROGEN RHIZODEPOSITION IN PEAS

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The release of nitrogen (N) from legume roots (N-rhizodeposition) contributes substantially to the N nutrition of subsequent crops. Quantity and quality of rhizodeposition is influenced by plant species, but also by the soil microorganisms (especially mycorrhiza) being present. Conversely, the rhizodeposits strongly influence the microbial turnover in the soil and therefore the mineralization of nitrogen. For a better prediction of available N in legume based crop sequences, but also for intercropping systems, our knowledge about the temporal dynamics of rhizodeposition (C and N), as well as the influence of mycorrhiza on this process has to be improved. The objective of the present study therefore was to investigate the temporal dynamics of N release from roots of peas (*Pisum sativum* L.) being infected with mycorrhiza compared to roots without mycorrhizal infection.

Materials and Methods

A pot experiment was carried out under controlled conditions in the greenhouse, where pea plants (*Pisum sativum* L. cv. Frisson and non-mycorrhizal isoline P2) were infected with Mycorrhiza (*Glomus* sp.). Pea plants were labeled fortnightly (beginning at BBCH 13: 3 leaves unfolded or 3 tendrils developed) with a solution of 2% ^{13}C-glucose (99 atom%) and 0.5% ^{15}N urea (95 atom%) using the cotton wick method (after Russell & Fillery, 1996 and Wichern et al., 2010). Sampling took place on four dates depending on the BBCH stage, beginning at BBCH 59 (first petals visible, flowers still closed) (Table 1). Rhizodeposition of C and N was estimated according to Janzen and Bruinsma (1989), based on the assumption, that root and rhizodeposit label distribution was equal in space and time, at the four growth stages. Furthermore, the percentage of the microbial biomass derived from rhizodeposition, was quantified and compared between the variety Frisson and the non-mycorrhizal pea mutant P2.

Results and Discussion

It was found that below-ground plant derived N (BGP-N) = consisting of root N and N derived from rhizodeposition as a percentage of total plant N decreased during plant growth in both pea varieties (Frisson and P2) (Table 2). It followed the same pattern as the root-to-shoot ratio, which was higher at an early vegetative growth stage. Moreover, also the mycorrhization of the variety Frisson decreased during plant growth with a maximum infection at early vegetative growth (Table 3). Mycorrhization was related to the root-to-shoot ratio of the plants, also showing a stronger contribution at early vegetative growth. Nitrogen rhizodeposition as a percentage of total plant N was low compared to previous experiments (Table 4). This can be explained by a very low root-to-shoot ratio, often observed in pot experiments. Below-ground plant N dynamics follow a different pattern compared to above-ground plant N. Plants invest relatively more N into the below-ground biomass at an early growth stage compared to later growth stages. This reflects the necessity for plants to invest in root development at early growth to have the ability of using the available resources (water and nutrients) efficiently for the generative growth. Genetic improvement of legume species and varieties better adapted to water and nutrient stressed environmental conditions, might make use of this observed pattern of root plasticity. A prolonged period of intensive symbiosis with mycorrhiza might further strengthen the plants ability to withstand drought conditions and increasing the nutrient use efficiency. Consequently, legumes with a better adaptation to environmental stresses can better contribute to the N nutrition of subsequent and accompanying crops.

Table 1: ^{15}N enrichment of various plant parts in Frisson (*Pisum sativum* L.) and P2 (*Pisum sativum* L.) at 4 different growth stages after ^{15}N labelling using the cotton wick method

	^{15}N excess (at%)			
	BBCH 59	*BBCH 69*	*BBCH 79*	*BBCH89*
Frisson				
Grain		2.73 ± 0.6	1.67 ±0.3	2.43 ± 0.5
Stem and leaves	3.76 ± 1.7	3.62 ± 0.7	2.72 ± 0.5	4.12 ± 0.8
BGP-N	1.76 ± 0.6	2.35 ± 0.7	1.70 ± 0.7	2.16 ± 0.3
P2				
Grain		4.82 ± 1.2	3.23 ± 1.1	5.08 ± 0.8
Stem and leaves	4.30 ± 0.9	4.95 ± 1.1	3.65 ± 1.2	4.97 ± 0.6
BGP-N	2.02 ± 0.6	1.94 ± 0.6	1.72 ± 0.6	2.14 ± 0.4

Table 2: Total plant-N, grain-N, stem & leaves- N and below ground plant-N (BGP-N = root-N + N derived from Rhizodeposition [NdfR]) in Frisson (*Pisum sativum* L.) and P2 (*Pisum sativum* L.) at different growth stages

	BBCH 59	*BBCH 69*	*BBCH 79*	*BBCH89*
Frisson	(mg pot^{-1})			
Total Plant-N	110.84 ± 22.9	292.63 ± 59.2	467.45 ± 60.8	646.25 ± 141.8
Grain-N		43.51 ± 16.9	94.34 ± 34.4	464.02 ± 108.6
Stem and leaves-N	85.42 ± 20.8	205.64 ± 30.2	332.95 ± 26.3	142.14 ± 55.4
BGP-N	25.42 ± 5.4	41.88 ± 11.0	40.74 ± 8.6	40.08 ± 11.0
NdfR	20.55 ± 5.1	35.92 ± 9.4	32.43 ± 7.3	32.37 ± 9.5
P2	(mg pot^{-1})			
Total Plant-N	105.22 ± 18.6	217.49 ±54.4	316.58 ± 57.9	293.91 ± 63.0
Grain-N		59.89 ± 13.7	90.55 ± 28.2	220.63 ± 43.2
Stem and leaves-N	85.18 ± 16.4	119.31 ± 41.5	186.31 ± 47.2	39.27 ± 10.8
BGP-N	20.04 ± 6.9	38.29 ± 10.9	39.72 ± 13.1	34.01 ± 15.1
NdfR	16.53 ± 7.1	34.30 ± 11.4	34.75 ± 14.1	29.63 ± 14.7

Table 3: Mycorrhization and root to shoot ratio in Frisson (*Pisum sativum* L.) and P2 (*Pisum sativum* L.) at 4 different growth stages

	BBCH 59	*BBCH 69*	*BBCH 79*	*BBCH89*
Frisson				
Mycorrhization (in %)	29.6	23.0	16.5	16.5
Root to shoot ratio	0.09	0.03	0.02	0.04
P2				
Mycorrhization (in %)	0	0	0	0
Root to shoot ratio	0.06	0.04	0.03	0.05

Table 4: Quantities of below-ground plant N (BGP-N) and N derived from rhizodeposition (NdfR) of peas.

Plant species	*Growth stage*	*BGP-N (in % of total plant N)*	*NdfR (in % of total plant N)*	*NdfR (in % of BGP-N)*	*BGP-N (mg/plant)*	*total N (mg/plant)*	*Experimental conditions*	*References*
Pisum sativum	Until flowering	27	4	15	44	165	Split-root	Jensen, 1996
	Until maturity	14	7	48	40	277		
		15	12.6	82	56	360	Cotton wick	Mayer et al., 2003
		16	10.5	65	64	397	Split root	Schmidtke, 2005
	Whole growth period	33.5	29.2	87			Cotton wick/column exp.	Wichern et al., 2007
P. sativum cv. Frisson	Until pod filling	27.3		38.1			Cotton wick	Mahieu et al., 2008
	Until maturity	26		45.2				
		6.2	2.5	80.7	20.0	323.1	Cotton wick	Schmidt et al., unpublished
P. sativum cv. P2	Until maturity	11.4	4.9	87.1	16.8	147.0		

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PHENOTYPIC AND GENOTYPIC CHARACTERIZATION OF INDIGENOUS RHIZOBIA NODULATING ALFALFA IN MEDITERRANEAN REGION OF CROATIA

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An essential element of agricultural sustainability is the effective management of nitrogen in the environment. This usually involves at least some use of biologically fixed nitrogen because N from this source is used directly by the plants, and therefore is less susceptible to volatilization, denitrification and leaching. Despite that, seed inoculation with *Sinorhizobium meliloti*, a nitrogen-fixing microsymbiont of alfalfa, has not become yet widespread procedure in the production of this important forage crop in Croatia. Selection of the most suitable strain represents one of the main presumptions for successful inoculation due to the fact that rhizobial strains strongly differ in their effectiveness, competitiveness and compatibility. However, the presence of adapted and competitive indigenous alfalfa rhizobia in soil can reduce the inoculation response even with the highly efficient commercial strains. Considering this, composition and characteristics of rhizobial field populations are of great agricultural imporatnce in alfalfa production. The main objective of the present study was to characterize indigenous *S. meliloti* strains isolated from different soil types in Mediterranean region of Croatia.

Materials and Methods

The soil samples for rhizobial isolation were collected from different field sites within the Zadar County which covers the central part of the Adriatic cost. Greenhouse pot experiment was established in order to obtain nodules for isolation procedure. The study included 20 isolates, reference strains of *Sinorhizobium medicae* (LMG 18864) and *S. meliloti* (2011) as well as *S. meliloti* type strain (30135). In order to perform phenotypic characterization of indigenous strains, growth characteristics under different temperature conditions, pH values, salt and heavy metal concentrations as well as carbon source assimilation were determined. Rhizobial isolates were characterized by different PCR fingerprinting methods. PCR-RFLP of 16S rDNA was used for identification at the species level while RAPD and ERIC-PCR were used for strain differentiation and detection of genetic diversity within indigenous rhizobial populations.

Results and Discussion

Stress tolerance assays revealed significant variations in pH tolerance while almost all isolates showed similar tolerance to elevated salt concentartions and growth temperatures. The obtained results has shown that almost all rhizobial strains tolerate alkaline conditions while their growth was mainly inhibited in acidic conditions. However, one third of total number of isolates showed tolerance to acidic pH values and their growth was not inhibited even at pH 4.5. Most of isolates showed good growth at 37C while at 42C the growth was mainly inhibited. Different impact of heavy metals on rhizobial growth was determined. Most of the tested strains showed good growth on nutrient media containing zinc and manganese while addition of cadmium and copper mainly resulted in partial or complete growth inhibition. The results of 16S rDNA PCR-RFLP clearly showed that most of isolates obtained from alfalfa nodules could be regarded as *S. meliloti* while none of the isolate was identical with *S. medicae* type strain. Three isolates were significantly different from both *S. meliloti* and *S. medicae* type strains. Dendrogram derived from RAPD and ERIC-PCR profiles revealed considerably genetic diversity among rhizobial isolates (Fig.1). Only a few strains were identical or nearly identical to each other.

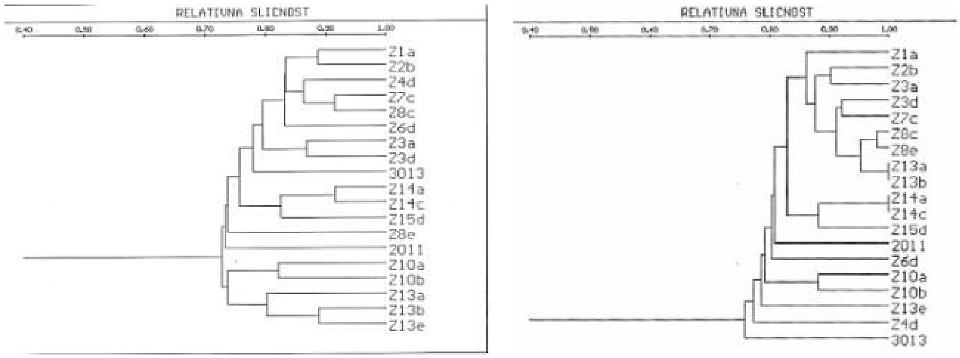

Figure 1. Dendrograms of S. meliloti field isolates derived from RAPD-PCR and ERIC-PCR patterns (from left to right)

Conclusions

In order to select strains for high-quality inoculant production, further investigations are needed to evaluate symbiotic properties of rhizobial isolates such as symbiotic efficiency and compatibility with different alfalfa cultivars. The results confirmed the presence of other bacterial species within the nodules. Therefore, the assessment of hidden diversity within the nodules shoud be verified in future studies. These results may be relevant to programs directed towards improving nitrogen fixation efficiency and crop productivity through alfalfa inoculation with locally adapted and genetically defined strains.

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INITIALIZING THE DSSAT-CENTURY MODEL: INVERSE CALIBRATION OF CARBON POOLS FROM APPARENT SOIL N MINERALIZATION

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The CENTURY soil organic matter model was adapted for the DSSAT (Decision Support System for Agrotechnology Transfer), modular format in order to better simulate the dynamics of soil organic nutrient processes (Gijsman et al., 2002). The CENTURY model divides the soil organic carbon (SOC) into three hypothetical pools: microbial or active material (SOC1), intermediate (SOC2) and the largely inert and stable material (SOC3) (Jones et al., 2003). At the beginning of the simulation, CENTURY model needs a value of SOC3 per soil layer which can be estimated by the model (based on soil texture and management history) or given as an input. Then, the model assigns about 5% and 95% of the remaining SOC to SOC1 and SOC2, respectively. The model performance when simulating SOC and nitrogen (N) dynamics strongly depends on the initialization process. The common methods (e.g. Basso et al., 2011) to initialize SOC pools deal mostly with carbon (C) mineralization processes and less with N. Dynamics of SOM, SOC, and soil organic N are linked in the CENTURY-DSSAT model through the C/N ratio of decomposing material that determines either mineralization or immobilization of N (Gijsman et al., 2002). The aim of this study was to evaluate an alternative method to initialize the SOC pools in the DSSAT-CENTURY model from apparent soil N mineralization (Napmin) field measurements by using automatic inverse calibration (simulated annealing). The results were compared with the ones obtained by the iterative initialization procedure developed by Basso et al., 2011.

Material and Methods

The initialization method developed in this work (Met.2), was compared with the Basso et al. (2011) procedure (Met.1), by applying both methods to initialize the SOC pools of a 4-year field experiment in a semiarid irrigated area of Madrid (40º03'N, 03º31'W, 550 m.a.s.l.). The main crop of the experiment was maize alternating with three cover crop (CC) treatments: barley, vetch and fallow. An additional control treatment was also included without fertilization to determine Napmin during the 2008 and 2009 maize period. The CENTURY model was initialized for the control treatment through the two methods, obtaining two different sets of SOC3 pools. Crop coefficients and soil parameters were also calibrated.For the model validation in the fallow, barley and vetch treatments, the obtained values of SOC3 from the two initialization methods were used separately.

Results and Discussion

The resulted SOC3 fractions were lower for Met.1 than for Met.2 in the top three layers. Since the total SOC was the same for both methods, a smaller SOC3 fraction would mean higher fractions of the active and slow pools (SOC1 and SOC2) in Met.1 than in Met.2. Method 2 improved the simulation of Nmin in the control plots as the Napmin was calibrated with actual conditions (78 kg N ha-1 in 2008 and 79 kg N ha-1 in 2009). Method 1 resulted in overestimation of the observed Napmin and Nmin in the control plots (180 kg N ha-1 in 2008 and 155 kg N ha-1 in 2009). The simulated N dynamics in the fallow, barley and vetch treatments presented higher Nmin in Met.1 than in Met.2 due to the higher proportion of the SOC1 and SOC2. Simulation of N leaching in the soil was similar for both methods showing a decrease in the nitrate leaching when replacing bare fallow with cover crops during the intercropping period. The Met.1 provided higher SOC1 and SOC2 fractions resulting in a higher Napmin and smaller cumulative SOC than Met.2. In both methods, Napmin was slightly higher in the CC treatments than in the bare fallow while SOC was smaller in the fallow than in the CC treatments.

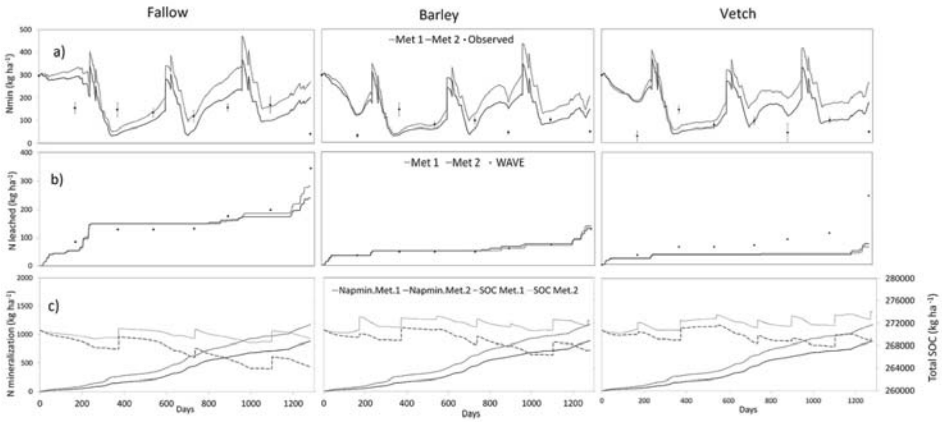

Fig.1. (a) Nmin, (b) N leached (kg ha^{-1}) leaching and (c) Napmin and soil organic carbon (SOC) evolution for the fallow, barley and vetch treatments comparing the two methods of CENTURY initialization: Met 1: Century initialization following the iterative procedure; Met 2: Century initialization based on field Napmin measurements.

Conclusions

The automatic inverse calibration of the initial carbon pools from Napmin measurements demonstrates to be an effective initialization method for several reasons: it is based on field observations that can be easily measured; it ensures a good simulation of the Napmin; it makes a close estimation of the N leaching and the Nmin. Therefore, it leads to reliable simulations of all the components of the soil N balance keeping a proper simulation of crop growth. Thus, when Napmin measurements are available, we recommend to use Met.2 to initialize the CENTURY model. This work highlighted the importance of the initialization procedure on the CENTURY model performance when simulating SOM dynamics, as the results can vary significantly with one initial SOC fractions or another.

Acknowledgements

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BREEDING OF OILSEED RAPE FOR SUSTAINABLE AGRICULTURE: CHARACTERIZATION OF A BRASSICA NAPUS DIVERSITY SET FOR NITROGEN USE EFFICIENCY TRAITS.

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The element nitrogen (N) is essential for plant growth and agricultural crop productivity and is the plant nutrient that has to be applied as fertilizer in the largest quantities. Unused nitrogen, however, can escape from the agricultural production system either by nitrate leaching into groundwater, runoff from the soil surface, or due to emissions of nitrous oxide or volatile ammonia. In the agro-ecological context these events can have potentially serious negative side effects on neighboring ecosystems. Furthermore, since mineral fertilizer production by the Haber-Bosch process is energy dependent, carbon dioxide emissions influence the greenhouse-gas balance in a negative way; simultaneously rising energy costs result in increasing fertilizer prices, thus diminishing the economic margin of farmers. In future the question will arise how a growing world population can match its demand without magnified impacts on the environment. Besides a more adjusted fertilizer management, a greater emphasis on breeding and cultivation of varieties with improved nitrogen use efficiency (NUE) is an important component towards a more sustainable agriculture.

Oilseed rape (*Brassica napus* L.) is Europe`s most important oilseed crop and the second most important in the world. Its high quality plant oil for human nutrition purposes, the industrial use as substitute for fossil oil and not at least its high value protein as animal feed, has resulted in significant production increases: Today around 40 Mio tonnes of oilseed rape are produced per annum in the European Union on an average cultivation area of 8.6 Mio ha (FAOSTAT). As the dominating dicoltyledon crop, winter oilseed rape has assumed an integral role in cereal crop rotations. However, with its relatively high acquisition of nitrogen during vegetative growth stages, but a comparatively low nitrogen seed yield, oilseed rape cultivation is often associated to an N-balance surplus. The objective of this study was to assess genetic variation for NUE-influencing traits and evaluate its potential use for breeding programs to improve NUE in oilseed rape.

During the 2012-2013 growing season a set of 30 highly diverse *B. napus* genotypes, including old varieties and novel resynthesized rapeseed lines, was investigated in Mitscherlich pots for responses to divergent nitrogen fertilization levels (0.75 g, 1.25 g, and 2.25 g N pot^{-1}). At flowering time (developmental stage BBCH 67-69), stems, leaves, flowers and pods of all plants were harvested separately to determine the respective nitrogen contents in the particular plant segments. Later, at seed maturity, the nitrogen content of seeds and plant residues was also analyzed. In this manner the experiment delivered a first understanding of genetic differences for both nitrogen uptake efficiency (NupE) and nitrogen utilization efficiency (NutE).

The results indicated a huge phenotypic variation within the materials for N uptake until flowering, resulting in a difference of 25% for NupE between the best and worst genotype at this stage. Moreover, the genotypes differed by around 30% in their utilization of uptaken N during the generative phase. They therefore showed notable differences in their N quantities in the different plant organs. Since uptake and utilization seem to be independently inherited trait complexes, the results suggest a great potential to create varieties with the combined effects of more efficient N-uptake and increased N-utilization.

Furthermore, since the variation in root morphology is an important aspect to consider for enhanced N uptake, the same genetic material was grown in 90 cm deep containers filled with 130 kg of soil and fertilized with an equivalent to 75 and 235 kg N ha^{-1}. This container experiment builds a compromise between field and pot experiments and allows the simulation of a field-simulating plant-soil-interaction under controlled conditions. After the harvest of above-ground plant material, roots of 120 containers were washed out and root phenotypes and responses to nitrogen were assessed.

Within the diversity set a huge genetic variation was discovered regarding the length, biomass and morphology of the roots. Interestingly, both negative and positive genotype responses of root growth to increased nitrogen fertilization were observed. In addition, collection of all aborted leaves between flowering and seed harvest showed a more than fourfold difference in leaf nitrogen loss between extreme genotypes within the first 10 days of leaf abortion. Particularly higher yielding genotypes exhibited a more continuous senescence and loss of leaves, which can be seen as an advantage in terms of nitrogen remobilization.

Overall, we demonstrate the broad variation in NUE influencing traits, identifying genetic differences that can be used for NUE breeding. In future the cultivation of more efficient varieties might allow farmers to harvest more with less fertilizer. This will be beneficial for the environment and delivers economic advantages for the entire society.

EVALUATION OF THE RELATIONSHIP BETWEEN DIFFERENT HYPERSPECTRAL INDICES AND LEAF N IN WHEAT USING MIXED MODEL THEORY

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Hyperspectral (HS) data of vegetation provide a wealth of detailed information to identify plant nutritional status, but data analysis is fundamental for exploiting their full potential. The discrete waveband approach (Heege, 2013), through computation of narrowbands vegetation indices (VIs), is the most straightforward strategy used to synthetize information from plant spectral signatures. However, saturation effects are known to occur for some VIs over specific LAI values; in addition, bands capturing most of information of crops characteristics may vary across growing cycle modifying VI efficacy. Conversely, full spectrum approach, through multivariate analysis, could synthetize whole plant spectral response. In any case, the statistical function is still depending on growth stage and plant health.

In the study of the relationship between HS data and biochemical parameters, ordinary least squares models (OLS) are commonly employed but their use requires important assumptions to be satisfied. However, residuals are often spatially correlated and, when spatial dependence is not taken into account, type I error tends to increase leading to results misinterpretation and improper management decisions. Generalized least squares models (GLS) with correlated errors allow spatial correlation components to be assessed and filtered from the residual term (Rodrigues et al., 2013).

The aim of this study was to investigate the efficacy of different approaches for HS data analysis in estimating leaf N in wheat. To reach this aim, models accounting for spatial correlation were used and compared to OLS models.

Materials and Methods

The research was carried out in southern Italy at the CRA-CER. Durum wheat was grown in rainfed conditions under varying N supply. The experimental design consisted of 10 un-replicated treatments, ranging from 0 to 180 kg N ha^{-1}. Canopy reflectance was surveyed at different growth stages in 100 georeferenced locations, using a high resolution radiometer (325-1075 nm range). Data used in this study were collected at beginning of stem elongation, booting and heading. Leaf N was quantified on samples collected at same locations and dates.

Leaf N concentrations were tested for normality and spatial autocorrelation. At beginning of stem elongation, leaf N data showed a large departure from normality, thus were transformed into normal scores through Blom's algorithm using PROC RANK of SAS.

Spectral reflectance data were processed through the discrete wavebands (VIs) and full spectrum approaches. Three VIs, chosen among the most informative and widely used, were computed: NDVI-normalised difference VI; REIP-red edge position index by Guyot; NDRE-normalised difference red edge. Full-spectrum indices were extracted through partial least square regression (PLS). PLS was applied to reflectance data restricted to 395-1004 nm interval, considered noise-free, after averaging over 10 nm. Significant X-scores were selected through cross validation and van der Voet's test.

The VIs or the significant X-scores were used as predictors in the mixed effect model while leaf N was used as response variable. In the spatial models, error variance was split into structured variance (partial sill) and residual effect. The covariance function was modelled using an isotropic spherical model. Non-spatial models, including same fixed effects of the spatial models, were also computed. All models were computed using PROC MIXED of SAS.

Spatial and OLS models were compared using the likelihood ratio test (Castrignanò et al., 2005) by calculating the -2 Res Log Likelihood. At each stage, selected models were compared through cross validation.

Results and Discussion

According to cross validation and van der Voet's test, three PLS factors were selected at booting, whereas only one at beginning of stem elongation and heading. However, at booting only the first factor was retained in the mixed model.

A different behavior was observed with the advancement of crop growing cycle with regards to both spatial correlation structure of errors and relationships between spectral indices and leaf N.

Overall, a significant spatial covariance structure was observed at the first growth stage, as evident from the significance of the likelihood ratio test and the partial sill, which represents the spatially correlated component of error variance. On the contrary, for the later stages the autocorrelation of errors was significant only at booting for full spectrum and NDVI models.

As regards the fixed effects, at beginning of stem elongation, VIs were not significant for spatial models, whereas they were significant for OLS models when REIP and NDRE were used as predictors. This result underlines that, when spatial dependence is not taken into account, type I error may lead to misinterpretation of results. At booting and heading, the fixed effect was significant for all models. Finally, the fixed effect was significant even at the earliest stage for full-spectrum models.

Cross validation statistics showed that VI and full-spectrum models performed similarly in terms of prediction bias, accuracy and precision at booting and heading, whereas full-spectrum model gave the best results in terms of unbiasedness at the first growth stage.

Conclusions

The detailed information given by HS data requires efficient analysis techniques. In this study, the full-spectrum index was always a significant predictor in leaf N models. At beginning of stem elongation, a critical stage for fertilization management decision, the use of models which take into account spatial correlation can help reduce the probability to make type 1 error and thus allow a better interpretation of the results.

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COMPARING NITROGEN FLUXES IN DIFFERENT CROPPING SYSTEMS DEDICATED TO LIGNOCELLULOSIC BIOMASS PRODUCTION

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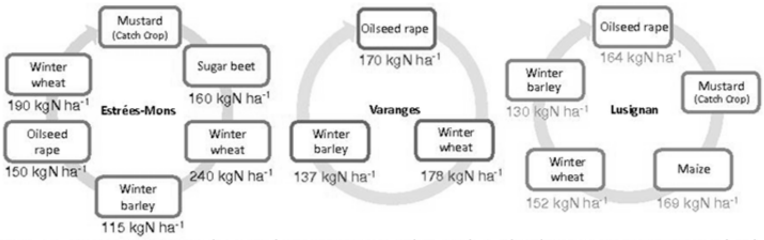

Figure 1: Crop sequences and N fertilisation rates used at each of the three sites in scenarios S1-S2.

	S3	S4	S5	S6
Harvest date	October 15	October 15	March 1	March 1
Fertilisation rate (kg N ha^{-1} yr^{-1})	0	80 to 120	0	50

Table 1: Harvest dates and N fertilisation rates used in all sites in scenarios S3-S6.

	Estrées-Mons	Lusignan	Varanges
S1	0.0	0.0	0.0
S2	4.6	3.4	4.0
S3	16.6	18.2	16.0
S4	21.7	18.9	18.1
S5	17.7	16.0	14.5
S6	18.4	16.1	15.6

Table 2: Annual lignocellulosic biomass production (t DM ha^{-1} yr^{-1}) simulated for the 6 scenarios and the 3 sites (average of 20 years).

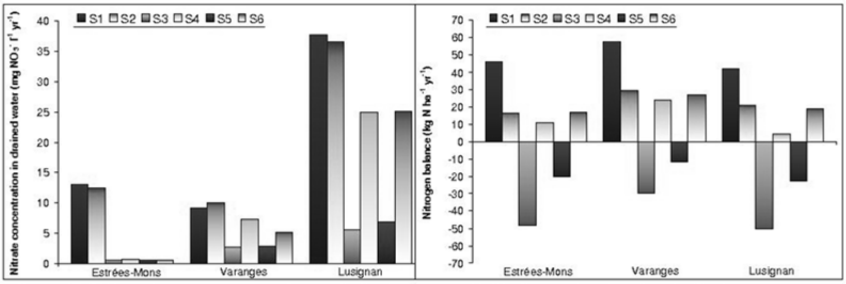

Figure 2: Mean nitrate concentration of drained water and N balance simulated at the 3 sites.

In order to reduce anthropogenic greenhouse gas emissions and replace fossil fuels, policies support the use of crop biomass to produce biofuels. Modelling biomass production and the environmental impacts of cropping systems dedicated to biomass production over the long term is needed in order to evaluate their sustainability. This study aimed at comparing lignocellulosic biomass production and environmental impacts (nitrate concentration in drained water and N balance) of cropping systems based on annual or perennial crops.

Materials and Methods

The STICS model (Brisson et al., 2008) was used in this study in order to simulate both biomass production and environmental impacts of different cropping systems in three sites in France: Estrées-Mons (Northern), Varanges (Eastern) and Lusignan (Western France). The sites varied in climate (temperature, rainfall, solar radiation) and soil characteristics (texture, depth, available water content). Six scenarios were simulated and compared: Scenario S1 with annual crops and no lignocellulosic biomass production (reference scenario); Scenario S2 with annual crops and systematic removal of cereal straw (for biomass use); Scenarios S3-S6 with perennial biomass crops (*Miscanthus* × *giganteus*). The cropping systems with annual crops were selected as representative of local cropping systems (Figure 1). Scenarios S3-S6 varied in harvest dates and N fertilizer rates (Table 1). First year biomass production was not harvested and returned to the field. In scenarios S4 and S6, N fertilisation rates were adapted to roughly compensate the amount of N exported at harvest. In the 6 scenarios, simulations were run continuously during 20 years using the last 20 years meteorological data combined randomly.

Results and Discussion

On average, lignocellulosic biomass production was highest at Estrées-Mons; perennial cropping systems produced the greatest quantity of lignocellulosic biomass (Table 2). Early harvest of *M. giganteus* resulted in a higher biomass production if N fertilization was applied (S4). Nitrate concentration of drained water was highest at Lusignan (Figure 2), the site with the highest rainfall and drainage. Drainage and leaching were little affected by the straw management, either returned or removed (S1 vs S2). Compared to annual systems, perennial cropping systems improved water quality. This is due to the highest nitrogen use efficiency of perennials thanks to rhizomes of *M. giganteus* which allow N recycling (remobilisation and storage) and hence limit its N fertilizer requirements (Strullu et al., 2011). These internal N fluxes are taken into account by the model (Strullu et al., 2014). Concerning the N balance calculated as the difference between N inputs (N fertilization + N deposition) and N outputs (N exported at harvest), the removal of cereal straw lead to a two fold decrease of the N balance in annual cropping systems. In perennials, scenarios S3 and S4 lead to a negative N balance whereas scenarios S4 and S6 (fertilized crops) had a slightly positive N balance. Hence, N fertilisation is required to maintain soil fertility and biomass production of perennial cropping systems on the long term.

Conclusions

We showed that perennial systems with *M. giganteus* can ally high productivity and water quality, due to lower N fertilizer rates than annual cropping systems. Perennials could be used efficiently to protect environmentally sensitive environments. Other scenarios have to be compared and an evaluation at regional scale is needed in order to find equilibrium between food, feed and lignocellulosic biomass to allow a sustainable development of the biomass industry.

Acknowledgements

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NITROGEN UPTAKE AND REMOBILIZATION BY FIELD-GROWN ORANGE TREES

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Oranges represent an important commodity in many Mediterranean Countries. Modern agricultural management techniques, including mineral supply to crops, should reconcile economic and ecological issues. The nitrogen-fertilizers supply at rates and timings that fit the tree nitrogen (N) demand decreases the risk of N leaching and volatilization in orchards. However, not only should yearly N demand data in Citrus be available (Roccuzzo et al., 2012), but they should also be integrated by information about the dynamics of N uptake during the vegetation period. At the moment, this information is mainly available for potted trees (Martínez-Alcántara et al. (2010), while very little is known for field-grown trees.

In this study we followed the uptake of nitrogen provided by fertilizers to orange trees and the within-tree N remobilization the following spring. Trees were fertilized with mineral nitrogen labeled with 15N. All trees received the same amount of N, but the labeled fertilizer was supplied to different set of trees in different periods.

Materials and Methods

We followed the tree uptake and the within-tree allocation of nitrogen derived from fertilizer (Ndff) in orange [Citrus sinensis (L) Osbeck] trees from March 2009 to February 2010. The trees were planted in 1998 at a density of 417 trees/ha in a commercial orchard in the province of Catania, Italy (37° 21' N, 14° 50' E). All trees received the same amount of total nitrogen (150 kg N ha-1 as NH4NO3) in 10 monthly applications starting from March (treatment A), from May (treatment B), from August (treatment C), and from November (treatment D). At monthly intervals, samples of living and abscised leaves, new shoots, old wood and fruits were collected. In winter 2009-2010 coarse and fine roots were sampled at a depth of 60 cm and a distance from the trunk of 1.2 m. The biomass of tree organs derived from direct measurements and estimates by allometric equations (Roccuzzo et al, 2012). Samples were analyzed for total nitrogen concentration and the 15N abundance, which were multiplied by their biomass to obtain the total and the labeled N. Nitrogen derived from fertilizers (Ndff) was calculated by dividing the amount of labeled N by the 15N abundance in the fertilizers. The tree-internal N remobilization in spring (from February to June 2010) was studied by quantifying the amount of 15N in the leaves and branches present in winter 2009/10 and following its appearance in the new shoots, flowers and fruits in 2010.

Results and Discussion

As expected, the tree biomass and N concentration were similar in the 4 treatments. In February 2010 the fruit production averaged 23 Mg FW ha-1 (3.5 Mg DW ha-1).

Total N uptake estimated by the N increase in above ground tree organs averaged 83 kg N ha-1 and about 35% (30 kg of N) of it derived from the fertilizer N supplied in 2009. When available throughout the season (treatment A), the 15N fertilizer was absorbed by trees at a fairly constant rate from May to November 2010, whereas no fertilizer uptake was recorded from November 2009 to February 2010. The later the trees received the fertilized, the lower was the recovery of fertilizer-N in above ground organs: 27% for trees of treatment A (15N from March), a value similar to that reported by Boaretto et al. (2006); 15, 14 and 12% in treatment B (15N from May), C (15N from August) and D (15N supplied in November), respectively.

The allocation of the Ndff to the different organs depended on the timing of uptake. The earlier the 15N fertilizer was given to trees, the higher was its allocation to fruit, while the late absorbed N was mainly allocated to branches and trunk. The N content in both fine and coarse roots derived for approximately 30% from fertilizer N, but the amount of Ndff in roots could not be estimated as root biomass data were not available.

The leaves present in 2009 represented the main winter storage organ for N: its N concentration decreased between February and April from 2.6% to 1.7% and its 15N content in the same period decreased by 50%. Most of the remobilized N reached the shoots, while the contribution of remobilization to fruit N content was minimal. Our field data are consistent with evidences obtained with orange trees in pots by Martínez-Alcántara et al. (2010).

Conclusions

In this study, we quantified the uptake and the remobilization of nitrogen on the same adult trees under field conditions. Out of a total estimated uptake of 83 kg N ha-1, about 35% derived from the fertilizer N supply and about 18% was accounted by the internal N remobilization from winter storage.

Even if the N fertilizer supply was split into several applications during the season, the efficiency of its recovery in the above ground organs was relatively low. It should also be stressed that we could not assess the amount of Ndff retained in the root system, although, roughly 30% of root nitrogen derived from the fertilizer. Root N uptake occurred at similar rate from March until November but no winter N uptake was observed. Most of the N recovered in the developing shoots in May derived from its remobilization from old leaves before they started their senescence and abscission.

Acknowledgements

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MODELLING WATER AND N IN LISBON URBAN FARMS

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In Lisbon a recent survey identified around 77 ha of cultivated allotments under the municipal regulations (Cabannes & Raposo, 2013) where soil fertility is maintained by large amounts of organic fertilizers. It is important to assess the environmental impacts of the current practices where plots tend to be small leading to over application. Knowledge on the availability of mineral N is important for the optimization of benefits (Gutser et al., 2005). The C:N ratio of the material affects the process as well as abiotic factors such as soil water and temperature. So, simulation models coupling the conceptual descriptions of water, heat and C-N dynamics provide a unique means of addressing these issues (Gabrielle et al., 2005). The objectives of the study were to characterize the actual agricultural practices related to irrigation and fertilization in case-study urban agriculture allotments (UA and to propose alternative practices to minimize N losses. The methodology integrates field experiments and modelling. The agricultural system Root Zone Water Quality Model (RZWQM) (Ahuja et al., 2000) was used.

Materials and methods

During November 2011, an enquiry was carried out to a group of 23 allotments in Lisbon. A group of farmers was selected based upon their willingness to communicate. Two study case allotments were chosen for the present study: Granja Conv (GC) where mineral fertilisers are complemented by organic amendments; and Ajuda Org (AO) where only organic fertilizers are applied. GC is installed in moderately permeable soils, while the Ajuda soils present low infiltration rates. Soil samples were collected for the soil physical properties and OM. Soil water contents were determined by the gravimetric method. Samples from the irrigation water sources were analysed for $N\text{-}NO_3$.

The crops were irrigated year round. The organic amendments were applied in early spring at the time of planting. A mineral fertilization of 150 kg N ha^{-1} was applied in GC 20 days after planting. The organic C and NH_4^+ contents and the C:N of the amendments were calculated according with the proportion of each component (Abbasi et al, 2007).

Results and discussion

In GC (tomato- pumpkin- fava bean) 300 mm of water were drained, representing 26% of total inputs. N inputs included the N released from organic amendments and from the chemical fertilizer and the N transported with the irrigation water. No leaching occurred during the tomato cycle due to its deep roots and high uptake potential. However, due to the slow N release the tomato plants suffered some deficit of N. About 36% of the total N provided was lost. Nitrate storage in the soil increased during the cultural year by 91 kg ha^{-1}.

In AO (cabbage-pumpkin, lettuce-lettuce) drainage and runoff corresponded to 12 and 4% respectively of the total inputs. The N concentration in irrigation water was high (46.8 mg NO_3^- L^{-1}) imposing an important N load on the system. Due to the low C:N, N is provided at a rate higher than crop uptake. NO_3-N accumulates and storage reaches 500 kg ha^{-1}. The N budget for the 1-year cycle shows that 40% of the N inputs are lost. During the cultural year, the nitrate storage in the soil increased by 480 kg ha^{-1}.

For both cases the N inputs were higher than crop uptake, generating N surpluses lost by different processes. The main causes are the non-fertilizer sources of N and the C:N of the organic amendments. In GC the main N loss process is the convective transport with the drainage water. The water balance calculations indicate that 300 L m^{-2} $year^{-1}$ are drained out of the root zone, with an average N concentration of 358.7 mg NO_3^- L^{-1}. Other loss processes are not significant due to the rapid N movement in depth. Since drainage is reduced in the Ajuda soils, they tend to accumulate water and nitrates in the profile. The low infiltration rate introduces the potential for runof losses for both water and N. Nevertheless the major loss process in AO is the gaseous loss. In a medium to long term basis, the nitrate accumulated in the lower soil layers will eventually be leached into groundwater.

Conclusions

– RZWQM predicted soil water with an average RMSE of 5.5 %, adequate to the proposed objectives

– N availability per ton of manure was estimated as 9.2 and 20.1 kg ton^{-1} $year^{-1}$ for GC and AO;

– Annual deep percolation was 300 mm for GC, with an average concentration of 359 mg NO_3^- L^{-1};

– Ajuda presented a large N surplus which is accumulating in the lower depths (518 kg NO_3^- ha^{-1}). The impact of the cumulative surplus consists in a potential groundwater contamination risk.

– To make efficient use of the N in the manure, there is the need to understand the dynamics of mineral N release from the organic forms, so modelling is suggested.

The following techniques for the reduction of leaching are advised:

– Adjust manure applications to the EU limit of 170 kg N ha^{-1}

– Use mechanical manometers to estimate the irrigation opportunity;

– Reduce N surplus by considering the N load in the irrigation water and the N resulting from mineralization.

– Adjust the C:N of the organic amendments.

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MANAGING SPATIAL AND TEMPORAL UNCERTAINTIES IN ACTUAL AND FORECAST RAINFALL FOR OPTIMAL IN-SEASON NITROGEN FERTILIZER RATES RECOMMENDATIONS IN CORN

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Corn growers do not appreciate the temporal processes involved in determining seasonal N needs and use "insurance levels" based on the worst-case scenario. Key relationships, involving the interaction of seasonal weather with soil texture, that condition the impact of nitrogen (N) rate on corn yield production have been formalized in Tremblay et al. (2012). These relationships have been quantified to optimize in-season N applications at a North American scale based on soil texture and rainfall (actual + forecast accumulation and distribution over time) and inserted into "SCAN", a decision-support system to optimize in-season N fertilization. Indeed, a study of the Shannon Diversity Index (SDI) revealed that not only rainfall accumulation, but also its distribution over time has as an impact on corn yield response to N rate. A new parameter (named "Abundant and Well-Distributed Rainfall" [AWDR]) was created from the product of rainfall accumulation (PPT) with SDI. This new parameter had a higher correlation with the N rate effect than either PPT or SDI alone, and therefore constituted a promising integrative parameter for rainfall. However, there is uncertainty associated to: 1) the spatial accuracy of interpolated actual rainfall (established between day -15 to day 0 of in-season N fertilizer application) and 2) the forecast rainfall events (quantity at each day in a period theoritically established from day +1 to day +30). This uncertainty requires the use of a security margin in N fertilizer application (extra amount of N) to cover for the risk of error in the estimation of actual and forecast rainfall. On one hand, any too conservative evaluation of rainfall will result in yield losses due to N shortage, particularly on soils characterized by clayey surface textures. On the other hand, the N security margin should be reduced to a minimum in order to avoid losses of N harmful to the environment (water and air). The goal of this study was to evaluate the temporal and spatial uncertainties related to the PPT, SDI, and AWDR in the -15 to +30 days around in-season N fertilizer application), to examine the consequences on N rates recommendations for soils of different surface texture properties and to propose a strategy for handling these uncertainties.

Weather stations with rain gauges were distributed at key locations in the region and were used as a reference. The variograms obtained from the 28 weather stations collecting daily parameters in the Montérégie region of Quebec in 2013 revealed that for a period of 45 days (-15 to +30 around in-season fertilizer application), the error of estimation amounts to 15 mm of rain within distances from 20 to 40 km, and to 22 mm for distances around 60 km. This corresponds to errors between 8 and 11% for the rainfall parameter used to calculate optimal N rates in the SCAN model. Another set of data was extracted for the years 2004 to 2013 from 13 weather stations in the Montérégie region. The comparison of variograms from the different years shows that there are no clear trends among years for a location to be "wetter" or "drier" than other locations.

The current official rainfall forecast system available from the Meteorological Service of Canada does not yet provide sufficient detailed information for the total 30 day period that would be theoritically useful for N rates calculations. However, the North American Ensemble Forecast System (NAEFS) produces 15 day probabilistic forecasts at a spatial resolution of about 100 km. The archived NAEFS forecasts (June and July) of the years 2012 and 2013 were tested on their reliability for optimal N rates calculations. When only the 10 day forecasts were evaluated, they were found to introduce a 10% (in 2012) or 20% (in 2013) error in the rainfall parameter used to calculate optimal N rates in the SCAN model. However, when the (known) -15 day period is integrated into the calculation (-15 to +10 days), the errors corresponded to 6 and 10%, in 2012 and 2013 respectively.

A simulation was conducted to determine the consequences of the rainfall parameter's uncertainties on the recommended N rates by the SCAN model. The simulations were made on rainfall conditions within 35% and 85% of the maximum historical rainfall experienced. They covered two types of soils (medium/coarse textures and fine textures). The uncertainties generated by a 10% uncertainty (considered reasonable as per the demonstration above) in the rainfall parameter leads to a 5 kg N ha^{-1} difference in the recommendation for the more tolerant soils (medium/coarse) and a 10 kg N ha^{-1} difference for the less tolerant soils (fine). On average, for the conditions prevailing historically in the targeted region, the error margin in the optimal N rate recommendation would reach 7 kg N ha^{-1}.

Farmers are reluctant to risk yield loss from N rates that are too low. High N application rates to reach yield potential are justified only under particular soil/seasonal weather circumstances and yet the farmers tend to aim systematically for those infrequent rewards. Using the N responses relationships established in the SCAN system and the rain (actual and forecast [+10 days]) information available in real time, a security margin of 5 to 10 kg N ha^{-1} would be warranted depending on soil type in order to secure yield production in the case of unexpected rainfall patterns. This constitutes a considerable improvement on the current situation at the North-American level where growers have no option to adjust N rates to seasonal conditions which have been documented to range within 100 kg N ha^{-1} among years. The work to reduce spatial and temporal uncertainties in actual and forecast rainfall is going on.

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ARE FUNCTIONAL TRAITS RELEVANT TO CHARACTERISE PLANT STRATEGIES OF COVER CROP SPECIES?

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Cover crops are used during fallow period between two main crops to reduce nitrate leaching, soil erosion, increase carbon storage and provide "green manure" effect (increase nitrogen (N) availability for next crops) [1]. Many plant species from various botanical families can be used to provide these ecosystem services related to N management. However the rules leading to correctly choose which species to produce attempted catch crop or/and green manure services are still to define. We tested the relevance of a trait-based approach to address this issue. Contrary to "classic" agronomical methods which characterise precisely a small number of species, a trait-based approach would allow to quickly characterise a large number of species based on their resource use strategies. For example, species with high Specific Leaf Area (SLA) and low Leaf Dry Matter Content (LDMC) correspond to species with high rates of resource acquisition, early growth, high level of primary production, for which we expect a high and early rate of N acquisition. Another approach is to classify species according to the CSR scheme proposed by Grime (1977), according to their competitive ability (C), stress tolerance (S) and ruderality (R). We tested the relevance of using functional traits and associated methods to characterise and explore the wide variability of cover crop species to provide ecosystem services for N management.

Materials and Methods

Thirty-six cover crops species were selected, among six botanical families, according to their ability to rapidly catch soil nitrate and produce green manure effects (low C: N ratio) during the limited length of fallow period. All species were grown in the same conditions in field under non-limiting water and N conditions in order to test whether species rankings based on traits are reliable across environments. Two field experiments were conducted in autumn 2012 at INRA site in Auzeville (southwest of France) and at Agroscope site in Nyon (Switzerland). For each species we measured leaf functional traits, i.e. SLA, LDMC and Leaf Nitrogen Concentration (LNC), on 20 individuals in vegetative stage of each species.

Results and discussion

The rankings of species for SLA, LDMC and LNC were found to be conserved between the two experimental sites, highlighting the strength of the functional characterisation (results not shown). We therefore merged the two data sets for the added analysis based on concepts developed in ecology. Figure 1 presents the positioning of the 36 species on the relationship between SLA and LDMC [3]. All species present high SLA and low LDMC, they are characterised by a fast resources capture strategy. Consequently, this representation did not appear relevant to precisely distinguish differences in their strategies. Crucifers show low SLA and LDMC, which is not consistent with the negative SLA-LDMC correlation established on wild species from natural ecosystems by Garnier et al. (2001). This can be explained by a domestication effect which would have strongly modified values and relationships between traits. Indeed, species domestication tends to maximise resource acquisition and especially N capture, in comparison to corresponding wild species. We also tested the CSR method [2] as an approach to ordinate species. The Figure 2 shows that cover crop species are not classified in the same strategy, even if they are annual and domesticated. Crucifers are classified as competitive; it was also the species providing the quickest and highest biomass production and the highest uptake of soil mineral N in field experiments. Grasses and legumes, which produced less biomass and uptake less mineral N, are classified as intermediate C-R type in this triangle. Even if Grime's triangle help to explore cover crop species diversity, this method is not precise enough to characterise species in function of related N ecosystem services produced.

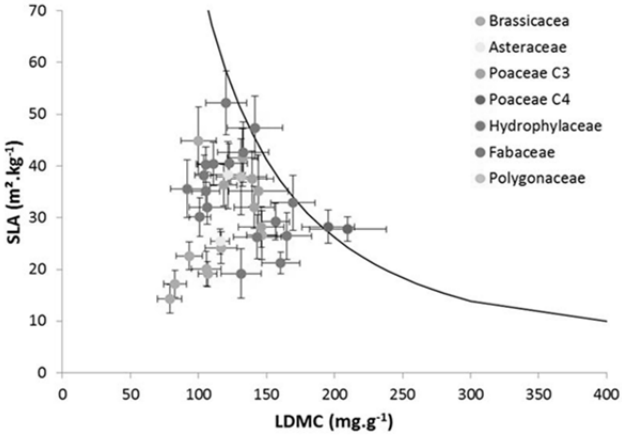

Figure 1: Relationship between SLA and LDMC for 36 cover crops species

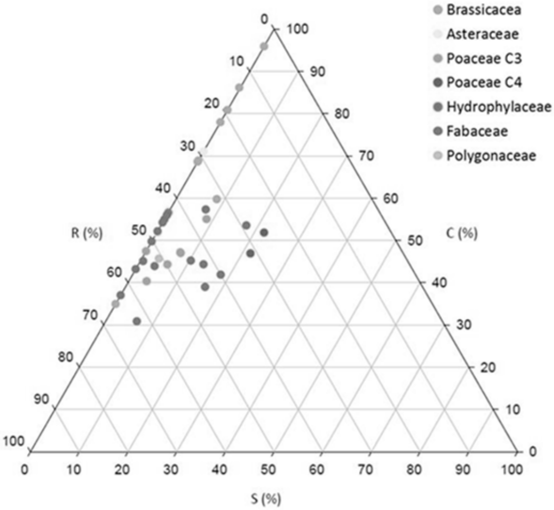

Figure 2: CSR triangle, species ranking on three axes Competition (C%), Stress tolerance (S%) and Ruderality (R%).

Conclusions

Leaf functional traits and associated methods based on plant strategies can help to explore ex ante the cover crop species diversity in order to express their ability to produce ecosystem services such as those related to N management. Even if domestication effect modified cover crops traits values and traits syndromes, some differences between species have been pointed out by functional approach, corresponding to well-known agronomical characteristics. In order, to increase the precision of species characterisation, we propose to combine functional traits and, what we propose to call "cover traits" measured at the cover level (e.g. height, growth rate, root depth, etc…).

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DESIGN OF BISPECIFIC LEGUME/NON-LEGUME COVER CROP MIXTURES TO PRODUCE VARIOUS ECOSYSTEM SERVICES RELATED TO NITROGEN MANAGEMENT

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In France and Europe, groundwater pollution is caused by nitrate (NO_3^-) leaching because of high fertiliser-N inputs and soil mineralization of organic matters. To reduce this pollution, Nitrate Directive (1991) and French Government circular (2008) established an obligation of cover cropping during long fallow period in vulnerable areas. Cover crops are used to produce various ecosystem services (ES) such as i) catch the largest possible amount of soil nitrate before the drainage period (catch crop), ii) allow the highest N release for the succeeding main crop (green manure), and iii) increase soil organic-N content in soil and produce C sequestration [1]. Mixing or intercropping (IC) different species could be an efficient way to maximise ES thanks to complementarity between legume and non-legume species. Crucifers, grasses or other non-legume families would be more efficient to catch mineral N (minN), rather than legume species increase the green manure effect thanks to atmospheric-N symbiotic fixation. Designing efficient species mixtures is needed in order to favour complementary between the two species. There is a need of conceptualisation and optimisation of generic rules for choosing species in mixtures depending on the length and management of the fallow period.

Materials and methods

A field experiment was conducted in autumn 2012 at INRA site in Auzeville, southwest France. Twenty-five IC of 1 legume/1 non-legume and 10 corresponding sole crops (SC) have been studied. Species were chosen by preliminary expertise for their early growth rate in order to quickly produce targeted ES. Each non-legume was intercropped with each legume with an IC density of ½ SC densities for both species. All modalities were sown on 16 August in field plots repeated in 3 randomized blocks. Before sowing, soil minN content was 50 kg $N.ha^{-1}$ in 0-120 cm depth. Samplings were done in November in order to analysed soil minN content, N acquired amount for each species in IC and SC, N2 fixed by legumes and C: N ratio of the different covers. The “Land Equivalent Ratio” (LER), an indicator of species performances in mixture, was calculated. LER is the sum of partial LER for each species which is calculated as the N acquired amount by the species reached in IC divided by those of the same species obtained in SC [2].

Results and Discussion

Concerning soil minN content (Fig 1), the mean for all modalities was 36 kg Nha^{-1}. It was significantly lower than the bare soil (93 kg $N.ha^{-1}$), indicating that all modalities acquired nitrate. Globally legumes SC acquired less minN than non-legumes. However, some legumes SC such as vetch and crimson clover are able to catch approximately the same minN amount than grasses SC. Mixtures showed minN amounts lower than legumes SC but they were closed to non-legumes SC minN contents. Even if in IC there was only the half density of non-legume, legumes up taken also soil minN, leading IC to be as much efficient as non-legumes SC. Figure 2 shows that almost all IC reached a total LER > 1, meaning that IC have better resources valorisation compared to SC. Ryegrass/vetch, foxtail millet/pea and ryegrass/pea IC presented LERp of both species >0.5, meaning that both species are advantaged in mixtures: interspecific competition was lower than intraspecific competition.

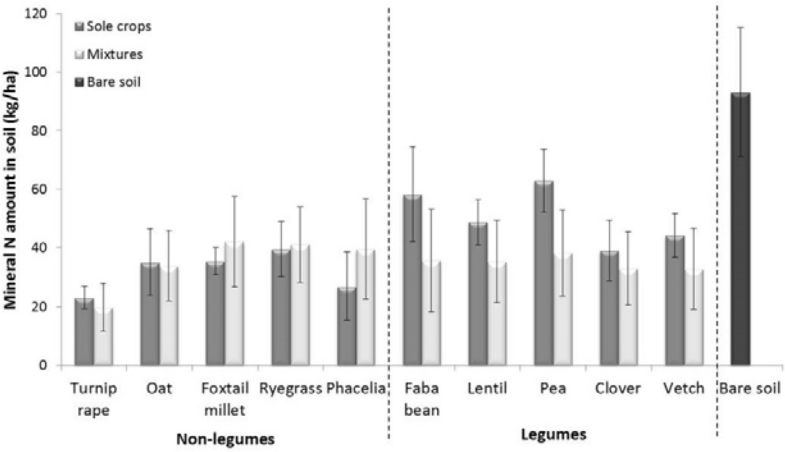

Figure 1: Soil mineral nitrogen amount in November for species in mixtures, sole crops and bare soil

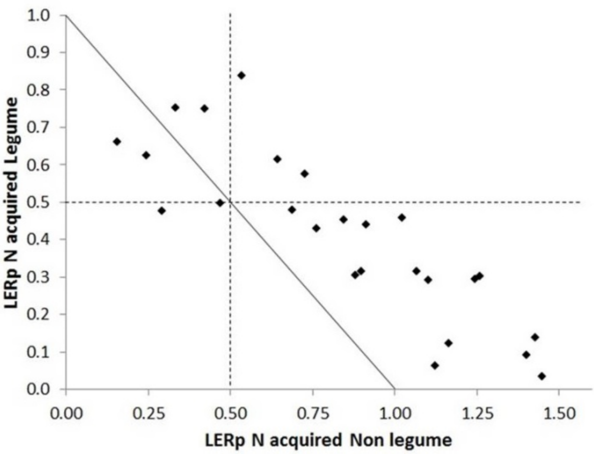

Figure 2: LER of mixtures in function of partial LER of legumes and non-legumes

They jointly maximised catch crop and green manure effects thanks to a good complementarity between species and probably some facilitation effects. However, numerous IC advantaged catch crop effect, because non-legume species were globally more competitive (LERp>0.5) than legume (LERp< 0.5). For example, turnip rape was advantaged in IC because it is a crucifer very competitive and with probably allelopathic negative effects on its intercropped species. Two IC favoured green manure effect because the legume dominated the non-legume species, i.e. ryegrass/faba bean and phacelia/pea. The legumes obtained the largest amount of N acquired though they presented the highest level of minN in November; this means that symbiotic N2 fixation was efficient even for a short period of growth. Moreover, the C: N ratio is an indicator of the mineralisation rate of the cover crop residues [3]. IC with a legume decrease the C: N ratio and maximise the green manure effect compared to grasses or crucifers SC. The study of species complementarity and ES maximisation can help to design choice of species in IC according to the objectives of fallow period management. For example, after a main crop without any fertilisation-N and thus low minN content at harvest, it could be interesting to choose a mixture maximising green manure effect.

Conclusions

Strong differences of behaviour of species cropped in IC have been shown, indicating that a consistent work is needed to make a relevant choice of species associated. Moreover, interspecific interactions between species evolve in time; it could be interesting to adapt IC to the length of fallow period. A relay intercropping with a non-legume very sensitive to frost and a legume slower to growth but frost resistant could be a way to maximise ES according over time.

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ENHANCED NITROGEN AVAILABILITY HAS AN IMPACT ON EXTRACELLULAR ENZYME ACTIVITY IN THE RHIZOPLANE OF CISTUS LADANIFER BUT NOT IN THE SURROUNDING SOIL.

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Increasing deposition of reactive nitrogen (N) is a multifaceted global problem, recognized as a major threat to ecosystems' stability. NHy derives mainly from agricultural activities, while NOx originates in urban/industrial activities. Increased N availability can alter nutrient cycling and lead to various feedbacks between soil microbiota and plants, disrupting ecosystem function. Even though it is accelerating in all parts of the world, most studies of the problem have been conducted in northern European and American ecosystems. To gain more insight into the effects of increased N availability on Mediterranean ecosystems, an N-manipulation field experiment has been carried out in the Arrábida Natural Park (Portugal) since 2007 (Dias et al., 2011). Since Mediterranean ecosystems are co-limited by N and phosphorus (P), increasing N availability is expected to exacerbate the limitation of P. An increase in phosphatase activity could help the system to overcome this limitation. However, the production and maintenance of phosphatase activity will in turn require more N that can, if not inorganic, only be supplied by the degradation of N-containing macromolecules. In order to explore this hypothesis, the extracellular enzyme activity (EEA) patterns of the dominant plant species (C.ladanifer) roots and surrounding soil were compared.

Materials and Methods

The study site is located in Arrábida Natural Park, Serra da Arrábida, Portugal (38°29'N – 9°01'W). Vegetation consists of a dense maquis with skeletal soil (15-20 cm deep). An on-going nitrogen addition experiment was started in January 2007 using 3 treatment types: 40 and 80 kg N ha-1 yr-1 of ammonium nitrate (NH4NO3) (40 AN and 80 AN) and 40 kg N ha-1 yr-1 of ammonium (1:1 mix of NH4Cl and (NH4)2SO4) (40 A). Each treatment has 3 replicates (400 m2 plots). From the resulting 12 plots (3 control plots), main root systems and attached bulk soil (ca. 20cm x 20cm volume) of 3 C. ladanifer plants with ca. 50 cm height were sampled in June 2013. Samples were transported as bulk agglomerate into the lab and stored in the dark at room temperature (ca. 23°C) until analysis. Soil was sampled from an interior part of the agglomerate, homogenized and used for enzymatic analysis. The roots were washed and were never stored more than 24 hours before analysis to avoid alteration of activity profiles. A novel technique for EEA profiling was used to assess the activity of acid phosphatase and N-related enzymes (N-Acetylglucosaminidase and Leucine aminopeptidase) in roots and soil. This technique allows sequential analysis of several enzyme activities on the same sample and makes use of high throughput filter bottom well plates and synthetic fluorescent substrates. The fluorescence emitted by the respective enzyme product was calibrated against a standard curve and calculated per g dry weight of sample. Finally, the proportions of root and soil were determined per 100 g agglomerate to extrapolate the total turnover of each compartment.

Results and Discussion

Interestingly, the total EEAs expressed per 100 g agglomerate are similar between soil and root compartment, even though the latter only contributes 2% to the total. This highlights the importance of the root surface (rhizoplane) as a compartment for nutrient turnover and plant nutrition. Moreover, the Pearson correlations in graph 1 show that the clear relationship observed in the control between N-related enzymes and phosphatase activity is lost with N addition. In contrast, this relationship is maintained in the soil (graph 2), except for that subject to the 80 AN treatment. As the great majority of the applied N has moved from the abiotic soil compartment into the biotic compartment during the spring months (Dias et al., 2012), the plant probably has enough N available to invest in rhizoplane phosphatases or provide microorganisms with the nutrients needed to exhibit higher activities. In contrast, the soil microorganisms are relying on N-containing polymers, as the inorganic N is mainly stored in the plants. The only treatment that has shown significantly higher inorganic N levels in previous studies (Dias et al., 2012) is the 80 AN treatment, thus possibly explaining the diminished relationship.

Conclusions

Despite being very little represented in the soil-roots matrix, the rhizoplane EEA contributes as much as all the surrounding soil to nutrients turnover. The additions of N decoupled the relation between activities of phosphatase and N-related enzymes in the rhizoplane but not in the bulk soil.

AcknowledgementsThis study was supported by the Fundação para a Ciência e Tecnologia (FCT) through the project PTDC/BIA-ECS/122214/2010. We are grateful to Arrábida Natural Park for making the experimental site available and allowing the N manipulation experiment to which this abstract refers.

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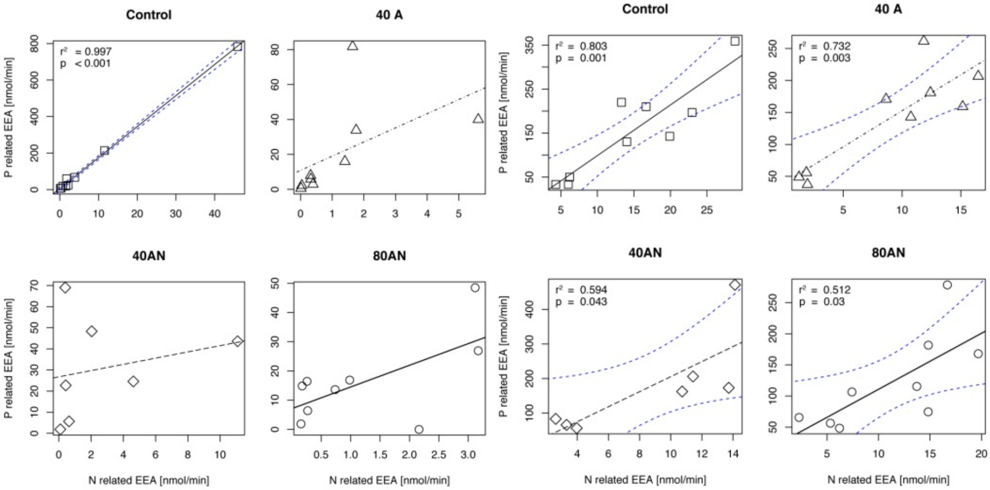

CLIMATE CHANGE AND NITROGEN FERTILIZATION FOR WINTER DURUM WHEAT AND TOMATO CULTIVATED IN SOUTHERN ITALY

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Climate change (CC) is playing an important role on agricultural productions. The climate modifications will have a great impact also on relationships between the agricultural processes and principal cycles of environmental and productive factors, as water and N. The interest of this paper is focused on "Capitanata area", a plain of about 4000 km^2 located in the northern part of the Apulia Region in Southern Italy, characterized by farms with highly productive soils cultivated in intensive and irrigated regime and with the winter durum wheat (TO: *Triticum durum*L.) and tomato (TO: *Lycopersicon aesculentum* Mill.). The main interest was to evaluate WW and TO responses under several future climatic scenarios for productive parameters. A complete description of the results is reported in Ventrella et al. (2012). We present an analysis related to the effects of CC on N utilization and management in WW and TO cultivation in Southern Italy.

Materials and Methods

The details of the geostatistical analysis and the downscaling techniques are descripted in Ventrella et al. 2012. Here we considered just the central area of the Capitanata area. Two IPCC future climate scenarios (B1 and A2, considered as "family scenario") were used for evaluating the impact of future CC, taking into account the progressive increase of atmospheric CO_2 concentration respect to the pre-industrial level. In this paper the Hadley Centre Coupled Model (HadCM3) is considered dividing the future climate scenarios in three time slices (I: 2011-2040; II: 2041-2070 and III: 2071-2100) and comparing them with a generated baseline scenario (REF: 1970-2000). DSSAT, a cropping system model that allows to predict and to interpret the behavior of the agronomic system, was used in order to simulate the cultivation of WW and TO in a two-year crop rotation. The soil water balance was considered according to the cascading method and the organic matter turnover and N balance were simulated adopting the CENTURY-based module available into DSSAT. In the experimental layout, we considered a fertilization of N consisting in 120 and 200 kg ha^{-1} for WW and TO, respectively, in two time applications (pre-sowing and top dressing). In order to assess the impact of CC on N fluxes of the system "soil-plant" the following indicators were considered: yearly uptake and leaching of N (kg of N ha^{-1}), N Use efficiency for above-ground biomass (kg of dry matter / kg of N uptake). For each of these parameters the SAS box-plot procedure was applied in order to describe the statistical distribution and the relative graphs were carried out.

Results and Discussion

Figures 1 and 2 report the box-plots related to statistical distribution for WW and TO, respectively, for the three indicator considered in this paper: uptake and leaching of N and NUE. For each box-plot, the edge delimits the Interquartile Range (IQR) as defined by the 25th and 75th percentiles and include the mean (symbol) and median (line). The endpoints of upper and lower whiskers indicate the maximum and minimum observations in the range of 1.5*IQR above and below the 75^{th} and 25^{th} percentile. The outliers beyond the 1.5*IQR are indicated by symbols. A first consideration is that the amounts of uptake and leaching are significantly lower for the cultivation of WW compared to those of TO typically submitted to intensive irrigation regime. The WW averages of uptake and leaching are approximately 60 and 30 kg ha^{-1}, against the corresponding average values of TO which reach thresholds of 300 and 100 kg ha^{-1}. For such indicators related to WW, significant trends, going from reference period to future scenarios, are not evident, both for scenario B1 to A2. The main difference between uptake and leaching is that for the second indicator the distribution tends to be more positively skewed, with the box shifted significantly to the low end and with more numerous outliers. Such consideration about the statistical distribution are much more noticeable in the case of TO. Moreover, for such spring crop, a significant trend is evident going from reference period to the end of this century, with the average uptake that increases of about 25% while for leaching much more significant increases can be observed. As expected, and contrary to the trends of the indicators described above, NUE for biomass is significantly higher for wheat with values often higher than 60 kg kg^{-1}, while for TO the NUE is usually lower than 50 kg kg^{-1}. Moreover, for the spring crop a decreased linearly trend is evident with a threshold value of about 30 kg kg^{-1} expected in the last 30 years of this century.

Conclusions

The CC forecasted for this century, are expected to determine important variations on the nitrogen cycle and management, depending on type of crop and cropping systems. The winter crop as WW, in the Mediterranean environment, seems to be minus sensitive to CC compared to the spring crops like TO, whose crop cycle is reduced by the warming induced by CC. In this paper such a consideration was confirmed for all three indicators related to Nitrogen cycle and management. For TO cultivation, the statistical distribution are expected to be more asymmetrical because of more frequent extreme values in particular for leaching fluxes due to the heavy rainfalls characterizing the future climate.

Acknowledgement

The results were obtained within the international research FACCE-JPI project MACSUR (Modelling European Agriculture with Climate Change for Food Security).

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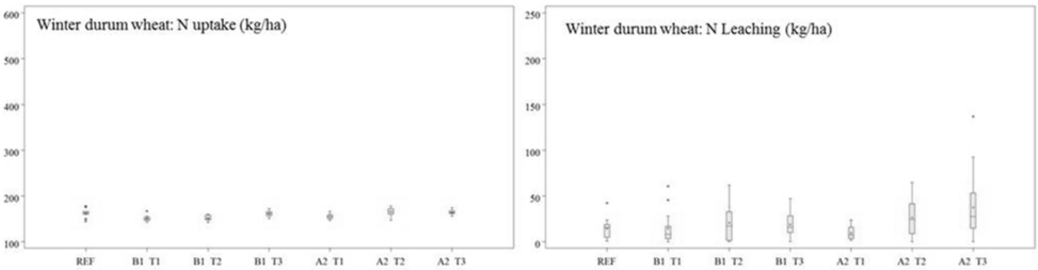


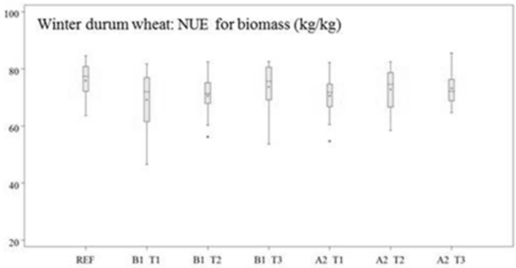
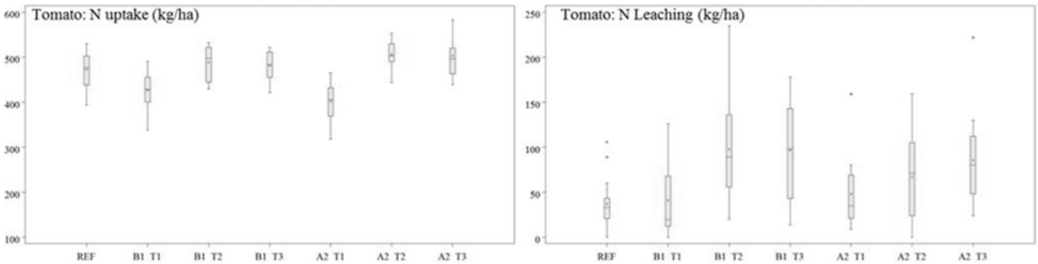


Figure 1 – Statistical distribution of simulated N indicator related to climate change scenarios for winter durum wheat cultivation in Southern Italy . REF is the reference period (1970-2000), B1 and A2 are the IPCC scenarios, T1, T2 and T3 refer to 30-year-periods from 2010 to 2100.



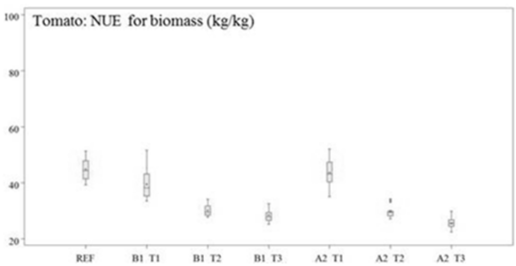


Figure 2 – Statistical distribution of simulated N indicator related to climate change scenarios for tomato cultivation in Southern Italy . REF is the reference period (1970-2000), B1 and A2 are the IPCC scenarios, T1, T2 and T3 refer to 30-year-periods from 2010 to 2100.

THE EFFECT OF NEW N FERTILIZERS ON YIELD ND GRAIN NITROGEN IN WINTER WHEAT

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To improve the efficiency of N fertilizers, urease inhibitors (that limits urease activity) and nitrification inhibitors (NI) (that restrict the microbial conversion of ammonium to nitrate) had been used in agriculture over the last few decades. Because new fertilizers containing these compounds had been developed recently, it is necessary to quantify its effectiveness in different crops, soil and climate conditions. Previous works with the nitrification inhibitor DMPP resulted in an improvement of N recovery in irrigated wheat (Villar & Guillaumes, 2010). A recent work by Liu et al. (2013) has demonstrated that NI enhance yield, nitrogen use efficiency and reduces N_2O emissions in a wheat-maize system. The objective of this study was to quantify the effect of new N fertilizers containing urease inhibitor or nitrification inhibitor on wheat yield and wheat protein.

Materials and Methods

This study was conducted at a commercial farm (Mas Lleó, Lleida, Spain) during 2012-2013 crop season. The soil was a Petrocalcic calcixerept (SSS, 1999). The soil texture was loam (USDA). Winter wheat (*Triticum aestivum* L., cv. Gades) was planted on 28th Nov. Harvest occurred on 10th July 2013. Rainfall during this period was 396 mm, and ETo 716 mm.

A sprinkler system provided 90 mm of irrigation water. The experimental design was a randomized complete block with four replications. Individual plots were 7 m wide and 9 m long. 9 treatments were tested (Table 1). Nitrogen was applied only at early-tillering (26th Feb). Apparent nitrogen recovery (NREC) is the additional N uptake per unit of added nutrient (kg kg^{-1}) and was calculated as described by Greenwood and Draycott (1989). Grain yields were adjusted for moisture of 135 gkg^{-1}. Volumetric grain weight was measured on a grain sample of each plot using a grain analysis computer mod. GAC II (Dickey-John, USA). The weight of thousand kernels was determined with seed counter Numigral (Sinar Technology, UK). N concentration was determined by Kjeldahl digestion. Statistical analysis of the results was performed using the JMP-SAS software (JMP®, Version 9. SAS Institute Inc., Cary, NC, 1989-2010). Means were separated using Student's t-test with 0.05 of significance level.

Results and Discussion

Residual soil N (NO3-N content) at the beginning of the experiment (0-40cm) was on average low (12.0±6.27) without significant differences at a depth of 0-40 cm. Residual soil N (NO_3-N content) at harvest (0-40cm) was on average very low (8.8±9.65) without significant differences at a depth of 0-40 cm. Table 1 presents the main results of this experiment. Grain yield, grain N uptake and total N above ground biomass were significantly ($p < 0.05$) lower for the unfertilized treatment. The inhibitors treatments significantly ($p < 0.05$) increased N uptake, both grain and above ground biomass. Higher dose of fertilizer (90 kg Nha^{-1}) resulted also in higher grain yield and N uptake. No significant differences were found between treatments for harvest index (HI) ($p = 0.3383$). On average HI was 0.45.

Table 1 ANOVA (p>F) for wheat yields, volumetric grain weight, thousand kernel weight, grain N uptake, straw N uptake, total N uptake, and apparent recovery of total N.

Source	N rate $kgha^{-1}$	Grain yield (13.5% moisture content) ($kgha^{-1}$)	Volumetric grain weight ($kghL^{-1}$)	Kernel weight (mg)	Grain N uptake ($kgha^{-1}$)	Straw N uptake ($kgha^{-1}$)	Total N above ground biomass ($kgha^{-1}$)	Apparent recovery of total N (%)
Control	0	5743 d	81.0	41.3	109.50 d	29.3	138.79 d	-
Urea	72	6277 cd	80.2	42.1	128.02 c	37.2	165.26 c	26
Urea	90	7105 a	80.6	40.8	144.03 a	42.2	186.21 a	38
UTEC	72	6706 abc	80.4	41.9	134.84 abc	37.4	172.22 abc	35
UTEC	90	6894 ab	80.3	40.6	140.46 ab	42.5	182.91 ab	34
CAN	72	6502 bc	80.6	40.4	128.34 c	38.5	166.86 bc	26
CAN	90	6638 abc	80.0	40.2	134.27 abc	41.7	175.99 abc	28
CAN NI	72	6932 ab	80.3	41.0	136.29 abc	39.6	175.87 abc	28
CAN NI	90	7051 ab	80.5	41.1	142.69 a	45.5	188.22 a	37
Treatment		0.0012	0.5446 NS	0.5646	0.0001	0.2098	0.0001	0.6641
Contrast Higher dose vs lower dose Inhibitors vs No inhibitors		 0.0345* 0.0757	 NS NS	 NS NS	 0.0057* 0.0209*	 NS NS	 0.0016* 0.0225*	 NS NS

Control unfertilized; UTEC Urea plus Urease inhibitor NBPT; CAN Calcium Ammonium Nitrate 27; NI New Nitrification inhibitor.

The highest volumetric grain weight (VGW) was inversely related with lower yield, although not significant results were found. Average VGW was 80.4 $kghL^{-1}$. On average, the kernel weight was 41 mg and differences were not significant between treatments. The straw N uptake was, on average, 39.3 $kgNha^{-1}$ without significant differences between all treatments. Straw N uptake was 22.8 % of total N uptake. The quantities of nitrogen fertilizer used in this experiment should be considered slightly limiting and probably more irrigation water was needed. The apparent recovery of N (NREC) by the crop was not significantly different between treatments and, on average, lower than 40%.

Conclusions

The new inhibitors should be considered as potentially useful tools to improve N effectiveness of mineral fertilizers as urea and calcium ammonium nitrate. Further research is required using higher Nitrogen doses and crop yield objective in order to assess the potential use of these new compounds.

Acknowledgements

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EFFECTS OF FERTILISER NITROGEN MANAGEMENT ON NITRATE LEACHING RISK FROM GRAZED DAIRY PASTURE IN NEW ZEALAND

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Current and proposed regulations across the world, including New Zealand, are increasing the pressure on intensive pastoral dairy farming to adopt systems and technologies that increase productivity to maintain economic viability, while reducing environmental impacts, including nitrogen (N) leaching. Increasingly N fertiliser is being used, with typical application rates on dairy farms of 100-200 kg N/ha per annum. The efficiency of such nitrogen fertilisation is influenced by environmental and management practices. Generally, N efficiency decreases as N fertiliser inputs increase, and N losses to the environment increase. Risk of loss of N fertiliser by direct leaching can, to a large extent, be managed by good fertiliser practice. The main source for N losses in grazed pastures is leaching from urine patches. Nitrogen fertiliser inputs indirectly affect this by providing more forage, enabling higher stocking rates, higher consumption, higher total plant N concentration, and thus more N excretion. The objective of this study was to estimate the relative importance of both the direct and indirect effects of N fertiliser application on the risk of N leaching. This assessment was based on a combination of experiments and deterministic modelling. We investigated the effect of N fertilisation rate and timing on (i) pasture yield, herbage N content, and risk of direct loss of fertiliser N by leaching, and (ii) the fate of the consumed herbage N and consequences for indirect leaching.

Materials and Methods

Four N response experiments were established in the 2010/11 dairy production season at AgResearch's Tokanui Research Dairy Farm, in the Waikato region of New Zealand. Nitrogen was applied at rates of either 0 (control), 25, 35, and 55 kg N/ha, and at of eight different application timings. Data from these experiments were used to calibrate the biophysical, process-based Agricultural Production Systems Simulator model, APSIM (Keating et al., 2003). Simulations were then run for a 20 year period to assess implications of N fertiliser management on leaching risk. Simulations were also undertaken to estimate the effects of N fertilisation on urinary N concentration and consequences for indirect N leaching from urine patches.

Results and Discussion

As expected, pasture showed highly significant increases in growth after each fertiliser addition ($P < 0.001$), with an average N fertiliser responses (NR) ranging from 4 to 16 kg DM/kg N applied, and an indication that NR is higher in spring. N fertilisation also increased pasture N concentration. APSIM simulated pasture growth and N fertiliser response across the season reasonably well compared with the experimental data. Both the data and simulations suggest a high importance of environmental conditions on NR. APSIM simulations of pasture N concentrations also agreed reasonably well with the measured values and the seasonal variation.

Long term modelling with APSIM and applying N fertiliser in four splits at annual rates of 0, 140 and 220 kg/ha shows a large variation in annual pasture yields between different years due to the variations in rainfall and temperature. Annual pasture yields without fertilisation ranged from 6.2 to 12.5 t DM/ha. Fertilisation increased annual pasture yields up to 15.2 and 16.4 t DM/ha. The risk of direct N leaching from the applied fertiliser was, in general, small over the 20 years simulated. However, in some years substantial N leaching was simulated, with highest values obtained for the 1998/99 period with 26, 48 and 61 kg N/ha with increasing rate of N fertilisation. This is a combined effect of low pasture growth in the 1998/1999 period leading to accumulation of N in the soil, and rainfall of nearly 200 mm following fertiliser application in July 1998, with drainage water carrying the applied fertiliser beyond the pasture roots. Indirect leaching from N return via excreta, is also likely to increase with N fertilisation. Based on simple assumptions indirect leaching losses were estimated to equate to 32, 38, and 40 kg N/ha for the three different application rates.

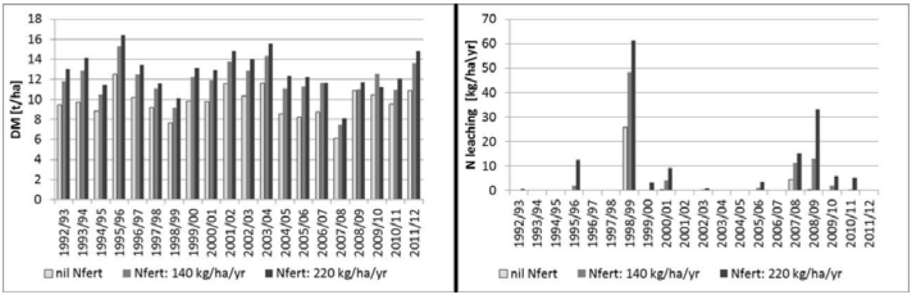

Figure 1. Annual dry matter production (left) and N leaching (right) simulated by APSIM for 20 different years with different N fertiliser application rates, applied in even splits in January, April, July and October to a ryegrass clover pasture in the Waikato region of NZ.

Conclusion

The APSIM model was able to represent the experimental data reasonably well. Running APSIM for 20 years demonstrated the variation in yield and also that N leaching risk of 'direct N leaching' from applied fertiliser is generally low. The highest risk of N loss is from indirect leaching due to higher pasture growth rates, enabling higher stocking rates and thus increasing returns of urinary N patches. The challenge remains to minimise these indirect loss of N from the urine patches.

Acknowledgments

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CROP RESPONSE AND NITROGEN LOSSES AS AFFECTED BY THE REDUCTION OF THE SOIL MINERAL-N TARGET VALUE FOR ORGANIC GREENHOUSE TOMATO

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Mineral balances in organic greenhouse crops revealed considerable gaps in the N balance (Voogt et al.,2011). These gaps are likely caused by nitrogen losses, due to leaching or denitrification (Visser et al, 2006). Nutrient losses by leaching should be reduced as much as possible as growers have to comply with the regulations for surface and groundwater protection. In organic greenhouses the target values for soil mineral N are the same as for conventional production. However, the rationale and basic principles for these values does not apply for organic, moreover never were established for aiming at low leaching. Calculations with the model Osmansoil and Linfert (Van Evert et al, 2006) showed that it would be quite possible to match N supply and N crop need while aiming at lower mineral N levels in the soil, using combinations of slower and faster mineralizing fertilisers. The objective of this study was to test the approach of model based organic fertiliser N supply with lower target soil mineral N in commercial practice.

Materials and Methods

An experiment was carried out at a commercial organic greenhouse with tomato, with a light, organic rich clay (18 % lutum, 8 % O.M), which was 8 years under certified organic production. Irrigation was based on radiation (2.1 l m^{-2} per 1000 J cm^{-2} day^{-1}), both with sprinkler and drip irrigation. Three treatments were laid out in three parallels each plot covering 8 m^2 and with 20 plants/plot. The treatments were: Control (C), Low N (LN), Extra Low N (ELN), with fertiliser management according to the growers experience (C), supply aiming at 50 % (LN) and 75 % (ELN) reduction of the mineral N target. The standard N mineral N values for C, LN and ELN were 5, 2.5 and 1.5 mmol N l^{-1} (1:2 volume extract, Sonnveld and van den Ende, 1971) respectively, which corresponds with 250, 150 and < 100 kg N ha^{-1} yr^{-1} . All treatments received base dressings with compost and dried organic fertilisers (Table 1). For LN en ELN the quantity was based on pre-planting soil samplings at two soil layers, 0-25 cm and 25-75 cm respectively. For the calculations of the required fertiliser supply an excel based computing programme was developed combining the output of modelled N dynamics (mineralisation, denitrification) by Osmansoil and Linfert (Van Evert et al, 2006) and a simple N uptake model, using Intkam for growth and linear N quantity in the dry matter, resulting in soil N-min quantity. Top dressings for LN and ELN then were scheduled according to the evolution of the N-min. 12 Week old tomato plant were planted January 12th and the crop was finished at November 15th. Yield and fruit quality was monitored for the whole growing period. Soil samples were taken at least every 3 weeks, at 0-25 and 25-50 cm. Irrigation, fertiliser supply as well as relevant climatic data were monitored. Samples of young full grown leaves, old leaves and fruits were analysed for macro elements.

Results and discussion

Soil mineral N increased to 500, 210 and 190 kg ha^{-1}, and decreased eventually to 100, 70 and 38 for C, LN and ELN respectively (Fig 1). Despite the significant differences in total mineral N neither yield nor fruit quality was affected by the treatments, as was also the case for total biomass. Plant samples showed only slight differences in N content, so the differences in total N uptake were only limited. The calculated leaching was almost zero since the calculated irrigation surplus was negative. Calculated denitrification was slightly higher for C. The differences between predicted and measured soil mineral N were 10 – 20 kg ha^{-1} for C during whole season, but increased seriously for LN and for ELN in particular in the last months of the growing season to 220and 280 kg ha^{-1} for LN and ELN respectively (Fig 1). Consequently a large gap remains between the calculated N-uptake and the delivery by N-supply and mineralisation. This could be explained either by contribution from the soil buffer below 50 cm, which was not taken into account, or the contribution from mineralisation is higher than estimated by our models.

Conclusions

The approach of model based fertiliser supply with low base dressing and scheduled top dressings resulted in significant lower N-min contents in the soil during the growing cycle, without affecting the yield or product quality. These lower N-min contents reduce the risks for N-leaching and denitrification losses. However a peak in Mineral-N during the first months of the crop appeared in all treatments, which need further fine tuning.

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Table 1. Supplied base and top dressings for the tomato crop

Fertilizer	N P K	C	LN	ELN
	g / kg	T/ ha	T/ ha	T/ ha
Base dressing				
Green compost	5 - 1.5 - 5	93.8	46.9	0
Chicken manure	22 - 9 - 19	3.5	0	0
DCM Ekomix 1	90 - 30 - 24	1.0	0	0
Patentkali	0 - 0 - 448	0.8	0.8	0.8
Top dressing				
Aminosol	90 - 10 - 0	2.6	3.8	3.4
Patentkali	0 - 0 - 448	0	1	1
DCM Ekomix 7-4-12	70 - 28 - 109	1.5	1.0	1.0

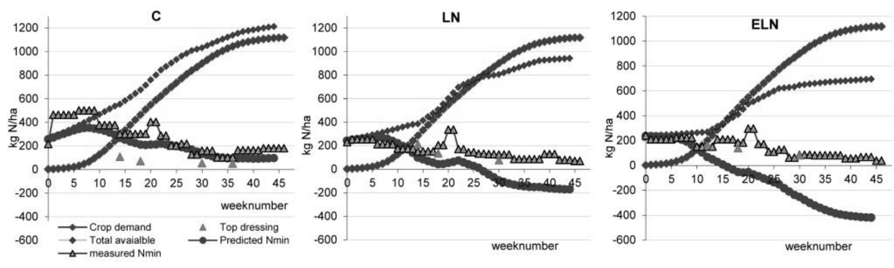

Figure 1. Cumulative N crop demand and available N, the predicted and measured Nmin in the soil (50 cm)and the applied N as top dressing, in a long term tomato crop, for treatments C (control),LN (low N) and ELN (extra low N).

EFFECTS OF POLYACRYLAMIDE AND DICYANDIAMIDE ON CARBON AND NITROGEN MINERALISATION

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Soil conditioners are increasingly used for sustainable soil management in agriculture, e.g. to improve water holding capacity, to decrease soil erosion or to optimise nitrogen (N) use efficiency. Anionic polyacrylamide (PAM) is a polymeric soil conditioner altering soil physical properties contributing to the above mentioned aims, as does dicyandiamide (DCD), which reduces biological activity of nitrifiers in the soil. However, it seems that PAM is reducing the microbial biomass in the soil but not influencing their metabolic activity (Sojka et al., 2006). As a consequence, PAM did not increase mineralisation (Awad et al., 2012). The question arises, if addition of PAM increases mineralisation and if nitrification under these conditions can be effectively managed using DCD in combination with PAM. The present study therefore investigates the short term effects of PAM and DCD on carbon (C) and N mineralisation and on the soil microbial biomass C content (SMB), as well as its activity after addition of maize straw and ammonium-sulphate fertilizer (AS) in an incubation experiment.

Materials and Methods

A sandy soil was incubated for 28 days at 22 °C at 50% water holding capacity in the dark. Maize straw, ammonium sulphate and DCD were added to the soil, in addition to PAM, which was added at a rate of 100% and 200% of the recommended application rate. Soil respiration was monitored throughout the experiment and microbial biomass C and nitrogen mineralisation was determined at the end of the incubation experiment. Microbial biomass C in the soil after incubation was estimated using chloroform fumigation-extraction (Brookes et al., 1985). CO_2 evolving from the soil was captured in 1 M NaOH and titrated with 1 M HCl after addition of $BaCl_2$ and phenolphthalein indicator. Specific CO_2 evolution of microbial biomass, the metabolic quotient ($q$$CO_2$), was calculated from the basal respiration: (µg CO_2-C d^{-1} g^{-1} soil evolved during the last 12 days of incubation in the control samples) / (µg microbial biomass C g^{-1} soil at the end of the incubation experiment) × 1000 = mg CO_2-C g^{-1} microbial biomass C d^{-1}.

Results and Discussion

Maize straw addition increased cumulated soil respiration and SMB, whereas NH_4^+ addition resulted in a decrease. Addition of DCD resulted in a decreased basal respiration but did not decrease the SMB. As a consequence, the $q$$CO_2$ was reduced (Fig. 1B). As expected, DCD addition reduced nitrification. The reduced soil respiration and nitrification indicates a reduction of the nitrifier activity in the soil. On the other hand, addition of PAM increased mineralisation and nitrification, especially when applied without fertilizer, which is in contradiction to other studies (Awad et al., 2012). At 100 % PAM application the maize derived soil respiration was reduced. At 200% PAM, effects are not as clear. PAM increased nitrate content in the absence of maize straw, indicating that PAM may increase nitrification. PAM increased the $q$$CO_2$

(Fig. 1A), indicating either microbial use of PAM or an increased relative activity due to stress or less efficient substrate use in the presence of PAM.

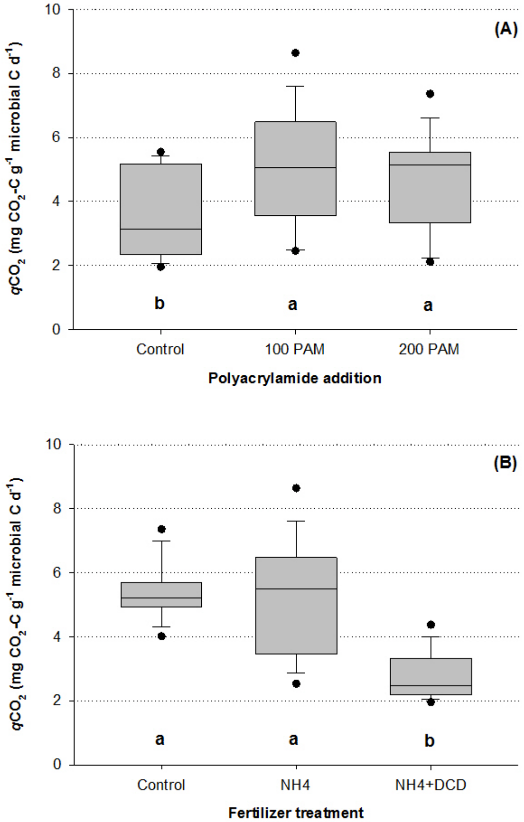

Figure 1. Metabolic quotient (***q***CO_2) of treatments without maize addition relative to fertilizer treatment (A)**;** Control = no fertilizer (n= 14), NH4 = ammonium sulphate (n=15) and NH4+DCD = ammonium sulphate and dicyandiamide (DCD) (n=15**),** and (B) polyacrylamide addition**;** Control = no PAM application (n= 14), 100 PAM = application of PAM 100% recommended rate (n=15) and 200 PAM = application of PAM 200% recommended rate (n=15). Different letters indicate significant differences ($p < 0.0001$, Tukey's HSD).

Conclusions

PAM increases mineralisation and nitrification in the soil, even though reducing the size of the microbial community. It remains unclear, if PAM is used as a substrate by soil microorganisms or stressing them. Furthermore, our study proved DCD to be an effective tool to reduce nitrification even under conditions, where mineralisation is increased by addition of PAM.

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ESTIMATION OF BELOW-GROUND NITROGEN IN CATCH CROPS

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Catch crop cultivation in temperate farming aims at reducing nitrate leaching by immobilization of residual nitrogen (N) in plants. To quantify the N stored in catch crops their biomass has to be assessed. Usually the N content of the above-ground biomass and the main roots is assessed. However, from other studies it is known, that besides roots the below-ground plant biomass also consists of rhizodeposits (Wichern et al., 2008). Rhizodeposits contain inorganic N, N in root exudates and N in root hairs, fine roots, root fragments, root border and sloughed cells (Uren, 2001). In legumes, rhizodeposition contributes about 16.5% to total plant N and in non-legumes 10% (Jones et al., 2009). In canola (*Brassica napus* L.), N rhizodeposition was 29% of total plant N (Arcand et al., 2013). For most catch crops rhizodeposition has not yet been estimated. The objective of the present study was to estimate the below-ground N in catch crops, including the rhizodeposition to assess their N storage ability.

Materials and Methods

In a pot experiment under controlled conditions, oil radish (*Raphanus sativus* L. *var. oleiformis* Pers.), tillage radish (*Raphanus sativus* L.) and winter turnip rape (*Brassica rapa* L. *var. silvestris* [Lam.] Briggs) plants were leaf-labelled with multiple ^{15}N-urea (99at%) applications. Plants were labelled 5 times during vegetation with intervals between 5 and 7 days. Labelling started at the 3- or 4-leaf stage and ended after 11 weeks. For labelling, leaves were partly placed into a vial containing the ^{15}N-urea solution. Absorption took place rapidly within the first days. Distilled water (1 ml) was then added to the empty vials to allow uptake of remaining ^{15}N. After harvest, plants were separated into above-ground plant parts and main roots or tap roots (thick roots). The remaining roots in the soil were separated into medium (< 1 mm) and fine roots (< 0.5 mm) by wet sieving. Oven-dry plant (72 h, 60 °C) and soil material (24 h, 105 °C) was analysed for total N and the isotope ratio ^{15}N/^{14}N was determined using isotope ratio mass spectrometry. The percentage of rhizodeposition derived total soil N (NdfR) was calculated according to the equation of Janzen and Bruinsma (1989). In addition to the pot experiment, a field experiment with unlabelled plants was set up to transfer the results from the pot experiment to the field scale. To estimate overall biomass and the shoot-to-root ratio, above-ground biomass and main roots and fine roots were separated after harvest. In this experiment, also the influence of additional mineral N fertilization on biomass distribution was tested by adding 0, 40 and 80 kg N ha^{-1} as calcium ammonium nitrate. The contribution of rhizodeposition to total N in the field was estimated by transferring the root-N-to-rhizodeposition-N ratio of the pot experiment to the field experiment.

Results and Discussion

In the pot trial, between 10 and 15 % of the total assimilated nitrogen of the catch crops was found as rhizodeposits (Figure 1). This is in the range of values from other studies on legumes and non-legumes (see Wichern et al., 2008; Jones et al., 2009).

However, the values of N rhizodeposition for *Brassica rapa* L. in our study were substantially lower compared to the values for *Brassica napus* L. in another study (Arcand et al., 2013). Reasons for this difference might be associated with the experimental conditions and with the traits of the different species. In the field experiment the shoot-to-root ratio was lower compared to the pot experiment as reported by others (Wichern et al., 2008). Thus, the contribution of rhizodeposition to below-ground N under field conditions is higher. Fertilization in the field trial mostly influenced the formation of above-ground plant biomass, slightly altering the shoot-to-root ratio.

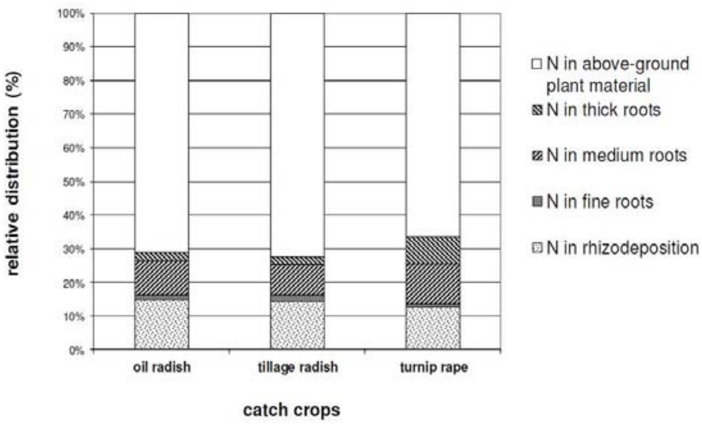

Fig. 1. Relative distribution (%) of plant N in above-ground and below-ground biomass of catch crops. Values show means (n=7).

Conclusions

Our results indicate, that catch crops store more N than previously assumed. Future studies have to verify these findings and investigate the N release from catch crop residues in different cropping systems and under different environmental conditions. Moreover, the availability of N mineralised from catch crops for subsequent crops has to be assessed as well as the possible risks of nitrate leaching.

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HOW ACCURATELY CAN WE ESTIMATE BELOW-GROUND NITROGEN OF LEGUMES?

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Symbiotic nitrogen (N) fixation of legumes is the main input of reactive N in many agricultural and natural ecosystems (Peoples et al., 2009) and especially in organic farming systems. The amount and fate of this leguminous N input determine its availability to subsequent or companion crops (Mayer, 2003). It also influences the amount of N lost in legume-based crop sequences and therefore the negative environmental effects of legumes. The N input by N fixation of legumes is often assessed based on above-ground biomass. However, it has been shown, that this approach underestimates total N input by N fixation, as the below-ground biomass N (BGN) is a substantial part of N input (Peoples et al., 2009). Consequently, the amount of BGN, which consists of root-N and N lost from living roots (rhizodeposition) has to be accounted for in N balances. Rhizodeposition includes not only root exudates, secretions and other soluble compounds, but also includes decaying fine roots, root hairs and root fragments, which cannot be easily separated from the soil (Wichern et al., 2008). More efficient nitrogen (N) use in legume-based agricultural systems requires a better understanding of the processes related to the below-ground plant biomass. Therefore, BGN input and its turnover throughout cropping sequences have to be quantified accurately. This contribution raises the question how accurately we currently predict the BGN input by legumes. The objective is to critically discuss our methodological limits and uncertainties and the consequences for N balances.

Results and Discussion

Stable isotope techniques to quantify N fixation and the BGN of legumes have been used in research for some decades now and have provided useful estimations. However, quantification of BGN is still often biased by the testing conditions. Many quantitative results have been derived from pot experiments. These are disputable, because a low available soil volume might result in a reduced root-to-shoot ratio and therefore in an underestimation of the contribution of BGN to total N (Figure 1). Therefore, future research aiming at quantifying BGN should be conducted under field conditions. Estimating BGN in the field also allows a more accurate estimation of the N taken up by e.g. catch crops and its availability to succeeding crops, assisting in determination of the requisite N fertilization of the following crop. Combining lab-derived models with field measurements might hold some future potential. However, labelling approaches used in many of the studies have flaws. In particular, the prerequisite for estimating rhizodeposited N, namely the homogeneous ^{15}N enrichment of roots and rhizodeposits, has not been verified yet. It is also unclear how the ^{15}N enrichment of roots and the closely related rhizodeposition changes over time with the use of one of the various labelling approaches. Therefore, methodological uncertainties have to be diminished to better quantify BGN.

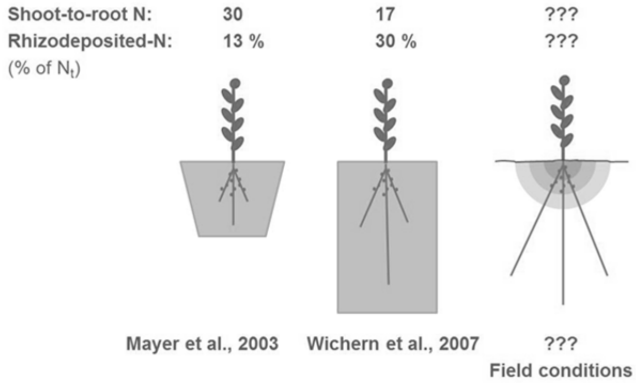

Figure 1. Influence of shoot-to-root ratio in experiments as influenced by available soil volume on the prediction of N rhizodeposition (% of total N). Mayer et al., 2003 and Wichern et al., 2007 used the same plant species, a similar labeling approach and comparable growing conditions except for a difference in available soil volume (pot vs. soil columns).

Conclusions

Results from quantitative estimations of BGN input are disputed as they differ substantially depending on experimental setup and labelling approach. Especially the available soil volume seems to influence the amount of BGN in relation to above-ground biomass. Additionally, approaches for estimating BGN based on ^{15}N labelling methods are criticised for not being able to guarantee homogeneous labelling of plants. Future research should aim at combining stable N isotope techniques with carbon isotope approaches and molecular techniques to elucidate the fate of BGN and its contribution to the N uptake of succeeding crops.

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SOIL CARBON AND FERTILITY AS AFFECTED BY THE SOURCE OF APPLIED NITROGEN UNDER NO-TILLAGE MANAGEMENT IN A VOLCANIC TEMPERATE SOIL

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The function of volcanic soils is limited by intensive soil management. Reducing tillage and a rational plan of fertilizer application are some of the strategies of soil management preventing soil degradation. These management techniques may influence the concentration of organic C and N, which are good indicators of soil quality and productivity due to their favourable effects on physical, chemical and biological properties (Angás et al. 2006; Zagal et al., 2009). In Chile, cereal production can be found in the central valley and on soils of volcanic origin (Andisols). About fifty percent of the Chilean agriculture production occurs in these soils, which have very special characteristics as the presence of allophone and imogolite, high contents of organic matter, low bulk density and pH, high phosphorous fixation capacity and high water holding capacity (Aguilera et al. 1997). We assessed the effect of the application of different sources of nitrogen (N) fertilizer on crop yield and its impact on biological and chemical properties of a volcanic soil under no tillage system. Furthermore, we aimed to evaluate the sensitive response of these soil properties to the N-fertilizer sources.

Material and Methods

A cereal crop rotation under no-till system was maintained for 13 years on a volcanic soil in the South of Chile (71°53'S, 36°55' W). Fertilizer treatments were tested as NO_3^--N and NH_4^+-N sources in dose of 150 Kg ha^{-1}, plus the combination of NH_4^+-N fertilizer with $CaCO_3$ (500 and 1000 Kg ha^{-1}, respectively), and a control treatment (N=0), which were arranged in a complete random design (n=3). Crop yield was measured every season for 13-y. Soil pH, and chemical properties were measured at the end of the experiment at two depths (0-5 and 5-10 cm), as well as labile organic matter fractions, soil microbial biomass (Cmic, fumigation-extraction method) (Vance et al. 1987); C mineralization (Cmin, closed incubation) (Alef and Nannipieri 1995) and dissolved organic C (DOC, extraction-wet digestion and colorimetric determination) (Haynes 2000).

Results and Discussion

Mainly, the crop yield decreased ($P < 0.05$) by the end of the experiment due to the application of NH_4^+-N fertilizer, and the same trend was observed in soil pH. Changes in Cmic, Cmin and DOC were detected at 5-cm ($P < 0.05$) due to the N-sources applied.

The use of NH_4^+-N fertilizer produced a decrease of 0.07 pH units per year. Despite of different source of N fertilizer and its effect on pH, no effect of treatment were observed at P availability. Besides, depth had a significant effect at P availability, with a concentration in the first 5 cm depth as consequence of phosphorus fertilizer, reflecting the low mobility in the soil profile. After 13 years of using NH_4^+-N sources the total exchangeable bases ranged 0.80 and 2.35 and Al saturation (%) reached 53% and 10% at 0 – 5 cm and 5 – 10 cm, respectively. These levels of Al saturation are very toxic for the studied crops (Inostroza-Blancheteau et al. 2008) and the main factor influencing the low yields obtained here.

Conclusions

Biological parameters as Cmic, Nmic and Cmin, and DOC were more sensitive to soil changes as consequence of treatments than total soil carbon and nitrogen. Therefore their use as soil quality indicators is recommended, according to the soil functions to be evaluated. In no till crop systems, the main effects in biological, biochemical and chemical properties of this volcanic soil occur in 0-5 cm depth. The relatively rapid acidification of ammonium fertilizers applied alone, has a detrimental effect in biological indicators as Cmic, Nmic and Cmin through a decrease in soil pH, which affects crop yields, aluminum saturation, ECEC and likely soil organic matter quality. The use of lime to neutralize the acidifying effect of urea and MAF has beneficial effects that go beyond the changes in the soil pH.

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GENE EXPRESSION-BASED DIAGNOSTICS: A NEW WAY TO QUANTIFY PLANT NITROGEN STATUS?

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Matching the supply of N to the crop N demand is important for meeting yield goals and minimizing environmental N losses (Zebarth et al. 2009). In-season assessment of crop N sufficiency is commonly achieved using chemical (Olfs et al. 2005) or optical (Goffart et al. 2011) tests. Plants respond to abiotic stresses through changes in gene expression. Distinct plant gene expression patterns for single and combined abiotic stresses and nutrient deficits have been documented (Hazen et al. 2003). Here, using the potato (Solanum tuberosum L.) crop, we propose use of gene expression in leaf tissue as a novel approach to measuring the crop N sufficiency, and for the purpose of guiding fertilizer N management. In a field experiment, potatoes (cultivar 'Shepody') were grown at six fertilizer N rates (0-250 kg N ha-1) which ranged from deficient to excessive. Sampling was performed weekly for 7 weeks. Leaf disks were collected from the terminal leaflet of the last fully expanded leaf weekly for gene expression analyses. RNA was extracted from the leaf disks and quantification of gene expression performed using nCounter. Quantification was performed for 22 genes related to N uptake/transport, N assimilation, amino acid metabolism, and plant stress response. An additional five house-keeping genes were quantified for use in normalizing gene expression results. Leaf chlorophyll content, measured using a SPAD meter, and petiole nitrate concentration were also measured. An ammonium transporter gene (AT1) exhibited an increase in expression under deficient N supply (Figure 1). This gene was highly expressed, and was consistently expressed across sampling dates (Figure 1), which makes it suitable for use as a diagnostic tool. AT1 gene expression was also closely related to root zone soil nitrate concentration (Figure 2). Expression of the AT1 gene on all sampling dates was highly negatively correlated (r = -0.86 to -0.97) with total tuber yield. Expression of the AT1 gene in leaf tissue was as good as, or better than, current optical or chemical measures of potato N sufficiency. In a growth room experiment, potato plantlets from tissue culture were grown hydroponically to compare N supplied in different forms (ammonium only, nitrate only, nitrate plus ammonium) at either abundant (7.5 mM) or deficient (0.75 mM) nitrate supply. Plants were grown under abundant N supply in a vermiculite and perlite media, and then the treatments imposed over a two week period at three stages of growth (tuber initiation, flowering and early tuber bulking). Samples collected at the end of the two week period were used for determination of gene expression in leaf tissue, petiole nitrate concentration and leaf chlorophyll content. Shoot growth and N accumulation responded to N supply regardless of N form. The form of N supplied strongly influenced chemical and optical measures of potato N status, with petiole nitrate most responsive to N supplied in nitrate form and leaf chlorophyll most responsive to N applied in ammonium form. In contrast, expression of AT1 to N supply was relatively consistent regardless of the N source. This suggests that expression of AT1 primarily reflects the internal demand of the plant for N. The findings of these experiments suggest that gene expression can be an effective diagnostic tool for assessment of crop N sufficiency. Current research is focused on identification of genes which can be used to quantify plant P and K sufficiency. The long term goal is to develop a single platform which can simultaneously quantify plant responses to a series of abiotic stresses.

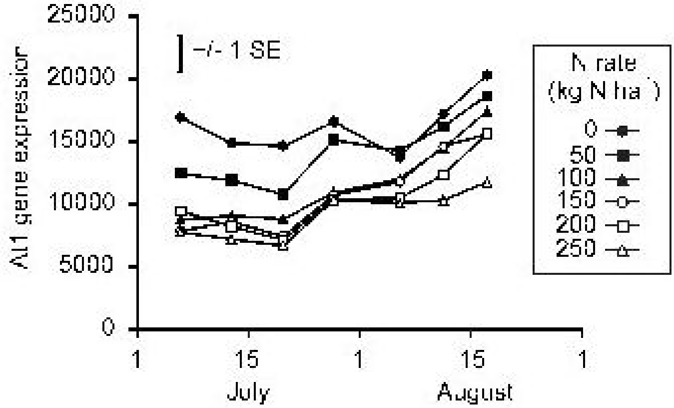
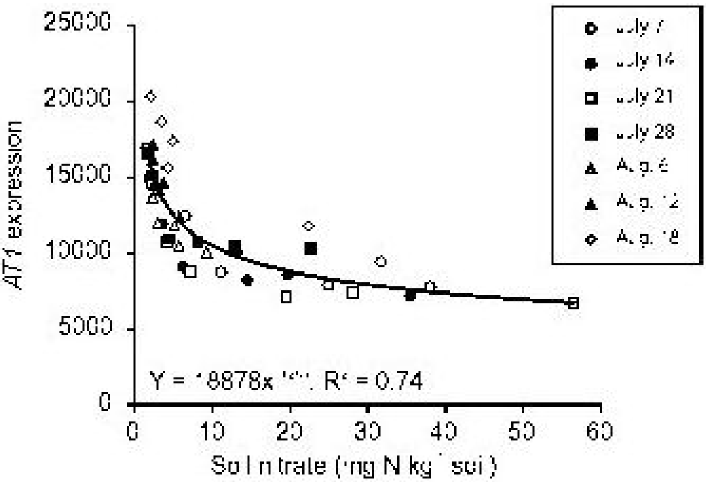


Figure 1. Effect of fertilizer N rate on expression of AT1 in potato leaf tissue in seven sampling dates through the growing season.



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PREDICTION OF SOIL NITROGEN SUPPLY USING A SIMPLE KINETIC MODEL

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Matching the supply of nitrogen (N) to the crop N demand is important for optimizing crop yield and minimizing environmental N losses (Zebarth et al. 2009). Uncertainty in the supply of N from the soil is one of the major factors limiting accurate fertilizer N recommendations (Lobell 2007). Although many indices of N availability have been evaluated, and have often shown promising results, no single index of soil N availability has been found to be robust across soil types, cropping systems and climatic zones (St. Luce et al. 2011). Simple kinetic models can be used to predict soil N mineralization as an aid to improved fertilizer N management, but require long-term incubations to obtain the necessary parameters (Dessureault-Rompré et al. 2012). Here we examine the feasibility of predicting the necessary parameters to use a simple kinetic model to predict soil N supply (SNS) in potato production in the humid soil moisture regimes of Eastern Canada. Replicated samples from 92 soils were used in a long-term (24-wk) incubation to determine mineralizable N parameters. The field-based estimate of soil N supply was the sum of N uptake in the unfertilized potato crop (vines plus tubers) plus residual soil nitrate, referred to as plant available soil N supply (PASNS). A first-order (F) kinetic model was examined of the form $N_{min} = N_0\,[1-\exp(-kt)]$ where N_{min} is the cumulative amount of N mineralized at time t, N_0 is potentially mineralizable N, and k is the mineralization rate constant. Use of measured values of k and N_0 resulted in adequate prediction of soil N supply (Table 1). However, use of estimated parameters (predicted N_0 and fixed-k) resulted in poor prediction of SNS as indicated by low values of R^2, and slopes of regression equations well below one. As a result, a zero- plus first-order (ZF) kinetic model was examined of the form $N_{min} = k_S t + N_L[1-\exp(-k_L t)]$ where N_{min} is the cumulative amount of N mineralized at time t, k_S is the rate constant of a non-depleting zero-order pool (“stable N pool”) in which N is mineralized at a constant rate, and N_L is a depleting first-order pool (“labile N pool”) with mineralization rate constant k_L. The ZF model provided satisfactory prediction of SNS using both measured and predicted values of k_S and N_L (Table 1). In both cases, SNS predicted from the ZF kinetic model for 0-30 cm depth were similar to or slightly greater than field-measured PASNS (Figure 1), which would be expected given that some losses of N during the growing season through leaching and denitrification can be expected which would not be reflected in the predicted SNS. Results of this study suggest that it is practical to predict the necessary parameters from soil properties to implement the ZF kinetic model in order to predict SNS on an individual field basis.

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Table 1. Regression of predicted soil nitrogen supply (SNS) against measured plant available soil nitrogen supply (PASNS) for the first-order (F) or zero- plus first-order (ZF) kinetic models using measured or estimated mineralizable N parameters

Model	Depth	Predicted SNS (kg ha^{-1})	% difference (absolute value) from 1:1 line	R^2 adjusted	Slope
F model, measured	15	111(±37)c†	20 (±22)a	0.45	0.79
	20	148 (±49)d	27 (±23)a	0.45	1.05
	30	217 (±72)e	49 (±45)bc	0.48	1.60
F model, estimated	15	81(±16)a	27 (±16)a	0.18	0.23
	20	108 (±21)bc	22 (±14)a	0.18	0.30
	30	164 (±33)d	61 (±32)c	0.17	0.46
ZF model, measured	15	77 (±22)a	28 (±16)a	0.44	0.46
	20	102 (±29)bc	19 (±12)a	0.44	0.62
	30	155 (±42)d	47 (±32)bc	0.48	0.94
ZF model, estimated	15	63 (±17)a	41 (±13)a	0.41	0.36
	20	84 (±23)ab	24 (±13)a	0.41	0.48
	30	126 (±32)c	24 (±22)a	0.49	0.73
PASNS (kg ha^{-1})		109 (±32)c			

†Means in the same column followed by the same letter are not statistical different at $p < 0.05$ based on Tukey's Honestly Significant-Difference Test.

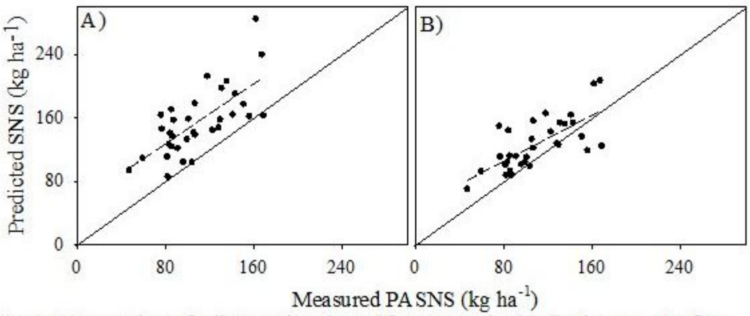

Figure 1. Comparison of soil N supply estimated for 0-30 cm depth using the zero- plus first-order kinetic model using A) measured and B) predicted values of mineralizable N parameters (solid line indicates 1:1 line, dashed line indicates regression line).

EFFECTS OF FERTILISATION AND IRRIGATION ON YIELD AND NITROGEN USE EFFICIENCY OF POTATOES

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Normally, farmers prefer to apply excessive nitrogen fertilizer at planting in potato production with the hope of maximizing return, and often, the rate of nitrogen they apply exceed the potato need, nitrogen not taken up by the crop, may potentially contribute to ground and surface water pollution through nitrate leaching and soil erosion (Almasri and Kaluarachchi, 2007), which may most often occur at and after harvest of crops (Demyttenaere et al., 1990). This is especially true when it comes to South Jutland, Denmark, with coarse-textured meltwater sand that contains ca. 76% coarse sand. In recent years, traveling Gun Irrigation (GI) system has been widely used in Denmark in potato production. Nevertheless, the 'big gun' sprays a huge stream of water high in the air during irrigation, which is extremely susceptible to wind and evaporation losses(Kendy et al., 2006), furthermore, with most proportion of applied water allocated at furrows, the sprinkled water is in risk of percolation, which may contribute to nitrate leaching. Since giving all nitrogen at potato planting has a high risk of nitrate leaching, it has been reported that access nitrogen status is necessary if one want to get temporal precision in nitrogen application (Rodrigues et al., 2005).This means that during the crop growth, the crop nitrogen status should be monitored and that supplementary nitrogen should be applied. From this point of view, Split nitrogen dressing based on nitrogen status evaluation are actually a promising way to meet crop need with optimal rate. In the proposed study, we tried to evaluate response of nitrogen use efficiency (NUE) to two irrigation systems, gun irrigation and drip irrigation, respectively. In addition, with the assumption that fertigation has a higher NUE. The effects of fertilization were investigated in terms of yield, NUE and nitrate leaching under different irrigation and fertilisation rates. A specific and targeted fertigation strategy would be developed to obtain a sustainable and environmental friendly potato production in Denmark. This would benefit farmers by reducing their cost and maximizing yield and quality, and it is crucial to the environment as more groundwater would be exempt from nitrate pollution.

Material and methods

The experiment was conducted at Jyndevad Research Station in South Jutland, Denmark. The soil is a coarse-textured meltwater sand that contains ca. 76% coarse sand (0.2-2.0 mm) The seed tubers were preheated at 14-16°C until 1 mm sprouts. Seed tubers were planted at 15th May, 2013. defoliated from 16th Aug to 3th Sep. harvested at 16th sep, plant sampling activities were done during the growth season for 5 times, with the aim to analyze nitrogen uptake and yield of potatoes. **Experiment1** contained three treatments: GI system (including two sub-systems) and DF system for comparison test, which aimed to display effects of GI and fertilization systems on growth, yield, and soil Nmin distributionof potato, as well as nitrate leaching after potato growth season. Table1 schematic of GI and DF system

Irrigation system	Irrigation Criteria	Irrigation frequency	Fertilization Criteria at planting	Dressing time
GI-Figaro GI-Daisy	25 mm water	Whenever water deficit equals 25 mm according to Daisy model	120 kg N ha^{-1} 30 kg P ha^{-1} 180 kg K ha^{-1}	No
DF-Daisy	90% θ_f –SWC_t according to Daisy model	Every two days	30% of calculated N demand 30 kg P ha^{-1} 180 kg K ha^{-1}	When critical N level is reached based on Daisy model,20 kg N ha^{-1} portions was applied

In **Experiment 2** two factors were implemented: irrigation and nitrogen regime.

There were 14 treatments in total, which were partial combination of irrigation and fertilization rates: *Irrigation amount* - I0: No irrigation; Id: Deficit irrigation; I1: Full irrigation; IFigaro: Irrigation will be guided with AquaCrop model; IDaisy: Irrigation will be guided with Daisy model;

Nitrogen regimes: 0 kg N/ha (N0), 60 kg N/ha (N1), 100 kg N/ha (N2), 140 kg N/ha (N3), 180 kg N/ ha(N4), dynamic N (Nd) fertilization rate (Nd1) and (Nd2), as well as treatment using slurry as basic dressing and afterwards as (Nd2)

The summary of all treatments were: I0N0, I0N3, IdN3, IdNd1,I1N0, I1N1, I1N2, I1N3, I1N4, I1Nd1, I1Nd2, I1Norg, IFigaroN3, IDaisyNd1

Timing and amount of fertilization

42kgN/ ha was applied in all treatments needing N at planting, the rest of N was added with fertigation in several doses after emergence. Fertigation in Nd treatments was guided with Daisy model.

I1N2, I1N3, I1Nd1, I1Nd2, I2N3, IFigaroN3 received labeled ammonium-15N nitrate-15N at 5 atom % enrichment.

These microplots included 6 plants (protected and 2 guard plants). Microplots received nutrient solutions through an independent drip tubes.

Measurements

Canopy reflectance data; yield, LAI, DM, N analysis in plants during 5 times plant sampling, nitrate leaching. Nitrate leaching will be monitored after harvest

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FLUXES OF MINERAL NITROGEN IN SOILS OF LITHUANIA DURING THE FROST-FREE SEASON

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Statistical data as well as the available information regarding the agricultural practices indicate that agricultural producers increase nitrogen fertilisation in order to achieve higher crop yields. Due to this fact the use of nitrogen fertilisers becomes an environmental problem on the global scale, since an excessive application of nitrogen fertilisers results in pollution of ground and surface waters. An excessive use of nitrogen fertilisers results in higher levels of nitrates in agricultural production (Staugaitis et al., 2007). One of possible ways to solve this problem is an optimised nitrogen fertilisation based on the determined content of mineral nitrogen (N_{min}) in soil, i.e. the calculation of the optimal rates of nitrogen fertiliser must be based on the knowledge available regarding the N_{min} content and fluxes in different soil types during the frost-free season (Cassman et al., 2002).

Materials and Methods

The research was conducted in 2011-2013 in Western Lithuania (main soil types, Luvisols and Albeluvisols). Soil samples for N_{min} determination (two replications) were collected in spring (March-April) summer (June) and autumn (October-November) from the 7 plots of 20 m^2 size distributed throughout the experimental field. One composite soil sample (4-6 soil probes) taken from 0-30, 30-60 and 60-90 cm layers. The thoroughly mixed composite soil sample is kept at 1-3^{o}C during transportation and up to the time of the laboratory testing procedures. Nitrogen content of soil ($N\text{-}NO_3$+$N\text{-}NH_4$) was determined in accordance with LVPD-05:2012. Soil dried samples of soil were agitated for 1 hour in the 1M KCl solution at the ratio 1:2,5.

Results and Discussion

Autumn season (Fig.1; A). After the harvesting an average level of N_{min} in 0-90 cm layer of soil was 21.69 mg kg^{-1}. *Fluvi-Epihypogleyic Cambisols* and *Haplic Luvisols (moderately eroded)* contained the highest N_{min} levels – 31.87 mg kg^{-1} and 25.83 mg kg^{-1} respectively. The highest levels of N_{min} were found in 0-30 cm layer of soil; it can be assumed that nitrogen supply was excessive and therefore it was not taken up by the plants. *Spring season* (Fig.1; B). An average level of N_{min} was 19.14 mg kg^{-1}. High precipitation rate and higher than usual air temperatures during the winter resulted in leaching of mineral nitrogen from the eroded soils. Content of N_{min} in *Haplic Luvisols (moderately eroded)* decreased dramatically (14.77 mg kg^{-1}) – it was washed down the slope and leached into the ground waters. At the same time the deluvial soils became enriched with nitrogen (28.42 mg kg^{-1}). The deluvial soils neighbouring the *Hapli-Calc(ar)ic Luvisols* located on flat fields gained the highest levels of N_{min}– 31.83 mg kg^{-1} (by 8.08 mg kg^{-1} higher than in autumn). *Summer season* (Fig.1; C). *Hapli-Calc(ar)ic Luvisols* contained the lowest levels of N_{min} – 10,27 mg kg^{-1}. The soil types which contained higher levels of N_{min} in spring maintained their positions in summer as well, yet the increase in N_{min} content was determined in 30-60 and 60-90 cm soil layers

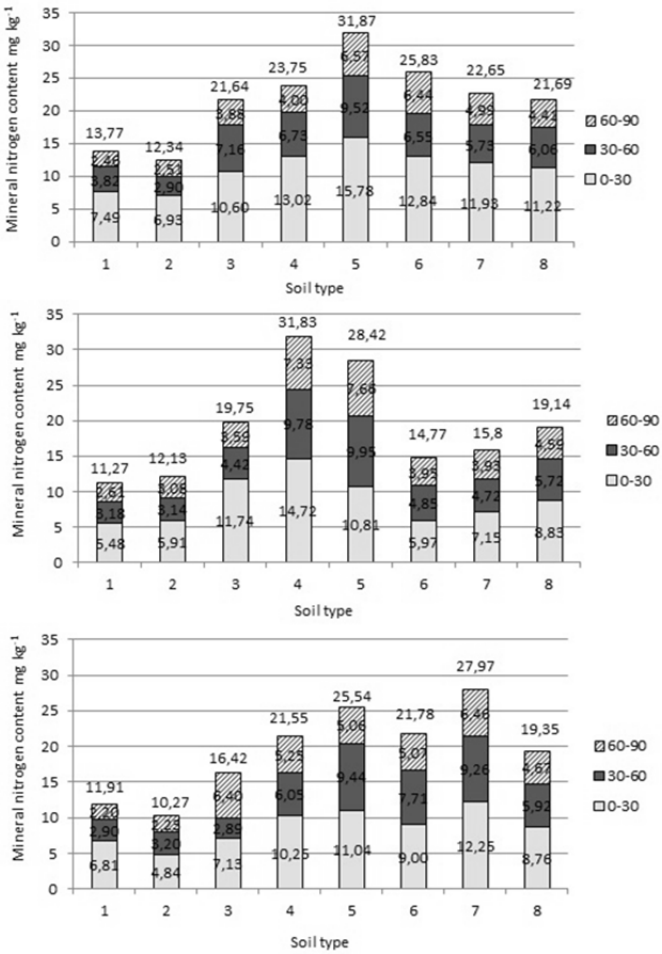

Figure 1. Fluxes of mineral nitrogen content in different soils: in autumn (A), spring (B), summer (C), 2011-2013 m. Note: 1. Endocalcaric Gleysols (GLk-n); 2. Calc(ar)i-Endohypogleyic Luvisols (LVg-n-w-ha); 3. Eutri-Endohypogleyic Arenosols (ARg-n-w-eu); 4. Hapli-Calc(ar)ic Luvisols (LVk-ha); 5 Fluvi-Epihypogleyic Cambisols (CMg-p-w-fv); 6. Haplic Luvisol (moderately eroded) (LVh-em); 7. Orthi-Haplic Luvisols (LVh-or); 8. Average.

Conclusion

Fluxes of N_{min} content in Luvisols and eroded Albeluvisols located on an irregular relief during the frost-free season were irregular. A three-year average data suggest that the highest level of soil N_{min} was determined in autumn – 21.69 mg/kg, summer level was lower – 19.35 mg/kg followed by the spring level – 19.14 mg/kg. The highest autumn levels of N_{min} in 0-90 cm soil layer were found in Fluvi-Epihypogleyic Cambisols, spring levels – in Hapli-Calc(ar)ic Luvisols, summer levels – in Orthi-Haplic Luvisols, the lowest autumn levels of N_{min} – in Calc(ar)i-Endohypogleyic Luvisols, spring levels – in Endocalcaric Gleysols, summer levels – in Calc(ar)i-Endohypogleyic Luvisols.

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ASSESSMENT OF LEAF N PERCENTAGE FROM COLOR IMAGING: A CONCEPTUAL MODEL

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Techniques that measure the N status of plants can aid in management decisions that have economic and environmental implications. This study was conducted to identify and quantify N deficiencies in plant by usingcolor imaging of canopy with the possibility for use as a management tool.

Materials and method

The experiment was conducted in 3 replicates in a lay out after the method of De Malach et al (1996) in a complete randomized design. In order to apply the required concentration of nitrogen two trickle laterals and their emitters were connected to two sources of water one was a fertilizer stock solution and the second supplies water from a tap water source. The two laterals are coupled together to form a double emitter source.

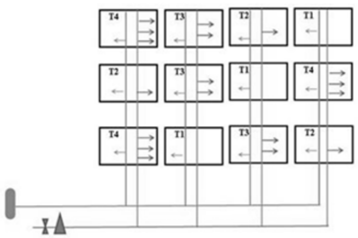

Fig 1

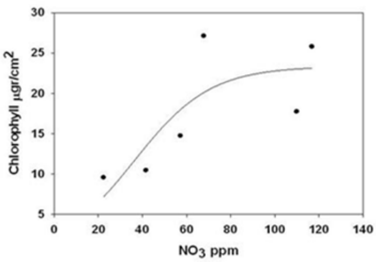

Fig 2

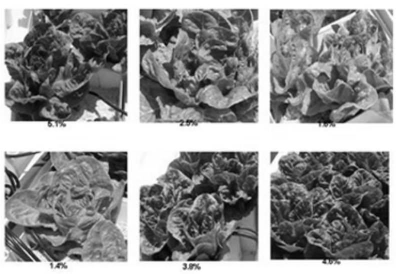

Fig 3

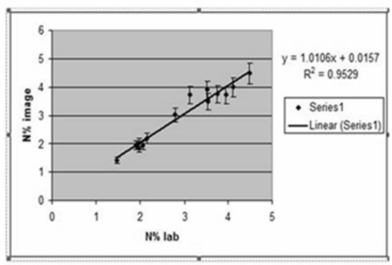

Fig 4

Tap water emitter
Fertilizer tank
Tap water control valve and a meter

Fig. 1 double-emitter source (des) for nitrogen concentration experiments.

Drainage water was collected every day and Nitrate concentration was measured from each container using Thermo scientific NO^{-}_{3} electrode. Chlorophyll content was estimated according to Lichtenthaler HK (1987). Leaf chlorophyll a (Chl a), chlorophyll b (Chl b), and total carotenoids were extracted with 80% aqueous acetone and the absorbance values of the extract at 470, 663.2, and 646.8 nm were determined with a spectrophotometer (UVmini-1240, Shimadzu, Japan). Chlorophyll and carotenoid concentrations were calculated using the equations previously described by Lichtenthaler (1987).

Results and discussion

Major segment of chlorophyll molecule in all plants is based on Mg^{++} in the center of the molecule surrounded by four N atoms. Theretofore a legitimate test is to compare chlorophyll content in the leaf and nitrite. This comparison is shown in Fig. 2. According to this figure, content of chlorophyll in corn leaves can be described by a simple function which presents chlorophyll saturation at about 100 ppm of nitrate.

Evaluation of the greeness of plant canopies can be assessed and quantified by using technology of color imaging. For example, laboratory obtained %N in leaves of several crops revealed increase in chlorophyll concentration as a function of total NO^{-}_{3} in the irrigation water. This increase was identified up to a certain threshold (Fig.2).

Fig. 2. Chlorophyll (μgr/cm^2) as a function of nitrate concentration (NO_3 ppm).

The threshold results from the Nitrogen partitioning among three compartments that together comprise the leaf: the leaf structure, the leaf protein (enzymatic) complex, and the leaf photosynthetic (chlorophyll) system. Each compartment is characterized by its dry matter (DM) weight (% of total leaf DM weight) and the %N in its DM (%N_{chl}, %N_p and %N_s). But 75-80% of the N belong to the chlorophyll molecule. Therefor evaluation of the greeness of plant canopies can be assessed and quantified by using technology of color imaging with a minor correction for the other compartments.

The image processing technology (Software) is based on the correlation between green color (crop greenness), chlorophyll amount, and %N. The green level or greenness of the leaves is increased (or decreased) according to their nutritional status. (Fig. 3)

Summary

A comparison between lab results and our image-based algorithm for N% in the leaves of wheat in a commercial field was found acceptable within 5-15% of the lab results(Fig.4).

Current achievements:

1. Calculation is done within seconds for images up to 3M
2. Calculated results are within 15% of leaves lab tests. Lab tests by themselves allow deviations of 6-15% depending on lab.
3. Minimal limitations for image sampling.
4. The algorithm works under both cloudy and sunny weather, most hours of the day, various picture taking angles and distances.

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Topic 2

Agronomic and environmental impacts
of recycling organic residues

THE POTENTIAL OF UNDERSOWN CATCH CROPS AND LEAVING VEGETABLE CROP RESIDUES INTACT IN INTENSIVE VEGETABLE ROTATIONS

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Crop residues of field vegetables are often characterized by large amounts of biomass with a high N content. These residues have been found to mineralize rapidly even at low soil temperatures (De Neve & Hofman, 1996). Furthermore vegetable crops are often the last crop before winter leaving behind considerable amounts of N prone to nitrate leaching. In order to meet water quality objectives set by the Nitrates Directive sustainable on-field management strategies are needed. A first set of field experiments assesses the potential of catch crops undersown in a vegetable crop. A second set of field experiments compares (i) leaving the crop residues and rooting system intact on the field, (ii) total removal of crop residues and (iii) conventional incorporation of crop residues. The potential to reduce the risk of N losses during winter as well as the feasibility of the different management options is evaluated.

Materials and methods

The field experiments were all located in the intensive vegetable growing region in Flanders (Belgium). All field experiments were designed in fully randomized blocks with four replicates. The potential of catch crops undersown was evaluated in two field experiments. Italian ryegrass (Lolium multiflorum), winter rye (Secale cereal) or phacelia (Phacelia tanacetifolia) were sown 4 week after planting of a cauliflower crop (Brassica oleracea var. botrytis) and compared to a treatment without understory in each of the locations. Total removal or leaving the crop residues intact was performed with cauliflower (Brassica oleracea var. botrytis) in four locations and with headed cabbage (Brassica oleracea convar. capitata var. Alba) in three locations. During the experiments soil samples were taken monthly with an auger in three layers: 0-30 cm, 30-60 cm, 60-90 cm and analysed for ammonium-N and nitrate-N. Total crop yield and crop residues (leaves and stalks) was determined at harvest. Plant samples (4 subsamples per treatment) were dried, ground and analysed for dry and organic matter, N and P content. N dynamics were simulated by means of the EU_Rotate_N model, which was calibrated using the measured soil mineral N content, crop yield and crop N uptake.

Results and discussion

Evaluation and simulation of the results of the field experiments is ongoing and will be presented at the N Workshop, but some trends can already be identified. Total soil mineral N content was generally greater following incorporation of vegetable crop residues compared to leaving them intact on the field or total removal. Intact crop residues of headed cabbage appeared to continue taking up N from the soil, whereas intact crop residues of cauliflower decomposed and increased soil N content. Removal of crop residues logically resulted in reduced mineral N concentrations in soil compared to the other options but the practical feasibility is very dependent of soil and weather conditions. At several locations only a small increase in soil mineral N content could be observed despite incorporation of a large amount of N (up to 297 kg N ha-1) during a period with relatively high soil temperatures (±10°C). This may indicate immobilization of N in soil organic matter or considerable gaseous N losses. Results of N uptake by catch crops undersown in a vegetable crop will be shown at the conference.

Acknowledgment

We greatly acknowledge VLM (Vlaamse Landmaatschappij– project TWOL2012/MB2012/1) for funding this research.

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THE YIELD FORMATION IN ORGANIC CULTIVATED SPELT BASED ON N SUPPLY WITH ORGANIC FERTILIZERS

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N supply in organically cultivated winter cereals, especially in temperate climate is associated with limitation of available nitrogen in the soil, because of limited N mineralization from organic fertilisers during late winter and spring time. For example, in organic wheat, nitrogen is one of the key limiting factors responsible for irregular productivity and low quality (David et al, 2005), 5 to 50 % less than conventionally managed crops (Nieberg and Schulze Pals, 1996). We know from cultivation practice that spelt (*Triticum aestivum* ssp. *spelta*) as a low input cereal, which lodging at high N supply, but in literature exist the lack of data about fertilisation with organic fertilizers in organic cultivation system. For those the aim of present study is to analyse available N from the soil and effect of added N with organic fertilizers on grain yield, plant height and spike characteristics.

Materials and Methods

The trials with spelt variety Ebners Rotkorn designed as a random block (5 m^2 plots) with for organic fertilisers (oil pumpkin cake 9.6 % N, biogas digestate approx 4.6 % N, biosol 6.0 % N and cattle slurry 5.0% N with rates 0, 30, 60 and 90 kg N ha^{-1}, respectively) were carried out at University Agricultural Centre Maribor, Slovenia in the years 2011 and 2012 Sandy-clay soils contain 41 kg Nmin before top dressing, pH was 6.5 with optimal content of potassium and phosphorus after 3 years clover grass mixture as previous crop. The cultivation system was provided according to organic farming regulation. The effects of fertilization treatments on grain yield, plat height and spike characteristics were analysed.

Results and Discussion

The height of plants varied significantly from 134 cm (digestate) to 140 cm (oil pumpkin cake) among fertilisers, and moisture in the grain between the years 2011, 2010 from 17,6 to 19,0%, respectively. The significant differences exist between grain yields and spikes comparing 0 treatment and N rates (Table 1), but with small differences in yield, because of available nitrogen from clover – grass mixture as previous crop.

However, the spelt is low input cereal, but in case of available nitrogen from previous crop the yields can be higher than in average production (2.0-2.5 t ha^{-1}). In this case also the differences among N treatments have low impact on the yield increasing.

Conclusion

We can conclude that in organic cultivation of low input cereals like spelt at optimal available nitrogen from previous clover-grass mixture, the yields can be twice higher than in usual organic production. Also in this case added organic fertilizers significantly increased the grain yield significantly.

Table 1: Hulled grain yield and spike length in spelt depending on years, organic fertilizers and N rates

	Yield of hulled grains 14 % moisture kg 5 m^{-2}	Yield of hulled grains 000 kg ha^{-1}	Spike - length (cm)
Y- Year	ns		**
F- Fertiliser	ns		ns
N- N rate	*		*
YxF, FxN, YxFxN	ns		ns
Y 2011	2.10±0.36	4.20	10.3±0.8a
2012	2.00±0.47	4.00	11.0±0.6b
F Oil pumpkin cake	2.01±0.45	4.02	10.8±0.7
Biogas digestate	1.99±0.41	3.98	10.6±0.7
Biosol	2.03±0.44	4.06	10.8±0.7
Cattle slurry	2.20±0.39	4.40	10.5±1.0
N 0	1.89±0.31b	3.96	10.3±0.5b
30	2.05±0.49ab	4.10	10.8±0.8a
60	2.10±0.41ab	4.20	10.7±0.7a
90	2.17±0.43a	4.34	10.8±0.7a

Means in the same column followed by the same letter are not significantly different at $P<0.05$

Acknowledgement

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NITROGEN RECOVERY FROM LOW-EMISSION APPLIED DAIRY SLURRY BY THREE FORAGE GRASSES: A LONG-TERM TRIAL

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Efficient use of manure N is important economically and because unused N is lost to the environment. Measures that reduce N losses but do not result in greater crop N uptake will increase alternate loss pathways. Manure N efficiency can be evaluated best in steady-state long-term trials in which the recovery of both the new and legacy N is determined. There have been few long term trials on slurry manure in which low-emission applicators have been used and responses of different grasses to manure and fertilizer is not well known. This long-term study compares recovery of N from surface-banded dairy slurry and fertilizer by three perennial grasses: perennial ryegrass (*Lolium perenne* L.), orchardgrass (*Dactylis glomerata* L.) and tall fescue (*Festuca arundinacea* Schreb.).

Materials and Methods

The experimental site, located on silty loam soil in south coastal British Columbia, Canada, has moderate temperatures and high rainfall (~1500mm). The trial was established in 1996 on a tall fescue stand that received surface-banded dairy slurry and commercial fertilizer (Bittman et al. 2006). Dairy slurry (59 g dry matter, 2.5 g total N and 1.31 g ammonium-N kg^{-1} fresh material) was obtained from high producing dairy farms using sawdust bedding. The nutrients were applied in 4 equal doses: in late March and after the 1st, 2nd and 3rd of four harvests. In 2003 the tall fescue was ploughed down and plots of tall fescue, perennial ryegrass or orchardgrass were seeded in a split-split plot design (4 replicates) with nutrients as subplots and grasses as sub-subplots (years were main plots). Treatments carried out from 2004-2010 included annual total N rates of 194 or 388 kg ha^{-1} as ammonium nitrate (Fert-low and Fert-high), with other nutrients based on soil test, and 351 or 676 kg ha^{-1} as dairy slurry (Man-low and Man-high). There was also a control treatment and a treatment with alternating Man-low and Fert-high (Man/fert). Harvested herbage was sampled for determination of dry matter and N concentration. Apparent N recovery (ANR) was calculated as '(N uptake – N uptake by control)/N applied' while unadjusted N recovery was calculated as N uptake/N applied. Analysis of variance was done as a mixed model with replicates being random and means separation was according to the Fisher protected LSD test.

Results and Discussion

Orchardgrass had significantly higher ANR than tall fescue and perennial ryegrass for both fertilizer treatments, with the highest value (0.70) for Fert-low (Table 1). In contrast, tall fescue had significantly higher ANR rates than the other grasses with both manure treatments but especially the Man-high treatment. Perennial ryegrass ANR was substantially lower than the other grasses and this may be attributed to lower ryegrass yields (not shown). Greater ANR from manure by tall fescue may be due its higher yield and deeper roots (Garwood and Sinclair 1979) but the reason for poorer recovery of fertilizer N by tall fescue is not known. Differences amongst grasses in response to N sources is a novel observation which suggests that N application as fertilizer or manure should be adjusted according to forage grass species and that tall fescue may be used to reduce N leaching from fields receiving high doses of manure. Values for ANR from manure N by tall fescue (0.46 for Man-low and 0.38 for Man-high) were similar to those previously reported for this trial for 1996-2002 (Bittman et al. 2006) (Table 1). Our study shows a lower ANR for manure than fertilizer despite the greater abundance of legacy soil N in the manured soils (data not shown). The N fertilizer replacement values (NFRV, defined as ratio of apparent recovery of manure to mineral fertilizer) averaged ~60, 70 and 80 kg N per 100 kg manure-N applied for, respectively, orchardgrass, perennial ryegrass and tall fescue. Such values are consistent with long-term NFRVs for manure (Schröder et al 2013, Webb et al 2013). Estimates of ANR are influenced by N recovery values in the respective control plots (Table 1) which are deducted to adjust for extraneous N inputs such as ammonia deposition. While wet deposition would be similar, dry deposition from nearby emission sources may be higher in unfertilized than fertilized grass due to lower apoplastic NH_3. Also unfertilized plots may have some N fixation from volunteer legumes (although chemically controlled periodically) and perhaps non-symbiotic fixation. Therefore the values of ANR in this multi-year study may underestimate the true recovery of applied N. The true recovery rate for tall fescue with Man-low, for example, is likely between the ANR value (0.46) and the unadjusted recovery value (0.70). Note that only a small amount of the recovered N (~20 kg ha^{-1} or 6%) is attributable to extraneous inputs (mainly wet deposition).

Table 1. Proportion of applied N recovered in harvested herbage in 2004 -2010 on a trial initiated in 1996. Here N recovery efficiency is adjusted for N uptake in control plots.

	Total	Organic	Mineral	Orchardgrass		Tall fescue		Perennial ryegrass	
	-----------kg ha^{-1}--------			------------------N recover efficiency ratio--------------					
Fert-low	194	0	194	0.70	A	0.58	B	0.50	CD
Fert-high	388	0	388	0.53	C	0.49	D	0.48	DE
Man-low	351	157	194	0.43	G	0.46	EF	0.37	H
Man-high	676	302	374	0.32	I	0.38	H	0.30	I
Man/fert	526	146	380	0.48	DE	0.50	CD	0.43	FG

Proportion of applied N recovered in harvested herbage in 2004 -2010 on a trial initiated in 1996. Here N recovery efficiency is adjusted for N uptake in control plots.

	Total	Organic	Mineral	Orchardgrass		Tall fescue		Perennial ryegrass	
	-----------kg ha^{-1}--------			------------------N recover efficiency ratio--------------					
Fert-low	194	0	194	1.20	A	1.05	B	0.89	C
Fert-high	388	0	388	0.78	D	0.72	E	0.67	GH
Man-low	351	157	194	0.69	FG	0.70	EF	0.57	I
Man-high	676	302	374	0.45	K	0.51	J	0.40	L
Man/fert	526	146	380	0.66	H	0.66	GH	0.57	I

Conclusion

That ANR and NFRV differ among grass species may be due in part to growth differences among species under control conditions, perhaps because of different abilities of species to cope with low soil nutrients or to absorb ammonia from the air. The grass species may also have different preference for nitrate vs ammonium ions in the soil. While some of the unused N is accumulating in the soil, there was no clear evidence for gradually increasing rates of N recovery due to accumulating legacy soil N in this forage system. A better understanding of actual N recovery is needed for nutrient balances.

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TEMPORAL EVOLUTION OF THE IMPACT OF A WOODY BIOCHAR ON SOIL NITROGEN PROCESSES – A 15N TRACING STUDY

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Biochar addition to soils has been proposed as a means to increase soil fertility and carbon sequestration. However, its effect on soil nitrogen (N) cycling and N availability is poorly understood. Furthermore, there is a need for biochar N studies that aim at investigating long-term analogues. In order to investigate the temporal evolution of biochar effect on the gross rates of simultaneously occurring N transformations, we conducted two 15N tracing studies in combination with numerical data analysis using sandy loam soil from a field trial amended with a woody biochar. For the first experiment we sampled immediately after biochar application; for the second experiment one year later.

Materials and Methods

Soil (sandy loam) was collected from a field trial, established in October 2011 at Merelbeke, Belgium. The experimental design was completely randomized. There were two treatments, including a control and biochar treatment, each in four replicates. The biochar dose applied was 20 t ha-1. The biochar feedstock was a mixture from hard- and softwood. Soil was collected twice from the 0-25 cm layer of each plot in the field trial: one day after biochar incorporation (October 2011), and one year after biochar application, in October 2012. Plastic tubes were filled with 70 g oven-dry soil and incubated at 20°C. A water solution containing NH4+ and NO3-, in which one of the N moieties was labeled with 15N, was applied to the soil. Like in the field trial, there were two treatments (a control and biochar treatment) and four replicates (the field repetitions) per treatment. Soils were extracted 0.25, 2, 5, 28, 96 and 216 h after label addition with 120 ml 1 M KCl and shaken for 60 min. The extracts were analyzed for NH4+, NO3- and their respective 15N contents. Multiple gross N transformation rates were quantified for each treatment using a numerical 15N tracing analysis tool (Müller et al., 2007). The N pools considered in the tracing model were NH4+, NO3-, an organic N pool (Norg), and a pool related to the adsorption of NH4+ (NH4+ads) and, for 2012, adsorbed NO3- (NO3-ads).

Results and Discussion

In the short-term experiment (October 2011), gross mineralization of the organic N pool to the NH4+ pool (MNorg), gross immobilization of NH4+ and NO3- into Norg (INH4 and INO3), net mineralization (MNorg – INH4) and gross nitrification (ONH4) rates were significantly ($P < 0.05$) higher in the biochar treatment compared to the control (Table 1). In 2012, one year after biochar application, differences in gross transformation rates between the biochar and control treatments were significant ($P < 0.05$) but small (Table 1). In contrast to the 2011 experiment, NO3- adsorption and release (ANO3 and DNO3a) took place in 2012 in both treatments. The most important gross rates MNorg, ONH4, ANO3 and DNO3a were lower in the biochar than in the control treatment. Net nitrification (ONH4 – DNO3) and NO3- adsorption

(ANO3 – DNO3a) rates were not significantly affected by biochar (P > 0.05). The general trends observed in the short-term experiment of Nelissen et al. (2012), in which two maize biochars were applied to a loamy sand soil one day before 15N addition, were confirmed in the 2011 experiment (which was started just after biochar application), being (i) stimulation of gross N mineralization and immobilization and (ii) stimulation of gross (and net) nitrification. An increase in gross N mineralization could be explained by stimulation of microorganisms degrading more recalcitrant soil organic matter (SOM) 1, resulting in positive priming of native soil organic C. An increase in gross immobilization could be due to biochar's labile C fraction, resulting in microbial demand for inorganic N present in the soil solution. However, biochar may have induced other micro-organism stimulating processes, e.g. a change in soil pH, microbial protection in biochar pores, bacterial adhesion or sorption of compounds that would otherwise inhibit microbial growth, and these could possibly increase the total microbial abundance or activity, thereby consuming more N and thus immobilizing N biotically. An increase in gross nitrification can be due to soil pH elevation (Nelissen et al. 2012) and increased NH4+ substrate supply for autotrophic nitrifiers due to the faster mineralization rate with biochar application. In contrast to the short-term response, the accelerating effect of biochar application on the N cycle had disappeared after one year. This indicates that biochar's stimulating effect on key gross N transformation rates is temporary, probably due to the fact that the biochar properties changed during the year: biochar's labile C fraction has been mineralized, and biochar's pH effect is likely transient.

Table 1: Gross rates (mean and standard errors (SE)) of soil N transformation processes in the control and biochar-amended treatments in 2011 and 2012.

Abbreviation	Description	Kinetics[a]	N transformation rate ($\mu g\ N\ g^{-1}\ day^{-1}$)							
			2011				2012			
			Control		Biochar		Control		Biochar	
			Mean	SD	Mean	SD	Mean	SD	Mean	SD
M_{Norg}	Mineralization of N_{org} to NH_4^+	0	0.731	0.028	0.980	0.043	0.784	0.036	0.755	0.010
I_{NH4}	Immobilization of NH_4^+ to N_{org}	1	0.003	0.002	0.138	0.044	-		-	
I_{NO3}	Immobilization of NO_3^- to N_{org}	1	0.086	0.043	0.526	0.072	-		-	
O_{NH4}	Oxidation of NH_4^+ to NO_3^-	1	0.801	0.022	0.902	0.037	0.878	0.033	0.839	0.014
D_{NO3}	Dissimilatory reduction of NO_3^- to NH_4^+	1	0.015	0.002	0.013	0.002	0.018	0.003	0.008	0.000
D_{NH4a}	Desorption of NH_4^+ from exchange sites	1	0.008	0.001	0.010	0.001	0.001	0.001	0.003	0.001
A_{NO3}	Adsorption of NO_3^- on exchange sites	1	-		-		2.640	0.582	2.317	0.135
D_{NO3a}	Desorption of NO_3^- from exchange sites	1	-		-		1.937	0.517	1.612	0.100

aKinetics: 0 = zero order, 1 = first order; Norg = soil organic N, NH4+ = ammonium, NO3- = nitrate; - = transformations not considered in final model

Altogether, our results show that application of a woody biochar type to a C-poor sandy loam soil accelerated N cycling just after biochar addition through stimulating soil microbial activity. One year after biochar addition, results show that these effects are temporary, probably due to the transient effects of biochar pH and labile C fraction.

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NITROGEN MINERALIZATION UNDER DIFFERENT FERTILIZER AND TILLAGE STRATEGIES

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Spain is ranked as the second European country for pig (S*us scrofa domesticus*) production. Annually, it produces 26 million head of pigs. The use of pig slurry as a fertilizer is a common practice in livestock areas. Slurry N dynamics are affected by slurry characteristics, soil factors, and environmental conditions (Griffin et al., 2002). The plant available N is the sum of NO_3^--N and NH_4^+-N (not including that lost through volatilization or denitrification) plus the organic N that is mineralized for a given time period (Gilmour and Skinner, 1999). Nitrogen availability to crops during the growing season is essential for improving fertilizer-use efficiency and minimizing the adverse impacts of N losses on the environment. Estimates of net N mineralization can be made by incubating manure-amended soil under controlled conditions. The aim of this study was to monitor microbial activity and to assess available mineral N in a soil where different fertilization strategies (which include pig slurries) were combined with different tillage systems (minimum and no-till).

Materials and Methods

A broad N fertilization experiment in winter cereal crops was established (41º52' 29"N; 1º 09'10"E, 443 m.a.s.l., Lleida, NE Spain). Soil was classified as Typic Xerofluent. The texture of the surface layer (0-30 cm) was silty loam. This soil layer has a low organic matter content (average value is close to 2%). The fertilization treatments were maintained until 2012 In the 2008/09 cropping season, plots were divided in two tillage strategies: half under minimum tillage half under no tillage. In the 2011/2012 cropping season, the soil of four high yielding fertilization treatments and a control (no N applied), all of them combined with the two tillage systems, were sampled. This means that eight management strategies (combining fertilization and tillage systems), plus two unfertilized controls (one for each tillage system) were sampled. The four fertilization treatments were: i) 30 t ha^{-1} of slurry from fattening pigs (30PF-0: 70 kg Norg-N ha^{-1} plus 106 kg NH_4^+-N ha^{-1}) applied at sowing (Oct. 2011); ii) 120 kg N ha^{-1} as ammonium nitrate (0-120M) applied at cereal tillering (Feb. 2012); iii) 40 t ha^{-1} of slurry from fattening pigs (0-40PF: 43 kg Norg-N ha^{-1} plus 127 kg NH_4^+-N ha^{-1}) applied at cereal tillering, and iv) 90 t ha^{-1} of slurry from sows (0-90SS: 49 kg Norg-N ha^{-1} plus 123 kg NH_4^+-N ha^{-1}) applied at cereal tillering. The control treatments (no nitrogen, 0-0) and the mineral N treatment received P and K potassium at sowing (96 kg P_2O_5 ha^{-1} and 107 kg K_2O ha^{-1}). At sowing, the slurry was buried as soon possible (< 24h) after application; at tillering, it was left over the soil surface. In May 2012, the 30 plots (ten treatments in each of the three blocks) were sampled in the first 10 cm depth of soil. The incubation (140 g of air-dried and 2 mm sieved soil) was carried out in 2 L hermetic glass jars. Three replicates were incubated for each sample and arranged randomly within the aerobic incubation chambers. Soil incubation was maintained during 52 days at 28±2ºC in the dark, under no-leaching conditions. Soil was adjusted at 65% of its water-holding capacity. Soil sampling was done at 0, 2, 9, 16, 23, 30, 37, and 52 days after incubation and analysed for NH_4^+-N and NO_3^--N content. Soil respiration was measured (Alef, 1995) accounting for the release of CO_2 by an NaOH trap. The statistical analysis was made using the SAS statistical package (SAS Institute, 1999-2001).

Results and Discussion

Accumulated respiration (52 days) was in the range from 442 to 588 mg CO_2-C kg soil^{-1} and a significant linear relationship between the CO_2 produced and the mineral N soil content was found (data not shown). Soil respiration and mineral available N were not affected, during the soil incubation period, by previous tillage management. The soil NH_4^+-N content during the incubation period was low (between 1.4 to 2.3 mg kg^{-1}) and no significant differences between fertilization treatments or tillage systems were found. The predominant mineral N form was nitrates (NO_3^--N). Initially, from the start of incubation up to 9 days later, soil N from the mineral N treatment (120M) was significantly higher than for the control or slurry treatments. But after 16 days onwards, slurry fertilization strategies equalled the mineral one, all of them being significantly higher than a control treatment (Fig.1). This fact indicates a sustained release of N in slurry treatments (from a transitory non-available form) which can catch up with mineral fertilization, both becoming similar in terms of N crop availability.

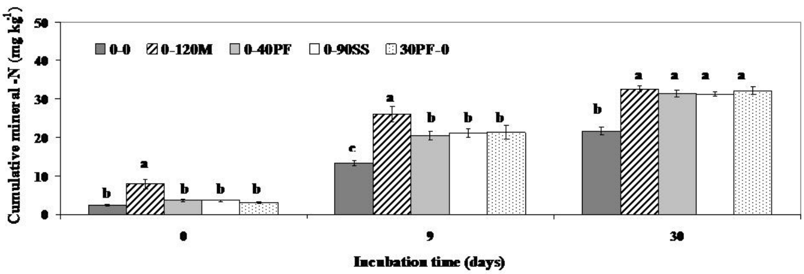

Figure 1. Initial and cumulative mineral–N (NH4+-N plus NO3--N) for each fertilization treatment and at two sampling periods during the incubation period. M: 120 kg ha^{-1} yr^{-1} of mineral nitrogen. FS: slurry from fattening pigs. Numbers indicate the average theoretical applied rate: 30 t ha^{-1} yr^{-1} (at sowing) or 40 t ha^{-1} yr^{-1} (at cereal tillering). SS: slurry from sows applied at cereal tillering (90 t ha^{-1} yr^{-1}). Within columns and incubation time, means followed by the same letter are not significantly different according to Duncan Multiple Range Test at the 5% level of significance.

Conclusions

In May samplings (at the grain filling cereal stage), soil N extraction by KCl in plots receiving slurry treatments at sowing or at cereal tillering did not detect the mineral N that can be potentially available to plants. The incubation period experiments showed that in these plots the NH_4^+-N immobilization was transient, and the final N crop availability was similar for the mineral fertilized plots and for the ones receiving slurries.

Acknowledgements

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PHYSICOCHEMICAL CHANGES AND NITROGEN LOSSES DURING COMPOSTING OF ACACIA LONGIFOLIA WITH PINE BARK

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Taking into account the high availability of Acacias (invasive Fabaceae species) and pine bark, a valorisation approach for these materials may be composting to produce horticultural organic amendments and substrate components. Acacias have large recalcitrant lignin, polyphenol and cellulose contents (Baggie et al., 2004) that do not contribute to raise composting temperatures. However, they are also rich in N whereas other conditions, including particle size, pile dimension and turning frequency affect heat loss and pile temperature (Brito et al., 2008). The objectives of this work were to investigate the physicochemical characteristics and to model the breakdown of OM and N losses during composting of acacia shrubs with pine bark, with different pile turning frequency, and to determine if compost sanitation temperatures are effective at pathogen inactivation and seed destruction.

Materials and Methods

Acacia longifolia shrubs (particles < 80 mm) collected in Portugal (at 40º25' N 8º44' W) were composted (60%/40% v/v) with pine bark (particles < 150 mm) in big piles (100 m3) with higher (A) and lower (B) turning frequency, during 455 days. Temperatures were monitored automatically with thermistors positioned at depths of 0.5, 1.5 and 2.5 m. Physicochemical characteristics were periodically determined by European standard procedures. OM and N losses were calculated from the initial and final ash and N contents according to the formulas of Paredes et al. (2000). Mineralization of OM during composting, determined by the OM lost, was described by the following two component kinetic model:OMm = OM1 [1 - exp(-k1t)] + OM2 [1 - exp(-k2t)]where OM1 and OM2 are pools of mineralisable OM (OMm), k1 and k2 the respective rates of mineralization (day-1), and t the time (days). Data referring to OM losses during composting was fitted to the kinetic equation by the non-linear least-square curve-fitting technique (Marquardt–Levenberg algorithm). The same procedure followed for OM was carried out to describe N losses.

Results and Discussion

The temperatures rose rapidly up to >70ºC in both piles and were >60ºC for several months, suggesting pathogen inactivation, seed destruction and high amount of biodegradable OM as found here by the potential mineralisable OM of the composting biomass (600-620 g kg-1 of initial OM). A two-component first-order exponential equation provided a reasonable description of OM mineralisation of composting biomass, and showed that the pool sizes for labile and recalcitrant OM were similar and in the ranges: 316-331 g kg-1 and 291-299 g kg-1 of initial OM, respectively. Concentrations of conservative nutrient elements (eg P, K, Ca, Mg, Fe) increased in proportion to OM mineralisation, enriching the compost as a nutrient source for agricultural use. Nitrogen concentrations were also increased to a degree, but were much more dynamic and losses were difficult to actively control because high temperature conditions promoted significant N volatilization. Total N content significantly increased ($P < 0.05$) with composting from an initial value of 8.5 g kg-1 DM to final values of 9.5 g kg-1 in pile A and 9.4 g kg-1 in pile B. An increase in total N content during composting has been widely reported (de Bertoldi and Civilini, 2006; Brito et al., 2008), and is due to a lower rate of N loss than OM loss. As expected, NO3--N content in composting piles during the thermophilic phase was negligible because the bacteria responsible for nitrification are strongly inhibited by temperatures greater than 40 °C. This implies that the risk of N leaching was insignificant during this composting period. However, high temperature and high pH conditions during the thermophilic stage probably promoted intense ammonia emission, which would explain high N losses (470 and 450 g kg-1 of initial N for pile A and B, respectively) found during composting, mostly at the initial phase of the process when OM degradation and ammonia production was at most rapid. Differences in N losses between piles were not significant. The C/N ratio declined from 56 at the beginning of composting to final values of 40-41 showing a higher OM mineralization compared to N loss (de Bertoldi and Civilini, 2006). This study indicates that composting Acacia longifolia with pine bark can produce valuable organic amendments with high OM content, and low electrical conductivity (< 0.7 dS m-1). However, a long period of composting is required to achieve advanced compost maturation.

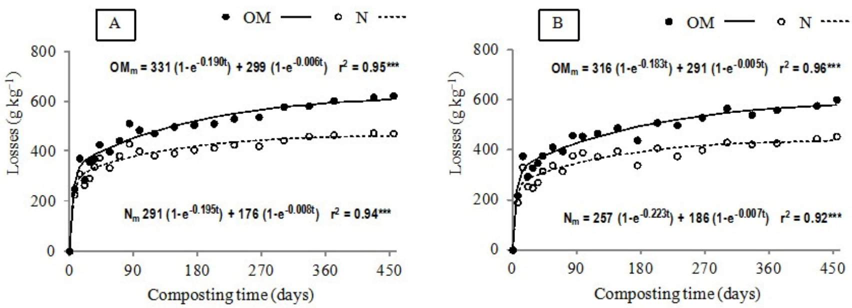

Figure 1. Organic matter (OM) and nitrogen (N) losses (g kg-1) of composting Acacia longifolia with pine bark, with higher (A) and lower (B) turning frequency. *** ($P < 0.001$)

Conclusion

This work highlights that ground and screened acacia shrubs and pine bark have sufficient biodegradability and structure to allow effective composting with increased OM losses compared to N losses.

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NITROGEN LOSSES AND TRANSFORMATIONS DURING STORAGE OF SOLID MANURES OF CONTRASTING N AVAILABILITY

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Manure management is a significant source of ammonia (NH_3) and nitrous oxide emissions (N_2O). A good understanding of the N losses and transformations throughout the manure management continuum is required to produce accurate emission inventories, to develop mitigation practices and to provide guidance on the potential nutrient value of manure for crop growth. This is particularly important for farmyard manure (FYM), where excreta are intimately mixed with bedding materials and a proportion of the more readily available N in the excreted urine is likely to be immobilised. During subsequent storage, the processes of mineralisation and immobilisation may occur, and readily available N will be lost through the processes of NH_3 volatilisation, denitrification and leaching. The objective of this study was to compare these N losses and transformations in FYM storage from manures with contrasting readily available to total-N ratios, specifically pig and cattle FYM.

Materials and Methods

Three experiments were conducted in which pilot-scale heaps of pig and cattle FYM of 3-5t were established in specially constructed bunkers of approximately 3.5 x 3.5m as described by Chadwick (2005). Two experiments were conducted at ADAS Gleadthorpe, starting in October 2010 and July 2011, and one experiment at North Wyke starting in July 2010. Manure was collected immediately prior to each experiment directly from pig or cattle housing close to the experimental location. Three replicate heaps of each manure type were established on each occasion and stored for a 3-month period. The fresh weight of FYM in each heap at the start and end of each storage period was recorded and samples were taken for analyses including dry matter content, pH, total N, ammonium-N and nitrate-N. Gaseous emission measurements were made periodically through the storage period by covering the entire manure heap with a large dynamic chamber for a 2-hour period. For NH_3 volatilisation, the mean concentration of NH_3 in the air entering and leaving the chamber was determined for each measurement period by drawing subsamples of air at controlled rate through absorption flasks containing 0.02M orthophosphoric acid. The emission rate from the heap was then derived as the difference in the concentration of air leaving and entering the chamber multiplied by the total volume of air flowing through the chamber over the sampling period. For N_2O, several samples of air entering and leaving the chamber over the 2-hour period were taken by syringe and stored in evacuated vials for subsequent concentration analysis by gas chromatography. Emission rates were determined as for ammonia. Gaseous emission measurements were made more frequently in the first two weeks of each storage period as this was when greatest fluxes were expected. Cumulative emissions of NH_3 and N_2O were estimated by linear interpolation between measured emission rates. An estimate of total N losses by denitrification was made using the acetylene inhibition technique following the procedures of Moral et al. (2012). On each sampling occasion, three manure samples (approx. 500g) were taken from each heap. Each manure sample was split between two Kilner jars (1l) to which airtight lids with sampling ports were fitted. In one jar from each pair, 60ml of the air was replaced with 60ml acetylene. Jars were incubated for 60 minutes, with gas samples being taken at 0, 30 and 60 minutes to determine N_2O concentration by GC analysis. The N_2O flux was determined from the increase in headspace concentration during the incubation period. The N_2 flux was determined as the difference in N_2O flux between the jar incubated with acetylene and that without acetylene for each pair of jars. Leachate from the stored FYM heaps drained to individual collection tanks. Volume of leachate was measured periodically and subsamples taken for analyses including total N, ammonium N and nitrate N.

Results and Discussion

FYM types differed significantly in terms of their total N and ammonium N contents and, particular, in the ratio of readily available N to total N with average ratios of 28 and 12% for pig and cattle FYM, respectively (Table 1). There were no consistent differences in dry matter content or pH between FYM types.

Table 1. Initial manure composition (fresh weight basis)

		Dry matter (%)	pH	Total N (kg t^{-1})	NH_4^+-N (kg t^{-1})	NO_3^--N (kg t^{-1})	RAN:TN[a] (%)
Jul 2010	Pig	24.8	8.6	7.41	2.32	0.00	31
	Cattle	21.4	8.9	5.20	0.58	0.00	11
Oct 2010	Pig	19.1	8.4	6.15	1.62	nd[b]	26
	Cattle	29.7	8.6	5.90	0.54	nd[b]	9
Jul 2011	Pig	21.2	7.8	7.86	2.00	0.00	25
	Cattle	27.7	8.0	6.37	0.94	0.01	15

[a] ratio of readily available N to total N; [b] not determined

There was a significant mass loss of FYM during storage, ranging from 39 to 63% of the initial heap dry matter. Total N loss ranged from 23 to 60% of initial heap N content (Table 2), with almost all of the ammonium N content being lost and some accumulation of nitrate N. There was no consistent effect of FYM type on net mineralisation, estimated as the loss of organic N (total N minus the ammonium and nitrate N) during storage, with a mean value of 37%, although some evidence that this was higher for the summer storage experiments.

Table 2. Quantities of N in the FYM heaps at the start and end of each storage period

		Start (kg)			End (kg)		
		Total N	NH_4^+-N	NO_3^--N	Total N	NH_4^+-N	NO_3^--N
Jul 2010	Pig	31.1	9.70	<0.01	16.6	2.40	0.17
	Cattle	15.5	1.76	<0.01	8.1	0.03	0.18
Oct 2010	Pig	28.3	7.45	nd	16.5	0.55	nd
	Cattle	28.4	2.62	nd	21.8	0.57	nd
Jul 2011	Pig	30.4	7.74	0.01	13.6	0.21	1.51
	Cattle	17.8	2.64	0.02	7.2	0.08	0.14

Conclusions

Total N losses during FYM storage were not consistently influenced by FYM type or the proportion of readily available N to total N at the start of the storage period. Further conclusions regarding the forms of N loss during storage, including gaseous emissions and leachate losses will be presented at the conference.

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ASSESSING AMMONIA EMISSIONS FROM LIQUID MANURE STORAGE: A MODELLING APPROACH APPLIED TO DANISH AND ITALIAN CONDITIONS

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Ammonia is a recognized pollutant gas mainly emitted from livestock manure (Bouwman et al. 2005), which may cause problems for human health, acidification and eutrophication of natural ecosystems (Sutton et al. 2011). Pollution caused by ammonia is a serious concern in European regions with a great concentration of animal production units. The emission of ammonia depends on manure removal method from livestock buildings, storage of manure in outdoor structures and manure spreading on agricultural land (Oenema 2007). This study has been focused on ammonia volatilization from liquid manure storages, with the aim of comparing the existent emission factors for Italian and Danish manure management. These two countries, situated in different agro-ecological zones, have a high potential for ammonia emissions. European Directives provide general guidelines to regulate the construction and management of manure storages (tanks or lagoons), but regulations in Italy and Denmark have led to different agricultural practices and abatement strategies. Thus, a method to reduce emissions being efficient in Denmark may be inefficient in Italy. Therefore, a modelling approach is suggested to assess ammonia emissions, because the high uncertainty level in measurement techniques and a number of variables that influence the process of volatilization (Shah et al. 2006). For this reasons, a simple model-based decision support system has been developed to predict emissions from storages in Denmark and the northern part of Italy.

Material and Methods

The proposed mechanistic model is written in a simple declarative language, SEMoLa (Danuso 1992), and it has been used for scenarios assessment based on climate, management techniques and regulations. Driving environmental variables that are loaded with meteorological dataset are: hourly temperature, hourly wind speed and rainfall. As suggested by a literature review (Ni 1999), the model has been developed in a number of modules or sub-models. It is generally acknowledged that there is a lack of information, among the cited models, about the sub-model of manure production. In the module of manure production stands the aim of the presented modelling work: the development of a simple decision support system, usable by farmers and stakeholders. Processes considered in the module are: excreta production rates by livestock, techniques of slurry removal from animal houses and slurry removal from storages. The model is able to compare different storage structures (type of covers, size and types of storages) and different slurry removal strategies, which are selectable by model users. It is possible to calculate the size of storage (tank or lagoon) per livestock building, which is dependent on regulation, consistence of livestock, type of animals in animal houses and management techniques. These storage structures have a specific depth and consequently a diverse ratio volume/surface, which is a crucial factor that determines ammonia emissions. Meteorological datasets of

Silkeborg weather station (Denmark) and Capralba weather station (Italy) have been used for simulations. Simulations consider two types of structure for an animal house of 1000 fattening pigs: lagoon with depth 2.6 m and tank with depth 3.79 m, both having the same storage capacity of 3183 m^3, and an emitting area of 1224 $m^2$and 840 m^2 respectively.

Results and discussion

As presented in figures, total yearly emission has been 2853 kg NH_3 for the Italian lagoon, 2033 kg NH_3 for the Italian tank, and 1300 kg NH_3 for the Danish tank. Simulations indicate an increase in ammonia emissions around 38 % when lagoon is preferred to tank as storage structure. A crucial factor to reduce ammonia volatilization is the cover of slurry: it is achievable a reduction of 89% in ammonia emissions using a PVC sheet, and of 50% when a natural crust is formed on slurry surface. For the north of Italy, the proposed model simulates a mean daily emission of 1.05 g m^{-2} in winter, and a mean daily emission of 13.27 g m^{-2} in summer. For Denmark, the model simulates a mean daily emission of 0.89 g m^{-2} in winter, and a mean daily emission of 7.27 g m^{-2} in summer. These simulated values are coherent with a range of values measured (Hristov et al. 2010), which vary from 0.13 g m^{-2} per day in winter to 15 g m^{-2} per day in summer.

Conclusion

Seasonal climatic variability is accounted in the model and, for the North of Italy, it has been shown that ammonia emissions during the summer can be really high. Considering the high ammonia volatilization from lagoons, these storage structures should be avoided, even if they are more cost effective than tanks. Further development of this work will be the validation of the model under Mediterranean condition where, despite the importance of manure management, few studies have been done.

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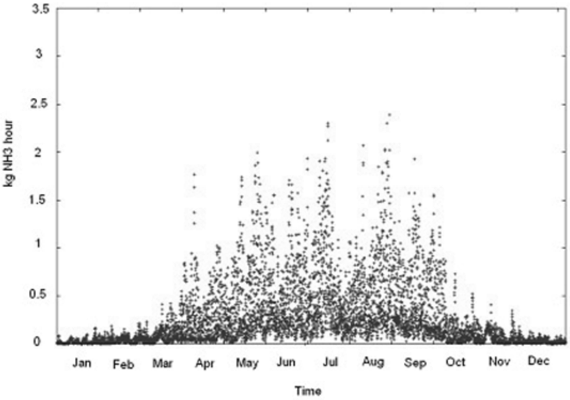

Fig 1: Hourly emission of ammonia from a tank in Capralba for year 2011 (Po Valley)

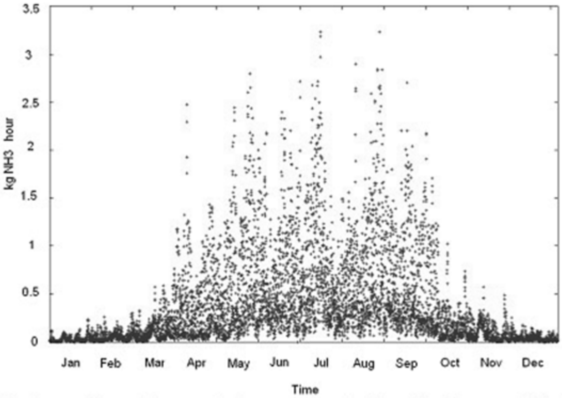

Fig 2: Hourly emission of ammonia from a lagoon in Capralba for year 2011 (Po Valley)

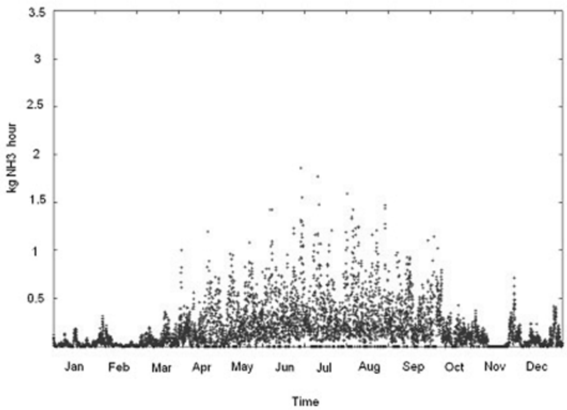

Fig 3: Hourly emission of ammonia from a tank in Silkeborg (Denmark) for year 2011

LOSSES OF NITROGEN DURING THE DECOMPOSITION PROCESS OF ANIMAL MANURE IN STATIC PILES WITHOUT FORCED AERATION

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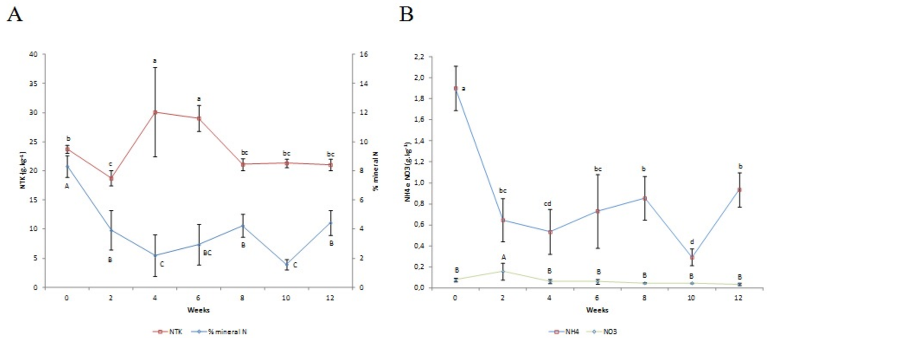
During waste decomposition, ammonia volatilization and denitrification are responsible for the greatest losses of nitrogen in the process (BRITO et al., 2008). Nitrogen losses are enhanced by factors such as temperature and the intensity of gas exchange occurrence, pH increase among others factors. However, a major concern during the decomposition process is to control N losses, since they reduce the composit agronomic value (HAO et al. 2004). During the composting process with forced aeration, it was observed that it is a key-factor for such decomposition. A correct aeration controls temperature, removes the excess of moisture and CO_2 and provides O_2 for biological processes (BERNAL, ALBUQUERQUE, MORAL, 2009). In this process, there have been reports of significant N losses by volatilization (GUARDIA et al., 2010). However, when the decomposition process occurs without forced aeration, the dynamics of nitrogen transformations can be changed. With the aim to monitor the dynamics of N transformations in piles without forced aeration, with cattle and sheep manure, this research was carried out.

Material and Methods

This trial was carried out in Cascavel city, State of Paraná – Brazil at Western Paraná State University – UNIOESTE. Cattle and sheep manure wastes were used from feedlot systems. The same parts of waste were used to prepare four piles without forced aeration at 50% ratio by dry weight. Cattle manure and the sheep litter showed 80% and 67% moisture, respectively. The piles essay without aeration was carried out in an area with just a waterproof canvas as flooring. The piles were not plowed and remained under weather conditions (rain, sun and wind). On average, their initial weights were 487 kg of natural matter and 128 kg of dry matter. Concentrations of total Kjeldahl nitrogen (NTK) were determined according to Malavolta et al. (1997) methodology, while NH_4^+ and NO_3^- concentrations were based on the methodology adapted from Bremner and Keeney (1965), based on samples that were taken at the beginning of the process and at each 15 days until the 12th week. The percentage of mineral N was calculated using the formula: % mineral N = $(NO_3^- + NH_4^+)/NTK + NO_3^-$. Data were submitted to the analysis of variance and the LSD average comparison test at 5% significance.

Results and Discussion

There was no nitrogen mineralization during waste decomposition process in piles without forced aeration process. Considering NTK values, at the beginning and end of the process, it was observed that there was no statistical difference, although it was observed between the fourth and sixth weeks (higher values). Regarding mineral N, which was initially represented by 8% NTK, there was a significant decrease during the process (Figure 1A), also consistent with N-NH_4^- loss (Figure 1B), while NO_3^- levels hardly changed. This finding allows us to infer that N-NH_4^- has not been converted to NO_3^- or it may have been transformed into N_2O by denitrification process due to anaerobic conditions provided by high moisture in the piles, almost 90% (GUARDIA *et al*., 2010). Thus, the nitrifying bacteria did not produce nitrate, except in the first weeks. Such fact is explained by the presence of oxygen in the piles (Figure 1B), as the preparation was done in layers and sheep manure bed had some straw. Therefore, there was oxygen inside the piles even for a short period of time. With the exposure to rain, the piles showed some moisture increase and got compacted further, promoting an anaerobic phase. This fact was commonly found in piles without aeration or turnings.



Same lowercase letters represent equal averages for TKN and NH_4^+, whereas the same uppercase letters represent equal averages for mineral N% and NO_3^- at 5% significance level by LSD test.

Figure 1A. Monitoring of total nitrogen and nitrogen mineralization over time. B. Concentrations of NH_4^+ and NO_3^-.

If N-NH_4^- has not been converted to NO_3^-, it may take NH_3 form, that is lost by volatilization, especially during the first two weeks when the temperature reached average values of 40°C and alkaline pH is nearly 8.65. Moreover, as NKT concentration remained the same, based on the beginning and the end of the process, it can be inferred that not all N-NH_4^- was converted to NH_3 or was lost by volatilization, but probably has been immobilized or transformed into organic forms (LU *et al*., 2013), so, NTK concentrations were kept.

Conclusions

During the process of cattle manure and sheep litter decomposition in piles without forced aeration, there was no N mineralization as there was no nitrate concentration.

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NITROGEN MINERALIZATION DURING CO-ANAEROBIC DIGESTION OF WASTE FROM CATTLE AND SHEEP

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Data from the National Association of feedlots (ASSOCON) show that livestock production in Brazil, in 2012, was 3,866,531 million heads, while data from ANUALPEC portray that sheep production in 2006 was 17,105,572 million heads. This panorama depicts the environmental problem given the amount of waste generated during production, as reflected in an energy and nutrient source for crop production, considering the significant amount of nitrogen in the material. However, for there to be N uptake by the plant, it must be in its inorganic form, that is, in the form of ammonium ($N\text{-}NH_4$) or nitrate ($N\text{-}NO_3$), which are, according Parron et al. (2003), assimilated by microbial population in the soil and by plants. Thus, this work aimed to evaluate the transformation of TNK (total nitrogen by Kjeldahl) in inorganic forms, $N\text{-}NH_4$ and $N\text{-}NO_3$, during anaerobic co-digestion of waste from cattle and sheep.

Materials and Methods

Anaerobic co-biodigestion was conducted at the Laboratory of Analysis of Agroindustrial Waste of UNIOESTE – Campus Cascavel – Paraná – Brazil. Four sets of digesters bench, made of PVC and compounded by a fermentation chamber (capacity of 7 L), gas meter and mini-biodigesters (PET bottles) with a capacity of 500 ml were used.

The substrate was obtained by mixing manure from cattle and sheep, considering 5% of total solids in a ratio of 1:1 in the dry matter. The four fermentation chambers were stocked with six liters of substrate and 40 PET bottles with 500 mL.

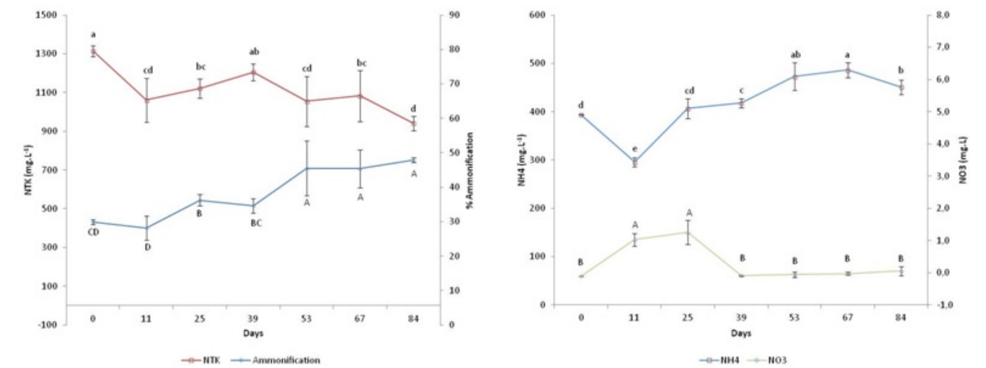
Weekly, four PET bottles were opened for determination of total nitrogen by Kjeldahl, ammoniacal nitrogen, nitrate and pH, performed according to *Standard Methods for the Examination of Water and Wastewater* (APHA, 2005). The efficiency of conversion from organic nitrogen to ammoniacal,that is, the percentage of ammonification, was calculated from the equation (eq. 1).

Eq. (1) $A(\%) = (1- (TNK_i – NNH_4{}^+{}_f)/TNK_i) \times 100$

The results obtained were subjected to analysis of variance and the averages were compared with each other by LSD test at 5% probability.

Results and Discussion

According Florentine at al. (2010) the anaerobic digestion is a natural process that occurs in anoxic environment, whereby certain groups of bacteria promote self-regulated and stable fermentation of organic matter. In Figure 1A, we observed that after 53 days of co-digestion process, 48% of the TNK was transformed by ammonification in mineral-N. This process ensues because of the absence of oxygen. Considering the concentration of NH_4^+ up to 53 days, it is observed an increase of 20.1%. This information may influence the choice of the retention time. Concerning the nitrification process (Figure 1B), it appears that it occurs only at the beginning of the process, because according to Siqueira et al. (2006), nitrification is a process that converts NO_2^- into NO_3^-, setting an oxygenated environment.



Same lowercase letters represent equal averages for TNK and NH_4, whereas equal capital letters represent equal averages for % of ammonification of NO_3 to 5% of significance by LSD test.

Considering the biofertilizer obtained until 53 days, 44.8% of N will be available to the plant, making it interesting for crop production.

Conclusions

It possible to obtain an organic fertilizer with a significant amount of mineral-N after submitting cattle and sheep waste to an anaerobic co-digestion process.

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NITROGEN MINERALIZATION IN SOLID FRACTION OF CATTLE AND SHEEP LITTER WASTE

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Vermicomposting is widely used in the treatment of both solid and semisolid agroindustrial waste; the advantages of its use are the low operational and capital cost, operation simplicity and high efficiency (Carvalho et al., 2009). The moisture content of the solid fraction of cattle waste and sheep litter taken from feedlots lays around 80% and its electrical conductivity is relatively low, because its minerals are dissolved in the liquid fraction. This factors allow the usage of vermicomposting in the treatment of these waste. Vermicomposting concentrates nutrients as, for example, the Total Kjeldahl Nitrogen (NKT), fact that can be justified by the mineralization of the organic matter performed by warms (Dores-Silva et al., 2011). However, the loss of mass, especially in the form of carbon, leads to an increase of the concentration of nutrients over time, giving a false impression of mineralization. The most accurate way to affirm that there is mineralization during the process is to monitor the inorganic forms of N, NH_4^+ e NO_3^-, which are the results of the process of ammonification and nitrification (Aquino et al., 2005). The aim of this paper was to observe the forms of N for the period of 12 weeks during the process of vermicomposting in order to know if there is mineralization.

Materials and Methods

Samples of cattle waste and sheep litter were taken from feedlots systems, homogenized and analyzed in order to have determined the total solids, aiming to obtain a mixture of 1:1 in the natural matter. The waste were mixed with water and placed in fiber boxes remaining there for 24 hours. After that, it had the liquid and solid phases separated by the use of a 5 mm mesh. The material kept in the mesh was drained for 24 hours and then have its moisture content determined. The vermireactors were made of wood, solid walls and opened bottom, with dimensions of 0.15 x 0.28 x 0.40m of height, width, and length respectively. In order to avoid the escape of worms and the entrance of predators, the vermireactors were covered with a dark canvas. 300g of dry matter was used in each of the four vermireactors (experimental units), so the height of the waste would not surpass 10 cm; 10 adult worms of the specie *Eisenia fétida* were inoculated. Moisture content was kept at 75%. Concentrations of NKT were determined according to Malavolta et al. (1989) and concentrations of ammonium and nitrate (NH_4^+ e NO_3^-), according to Bremner; Keeney's (1965) adapted methodology in samples collected in the beginning of the process and at each fifteen days over the first 12 weeks.

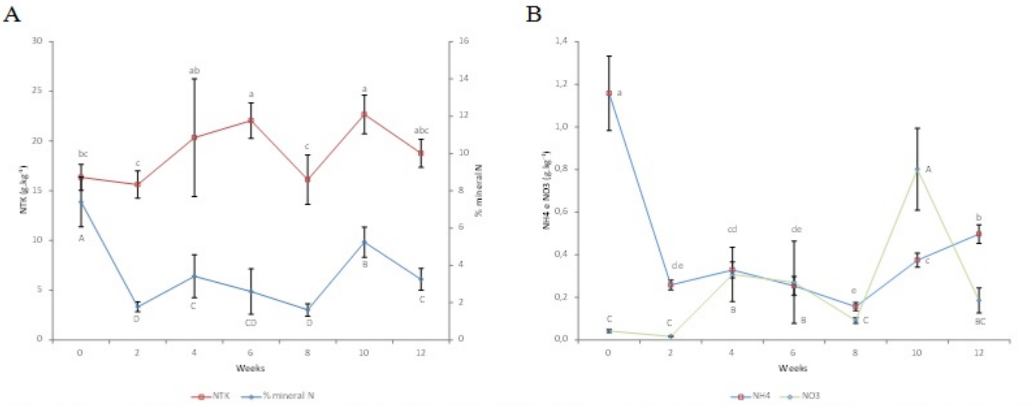
A % of N mineral was calculated according to the formula:

$$\%N\ mineral = (NO_3^- + NH_4^+)/NTK+NO_3^- \times 100$$

Data was analysed anova and the LSD test for comparison of means at 5% significance.

Results and Discussion

Figure 1A shows an increase in the concentration of NKT during the 12 weeks of vermicomposting ($p < 0,05$) which is explained by the loss of mass (25,75%), specially the C (10,16%) in the period.



The use of equal low case letters represent equal means for the NKT and NH_4^+, while equal capital letter represent equal means for the % of N mineral and NO_3^- at 5% significance on LSD test.

Figure 1 Means of NKT, % of N mineral (A), NH_4^+ and NO_3^- (B) over time.

The % of N mineral reduced from the first to the last week ($p < 0,05$). This occurred due to the high reduction of NH_4^+ on the two first weeks of vermicomposting (Figure 1B), fact that is explained by the auto-regulation of the environment caused by the microorganisms, releasing N in the form of ammonium (NH_3). When released, the NH_3 caused the death of the worms. Latter, new worms were inoculated, this time, together with a stabilizing material which serves as a refuge to them. Once the concentration of ammonium was reduced, the worms reproduced normally. In the vermicomposting an average of 80 young, 10 adults and 67 eggs per experimental unit were found. In general, the process of vermicomposting allowed nitrification. However, the concentrations of NO_3^- did not follow a defined patter, demonstrating high picks (10 weeks) and low picks (8 and 12 weeks). The variation in the forms of N is related by Aquino et al. (2005) as a consequence of the immobilization in the bodies of the worms and by nitrification. Another source of variation is the death of the worms, which decompose rapidly, and may lead to variations in the inorganic forms of N during vermicomposting.

Conclusions

The formation of nitrate (nitrification) and the increase in the concentration of NKT happens during vermicomposting of the solid fraction of cattle waste and sheep litter.

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BIOCHAR FROM DIFERENT RESIDUES HAVE A DISTINCT IMPACT ON SOIL N DYNAMICS

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The use of biochar, a carbonaceous material obtained by pyrolysis of biomass, is known to interact with key processes involved in soil N cycling: mineralization, denitrification, nitrous oxide emissions and N fixation [1]. The aim of this work is to study the impact of biochars from different lignocellulosic residues upon the N mineralization dynamics and N availability in an agricultural soil amended with either sheep manure or mineral fertilisation.

Material and Methods

Table 1: Properties and elemental composition of biochars

	OAK	GHW	PRE	CEL
Ash (%)	8.4	16.7	76.7	50.0
VM (%)	24.4	25.0	11.7	26.4
Lignin (%)*	31.3	24.9	13.0	24.4
Cellulose (%)*	52.8	56.9	22.8	55.8
Hemicellulose (%)*	14.7	6.5	< 0.5	0.6
pH	9.6	9.5	10.2	9.4
EC (dS $m^{-1)}$	0.58	1.27	1.14	2.25
C_T (%)	71.53	63.95	17.55	37.37
N_T (%)	0.33	1.14	0.90	1.25
P (g/kg)	0.07	0.23	0.55	0.42
K (g/kg)	0.90	3.29	1.07	0.82

VM: Volatile matter; EC: Electrical conductivity; CT: total carbon; NT: total nitrogen; *Ash free bases.

Biochars were produced by the Energy Research Centre in the Netherlands (ECN) by slow pyrolysis at 400ºC. Four biochars (table 1) were selected from feedstock with different lignocellulosic composition: Oak biochar (OAK) as reference lignocellulosic biomass; Greenhouse waste biochar (GHW) from paprika crop residue; Press cake biochar (PRE) from an anaerobic digestate of the organic fraction of municipal solid wastes and CellMatt biochar (CEL) from an autoclaved organic fraction of municipal solid wastes. Sheep manure was used as organic amendment (N: 1.9 %; P: 1.56 %; K: 2.44%) and Diammonium phosphate (DAP) was used as mineral fertiliser (total N: 18 %; P: 26 %).

A haplic calcisol soil (loam-sandy texture) from an organic olive orchard in Murcia Region (SE Spain) (coordinates 38°23' N 1°22' W) was used for the incubation experiment (pH: 8.1; electrical conductivity: 0.51 dS/m; Total organic C:1.7 g/100g; Total N: 0.3 g/100g). The soil incubation experiment was performed in aerobic conditions with the following treatments: S: control soil; S+B 1%: soil with biochar at 1% (dry weight basis); S+B+M 1%: soil with biochar 1% and sheep manure 1%; S+M 1%: soil with sheep manure 1%; S+B+F 1%: soil with biochar 1% and mineral fertiliser (DAP) 1%; S+F 1%: soil with fertiliser 1%. Three replicates of each treatment were incubated in 100 ml glass containers containing 40 g of soil with the respective amendments during 30 days at 25 °C and 40% of its water holding capacity (WHC). Extractable NO_3^--N and NH_4^+-N contents were determined after three and 30 days of incubation. NO_3^--N was determined by ionic chromatography (HPLC) in the 1:20 (w:v) aqueous extract and NH_4^+-N by a colorimetric method based on the Berthelot´s reaction in the 1:20 (w:v) 2M KCl extract.

Results and Discussion

The levels of soil NH_4^+-N were not affected by the addition of biochar, due to the low supply of mineral N from the biochar and its low mineralization rate in soil. However, the amount of mineral N after 30 days of incubation was affected by the lignocellulosic composition of the biochar feedstock. PRE and CEL biochars, with a low organic matter content (high ash) showed higher amount of available–N than the rich lignocellulosic materials and soil control.The interactions of biochar on NH_4^+-N were observed in the treatments with either the organic amendment (manure) or the mineral fertiliser (DAP). A reduction of the amount of NH_4^+-N was observed after three days of incubation in the treatment prepared with CEL and OAK biochars and manure (S+B+M 1%) or mineral fertilisation (S+B+F 1%), compared to the soil amended with manure (S+ M 1%) or fertiliser alone (S+F 1%). The low availability of mineral N in both treatments containing CEL and OAK biochar may be due to a lower mineralisation rate of the organic amendment in the soil or a fast conversion of NH_4^+-N to NO_3^--N. After 30 days of incubation all biochar amended soils showed similar NO_3^--N levels than the treatment with only manure (S+M 1%) (Figure 1). A similar impact was observed in the treatments prepared with fertiliser and biochar (S+B+F 1%), where the addition of either OAK or CEL biochar originated a reduction of NH_4^+-N, suggesting that the soil N dynamics at the beginning of the incubation were different for each biochar.

Conclusions

The different lignocellulosic composition of the feedstock used for the preparation of the biochar affected the initial N dynamics in the agricultural soil amended with either an organic amendment or mineral fertiliser. However, the levels of NO_3^--N are similar or slightly lower than the control after 30 days of incubation. From an agricultural point of view, this phenomenon may reduce the risk of NO_3^--N lixiviation to groundwater when using N-rich organic amendments.

Acknowledgement

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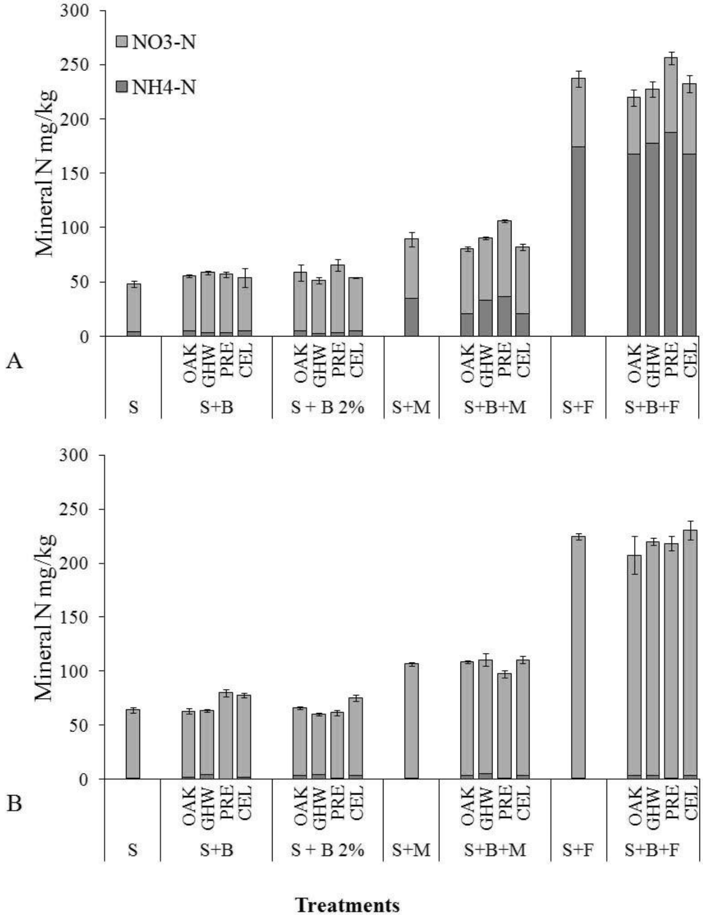

Fig. 1. Mineral N (NH_4^+-N and NO_3^--N) in the soils amended with the different treatments after 3 and 30 days of incubation (A and B, respectively).

THE FATE OF CROP AVAILABLE NITROGEN FROM CONTRASTING ANAEROBIC DIGESTATE AND PIG SLURRY APPLICATIONS TO ARABLE LAND

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Anaerobic digestate (also known as biofertiliser) is produced from the digestion of biodegradable materials (e.g. food waste, livestock manures or purpose-grown crops) in the absence of oxygen, releasing biogas that can be used to provide heat and/or power. Food-based digestate is potentially a valuable source of crop available nitrogen (N), with typically 80% of its total N content present in a readily available (i.e. ammonium N) form. N availability from livestock manures has been widely researched, however, digestate is a 'new' organic material and there is uncertainty about how much of the readily available N will be utilised by crops and the potential for N losses to air (ammonia and nitrous oxide emissions) and water (nitrate leaching). This paper reports results from a field experiment which quantified ammonia and nitrous oxide emissions to air, nitrate leaching losses to water and crop available N supply from autumn and spring applications of food-based digestate and pig slurry.

Materials and methods

An experiment was set up in harvest season 2012 on a sandy loam textured soil at Wensum (Norfolk). Food-based digestate and pig slurry were applied by surface broadcast and trailing hose techniques to arable stubble in August 2011 before the establishment of winter wheat, and top-dressed to the growing winter wheat crop in February 2012 (Table 1). There were 3 replicates of each treatment and an untreated control arranged in a randomised block design. On each treatment, ammonia emissions were measured using windtunnels for 7 days and nitrous oxide emissions were measured with static chambers (5 per plot) for 12 months after application. Nitrate leaching losses were measured from the autumn treatments using porous cups (6 per plot) installed at 90 cm depth with water samples taken after every 25 mm of drainage. Yield and grain offtake measurements were made at harvest. Additional plots receiving manufactured fertiliser N applications at six incremental rates (0-250 kg/ha N) were used to quantify the fertiliser N replacement value (based on crop yields) of the different liquid organic material applications.

Manure type	Application date	Application rate (m^3/ha)	Dry matter (%)	pH	Total N (kg/ha)	Ammonium-N (kg/ha)
Food-based digestate	30/08/2011	32	5.4	8.8	245	201
Food-based digestate	22/02/2012	30	4.4	8.7	207	185
Pig slurry	30/08/2011	41	7.5	7.5	122	92
Pig slurry	22/02/2012	38	2.7	8.0	98	85

Results and discussion

The proportion of readily available N to total N was similar for both organic materials (82-89% for food-based digestate and 75-87% of total N applied for pig slurry). There was no effect (P>0.05) of application method on any of the N loss pathways following food-based digestate or pig slurry applications. Ammonia emissions following autumn application (at 58% of total N applied for food-based digestate and 39% of total N applied for pig slurry) were greater (P<0.05) than following spring application (19% of total N applied for food-based digestate and 21% of total N applied for pig slurry). The higher emissions in the autumn were probably a reflection of soil conditions at the time of application when the ‘dry’ (i.e. hydrophobic) soil surface of the arable stubble restricted infiltration into the soil, and may also have been influenced by the higher pH of the food-based digestate than pig slurry. In spring, the applications infiltrated rapidly into the cultivated soil, reducing the potential for ammonia losses (Figure 1). There was no effect (P>0.05) of application timing on nitrous oxide emissions with losses ranging between 0.09-0.80% of total N applied for food-based digestate and 0.20-0.58% of total N applied for pig slurry. Over- winter drainage volumes were estimated at 92 mm, and mean nitrate leaching losses were 15% of total N applied from the food-based digestate treatment and 17% of total N applied from the pig slurry treatment.

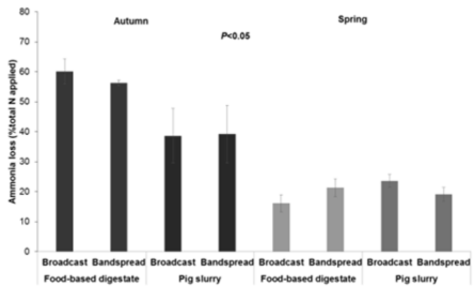

Figure 1. Ammonia emissions following autumn and spring food-based digestate and pig slurry applications to arable land at Wensum (harvest year 2012)

Grain yields on the spring treatments were greater (P<0.05) than on the autumn treatments with increases of *c.*1.5 t/ha for the food-based digetsate and *c.*1.0 t/ha for the pig slurry treatments. The mean N use efficiency (compared with manufactured fertiliser N applications) of the spring timings (81% for food-based digestate and 77% for pig slurry) was greater (P<0.05) than for the autumn timings (23% for food-based digestate and 40% for pig slurry). The greater N use efficiency of the spring applications reflected the lower N losses via ammonia emissions and nitrate leaching losses compared with the autumn application timings.

Conclusions

There was no effect of application method on ammonia or nitrous oxide emissions from either the food-based digestate or pig slurry applications. Ammonia emissions were greater following autumn applications to a ‘dry’ arable stubble which restricted infiltration of the liquid organic materials into the soil. Nitrous oxide emissions from all the food-based digestate and pig slurry applications were below the IPCC default value of 1% of total N applied. N use efficiency was higher from the spring applications reflecting lower N losses via ammonia volatilisation (as the liquid organic materials infiltrated rapidly into the soil) and nitrate leaching compared with the autumn applications.

Acknowledgements

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Cordovil: Expresso coffee residues: a valuable source of nutrients

NITROGEN DYNAMICS IN POULTRY LITTER BY SIX SUBSEQUENT LOTS

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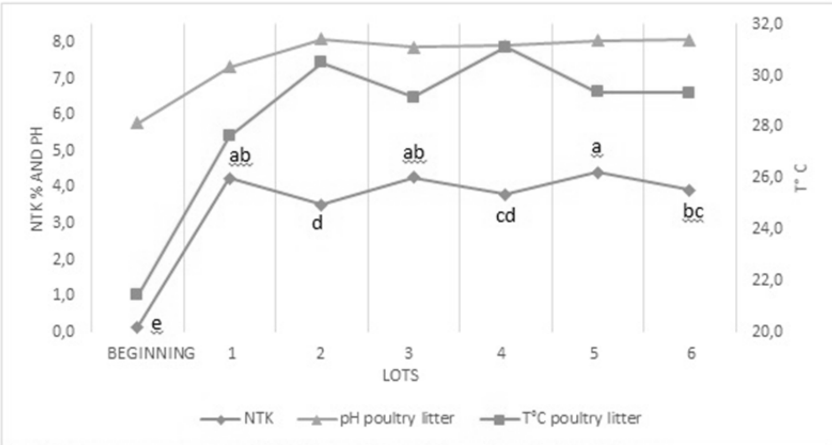
In 2012 the State of Paraná-BR produced 25% of total nationwide production of chickens that has a little more than a billion heads. The west region of the state contributed with almost 70 million of heads according to IBGE (Brazilian Institute of Geography and Statistics). Another expressive activity in this region is the planting of soybeans and corn as the high request for nutrients, as nitrogen, an important macronutrient acting in physiological plants processes, just as photosynthesis, briefing development and root activities, and ionic absorption of other nutrients, growing, cellular and genetic differentiation. This way, allying poultry production, which byproduct is poultry litter, reused up to twelve times, containing animals excreta that are, after all, rich in nitrogen, there's a lock in productive cycle with demand and supply of nutrients, culminating in the reduce at production's cost. After this, aimed to evaluate nitrogen's dynamics into poultry litter at poultry rearing focusing at its usage as agricultural fertilizer.

Materials and Methods

Samples of poultry litter were collected in an aviary located at western region of Paraná-BR with a capacity for housing 25000 chickens. The climate classifications, according to Köppen, is Cfb. The lots of chickens are housed in the aviary on average by 45 days, with sanitary intervals of 15 days. The pH data and temperature at poultry litter are collected in 27 points, distributed at the aviary. The aviary was divides in four sectors, where were collected samples of the poultry litter in different points, forming four composite samples, analyzed separately. To gauge the pH and temperature of the poultry litter, was used a pH and temperature meter model TESTO 205. The total nitrogen (NTK) was analyzed with the assistance of the Kajeldahl distiller according to the methodology proposed by Malavolta et al. (1989). The experimental design used was randomized blocks. For the average comparison test, there was used o LSD test at 5% of significance, to correlate pH and temperature of poultry litter was employed the Pearson correlation test at 5% of significance.

Results and Discussion

It can be observed at Figure 1 that after the accommodation of the sixth consecutive lots of chickens, there is no increase of NTK concentration. There occur small variations at nitrogen percentages from one lot to another, which are negatively correlated (r= -0,68) with the poultry litter temperature, indicating thus and inverse proportionality, which means, when the temperature of poultry litter raises the NTK decreases. There is yet the dominant N at poultry manure is the ammonium ionic form (NH_4^+), which is easily converted to ammonia (NH_3) at alkaline pH ranges (MARÍN, 2011). This way, it is inferred that after the first lot, the pH of poultry litter get ups 7.0, getting closer to 8.0 in the following lots, influencing straight to N loses by NH_3 volatilization. These parameters, allied to urease enzyme activity that occurs more sorely in ways containing vegetable residues with a humidity close to 20%, as in case of chicken litter, end up causing environment's self-regulation, because poultry excrete the same extent that nitrogen is lost to the environment. Countless works are made aiming nitrogen retention at poultry litter, focusing its usage as agricultural fertilizer.



Equal letters represent averages equal NTK% between lots in 5% significance level by LSD test.

FIGURE 1. Average pH, poultry litter temperature (°C), and NTK percentage average per lot.

Oliveira et al. (2003) concluded that agricultural gypsum ($CaSO_4$) can be added to the poultry litter, promoting pH's reduction and consequent increase in nitrogen retention. Loch et al. (2011) notice the same effect with the addition of aluminum sulfate, also due to the reduction of pH at poultry litter. Besides causing impoverishment of poultry litter the loss of nitrogen to the environment at the ammoniac form cause has a straight influence over health as much as chicken' as humans', contributing for the development of breathing illness, culminating in damage for the farmer.

Conclusions

Accumulation of NTK at poultry litter does not occur over six consecutive batches.

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NITROGEN MINERALISATION BY VERMICOMPOSTING IN CATTLE MANURE AND SHEEP LITTER

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Dores-Silva et al. (2013) aiming to determine which process (composting or vermicomposting) is most effective over the short term in stabilize organic wastes concluded that vermicomposted materials showed higher stability, proving a superior tool for stabilization of organic wastes. The use of vermicomposting for treatment of agroindustrial solid and semisolid wastes has the advantages of low capital cost, simplicity of operation and high efficiency (Carvalho et al., 2009). The vermicomposting concentrated nutrients, such as Total Nitrogen by Kjeldahl (TNK). Fact justified by mineralization of organic matter carried by worms (Dores-Silva et al., 2011). However, it is important to determine the concentration of inorganic forms of N is obtained in the vermicomposting, because are these forms important to plant grow. In order to find whether there is mineralization in vermicomposting, inorganic forms of N, NH_4^+ and NO_3^- were monitored, which are the result of the processes of ammonification and nitrification.

Materials and Methods

The cattle manure and sheep litter were removed of confinement systems. Samples were taken of the residues which were homogenized and brought to laboratory for determination of dry weight. Subsequently, it was calculated the required amounts of natural matter to obtain a ratio of 50/50% of each residue in dry matter.

The residues were mixed and composted for 30 days with weekly turnings. It used 0.3 kg of dry matter in each of the four vermireactors (experimental units) so that the height of residue did not exceed 10 cm. The vermireactors were built of woods with dimensions of 0.15 x 0.28 x 0.40 m in height, width, and length, respectively, and cast background. To prevent escape of earthworms and entry of predators, the reactors were sealed with porous dark screen.

Concentrations of TNK were determined according Malavolta et al. (1989), and the concentrations of ammonium and nitrate (NH_4^+ and NO_3^-), using suitable methodology from Bremner; Keeney (1965), sampled at the beginning of the process and every 15 days until the 12th week.

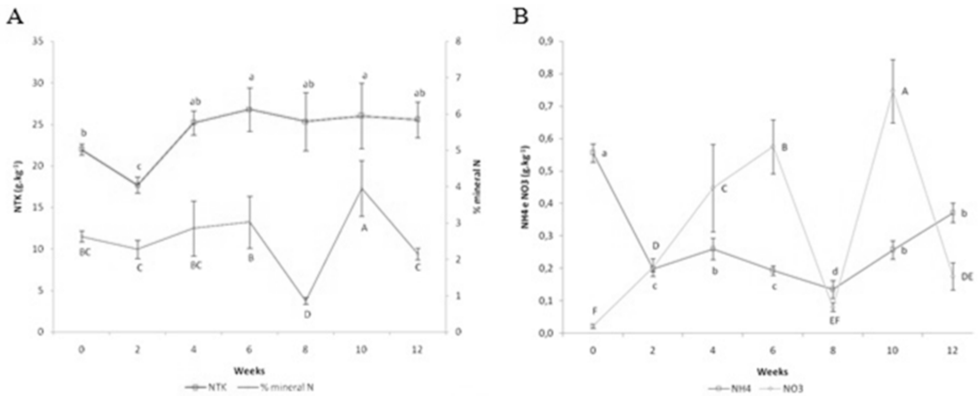
The percentage of mineral N was calculated using the formula:

$\%\text{mineral N} = (NO_3^- + NH_4^+)/TNK + NO_3^-$

The results obtained were subjected to analysis of variance and the averages were compared with each other by LSD test at 5% of significance.

Results and Discussion

Throughout the vermicomposting process, transformations of nitrogenous compounds occur. Organic complexes are mineralized to inorganic forms (Dores-Silva et al., 2011). For Aquino et al. (2005), every activity there is a component of mineralization of N immobilization in which organisms immobilize inorganic fractions for growth and maintenance of their biomass. It is observed in Figure 1, the variation in the forms of N during 12 weeks of vermicomposting process. The concentration of TKN change significantly until the second week thereafter remained constant (Figure 1A). There were variations in % of mineral N during the process, however, at the end of twelve weeks, the mean value was equal to the initial ($p \geq 0.05$). Variations in % of mineral N were caused by the sudden change in the concentration of NO_3^-(Figure 1B). According Aquino et al. (2005), variations in the forms of N are results of nitrification, immobilization in the body of the worm and release after the death of it.



Same lowercase letters represent equal averages for TNK and NH_4^+, whereas same capital letters represent equal averages to % of N mineral and NO_3^- by LSD test at 5% of significance.

Figura 1 (A) TNK averages, % of mineral N, (B) NH4+ and NO3- over 12 weeks.

Although throughout the process NO_3^- showed no defined behavior, the final concentration of NO_3^- was greater than the initial ($p < 0.05$), confirming the hypothesis that there is nitrification in vermicomposting, as exposed by Aquino et al. (2005). The concentration of NH_4^+ decreased rapidly, from the beginning until the second week ($p < 0.05$). This caused by the transformation of NH_4^+ into ammonia (NH_3), which is volatile and die away to the atmosphere. This process is commonly performed by microorganisms in decomposing of organic materials as a way of adapting the environment to their ideal conditions for growth and reproduction.

Conclusions

There are nitrification and ammonification in the vermicomposting process, and increase of $NO_3^-$and NH_4^+ reduction.

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NITROGEN MINERALIZATION IN BIOLOGICAL PROCESSES OF BEEF CATTLE WASTE STABILIZATION

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The efficiency of organic fertilizers to dispose nutrients, especially N, is directly related to inorganic form. The most appropriate way to state whether there is mineralization in the process is to monitor the inorganic forms (ammonium and nitrate). Thus, in order to get biological stabilization of beef cattle waste, it has been managed some composting processes as vermicomposting and anaerobic digestion, or they are left to be decomposed in static piles without any management or aeration. Whereas the result of stabilization processes is the produced organic fertilizer, it becomes an important input to evaluate its agronomic quality based on the chosen process. Each process has advantages and disadvantages, so, this study aimed at evaluating inorganic forms and their percentage in relation to NTK in final products of four stabilization processes of beef cattle waste.

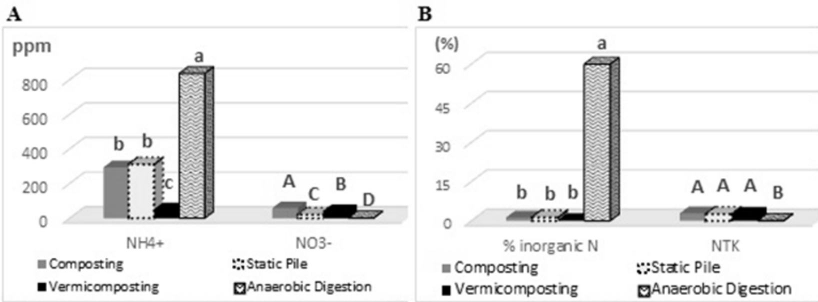
Material and Methods

Feces and urine of cattle, raised in loose housing confinement (with dry scraping management of paddocks) system, were tested. Five composting piles and five static piles were built with 500 kg of cattle manure in raw matter. Manure had 70% moisture yielding 150 kg of dry matter. Composting piles were made on cement floor in a covered yard, were weekly turned over and moisture maintained at 60%. Static piles were not turned over and remained in an outdoor yard with floor, under environmental conditions. Five worm-reactors were built in wood, with solid walls and hollow depths. Dimensions were 0.15 x 0.28 x 0.40 m in height, width and length, respectively. To prevent earthworms escape and predator invasion, the worm-reactors were covered with a dark screen. In each worm-reactor, 0.3 kg dry matter manure was introduced so that the height of manure did not exceed 10 cm, avoiding anaerobic conditions. Ten adult earthworms *Eisenia foetid* species were inoculated in each reactor. The moisture was kept at 75% until the end of the process. For the anaerobic digestion process, five sets of biodigestors were used under batch system. They were made of PVC and divided in the fermentation chamber (7 L capacity) and gasometer. The supplying substrate was obtained by the waste mixing with water, totalizing 6% total solids. In all cases, the duration was 126 days. In solid samples, total Kjeldahl N (TKN) (Malavolta et al. 1989), and ammonium ($N\text{-}NH_4^+$) and nitrate ($N\text{-}NO_3^-$) were determined according to the adapted methodology from Embrapa (2009). For liquid samples, the parameters were determined according to Standard methods for the examination of water and wastewater (APHA, 2005). Inorganic N percentage was calculated using the following formula: % inorganic N = $(NO_3^- + NH_4^+)/NTK + NO_3^-$.

Thus, the results were obtained by the averages comparison analysis and Tukey at 5% significance level.

Results and Discussion

The composting processes, static pile and vermicomposting are less efficient in mineralizing and keeping greater amounts of N under inorganic forms (Figure 1 B). The high temperature in thermophile stage during composting and alkaline pH (Bernal et al, 2009; Lu et al, 2013), aeration (Gómez-Brandón et al, 2008) were promoted by turnover or by earthworms (vermicomposting) or denitrification (Brito et al., 2008) under oxygen low concentration conditions (static pile) contributed on losses of nitrogen processes.



Same lowercase letters represent equal averages for NH_4^+ (A) and % inorganic N (B). Equal uppercase letters represent equal averages for NO_3^- (A) and NTK (B) at 5% significance level by Tukey test.

Figure 1 NH_4^+ and NO_3^- averages (A); inorganic N and TKN Percentage (B) after 126 days in the processes of: Composting, Static Piles, Vermicomposting and Anaerobic Digestion.

Anaerobic digestion produced a biofertilizer with the highest concentration of inorganic N, mainly in NH_4^+ form, due to anaerobic conditions. Organic N is converted to ammonia nitrogen during the ammonification process and this form can be assimilated by plants (Silva et al., 2012). Therefore, to produce large amounts of ammonium and minimal loss of N during the process, the anaerobic digestion showed the highest percentage of mineralization ($p < 0.05$) when compared to NTK (Figure 1 B). The weekly turnover for composting and the continuous one for vermicomposting introduced oxygen in the waste. Since there were the greatest offers of O_2, there were the highest concentrations of NO_3^-, among the four biological stabilization processes. The composting followed by vermicomposting were the most effective ($p < 0.05$) to generate a product rich in nitrate.

Conclusions

The anaerobic digestion process mineralizes the greatest amounts of nitrogen under ammonium form in beef cattle waste. Composting generates a product with the highest nitrate concentration (N-NO_3^-).

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USING CARBON: NITROGEN RATIO AS AN INDICATOR OF THE NITROGEN REPLACEMENT VALUE OF ORGANIC RESIDUES

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Apart from manure, various new organic fertilizers based on residues are now being used in agriculture. It is important to apply the right amount in order to get the desired effect on yield and avoid undesired leaching of excess nitrogen (N) to the environment. This requires knowledge of the proportion of plant-available N in the organic fertilizer, which can be expressed as the N fertilizer replacement value (NFRV). The NFRV can be measured in crop experiments, but this is time-consuming and cannot be done for every single residue. A standardized laboratory method to determine the expected NFRV applicable to all kinds of residues would be useful. One proposed method is to use the carbon:nitrogen (C/N) ratio, which in pot experiments with ryegrass has been shown to display a linear relationship to NFRV (Delin et al., 2012). A similar relationship has been shown in other studies, but for a narrower range of fertilizer types (Sørensen et al., 2003; Sørensen & Fernández, 2003; Gale et al., 2006; Antil et al., 2009). Before farmers and agricultural advisors begin adopting C/N ratio to estimate NRFV, it must be tested in field experiments under different conditions. Thus this study evaluated the relationship between C/N ratio and NFRV in field experiments on spring oats.

Materials and Methods

Four oat experiments were conducted in two years (2012-2013) at two locations in Sweden, a sandy soil in Halland and a clay soil in Västergötland. The experiments had 12 treatments randomized within four blocks, which included eight organic residues commonly used as fertilizers and four different rates of mineral N fertilizer. The organic fertilizers were vinasse (by-product from yeast production), chicken manure, cattle slurry, pig slurry, two meat bone meal (MBM) products and two digestate products from biogas plants. Grain yield was measured with a combine harvester in 20 m^2 plots and grain samples were analyzed for N content. By comparing grain yield in the organic amendment treatments with crop response to mineral N fertilizer, NFRV was calculated. The NFRV obtained were plotted against C/N ratio and the curve compared with the relationship found in pot experiments.

Results and Discussion

The relationship obtained for NFRV and C/N ratio in the field experiment appeared to have a similar regression line to that obtained in the pot experiment (Figure 1). However, the relationship was unable to explain as much of the variation in NFRV in the field experiments (r^2=0.34) as in the pot experiments (r^2=0.84). Since C/N ratio did not vary significantly within each type of fertilizer, it could not always explain the variation in NFRV within a single type of fertilizer. In those cases, using mean NRFV for the fertilizer (Figure 2) would be just as good as analyzing the C/N ratio to estimate the NRFV. However, for digestates and cattle slurry there was a strong relationship between C/N ratio and NFRV. A strong relationship between NFRV and C/N ratio was also found by Sørensen et al. (2003) for cattle slurry and by Sørensen and

Fernández (2003) for pig slurry. Thus C/N ratio probably provides just as good advice concerning NRFV in slurries and digestates as ammonium content, if not better. The advantage with C/N ratio compared with ammonium content is that it can be interpreted similarly regardless of the type of manure or residue in question. For newly introduced products that have not been tested in crop experiments, the C/N ratio provides good information on what NRFV to expect.

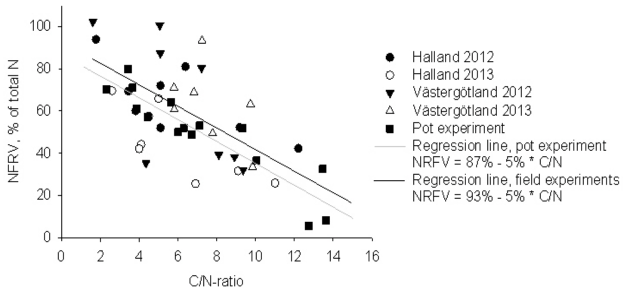

Figure 1. Nitrogen fertilizer replacement value (NFRV) as a function of carbon:nitrogen (C/N) ratio in the field experiments compared with pot experiments.

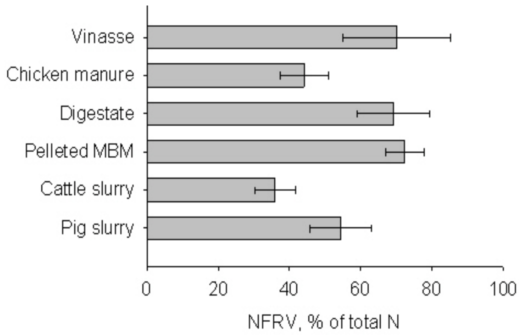

Figure 2. Average nitrogen fertilizer replacement value (NFRV) for the different types of fertilizers used in the field experiments. Error bars indicate standard error.

Conclusions

The NFRV of manure or other residues can be estimated based on C/N ratio. Since NFRV in the field is dependent on many other variables, the estimation will be rather rough, so only large differences in C/N ratio will be important.

Acknowledgements

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SOIL RECEIVES, SOIL REPORTS: SOIL BACTERIAL COMMUNITIES AS RAPPORTEURS OF INTEGRATED ECOSYSTEM RESPONSES TO INCREASED NITROGEN AVAILABILITY

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Without the use of nitrogen (N)-containing fertilizers, the human population would be approximately half of what it is. Due to the N cascade, even the N added as fertilizers and used by the plants will later affect the air, water and soil, climate and ecosystems' stability and biodiversity, which in Europe costs 70-320 billion € per year. Within the European Union, the Mediterranean Basin (a biodiversity hotspot) is the main producer of fruits and vegetables, an activity that causes N pollution and threatens the biodiversity. To understand the effects of increased N availability on a N-limited and highly biodiverse ecosystem, a unique N-manipulative experiment in Europe (N dose and form) was set up in 2007 focusing on the structural and functional changes in the of above and belowground communities [e.g.(Dias *et al.* 2012)]. However, it has been difficult to understand the links and mechanisms through which increased N availability leads to structural and functional changes. Attempting to link N-driven changes in soil processes and changes in soil microbial communities, we focused on soil bacterial communities. In particular, we aimed at bacterial groups that are involved in key processes of the N cycle such as nitrification and N fixation, hopping that they could indicate the N status of the receptor ecosystem.

Materials and Methods

The study site is in Serra da Arrábida (Portugal, 38°29' N, 9°01' W), which is within the sub-humid thermomediterranean bioclimatic domain. It belongs to the Natura 2000 network (PTCON0010 Arrábida/Espichel) and estimated ambient N deposition is < 4 kg N ha^{-1} yr^{-1}. The soil is 15-20 cm deep and has a silt-sand-loam texture (Dias *et al.* 2012). The vegetation consists of a dense maquis, established through a secondary succession after a fire event in the summer 2003. The dominant plant species was a Cistaceae, *Cistus ladanifer* L. (Dias *et al.* 2012). *Genista triacanthos* Brot. (Fabaceae) is also abundant. Control plots received no N addition, while there were three N treatments: 40A received 40 kg NH_4^+-N ha^{-1} yr^{-1} as a 1:1 mixture of NH_4Cl-N and $(NH_4)_2SO_4$-N; 40AN received 40 kg NH_4NO_3-N ha^{-1} yr^{-1}; and 80AN received 80 kg NH_4NO_3-N ha^{-1} yr^{-1}. Fertilization started in January 2007. Each treatment has three replicates (400 m^2 experimental plots). Large-scale pyrosequencing-based analysis of 16SrRNA gene sequence was applied to monitor the impact of N addition in the soil bacterial communities structure. Sampling occurred in October 2012. In each plot, soil samples were collected under the canopy of five plants of *Cistus ladanifer* and five plants of *Genista triacanthos*. Two-way ANOVA was applied to determine significant interactions between plant species and treatment.

Results and Discussion

Analysis of the rarefaction curves (97, 95 and 90% similarity) showed that 95% similarity guaranteed that a high taxonomic richness was being attained by the surveying effort. Considering all the treatments more bacteria sequences were found in the *C. ladanifer* than in the *G. triacanthos* (+10%). When comparing the number of sequences per treatment no differences were detected with the exception of *G. triacanthos* 80 AN, where a higher number of sequences was detected (+ 25%). The dominant (representing ± 75% of the sequences) taxa across all samples were *Proteobacteria*, *Acidobacteria*, *Actinobacteria*. The abundance of *Firmicutes* and TM7 showed significant effects ($p< 0.05$) of both plant species and N treatments. For other groups there was a significant effect of the plant species (*Bacteroidetes* and *Cyanobacteria*) or the N treatments (*Acidobacteria* and *Gemmatimonadetes)*. Despite the complex matrix of soil bacteria the relative abundance of some bacterial groups appears to overcome the importance of the plant species thus indicating a bigger effect of the N availability.

Concerning the bacterial groups involved in N fixation and nitrification, there were also genera that responded significantly to the plant species (*Burkholderia* b - N φιξερ), the N treatments (*Bradyrhizobium*- N φιξερ) and to both (*Nitrosospira* - nitrifier). In terms of the bacteria involved in the first step of nitrification, only *Nitrosospira* was detected and only in the N treatments. As for the second step of nitrification only *Nitrospira* was detected also only in the N treatments. We detected 9 bacterial genera capable of N fixation but they occurred irrespective of the treatments. Data suggest that the presence of some nitrifying bacteria in the soil can indicate increased N availability.

It is interesting that even in an initial phase of the work, it was possible to relate the major differences in the structure of the bacterial community with the major limiting factors to productivity: 1) *Acidobacteria* – known for their capacity to immobilize polyphosphates – decreased with increased N availability, which may be associated with an increase of the P limitation; 2) *Actinobacteria* – known as regulators of microbial activity, and 3) *Gemmatimonadetes* – known as being tolerant to dehydration tended to represent a higher percentage of soil bacteria in response to N addition.

Acknowledgements

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MAXIMIZE THE PLANT NITROGEN USE EFFICIENCY OF ANIMAL SLURRY USING AN EFFICIENT TREATMENT PRIOR SOIL APPLICATION

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Animal slurry is widely used as organic fertilizer to supply plant nutrients, namely nitrogen (N) but it is well known that slurry application to soil can lead to strong losses of N by gaseous emissions (ammonia and nitrous oxide) and/or leaching (nitrate and organic nitrogen). These N losses have a negative impact on environment and significantly affect the plant N use efficiency of animal slurry.

Slurry injection has been proposed to minimize ammonia (NH3) emissions but this technique implies high financial investment and is not easily applied in some type of soils. Furthermore, a decrease of ammonia emissions may lead in some conditions to an increase of nitrous oxide (N2O) emissions or nitrate leaching.

On the other hand, several slurry treatments (prior to soil application) such as solid-liquid separation and acidification proved to be efficient to decrease NH3 and N2O emissions after soil application. An application of the liquid fraction or acidified slurry rather than untreated slurry using adapted conventional methods could therefore be as efficient as slurry injection. Indeed, the negative impact of slurry broadcasting by splash plate can be minimized if slurry is applied using a surface banding technique.

Thus the main question to be clarified in this study is:

'Is animal slurry injection in soil more efficient to reduce the associated environmental impacts and increase the plant N use efficiency than a combined approach of slurry treatment followed by surface banding application?'

To answer this question, the following objectives were established:

a. Estimate the leaching of nitrogen (nitrate and organic N) following untreated slurry injection or surface band application of pre-treated slurry;

b. Compare the nitrogen dynamics in soil as well as the plant production in soil amended by injection of untreated slurry or by surface banding application of pre-treated slurry;

c. Compare the N2O and NH3 emissions at soil surface in soil amended by injection of untreated slurry or by surface banding application of pre-treated slurry;

An overview of the main results obtained in this study will be shown and analysed in order to propose an efficient solution to maximize the plant N use efficiency of animal slurry.

Materials and Methods

Both laboratory and field experiments were performed to achieve our objectives. Two slurries were considered, cattle and pig slurry, and 6 treatments were established in 3 different soils (sandy and 2 sandy-loam soils):

i. untreated slurry injected at 10 cm (IWS),

ii. untreated slurry applied on soil surface followed by soil mobilization (WS);

iii. liquid fraction applied to the soil surface without mobilization (LF);

iv. acidified slurry applied to the soil surface without mobilization (AWS);

v. Acidified liquid fraction applied to the soil surface without mobilization (ALF).

vi. Control (no amended soil)

The liquid fraction (LF) was obtained by mechanical separation (screw press or centrifugation) and acidification to pH 5.5 was performed by addition of concentrated sulfuric acid.

Ammonia emissions were measured using a dynamic chamber method and nitrous oxide emissions were estimated using a closed chamber system.

Leaching was measured at field scale using ceramic suction cups and soil column experiments were conducted under controlled conditions.

The plant N use efficiency of animal slurry was evaluated considering two crops: oat and maize silage. Plant production was estimated in field and pot conditions.

Results and Discussion

Results obtained with soil column experiments showed that application of treated slurry by separation or acidification may lead to an increase of NO3 leaching relative to WS application but the differences relative to IWS were not so significant. Furthermore, as expected, slurry acidification may increase salts leaching.

At field scale, our results showed that slurry injection may be replaced by surface application of acidified slurry in order to minimize the NH3 emissions. However, high NH3 emissions were observed in LF treatment. Furthermore, in the case of sandy soil, surface application of acidified slurry followed by incorporation appears as a more efficient solution than injection of non-treated slurry to minimize NO3 losses.

Crops production and N use efficiency is higher in AWS treatment relative to WS but differences between IWS and AWS treatments were low.

Lower N2O emissions were observed in IWS relative to WS whereas AWS and WS led to similar emissions.

Conclusions

Our results indicate that surface application of acidified slurry is a good alternative to slurry injection to minimize environmental impacts and increase the the plant N use efficiency of animal slurry

Acknowledgements

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EFFECT OF LIMING AND ORGANIC FERTILISATION ON SOIL CARBON SEQUESTERED IN A SILVOPASTORAL SYSTEM UNDER *PINUS RADIATA* D. DON

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Global climate change caused by rising levels of CO_2 and other greenhouse gases is currently recognized as a serious environmental issue of the twenty-first century. The establishment of silvopastoral systems, a type of agroforestry system promoted by EU through the European Rural Development Council regulation 1698/2005, has been highlighted as a strategy for soil C sequestration under the Kyoto Protocol. In silvopastoral systems, the application of lime and the fertilisation with sewage sludge could increase tree growth and pasture production as well as have a considerable influence on carbon storage in soils (Mosquera-Losada et al., 2011a). The objective of this study was to evaluate the effect of liming and two sewage sludge doses (50 and 100 kg total N ha^{-1}) on the amount of total carbon stored at four soil depths (0-25, 25-50, 50-75, and 75-100 cm) compared to control treatment (no fertilisation) in a silvopastoral system under *Pinus radiata* D. Don.

Materials and Methods

The experiment was conducted in San Breixo Forest Community (Lugo, Galicia, northwestern Spain). A plantation of *Pinus radiata* D. Don was established in 1998 at a density of 1,667 trees ha^{-1}. In October 1999, an experiment with a randomised block design was carried out in 15 experimental plots (5 treatments x 3 replicates) of 96 m^2, each consisting of 25 trees arranged in a 5 x 5 frame with a distance of 3 m between rows and 2 m between lines. Each plot was sown in autumn of 1999 with a mixture of 25 kg ha^{-1} of *Lolium perenne* var. Brigantia, 10 kg ha^{-1} of *Dactylis glomerata* var. Artabro and 4 kg ha^{-1} of *Trifolium repens* cv. Huia after ploughing. The established treatments were two sewage sludge doses based on N application (T1: 50 kg total N ha^{-1} and T2: 100 kg total N ha^{-1}), with or without liming applied in 1999 before sowing (2.5 t $CaCO_3$ ha^{-1}). A no fertilisation (NF) treatment was also established as a control in the unlimed plots. Sewage sludge fertilisation was superficially applied during 2000, 2001, 2002 and 2003. To estimate the amount of soil total carbon a composite soil sample per plot was collected to 1 m, measured for depth, and divided in the field into four subsamples corresponding to different sampling depth classes of 0-25, 25-50, 50-75 and 75-100 cm (Moreno et al., 2005) in February 2010. In the laboratory, soil C was determined by using a LECO C.N.H.S. Elemental Analyzer (LECO, St. Joseph, MI) (Kowalenko, 2001). In order to facilitate comparison with most soils from the literature, the mean bulk density of soil at each sampling depth was used to convert the soil C concentration to Mg C ha^{-1}. The data were analysed using ANOVA (proc glm procedure). Means were separated by using LSD test, if ANOVA was significant (SAS, 2001).

Results and Discussion

In this experiment, the ANOVA analysis showed that in all treatments the soil total carbon was significantly higher in the upper soil layers than at lower depths ($p<0.001$). However, it was not clearly observed a treatment effect on this soil variable ($p>0.05$)

(Fig. 1). The predominance of soil organic carbon in upper horizons is consistent with worldwide trends and can be explained by the vertical carbon gradient resulting from surficial litter deposition but also by differences in root distribution. Several authors, including Mosquera-Losada et al. (2011a), have shown that fine roots located in the upper few centimetres of soil are the main source of organic matter within a soil carbon pool. However, it is also important to be aware of the amount of carbon stored in deeper soil layers, which may reflect inputs from tree roots (Howlett et al., 2011). Indeed, the role of deep root systems in the storage of carbon in deeper soil layers is one of the main premises on which the carbon sequestration potential of agroforestry is based compared with traditional agricultural systems (Howlett et al., 2011; Mosquera-Losada et al., 2011a). Finally, the soil total carbon was not modified by the treatments applied probably due to several factors. Firstly the inputs of organic matter were high in all treatments which could mask the effect different treatments applied, secondly the residual effect of treatments could have been reduced and finally the low soil pH (water soil pH: 4.28) which could have limited the incorporation of liming and sewage sludge to the soil as observed previously Mosquera-Losada et al. (2011b) in the same study when the nutrient availability for pasture and the trees were evaluated.

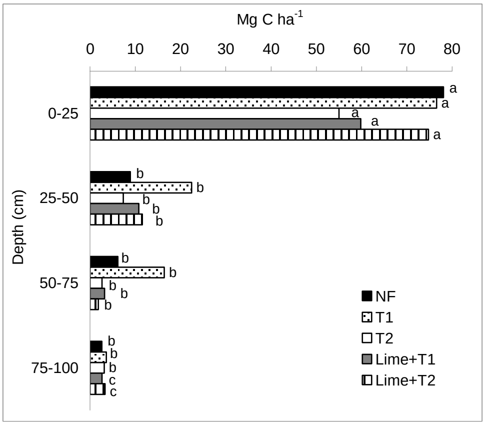

Fig 1. Total carbon storage in the soil expressed as Mg ha^{-1} under each treatment at four soil depths (0-25, 25-50, 50-75 and 75-100 cm). NF: no fertiliser; T1: low sewage sludge dose (50 kg total N ha^{-1}); T2: high sewage sludge dose (100 kg total N ha^{-1}). Different letters indicate significant differences between soil depths within the same treatment.

Conclusion

Initial liming or fertilisation inputs in silvopastoral systems established in acid soils did not modify the carbon stored in the soil ten years after establishment probably due to the soil acidity, the time elapsed since treatments applications and the high inputs of pinwood..

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AMMONIA AND ODOUR EMISSIONS FOLLOWING DIGESTATE AND UREA FERTILISATION IN MAIZE

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The interest in anaerobic digestion has recently increased both because biogas is an extremely useful source of renewable energy, whilst digestate is a highly valuable fertiliser. In 2011 a three-years project funded by Regione Lombardia, aiming to investigate the agronomic potentiality of the digestate derived from anaerobic digestion of pig and dairy slurry, started. Digestate was compared with urea to assess its effectiveness in sustaining maize growth and reducing odour and ammonia (NH_3) emissions during field application. In order to assess the performance of the fertilisers, different application techniques were tested both at sowing and at V6 stage of maize. Two commercial farms producing digestate from a 1 MW biogas plants fed by pig and dairy slurry and biomass, located in Lombardy region (northern Italy), were selected to carry out the study. Here preliminary results are presented; they referred to the field trial established in a dairy farm in spring 2012 at maize sowing.

Materials and Methods

The experimental trial was performed in a 5 ha field considering the following treatments: i) unfertilised check plot (T1), ii) digestate with surface spreading (T2), iii) urea (T3) and, iv) a digestate incorporated into the soil (T4). Twenty-four hours after the fertilisation, all the field area was ploughed at 30 cm depth. Fertilised treatments were designed to receive similar amounts of nitrogen, namely 130 kg N ha^{-1}. Treated plots were organised in randomised block design with each treatment replicated two times in each block. The plots were previously characterised by selected chemical and physical soil properties in the 0 to 30 cm layer, subsequently monitored during maize growing season. The digestate was previously analysed for the main composition parameters. The measurements of NH_3 concentrations were performed using long term exposure samplers ALPHA [Tang et al. (2001)]. Samplers were exposed in three replicates at 50 cm height and placed in the centre of each plot to measure the NH_3 concentration from the fertilisers application. To measure the background level of NH_3 concentration (BKG), a sampling point was located away from the field and from any other known source of NH_3. The samplers were changed between 2 h during the spreading and incorporation days, and two times per day, after dawn and just before sunset, in order to follow the change of atmospheric turbulence. To evaluate the atmospheric turbulence, responsible for the gas dispersion, a three-dimensional ultrasonic anemometer sampling at 10 Hz (USA-1, METEK GmbH, Elmshorn, Germany), was placed in the field at height of 1.25 m. The flux of NH_3 emitted from the fertilised surface was finally determined by the use of inverse dispersion modelling (Carozzi et al., 2013). From each plot the gaseous samples for odour detection were collected using a flux chamber system based on the APAT method (APAT, 2003). A Plexiglass chamber having a surface of 0.196 m^2 was continuously flushed for 10 min with air. Output gas from the chamber was taken from the outlet port and stored in Nalophan bag with 80 liters volume to be used for olfactometric analysis (standardised

EN method n. 13725). An Olfaktomat-N 6 (six stations) olfactometer (PRA-Odournet B.V., Amsterdam, NL) based on the forced choice method was used as a dilution device and six panellists were employed during test (Orzi et al., 2010).

Results and Discussion

The digestate: 3.4 g N kg^{-1} of TKN, 2.0 g N $kg^{-1}$of TAN, TAN/TKN= 59%, DM= 7.4% and ph=8.10. Concentration peaks of NH_3 (µg m^{-3}) were reached immediately after the distribution of fertilisers: in the first three hours for T2 and T4 and after 12 h for T3 (Fig 1). After the first 18-24 h concentration gradually decreased reaching the BKG concentration, as a consequence of the deep incorporation by ploughing occurred in the day after the fertilisation. Surface spreading of digestate (T2) had the maximum NH_3 concentration value which was seven times higher than its contextual incorporation (T4). Urea showed its higher concentration level at 12 h after the application, highlighting the slowness of the hydrolysis process (Fig1). Values of odour emission rates (Tab 1) were well distinguished between treated and untreated plots, the surface spreading of digestate treatment determining the highest human odorous response.

Figure 1 – NH_3 concentrations measured in the field plots and in its background value from the time of application. To simplify the readers, treatment T1, standard deviation between the two blocks and the last five monitoring days are not reported.

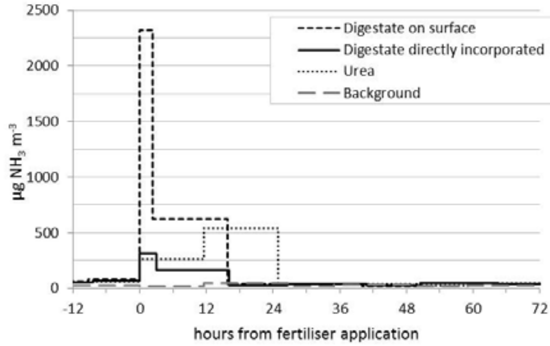

Table 1 - Mean NH3 concentrations and odour emission rate among treatments.

Treatment	Mean NH_3 concentration (µg m^{-3})	Odour emission rate (UO m^{-2}soil h^{-1})
BKG	27	310
T2	403	3318
T3	131	2893
T4	85	1645

Conclusions

While surface application of digestate resulted in high NH_3 concentration levels, its contextually incorporation is an efficient method to control and prevent the volatilisation process and, consequently, to increase the availability of N for crop production. The incorporation technique also caused lower NH_3 release compared to urea. According to the preliminary results, the treatment designed to reduce ammonia volatilisation would reduce also odour emissions.

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INTERACTIONS BETWEEN FREE-LIVING NEMATODES AND MICROBES IN ORGANICALLY AMENDED SOILS: EFFECTS ON N MINERALIZATION

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Free living soil nematodes, one of the most abundant and functionally diverse groups of soil fauna, have been shown to significantly contribute to N mineralization mainly by feeding on the primary decomposers (bacteria and fungi). The contribution of these nematodes to the decomposition of organic residue depends on the biochemical characteristics of the organic material such as the C:N ratio. However, most of the experiments that tested the effects of nematode grazing on N mineralization used simple organic amendments such as glucose, or dried alfalfa. Moreover, despite their abundance and diversity only few species of nematodes have been used in these studies. In order to examine how interactions between the entire free living nematode communities and microbes affect N mineralization in organically amended soil, we conducted an incubation experiment by applying different organic materials spanning a range of C:N ratios and presumed N availability.

Materials and Methods

Bulk soil was collected from an agricultural field and part of it was gamma irradiated with a 5 kGy dose, that selectively kills nematodes while leaving the microflora largely intact (Buchan et al., 2012). All free-living nematodes were extracted from unirradiated bulk soil using zonal centrifuge and reinoculated into an equivalent amount of irradiated soil. Three treatments: CTR (control sieved bulk soil), DEFN (defaunated with 5 kGy dose) and REIN (defaunated and reinoculated with nematodes) were compared. Cores were either left unamended (UNA) or received lignin-rich low N compost (COI), N-rich compost (COV), fresh manure (MAN) or chopped clover (CLO), all added at the same rate of 277 μg N g^{-1} dry soil which was equivalent to 126 kg N ha^{-1}. Cores were incubated at $18^{\circ}C$ at constant and optimal water content (50% WFPS) for three months and sampled destructively after 7, 21, 40, 68 and 97 days. Mineral N, microbial biomass carbon (C_{mic}) and nematode abundances were determined at every sampling time, while PLFA measurement and nematode identification were conducted at the first and last sampling times. The effect of nematode reinoculation on N mineralization was calculated by subtracting the mean total mineral N concentrations at time *t* in the DEFN treatment from the equivalent value in the REIN treatment for each amendment separately. Data was analysed using Two way ANOVA model (time and treatment as fixed factors).

Results and Discussion

Nematode reinoculation was successful in establishing viable nematode populations that resembled those of CTR cores in abundance and composition. Given the only difference between the DEFN and REIN treatments was addition of filtered nematodes in distilled water, it was assumed that all differences in mineralization dynamics are a result of nematode activity (feeding on primary decomposers and predation). Nematode reinoculation generally increased the amount of N mineralized from the amendment, the maximal mineralization being greater and occurring earlier for amendments with high bioavailable N (CLO and COV) (Fig 1a). Nematode reinoculation also clearly stimulated nitrification in all amendments (Fig 1b). Results strongly support the hypothesis that increased nitrification was due to bacterial (nitrifiers) stimulation rather than an increased supply of NH_4^+, as in DEFN cores NH_4^+ was in plentiful supply but no increased nitrification occurred. C_{mic} in REIN was lower than DEFN in the unamended treatment and all amendments except MAN, indicating nematode reinoculation decreased C_{mic}. as a result of (over) grazing by microbivorous nematodes. Gamma irradiation increased NH_4^+ which is preferentially used by microbes over NO_3^- and organic molecules (Geisseler et al. 2010). Given the high NH_4^+ concentration (less nitrification) in DEFN cores, it could be expected that microbes made full use of NH_4^+, which could explain the higher C_{mic} in DEFN compared to REIN.

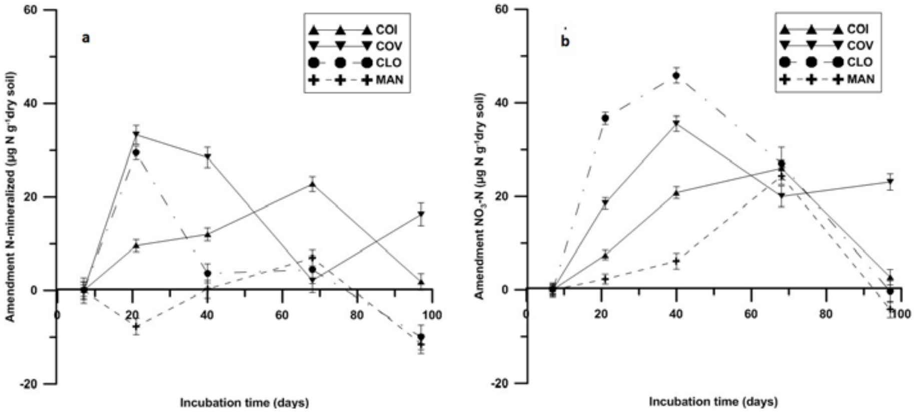

Fig 1. Amount of amendment-N mineralized (a) and Nitrification (b) as affected by reinoculation

PLFA data showed that all microbial taxa were more abundant in amended cores than in UNA cores, indicating the range of amendments used in this experiment stimulated a wide range of microbial groups. The B:F ratio was significantly lower in CLO and MAN showing that these N rich amendments preferentially stimulated fungi over bacteria compared to other treatments, in contrast to the previous findings and expected trends (Fostegard and Baath, 1996). Analogous to the PLFA data, CLO had the highest abundances of both bacterivores and fungivores nematodes. Abundances of bacterivores but also fungivores and omnivores were positively correlated to total mineral nitrogen concentrations.

Conclusion

Nematodes exert an important influence on N cycling, particularly on mineralization and nitrification as they interact with microbes. Contribution of these nematodes to the decomposition of organic amendments in this agricultural soil seems to be most strongly and rapidly expressed when the availability of the added organic N source is high. High bio-availability of N in clover positively stimulated both bacterial and fungal decomposition channels more than manure or compost with the same N content.

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STABILIZATION AND LOSSES OF N IN SOIL AMENDED WITH 15N-LABELED RESIDUES COMBINED WITH AN ORGANIC N FERTILIZER

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In agricultural systems, crop residues (CRs) incorporated into the soil can be an important source of nutrients useful to maintain soil fertility. Both CRs decomposition and nutrients release are affected by the chemical characteristics defining residue quality, mainly N content and amount of recalcitrant C compounds. The C:N ratio is the most commonly used index to predict residue decomposition and N release but this alone might not be sufficient since the presence of recalcitrant C may reduce these processes (Sakala et al., 2000). The addition of a mineral or organic N fertilizer can alter these processes, with high quality CRs the influence on decomposition is minimal because N is not limiting for microbial activity, whereas, with low quality CRs, N may increase microbial decomposition. The N released, if not taken up by plants, can be lost through denitrification or leaching, or stabilized in soil aggregates where OM is physically or chemically protected. In this study the decomposition of two 15N labelled cereals (barley and triticale) managed in Italy as CRs, with or without an organic N fertilizer, was followed over one year. The amount of residue N lost from soil and that recovered and stabilized in soil aggregates at the end of the year was quantified through the isotopic approach.

Materials and Methods

The study was carried out at the experimental farm of Cadriano, (Bologna University) for one year. Plastic tubes (10 cm diameter, 15 cm height), closed at the bottom with a metallic net, were filled with 400 g of soil (dry basis) and buried. The soil characteristics were pH 7.1, SOC 12 g kg^{-1}, TN 1.2 g kg^{-1}. The experimental design included a control soil (S), soil with barley residues (S+B), soil with barley and leather (S+B+L), soil with triticale (S+T), soil with triticale and leather (S+T+L). Barley (cv. Tidone) and triticale (cv. Oceania) were grown in the greenhouse and fertilized with 15N-labelled solution. At the barrel stage plants were harvested, leaves and stem cut into 1-2 cm sized pieces and evenly mixed. Residues and leather were added to soil at equivalent rates of 10 t ha^{-1} fresh material and 50 kg N ha^{-1} respectively. The tubes were arranged in a completely randomized block design with 3 replicates per treatment and 5 destructive samplings, 1, 3, 6, 9 and 12 months. At sampling the residues were recovered from soil, dried at 60°C and weighed to calculate the mass loss. Then they were grinded and analyzed for their C and N content and 15N abundance (CF-IRMS Delta Plus Thermo Scientific). At the last sampling soil was air dried, an aliquot was analyzed for C, N and 15N abundance, another (5 g) was physically fractionated by sieving in water (Elliott, 1986). Three aggregate size fractions were recovered: >250 μm (particulate organic matter POM); 53-250 μm (intra-aggregate light fraction, IALF); < 53 μm (mineral fraction, MF), characterized by increasing protection of SOM against microbial degradation: POM not protected OM; IALF physically protected OM, MF chemically protected OM. Once dried, the fractions were weighed and analyzed for C, N and 15N abundance. The residues derived N (NDFR) in soil and aggregates was calculated as follows: NDFR % = 15N atom excess soil / 15N atom excess residues * 100. The portion of N added with residues and recovered in soil and fractions was also calculated. Data were analysed using analysis of variance with three factors: time (5 levels); residues (2 levels) leather N (2 levels). Statistically significance ($P \leq 0.05$) differences between means were separated by Student Newman Keuls test. When interaction between time, residues and leather N was significant, twice standard error of means was used as the minimum difference between two statistically different means.

Results and discussion

Barley and triticale showed a very different degradation pattern and N release. About 77% of barley was degraded after one year, and most of the C and N losses were observed during the first month. This behaviour was consistent with the C:N ratio (24) of this residue. Triticale degradation was slow during nine months, then increased during the last three months, with a final mass loss of about 55%. The triticale behaviour was probably related to a higher C:N ratio (29) and the presence of more recalcitrant C components. The addition of leather, as a source of energy and N, partly reduced the barley degradation (72%) and significantly speeded that of triticale (64%). After one year however, similar NDFR were measured in all soils, and the recovered portion of residues N ranged between 56% (S+B) and 64% (S+T). The soil fractions showed similar NDFR values but, due to their different distribution in soil (POM >IALF>>MF) greater amount of residues N was in POM and IALF than in the MF. The presence of leather reduced the residues N stabilized in the finest fractions IALF and MF. The amount of residues N not found in soil, was supposed to be lost by leaching since the environmental conditions were not favourable to denitrification. These losses ranged between 27.6% of the N applied in S+B to 17.5% in S+B+L and 18.5% in S+T+L, whereas no losses were measured in S+T.

Conclusions

The different residues degradation and the effect of leather on this process affected the leaching losses of residues N. However, more than half of the N added was still in soil after one year. When residues were added alone about half of this N was stabilized in IALF and MF physically or chemically protected, whereas the presence of leather reduced this amount to about 30%.

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EFFECT OF DRYING TEMPERATURE ON NITROGEN MINERALISATION AND SOIL N2O EMISSIONS FOLLOWING ADDITION OF THERMALLY-DRIED SLUDGES

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Sewage sludge has long been used as an agricultural amendment (Singh et al., 2011). Thermal drying is a sludge processing technique that is used in a number of waste water treatment plants (WWTP) (Rigby et al., 2010). There are a number of potential advantages and disadvantages attributed to thermally-drying sludge compared to other sludge treatment processes (Table 1).

Table 1. Advantages and disadvantages of thermally-drying sludge

Advantages	**Disadvantages**
Drying results in a class A biosolid product	The economic and energetic costs of drying sludge can be high
Drying produces a biosolid product more physically, chemically and biologically stable than un-dried sludge	Thermal drying results in a loss of nutrients, particularly ammonium
Transportation costs for dried sludge are low due to high dry matter content (>85%)	The drying process generates dust

Sludge can be dried at a wide range of temperatures, with inlet temperatures from 65°C up to 480°C (Sullivan, 2012), but in practice is generally dried at temperatures under 200°C. Drying can have a significant effect on N contents of sludge. Sludge loses up to 80% of its ammonium content as ammonia (NH_3) via volatilisation during the drying process (Smith and Durham, 2002). Despite this loss, a greater percentage of N from thermally-dried sludges has been found to be available to soil compared with raw or anaerobically-digested sludges (Rigby et al., 2010; Silva-Leal et al., 2013; Smith and Durham, 2002; Tarrasón et al., 2008). Therefore thermally-dried sludges may be equally or more suitable as an N-based fertiliser for agriculture than un-dried sludges. However, the effect of different drying temperatures on the N mineralisation of dried sludge is unknown. Dried sludges may also affect soil greenhouse gas emissions such as nitrous oxide (N_2O). However, this has not been considered in previous dried-sludge application studies. Finally, the effect of organic fertiliser amendment on soil N cycling is likely to vary with soil water content. We conducted a laboratory incubation study with the aims to investigate whether thermally-drying sludge at several different temperatures (70, 130, 190, and 250°C) can influence the cycling of N via mineralisation or soil N_2O emissions at two water contents (pF 2 and pF 1). Our research objectives were to investigate i) the effect of a range of drying temperatures on dried sludge addition to soil and resulting N mineralisation and N_2O emissions and ii) whether the effect is the same at field capacity (pF 2) and at high water content (pF 1).

Materials and Methods

Soil was collected from an NPK-treated plot from the CRUCIAL field trial at Taastrup, Denmark (Poulsen et al., 2013). Anaerobically-digested sludge and in-house dried sludge was sourced from a WWTP in Randers municipality, Denmark. The WWTP dried sludge was dried on a belt drier at a temperature between 95 and 200°C. The lab-dried sludges were dried in a laboratory oven at the different temperatures (70, 130, 190 and 250°C) until the water content reached < 5%. Samples of 45g soil were pre-incubated at 15°C and pF 2 in the dark for two weeks before the sludges were added to soil at a 2% rate (dw). Water contents were then adjusted to pF 2 or pF 1. Inorganic N (IN) from soil and sludge was extracted with 1 M KCl and analysed with by flow injection analysis. Soil nitrous oxide emissions were analysed using the static chamber method over a 90 minute period. The percentage of N mineralisation was determined after 160 days.

Results and discussion

Drying temperature had a significant effect on ammonium contents within the sludge (Figure 1). Preliminary results indicated that there were significant differences in IN and pH content in soil after the application of different treatments. Inorganic N in soil decreased with the application of lab-dried sludges at higher temperature. While N_2O emissions were significantly higher in soil amended with lab-dried sludges at low temperature (70 and 130°C).

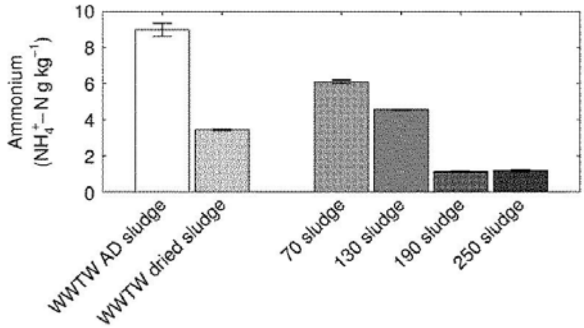

Figure 1. Ammonium contents within anaerobically-digested sludges: un-dried (WWTP AD sludge), dried in-situ via a belt-drying process (WWTP dried sludge), dried in the laboratory at 70, 130, 190 and 250°C. All of the treatments were statistically significant from each other ($p< 0.01$) except the 190 and 250 sludge ($p>0.05$).

Conclusion

Thermal drying of sludge at different temperatures appears to affect chemical properties of the end product, and we have indications that this affects the N mineralisation rate and N_2O emissions following addition to soil.

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THE SENSITIVITY OF AMMONIA OXIDATION TO SOIL AMMONIUM CONCENTRATION AS AN INDICATOR OF ECOSYSTEM N SATURATION

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As the human population continues to increase, so do the global impacts of release of reactive nitrogen (except di-nitrogen – N_2) into the biosphere. Since reactive N (Nr) is a limiting factor to life, its increased availability will impact all compartments of the receptor ecosystems. Soil microorganisms are among the most sensitive elements of ecosystems and are key participants in recycling of nutrients, in particular N. Therefore the assessment of microbial activities provides early signs of disturbance and is used to monitor soil quality and health (Nielsen and Winding, 2002). In fact, increased nitrification rates are usually used as an indicator of increased N availability. Nitrification is a 2-step process: 1) ammonia oxidation to nitrite; and 2) nitrite oxidation to nitrate (Norton and Stark, 2011). Ammonia can be oxidised by ammonia oxidizing bacteria (AOB), which are among the most responsive groups to increased Nr availability (Allison *et al.* 2008). Our objective was to evaluate the impact of increased N availability (form and dose) on the structure and functioning of the AOB community of a Natura 2000 site belonging to a Mediterranean ecosystem.

Materials and Methods

The present study was conducted in the Arrábida Natural Park, a Natura 2000 site south of Lisbon, Portugal (PTCON0010 Arrábida/Espichel). N availability was modified by the addition of 40 and 80 kg N ha^{-1} yr^{-1} in the form of NH_4NO_3 (40AN and 80AN, respectively) and 40 kg N ha^{-1} yr^{-1} as a 1:1 mixture of NH_4Cl-N and $(NH_4)_2SO_4$-N (40A). Each treatment was replicated in three 400 m^2 plots (Dias *et al.* 2011). Soil sampling was performed in October 2012, after 5 years of Nr applications. A composite sample was obtained for each plot, 3 samples per treatment. Three sequential enrichment cultures were prepared to allow a subsequent increase of the AOB population, using soil as the initial inoculum. AOB susceptibility to ammonia was studied by testing several ammonia concentrations in the medium (1, 10, 50, 100 and 500 mM of NH_4Cl-N). Growth was indirectly estimated by the nitrite accumulated along time (Griess method). DNA was extracted from the 3rd enrichment of AOB community cultures derived from soil samples and the *amoA* gene amplified. Cultures were considered active when there was accumulation of nitrite in the culture medium and amplification of the amoA gene.

Results and Discussion

AOB populations were not detected in all treatment replicates, indicating that they were not very abundant, although there was a tendency for AOB to be more abundant in the plot which received the 40A treatment (Fig. 1). Similarly, cultures from 40A and 80AN treatments cultures showed the highest nitrite accumulation rates, while those from 40AN had the lowest (Fig. 2 a). The very similar activities of 40A and 80AN cultures suggest that AOB community responds more to the quantity of Nr added to the soil as ammonia than to the total quantity of Nr added. On the other hand, the lower activity of 40AN culture, may reflect the competition between plants and AOB for ammonia (Verhagen *et al.* 1994). This is supported by the N-driven increase in plant growth observed in the 40AN field plots (Dias *et al.* 2011). Increasing the ammonium concentration of the growing medium from 1 to 10 mM stimulated the nitrite accumulation rate of all cultures and treatments. Further increasing the ammonium concentration to 100 mM only resulted in a complete inhibition of nitrite accumulation by the cultures from the control plots and a slight inhibition of nitrite accumulation by the cultures from N treated plots. Full inhibition of ammonia oxidation was observed at 500 mM ammonium (Fig. 2 b). Given that many AOB populations grow and are active at ammonium concentrations as high as 1000 mM (Hatzenpichler, 2012), these results indicate that the AOB populations found at the experimental site are more sensitive to ammonium concentrations than most of those described in the bibliography, indicating the potential of inhibition kinetics of ammonium oxidation by the substrate (ammonia) to indicate the degree of N saturation of the ecosystem.

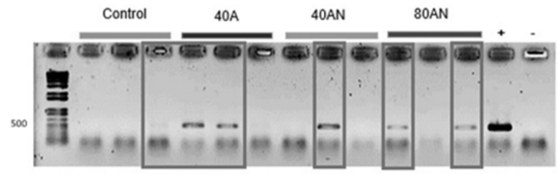
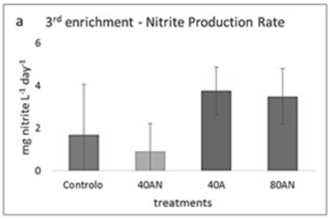


Fig. 1 *amoA* gene amplification for the 3rd enrichment AOB community cultures. Control AOB communities (blue); 40A treatment (red); 40AN treatment (green); 80AN treatment (purple). Positive control (+) and negative control (-). Samples outlined by green had positive amplification for *amoA*.



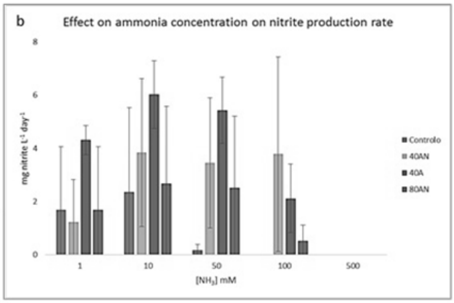


Fig. 2 Nitrite production rate for non-fertilized (control - blue) and fertilized soils AOB cultures (40A - red, 40AN - green, 80AN - purpe): a) in the 3rd enrichment; b) with different ammonia concentration in the medium (1 mM, 10 mM, 50 mM, 100 mM and 500 mM). Standard deviation is represented as error bars.

Conclusions

N-additions can alter the activity, abundance and population structure of AOB. AOB populations seem to respond to the ammonium availability and not to total N. The inhibition kinetics of AOB by the substrate (ammonia) may be a good descriptor of the degree of N saturation of the ecosystem

Acknowledgements

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BIOCHEMICAL VARIATIONS IN PLANT RESIDUES (OILSEED RAPE AND BARLEY STRAW) AFFECT N2O EMISSIONS AND ORGANIC MATTER DECOMPOSITION RATE

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Organic residues in soils are decomposed by soil microorganisms and residual nitrogen is nitrified to NO_3. Subsequently NO_3 can be denitrified to NO, N_2O or N_2. N_2O is formed in the processes of nitrification and denitrification. Nitrous oxide (N_2O) is one of the most important permanent greenhouse gases. Crop residues, cover crops or green manure can be important sources and/or drivers of N_2O emissions from agricultural soils (IPCC 2007). For example, denitrification strongly depends on the presence of available carbon (C). In this context, soil amendment with organic residues that contain readily-decomposable organic carbon compounds may trigger denitrification by enhancing O_2 consumption (thus contributing to the creation of anoxic micro-sites) and by providing energy for denitrifiers (Senbayram et al. 2009). However the bio-chemical properties of the organic residues vary significantly (e.g. C/N ratio, labile C content) and we still need further understanding how such variations affect N_2O release. The aim of this work was to determine to what extent contrasting biochemical compositions of soil-incorporated plant residues (oilseed rape and barley) affect N_2O release and the decomposition rates of these residues.

Materials and methods

Soil was collected in spring 2013 from the upper 15 cm soil horizon of an unfertilized field site. Soil samples (clay 25 %, silt 65.5 %, sand 9.5 %) were carefully air dried to allow sieving. Complete drying out was avoided to minimize mineralization after rewetting. A soil incubation experiment was carried out in a fully automated continuous flow incubation system. Briefly, oilseed rape or barley straw was mixed with soil prior to the experiment at a rate of 1.5 g DM kg^{-1} soil DM. Then 4.8 kg soil was repacked into each incubation vessel including control soils to a final density of 0.96 g soil DM cm^{-3}. Prior to the experiment, 1.2 gram of N applied to the soil surface in the form of ammonium-nitrate in the respective treatments following rewetting of soil to ca. 80 % water holding capacity (WHC) by carefully dripping dist. water on the soil surface. Overall, there were five soil treatments including non-treated control (**CK**), barley straw incorporation only (**BST**), oilseed rape straw incorporation only (**RST**), barley straw+N (**BST+N**), or oilseed rape straw+N (**RST+N**), all carried out in three replications. In addition, the entire set of treatments was replicated in a parallel microcosm system for soil sampling without online gas sampling. For online trace gas concentration analysis of N_2O and CO_2, a gas sample from each vessel outlet was directed to a Varian gas chromatograph (450-GC, Varian B.V., Middelburg, The Netherlands). The gas sample was then analyzed by the GC deploying a thermal conductivity detector (TCD) for CO_2, and an electron capture detector (ECD) for N_2O.

Results and discussion

During the first 10 days of the experiment, the soil NH_4 content decreased gradually in all N treated soils indicating significant nitrification. 25 days after onset of treatments, soil NH_4 contents in BST+N and RST+N were close to background levels. Soil NO_3 and NH_4 contents in CK, BST and RST were low and remained under 0.01 mg N g^{-1} soil DM. CO_2 fluxes in CK were significantly lower than in all other treatments. Among treated soils, cumulative CO_2 fluxes were highest in BST and RST and N addition decreased cumulative CO_2 fluxes significantly. However, the decrease in cumulative CO_2 fluxes was more pronounced in barley straw treated soils than in rape straw treated soils. Fluxes and cumulative N_2O emissions in non-fertilized control soils were low and were significantly lower than with all other treatments. Expectedly, addition of organic residues to soil caused higher N_2O emissions than in the CK, and the time course of this stimulation was almost identical in BST and RST. Additionally, cumulative N_2O emissions were also significantly higher in RST than BST. Addition of mineral-N to both organic residue amended soils increased N_2O emissions drastically. Cumulative N_2O emissions were 16 and 26 fold higher in RST+N and BST+N than in RST and BST respectively.

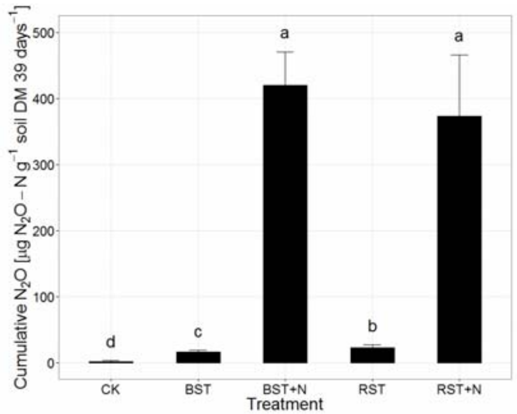

Figure 1. 39-day cumulative N_2O emissions from control soil, or soils amended with barley straw (BST), BST plus mineral N (ammonium nitrate), rape straw (RST) or RST plus mineral N. Bars labeled with the same letters are not significantly different ($p < 0.05$). Error bars represent the standard error.

Not only the decomposition rate but also N_2O emissions were significantly affected by variations in the biochemical properties of incorporated plant residues into soil. Here, rape straw clearly had a higher potential to promote N_2O emissions under N limiting conditions. Higher N_2O fluxes in RST than BST may partly be attributed to the lower C/N ratio of RST (46.61) as compared to BST (56.25). In this, our study confirms findings of Velthof et al. (2002) who reported a negative relation between the C/N ratio of straw and N_2O. With addition of mineral N to soil, there was no significant variation in N_2O emissions among organic residues.

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NITROUS OXIDE EMISSIONS FROM AN ORGANIC CROPPING SYSTEM AS AFFECTED BY LEGUME-BASED CATCH CROPS

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Legume-based catch crops (LBCC) may act as an important N source in organic crop rotations because of biological N fixation from the atmosphere in addition to soil N uptake. However, compared to non-LBCC there is a risk for higher nitrous oxide (N_2O) emissions in connection with harvest or spring incorporation which needs to be quantified. Here, we report the results from a one-year field study on a loamy sand soil in Denmark. The objective was to investigate N_2O emissions as affected by different LBCC compared with non-LBCC. The effect of two management strategies was also tested: early removal of the tops (*e.g.*, for use in biogas production or animal feeding) versus incorporation of whole-crop residues by spring ploughing.

Materials and Methods

The factorial experimental design comprised two factors arranged in three replicated blocks, leading to 36 plots (3 × 20 m in size). The catch crop treatment included three LBCC (red clover [CL], red clover/ryegrass [GC] and winter vetch [WV]), two non-LBCC (ryegrass [GR] and fodder radish [FR]) and a fallow control without catch crops [CO]. All catch crop treatments were either harvested on 30 Oct 2012 [H] or left untouched until spring ploughing [U]. Glyphosate was used to maintain a bare soil in the CO_H treatment, whilst the CO_U treatment was left for volunteer weeds. After mouldboard ploughing (ca. 20 cm depth) on 22 Apr 2013, all plots were sown to spring barley, and effects of catch crops on crop N uptake were quantified. There was no manure or fertilizer application during the monitoring period. The N_2O fluxes were measured biweekly from 10 Sep 2012 to 10 Sep 2013 using static chambers, with lower frequency in winter, and higher after spring ploughing. Five gas samples were taken during 1 h; N_2O fluxes were estimated by HMR (Pedersen *et al*., 2010). Soil water and mineral N were determined on selected gas sampling dates. Soil temperature at 5 cm depth was measured around the gas collars in block 1 during each gas sampling campaign. The aboveground biomass of catch crops was sampled in two 0.5 m^2 subplots on 25 Oct 2012, before the first frost. Catch crop root biomass was sampled to 20 cm depth in a 0.0875 m^2 subplot from all H treatments. The spring barley grain yield was determined in a central area of each plot by a combine harvester on 21 Aug 2013, and the dry matter yield was measured by hand-cutting in a 0.5 m^2 subplot one week earlier. Dry matter and N accumulation of all the plant samples was determined subsequently. Linear interpolation of the daily N_2O fluxes across the sampling dates was used to calculate the cumulative emissions (Dobbie *et al*., 1999).

Results and Discussion

LBCC accumulated significantly more N than the non-LBCC in their tops ($p< 0.05$, Table 1) with higher N concentrations in late autumn 2012. It shows that LBCC can have a substantial N input to organically managed systems. Roots at 0-20 cm represented more than 40% of the biomass and 33% of the N in catch crops. High N_2O emissions generally occurred after tillage in spring in all treatments. However, the overall highest emission was observed after early harvest of FR in winter, 130 μg N_2O-N m^{-2} h^{-1}, leading to the highest annual emission and emission intensity on the basis of both DM and grain yield of spring barley (Table 2). This may be partly explained by the fact that FR is not a winter-hardy species and that its residues after harvest was decomposed fast during the freezing and thawing event in winter. Compared with non-LBCC, using LBCC does not necessarily emit more N_2O annually.

Table 1 Dry matter and N accumulation of catch crop tops (n=6) and roots (0-20 cm, n=3) determined in late autumn 2012. Different letters indicate significant differences ($p< 0.05$) within each column.

		Dry matter (Mg ha^{-1})		Total N (kg ha^{-1})	
		Top	Root	Top	Root
	Red clover	1.9 a	1.4 a	66 a	41 a
LBCC	Grass/clover	1.9 a	1.2 a	59 a	32 a
	Winter vetch	1.7 ab	1.2 a	67 a	32 a
Non-	Fodder radish	1.7 ab	1.3 a	40 b	26 a
LBCC	Ryegrass	1.3 b	1.3 a	32 b	23 a
Weeds	Control	1.4 ab		31 b	

Table 2. Aboveground dry matter production, grain yield and grain N yield of subsequent spring barley determined at harvest in Aug 2013, the annual N_2O emissions from 10 Sep 2012 to 10 Sep 2013 and the emission intensity. Different letters indicate significant differences ($p< 0.05$) within each column, n=3.

		Dry matter (Mg ha^{-1})	Grain yield (Mg ha^{-1})	Grain N yield (kg N ha^{-1})	Annual emissions (g N ha^{-1})	Emission intensity (g N Mg^{-1} grain)
Red clover	H	9.5 ab	4.0 a	61 a	790 bc	197 c
	U	9.6 ab	4.5 a	72 a	841 bc	188 c
Grass/clover	H	8.3 ac	4.0 a	61 a	815 bc	204 c
	U	10.2 a	4.5 a	70 a	835 bc	189 c
Winter vetch	H	6.2 de	3.3 b	46 b	673 bc	199 c
	U	7.8 bcd	4.1 a	65 a	691 bc	168 c
Fodder radish	H	6.2 de	2.7 bce	34 bc	1714 a	635 a
	U	6.5 ce	3.3 bc	43 bc	1180 ab	363 b
Ryegrass	H	5.3 e	2.8 d	37 c	527 c	188 bc
	U	6.0 de	2.6 cd	35 bc	509 c	202 c
Control	H	5.8 de	2.7 de	34 c	623 bc	235 bc
	U	6.3 ce	2.8 bd	34 bc	801 bc	297 bc

The spring barley following LBCC treatments shows significantly higher grain and grain N yields, which tend to be negatively affected by early removal of the catch crop tops in late autumn (Table 2). The data of soil mineral N dynamics will be available later to support the analysis of the emission data.

Conclusions

The use of LBCC (*i.e.*, CL, GC and WV) on a temperate loamy sand soil enhanced the performance of the subsequent spring barley with generally low annual N_2O emissions or yield-scaled emissions in an organically cultivated system. However, harvest of the catch crops may reduce the yield of the following main crop, unless the harvested N is recycled to the field in manure of digested biomass.

Acknowledgements

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DISTRIBUTION OF NITROGEN, PHOSPHORUS, POTASSIUM AND CARBON BETWEEN MANURES IN FUNCTION OF BARN CATEGORIES

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Fate of nitrogen (N), phosphorus (P), potassium (K) and carbon (C) in agriculture is a major concern for fertility or environmental reasons. In Wallonia (Belgium), different types of barns for cattle are recognized (table 1) leading to production of 4 manure types (Pain and Menzi, 2011): slurry (SL), liquid fraction (LF), farm yard manure (FY) and semi-solid manure (SSM). Distributions of nutrients between manure types after their excretion at barn have to be known for example to estimate losses to the environment (ex: IPCC., 2006; Hutchings *et al.*, 2013)).

Materials and Methods

Distribution between manure types for a given element x barn category was the mean of the distributions for the nine animal categories considered in the Nitrate directive. For a given element x barn system x animal category, the distribution was derived from the amount of the excreted element in the manure divided by the total element excretion. The total element excretion was the sums of the amounts of the element in the different manures. The amounts of the element in the different manures were calculated from the amount of the element remaining after manure storage to witch a proportion is added due to losses at barn and storage: [Concentration (kg/t FM) x production (m³) x density (t /m³)]/[1-proportion of losses (g/g of excreted)] = Excreted.

The N, P, K and DM concentration in manures before application and volume productions (i.e. after storage) were taken from the nitrate directive and Piazzalunga et al., 2012. One concentration of N per manure type was available, all animal categories included. Density of 0.7, 0.9, 1.0 and 1.0 t fresh manure/m³ were used for respectively FY, SSM, SL and LF. Proportion of losses for N and P, K and C are in table 2. For FY, seeped liquid fraction during storage on field was considered as lost while for, SSM, it was not because of the obligation to keep it on store with seeped fraction collecting system. For C, the distribution was estimated as equal to OM distribution.

Results and Discussion

The theoretical distributions of excreted elements are presented in table 3. Negative losses of OM and P relatively to their excretion were used for FY because of their lower losses compared to their supply with straw for bedding (table 2). No losses of P and K should normally occur in SSM storage systems with collection of seepage. However, the values used reflect more the reality than the theoretically null losses values used for SL and LF. Furthermore, intermediate results are the estimations of excreted N by the nine animal categories in the different barn systems. They show high variation (mean of the variation coefficient: 14%) probably reflecting the unaccounted variability of elements concentrations in link to the different manure types by using the few recognized generic values.

Table 1: Barn types (Nitrate directive: AGW, 2013; Nitrawal, 2013).

1. Cubicle house with and without bedding supply
2. With slatted floor or without bedding supply
3. Deep litter or fully covered by bedding with partial manure removal by scrapping at low frequency (>5 days)
4. Tied stall with bedding supply
5. Cubicle house with bedding supply
6. Fully covered by bedding with partial manure removal by scrapping at low frequency (< 5 days)
7. Cubicle house with partial bedding supply
8. Partially covered by bedding

Table 2: Coefficients used for the estimation of the excretion (g/kg excreted).

	Proportion of losses				Straw supply			
Manure	N	OM[a]	P	K	DM[b]	N	P	K
FY[c]	312	-191	-215	150	1690	168	215	229
SSM[c]	214	105	21	89	185	21	31	18
SL[d,e,f]	410	100	0	0	0	0	0	0
LF[d,e,f]	410	100	0	0	0	0	0	0

[a]OM: organic matter = DM[b]-ash; [b]DM =Dry matter; Source: [c] Mathot *et al.,* 2011, [d] Lambert *et al.,* 2006; [e]Vredenne *et al*., 2008; [f]IPCC 2006.

Table 3: Theoretical distribution (g/kg excreted) of the elements (C, N, P, K) in manure type (1 to 8 see table 1) at release by animal (mean (standard deviation) of the nine animal categories).

		Barn category							
Element	Manure	1	2	3	4	5	6	7	8
C	FY			1000 (0)			452 (45)		523 (37)
	SSM	577 (36)			948 (2)	963 (3)	507 (43)	577 (41)	
	SL	423 (36)	1000 (0)					423 (41)	477 (37)
	LF				52 (2)	37 (3)	41 (3)		
N	FY			1000 (0)			412 (45)		413 (39)
	SSM	496 (40)			917 (3)	941 (5)	519 (40)	496 (40)	
	SL	504 (40)	1000 (0)					504 (40)	587 (39)
	LF				83 (3)	59 (5)	69 (5)		
P	FY			1000 (0)			411 (45)		386 (38)
	SSM	500 (40)			1000 (0)	1000 (0)	589 (45)	500 (40)	
	SL	500 (40)	1000 (0)					500 (40)	614 (38)
	LF								
K	FY			1000 (0)			505 (45)		610 (37)
	SSM	573 (39)			846 (6)	887 (9)	390 (36)	573 (39)	
	SL	427 (39)	1000 (0)					427 (39)	390 (37)
	LF				154 (6)	113 (9)	104 (9)		

Conclusions

Even if aware of the limits of the approach, it is proposed, to use those distributions to help in modelling the fate of element in manure like as recommended by IPCC methodology.

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HISTORICAL DEVELOPMENT OF AMMONIA EMISSIONS AND NITROGEN FLOWS RELATED TO SWISS AGRICULTURE

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Reliable data about the historical development of ammonia (NH_3) emissions from agriculture is needed for a better understanding of changes in the environmental pressure resulting from NH_3 concentrations and nitrogen (N) deposition, and for the comparison of agricultural emissions with other anthropogenic emissions. To allow a meaningful interpretation of such results, the emission calculations should look back over at least one century and should take into account not only changes in animal numbers but also in livestock and manure management practice. The aim of our study was to perform such a historical analysis of agricultural NH_3 emissions based on a detailed review of statistics and publications on farming practice and using state-of-the-art N flow emission modelling.

Materials and Methods

The project consists of the following main modules:

1) Review on livestock numbers, animal performance and feeding practice between 1866 and 1990.

2) Review of manure management and fertilisation practice since 1860 using recommendations as well as practical and scientific literature.

3) Differentiated estimate of the development of N excretions of different livestock categories between 1866 and 1990.

4) Emission calculations using the N flow model AGRAMMON (Kupper et al. 2010) for selected years between 1860 and 1990 (1866, 1886, 1906, 1916, 1921, 1926, 1936, 1941, 1946, 1956, 1966, 1973, 1978, 1983, 1988) using the findings of modules 1) to 3).

5) Combination of the results of module 4) with the existing detailed emission inventories for 1990, 1995, 2002, 2007 and 2010 (Kupper et al. 2012).

6) Interpretation of the time series results of module 5) for N excretions and NH_3 emissions of different livestock categories and different source categories (housing, manure storage and application, mineral and organic residue fertilizers, crops).

7) Complementing the results of module 5) with non-agricultural NH_3 emissions and NO_X emissions from different source categories (since 1990 as reported to CEIP; before 1990 according to unpublished revised data from the Swiss Federal Office for the Environment).

Selected results

The number of dairy cows was only 6% higher in 2010 than in 1866 (maximum 1966 +66% relative to 1866). The number of fattening pig places was 308% higher in 2010 than 1866 (maximum +539% in 1978). Total N excretions from livestock increased by a factor 2.0 between 1866 and 2010 (from 65.0 to 129.4 kt N). The maximum was reached in 1978 with 147.8 kt N. Total emissions from agriculture and from livestock and manure management both increased by a factor 2.0 between 1866 and 2010 (total emissions from 24 to 48 kt N). A maximum of total agricultural emissions was reached in 1983 (factor 2.4 relative to 1866). The share of livestock production and manure management fluctuated between 87% (1988) and 94% (1926). Cattle always contributed around 80% of the emissions from livestock and manure management, the share of pigs increased from 4% to a maximum of 18% for 1983-1990.

From 1866 to 1995 the contribution of manure use, housing, manure storage and grazing to emissions from livestock/manure always was around 60%, 20%, 20%, < 2%, respectively. Since 1995 the share of housing emissions increased to 34% in 2010, while the share of manure use decreased to 46%. This development can mainly be explained by the rapid increase of loose housing systems and increased grazing for cattle which led to higher initial emissions in the housing area and thus to a reduced N flow through the manure chain.

Emissions from fertilizers (mineral and recycling) gradually increased from 0 to 4.9 kt in 1988 and since decreased by 50% thanks to nutrient balance legislation and the ban on sewage sludge use.

Total atmospheric emissions of reactive N, NH_3 plus nitrogen oxides (NO_X), were 89% higher in 2010 than in 1900. Thus, 2010 was comparable to the level around 1960. The maximum was reached in 1983 (factor 2.9 relative to 1900). The contribution of NH_3 to total atmospheric emissions of reactive N was >80% from 1900 to 1946, then declined to a minimum of 53% in 1983 and then increased again to 68% in 2010.

Conclusions

NH_3 emissions from Swiss agriculture changed considerably over the past 150 years. The emission increase was quite steady around +1% per year for 1866-1916 and between 1% and 2% for 1946 – 1978. A maximum emission level of around 57 kt N was reached for 1983-1990. During the 1990ies the reduction of animal numbers, especially pigs, lead to a reduction of emissions, which from the mid-nineties onwards was accentuated by nutrient balance restrictions. However, this was counterbalanced by the fast shift from tied to loose housing systems for cattle initiated by incentives for animal friendly systems. From 2000 onwards specific emission reducing techniques were increasingly used and helped to stabilize the emission level. From 2014 onwards, additional incentives for conservative resource use are expected to lead to a further emission reduction.

In our time series, 1983 can be considered as a turning point. In this year NH_3 emissions from agriculture, total NH_3 emissions, total NO_X emissions and total atmospheric emissions of reactive N reached a maximum while at the same time the share of agriculture to total atmospheric emissions of reactive N reached a minimum of 50%.

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UNDERSTANDING EMERGING MECHANISMS THAT UNDERPIN NITROGEN MINERALISATION AND THEIR IMPACT ON NITROGEN MANAGEMENT STRATEGIES

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In soil, the dominant form of nitrogen and other essential plant nutrients is soil organic matter (SOM) but this source is not in an immediately available. The transformation of these nutrients to readily available forms is mediated by the microbial community. Nonetheless, SOM is a chemically complex and heterogeneous substrate. Therefore, in environments where labile C is limited (e.g. without root exudation) more energy may be required to make stabilized SOM-C available than can be gained from its utilization; in this case mineralisation of stabilized SOM is thought to be a nonviable source of carbon and nutrients. Conceptual models interpret priming effects (PE) as a mutualistic relationship between plants and microbes to acquire nutrients from SOM. That is, microbes utilize root exudates as an energy source to decompose SOM and subsequently acquire nutrients. Many studies have identified PE's but few studies have investigated the underpinning mechanisms. Here, we investigate the mechanism, drivers and regulators underpinning PE's. The objectives were: 1) to assess the importance of biotic processes for nutrient cycling. 2) To assess if the addition of labile C to soil results in an increase in gross C and N fluxes from SOM and if this effect is soil specific, independent of nutrient availability 3) To demonstrate that PE's are a response by plants to acquire necessary nutrients from SOM 4) To investigate if plant growth stimulates N mineralisation and if the presence of fertilizer nitrogen and grazing reduces plant uptake of SOM derived N.

Materials and Methods

Two soils with very different productivities, which their chemical and physical properties could not explain, were chosen to investigate the importance of biological processes for nitrogen mineralisation. ^{13}C and ^{15}N stable isotopes were used to measure specific C and N fluxes from SOM. Gross nitrogen mineralisation was measured using ^{15}N pool dilution. Glucose was added daily to quantify the effect of root exudation on C and N mineralisation within a rhizosphere environment. Total soil CO_2 efflux was measured and ^{13}C isotope partitioning was applied to quantifiably distinguish between SOM and glucose derived components. Also, ^{13}C was traced through the microbial biomass (chloroform fumigation) to separate pool-substitution effects (apparent priming) from altered microbial utilisation of soil organic matter (real priming effects). Also, in a planted system, continuous ^{13}C-labelling of plants was applied to study effects of nutrient availability (fertilization) and grazing on plant soil interactions mediating microbial mineralisation of SOM and cycling of nutrients into plant available forms.

Results and Discussion

Our results demonstrated higher gross C and N fluxes for the high productivity soil. Addition of glucose increased gross C and N fluxes from SOM. This indicates that biotic processes play a vital role in nutrient cycling. Furthermore, the C-to-N ratio of the flux from 'primed' SOM was much lower than that of the basal flux (Fig 1). This demonstrates that the release of labile carbon from plant roots functions as a nutrient acquisition response, increasing mineralisation of SOM. Also, this would indicate that the priming response targets specific organic matter pools. One suggestion is that the response is regulated by microbial production of enzymes which is activated by the utilization of labile C. Within the rhizosphere, organic nitrogen accessibility may take precedence over mineral N status in determining the direction and magnitude of priming. Addition of N fertiliser resulted in negative priming of SOM but overall, and in both soils, the plant accessed more organic-N. Grazing and priming were closely coupled, grazing increased SOM priming. This could be interpreted as a response supporting recovery from defoliation, providing nutrients for re-growth of plant tissue.

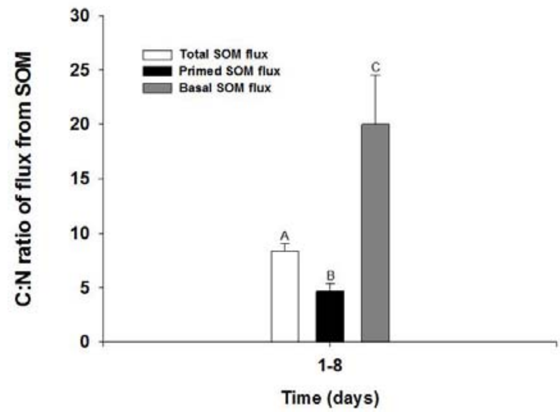

Figure 1. C-to-N ratio of C and N fluxes from SOM for the high productivity soil (HPS). Different letters indicate significant (< 0.05) differences between treatments. Mean values (n=4) of experimental data are reported with standard error bars.

Conclusions

Here, using quantitative methods, we clearly demonstrate that priming effects are an integral component of N mineralisation. Establishing an understanding of the mechanisms underpinning gross N mineralisation creates a platform for the furtherance of nitrogen management strategies. Furthermore, with an understanding of plant soil interactions there is the capacity to manipulate plant-soil interactions for the optimisation of multiple soil functions, such as resource capture and nutrient cycling; this allows for increased yet, sustainable crop production whilst reducing fertilizer use and GHG emissions.

NITROGEN DYNAMICS IN SOILS OF CENTRAL AFRICAN SMALLHOLDER FARMS

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Investigations on the fertility status of soils in the Western Plateau of Rwanda (Gatumba area) have shown that pH, organic C and total N contents as well as cation exchange capacity were low not only in naturally occurring soils but also in Technosols from mining dump material (mainly pegmatite of tantalum mining areas; Reetsch et al., 2008; Knoblauch, 2012). Technosols are frequently used by farmers for crop production. Some exceptions are humic-rich soils such as Umbrisols and colluvic soils that receive soil organic matter (SOM) from supply of eroded topsoil material.

Materials and Methods

In 2011, soil samples were taken in the Gatumba area for specific investigations on nitrogen (N) dynamics. The soils included were i) soils from farmers' fields, ii) soils from field experiments and iii) soils from mining materials (Technosols from pegmatite dump material) with different amendments. In detail, laboratory experiments on N mineralization at 35°C according to Stanford and Smith (1972) and on nitrification (25°C) following ammonium (NH_4^+) fertilizer application were conducted.

Results and Discussion

Results from incubation experiments on nitrogen mineralization yielded an extremely low N mineralization potential for fresh pegmatite (2 µg N kg^{-1} soil after 177 d). However, amendments of organic materials such as *Tithonia diversifolia* leaves in combination with different soil materials significantly increased the N mineralization potentials (Figure 1). P: Pegmatite alone; T+P+Bt: Tithonia+ Pegmatite+Bt material from Lixisol; T+P+Bt+E: Tithonia+Pegmatite+Bt+E material from Lixisol

N mineralization potentials in Rwandan naturally developed soils (smallholder farms) were higher (range: 60-92 µg N kg^{-1} soil) compared to the Technosol on fresh pegmatite (Fig 2). P5: Cambic Fluvisol; P8: Umbric Leptosol; P9: Vertic Umbrisol. The laboratory experiment on nitrification showed that after amendment of ammonium fertilizer there was only a weak nitrification activity in soils from farmers' fields as well as from Technosols (Fig 3). This might have been mostly due to the low pH (range: pH 4.1-4.7) which is representative for most Rwandan soils. Some soils with pH >4.9 showed significantly higher nitrification rates (not shown here).

Conclusions

Our results showed a wide range of N mineralization potentials in Rwandan soils. Amendments of *Tithonia* residues had a strongly positive influence on N mineralization potentials of Technosols.

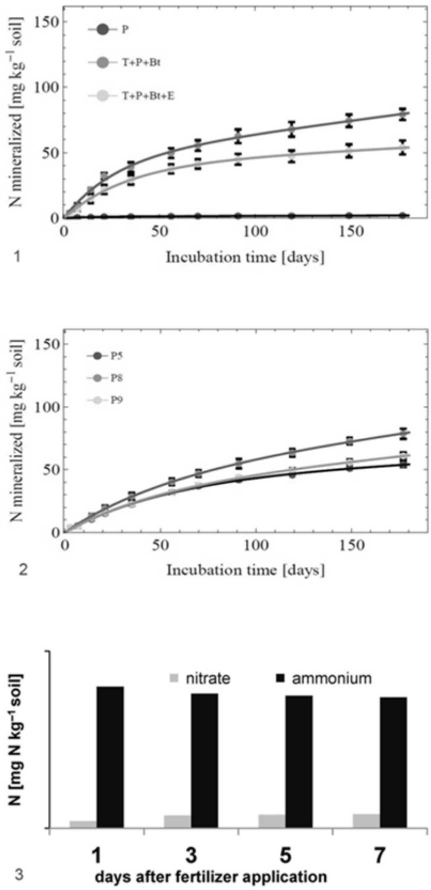

Figure – 1: Cumulative N mineralization in pegmatite alone and with different amendments. 2: Cumulative N mineralization in typical soils of smallholder farms in the Gatumba area. 3: Nitrification in soils of the Gatumba area after NH_4^+ fertilizer application (means of 9 soils)

Cumulative N mineralization in some naturally developed soils was not much lower compared to arable soils in Germany. This may indicate a potential for increasing N mineralization capacities of Rwandan soils. Nitrification potentials were extremely low in acidic soils. A positive effect of liming could be observed. This demonstrates that determination of reactive nitrogen considering a range of soils of sub-Saharan Africa needs to include nitrate plus ammonium. There should be a high priority of improving SOM and total nitrogen conditions for better N availability.

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ABATEMENT MEASURES OF AMMONIA EMISSIONS IN THE MANURE MANAGEMENT CHAIN – A QUANTITATIVE ANALYSIS

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Abatement of ammonia (NH_3) emissions from animal excretions are of particular importance in improving environmental quality and increasing nitrogen (N) use efficiency in the whole animal production chain. Various techniques have been developed and tested to mitigate NH_3 emissions from different stages of the manure management chain, and to examine possible side-effects. There is a need of synthesis information to provide farmers and policy makers with comprehensive and quantitative information on the mitigation potential of these techniques, in order to properly implement these techniques in practice.

Here, we present a review and statistical analysis of recent literature with the aims to 1) estimate the reduction potential of low-NH_3-emission techniques used for manure management, and 2) to identify possible interactions on greenhouse gas (GHG) emissions, notably nitrous oxide (N_2O) and methane (CH_4).

Materials and methods

We reviewed and examined more than 150 peer-reviewed articles, published since 1995, in which NH_3 and GHG emissions from animal manure were measured and the mitigation potential was quantified. Data from laboratory-based studies were also taken into account, because there are insufficient data from field-based studies for a solid quantitative analysis. Comparisons were made for each study between emissions from the control treatment and from treatments with mitigation measures. We examined the reduction potentials of these abatement measures for each stage of manure management chain, namely animal feeding, animal housing, slurry storage, solid manure storage, and field application through statistical analysis.

Results and discussion

Reducing dietary crude protein content in diets, with or without additional supplementation of essential amino acid significantly decrease total N excretion, especially urinary N excretion. On average a 7-12% reduction in NH_3 emissions can be achieved by lowering the crude protein content with 10 g per kg feed. Emissions from pig or cattle housings with (fully or partially) slatted floors were lower than that with deep litter, although the trend was not always consistent. Variations in thickness of bedding and removal frequency of slurry from housing affect the emissions. Methane emissions appeared to be higher (by a factor of 0.6) and N_2O emissions lower (by a factor of 0.7) in slatted-floor barns, compared with deep litter. Covering slurry storage using chopped straw, woody lid, clay granules or plastic film and acidifying slurry to a pH less than 6 reduced NH_3 emission by 65-99%. Acidification also decreases CH_4 emissions, due to the inhibition of methanogenesis at acidic condition. Covering slurry with chopped straw increases risk of N_2O emissions. About 45-75% of total carbon (C) and 30-70% of total N tend to be lost during composting, particularly if manure heaps are frequently turned over. Losses of C and N from compacted and covered manure heaps were less than 40 and 25% relative to turned heaps, respectively. Compaction increases anaerobic conditions in solid manure heaps, leading to higher CH_4 emissions.

Emissions of NH_3 were reduced by 45-65% when slurry was applied via trailing hoses or trailing shoes compared to surface spreading. Large reduction in NH_3 emission (by 75-95%) can be achieved when slurry is injected and solid manure is incorporated into the soil, but the effectiveness largely varies because of differences in slurry composition, climate conditions and vegetation types. Our analysis revealed that injection of slurry into soil increased N_2O emissions compared to surface spreading. Crop N uptake was higher following injection than following surface spreading.

Conclusion

We assessed the mitigation potential of low-NH_3-emission measures at different stages of the manure management chain, based on an extensive literature study and statistical analysis. The effects of the measures highly depend on the environmental conditions, manure characteristics and technical operations. Our analyses indicate that an accurate assessment of low-emission measures requires a chain approach, in which the effects of the measures are examined at all stages of the manure management chain. The results of our literature review and statistical analyses will be used to update the integrated assessment tool MITERRA-Europe to assess the perspective of emission mitigation options in the whole manure chain in EU-28 through scenario and sensitivity analyses.

Acknowledgement

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N MINERALIZATION FROM GREEN MANURES IN A LABORATORY INCUBATION STUDY

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The actual environmental concerns, decline in soil fertility and rising cost of chemical fertilizers instigate for finding new ways to supply nutrients for crops (Kramberger *et al*., 2009).Grren manures can be utilized for supply N to subsequent crops (Gaskell and Smith, 2007). To utilize green manures as an N source, a more comprehensive understanding of N mineralization process is essential to synchronizing the release of N green manures with uptake by following crops (Nakhone and Ali Tabatabai, 2008). In bibliography, a limited number of plant residues decomposition has been studied. The main propose of this study was to evaluating, under laboratory conditions, N mineralization after incorporation of green manures residues into a soil.

Material and methods

Soil and crops residues used in incubation experience were collected at Agrarian School of Viseu (Portugal). Crops residues experimented were: balansa clover (*Trifolium michelianum* Savi), yellow lupine (*Lupinus luteus* L.) and ryegrass (*Lolium multiflorum* Lam.) roots ans aerial biomass. In incubation experiment, nine treatments were included with four replicates: residue – unamended treatment (control) (C); aerial balansa clover (ABC); root balansa clover (RBC); aerial yellow lupine (AYL); root yellow lupine (RYL): aerial ryegrass (AR); root ryegrass (RR); mixture of yellow lupine (MYL); mixture ryegrass (MR) and a container without soil and plant residues – blank (B). At each treatment 140 mg N kg^{-1} of crop residue was incorporated homogeneously into the soil (equivalent to 50 g oven dried soil), except treatmento RBC where was added 100 mg N kg^{-1} because had insufficient amount of roots.Treatments were placed individually in a hermetically glass pot and incubated at constant temperature (25º C) for 196 days. N mineralization was measured at 0, 1, 3, 7, 10, 14, 21, 35, 56, 84, 112, 154 and 196 day. At outset of experiment were prepared treatments for to be changed at the day of N mineralization measures.

Results and discussion

At the end of incubation experiment (196 day), the cumulative mineral N was higher in soils amended with legumes, comparatively with non legumes (Fig. 1). Cumulative N mineral of MYL are smaller than aerial residues and root residues separately, and are significantly lesser than roots residues. N mineralized content ranged between 55% for RYL and 13% for RR. Ryegrass differs statistically of legumes. MYL is significantly lesser than roots and aerial biomass separately. For ryegrass, mixture not statistically differs for aerial biomass, but is significantly higher than roots. RBC and ABC have a similar percentage of mineralized N. Legumes residues decompose more rapidly than non leguminous (ryegrass) like in a study developed by Li *et al*. (2013). Addition of ryegrass (RR, AR, MR) plant residues produced a rapid N immobilization. In experiments developed by Cayuela *et al*. (2009) with wheat straw also verified rapid N immobilization after residues incorporation. Aerial biomass of legumes not differs statistically, but roots of yellow lupine are significantly bigger than roots of balansa clover. MYL showed less 20% N mineralization than AYL and 32% lesser than RYL. Ryegrass mixture not differ statistically by AR or RR, but RR is statistically lower than AR, corresponding at 36% lesser cumulative mineral N. Frankenberger and Abdelmagid (1985) cite factores like N content of residues, lignoprotein complexes and their resistance to microbial decomposition and toxic metabolites associated with decomposition, inhibiting N mineralization for justify differences among fractions of plant residues in releasing N. In Yellow lupine treatments, with smaller C/N ratio not observed N immobilization. Contrary, ryegrass (RR, AR and MR), with higher C/N ratio, is the treatment where is verified more immobilization. Addition of residues with high C/N ratio to soil induced net N immobilization during their decomposition in soils (Li *et al*., 2013). Roots yellow lupine has a bigger C/N ratio than AYL and has a larger cumulative N mineral, although not differ statistically.Abiven *et al*., (2005) affirm that root decomposition is not always lower than aerial plant residues.

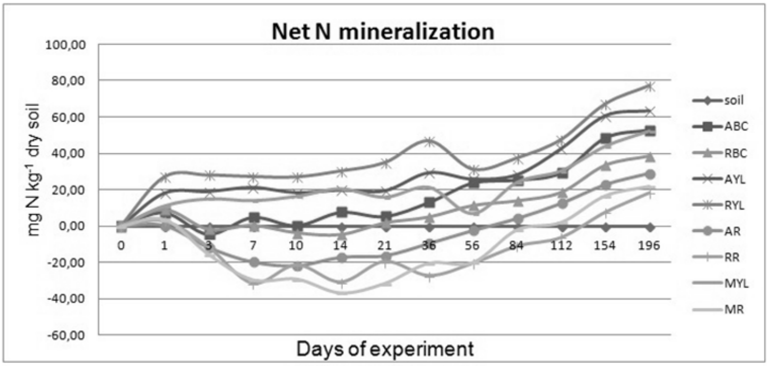

Figure 1. Net N mineralization (mg N kg^{-1} dry soil).

Conclusions

Our observation showed that plant residues have a significant impact on N mineralization rate. Legumes residues have a bigger N mineralization rate those non legumes. Not observed differences between aerial biomass and roots N mineralization for each green manure. In green manures evaluated, was the yellow lupine (roots or aerial biomass separately) that showed the best like a nitrogen source. Further studies must be carried out in order to determine the relationship of plant residues constituints and their N mineralization.

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MINERALISATION NITROGEN DURING ANAEROBIC CO-BIODIGESTION FROM LIQUID FRACTION OF WASTE FROM CATTLE AND SHEEP

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Productivity growth in livestock production systems has major importance in the country's economy. However, this increase brings an increased production of waste that if is not stabilized properly can cause serious damage to the environment (ORRICO JUNIOR et al., 2012). The stabilization of the waste by the anaerobic biodigestion process is interesting because it also promotes energy and nutrient recycling. Variations of this process, as anaerobic co-biodigestion from the liquid fraction of waste improve the efficiency of biogas production (ROZATTI et al., 2013), and may increase mineralization of some nutrients, as N. Lopes et al. (2013) concluded that in the first 14 days of anaerobic biodigestion of manure from beef cattle there was a percentage of around 14 % of ammonification. After this period, the conversion from organic N to ammoniacal N remained almost constant, in the range of 27 %. The objective is to assess the dynamics of N mineralization during the process of anaerobic co-biodigestion from liquid fraction of manure from cattle and sheep.

Material and Methods

The test of anaerobic co-biodigestion was conducted at the Laboratory of Analysis of Agroindustrial Waste belongs to WESTERN PARANA STATE UNIVERSITY – UNIOESTE – Campus Cascavel – Paraná – Brazil. Four sets of digesters bench, made of PVC and compounded by a fermentation chamber (capacity of 7 L), gas meter and mini-biodigesters (PET bottles) with a capacity of 500 mL were used. The substrate was obtained by mixing cattle and sheep manure, in 1:1 ratio, considering the dry matter. To this mixture was added sufficient water to make the liquid fraction, after separation of the phases, presents 5% of total solids. The separation was performed 24 hours after mixing with water, in a 3 mm mesh sieve. Each one of the four fermentation chambers were stocked with six liters of substrate and 40 PET bottles with 500 mL.Weekly, four PET bottles were opened to determinate total Kjeldahl nitrogen, ammoniacal nitrogen, nitrate and pH, managed according to *Standard methods for the examination of water and wastewater* (APHA, 2005). The efficiency of nitrogen conversion from organic to ammoniacal form, that is, the percentage of ammonification was calculated from the equation (eq. 1).

Eq1: $A\ (\%) = (1\text{-}NTK_{j}\ _{-}NNH^{4+}f)x100$

The results obtained were subjected to analysis of variance and the averages were compared with each other by LSD test at 5% probability.

Results and Discussion

Approximately 88% of the TNK is converted to N-NH_4 during anaerobic co-biodigestion of the liquid fraction of the manure from cattle and sheep (Figure 1A), in a process called ammonification. This form of N is assimilable by plants, making biofertilizer important as a supplier of this nutrient (SIQUEIRA and MOREIRA, 2006). The dynamics of formation of N-NH_4 is on Figure 1B, and it can be observed that the maximum concentration of this element takes up to 53 days, this being some important information in decision making on the hydraulic retention time (HRT).

Another form of labile N is nitrate. However, in the process of anaerobic biodigestion, given the low concentration of oxygen, there is little activity of nitrifying bacteria. Chernicharo (1997) notes that, under anaerobic conditions, nitrogen in the form of nitrite and nitrate are not available for bacterial growth, since it is reduced to N gas and released into the atmosphere. It can be seen from Figure 1B that there was nitrification at the beginning of the process (up to 25 days), and after NO_3^- levels decreased significantly.

Conclusions

It occurs conversion from organic nitrogen to mineral (ammonium) during anaerobic co-biodigestion from the liquid fraction of waste from cattle and sheep.

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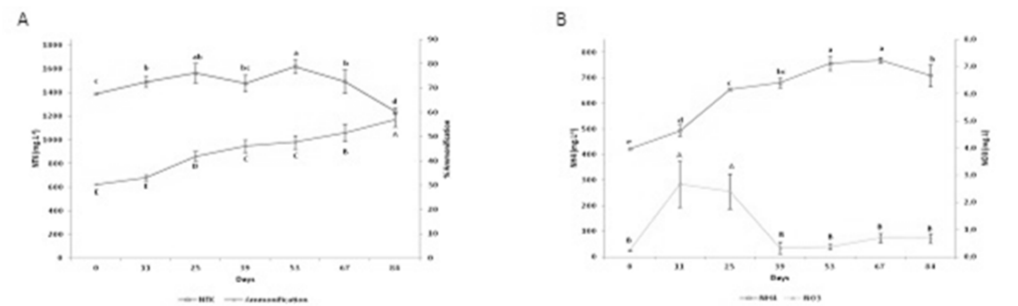

Lowercase letters equal to NTK and NH_4 and capital letters equal to % of ammonification and NO_3, do not differ at 5% of significance by LSD test.

Figure 1. Averages of TNK, % of ammonification (A), NH_4^+ and NO_3^-(B) in time.

MINERALIZATION OF ORGANIC FERTILIZERS PACKED IN POROUS CAPSULES DURING AUTUMN-WINTER SEASONS

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The food that the poultry receive in the aviaries is rich in protein, amino acids, vitamins and carbohydrates and the excess of nutrients ingested by the poultry is excreted though feces and urine. In this way, poultry litter, which is rich in nutrients, is often used as soil fertilizer, once it has satisfactory quantities of nitrogen, phosphorus and potassium. However, poultry litter and other organic materials submitted to biological processes of recycling have its mineralization speed varying according to the seasons of the year. The availability of nutrients in each organic fertilizer may not match the season in which the demand of nutrients is greater. In this study we assessed the mineralization of carbon and nitrogen in organic fertilizers (compost, biodynamic compost, vermicompost, vermicompost + P, poultry litter) packed in porous capsules during autumn/winter seasons. Each organic fertilizer must be analyzed before its application to the soil once the mineralization rate may vary for each. Therefore, it is fundamental to have knowledge of the rate of the mineralization of the organic materials when applied to the soil in order to proceed with the recommendations of organic fertilizers for different seasons of the year and regions. Assess the availability of nitrogen from different organic materials is the aim of this paper.

Material and Methods

The experiment was carried at the *Núcleo Experimental da Engenharia Agrícola – NEEA*, in Wester Paraná State University – *UNIOESTE*, Campus of Cascavel. The soil classified as typical dystroferric red latosol (EMBRAPA, 2006). The block design was completely randomized, with four replications in a factorial (5x5) consisting of five sample collecting moments in autumn-winter (7th, 14th, 35th, 65th and 100th days) and five organic fertilizers: pelletized vermicompost (V); pelletized vermicompost + Arad phosphate (VP); compost (C); biodynamic compost (CB), poultry litter (CF) from three lots. Porous capsules (ceramic), with an outside diameter of 5.3cm and 4cm, internal length of 9.8cm and weighing 174.33g were used. The porous capsules have micro porous wall of 1.3cm, with retention of particles above 1.0 microns. Before installing the experiments, the capsules were first immersed in water for 24 hours, then immersed in deionized water for 24 hours and then air dried. The filling of capsules was performed with dry organic materials in an forced air circulation at 65 °C. Each porous capsule was filled with 20g of organic material. The installation of the capsules in the soil was at a distance of 0.10 x 0.20m to 0.07m depth and covered with a layer of 0.02m in the soil. The sampled capsules were taken to the laboratory and the organic material contained in it was removed and dried in an oven with forced air circulation at a temperature of 65 °C to constant weight to determine the remaining mass. The amount of remaining material during 100 days was evaluated by the following formula: remainder (g) = (initial mass – final mass). For the organic fertilizers the remaining carbon and nitrogen were determined by Malavolta; Vitti; Oliveira (1997).

Results and Discussion

Among the organic fertilizers the poultry litter showed greater decomposition of carbon and nitrogen (Figure 1). For the poultry litter, there was a greater amount of carbon released from the capsules from the 35th day. On the other hand, the decomposition of the poultry litter and nitrogen availability began after 7 days of the incubation of the capsules. The ceramic capsule provides a proper incubation chamber for determining the mineralization of the nitrogen of biosolids through residual techniques (Henry et al. 2000). In addition, the porous capsules allow the passage of water, oxygen and microorganisms; these conditions favor the decomposition (Pereira, 2011). During autumn-winter seasons, at the 100th days, the poultry litter gave 30 and 40% decomposition of carbon and nitrogen, respectively. In general, the weight loss may be directly proportional to the amount of nitrogen of the organic material (Cotrufo et al., 2009). The compost and the biodynamic compost due to its high stability showed similar behavior for both carbon and nitrogen and demonstrate mineralization of 7 and 10% carbon and 1.7% and 4.0% nitrogen, respectively. The vermicompost and vermicompost + Arad phosphate showed intermediate condition for the decomposition of carbon and nitrogen, 16 and 13% carbon and 21 and 17% nitrogen, respectivelly. These fertilizers still have high amount of nutrients to decomposition when excreted by the earthworms due to its rapid digestion.

Conclusion

Poultry litter is more efficient in providing N during the autumn-winter season.

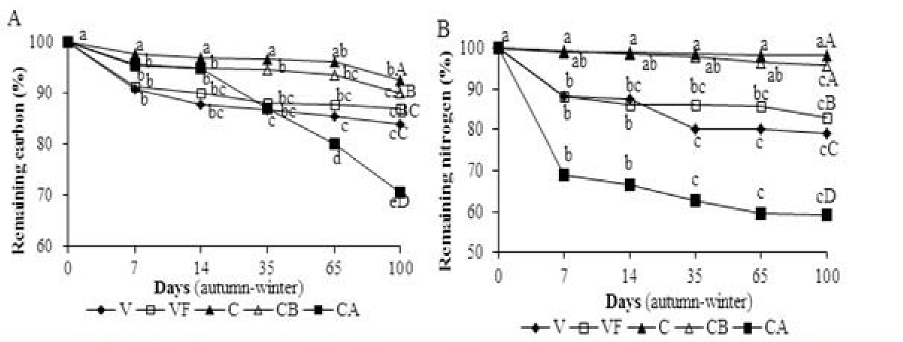

Figure 1. Remainings of carbon and nitrogen from organic fertilizers packed in porous capsules.

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NITROGEN DYNAMICS IN TWO SOILS WITH DIFFERENT TEXTURE TREATED WITH LIVESTOCK-DERIVED ORGANIC MATERIALS

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The use of livestock-derived organic materials (LOMs) could partially offset the need for mineral fertilizers, giving both economic and environmental benefits. The LOMs, other than C, contain variable amounts of organic N, whose fate in soil depends on the mineralization rate of the different organic pools, which, in turn, is influenced by the degree of stability of the materials. Therefore, the aim of this work was to evaluate the effect of the addition of differently stabilized livestock-derived organic materials on N dynamics in two soils with different texture.

Materials and Methods

The soils used in the experiment were two agricultural soils collected near Perugia (Central Italy). The first soil, classified as Typic Ustifluvent (Soil Survey Staff, 2010) with a silty loam texture (SL), was sampled within an agricultural field located in Casalina Deruta (PG). The second soil, a Typic Haplustalf with a silty clay loam texture (SCL), was sampled in Petrignano (PG).

Aliquots of the two soils were amended with pig slurry (PS), solid fraction of the digestate produced by anaerobic fermentation of the same pig slurry (DPS), and compost obtained by the aerobic stabilization of the solid fraction of digested pig slurry (COM). The main characteristics of these organic materials were reported in Table 1. The organic materials were added to 200 g of SL and SCL soil and their amount was calculated on the basis of the N concentration in the soluble fraction of the three amendments and considering a soil layer with a thickness of 15 cm to simulate a spreading equivalent to 340 kg ha^{-1} of N, which is the maximum N application allowed by the European "Nitrate Directive". Controls (CNT), made of soil samples without any LOM adding, were prepared accordingly. The amended and control samples were incubated for 45 days at 25°C and 60% of water holding capacity. During the incubation period, three pots for each treatment and soil type were sampled after 3 h, 5, 12, 20, 30 and 45 days, and analyzed for their content of NH_4-N, NO_3-N and microbial N (N_{mic}). Moreover, Total and organic N contents were determined at the beginning and at the end of the experiment, both in treated and untreated samples.

Results and Discussion

Considering the SL samples, the concentration of NH_4-N had a decreasing trend for all the treatments during the incubation period. With the exception of significant higher values ($p< 0.05$) occurred after three hours and 5 days of incubation for the samples amended with PS (Table 2), all the SL samples had a low NH_4-N concentration that did not significantly differ from that of the control. The SCL samples did not show any definite trend throughout the experiment. However, also in this case, the samples amended with PS had higher NH_4-N concentration ($p< 0.05$) than the other treatments and control for the first 5 days of incubation. The initial high values of NH_4-N concentration in SL and SCL amended with PS was likely due to an exogenous source of ammonia derived from the added organic materials. This was probably related to the less stabilized organic matter added with pig slurry, which led to a rapid transformation of organic N to NH_4-N. Afterwards, the subsequent rapid decrease of NH_4-N in SL soil might be due to its texture, coarser than SCL, and the resulting major oxidative conditions, which promoted nitrification process.

Table 1. Chemical characteristics of LOMs

Parameter	PS	DPS	COM
TOC g kg^{-1}	345 ± 32	342 ± 4	318 ± 10
TN g kg^{-1}	44.2 ± 6.7	29 ± 0.2	31.8 ± 1.4
NO_3- N g kg^{-1}	-	-	-
NH_4-N g kg^{-1}	16.8 ± 1.4	7.7 ± 0.9	6.3 ± 1.1
N_{org} g kg^{-1}	27.4	21.3	25.5
C/N	7.8	11.8	10

TOC: Total Organic Carbon; TKN: Total Kjeldahl Nitrogen

Table 3 showed that the SL soil amended with PS and DPS, after 5 days had a higher NO_3-N content than the control ($p< 0.05$), unlike samples treated with compost. Moreover, the greatest concentration of NO_3-N was observed in SL samples amended with PS in correspondence with one of the lowest value of NH_4-N concentration (45 days), suggesting that the coarser texture allowed oxidative transformation of N contained in the pig slurry. With regards to N_{mic}, the obtained results showed that, at the end of the experiment, in both SL and SCL, N_{mic} was present in higher amount in the samples amended with DPS (18.22 and 28.87 mg kg^{-1} d.w. in SL and SCL, respectively) and this might be attributed to the presence of an autochthonous microflora in the digested pig slurry. Also the increase of organic N was much more noticeable in SCL soil, particularly in the samples amended with PSL at the end of the experiment (545.3 and 1165.94 mg kg^{-1} at the beginning and 45 days, respectively).

Table 2. Amount of NH_4-N (mg kg^{-1} d.w.) in amended and unamended SL and SCL soils during incubation time.

Treatment	3h		5 days		12 days		20 days		30 days		45 days	
	SL	SCL	SL	SCL	SL	SCL	SL	SCL	SL	SCL	SL	SCL
CNT	12.75	19.58	6.9	25.15	4.87	16.4	1.62	25.76	1.24	14.73	2.32	21.03
PS	52.94	37.97	23.83	36.65	3.08	19.13	4.26	26.09	1.44	12.14	2.24	21.17
DPS	13.16	22.7	7.49	27.78	3.4	18.01	3.24	23.22	1.13	11.3	2.22	23.35
COM	13.41	19.97	8.37	26.13	3.9	18.02	1.73	23.69	0.31	14.27	1.43	26.1

CNT= control; PS=pig slurry; DPS= digestated pig slurry; COM= compost obtained by digestate.
The standard error of the mean from three-way ANOVA was 2.26 (48 DF). Corresponding to a L.S.D. ($p<0.05$) of 6.42.

Table 3. Amount of NO_3-N (mg kg^{-1} d.w.) in amended and unamended SL and SCL soils during incubation time.

Treatment	3h		5 days		12 days		20 days		30 days		45 days	
	SL	SCL	SL	SCL	SL	SCL	SL	SCL	SL	SCL	SL	SCL
CNT	16.6	16.8	44.64	31.89	52.64	46.81	56.42	57.47	39.35	18.08	51.83	35.74
PS	15.58	22.74	54.01	32.2	64.21	44.3	74.48	34.51	62.3	15.81	96.11	33.42
DPS	26.12	7.63	55.23	28.6	34.09	35.34	26.6	19.75	18.89	52.73	33.25	27.94
COM	19.32	4.34	26.44	46.09	51.1	18.06	41.16	27.26	14.89	16.51	12.38	7.98

CNT= control; PS=pig slurry; DPS= digested pig slurry; COM= compost obtained by digestate.
The standard error of the mean from three-way ANOVA was 1.83 (48 DF). Corresponding to a L.S.D. ($p<0.05$) of 5.2.

Conclusions

The present study showed that the different physical properties of the soils used in the laboratory experiment could drive the N transformation after the addition of different stabilized organic materials. In fact, SL underwent a greater depletion of NH_4-N than SCL, where not all the NH_4-N was oxidized to NO_3-N. This was ascribed other than lower oxidative condition due to coarser texture, to a possible fixation of NH_4-N on clay surfaces since the early hours of incubation. This suggested the importance of the stability of LOM, since the different forms of N added to soils might have different effect on N mineralization and hence different environmental implications. Therefore, the results showed that the biological processes can represent for farmers a solution to mitigate N losses from agricultural soil to the water bodies and to the atmosphere, which can represent one of the major threats to the environment. Therefore, the choice of the right process for organic matter stabilization, i.e. composting vs anaerobic digestion may significantly reduce N losses from the organic amendment as the increase availability of N in the anaerobic digestate facilitates a prompt N uptake by plants. For these reasons, it might be interesting further investigation and to verify the findings in a field scale.

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INTERACTIONS BETWEEN THE CHEMICAL QUALITY OF CROP RESIDUES AND THEIR LOCATION IN SOIL: HOW NITROGEN AVAILABILITY CONTROLS MINERALIZATION OF C AND N?

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Nitrogen (N) availability can control the kinetics of decomposition of plant residues and the net mineralization of N in soils, due to the high microbial N requirements during decomposition (e.g. Recous et al., 1995). In field conditions, overall N availability for decomposers depend on the soil mineral N content, the amount and type of plant residue, and particularly their tissue N content (i.e. C:N ratio) and the location of plant residues (incorporated or at the soil surface, as in no-tilled systems). Indeed initial residues N content can be a good predictor of the net N mineralization during decomposition. However, the studies in field experiments or laboratory incubations aimed at varying the availability of N during decomposition (either by adding N to soil, or by varying type of plant residues) showed inconsistent effects on decomposition. Therefore, the aim of our study was to investigate for a range of crop residues, the effect of the availability of N on the C and N mineralization. The availability of N was manipulated by varying the initial quality of the residues, the residues placement, and the supply of mineral N to the soil.

Materials and Methods

The shoots of ten species of plants were collected at flowering and harvest for cover crops and main crops species, respectively, and characterized chemically (Table 1). The residues were dried at 40°C and leaves and stems were cut into pieces of 1 cm length. Mixture of leaves and stems were prepared with a ratio leaf:stem similar to the ratio in dry biomass observed in field. The soil used was a Typic Hapludalf collected from the 0-10-cm layer. Two initial soil mineral N concentrations were used, i.e. 9 mg N kg^{-1} dry soil (low N availability; 9N) and 77 mg N kg^{-1} dry soil (high N availability; 77N), obtained by adding KNO_3-N prior to incubation. The residues added at a rate of 0.6 g dry matter (DM) per pot (equivalent to a basis of 5.0 g DM kg^{-1} dry soil) were applied either on the surface (S) of soil samples or incorporated into the soil (I). C mineralization was assessed by quantifying continuous CO_2 release using NaOH trapping, at 2, 4, 7, 10, 14, 21, 28, 35, 50, 70, 90 and 120 days after start of incubation. The soil mineral N content was measured destructively, at day 0 and at 7, 14, 21, 35, 63, 90 and 120 days of incubation.

Results and Discussion

The type of residues did modify significantly the kinetics and rates of C and N mineralization in soil (not shown) as expected, and strongly interacted with soil mineral N availability and localization. For example, the residue of wheat (Fig. 1a) showed +6% (S) and +8% (I) C-CO_2 evolved with high soil mineral N (77N) compared to low soil mineral N (9N). For rape residues (Fig. 1c) there was no significant difference in C-CO_2 evolved whatever the soil N treatment and the residue location. This indicates that the increase of soil mineral N for high C:N wheat residues removed a N limitation of decomposition while the initial availability of N was sufficient for rapeseed residue for its optimal decomposition. Regarding soil N dynamics, we observed with wheat (Fig. 1b) that net immobilization of soil N was all the more important that the availability of N increased, with the following ranking: I-77N> I-9N> S-77N> S-9N. For oilseed rape residues (Fig. 1d), it is noticeable how N mineralization was strongly influenced by residue location: residues decomposing at the soil surface induced net mineralization of N compared to incorporated residues. The amount of mineral N in soil (9N or 77N) also influenced the net N mineralization. These results suggest that microorganisms had more efficient use of N (immobilized N by gram of decomposed C) and/or lower N requirements when residues were left on the surface due to shift in microbial populations with surface decomposition.

Table 1. Initial composition of the crop residues

Species	SOL[(a)]	HEM	CEL	LIG	C	N	Csw	Nsw	POL	C:N
	g kg^{-1} DM									
Soybean	349	122	386	143	450	11.7	94	10.5	21.7	38
Maize	141	323	469	67	452	4.3	46	3.5	6.4	105
Sunflower	322	79	485	114	428	9.6	84	8.0	37.4	45
Gray mucuna	464	119	318	99	451	29.4	168	7.1	21.4	15
Showy rattlebox	417	90	408	84	445	22.4	45	7.4	23.5	20
Black oat	290	246	417	47	447	12.2	104	10.4	15.2	37
Vetch	571	88	272	69	453	35.2	219	19.3	26.3	13
Wheat	326	257	356	61	437	4.9	94	4.3	24.5	89
Oilseed rape	394	152	359	95	421	19.1	60	15.0	14.8	22
Barley	271	260	407	62	441	5.3	109	4.7	12.1	83

[(a)] SOL = Soluble fraction (Van Soest); HEM = Hemicellulose; CEL = Cellulose; LIG = Lignin; C = Total carbon; N = Total nitrogen; Csw = Water-soluble carbon; Nsw = Water-soluble nitrogen; POL = Soluble polyphenols.

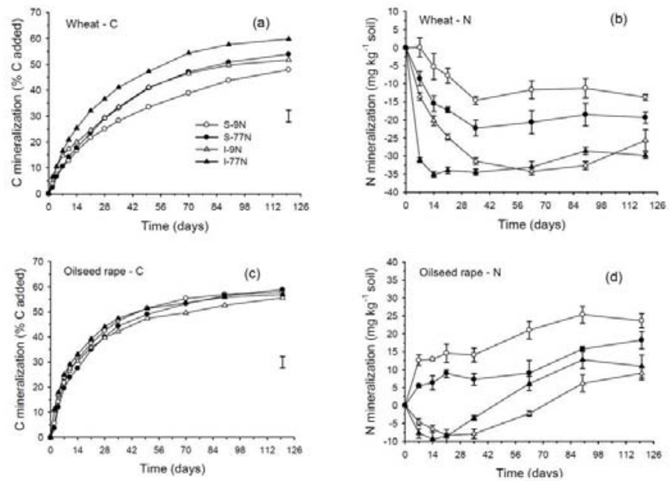

Figure 1. Cumulative CO_2 mineralization (a, c) and net changes in soil mineral N contents (b, d) of the wheat and oilseed rape residues during decomposition in soil at 25°C during 120 days. Bars are minimum significant difference between treatments (P<0.05) (a, c) and standard deviation values (n=3) (b, d).

Conclusions

The results indicate that the effects of soil-N and residue-N availability on C and N mineralization are strongly dependent of the location and type of residue in the soil.

Acknowledgements and References

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NITROGEN MINERALIZATION FROM A SOIL AMENDED WITH SEAWEED AND FISH WASTE COMPOST

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When seaweed and fish wastes are not properly managed, they may constitute an important waste in coastal areas. However these materials represent a natural resource with great potential to be used in agriculture. Seaweed and fish wastes can be composted to produce fertilizers, providing an appropriate method of waste management from both economic and environmental points of view. Among its properties, it is worthy to note the high content in organic matter and nutrients (Illera et al., 2013). Nevertheless, nutrients present in compost are mostly in organic form and need to be mineralized before being available to the plants. The mineralization process differs between composts since it depends on many factors including C/N ratio, composting conditions, maturity and compost quality (Amlinger et al., 2003). For this reason, it is necessary to study the mineralization dynamics of each compost, in order to establish guidelines for its use, maximizing the use of the resource and avoiding potentially losses of nutrients. In this study the release of inorganic N (NO_3^- and NH_4^+) and the dynamics of N mineralization from seaweed and fish wastes compost were studied during a 90-days incubation experiment.

Material and methods

The compost (table 1) was obtained from (a) seaweed; (b) fish waste (Trachurus trachurus L.); and (c) pine bark (1:1:3 (v/v)) (Illera-Vives et al., 2013). Incubation experiment was set in a non-amended soil coming from the upper layer of a Humic Cambisol. Soil only (C) and three different doses of the compost 39 (C1), 49 (C2) and 66 (C3) tons ha^{-1} were studied (corresponding to 610, 763 and 1017 mg of total N kg^{-1}soil, respectively). The soil moisture was adjusted to 60% field capacity. For each treatment, an amount equivalent to 100 g of air-dried soil was weighed, mixed with the corresponding amounts of compost. This process was repeated in triplicate for each sample. An aerobic incubation was performed over 13 weeks at 25°C. The containers were periodically opened to allow aeration, and water content was controlled. On days 0, 1, 2, 4, 7, 14, 21, 35, 49, 63, 77, and 90 after fertilizers addition, 25 g of soil were sampled from each container and soil mineral N (NH_4^+-N and NO_3^--N) was extracted with 75 ml of KCl (2M) and quantified as described by Houba et al. (1989). Results of the study were subjected to one-way analysis of variance, followed by Duncan test. The first order kinetic model (Stanford and Smith, 1972) was used to describe the N mineralization ($N_{min} = N_i + N_0 \times (1 - e^{(-k\times t)})$) where N$i$ is the initial amount of N, No is the amount of potentially maximum N present in the soil, k is the mineralization rate constant (d^{-1}) and t is the time (days).

Results and discussion

The concentration of nitrogen in the soil at the beginning of the experiment (Ni) was high in all treatments, with significant differences between doses, due to the large amount of available mineral N present in the compost. Soil mineral N increased over the time, keeping these significant differences during the entire experiment (Figure 1).

The release of mineral N shows important increases even in the last dates; only C1 showed a tendency to stabilization within 60 days. After 90d, soil N concentration ranged from 22.99 mg N kg^{-1} in the control to 231.59 mg kg^{-1} in the treatment with the highest dose of compost (figure 1). The R^2 obtained fitting the first order kinetic model with values around 0.7 for every compost treatments, being lower in the control. However, the fitting values were significant in all cases ($p \leq 0.05$), indicating that this model is appropriated to describe the obtained data (Table 2). Potentially Mineralizable Nitrogen (PMN) shows negative values at the first dates, indicating some N immobilization in the first stages of the process. However, this phenomenon disappears after few days, growing steadily since then, until the end of assay. At the end of the experiment almost 25% of the total N applied was available (PAN), and 9% of organic N was mineralized in C2 and C3; these results are consistent with results obtained by other authors (Bernal et al, 1988); C1 showed less efficiency, but in all cases the dose did not affect significantly the PAN or PMN of the compost.

Table 1. Chemical characteristics of compost and soil. Mean values ± standard deviation of three replicates.

Compost		Soil	
pH 1:5	6.72±0.28	pH ext.sat	4.18±0.09
EC 1 :5 (dS m^{-1})	2.21±0.26	EC sat ext. (dS m^{-1})	6.30±1.69
C (mg kg^{-1})	48.08±0.18	C (mg kg^{-1})	2.28±0.35
N (mg kg^{-1})	2.10±0.01	N (mg kg^{-1})	2.25±0.04
C/N	22.89±0.08	C/N	7.27±0.57

Table 2. Model parameters of determination estimated using the first-order exponential model. Mean values ± standard deviation of three replicates.

	Ni[a]	No[b]	k[c]	R^2	p valor
C	7.54 ± 0.49a	34.44 ± 7.31a	0.018 ±0.00ab	0.56± 0.03	0.05
C1	108.04 ± 6.25b	72.48 ± 23.98a	0.028 ±0.02b	0.69± 0.06	0.05
C2	133.37 ± 9.87c	179.37 ± 35.90b	0.007 ±0.00ab	0.70± 0.07	0.001
C3	170.90 ± 5.83d	632.20 ± 94.65c	0.002 ±0.00a	0.74± 0.15	0.001

[a]Initial Available Nitrogen [b]Potentially Mineralizable Nitrogen [c]Mineralization rate constant. Different letters after the parameters mean significant differences between treatments, calculated by the Duncan test at p<0.05

Table 3. Potentially available nitrogen (PAN) and potentially mineralizable nitrogen (PMN) from the compost predicted by the model. Mean values ± standard deviation of three replicates.

	2		21		49		90	
C1	16.79±0.91	a	19.07±0.63	a	20.63±1.39	a	21.64±2.50	a
C2	16.68±1.24	a	18.44±1.42	a	20.92±1.96	a	24.14±2.74	a
C3	16.19±0.55	a	17.57±0.27	a	19.88±0.50	a	23.54±1.26	a
C1	-0.99±0.19	a	1.46±0.70	a	3.13±0.63	a	5.13±0.51	a
C2	-1.11±0.17	a	1.26±0.83	a	4.59±1.62	a	8.91±2.52	a
C3	-0.67±0.08	b	0.81±0.46	a	3.29±1.09	a	7.21±1.94	a

Different letters after the parameters mean significant differences between treatments, calculated by the Duncan test at p<0.05

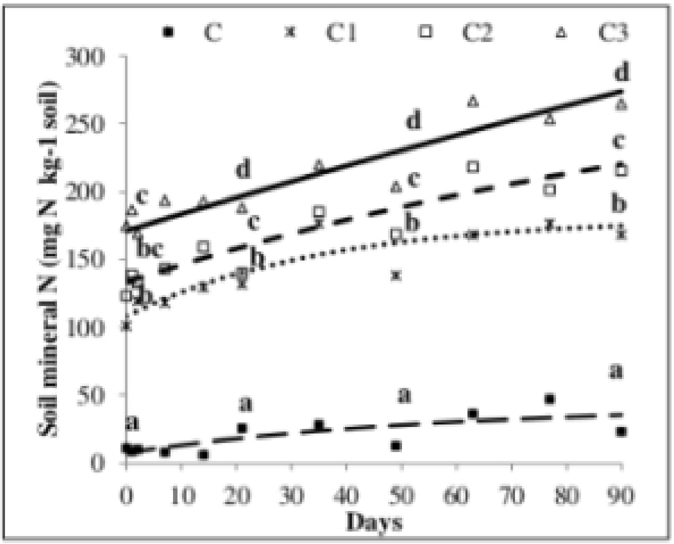

Figure1. Soil inorganic N (mg N kg^{-1} soil) observed during the 90 d incubation in the treatments studied (C1:39, C2:49 and C3:66 tons ha^{-1}). Different letters in the same date, mean significant differences between treatments, calculated by the Duncan test at p<0.05.

Conclusions

Composting of seaweed and fish waste has a relatively high content of mineral N readily available after soil application, which gradually increases as a result of organic N mineralization of during more than three months. The increase of the doses represented a significant increase in mineral N in the soil without affecting the efficiency of the mineralization process.

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NITROUS OXIDE EMISSION FACTORS FOR DAIRY COW EXCRETA IN EASTERN CANADA

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In Canada, N2O emissions resulting from the deposition of urine and dung by farm animals on paddocks, ranges and pastures are estimated at 8 Gg N or11.5% of national emissions. Amounts of N2O emitted from soils are generally proportional to N inputs but also depend on soil redox potential and organic carbon availability. Accordingly, several factors affecting soil redox potential such as gaseous diffusion, water content and compaction at the time of urine application to grassland soils have been shown to impact N2O emissions. However, the proportion of urine N lost as N2O was not always clearly related to soil type (Di and Cameron, 2007) or time of year (Šimek et al., 2006). Impacts of time of year and soil type may therefore vary depending on local climatic conditions, management practices, and soil properties. Nearly all studies reporting soil N2O emissions from grazed land were carried out in Western Europe, United Kingdom and New Zealand, and it is unclear how their results apply to other regions. This is a source of uncertainty for the Canadian national inventory of greenhouse gas emissions and in-country measurements are needed to improve Canadian estimates. The objectives of this study were to quantify the N2O emission factors (EF) following deposition of dairy cow urine and dung onto two grassland sites in Eastern Canada and to determine the impact of excreta type, urine-N rate, time of year and soil type on annual emissions.

Materials and Methods

The study was conducted from September 2009 to October 2011 near Québec City, Canada on a clay soil (0.58 g clay g-1; bulk density, 1.05 g cm-3; pH, 5.7) and a sandy loam soil (0.19 g clay g-1; bulk density, 1.27 g cm-3; pH, 5.2). Perennial grasses were grown at both sites and were cut to 0.05 m in May, June, August and October when it reached 0.25 m to simulate periodic grazing. The same experiment was conducted twice on the two soil types, each time with three dates of dejection application. The four treatments were unamended control plots, circular dairy cow 0.1-m2 urine patches at 50 (U50) and 100 (U100) g N m-2, and circular dairy cow 0.1-m2 dungs (17.5 kg FW m-2). Dates of urine and dung applications included spring (31 May 2010 and 6 June 2011), summer (5 July 2010 and 4 July 2011) and fall (28 Sept 2009 and 27 Sept 2010), for a total of six application times on each soil type. Soil-surface N2O fluxes were measured at least once a week during the snow-free periods using non-flow-through non-steady-state chambers (Rochette and Bertrand, 2008). ´

Results and Discussion

Mean EFs for dairy cattle urine were three times greater in the wetter clay (1.06%) than in the dryer sandy loam (0.29%) soil (Fig. 1). This is in agreement with previous reports of increasing EFs in urine-treated soils with decreasing soil aeration in response to differences in texture (Clough et al., 1998), compaction (Uchida et al., 2008), drainage (de Klein et al., 2003) or water content (Thomas et al., 2008). The excreta deposition date did not affect EF on the sandy loam soil but values were greater in summer (1.5%) than in spring (1%) and fall (0.5%) in the clay soil. The greater EF estimates in summer on the clay soil were not related to higher water contents and it is hypothesized that temperature was the main factor controlling N2O production and emission under the relatively humid conditions observed in this soil. Nearly identical mean EFs were observed for both urine rates on the two soils. This indicates that overlap of urine patches that results in increased urine-N load may not result in disproportionate increases in N2O emission and associated biases in estimates based on a unique EF. The mean EF estimates for dung were one order of magnitude smaller than for urine. Application date did not affect the emission factor for dung. However, the impact of soil type on N2O emission following dung application was opposite to that of urine with losses accounting for 0.07% of applied N in the clay soil and 0.15% in the sandy loam soil.

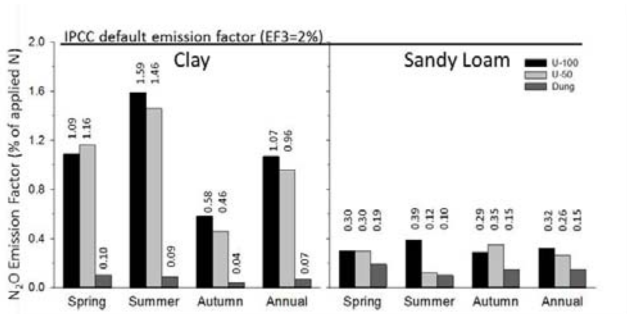

Figure 1. N_2O emission factors following application of dairy cow excreta to two soils. Values are averages of two runs of the experiment.

Conclusions

Our results suggest that the default IPCC EF for total excreta N (2%) overestimates emissions in Eastern Canada. They also indicate that estimation of soil N2O emissions following deposition of excreta by grazing dairy cows in Eastern Canada 1) should account for soil type, 2) should use EFs specific to urine and dung, and 3) could use a unique emission factor for urine rates up to 100 g N m-2.

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FIELD EVALUATION OF THE IMPACT OF BIOCHAR AND COMPOST AMENDMENTS ON NITROUS OXIDE EMISSIONS FROM SOIL

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Agricultural soils are a major source of N2O emissions. Increasing awareness on greenhouse gas emissions due to climate change has focused scientific research on strategies to reduce their release to the atmosphere[1]. Biochar amendment has shown to affect carbon and nitrogen transformation and retention processes in soil. In particular, numerous laboratory studies have shown that biochar could be used as a strategy to mitigate N2O emissions from agricultural soils.
The aim of this experiment was to elucidate the impact of biochar, compost and a mixture of both on N2O emissions under field conditions.

Materials and Methods

The experiment has been performed in an organic olive orchard in South East Spain (coordinates 38°23' N 1°22' W). It is an area with a high insolation rate (200 w/m2), low precipitations (250 mm/year) and the mean annual temperature is 16.25°C.

The soil is a Haplic Calcisol with a loam-sandy texture (16.2% clay; 27.0% silt; 56.8% sand). Compost was made from two-phase olive mill waste and sheep manure (50% volume). The biochar was produced from oak by slow pyrolysis at 650ºC. A mixture of compost and biochar was prepared in a proportion of 90/10 (dry weight). The main chemical properties of the soil, compost, biochar and the mixture are shown in table 1.

Table 1. Chemical properties of the soil, compost, biochar and mixture

	pH	EC (mS/cm)	TOC (%)	TN (%)	TOC/TN
Soil	8.0	0.52	1.68	0.24	7.13
Compost	8.7	2.70	32.87	2.16	15.23
Biochar	9.3	0.57	63.09	0.79	80.16
Mixture	8.9	2.45	33.29	2.02	16.50

EC: Electrical Conductivity; TOC: Total Organic Carbon; TN: Total Nitrogen

The experimental design was a fully randomized split-plot design with four treatments in triplicate: (i) control (no amendment), (ii) compost, (iii) biochar and (iv) the compost/biochar mixture. In all cases the application rate was 20 ton/ha. A buffer line of trees was established between each plot. Amendments were added in the beginning of May, as it is usual in local agricultural practices. Additionally, an organic nitrogen fertilizer (170 kg N ha-1) was applied in July. The static closed chamber method was chosen for the measurements of soil gas fluxes[2]. These chambers were placed on permanently installed PVC-soil collars with 300 mm diameter and sealed with a lid. Gas samples from each chamber headspace were taken after an enclosure period of 60 up to 90 minutes using syringes and transferred to evacuated 12 ml glass vials. During the first four months the gas sampling was performed every ten days. Additional gas samples were taken after precipitation (> 10 mm) and irrigation events.

After the irrigation period, sampling frequency was reduced to monthly measurements in the absence of rainfall events. Denitrifying enzymatic activity (DEA) was evaluated every three months[3].

Results and Discussion

Fluxes of N2O-N were not detected until the first irrigation event (45th day). The highest emission rates were reached by the mixture and compost treatments with values of 49.59 and 40.61 μg N2O-N h-1 m-2 respectively. A new peak was registered after N-fertilization (69th day), and again the mixture treatment reached the highest value of 14.97 μg N2O-N h-1 m-2. Irrigation events were followed by N2O-N emission peaks during summer (Figure 1). After four months N2O-N emissions were negligible and not different to the control soil. Three months after amendment, the denitrifying enzymatic activity (DEA) was significantly higher in the plots with compost and the mixture compared to the control and the biochar amended plots (Figure 2). After six months these two treatments still showed the highest DEA.

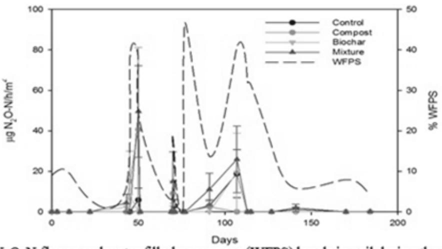

Figure 1. N_2O-N fluxes and water filled pore space (WFPS) levels in soil during the experiment

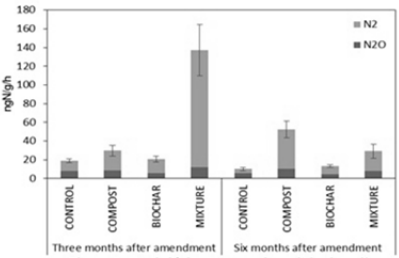

Figure 2. Denitrifying enzymatic activity in soil

Conclusions

N2O-N fluxes were very low even after amendments and nitrogen fertilization, which differs with other field experiments. DEA values were consistent with the N2O-N fluxes registered during the experiment, being the highest values those reached by the mixture and the compost treatments. N2O-N emissions from biochar amended plots did not differ significantly from the controls.

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IMPACTS OF LONG-TERM COMPOST APPLICATION ON SELECTED INDICATORS FOR SOIL ORGANIC MATTER DYNAMICS

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Organic fertilisation with compost contributes to soil fertility by returning organic matter and nutrients to the soil. Compost application helps to save resources, such as energy and raw materials for the production of mineral fertilisers. In Austria, biowaste has been collected separately for more than twenty years. More than 90 % of compost corresponds to the quality class A^+ and A according to the Austrian biowaste ordinance (set into force 1996), which renders this compost suitable for organic farming and agriculture in general. The Austrian Agency for Health and Food Safety has been running a field experiment in Upper Austria on a loamy silt soil since 1991 to investigate the influence of compost application on crop yields and soil parameters and to quantify the plant available nitrogen (N) in composts.

Materials and Methods

The field trial consists of a control plot (zero N), minerally fertilised plots (40 kg N, 80 kg N, 120 kg N $ha^{-1}yr^{-1}$) and plots amended with biowaste compost (from source-separated organic waste, OWC), green waste compost (GWC), cattle manure compost (FYMC) and sewage sludge compost (SSC), each treatment corresponding to 175 kg N ha^{-1}. Additional treatments were composts + 80 kg N (NH_4NO_3) ha^{-1} (OWC, GWC, FYMC, SSC 175N + 80 N). The crop rotation consisted of winter wheat, winter barley, maize and pea (without compost application) since 2003. In former years also spring wheat was included in the crop rotation. Soil samples taken after the harvest 2010 in 0-25 cm soil depth were analysed for soil organic carbon (SOC) and total N (N_t) with elementary analysis after dry combustion. The analysis of the potential N mineralisation with the anaerobic incubation method was done according to Keeney (1982), modified by Kandeler (1993). N_{min} samples were taken in several years in spring (March or April) in 0-90 cm soil depth. Active carbon was determined by $KMnO_4$-oxidation according to Weil et al. (2003). Furthermore, grain yields of winter barley (mean values of 4 years) and the crude protein contents of the corn (= N x 6.25) were investigated.

Results and Discussion

The results of the soil analyses show a significant increase of pH ($p < 0.05$) only with the application of sewage sludge compost. SOC and Nt (except for FYM compost) were significantly enhanced in all compost variants compared to the control in 2010. The Corg to Nt ratio was highest with sewage sludge compost application. This may be due to the fact that in the production process of this compost wood chips and bark had been added as bulking agents. These results were underlined by the findings for active carbon, a dynamic soil organic carbon fraction obtained by oxidation with 0.02M $KMnO_4$. Especially its share in SOC was able to differentiate between the different compost amendments (Tatzber et al., 2014).

N_{min} contents in 0-90 cm soil depth in spring revealed highly significant differences ($p < 0.01$) between the years of investigation and depended on the crop type. The highest N_{min} contents occurred with maize and winter wheat. N_{min} contents in the compost variants rose in the course of the years. Twelve years after the beginning of the experiments and thereafter the compost variants showed N_{min} contents as high as or higher than the highest mineral fertilisation rates. This fact was partly underlined with results of the N mineralisation potential measurements.

The grain yields of winter barley and the crude protein contents of the corn did not show significant differences between all compost treatments (correspondent to 175 kg N). The yields of the compost variants with additional mineral N proved to be significant and the crude protein contents slightly higher compared with exclusive compost application. However, the highest crude protein contents in winter barley occurred with 120 kg mineral N fertilisation.

The compost N utilisation by the crop derived from the yield curve depended on the crop and highly varied between the years of investigation. 57 kg compost N was available for maize, 35 kg N for spring wheat and 28 kg N for winter barley on the average of 4 to 6 crop yields, respectively. Pea apparently could not use any additional N supply (neither mineral nor compost N). In summary, the plant availability of compost N was rather low, at an average level of 27%.

Conclusions and perspectives

SOC and Nt contents in the soil rose – in most cases – significantly with long-term compost application compared to mineral N fertilisation. These changes were also reflected by the $KMnO_4$-determination of active carbon, indicating a dynamic SOC fraction. The development of plant available nutrients corresponded to the input by the composts. Consequently, compost application helps to save valuable resources. On the other hand, the development of available nutrients in the soil (and the composts) should be observed to avoid environmental impacts, especially with N (and P). As expected, compost proved to act as a slow release N source.This contribution will also highlight N dynamics in the soil – plant system after ending the application of compost.

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SHORT-TERM CARBON DIOXIDE AND NITROUS OXIDE EMISSIONS AND CHANGES IN PHYSICO-CHEMICAL PROPERTIES FROM SOILS AMENDED WITH DIFFERENT BIOCHARS

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Increased concentration of greenhouse gases in the atmosphere has drawn world's attention to look for novel technologies to combat with climate change and global warming. Biochar is considered as an important soil amendment as it improves soil productivity, carbon storage, infiltration and water holding capacity of soil [1] and has been shown to mitigate greenhouse gas emission in the field [2] through increased carbon sequestration and reduced nitrous oxide emissions. The uses of biochar as soil amendment modify the dynamic of carbon (C) and nitrogen (N) in soil after the application of fertilizers [3, 4]. The adsorption of nutrients and dissolved organic carbon (DOC) in biochar is fully associated with increased cation exchange capacity and surface area, and the acidic functional groups attached to the surface [3, 5]. Some researchers have found out that biochar can reduce N losses due to leaching, also reduce heavy metal contamination in soil solution [4, 6] and thereby enhancing crop quality. Soil N immobilization can be expected due to high C:N ratio of biochar [7]. The stability of biochar and its interaction with soil environment considerably varies with nature of feedstock, method of production (e.g. gasification, pyrolysis), residence time, heating rates and final process temperature [1]. A laboratory incubation experiment was carried out with a hypotheses stating that CO_2 and N_2O emission patterns from biochar amended soils are closely related to process conditions (pyrolysis and gasification), process temperature and properties of feedstock (manure and wood biomass) used.

Material and Methods

The experiment was carried out in glass jars (3 L) intended to represent the plough layer (0-30 cm) of two agricultural soils (NW Italy) with different physico-chemical characteristics: 1) a sandy soil (pH: 8.3, O.C.: 0.52%, N: 0.057%) and 2) a silt-loam soil (pH: 6.2, O.C.: 1.2%, N: 0.15%). The air-dried soil samples (1.5 kg for each replication, sieved at 2 mm) were amended (at 2% w/w) with five different types of biochars (PL- poultry litter and SM- swine manure biochar, both at 400 and 600 °C; and WC- wood chip biochar from gasification at 1000 °C), 4 replications per each treatment and incubated for 85 days (Table 1). De-ionized water was added to the soils in order to get the desired water filled pore space of 75%. After 20 days of incubation, all jars were fertilized with ammonium nitrate at a rate of 170 kg N ha-1. The measurements of CO_2 and N_2O were performed at different sampling dates using gas chromatography. The whole experiment was carried out in a randomized block design within a climatic chamber with controlled environment (20 ºC and 60% RH).

Results and discussion

Results from cumulative emissions show that soils amended with poultry litter biochar at 400 ºC emitted higher CO_2 and N_2O whereas wood chip biochar emitted significantly less than the rest of the treatments (Figure 1). The emission patterns are well related to the specific characteristics of biochars [4, 8]. In general, chars produced at higher temperature emitted less than at lower temperature. This could be the fact that chars produced at lower temperature contain higher percentage of labile carbon which is easily attacked by the soil microbes. This supports the previous findings of [2].

Biochar type	Total C	Total N	Ash	pH	surface area
	%	%	%		$m^2 g^{-1}$
PL 400	52.3	6.7	26.9	8	6.1
PL 600	50.3	4.1	37.6	11	6.3
SM 400	50.6	2.4	33.8	9	5.1
SM 600	57.8	1.8	35.3	9.9	13.2

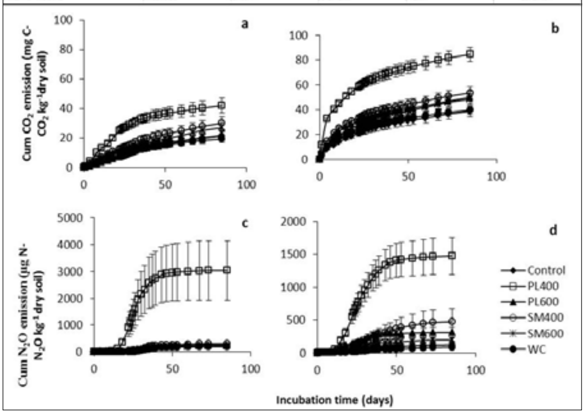

Figure 1. Cumulative CO_2 and N_2O emissions from sandy soils (plot a, c) and silt-loam soils (plot b, d) amended with different biochars during 85 days of incubation. Fertilization with ammonium nitrate starts after 20 days of incubation. PL400, PL600- poultry litter biochar at 400 and 600 °C; SM400, SM600- swine manure biochar at 400 and 600 °C; WC- wood chip biochar at 1000 °C. Error bars represent standard errors (n=4).

Conclusion and perspectives

The total cumulative emissions (CO_2 and N_2O) were strongly related to pyrolysis temperature as well as soil types. Pyrolysis conditions during biochar production process, quality and nature of feedstock seem to be important parameters in determining the gaseous emission patterns when used as soil conditioner. In depth research study on processes and mechanisms associated to these effects are necessary for better understanding and also exploring future use of biochar properties in order to meet the new mitigation strategies.

Acknowledgement

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NITROGEN ISOTOPIC COMPOSITION OF N-SOURCES, VEGETABLES AND SOIL FROM TWO ORGANIC FARMING SYSTEMS

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Crops and agricultural soils tend to show variations in $\delta^{15}N$ values regarding N-fertilizers types having different $\delta^{15}N$ signatures (Yun et al., 2006; Lim et al., 2010). Thus, the $\delta^{15}N$ signature of plants has been suggested as a tool to differentiate mode of production i.e. conventional vs. organic, despite some confounding factors (Inácio et al., 2013). The aim of this study was to elucidate factors affecting the $\delta^{15}N$ values of vegetables grown in two distinct organic systems.

Materials and Methods

Samples of soil, lettuce (Lactuca sativa) and organic inputs (composts, manures, cover crops) were taken from two distinct organic farms in Rio de Janeiro State, Brazil. Farm Org. A is in-conversion from a conventional to an organic system, and Org.B is a three-year organic farm. The different organic inputs (N-sources) used by each farm are shown in Table 1. A combustion elemental analyzer (EA) and an isotope ratio mass spectrometer (IRMS) were used to determine total N and C (%) and $\delta^{15}N$ (‰) values of dried and finely ground samples, respectively.

Results and Discussion

Farms Org.A and Org.B differed markedly in terms of $\delta^{15}N$ values of organic inputs (Table1), plant (lettuce) and soil (Fig. 1). Plant and soil $\delta^{15}N$ values of farm Org.A increased during the first year of conversion to organic farming reflecting the $\delta^{15}N$ values of compost applied to soil (Fig. 1). The $\delta^{15}N$ signatures of manure also varied during the 12-months and the compost signature reflected this variation of the primary fertilizer source. Farm Org.A showed higher $\delta^{15}N$ values for lettuce than farm Org.B due to the differences of $\delta^{15}N$ values of N-sources applied. However, lettuce $\delta^{15}N$ values in farm Org.B suggest two hypothesis: (i) legumes used as mulching or green manure, with high BFN inputs seem not to be the main N-source despite its high N-content, while other N-sources were available with higher $\delta^{15}N$ signatures, such as fermented product (Bokashi), or (ii) the management of legume biomass might be favouring N loss e.g. by NH_3 volatilization and/or N_2O/N_2 emissions, which are ^{15}N fractionation processes that leads to the enrichment of available N in soil. These hypotheses will be tested by controlled experiments in which different N-sources will be individual treatments.

Conclusions

The array of N-sources available for different organic farms shows a wide range of $\delta^{15}N$ signatures (from +0.0 to +14.9‰). However, organic amendments and fertilizers (manure, composts and fermented products) seem to influence the $\delta^{15}N$ signatures of plants and soil more than BNF inputs (legumes mulching and green manure), which might imprint lower $\delta^{15}N$ values in vegetables than were found.

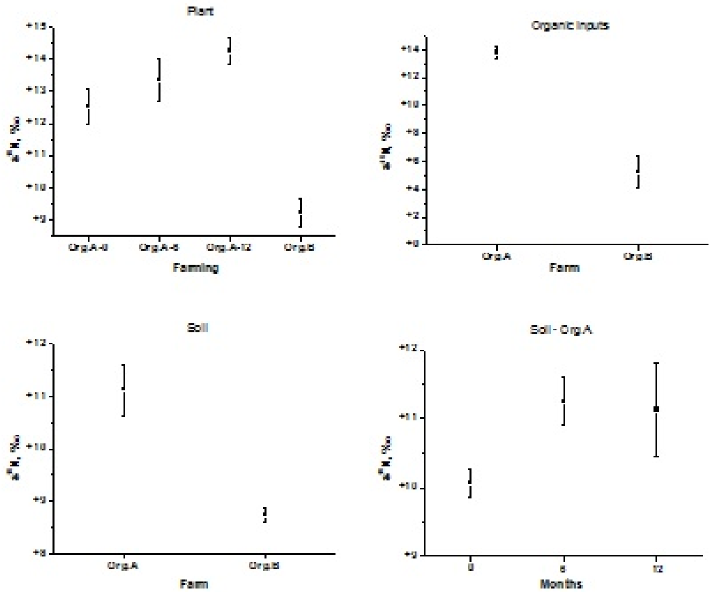

Figure 1. Mean and standard error bars of nitrogen isotopic composition ($\delta^{15}N$) of lettuce (*Lactuca sativa*), organic inputs (manures, compost, cover crops) and soil samples of two organic farms. Org.A is an in-conversion organic farm (first year) based on the use of manure and compost as N-sources. Org.B is a three-year organic farm based on legumes and composting of plant residues without the use of manure. Org.A-0, -6 and -12 represent months of organic farming.

Table 1. Nitrogen content and isotopic composition of organic inputs from two organic farming systems in Rio de Janeiro State, Brazil.

Farm	Input[a]	n	N-total (%)	$\delta^{15}N$ (‰)
Org.A	Horse manure	2	1.8 and 1.2	+11.8 and +14.6
	Poultry manure	1	3.0	+13.9
	Compost	3	1.7	+13.1 to +14.9
Org.B	Gliricidia	1	4.5	+0.0
	Mucuna	1	6.2	+2.5
	Crotalaria	1	4.6	+1.4
	Elephant grass	1	0.6	+4.3
	Maize	1	3.0	+10.0
	Fermented product	1	4.0	+7.9
	Vermicompost	1	2.7	+8.8
	Mulch	2	3.6	+6.5 and +7.6
	Green-manure	2	2.1	+2.2 and+5.4

[a]Horse manure, samples of March and September-2012; Compost made from manures + vegetables residues + straw, samples of March/July-2012 and March-2013; Fermented product made from wheat bran + castor bean mill cake; Vermicompost made from dairy manure; Mulch is a mixture of chopped residues of Gliricidia (*Gliricidia sepium*) + Elephant grass (*Pennisetum purpureum*); Green-manure is a mixture of Mucuna (*Mucuna aterrima*) + Maize residues incorporated into soil at the end of summer before vegetables growing.

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Silmara Bianchi and Renato Moutinho who analyzed the samples by EA and by IRMS, respectively.

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PART OF GRASSLAND IN LEY-ARABLE ROTATIONS IS A PROXY FOR PREDICTING LONG- TERM SOIL ORGANIC MATTER DYNAMICS

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Ley-arable rotations, common in dairy systems, have long been known to ensure good productivity and limit the decrease of soil organic matter (SOM) frequently observed in permanent arable land (Conijn *et al*., 2002). This paper deals with the long-term effect on SOM of rotations differing in the ratio of grass to grass+crops (G/G+C), to answer the following questions: how does the G/G+C ratio affect SOM? Which combination could ensure carbon storage? Can SOM dynamics be predicted with a simple model previously evaluated for arable crops?

Material and methods

Fodder ley-arable rotations were studied in a 27-year experiment set up in western brittany, france, on a loamy sand and slightly acidic soil in oceanic conditions (1110 mm rainfall, 11.8°c mean temperature). eight of them combined maize (silage) with grass (cut): continuous maize, maize/*lolium multiflorum l.* for 6 or 18 months, maize/*lolium perenne* l. for 3.5 years, or permanent *l. perenne* and *festuca arundinacea* l. the other rotations were biennial, including maize and *l.multiflorum* as a catch crop after wheat, barley or grain legumes (details in simon, 1992). their g/g+c ratios, calculated as the relative duration of both crop types, varied from 0 to 1.

The topsoil (0-25 cm) was sampled 7 times during the 27 years and analysed for total n and c (dumas). Soil density was measured at the beginning and end of the trial.

Data were used to test the amg model (andriulo *et al.*, 1999), which simulates the evolution of humified om on an annual time step, assuming that som consists in a stable and an active pool. Its parameters are the initial size of the active fraction, the annual c input, the humification rate of fresh residues and the mineralisation rate of the active fraction. Amounts of annual c inputs derived from above- and below-ground plant residues, slurry and rhizodeposits to soil were estimated from the literature (e.g. Nguyen, 2003; whitehead *et al.*, 1990) or measured data.

Results and discussion

Final soil status: after nearly 30 years of constant practices, c and n stocks had decreased by 3% (permanent grass) to 30% (continuous maize) (fig. 1a) (observed data). Initial soil organic carbon (soc) was 88 (±3) t c.ha-1, while final soc varied from 65-85 t c.ha-1 for continouous maize and permanent grass respectively. We determined 4 groups for the modelling approach, g1 to g4, that varied in their g/g+c ratios: 0.00, 0.56, 0.76 and >0.90, respectively. Values used for c inputs (all in t c.ha-1.yr-1) were 0.77 (grass and slurry, c3 inputs) and 1.82 (maize, c4 inputs) for g1, 1.82 (c3) and 1.82 (c4) for g2, 3.50 (c3) and 0.88 (c4) for g3 and 3.96 (c3) and 0.43 (c4) for g4 (Vertès and Mary, 2007).

Long-term SOC dynamics: The simulation was run first using standard parameters calibrated with arable crops (Saffih and Mary, 2007). The model predicted a decrease in SOC in all situations and provided satisfactory simulation of SOC in rotations dominated by crops (G/G+C < 0.6) but overestimated (by about 11%) the

SOC decrease in grass-dominated rotations (Vertès and Mary, 2007). The C inputs from grasslands were then calculated to obtain a good prediction of SOM dynamics in all cases (Fig. 1a), which led to optimal values of about 7 t C $ha^{-1}.yr^{-1}$ (Fig. 1b) for grass, corresponding to the highest values reported in the literature (e.g. Paustian *et al.*,1990).

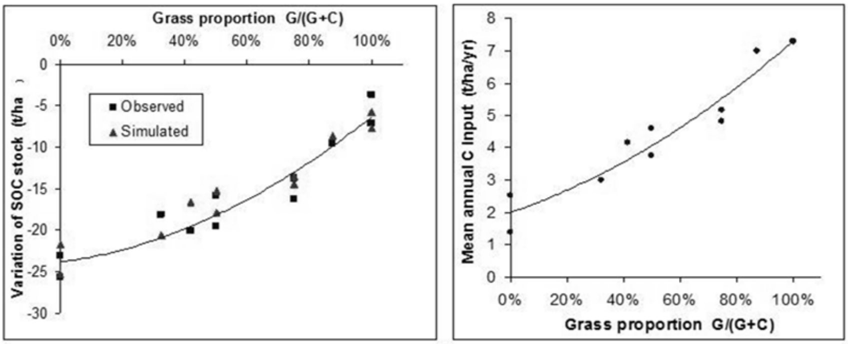

Figure 1: effects of the grass/grass+crop ratio on a) long-term soil organic matter evolution in a ley-arable rotation trial and b) optimal mean annual c inputs

In our experimental conditions, a large decrease in SOM was observed in arable rotations, and even permanent grassland barely ensured the stability of SOC. The two main factors explaining this result are i) the high initial SOM content, which favours high mineralisation rates, as assessed by measurement in incubated soils at the end of the experiment (data not shown) and ii) the lower C restitutions in cut compared to grazed grasslands, where about 75-95% of ingested organic matter returns to soil as dung (and urine). Final SOM status was consistent with local statistical references (Simon et al., 1992) for common rotations, where a similar decline was observed, especially in soils with high initial SOM content. The AMG model was able to satisfactory predict SOC evolution in crop-dominated rotations and predicted that SOC is not at equilibrium after nearly 30 years of constant agricultural practices. The large uncertainties about C inputs from grassland may explain why the model overestimated SOM decline in grass-dominated rotations: reliable data are crucial to evaluate simulation models that predict SOM changes, including transitions from arable crops to grasslands and vice-versa. Finally, changes in SOM quality (data not shown) led to predictions that C and N mineralisation rates should be more than proportional to the total C and N contents of soil (Accoe *et al.*, 2004).

Conclusions and perspectives

The G/G+C ratio was shown to be a good proxy to explain observed long-term SOM dynamics and could be used in SOM-evolution modelling and determination of optimum combinations. Although no net C (or N) storage was observed in either simulated or experimental conditions, a hierarchy between rotations based on this ratio was in agreement with main conclusions from the literature (Soussana *et al.*, 2010) about the positive effect of grasslands on SOM dynamics.

COMPARISON OF MICROMETEOROLOGICAL TECHNIQUES FOR ESTIMATING AMMONIA EMISSION FROM COVERED SLURRY STORAGE AND LANDSPREADING OF CATTLE SLURRY

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The micrometeorological mass-balance integrated horizontal flux (IHF) technique has been commonly employed for measuring ammonia (NH_3) emissions in on-field experiments. However, the inverse-dispersion modeling technique, such as the backward Lagrangian stochastic (bLS) modeling approach, is currently highlighted as offering flexibility in plot design and requiring a minimum number of samplers (Ro et al., 2013). The objective of this study was to make a comparison between the bLS technique with the IHF technique for estimating NH_3 emission from flexible bag storage and following landspreading of dairy cattle slurry. Moreover, considering that NH_3 emission in storage could have been non uniform, the effect on bLS estimates of a single point and multiple downwind concentration measurements was tested, as proposed by Sanz et al. (2010).

In the current study, two experiments were performed in northern Spain: (I) in an outdoor flexible bag storage system and (II) in a plot. In study I, 2,000 m^3 slurry were stored into an impermeable plastic bag and produced biogas was discharged to the atmosphere through 6 chimneys located at the centre of the lagoon. One vertical mast was placed in each side of the storage, which was considered as upwind or downwind depending on main wind direction recorded during the exposure session. Masts supported five passive flux samplers (PFS) coated with oxalic acid (Leuning et al., 1985) and mounted logarithmically at heights of 0.30, 0.48, 1.00, 2.05 and 3.05 m above the soil surface. Furthermore, for single (S) and multiple downwind points (M) testing, two more masts (PFS at 1.00 m height) were placed in each side of the storage. Samplers were replaced every day during 10 measuring days and were immediately transported to the laboratory to be leached with 60 ml of deionised water and analyzed for NH_4^+-N by spectrophotometry. In study II, 145 m^3 slurry from storage were applied to 2 ha land at 323 kg N ha^{-1} by means of a tanker fitted with trailing-hoses. One day after slurry application, the soil was turned with a plough machine. A mast was placed towards downwind of the plot supporting four PFS placed at 0.48, 1.00, 2.05 and 3.05 m height and were changed daily during 3 days before fertilization and 2.5, 9.5, 22.5 h after slurry application. Thereafter, they were changed daily during the following 4 days. For study I and II, a background mast was located 150 m upwind from the storage and fertilized soil. For NH_3 emission determination, the IHF method was used as reference technique. On the other hand, commercial software (Windtrax 2.0, Thunder Beach Scientific, Canada) was used for the determination of NH_3 emission by applying the bLS model. Inputs within measurement periods were average wind speed at heights of 0.30, 0.48, 1.00 and 2.70 m, wind direction at 2.70 m, air temperature, the default surface roughness length for base soils (z_0 = 0.01 m) and NH_3 concentration (µg N m^{-3}). A post-data filtering criteria was performed (friction velocity, $u^* \geq 0.15$ m s^{-1}; Obukhov stability length, $|L| \geq 10$ m) in order to avoid extreme atmospheric stability situations (Ro et al., 2013).

Ammonia emissions estimated by the IHF technique in storage and land (Table 1) were considered low compared to previous studies (VanderZaag et al., 2010; Sanz et al., 2010), indicating that flexible bag storage and slurry application to soil through narrow bands followed by incorporation to soil could have mitigated NH_3 emissions in our research. In study I, NH_3 estimates in M were higher and presented better plot coverage (higher footprints) than in S (Table 1). The differences observed in NH_3 estimates between bLS and IHF techniques were considerably greater in the current experiments than those reported elsewhere by other authors. For instance, Sommer et al. (2005) observed NH_3 estimates from bLS averaged within 16-24% of the IHF estimates in land, whereas Sanz et al. (2010) reported bLS estimates averaging within 5-10% of the IHF estimates after pig slurry application. Our reduced accuracy was attributed to two factors: (I) the low NH_3 emissions observed through both techniques could have lowered the accuracy of the ratio between bLS and IHF estimates and (II) the low footprints observed in the bLS case could indicate that average wind direction within 24 h period did not allow the NH_3 emission plume to be effectively captured by PFS when using Windtrax 2.0, resulting in uncertain emission estimates under changing wind directions.

Table 1. Comparison of mean NH_3 emission from storage (study I), NH_3 emission at peaking time from land (study II) and cumulative NH_3 emissions and footprints by bLS and IHF techniques.

Study	Downwind point	Mean $NH_{3\,bLS}$ ($mg\ N\ m^{-2}\ d^{-1}$)	Mean $NH_{3\,IHF}$ ($mg\ N\ m^{-2}\ d^{-1}$)	Cumulative $NH_{3\,bLS}$ ($mg\ N\ m^{-2}$)	Cumulative $NH_{3\,IHF}$ ($mg\ N\ m^{-2}$)
I	S	0.12 (0.25)†	14.17 (28.76)†	0.93 (9-14 %)*	157.05
	M	0.34 (0.29)†		2.58 (27-36 %)*	
		$NH_{3\,bLS}$ peak ($g\ N\ ha^{-1}\ h^{-1}$)	$NH_{3\,IHF}$ peak ($g\ N\ ha^{-1}\ h^{-1}$)	Cumulative $NH_{3\,bLS}$ ($kg\ N\ ha^{-1}$)	Cumulative $NH_{3\,IHF}$ ($kg\ N\ ha^{-1}$)
II	S	7.26	271.45	0.10 (1-27 %)*	6.77

†Mean (standard deviation). *Footprint (min-max %): fraction of the source area where emissions were "measured" by the sensor.

In conclusion, ammonia emissions from slurry management were lower than expected, possibly due to convergence of NH_3 abatement strategies, in both the storage and fertilization stages, and insufficient plot coverage by PFS. Under non uniform NH_3 emissions, the use of multiple downwind points resulted in more accurate estimates and better plot coverage compared to a single point. Results from the current study indicated that 24 hours average concentration and wind direction values could result in uncertain bLS estimates under changing wind directions.

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DEVELOPMENT OF A MODELLING FRAMEWORK OF NITROUS OXIDE FOR BIOGAS DIGESTATE USED AS FERTILISER

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A major challenge in manure management is the sheer volume, odor and health risks posed by raw manure, and the need to safely dispose of this resource as it accumulates. The anaerobic digestion (AD) is to use animal manure and organic waste as feedstock to produce biogas. A number of studies (Moller and Walter, 2009a and 2009b) indicated significant changes in slurry composition through AD. So this processed manure by AD is known as the digestate which is seed and pathogen free without the odor by the time it exits the digester. Moreover, the digestate still has chemical and nutrient value because the digestate from the AD consumes only the carbon. The nitrogen, phosphorus, potassium and micronutrient components remain intact as an organic fertilizer. However, while the digestate is used as land fertilisers, CO2 and CH4 emissions are reduced greatly but N2O emissions may increase due to high N concentration. Thus, a systematic assessment is desirable for the digestate used as fertiliser. The objectives of this study are 1) to evaluate the potential for recycling nutrients of digestate in terms of nitrogen utilisation and losses to air and water, and 2) assess the potential of integrating AD into agricultural systems to improve nutrient efficiency and environmental value at the field scale.

Modelling Methods

Since the plant uptake of N from digested manures may be expected to be closer to that of commercial fertilisers, a modification of DNDC (Li, 1992a and 1992b; Wang et al, 2012) is made for digestate used as fertiliser. Digestate is inputted into soils, based on concept model of carbon and nitrogen pools, in which its C and N is distributed into relative pools of C and N (Fig.1). Four interacting submodels of DNDC: thermo-hydraulic, denitrification, decomposition, and plant growth are used to couple the abiotic and biotic drivers of carbon and nitrogen dynamics in soils. The plant growth submodel calculates daily root respiration, nitrogen and water uptake by vegetation, and growth rates are dependent on photosynthesis, water and nutrient limitation under climate and soil status.

Results and Discussion

The model was tested against sampling sites of Cae Banadl at Aberystwyth in Wales. The site plots were grazed by cattle. Further details of sites can be found in Cardenas et al. 2010 and Wang et al., 2012. Figure 2 shows a comparison of N2O emissions from different quantities of digestate applications at C/N =5. The modified DNDC model has captured successfully the effect of the digestate applications. The N2O emissions increase as the quantities of the digestate increase. Fig. 3 shows a comparison of N2O emissions between the quantities of synthetic fertiliser (NH4NO3) and digestate applications with different ratios of C to N. It can be seen that digestate does affect the emission of N2O. After C/N ratio is greater than a value, N2O emissions of the digestate is much lower than that of fertiliser. A similar emission is found at C/N=5 and =10. However, after a deeper digestion, the emissions of the digestate will be the same order of magnitude as the fertiliser. If C/N ratio reaches a value (e.g. 0.5), N2O emissions of the digestate is a little greater than that of fertiliser. This is in agreement with the previous studies (Moller and Stinner, 2009a and 2009b).

Conclusions

A process-based model based on DNDC has been developed for modelling of the digestate used for fertiliser to compare emissions of N2O between fertiliser, digestate and manure applications. The results show that emissions of N2O of the digestate increase as the C/N ratio decreases. The emissions of N2O can be greater than that of the fertiliser while C/N ratio is small (=0.5) after deeper digestion of raw manure. However, the emissions can be much lower than that of the fertiliser while C/N > 5. This implies that the digestate does not cause a significant increase of N2O emissions. However, further studies would be required to take account of other factors such as pH, dry matters, different slurry application techniques and CO2 emission.

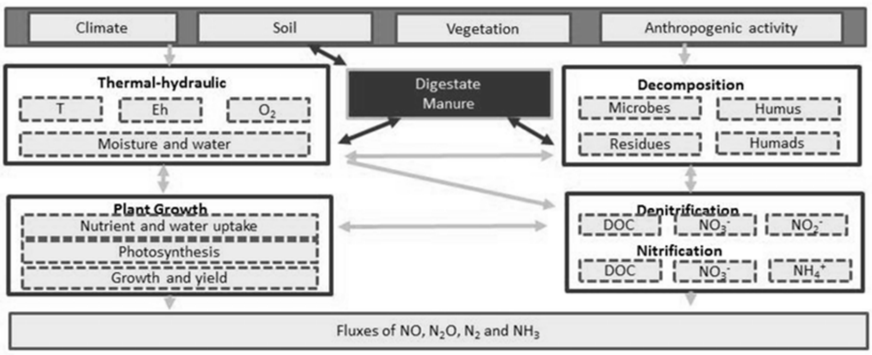

Fig.1 Schematic representation of digestate amendment

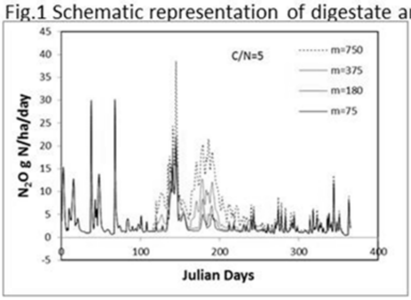

Fig. 2 N2O emissions of different quantities, m, (kg) of digestate applications at C/N = 5

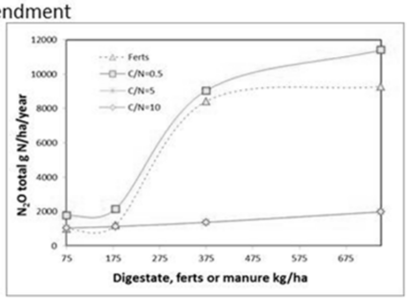

Fig. 3 A comparison of N_2O emissions between fertiliser and digestate with different C/N ratio

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NITROGEN DYNAMICS IN AN UPLAND RICE-STYLOSANTHES GUIANENSIS BASED CONSERVATION AGRICULTURE SYSTEM IN THE MIDWEST HIGHLANDS OF MADAGASCAR

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In Madagascar rice is a staple food. Most fertile lowlands are occupied by paddy rice. To meet growing food demand rainfed rice production has been extended onto the less fertile uplands called *tanety*. Most soils on the *tanety* are ferralsols characterized by low pH, low organic matter, nitrogen (N) and available phosphorus (P) content. Cultivating the latter sustainably is challenging. Smallholders have limited inputs. Since more than a decade conservation agriculture has been proposed to Malagasy small-scale farmers as a way to sustainably grow crops in the uplands. Conservation agriculture (CA) encompasses three agronomic practices: minimal soil disturbance, permanent soil cover and crop rotation (FAO, 2008). One leguminous plant suggested in local upland rice rotations is *Stylosanthes guianensis* (*Stylo*). Besides N_2-fixation, *Stylo* can produce high quantities of shoot biomass and has allelopathic effects on weeds such as *Striga asiatica*. However, till now adoption rates of CA on Madagascar have been low and the feasibility of CA for small-scale farmers has been questioned (Giller et al., 2009). Possible reasons of the low adoption could be the difficulty to attain a sufficiently high *Stylo* biomass production, limited landholding area per farmer, or an initial yield decrease under CA. Initial yield decreases may be even more pronounced in these low fertile soils following N immobilization. So far, most studies on CA have focused on yields and rice phenology as affected by climate change, weed pressure and erosion control. Yet, little research has been done on the N and P flows in CA systems to understand the potential and constraints of CA for small-scale farming in the Malagasy highlands. We examined in a researcher-managed field experiment the effect of soil preparation (direct seeding vs. tillage) and *Stylo* aboveground biomass management (mulch/removal) on upland rice yield and N and P dynamics. Regarding N our specific objectives were: to quantify N uptake and fertilizer use efficiency by upland rice from organic and mineral N sources; to quantify symbiotic N_2 fixation by *Stylo;* to establish the N balance of our crop rotation.

Materials and methods

A three-year field trial was installed in Ivory in the district of Mandoto, region of Vakinankaratra, at 900 m altitude, on a ferralsol that is representative for the region of the Midwest Malagasy Highlands. The crop rotation encompassed upland rice (NERICA 4) and *Stylosanthes guianensis* (CIAT 184). In season one rice and *Stylo* were inter-cropped. After the first rice harvest *Stylo* was left as fallow during season two. In season three *Stylo* was cut, removed or applied as mulch, and rice was grown. In season one and three, treatments were no fertilizer, cattle manure (5 t ha^{-1}) or mineral fertilizer (N, P, K: 70, 29, 40 kg ha^{-1} and urea: 37 kg N ha^{-1}) application. In season three, two different soil and residue management practices were applied, i.e., direct seeding into *Stylo* mulch (9 t ha^{-1}) or tillage and *Stylo* aboveground biomass removal. Micro-plots were installed to assess N uptake and fertilizer use efficiency by upland rice. Direct ^{15}N isotope-labelling was used to trace the fate of N added as mineral fertilizer and *Stylo*, while an indirect ^{15}N isotope-labelling technique was applied to trace the fate of N added with manure (Oberson et al., 2007). To quantify symbiotic N_2 fixation of *Stylo* the natural abundance technique was used (Unkovich, 2008)

Results, discussion and conclusions

First results show that, *Stylo* produced biennial an aboveground dry matter of 9 t ha^{-1} and had an atmospheric N_2 fixation rate of 50%, which results in an N input of 90 kg N $ha^{-1}a^{-1}$ by *Stylo* aboveground parts. Manure and mineral fertilizer inputs increased rice grain yields as compared to the non-amended control. The application of combined soil and residue management practices (tillage-removal vs. direct sowing/mulch) has not yet significantly affected rice grain yields. We could not see any N immobilization following the inputs of *Stylo* biomass. Overall increased rice N uptake in season three as compared to season one suggests increased mineral N contents in the soil, in spite that microbial biomass and mineral N dynamics in season three showed no significant treatment effects. The latter may result from the high variability within the data, which may be characteristic in the early transition phase of our rice-*Stylo* rotation. Lastly, upland rice production in Madagascar using CA practices including *Stylo* leads to a positive N balance during the first year of conversion. The project is on going and samples from the applied isotopic methods are being processed and analysed. Results from the micro-plot study using ^{15}N techniques to determine N use efficiency by rice and soil mineral and microbial biomass N dynamics will be presented and discussed at the N workshop.

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The project was funded by the Swiss National Science Foundation (SNF) and the Swiss Agency for Development and Cooperation (SDC).

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SUSTAINABILITY OF PRODUCTS FROM MANURE

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Manure processing is receiving an increasing interest in the Netherlands and in other EU countries or regions with an animal manure surplus. It is one of the options to reduce the surplus of manure minerals. The products are claimed to be more nutrient-efficient than unprocessed manure.

The ideas behind manure processing are (i) fractionating mineral nitrogen and phosphorous together with organic N in a liquid and a solid fraction, respectively, (ii) upgrading (concentrating) the liquid fraction in order to reduce transport costs, and (iii) characterization of the concentrated liquid fraction as a chemical fertilizer. If the latter would be acknowledged by the European Commission, the concentrate no longer needs to be regarded as an animal manure product. This would open the opportunity to apply amounts of concentrate representing more than 170 kg of nitrogen, the current EU-limit for animal manure application.

The proposed processes are generally: manure separation into a solid and a liquid fraction, sometimes in combination with anaerobic digestion, followed by upgrading (i.e. concentrating) the liquid fraction using ultrafiltration and reversed osmosis.

However, how sustainable is the application of the resulting products in terms of: energy consumption, greenhouse gas (GHG) emission, financial costs and fertilizer efficiency and which product or combination of products are suitable for Dutch arable farmers?

Manure processing

Although different methods of manure processing exist, most of them follow in general the following procedure:

1. Manure separation into a solid and liquid fraction; or in case of anaerobic digestion of manure or manure with co-substrates: Digestate separation into a solid and liquid fraction
2. Direct application of the solid fraction or application after drying
3. Further upgrading of the liquid fraction by filtration (ultrafiltration) producing a filtrate and a permeate, the filtrate returns to the liquid fraction and/or reversed osmosis of the permeate, producing a concentrate and water
4. Transport and application of the concentrate

All these steps require energy, except anaerobic digestion, which produces energy in the form of biogas.

The following products have been assessed to investigate their sustainability and suitability: cattle slurry, pig slurry, cattle- and pig-slurry after anaerobic digestion with maize as raw product and after processing.

The energy- and GHG-sustainability of the application of the solid fraction, the liquid fraction and concentrated liquid fractions of cattle and pig slurry, either with or without anaerobic digestion and with or without the application of co-substrates, was studied using a simplified LCA (Zwart and Kuikman, 2011).

The suitability of these products was assessed for different farming systems in each the five most important Dutch arable farming regions (3 at clay soils, 2 at sandy soils). Suitability criteria were: crop rotation, crop nutrient requirements, relative nutrient concentration in the manure-derived products, legislative aspects regarding the application of animal manure end chemical fertilizers.

The summarized conclusions are:

1. Liquid fractions and concentrates have a 25-100 fold lower N concentration than chemical fertilizer
2. Concentrates have a lower N-efficiency than chemical fertilizer
3. Manure processing results in a reduction of GHG emissions, due to a reduction in storage period
4. The GHG emission of raw slurries or FYM is per kg N-eff, higher than that of chemical fertilizer
5. Raw slurry processing increases the GHG emission per kg N-eff
6. In combination with anaerobic digestion the overall GHG-emission of processing becomes negative due to the potential saving of fossil energy GHG and reduction of GHG during slurry storage
7. At clay-bound farms, on average, 1/3rd of the effective N originated from raw manure, concentrate and chemical fertilizer each
8. At sandy soil bound farms the portioning was 35%, 50% and 15% for each of these sources, respectively

Aknowledgements

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Topic 3

Controlling reactive nitrogen losses

A DECISION SUPPORT SYSTEM FOR ENVIRONMENTAL STRATEGIES: EFFECT IN FARM N BALANCE

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Nitrogen (N) is a key element through farm production systems because of its major role for animal and crop production and the increasing environmental concerns [1]. The N retained by livestock is around 30-40% of the total ingested. The rest is excreted and, therefore, susceptible of being volatilized, leached or lost by direct runoff. In addition, economical balances of the farms can be improved through increasing the efficiency of farm N balance. However, it is still necessary to determine synergistic and appropriate mitigation options to efficiently reduce the environmental pollution from intensive livestock production. These mitigation strategies must be selected on the basis of their abatement potential and cost-effectiveness along the whole production process. Within Batfarm project, we present a novel tool to assess the mitigation potential of nitrous oxide (N_2O) and ammonia (NH_3) losses as a consequence of different strategies and techniques implemented in intensive cattle, pig and poultry farms.

Materials and Methods

Emission factors and water and energy consumptions from different mitigation strategies implemented on farms have been incorporated in the Batfarm software data base [2] [3]. Some of these data also come from on-farm measurements obtained at regional scale. Default values, which are modifiable by the user, have been included to develop versatile and user-friendly software. Regionalizable zootechnical data, climatic information and emission factors have been defined in order to simulate different situations within Portugal, Spain, France, UK and Ireland. As a result, N balance is calculated throughout the different stages of the animal production system (housing, storage, treatment and landspreading). All calculations are performed on a cumulative monthly and annual basis. With regard to the information related to economical feasibility of implementing the techniques selected, users will have the option of introducing economical parameters (investment cost, life of investment, interest rate, variable costs) to calculate the cost of particular strategies adopted for each case of installation. We present a case study for a farrow to finish swine farm which includes the production of weaner and fattening pigs. The farm is located in Spain under Continental Mediterranean conditions (annual mean temperature, 14ºC; annual rainfall, 354 mm, annual mean wind speed 0.7 m/s and annual mean relative humidity 63.4%). The number of sows housed (including gilts) is 270, and there are 1050 weaner places (from 5 to 22 kg) and 1200 places for grower-finishing pigs (from 22 to 110 kg). Two scenarios are compared in this case study: standard situation *vs* a situation where BATs have been implemented (Biphase feeding with synthetic aminoacids in grower-finishing pigs, frequent slurry removal every 2 weeks in grower-finishing pig houses and landspreading using trailing hose). In both scenarios, all the slurry produced in the farm is spread in the same type of crops after being stored in a single tank.

Results and Discussion

Main outputs of the model are: (i) Whole farm feed, energy and water consumption; (ii) Whole farm animal production; (iii) Whole farm NH_3, N_2O, CH_4 emissions; (iv) Whole farm solid and liquid manure production and composition; (v) Faecal Indicators Organisms presence at different farm production stages; (vi) Farm scoring (when two situations are compared). Regarding the case study described, there is a reduction of N excreted and gaseous emissions in the scenario where BATs have been implemented (Figure 1). Protein consumption is reduced from 484 to 402 g per kg of live weight (LW) of fattening pig. Nitrogen concentration in the slurry is also reduced from 4.1 to 3.5 g/kg. Total farm gaseous emission per kg of LW is lower in the scenario with BATs (18.9 g NH_3/kg LW and 1.2 g N_2O/kg LW) compared to the standard scenario (29 g NH_3/kg LW and 1.5 g N_2O/kg LW). Gaseous N losses during the slurry application are reduced by 40% (from 4086 to 2467 kg N) using the trailing hose system. Therefore the efficiency in N use is significantly increased by the use of these BATs. It is estimated that when BATs are applied, 67% of the N excreted by animals can be deposited in the soil for crop N intake. On the contrary, only 59% of the N excreted would reach the soil in the standard scenario.

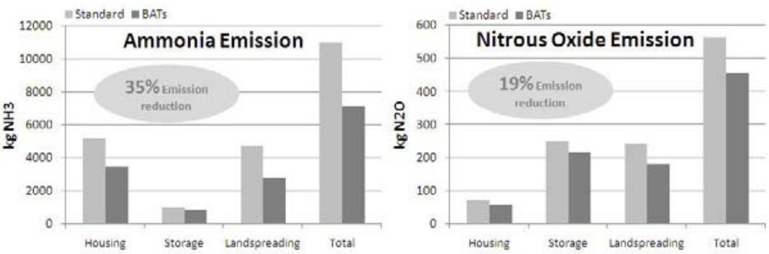

Figure 1. Gaseous emissions (NH_3, N_2O) in the different farm stages for the two scenarios.

Conclusions

This tool integrates existing information on the potential of different strategies and environmental techniques implemented to reduce gaseous (NH_3, N_2O) and N losses in intensive livestock farms. The BATFARM software enables the simulation and comparison of the combined effects of different environmental techniques throughout all the farm process. In addition, BATFARM Software takes into account specific management practices and climatic conditions. Nevertheless, further test will be necessary to validate the results provided by this tool.

Acknowledgements

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MODELLING MITIGATION OF AUSTRIAN NITROGEN EMISSIONS: ECOLOGICAL AND ECONOMIC CONSIDERATIONS

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The project FarmClim ("Farming for a better climate") assesses impacts of agriculture on N and GHG fluxes in Austria and proposes measures for improving N efficiency and mitigating emissions, including their economic assessment. Including stakeholders` views at a very early project state will contribute significantly to closing the science-policy gap in the field of climate friendly farming (Amon et al., 2014). This paper sfocuses on animal husbandry and crop production measures, and on N2O emissions from soils.

Material and Methods

FarmClim applies national inventory reporting methods to assess Austrian NH3 and GHG fluxes in order to develop flux estimates with implementation of mitigation measures. Based on scientific literature and on the outcome of the Austrian working group "agriculture and climate protection" a list of potential mitigation measures has been produced. Data cover resulting production levels as well as GHG mitigation.

In crop production, an optimisation potential remains with respect to N fertilization and nutrient uptake efficiency. Projected regional yield data and information on the N content of arable crops have been delivered from field experiments. These data complement official statistics and allow assessing the effect of increasing proportions of legume crops in crop rotations and reducing fertilizer input on a regional scale.

Economic efficiency of measures is a crucial factor for their future implementation on commercial farms. The economic model evaluates investment costs as well as changes in direct costs, labour costs and economic yield. Biophysical modelling with Landscape DNDC allows establishing a framework to move from the current approach of applying the IPCC default emission factor for N2O emissions from soils. We select two Austrian model regions to calculate N fluxes taking into account region and management practices. Hot spots and hot moments as well as mitigation strategies are identified.

Results and Discussion

N and GHG fluxes in Austrian agriculture – livestock production

Flux estimation and assessing potential mitigation measures in FarmClim follows the recently updated reporting guidelines (EEA, 2013), which require an N flow model for estimating agricultural NH3 and NOx emissions. Flux changes are consistently followed up in all process stages. The Austrian inventory is currently updated and results will directly feed into the FarmClim project. Three promising measures in animal husbandry have been identified. Phase feeding for pigs reduces N emissions and feeding costs. However, investments in technical equipment are necessary. An “optimised diet” adjusts the protein content of pig feed rations according to feed analysis. This is not as effective as phase feeding, but can be applied more widely and by this wider implementation also has the potential of contributing significantly to a reduction of N emissions. Dairy cattle diet: Increasing milk yield based on high quality roughage reduce GHG emissions. The calculation model allows the economic assessment of different feeding rations in regard to high and medium quality roughage.

Anaerobic digestion of animal manures reduces GHG emission, but is rarely used in practice in Austria, because of limited economic efficiency. To assess this measure, collectively owned facilities (3-5 medium sized farms) are assumed and their GHG abatement costs calculated.

N and GHG fluxes in Austrian agriculture – crop production

Intensity of fertilisation in arable land is reduced by 10% or 20%. For calculations high and medium yield capacity are assumed. The economic calculations are conducted for sugar beet, corn, wheat, raps, and barley. Possible higher yields and lower costs due to reduced need for mineral fertiliser are valued. Results will be available at the conference. Substitution of corn in crop rotation: The share of corn in crop production in Austria is still rising because of the high energy density compared to other agricultural crops. We assume a replacement of corn by soybeans in arable farming and by wheat in pig fattening. Costs are calculated on the level of one hectare arable land. The basis will be a comparison of gross margins.

Mapping emissions and improving inventory reporting

Two test regions have been identified for soil emission modelling. The “Marchfeld” is an intensively used agricultural area in North-East Austria with very fertile soils and dry climate. The area of central Upper-Austria is characterized by heavy gley soils and higher annual precipitation (890mm). Based on site parameters, vegetation characteristics, management and meteorology, the model is able to predict C and N bio-geo-chemistry in agricultural ecosystems at site and regional scale. This is the basis for assessing further mitigation specifically focussing on the hot spots and hot moments of N emissions on a regional scale.

Conclusion

The list of mitigation measures resulting from the project activities has been tailored to fit Austrian conditions in order to be attractive to stakeholders and farmers. Providing information on economic impacts to farms adds to the transparency of the approach taken. We expect that understanding the interest and the worries of farmers from the beginning supports creation of realistic output that can provide a strong incentive to urgently needed actions on improving farm N efficiencies.

Acknowledgements

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NITROGEN GAS EMISSIONS AND BALANCES OF ENERGY MAIZE CULTIVATED WITH BIOGAS RESIDUE FERTILIZER

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Despite increasing energy maize cultivation in combination with biogas residue (BR) application, little is known about the impact of this farming system on gaseous nitrogen (N) losses as well as N balances. However, there is an urgent need to address these knowledge deficits, since the application of BR is associated with a potentially higher ecological risk compared to other organic fertilizers. The reasons for this increased risk are the higher ammonium (NH_4^+) and total N concentration, the decreased organic matter (OM) content, and the enhanced pH value of BR compared with undigested organic fertilizers (Möller and Müller 2012). Our study will make a major contribution to fill these knowledge gaps by conducting field experiments at two different study sites in Germany.

Materials and Methods

The field experiments are carried out in the context of the collaborative research project "Greenhouse gas mitigation potentials in energy maize cultivation to produce biogas residues" The field site Dedelow, located in the lowlands of Northeast Germany is characterized by loamy sand (haplic luvisol). The Dornburg site is located at the eastern periphery of the Thuringian Basin and characterized by very clayey silt (haplic luvisol). *Zea mays* was cultivated at both sites. The impact of BR fertilization was examined by means of five increasing N application levels (50%, 75%, 100%, 125% und 200% N-BR; applied using trail hoses), compared to an unfertilized (0% N) and a minerally fertilized control (100% N-MIN; site-specific amounts of 160 and 150 kg N ha^{-1}, respectively). Seventy percent of the applied N-BR was assumed to be plant-available. Nitrous oxide (N_2O) measurements at three plots occurred daily immediately after fertilization and subsequently bi-weekly using the *non-flow-through non-steady-state* chamber measurement technique after Livingston und Hutchinson (1995, Fig. 1). Immediately after BR application, ammonia (NH_3) volatilization was measured intensively using the open dynamic chamber *Dräger-Tube* method after Pacholski et al. (2006). To quantify soil N dynamics, soil samples were taken both three times in spring after fertilization and once in autumn after harvest from 0-30 cm soil depth. In the laboratory, the sum parameter N_{min} was analyzed by means of $CaCl_2$ extraction. Subsequently to drying the harvested plant material, plant N content ($N_{harvest}$) was determined using elementary analysis (VARIO EL III, Elementar GmbH Hanau). With consideration of the measured N gas exchange as well as the determined N soil and plant parameters, N balances can be calculated using a simple difference approach. Values of N output ($N_{harvest}$, N_{N2O_cum} and N_{NH3_cum}) are subtracted from N input values ($N_{fertilizer}$, N_{min_spring} and N_{min_autumn}). All presented results refer to the time periods 01.04.2011 until 31.03.2012 and 01.04.2012 until 31.03.2013, respectively.

Results and Discussion

At both sites, our results showed small N_2O-N losses with maximum cumulative emissions of 4.9 kg N_2O-N ha^{-1} yr^{-1} and 3.1 kg N_2O-N ha^{-1} yr^{-1} in Dedelow and

Dornburg, respectively. The amount of applied N-BR had a moderate impact on the N_2O emissions. Only very high fertilizer amounts (125% and 200% N-BR) caused slightly increased cumulative N_2O emissions. However, the correlation was attenuated to a considerable degree by climate-induced inter-annual and seasonal variability of N_2O fluxes (Fig. 2 and 3). Maximum NH_3 losses in Dedelow amounted to 28 kg N ha^{-1} yr^{-1} and 6 kg N ha^{-1} yr^{-1} in 2011 and 2012, respectively. The considerable variability between these years likely results from differences in the time period between fertilizer application and incorporation. While it took more than 24 hours to incorporate the applied BR in spring 2011, the incorporation occurred within two hours following application in 2012. Thereby, NH_3 volatilization was reduced by one fifth (Tab. 1). Due to immediate incorporation at the treatments 50%-100% N-BR in Dornburg in 2012, a reduction in NH_3 losses of one third was achieved compared to 2011. For the highly fertilized treatments, BR application was split into two doses, with the second application taking place after planting. As an incorporation of the BR was thus not possible, the resulting NH_3-N losses were comparable to values from 2011 (Tab. 1). With increasing N-BR amount, N balances tend to increase from strongly negative values (accumulative deficit) to extremely positive values (accumulative profit) for treatments receiving high N-BR amounts (Fig. 4). Only applied N amount exceeding 200 kg N ha^{-1} resulted in accumulative profits, with no differences between field sites for increasing N levels. It is evident that N balances were more influenced by N uptake from plants than by gaseous N losses. In order to correctly interpret the N balances, it is mandatory to integrate other N fluxes into the balance calculation. Apart from soil-based mobilization and immobilization turnover processes and nitrate leaching, this applies specifically to gaseous N loss in the form of N_2. Therefore, in additional laboratory experiments, we measured the amount of N_2-N loss using the Helium incubation method of Butterbach-Bahl et al. (2002). Our results indicate that N_2 may be a potentially important pathway of N loss, with up to 5% of the total N output lost as N_2, corresponding to 15 kg N ha^{-1} yr^{-1}.

Conclusions

Our results showed that climatic conditions and incorporation time have a higher impact on N_2O emissions and NH_3 losses than the applied N amount. Plant N uptake affected the N balances more than gaseous N losses, but positive N balances only occurred when applied N-BR amounts exceeded 200 kg N ha^{-1}. As the results varied considerably between years, definite conclusion about the effect of BR application as well as site-specific and climatic effects on gaseous N losses and N balances cannot be drawn based on only two years of field measurements.

Figure 1: Manual chamber measurement system to determine N2O fluxes.

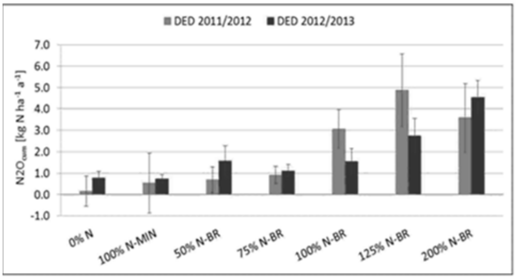

Figure 2: Cumulative N2O-N losses at the Dedelow site.

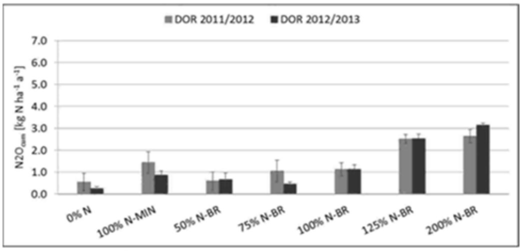

Figure 3: Cumulative N2O-N losses at the Dornburg site.

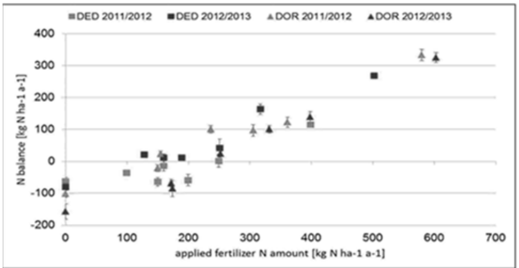

Figure 4: Calculated N balance depending on the applied N-BR amount.

Table 1: Cumulative NH3-N losses in Dedelow and Dornburg during both study years.

Treatment	DED 2011/2012	DED 2012/2013	DOR 2011/2012	DOR 2012/2013
	NH_3 losses in kg N ha^{-1}		NH_3 losses in kg N ha^{-1}	
50% N-BR	13.76	2.56	23.54	6.46
75% N-BR	28.11	3.67	25.56	9.42
100% N-BR	22.64	4.47	29.01	11.39
125% N-BR	24.97	5.79	25.18	25.65
200% N-BR	25.06	4.90	31.08	32.51

BATFARM SOFTWARE: A SUPPORT TOOL TO MITIGATE NITROGEN LOSSES AT DAIRY HOUSING

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Dairy farms contribute to nitrogen (N) pollution, especially in intensively managed farms with scarce land availability and high external inputs. Nitrogen pollution from dairy farms affects either air through the emissions of ammonia (NH_3) and nitrogen oxides (N_2O and NO) or water by nitrate (NO_3^-) leaching. Efficient use of N is one of the major assets of sustainable dairy farming as inefficient N use also affects the economic performance. However, it is necessary to determine appropriate mitigation options to reduce efficiently the environmental pollution from intensive farms. BREF documents, in which environmentally friendly best available techniques (BAT) are listed, have been published on intensive rearing of poultry and pigs. However, there is not official BREF document for dairy farming. BATFARM Project (http://www.batfarm.eu/) aims to set-up a model in which the application of different mitigation strategies will be simulated on dairy farms. This contribution is focused on the effect of such strategies on N balance and N gaseous losses (NH_3, N_2O) at dairy housing.

Materials and Methods

The model has been set-up by intermediary approaches between empirical and mechanistic models. Data from national inventories have preferably been considered. When necessary, literature values from scientifically tested commercial/experimental farms have been included. Strategies related to N mitigation have been classified as: Nutritional (crude protein fitting in total mixed rations, phase feeding and electronic collars for milking cows), Manure Management (manure removal system and removal frequency) and Technological (deep pit aeration, curved slat mats and slatted floors with valves). Ammonia losses have been assessed including the type of facility (freestall, tie-stall), type of floor (solid, slatted floor), type of manure (slurry, solid manure), herd management (confinement, grazing) and slurry temperature. IPCC default values have been used to estimate N_2O emissions. Each country has been divided in different climatic areas for monthly temperature values. In relation to the economical feasibility, a calculator will assess the cost of the strategies.

Dairy farms located in Bilbao (Sp), Lisbon (Por), Rennes (Fr), Glasgow (UK) and Dublin (Ire) have been simulated. Intensive farming has been allocated for Spain, Portugal and France. On the contrary, extensive farming with grazing activity has been simulated for UK and Ireland. Herd size is 60 cows, 15 calves and 20 heifers in all cases. Mean milk yield is 9,000 l/cow/year for intensive farms and 7,000 l/cow/year for extensive farms. Grazing season is simulated from April to October. Milking cow diet is based on total mixed rations (TMR) for the intensive system. On the contrary, milking cows from extensive farms are fed with grass silage based rations during confinement. Default values are used for body weights, N retention and manure production. Table 1 shows the abatement strategies (BAT) simulated:

Results and Discussion

Main outputs of the dairy housing sub-model are: (i) Herd N intake and excretion; (ii) NH_3 and N_2O losses from manure at dairy housing; (iii) Solid manure and liquid slurry production; (iv) Water consumption; (v) Efficiency indexes. Figure 1 shows NH_3 and N_2O losses after adopting 3 mitigation strategies at dairy housings distributed throughout EU-Atlantic region. It is estimated that up to 10-20% of emitting NH_3 could be abated in these case studies. Ammonia losses were significantly higher in intensive farms because of the larger N intake of high-producing herds. However, NH_3 losses per unit of milk produced ranged from 1.7 to 2.2 g NH_3-N/ kg milk in intensive farms while extensive farms ranged from 1.0 to 1.3 g NH_3-N/ kg milk. Different NH_3 emissions among regions which were grouped either as intensive or extensive were accounted for the mean temperature at each site. Nitrous oxide abatement ranged between 1-5% in this simulation. Emissions were higher for grazing based farm systems. Nonetheless, this simulation did not include N_2O emission from on-field manure application (calculated in the Landspreading sub-model of BATFARM Software).

Table 1. Nitrogen abatement strategies (BAT) at dairy housing for intensive and extensive farms.

Type of Farm	Standard Situation	BAT Situation
Intensive Farms	CP content TMR diet: 17.0%	CP content TMR diet: 16.5%
	Slurry Removal Frequency: 6 months	Slurry Removal Frequency: 1 month
	No Valves in Slatted Floor	Valves in Slatted Floor
Extensive Farms	Grass Silage and Grazing	Grass Silage and Grazing with Electronic Collars
	Slurry Removal Frequency 6 months	Slurry Removal Frequency 1 month
	No Valves in Slatted Floor	Valves in Slatted Floor

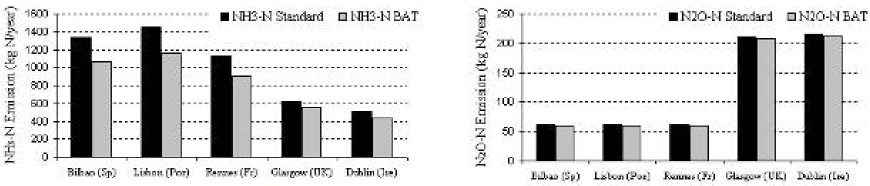

Figure 1. Annual ammonia (NH_3) and nitrous oxide (N_2O) losses at dairy housing from five EU-Atlantic regions in two scenarios.

Conclusions

BATFARM Software integrates existing information on the potential of different strategies and environmental techniques to reduce N gaseous losses (NH_3, N_2O) at dairy housings. BATFARM Software takes into account specific management practices and climatic conditions throughout EU-Atlantic regions. Further test will be necessary to validate the results provided by this tool.

Acknowledgements

This work has been co-financed by BATFARM Interreg-Atlantic Area Project (2009-1/071): "Evaluation of best available techniques to decrease air and water pollution in animal farms".

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DM YIELD AND N2O EMISSIONS AS AFFECTED BY 3,4-DIMETHILPHIRAZOLPHOSPHATE, A NITRIFICATION INHIBITOR APPLIED DURING WINTER IN NORTHWESTERN SPAIN

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Nitrogen (N) additions to cropland soils are the largest source of anthropogenic nitrous oxide (N2O). N2O is an important contributor to global greenhouse gas radiative forcing. Slurries and inorganic N fertilizer application may cause important emissions of N2O. It has been proved that the use of nitrification inhibitors (NI) may increase N use efficiency by crops and also decrease N2O losses. The objectives of the study were to determine in a field trial (after autumn and spring fertilizations) the effect of N fertiliser source on the crop yield and to evaluate the influence of the dimetil pirazole as NI, when is applied together with cattle slurry and ammonium calcium nitrate, on crop yield and N2O losses.

Materials and Methods

The experiment was carried out between October 2012 and May 2013 at the CIAM Research Centre (NW Spain), on a silt loam soil classified as Humic Cambisol. The average annual temperature in the study area is 13.3 ºC and the average annual rainfall 1128 mm (10-year average). Fertilizers were applied twice (on November 15th and on April 18th). Italian ryegrass was sown at 40 kg ha-1 on November 16th. The trial was carried out as a completely randomized block design with three replicates and seven treatments. Two treatments consisted on cattle slurry injected into the soil with or without DMPP as NI, and four treatments resulted of combinations of a first fertilization of injected cattle slurry: with or without DMPP, and a second application of mineral N fertilizer as calcium ammonium nitrate (CAN) or CAN stabilised with DMPSA (Dimethylpyrazole succinic acid, EuroChem Agro). There was also a nil N treatment as control. The plots were cut twice (on March 14th and on May 21st) and sub-samples were collected for determination of dry matter (DM) and Kjeldahl N content. Within each plot two closed chambers were placed (diameter: 25 cm; height: 36 cm; depth into the soil: 3 cm) to monitor N2O fluxes from November 15th to May 20th. Samples were collected 40 minutes after chamber enclosure and analyzed by a gas chromatography (Agilent 7890A) equipped with an electron capture detector for N2O detection. In addition, soil samples, for the measurements of moisture as Water Filled Pore Space (WFPS) and mineral N, and meteorological station data were taken to see the influence of weather and soil properties on N2O fluxes.

Results and Discussion

Fertilizer source had a significant effect ($P< 0.001$) on the total DM yield and N uptake by the crop. The addition of DMPP to cattle slurry (Table 1) increased significantly DM yield and N uptake in the first cut (on March 14th). Due to a rainy spring, the second fertilization was applied too late, on April 18th, and in the second cut (on May 21st), there were no significant differences on DM yield or N uptake in relation to the NI use, for either cattle slurry or mineral fertilizations.

Cumulative N2O from November to May (growing season) ranged from 0.808 to 1.518 kg N2O-N ha-1 (Table 2). Spatial and temporal variability in N2O emissions were high. In period I, from November15th to March 8th, the incorporation of DMPP with cattle slurry decreased the percentage of N2O lost in relation to the N applied (Table 2, EF). The application of CS and CAN were not followed by distinct peak of emission following the second application on April, and fluxes of N2O emission were low throughout the whole period II, from April 18th to May 20th (Tabla 2). High %WFPS contents, mean value of 78% WFPS for period II, suggesting that denitrification was the major process responsible for emissions, and N2 was probably the final reaction product. Such conditions might have decreased the efficiency of DMPP and DMPSA in reducing N2O emissions as is described in other experiments under laboratory conditions (Menéndez et al., 2012).

	Kg DM ha^{-1}			Kg N ha^{-1}		
Treatment	1st Cut	2nd Cut	Total	1st Cut	2nd Cut	Total
T1	0,74d	0,71c	1,45d	11,65c	7,95b	19,60d
T2	1,34bc	1,69b	3,02c	19,45bc	19,92b	39,37c
T3	1,80ab	1,57b	3,36c	27,10ab	17,40b	44,50c
T4	1,21cd	3,46a	4,68b	18,17bc	54,00a	72,16b
T5	1,37bc	3,61a	4,99ab	20,20bc	66,73a	86,93a
T6	2,07a	3,47a	5,54a	31,89a	62,96a	94,85a
T7	1,52bc	3,52a	5,04ab	22,79b	66,58a	89,38a

Values followed by different letters are significantly different ($p<0.05$, Duncan test)

Table 1. Effect of N source on dry matter (DM) yield and N uptake from italian ryegras during growing season. T1: Control; T2: Cattle slurry (CS)+CS; T3: CS-DMPP+CS-DMPP; T4: CS+Calcium ammonium nitrate (CAN); T5: CS+CAN stabilized with NI; T6: CS-DMPP+CAN; T7: CS-DMPP+CAN stabilized with NI.

	Period I				Period II				Growing season	
Treatment	Fertilizer	N Applied Kg N ha^{-1}	Total N_2O cumulative fluxes kg N_2O-N ha^{-1}	EF (%)	Fertilizer	N Applied Kg N ha^{-1}	Total N_2O Cumulative fluxes kg N_2O-N ha^{-1}	EF (%)	Total N_2O Cumulative fluxes kg N_2O-N ha^{-1}	EF (%)
T1	C	0	0.524 (0.197)			0	0.067 (0.017)		0.591 (0.197)	
T2	CS	59	1.306 (0.639)	1.32	CS	93	0.170 (0.060)	0.11	1.476 (0.603)	0.58
T3	CS+DMPP	62	0.719 (0.424)	0.32	CS+DMPP	82	0.109 (0.023)	0.05	0.822 (0.420)	0.16
T4	CS	61	1.332 (0.400)	1.32	CAN	70	0.109 (0.037)	0.06	1.441 (0.382)	0.65
T5	CS	62	1.377 (0.869)	1.38	CAN-NI	70	0.141 (0.037)	0.11	1.518 (0.874)	0.70
T6	CS+DMPP	81	0.697 (0.416)	0.21	CAN	70	0.143 (0.087)	0.11	0.840 (0.341)	0.16
T7	CS+DMPP	58	0.619 (0.209)	0.16	CAN-NI	70	0.189 (0.018)	0.17	0.808 (0.194)	0.17

Data are the average of three replicates

Table 2. Average cumulative N2O-N emission (standard deviation in parentheses) during the periods studied. Period I: from November 15th to March 8th and period II: from April 18th to May 20th. C: Control; CS: Cattle slurry; CS+DMPP: Cattle slurry with DMPP; CAN: Calcium ammonium nitrate; CAN-NI: CAN stabilized with the NI.

Conclusion

Our results suggest that in our climate conditions the use of cattle slurry had the same impact on N2O emissions in comparison with mineral fertilization with calcium ammonium nitrate. During the growing season of italian ryegras the lowest emissions factors of the N fertilized treatments studied were found after cattle slurry+DMPP applications in autumn. The use of the inhibitor increased N efficiency by the crop.

Acknowledgements

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MITIGATION OF NITROUS OXIDE FOLLOWING GRASSLANDS CULTIVATION

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Grasslands managed by agriculture are regarded as beneficial for the greenhouse gas balance owing to their perennial nature and carbon (C) sequestration capacity (Hutchinson et al., 2007). However, grasslands renovation by tillage disturb the balance by increasing nitrous oxide (N2O) emissions (Mori and Hojito, 2007). Ploughing is practiced to improve quality and productivity of aged grasslands, and this stimulates the C and nitrogen (N) mineralization of freshly incorporated residues and microbial activities, giving an oxygen-limited environment conducive to N2O emission (Velthof et al., 2010). Composition of residues, and the extent of oxygen limitation, is likely to influence the level of N2O emissions, hence the proportion of clover in the grassland, and tillage depth, could be controlling factors. The objective of this study was to investigate shallow tillage as a potential N2O abatement option for grasslands with different botanical composition. We hypothesized that: i) rotovation (shallow incorporation) two weeks prior to ploughing, by partly un-coupling C and N turnover, would reduce N2O emissions; and that ii) legumes stimulate N2O emissions to a greater extent than grass due to faster decomposition and net N mineralization.

Materials and Methods

A 56-day field study was conducted on a sandy-loam soil in Denmark ($56^{0}29'$N, $9^{0}34'$E) in spring-summer 2013. The experimental design was a split-plot, where whole-plots included: i) rotovation to 6 cm (RT, two weeks only); ii) rotovation with ploughing to 20 cm depth after 2 weeks (RTP); iii) direct ploughing to 20 cm depth (PL);and iv) undisturbed grassland as reference (CTL). The split-plots included: i) ryegrass (Lolium perenne L.); ii) clover (Trifolium repens L.); and iii) a mixture of ryegrass and clover (grass-clover). Chamber support frames were installed immediately after the rotovation to measure N2O. Static chambers, equipped with an internal fan, were used for taking five gas samples during an hour. Soil mineral N, moisture, plant dry matter and C:N were analysed. The fluxes were calculated using HMR (Pedersen et al., 2010), and data were analysed in R with a mixed model. 3.Results and Discussion During the first three weeks of monitoring, emissions from all treatments were similar, possibly owing to a lower temperature. The emissions of N2O then sharply increased from day 21 onward to 49, a period with accumulated rainfall of 56 mm. Higher cumulative N2O was observed from ryegrass in the PL treatment (2.2 ± 0.2 kg N2O -N ha-1), followed by RTP (1.6 ± 0.4 kg N2O -N ha-1 including emissions from RT). The PL treatment emitted 34, 30 and 38% more N2O compared to RTP in ryegrass, clover, and grass-clover, respectively. Ploughing of grasslands increased N2O emissons ($p< 0.01$) compared to CTL. Relative to PL, RTP reduced N2O emissions. There was a significant interaction between vegetation type, ploughing method and sampling day ($p< 0.01$) with respect to N2O emissions. Relatively lower emission of N2O in the RTP treatment compared to PL might be a result of crop residues being mineralized under aerobic conditions, which is supported by the dynamics of mineral N (data not shown). This will leave less available organic C when ploughed in RTP, whereas in the PL treatment both moisture and carbon availability can stimulate the demand for O2, and N2O emissions (Johansen et al., 2013). In this study, emission factors for ryegrass and clover were 2.0 and 2.2% in the

PL treatment and 1.5 and 1.6% in RTP, during the 56-day period, suggesting that clover had a higher potential for N2O emissions. Emissions of N2O were less in the clover treatment compared to the other vegetation, even though, higher or similar amounts of mineral N accumulated in the soil. This indicates that remaining degradable C is important in addition to mineral N for N2O emissions (Petersen et al., 2008). The multiple regression analysis showed that N2O emissions were controlled by the interaction of ploughing method with soil nitrate and soil moisture.

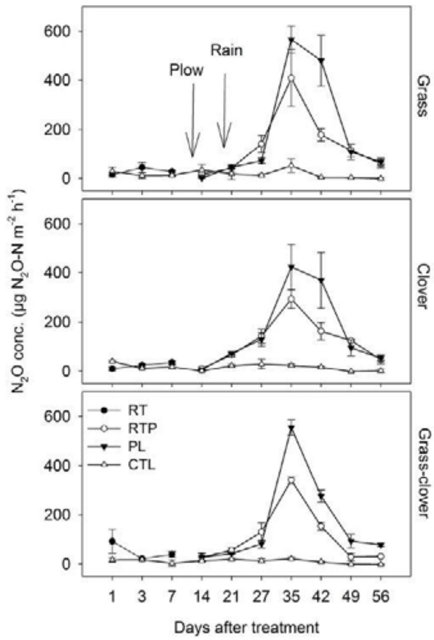

Fig.1 Temporal dynamics of N2O in RT, RTP, PL and CTL. Data points show means and error bars denote the standard error for n=3 (for CTL, n=2).

Conclusions

Rotovation of grasslands prior to ploughing reduced the emissions of N2O. The sward type was non-significant in mitigation of N2O emissions, but the potential for N2O emissions would be higher from clover. The most important drivers of N2O emissions were soil moisture, NO3- and ploughing methods.

Acknowledgements.

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SHORT AND MEDIUM TERM EFFECTS ON NITROGEN LEACHING OF THE INTRODUCTION OF A PEA OR AN OILSEED RAPE CROP IN WHEAT-BASED SUCCESSIONS

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Legume crops are a major source of natural nitrogen (N) input in cropping systems. They enable to reduce some environmental impacts associated with the synthetic fertilizers. Pea or oilseed rape crops enable to increase the grain yield of the succeeding wheat, while decreasing its need of synthetic N. However, several studies have highlighted higher soil mineral nitrogen (SMN) and leaching risks just after pea or oilseed rape crops compared to cereals. Nevertheless, rare other studies (Thomsen et al. 2001; Hauggaard-Nielsen et al. 2003) suggested lower SMN and leaching risks during the second winter after pea or oilseed rape cultivation. In this study, we analyzed the SMN and leaching risks during the first and second winter after winter pea, oilseed rape and wheat crops and assessed the consequences on N leaching at the crop sequence scale.

Materials and Methods

Two field experiments were carried out from 2007 to 2011 at INRA-Grignon and CETIOM Experimental units, Paris Basin. Soft wheat, winter pea and winter oilseed rape were sown every year in order to compare 8 crop sequences. The soil remained bare in autumn. All plots were statistically randomized with 4 replicates. The SMN was measured three times over the winter periods in the Grignon trial and only after harvest in Holnon. Effects of the harvested and preceding crops on the SMN during autumn were studied through linear and mixed models using R statistical software. Soil nitrogen fluxes below the rooting zone were then predicted for various crop sequences using the LIXIM model (Mary et al., 1999). Simulations, run on 20 climatic years, were initialized with post-harvest measurements of Grignon trial. Prediction of N leaching was made either with bare soil in autumn or sowing of a catch crop after pea and volunteers after oilseed rape.

Results and Discussion

Despite higher SMN at the beginning of winter (+26 kg N ha^{-1}) and N leaching (+5 kg N ha^{-1}) after pea compared to wheat, crop sequences with pea do not increase the average yearly N leaching compared to those with only wheat. Yet, effects on first and second winter after the pea crop compensate each other. SMN in autumn are significantly lower after wheat with a pea preceding crop compared to wheat with a wheat as preceding crop: -24 and -30 kg N ha^{-1} after harvest respectively for Grignon and Holon trials and – 18 kg N ha^{-1} at the beginning of the winter for Grignon trial (Fig. 1). Similarly, N losses are 7 kg N ha^{-1} lower for a pea preceding crop compared to a wheat preceding crop.

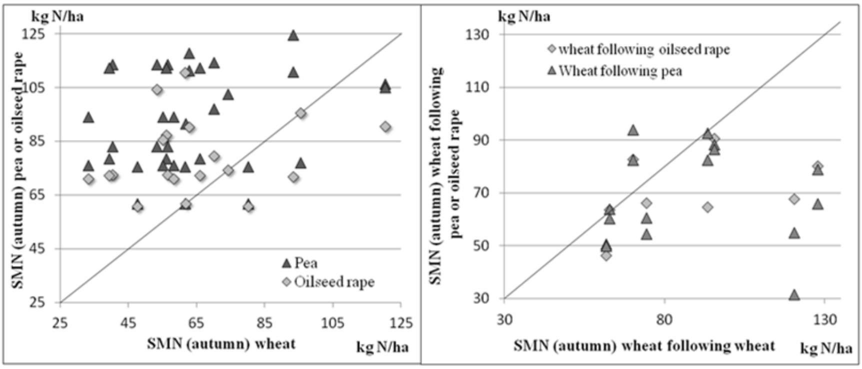

Figure 1. Soil mineral nitrogen (SMN) in autumn after pea, oilseed rape or wheat crop (left) and soil mineral nitrogen in autumn after wheat with pea, oilseed rape or wheat as the preceding crop (right)

The crop sequences with oilseed rape lead to lower N leaching values than those with only wheat (Fig. 2). This is due of the high N absorption capacity of oilseed rape sown early in autumn and of a preceding effect similar to the one of pea (-9 kg N ha^{-1} for N losses with oilseed rape preceding crop compared to wheat). The reduction of N leaching linked to the preceding effect of pea and oilseed rape could be explained by better N uptake conditions for the cereal with oilseed rape or pea as preceding crops, mainly due to a safer root system, as several root diseases can occur on a second wheat crop. Indeed, in our experiment, we observe a significant higher dry matter and yield for wheat with oilseed rape or pea as preceding crops. However this is probably not the only factor since, we observed no regular higher aerial N uptake for oilseed rape, and regular but not significant higher aerial N uptake for pea (+12 kg N ha^{-1}). Our simulation demonstrates also that the preceding crop effect on N leaching (-7 to -9 kg N ha^{-1} for pea or oilseed rape compare to wheat) is important to take into account in addition to the effect linked to the presence of catch crop or volunteers in intercrop period (in average, -17 kg N ha^{-1} compared with bare soil).

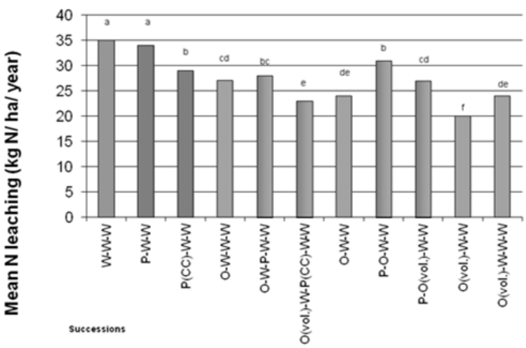

Figure 2. N leaching risks for various successions simulated with LIXIM model on 20 climatic years. W = wheat; O = Oilseed rape; P = pea; CC = catch crop; vol = volunteers. Bars with the same letters are not statistically different (α = 0.05, Tukey HSD).

Conclusions

Our results demonstrated that N leaching should be analyzed at the scale of crop sequences, in order to take into account the effects of the preceding crop, the crop in place and the intercrop management. At this multi-year scale, the introduction of a legume crop such as pea or oilseed rape in cereal-based successions led to a similar or lower risk of nitrate leaching. In addition, it should be reminded that pea enables to avoid the other forms of nitrate losses: no occasional nitrate losses during N fertilizers handling, no indirect nitrate losses due to long distances deposition of gaseous forms of N.

Acknowledgment

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NITROUS OXIDE EMISSIONS FROM FERTILISED UK ARABLE SOILS: QUANTIFICATION AND MITIGATION

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Cultivated agricultural soils are one of the greatest sources of nitrous oxide (N_2O) emissions (Regina et al., 2013), and with agricultural land covering 40-50% of the earth's surface they could have a large influence on future climate. The time and amount of fertiliser application can alter the magnitude of N_2O emissions, and various methods are being investigated to limit emissions from agricultural croplands. Split applications of fertiliser are encouraged to avoid surplus N (Maidl et al., 1996), and application is generally avoided following periods of high rainfall and soil moisture. The use of nitrification inhibitors (NIs) has also been investigated (Di and Cameron, 2012), but reported efficiencies are mixed, resulting in a requirement for more research before their use on arable crops is encouraged. A lack of measured data caused by practical and financial restraints means that UK N_2O emission estimates adopt the IPCC tier 1 methodology (IPCC, 1997), where an Emission Factor (EF) of 1% is applied to soils, regardless of soil type, climate, or location. Following this approach a linear increase in N_2O emissions with N application is assumed- but uncertainty in N_2O estimates is reported as the major source of uncertainty in national GHG inventories. The aim of this research is to improve the UK's agricultural inventory, by determining whether N_2O EFs should be location and fertiliser specific, and to understand how N_2O emissions are controlled by soil and environmental factors and fertiliser N rate. In addition, mitigation methods will be investigated to assess the impact of the NI Dicyandiamide (DCD), the use of split fertiliser applications, and the impact of different forms of N fertiliser. This work forms part of the UK's GHG Platform Programme.

Materials and Methods

Three cropland sites, one in Scotland and two in England were selected to enable measurements from arable land with contrasting soils and climates. Gilchriston (East Lothian, Scotland) is located on a sandy loam soil; Rosemaund (Hereford, England) on a clay loam soil; and Woburn (Bedfordshire, England) on loamy sand over sandy loam. Experiments took place for 1 year, from March 2011 to March 2012. Static chambers were installed at each site to measure N_2O from a range of fertiliser types and rates, and to assess the impact of DCD. The application rates and forms of all fertilisers used are displayed in Table 1. Five levels of ammonium nitrate (AN) were applied, with split applications over two or three dates (depending on location). Rates were based on those typical for the crop and location, and an additional split treatment was investigated, where application was spread over a greater number of days. Treatments were applied to plots randomly located in three blocks at each site, with five replicate chambers per plot. Daily N_2O measurements were taken in the weeks following application, with frequency decreasing throughout the year. Soil analysis of mineral N, moisture and temperature was also undertaken, allowing assessment of the impact of climatic and environmental variables on N_2O emissions.

Results and discussion

A significant difference in mean annual cumulative emission between the sites was observed. The greatest emissions were observed at the Scottish site (Figure 1), which experienced much wetter conditions (822 mm annual rainfall compared to 418 mm and 472 mm at Rosemaund and Woburn respectively). Treatment differences were apparent- however this effect was not consistent between sites, and in most cases was insignificant (Figure 1). There was no effect (P >0.05) of DCD, split fertiliser application techniques or fertiliser type on N_2O emissions at any of the three sites. The emissions at Rosemaund and Woburn were low on all treatments (generally < 0.5% total N applied) reflecting very dry conditions in the period after the fertiliser was applied. Although EFs were higher at Gilchriston (Figure 2), the IPCC default value of 1% was only exceeded for the 120 kg N ha^{-1} treatment, 120 kg N ha^{-1} split treatment and 200 kg N ha^{-1} treatment.

Fertiliser	Rosemaund (kg N ha^{-1})	Woburn (kg N ha^{-1})	Gilchriston (kg N ha^{-1})
AN 1	**60** (20, 20, 20)	**60** (20, 20, 20)	**40** (20, 20)
AN 2	**120** (40, 40, 40)	**120** (40, 40, 40)	**80** (40, 40)
AN 3	**180** (40, 70, 70)	**180** (40, 70, 70)	**120** (40, 80)
AN 4	**240** (40, 100, 100)	**240** (40, 100, 100)	**160** (40, 120)
AN 5	**300** (40, 130, 130)	**300** (40, 130, 130)	**200** (40, 160)
AN split	**240** (40, 50, 50, 50, 50)	**180** (40, 40, 40, 30, 30)	**120** (40, 40, 40)
Urea	**240** (40, 100, 100)	**180** (40, 70, 70)	**120** (40, 80)
AN + DCD	**240** (40, 100, 100)	**180** (40, 70, 70)	**120** (40, 80)
Urea + DCD	**240** (40, 100, 100)	**180** (40, 70, 70)	**120** (40, 80)

Table 1. Fertiliser application rates at the 3 experimental sites. AN: Ammonium nitrate. Numbers in bold indicate total fertiliser application rates; numbers in brackets indicate split application rates.

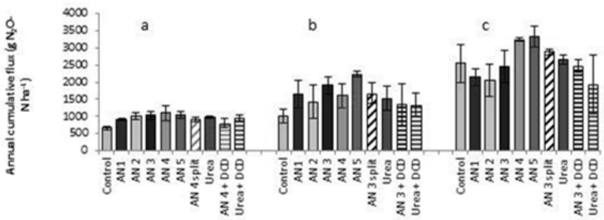

Figure 1. Annual cumulative N_2O emissions from a. Rosemaund, b. Woburn and c. Gilchriston

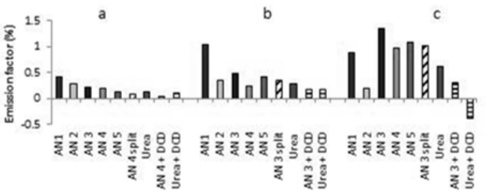

Figure 2. Emission factors for a. Rosemaund, b. Woburn and c. Gilchriston

Conclusions

A significant difference in mean annual cumulative emissions between sites indicates that location specific emission estimates should be considered, and reinforces the suggestion that N_2O emissions from N fertiliser depend on local climatic and soil characteristics. Further research is required under contrasting weather conditions to test the effects of DCD use, application timings and fertiliser type on N_2O emissions.

Acknowledgements

The authors are grateful to the UK Department for Environment, Food and Rural Affairs (DEFRA) and the Devolved Administrations for financial support.

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NITROGEN LEACHING FROM ORGANIC AGRICULTURE AND CONVENTIONAL CROP ROTATIONS

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Nitrogen (N) is an essential element for plant growth but when leached due to its high solubility in nitrate (NO_3), it has a major impact on hydrosystems contaminations. Indeed in the Seine basin, characterized by intensive cropping, most of the surface and groundwater is contamined by NO_3 (Directive n°91/676/CEE). Organic farming (OF) is already recognized as a good alternative for pesticide pollution, but its positive impact on NO_3 contamination is still controversial (Mondelaers et al. 2009). Only few articles have dealt with the impacts of OF on NO_3 leaching in Europe (Askegaard et al. 2005) but none in France, yet. The goal of our study is to investigate N leaching from different arable crops systems in OF and conventional farming (CF) in the Seine basin.

Materials and methods

So far, three regions have been studied: the Seine & Marne (S&M), Oise and Yonne which are characterized by clay-silt loam soil with drains, clay-silt loam without drains and sandy-silt loam respectively. In 2012-13, a network of ceramic cups has been installed in 8 commercial farms distributed in three different soil and climate conditions (Figure 1). OF systems differed in their nitrogen management, but had similar long crops rotations (average of 8 years including alfalfa). CF is dominated by the rotation wheat-oat-rape seeds and maize in those three regions. Exogenous fertilization averages on complete rotations were around 30 $kgN.ha^{-1}.yr^{-1}$ in OF and 150 $kgN.ha^{-1}.yr^{-1}$ in CF. Net input from legumes (alfalfa, faba beans or peas), due to biological nitrogen fixation, is around 100 $kgN.ha^{-1}.yr^{-1}$ (Anglade et al, in prep; Lorry et al. 1992). A total of 39 parcels in OF rotations and 9 in CF has been followed with 6 ceramic cups on each parcel during 6 winter months. Soil analysis (texture, nutrients, pH) were determined and water capacity retention were obtained with regional soil classification: 114 mm for S&M, 115 mm for Oise and 110 mm for Yonne. Climatic data was also gathered for determining water fluxes with rainfall and evaportanspiration.

Results and discussion

The OF rotations showed a gradient of sub-root concentrations: legumes with the lowest sub-root concentrations (6 $mgN.l^{-1}$), wheat post legumes without fertilization (15 $mNg.l^{-1}$) and with fertlization (21 $mgN.l^{-1}$), secondary cereals (12 $mgN.l^{-1}$) and a total average of 15 $mgN.l^{-1}$. In CF rotations, crops with catch crops showed the lowest values (8 $mgN.l^{-1}$), crops fertilized is in the middle (27 $mgN.l^{-1}$) and crops after legumes and fertilized are the highest (30 $mgN.l^{-1}$). This data matches past studies on CF which scored of 25 $mgN.l^{-1}$ (Machet & Mary, 1990). Soil mineral nitrogen before winter and soil organic matter were good indicators to predict sub-root concentrations in the different regions. The averages concentrations from the farms rotations were from 7 to 19 $mgN.l^{-1}$ in S&M, 10 to 39 $mgN.l^{-1}$ in Oise and 8 $mgN.l^{-1}$ in Yonne. However the conversion to leaching fluxes (concentrations x percolation water) changed the final contribution of the farms to NO_3 contamination. In 2012-13, efficient rainfalls (rainfall – evaportranspitranspiration) were in S&M, Oise and Yonne of 200 mm, 30 mm and 400 mm respectively. In consequence, the farm in Yonne with the lowest concentrations corresponded in fact to a higher leaching (30 $kgN.ha^{-1}.yr^{-1}$) than in Oise (around 10 $kgN.ha^{-1}.yr^{-1}$). In S&M, the leaching means varied from 10 to 36 $kgN.ha^{-1}.yr^{-1}$ for OF and from 26 to 41 $kgN.ha^{-1}.yr^{-1}$. Leahching data was significantly different between regions (Anova, R, pvalue = 0.0107*) (Figure 2).

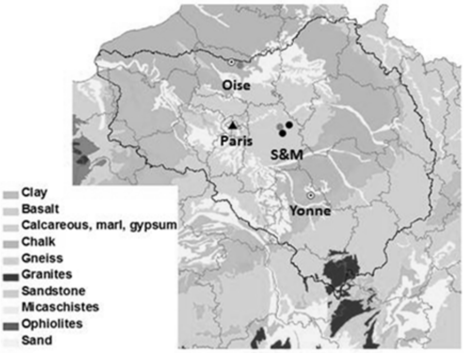

Figure 1 : Localization of the farms equipped (OF in white, CF in black and OF/CF in grey) on lithology map in the Seine & Marne (S&M), Oise and Yonne, in the Seine Basin.

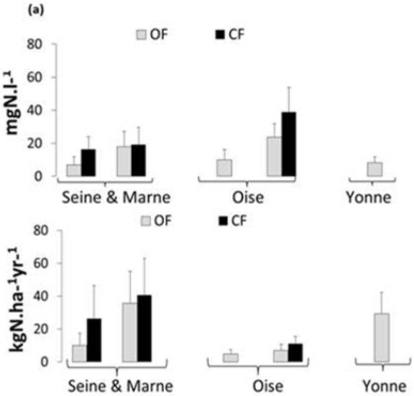

Figure 2: Sub-roots concentrations (a) and leaching (b) from the different farms studied in organic farms (OF) and conventional farms (CF) in the Seine &Marne, Oise and Yonne departments.

Conclusions

Overall, OF leads to lower nitric contamination in S&M from 10-50% and in Oise from 20-50%, but with important variations between two systems from a same department, due to different N management. The network of farms has been extended in 2013-14 to investiguate annual variations and a wider panel of N managements, with 8 CF and 8 OF, which make a total of 80 parcels and 500 ceramic cups.

Acknowledgements

Mr Collin, Gobard, Gilloots, Leturq and Lefèvre are acknowledged to allowing us to conduct our research in their fields. Thanks are due to Ile-de-France region (DIM-Astrea) and Seine Region Water Agency to support this ABAC project.

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TRENDS IN AGRICULTURAL AMMONIA EMISSION IN CANADA AND POSSIBILITIES FOR MITIGATION

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Canada emits almost 400 Gg of ammonia annually and the primary sources are production of beef cattle, crops (fertilizer application), pigs, dairy and poultry. Although a signatory to the Convention for Long Range Transport of Air Pollutants (CLRTAP) the issue of ammonia is considered to be bilateral between USA and Canada and there are currently no policies in Canada on abating ammonia emissions. Although farmers are not taking measures to abate emissions of ammonia, several practices are routinely used for economic or other reasons that help to reduce ammonia loss from farms including: fertilizer side-banding (injection), multi phase feeding, year-round grazing of beef cows and, in some regions, low emission application techniques for manure. Future emissions will be affected by economic trends and practicability of new abatement measures. The objective of this paper is to identify trends in Canadian emissions based on economic factors such as food consumption and agricultural exports and the potential for abatement measures.

Materials and Methods

Emission data has been previously computed by sector, sub-sector and ecoregions across Canada (Sheppard et al 2010, Sheppard and Bittman 2013). Emissions from the livestock sectors were amended by allocating emission from production of crops used to feed livestock based on farm survey data on livestock feeding practices in Canadian. Allocation of emissions to food consumption was calculated based on after-farm losses due to dressing, trimming and other losses at the processing, retail and domestic levels, according to data from Statistics Canada. Data on food and protein consumption, food import, and agricultural exports including commodities and live animals was also obtained from Statistics Canada.

Results and Discussion

Canadian emissions due to total food consumption and agricultural exports totalled 200 and 160 Gg, respectively, in 2006. Emissions that can be attributed to food imports assuming equivalent emission factors in source countries was c. 35Gg. Under Canadian production systems, emission in kg NH_3 per kg of actual food protein, after all trimmings and losses, varied from 4.5 for beef to 0.10 for poultry meat and on average much less for plant based foods (Table 1). Red meat (beef and pork) consumption accounted for 6.4 of 7.5 kg annual ammonia emissions per capita while emissions from all plant food totalled c. 0.3 kg emissions. The high emission from beef compared to other animals was due to high emissions from feedlots where cattle are fattened before market and to the large mass of reproductive animals that are needed to produce calves, even though in Canada reproducing cows and their unweaned calves are kept largely on pastures where emissions are very low. There may be scope for increasing the use of pastures as winter grazing technology improves and because of rising consumer preference for free range beef (and other animals) due to food quality and animal welfare benefits. Between 1981 and 2006 per capita protein consumption has increased by c. 5% while per capita ammonia emissions from food consumption has declined by c. 20% due to shift away from eating beef and towards more poultry products which have particularly low emissions (Fig. 1. Table 1). However, in terms of national emissions, the decline in per capita emissions has been offset by the growing population in Canada so overall food related emissions have risen gradually. There has also been increasing agricultural exports in Canada over the past decades and this is likely to continue due to world demand and possibly improving, if more variable, climate. Prospects for mitigation from imposed farm measures are limited as there are no national guidelines for ammonia. The costs for the most efficient abatement measures leading to 26% total reduction in ammonia would be about \$0.80 per kg NH_3 conserved (Sheppard and Bittman 2013). The current estimates are that a range of 20-140 Cd\$ of retail food value is produced for every kg of NH_3-N emitted so the cost of abatement measures would not be trivial.

Conclusion

It seems likely that contemporary food trends in Canada will continue the current pattern of decreasing per capita emissions, i.e. less total meat consumption, more consumption of plant based foods overall and pulse crops in particular; less red meat relative to poultry, dairy and fish; growing acceptance of consuming all parts of animals (nose to tail diets). These trends are due to greater concerns about health and environment, growing preference for Asian and Mediterranean cuisine, aging population, and a more urbanized population where slimness and plant based diets have growing social status. However exports of food, especially red meat, are not likely to decrease and the Canadian population will continue to increase mainly through immigration so total Canadian emissions will gradually rise.

Table 1 Annual ammonia emissions per unit food protein and per capita food consumption in Canada.

Food	NH_3 emission (kg NH_3 kg protein^{-1})	NH_3 emission (kg NH_3 person^{-1})
Beef	1.3	4.3
Dairy	0.21	1.1
Pork	0.43	1.0
Poultry	0.10	0.55
Eggs	0.18	0.17
Cereals	0.03	0.15
Vegetables	0.10	0.14
Other		<0.01
Totals		7.5

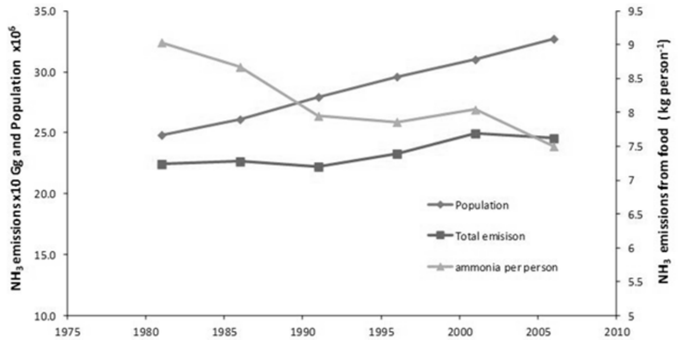

Fig 1. Annual ammonia emissions per capita and total Canadian based on food consumption and Canadian population from 1981 to 2006

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A DYNAMIC SIMULATION MODEL TO ANALYSE N2O EMISSIONS FROM BIOMETHANE PRODUCTION SYSTEMS

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Biomethane production in Germany as a source of renewable energy is based mainly on maize. For purposes of a more diverse agricultural landscape alternative crops are investigated. Besides a high dry matter production also the greenhouse gas (GHG) balance should be considered. In intensive and highly productive cropping systems the largest GHG source are N_2O emissions.

N_2O emissions originate from nitrification and denitrification in the soil. Soil water content and temperature of the soil, NH_4 concentration, NO_3 concentration, carbon turnover rate, soil pH are factors affecting these processes. Most of these factors vary during the growing season on a small time scale and are connected to other processes within the system of soil – plant – atmosphere. To evaluate the reasons for differences in N_2O emissions between different cropping systems, N fertilisation levels and years a dynamic simulation model was developed to include both the dynamics of influencing factors and the processes of nitrification and denitrification. The main objective was to build up the model in such a way that the influences on nitrification and denitrification reproduce measured data as accurate as possible.

Materials and Methods

In the seasons 2006/07 and 2007/08 a field trial was conducted at two sites in northern Germany. The data for this project include a maize monoculture at the more sandy site and maize monoculture as well as a crop rotation of wheat (for whole crop silage use), Italian ryegrass and maize at the more loamy site. All crops were tested in an unfertilised treatment and two treatments with different levels of mineral N fertilisation. In the selected treatments leaf area index (LAI), crop height and N in the aboveground biomass were measured at regular intervals throughout the growing season. Soil water content was measured by TDR probes and soil mineral nitrogen (SMN) in the different soil layers was measured before winter, at the beginning of spring growth and after harvest (Wienforth 2011). On average once a week N_2O fluxes from the soil were measured (Senbayram 2009). The soil water content as a factor influencing both nitirification and denitrification was simulated in a dynamic layered model based on a solution of the water content based formulation of the Richards equation. LAI and crop height as input variables for the Penman-Monteith equation for evapotranspiration were interpolated linearly between the measurement dates. Rooting depth and root distribution were estimated based on temperature sum since sowing and provide together with evapotranspiration a measure of water uptake by the crop. The amount of NH_4 as substrate for nitrification was derived from a coupled C and N mineralisation module based on four C pools. NO_3 is then transported by vertical water movement and taken up by the growing canopy. NO_3 also is the substrate for denitrification. Soil temperature was calculated by a simple heat conductance model depending on the soil water content (Fig.1).

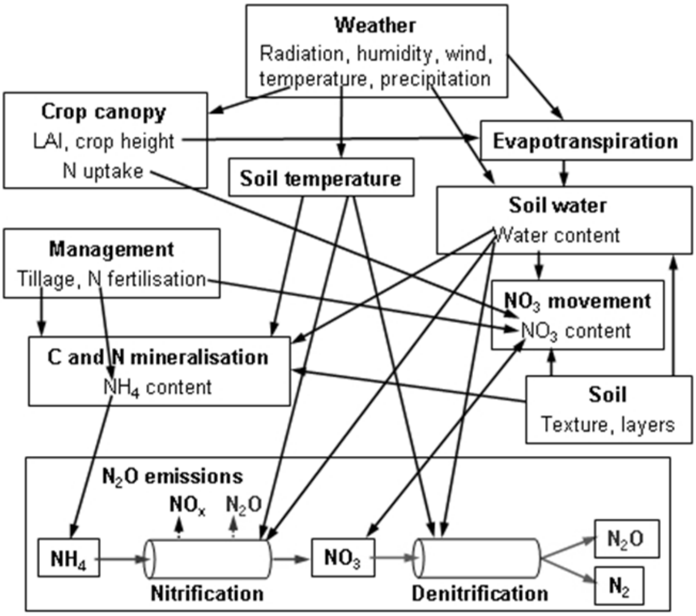

Figure 1: Simplified scheme of the coupled dynamic soil-plant-atmosphere model with processes and variables influencing N_2O emissions by nitrification and denitrification

Results and Discussion

The soil textures as parameters for the soil water dynamics were adjusted according to soil analyses and measured water contents. Due to the large soil heterogeneity at both sites there were large variations in the measured data between the four replications of each treatment. By dynamic modelling we were able to simulate soil water contents in the important layer of 0-30 cm depth quite accurately (RMSE = 0.031 cm^3/cm^3). The turnover rate for the pool of soil organic matter was adjusted for each fertilisation level separately while all other mineralisation parameters were taken from literature. With this parameterisation the model was able to simulate SMN in 0-30 cm well (RMSE = 21.6 kg N/ha). It should be noted that the model also provides plausible data for times without SMN measurements (main mineralization period in spring, fertilisation dates).

Conclusions

The presented model combines frequently available measured data (LAI, crop height, N uptake of the crop) and dynamic process oriented approaches (soil water dynamics, NO_3 dynamics, mineralization, nitrification and denitrification). This combination appears to be a good method to simulate the influencing factors for N_2O emissions from cropping systems as accurately as possible and to provide a tool to analyse the effect of different crop rotations and management measures on N_2O emissions.

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HOW DO DIFFERENT N SOURCES AFFECT N2O EMISSIONS FROM SOIL?

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Long-term food security may require fertilizer diversification and the simultaneous use of diverse N sources to meet population demands. This gradual transition from the intensive use of mined resources to a reliance on recycled and renewable nutrient pools (such as manures, sewage or stabilized organic residues) seeks to minimize pollution levels and land degradation. Such evolution requires the incorporation of new organic nitrogen sources into traditional N fertilizer practices. In order to assess the environmental impact of N source alternatives, a laboratory experiment was carried out to study the N_2O losses from different commercially available organic N sources.

Materials and Methods

Soil was collected from an ungrazed, unfertilized, freely draining Eutric Cambisol with a sward dominated by *Lolium perenne* L. and *Trifolium repens* L. situated in Abergwyngregyn, Gwynedd, North Wales (53º14'N, 4º1'W). On return to the laboratory the soil was sieved to pass 5-mm. The experiment contained six treatments including: (i and ii) two different commercially-available dissolved organic nitrogen (DON)-based fertilizers, (iii and iv) an amino acid mixture ± glucose addition (AA and AA+$C_6H_{12}O_6$), (v) ammonium nitrate (AN), and (vi) an unamended control treatment. The DON based fertilisers were Avant Natur® (AvN) and Avant Natur® 8-4-6 (AvN+NPK) (provided by COMPO Iberia S.L.). The amino acid mixture consisted of an equimolar mixture of fourteen amino acids. Within 48 h of collection from the field, 100 g portions of field-moist soil were placed in individual 110 ml polypropylene pots and amended with each of the different N fertilisers indicated above. The different fertilisers were dissolved in water respectively in order to obtain the solutions with the same concentration, which were applied at a dose of 50 ml kg^{-1} (equivalent to an N dose of 100 kg ha^{-1}) whilst distilled water was added to the control treatment. The bottles were then covered with Parafilm® (to conserve moisture but allow gas exchange) and placed in a climate-controlled chamber at 10ºC. At various times after the application of the treatments (0, 5, 10, 14, 21, 35, 49 and 63 d), four independent replicates from each treatment were removed for gas sampling. Pots were incubated in hermetic jars with a volume of 1 L at 10ºC for 1 h. Emission rates were calculated taking into account the increase in the concentration of each gas during incubation time (Menéndez et al., 2008). Gas samples (20 mL) from the atmosphere of the hermetic jars were stored in vials of 12 mL and later analyzed by gas chromatography (GC) (Agilent, 7890A) equipped with an electron capture detector (ECD) for N_2O detection. A capillary column (IA KRCIAES 6017: 240 ºC, 30 m x 320 mm) was used and the samples were injected by means of a headspace autosampler (Teledyne Tekmar HT3) connected to the gas chromatograph. Cumulative gas production during the experiment was estimated by averaging the fluxes of two successive determinations, multiplying that average flux by the length of the period between the measurements, and adding that amount to the previous cumulative total.

Results and Discussion

Daily fluxes ranged between 0.25 and 15.90 mg N_2O kg^{-1} dry soil (data not shown). As expected, the addition of the different sources of nitrogen significantly increased the cumulative loss of N_2O relative to the unfertilized treatment (Table 1). Maximum losses were observed in treatments where only organic N was added (AvN and AA). The AvN+NPK presented a significant reduction of N_2O emissions in comparison to the product without mineral nitrogen (AvN). Barrett and Burke (2000) described that N immobilization increased with soil organic matter content. In our case, not only N immobilization happened, the reduction with respect to AvN treatment suggests that the mineralization of the organic N applied was also reduced. In the same way, the addition of glucose to the amino acids also reduced slightly the losses, although not significantly.

Table 1. Cumulative emissions of N_2O up to day 63.

	μg N_2O kg^{-1} dry soil
AvN + NPK	3083 b
AvN	5238 a
AN	2498 bc
AA	4429 ab
AA + $C_6H_{12}O_6$	3771 ab
Control	1019 c

Different letters within a column indicate significantly different rates ($P< 0.05$; n=4)

Conclusion

The N_2O emissions that were induced by after the addition of DON differ in magnitude. The DON origin seems to play an important role on the magnitude of the losses. The commercial product Avant Natur resulted in higher N_2O losses. Nevertheless, its combination with mineral nitrogen (AvN+NPK) favoured N immobilization, resulting in a significant reduction of losses. To further advance our understanding, this trial should be performed in the presence of plants as we hypothesize that plant N metabolism would change N_2O emission rates.

Acknowledgments

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DENITRIFICATION RATE UNDER POTATO PRODUCTION: EFFECT OF THREE NITROGEN FERTILIZER SOURCES IN EASTERN CANADA

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Agriculture accounts for 72% of the Canadian nitrous oxide (N_2O) emission (Environment Canada 2012). Nitrogen (N) fertilizers are the main drivers that contribute to increasing N_2O emission (Gregorich et al. 2005). The denitrification process is the predominant source of N_2O emissions in humid environments (Burton et al. 2012). Rate of denitrification in soil is regulated by various environmental factors including soil water content, temperature, soil pH, redox potential, nitrogen oxide concentrations and availability of carbon. It is also influenced indirectly by many crop management factors such as fertilization, tillage, drainage and irrigation systems. Denitrification is generally promoted under high soil moisture conditions where oxygen is limited, and NO_3 and organic C are available for denitrifying. Large inputs of nitrogen (N) are required to optimize yield and quality of potato (*Solanum tuberosum* L.) and consequently may result in a high potential for N losses including N_2O emissions associated with the denitrification process (Cambouris et al. 2007; Zebarth and Rosen 2007).

This three-year study compared the effect of three fertilizer N sources [ammonium nitrate (AN), ammonium sulphate (AS) and polymer coated urea (ESN, Environmentally Smart Nitrogen)] band applied at 200 kg N ha^{-1} (N_{200}) and an unfertilized control (N_0) on denitrification rate (DR) from irrigated potato production. The site was located near Quebec City in eastern Canada. The site had well-drained sandy soils classified as Aquic Haplorthods with organic matter content of 5.2% and pH of 5.3. The DR was measured on four treatments and three blocks. The ESN was applied all at planting and AN and AS were split 40% at-planting and 60% at-hilling. The DR was measured biweekly from planting to harvest (10 sampling dates per year) at two positions (ridge and furrow) using the acetylene blockage technique (Drury et al. 2007).

In 2011, the DRs were significantly greater with N_{200} (2.12 µg N_2O-N kg dry soil^{-1} d^{-1}) than N_0 (0.76 µg N_2O-N kg dry soil^{-1} d^{-1}) (Figure 1). The three N sources (AN, AS and ESN) produced similar DR. The N source by position interaction indicated that DR measured in the hill varied with treatment N_0=AS2O-N kg dry soil^{-1} d^{-1}) compared with the furrow (0.71 µg N_2O-N kg dry soil^{-1} d^{-1}). The position by sampling date interaction analysis revealed that position had a greater effect on DR early in the growing season when soil nitrate concentration in the hill is elevated. In each year, DR varied significantly among sampling dates in response to intensive water input (precipitation & irrigation; >20 mm) occurring 24-72 hours before measurement. In fact, the highest seasonal mean DR (2.99 µg N_2O-N kg dry soil^{-1} d^{-1}) occurred in 2010 when the overall water input in the 72 hours before the measurements was 125 mm including four water input events of more than 20 mm (Figure 2). In 2011, the seasonal mean DR was 1.68 µg N_2O-N kg dry soil^{-1} d^{-1} and the overall water input in the 72 hours before the measurements was 137.6 mm, but with 1 event x > 20 mm. In comparison, the seasonal mean DR in 2012 was lower at 0.76 µg N_2O-N kg dry soil^{-1} d^{-1}, which was attributed to the very low overall water input in the

72 hours before the measurements at 47.2 mm, with one episode of more 20 mm of water input.

Our results confirm that source of N fertilizer can affect DR, but these effects are often small compared with differences resulting from environmental conditions and cultural practices such as water input occurring until 72 hours ahead of the measurements.

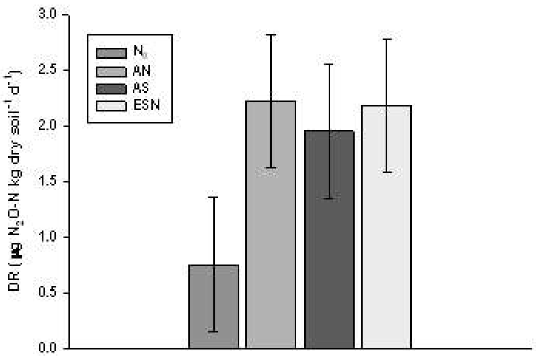

Figure 1. Denitrification rate (µg N_2O-N kg dry soil^{-1} d^{-1}) as influenced by the N fertilizer treatments.

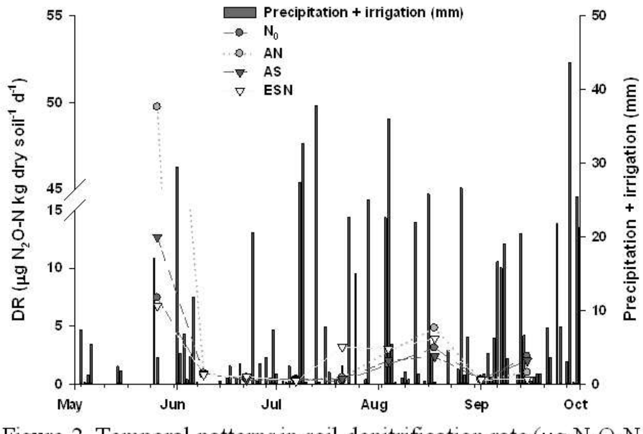

Figure 2. Temporal patterns in soil denitrification rate (µg N_2O-N kg dry soil^{-1} d^{-1}) in 2010.

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IMPACT OF A NITRIFICATION INHIBITOR DICYANDIAMIDE (DCD) ON N2O EMISSIONS FROM URINE APPLIED TO UK GRASSLAND

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The current UK GHG inventory predominantly uses the standard Tier 1 methodology of the Intergovernmental Panel on Climate Change (IPCC, 1997) for estimating N_2O emissions from soils, which is generalised and simplified. The UK's Greenhouse gas platform programme (www.greenhousegasplatformprogramme.org) is a large project with the aims of improving the methodology for calculating UK emissions. At present the default emission factor (EF) used for excreta deposited during grazing is 2%. The presence of livestock in grazed grassland is expected to influence gas emissions in a different way to synthetic fertiliser due to the returns of N and C in excreta. Urine, due to its N content and water deposited on the soil in each urination event has large potential for emissions. Determination of accurate EFs for urine deposition and implementation of mitigation strategies are of major importance for UK agriculture. In this study we measured N_2O emissions at three grassland locations in the UK, from urine applications to soil in the spring, summer and autumn of 2012 and investigated the effect of the nitrification inhibitor, DCD, added to the urine.

Materials and Methods

Experiments took place at 3 UK grassland sites: at Beacon Field (BF, clay loam) in SW England, Crichton (CRI, sandy loam) in SW Scotland and Hillsborough (HILLS, sandy clay loam) in Northern Ireland between March 2012 and September 2013. The treatments applied on each site were: urine, urine + DCD and a control. Emissions of N_2O were measured for a year using 5 static chambers per treatment-plot. Three plots were setup for each treatment in a randomised block design. Urine was applied with or without DCD at N rates between 353 and 480 kg N ha^{-1}. Gas sampling was frequent (daily) immediately after urine application but became less frequent as time went on and totalled around 30 sampling occasions in the year. Cumulative emissions were estimated using the trapezoidal method. Simultaneous soil samples were collected to measure soil moisture, ammonium and nitrate. The urine was analysed for pH, DM, total C, total N, urea, NH_4^+-N, NO_3^--N, hippuric acid, allantoin and creatinine. Climatic variables were monitored for the full year.

Results and Discussion

Emissions from the urine treatment were highest in spring at HILLS and BF, and in summer at CRI (Figure 1). Urine driven total emissions were largest at BF (13257 g N/ha). Total rainfall for the 12 months of measurements was similar at all sites (Table 1). Maximum temperatures were generally lower at CRI but the minimum was higher than at the other sites. The annual EFs varied between 0.05 at HILLS following the autumn application to 2.96 at BF in the spring. They were generally larger following the spring application except for CRI where it was larger following the summer application (Table 2). The EFs were generally lower than the IPCC default value, indicating a potential overestimation of emissions in the UK agricultural inventory of greenhouse gases. Application of DCD was effective in reducing N_2O emissions in the spring at all three sites. However it did not significantly reduce emissions at any of the sites in autumn and summer. Overall a mean reduction of 45% was achieved. The efficacy of the inhibitor in reducing N_2O emissions was affected by the season in which it was applied, resulting in small or no effects in the summer. In the autumn the efficiency varied from no effect to a small effect. The largest effect was in the spring when temperatures were cool. Effectiveness of DCD in inhibiting nitrification has been reported to depend on season of application in New Zealand (Moir et al., 2012) where the inhibitor activity was highest in autumn/winter/spring.

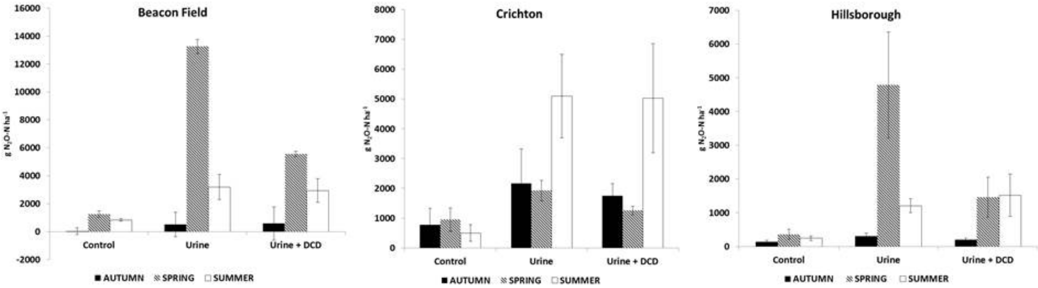

Figure 1. Cumulative N_2O fluxes from the three sites.

Table 1. Yearly weather at the three sites

Site	Season	Rainfall total, mm	Max/Min Temp ºC
BF	autumn	1243.41	-4.75/27.7
BF	spring	946.68	-4.75/18.8
BF	summer	1285.46	-4.75/29.6
CRI	autumn	1142.2	-1.7/22.2
CRI	spring	1325.9	-1.7/20.1
CRI	summer	1262.7	-1.7/19.3
HILLS	autumn	1092.6	-6.5/27.2
HILLS	spring	1191	-6.5/23.4
HILLS	summer	1130	-6.5/22.3

Table 2. Resulting emission factors for urine application with/without DCD

N_2O EF (%)	Season	Urine	Urine+DCD	DCD efficiency,%
	Spring	0.20	0.06	69
Crichton	Summer	1.09	1.08	1
	Autumn	0.32	0.23	28
	Spring	1.02	0.25	75
Hillsborough	Summer	0.28	0.38	-36
	Autumn	0.05	0.02	60
	Spring	2.96	1.00	66
BF	Summer	0.55	0.48	13
	Autumn	0.11	0.12	-9

Conclusions

There was variability in N_2O emissions between the three sites, possibly due to differences in soil type and climatic variation at the different locations. The effectiveness of DCD at reducing N_2O emissions was consistent with published results and showed a larger inhibitory effect in the spring and autumn and little or no effect in the summer.

Acknowledgement

UK Department for Environment, Food and Rural Affairs (DEFRA) and the Devolved Administrations for financial support.

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EFFECTS OF PERI-URBAN NH_3 EMISSIONS ON THE CONCENTRATION OF ATMOSPHERIC PARTICULATE MATTER IN THE CITY OF MILAN (ITALY): A PRELIMINARY STUDY.

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The prime emitting source of ammonia (NH_3) is agriculture, including animal husbandry and NH-based fertiliser application (Bouwman et al., 1997); the sector is estimated to be responsible to over 95% of NH_3 emissions in Italy (ISPRA, 2011). Once in atmosphere, NH_3 may be deposited as wet or dry form, or react with sulphuric (H_2SO_4), nitric (HNO_3), and hydrochloric (HCl) acids producing the relative salts that contributing significantly to produce secondary inorganic ambient particulate matter (PM) PM10 and PM2.5 (particulate smaller than 10 and 2.5 µm in aerodynamic diameter, respectively). NH_3 emissions assume a key role in the production of PM, especially in the urban areas, where atmospheric acids are naturally abundant, whilst NH_3 is limiting. Therefore it becomes essential to understand how peri-urban agricultural areas may affect the formation of PM in urban areas. NH_3 emissions in Italy are concentrated in the north (Skjøth et al., 2011), where most of the livestock are bred (ISTAT, 2011). In this framework, the agricultural lands around the city of Milan, one of the most polluted (Perrino et al., 2013) and populated areas of northern Italy and EU, are suitable to assess the influences of NH_3 emissions toward urban area. This preliminary study starts on the assumption that a direct determination of the influence of all the agricultural activities in urban areas is challenging mainly due to the complexity of the urban pollutants and the uncertainty of the NH_3 sources determination, therefore the aim is to quantify the levels of NH_3 concentrations in the urban fabric following spreading of fertilisers in the agricultural lands surrounding this area.

Materials and Methods

The study was performed for 11 weeks from 1st February to 19th April 2013, in a period chosen before and after the interdiction of the spreading of nitrogen fertilisers (91/676/EEC and subsequent amendments). The location, in the sub-urban north area of Milan (municipality of Cusano Milanino), is characterised by the presence of a 9 km^2 agricultural area isolated from wooded, productive and residential areas. Here a sampling point in the agricultural context, a field of 10 ha covered by winter wheat, and one at 2000 m far from this, in the urbanised area towards the city, have been identified. During the monitoring period, a nitrogen fertilisation of 33 kg N ha^{-1} as NH_4NO_3 was applied into the tested field at 14th March 2013. The measures were carried on by using time averaged passive samplers ALPHA (Adapted Low-cost Passive High Absorption, Tang et al. 2001) positioned at 2 m height into the agricultural field and on the top roof of a two-floors building for the urban sampling point. Samplers have been changed once per week and for each sampling point, a series of three replicates were employed. The operating principle of ALPHA samplers is based on the capture of gaseous NH_3 on acid support (13% of citric acid), protected by a membrane to avoid particle contamination and to establish a turbulence-free diffusion path between the membrane and the collection filter. These tools are designed to measure NH_3 air concentration from less than 1 μg m^{-3} to over 1 mg m^{-3} (Carozzi et al., 2013). After the exposure the filters were extracted in deionised water and the N-NH_4^+ content determined by the indophenol blue colorimetric method and spectrophotometer.

Results and Discussion

Figure 1 shows NH_3 concentration levels in the agricultural and in the urban sampling locations, together with the average temperature and the rain events. Results show a similar behaviour between the two NH_3 concentrations in agricultural and urban sampling locations, respectively, with a general fluctuating trend. No growing of the concentrations occurred after the period of interdiction of spreading 16th February), how would be expected. In the period before the fertilisation event (14th March), the NH_3 concentrations detected in the agricultural location resulted higher of 0.8 μg NH_3 m^{-3}, on average, than the urban location. After the fertilisation the gap between the two locations was progressively reduced to 0.3 μg NH_3 m^{-3} to reach, towards the end of the monitoring period, to a concentration of 1.8 μg NH_3 m^{-3} higher for the urban location than the agricultural one. This fact highlights that the NH_3 emissions affect both the source, the agricultural areas, that the surrounding urban areas. This behaviour can be explained by the weather conditions in the study period. Precipitation, inversely correlated with the NH_3 volatilization (Bouwman et al., 1997), was abundant during the trial (+57% than the same period from 1990; Figure 1), and was one of the fundamental cause of a general delay in the distribution of fertilisers in the studied area. Air temperature, strictly related to the NH_3 volatilisation phenomenon (Sommer and Hutchings, 2001), remained relatively low until the end of the monitoring period, where it grew positively accompanying NH_3 emission levels. Moreover, a significant score of the regression analysis ($P < 0.05$) was obtained between the values of NH_3 concentrations and the averaged air temperatures.

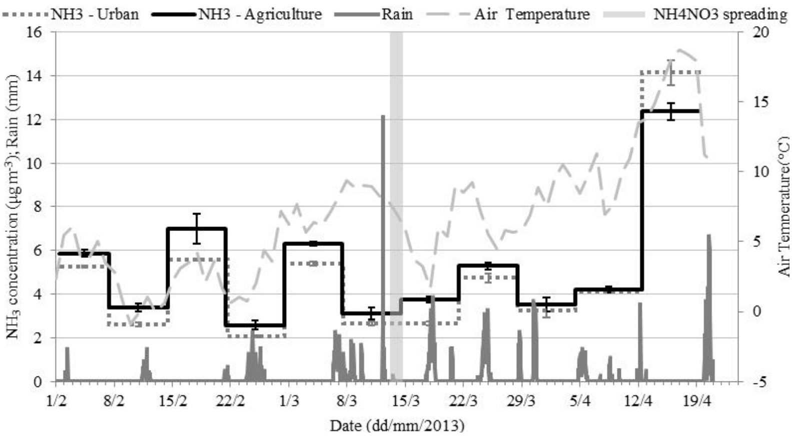

Figure 1. NH_3 concentrations in the agricultural and urban studied sites, together with rain and the daily averaged temperature in the monitoring period.

Conclusions

Despite preliminary and of short period, the study highlighted that NH_3 emissions from the agricultural areas may influence the surrounding urbanised areas. For the future, an improvement in the survey on multiple and different sites of the peri-urban and urban areas, is needed.

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DO HIGH NAPHTHALENE CONCENTRATIONS IN BIOCHAR JUSTIFY THE REDUCTION IN SOIL NITROUS OXIDE EMISSIONS?

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Biochar, a charcoal-like substance produced from the pyrolysis of residual biomass, has been found to decrease nitrous oxide emissions by an average of 54% in diverse soils and environmental conditions (Cayuela *et al.*, 2014). The causes and mechanisms of this reduction are still controversial. One of the proposed hypotheses is the release of polycyclic aromatic hydrocarbons (PAHs) that are present in biochar as a result of the pyrolysis process. Naphthalene is the major PAH found in biochar, comprising up to 60% of total PAHs. Naphthalene has been found to influence nitrification (Chang *et al.*, 2002) and some studies noted a correlation between PAH concentration in the biochar and the decrease in N_2O emissions (Wang *et al.*, 2013). The main objective of this research was to evaluate the influence of naphthalene (as main PAH found in biochars) in N_2O emissions after soil amending with two contrasting pine wood biochars, produced at a different pyrolysis temperature (350 and 550°C).

Materials and Methods

The soil, classified as Haplic Calcisol (FAO, 2006), was collected from the topsoil (0-25 cm) of an agricultural field (southern Spain), air-dried and sieved (< 2 mm) before its use. It had a loam texture (32% sand, 42% silt, and 26% clay), pH of 7.8, 501.1 g kg^{-1} $CaCO_3$, 8.1 g kg^{-1} total organic-C and 0.8 g kg^{-1} total N. Two biochar samples produced from pine wood by slow pyrolysis at 350 and 550°C (B350 and B550, respectively) were evaluated. B550 showed an initial higher PAH content (8.9 compared to 2.1 mg kg^{-1}; sum of the EPA's 16 priority pollutants). Biochars were homogenised and ground in a stainless steel mill to < 2 mm.

The incubations were carried out in 250 mL plastic jars, containing 100 g (oven-dry weight basis) of soil or biochar-soil mixture. The biochars were mixed thoroughly with the soil in a proportion of 2% (w/w on a dry weight basis) and a solution with KNO_3 was added to adjust moisture to 90% of the water filled pore space and apply 50 mg N kg^{-1} soil. The jars were incubated in darkness under aerobic conditions at 25°C for 21 days. The following treatments were conducted (4 replicates per treatment) in a randomized block design: control soil (unamended); soil+B350; soil+B550; soil+naphthalene; soil+B350+naphthalene; and soil+B550+naphthalene. Naphthalene was added to reach the maximum value allowed by the IBI (2014) for PAHs in biochar samples (20 mg kg^{-1}). Changes in the concentration of N_2O in the headspace of the jars were measured 60 min after closing the jars with a photo-acoustic gas monitor 1412i (Lumasense Technologies). Cumulative N_2O emission data were subjected to a two-way analysis of variance, considering biochar and naphthalene addition as factors.

Results and Discussion

All treatments showed the peak of N_2O emissions within the first 24 h of incubation (data not shown). After that, biochar-treated soils presented a second peak of emissions (much lower) and finally N_2O emissions reached a background level in all treatments after 7 days. Biochar type had a significant effect on cumulative N_2O emissions (P=0.000), showing the soil treated with B350 an increase in N_2O emissions (+44%), while that treated with B550 a decrease (-29%) compared to the control soil at the end of the experiment (Fig. 1). These results agree well with previous studies, indicating that biochars produced at high pyrolysis temperatures generally lead to the greatest N_2O reductions (Spokas and Reikosky, 2009). Although naphthalene addition reduced N_2O emissions (Fig. 1): -7% (control), -6% (B350) and -9% (B550) with respect to non-naphthalene treatments; this effect was not statistically significant (P=0.079). In addition, a no significant biochar×naphthalene interaction was found (P=0.972).

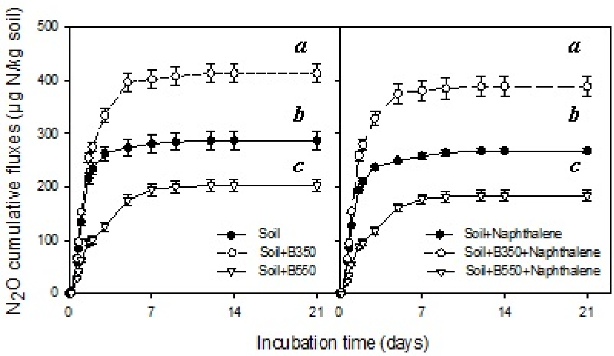

Fig. 1. Cumulative N_2O fluxes in control and biochar treatments, without and with naphthalene addition (mean value ± standard error). Letters show significant differences after 21 days according to the Tukey's test (P=0.05).

Conclusions

This study demonstrates that naphthalene (the most abundant PAH found in biochars) minimally contributes to reducing N_2O emissions in an agricultural soil amended with 2% biochar. The different behaviour showed by both biochars (produced at low and high pyrolysis temperature) seems to respond more to other compositional and/or structural properties of biochar than to naphthalene concentration.

Acknowledgement

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QUANTIFICATION OF NITROGEN LEACHING TO SHALLOW WATER TABLE IN LARGE SCALE IRRIGATION SCHEMES: A CASE STUDY IN THE MEDITERRANEAN LANDSCAPES

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Irrigated agriculture with fertilization remarkably increases land productivity and enhances crop diversification in the arid and semi-arid areas of the world, including the Mediterranean region. Since nitrate (NO3) leaching is frequently the most important nitrogen (N) loss process in irrigated agriculture, it has considerable potential for contaminating groundwater and surface water at the irrigation district level. Therefore, as indicated by Cavero et al. (2003), management of the irrigation systems, fertilization practices and land use are the key factor in dictating whether significant amounts of N are lost or not. In this regard, high NO3 concentrations in root zone drainage are of major concern in areas of extensive irrigation development, where much of the recharge to shallow water table and/or groundwater percolates through irrigated cropland. In irrigated areas, best management practices for N fertilization and irrigation water are being promoted to reduce NO3 losses to shallow water table. In order to take timely precaution for reducing N loss to shallow water table, it is important to figure out spatial and temporal changes in N dynamics at the district level. Therefore, Akarsu Irrigation District (AID, 9 495 ha) with excess use of N fertilizers was studied with the following objectives: (1) identification and assessment of spatial and temporal variability of NO3 concentrations in the shallow water table, (2) quantifying unitary NO3 loads based on the soil series characteristics, (3) driving hypsometric NO3 concentration curves for assessing the N fertilization practices in non-irrigation (rainy) season, start of irrigation, peak of irrigation and end of irrigation periods.

Materials and Methods

This study was conducted in the AID, located in the Mediterranean region of Turkey. In the present study, groundwater samples from 108 drainage observation wells, up to 3 m depth, were taken four times a year, i.e., in the non-irrigation (rainy) season, at the start of irrigation season, at the peak irrigation season, and at the end of irrigation season of 2012. Nitrate concentrations of water samples were determined in the laboratory by Standard Methods (1998). Geo-referenced data were processed through using spatial analyst tools of GIS (Cetin et al., 2012), and concentration distribution maps were drawn with the inverse distance weighting interpolation procedure. Zonal statistics of the themes were employed to derive hypsometric NO3 curves exclusive for the sampling periods. Nitrate loads were calculated by using NO3 concentrations, porosity of soil series, and shallow water thickness between water table and calculation base that is 3 m from soil surface.

Results and Discussion

In 2012, areal averaged NO3 concentrations of shallow water table were 27, 19, 15 and 10 mg l-1 in the rainy season (mid-January), at the start of irrigation season (April), at the peak irrigation season (mid-July) and at the end of irrigation season, respectively. Nitrate concentration was found the highest in rainy season due to the fact that wheat was fertilized in the rainy period, and winter rains caused to leach the fertilizer to the shallow water body. However, variability was the least (CV=40%) in the rainy period due to the limited agricultural practices and the highest (CV=74%) at the end of irrigation season (end of September) which is the end hydrological year 2012. The very high spatial variability may be attributed to the non-uniform irrigation applications, cropping pattern and fertilizer applications to diversified crops in the district. It seems pretty obvious that NO3 leaching to shallow water table is not time invariance, indicating anthropogenic degradation of the environment. Hypsometric salinity curves derived uniquely for the study area, and sampling periods (Figure 1) revealed that only negligible percentage of the area (less than 0.2%) had NO3 concentrations greater than 50 mg l-1. However, the percentage area of NO3 concentration greater than 30 mg l-1 was 42, 14, 9 and 3 in winter, at the start of irrigation season, peak irrigation season and end of irrigation season, respectively (Figure 1). Unitary NO3 load maps suggested that porosity of soil series was as important as the NO3 concentrations of the shallow water table. Nitrate loads showed a clustered behavior based on the cropping pattern.

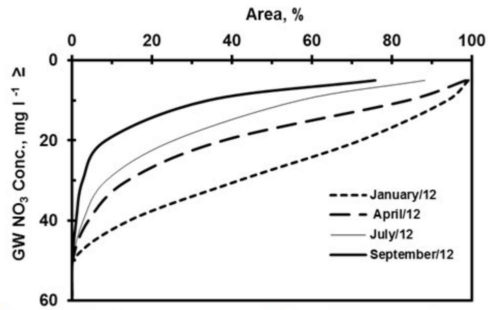

Figure 1. Hypsometric nitrate curves for the District in four sampling periods

Conclusions

The most significant part of NO3 leaching to shallow water table occurs in winter by rainfall. Although nitrate concentrations are below the critical threshold level of 50 mg l-1, some parts of the area are potentially risky, indicating the importance of NO3 monitoring during the hydrological year in the cropping system. Nitrate leaching is not time invariance, and depends on mostly cropping pattern, indicating the impact of fertilization doses on the leaching amount.

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OVERWINTER TRANSFORMATIONS AND FATE OF FALL-APPLIED NITROGEN AS A FUNCTION OF SOIL CONDITIONS – A MULTI-ECOZONE EXPERIMENT

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There is growing evidence that biological activity and N cycling are sustained in soils under freezing conditions (Maljanen et al., 2007; Drotz et al., 2010; Virkajärvi et al., 2010), and that a significant portion of N present in soils in the fall may be transformed and lost during the non-growing season (Clark et al., 2009; Chantigny et al., 2014). Climate models are currently predicting that, in North America, global warming will cause a reduction in snow cover and increase soil frost penetration and freeze-thaw cycles (Henry, 2008), which could hinder soil biological activities (Groffman et al., 2001). Information on the extent of N losses in the non-growing season as a function of winter conditions (e.g. snow cover depth, frost penetration) is scant, but could help in predicting how soils in northern regions may respond to global warming. Our objective was to determine if and to what extent fall-applied N may undergo transformations and be lost from agricultural soils during the non-growing season under contrasted winter conditions.

Materials and Methods

The NH_4-N fraction of pig slurry and dairy cattle slurry were enriched with ^{15}N, and ^{15}N-labelled ammonium sulfate was included as a no-carbon reference treatment. All N sources were applied to bare loamy soils in late fall (November) at four sites located in four Canadian ecozones: (1) Pacific Maritimes; Mean annual temperature [MAT], 10.5°C; Mean annual precipitation [MAP], 1755 mm; average snow depth [ASD], < 1cm); (2) Prairies; MAT, 5.7°C; MAP, 383 mm; ASD, 3 cm); (3) Mixed Wood Plain; MAT, 6.3°C; MAP, 914 mm; ASD, 13 cm); (4) Boreal Shield; MAT, 4.2°C; MAP, 1213 mm; ASD, 44 cm). All sites were cropped to spring barley (*Hordeum vulgare*) or spring wheat (*Triticum aestivum*), and N treatments were applied after harvest. The experiment was replicated in 2009-10 and 2010-11 at all sites. Soils were sampled (0-30 cm) on the week of N application, in November, and at intervals until next spring (May). The recovery of applied ^{15}N in soil NH_4-N, NO_3-N and organic (immobilized) N pools was measured. Soil temperature was also monitored at the 5, 20 and 50 cm depths.

Results and Discussion

Fall-applied $^{15}NH_4$-N was nitrified and immobilized throughout the non-growing season at all sites. Transformations occurred rapidly at the warmest site (Pacific Maritimes), closely followed by the site with deep snow cover (Boreal Shield) (Fig. 1). Transformations were more gradual at the other two sites (Mixed Wood Plain; Prairies) where colder soil temperatures and deeper frost penetration were recorded. In the following spring (May), residual ^{15}N was essentially recovered as NO_3-N and organic (immobilized) N, and in all cases recovery of fall-applied ^{15}N was low and varied from 10 to 50%. In general, more $^{15}NH_4$-N was immobilized and more ^{15}N was recovered in May with manure than ammonium sulfate, suggesting that the presence of fresh carbon in the manure stimulated N immobilization and retention in soil.

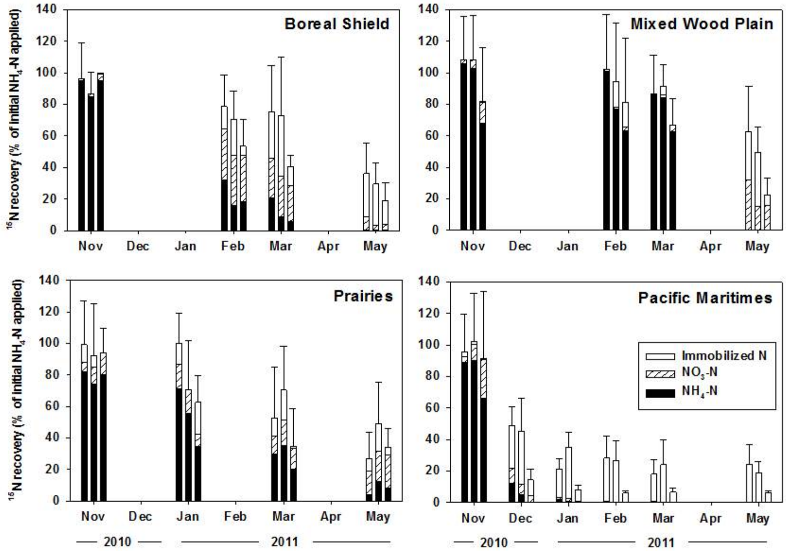

Figure 1. Recovery of fall-applied $^{15}NH_4$-N in soil N pools as a function of time after application to loamy soils at four sites with contrasted winter conditions (results for 2010-2011 are presented here). At all sites, for a given sampling date, the left column shows results for ^{15}N applied as pig slurry, the middle column is for dairy cattle slurry, the right column is for ammonium sulfate.

Conclusion

A substantial portion of fall-applied NH_4-N was immobilized, nitrified, and lost during the non-growing season, even at sites with the coldest soil conditions (Ottawa, Lethbridge). This suggests that soil microflora may adapt to more severe freezing conditions caused by global warming and decreased snow cover depth, thereby leading to maintained nitrogen cycling and potential loss.

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N2O LOSSES IN SOIL WITH SHALLOW GROUNDWATER: EFFECT DUE TO DEROGATION OF NITRATES DIRECTIVE. A CASE STUDY IN VENETO.

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Numerous authors have observed that low nitrogen concentration and percolation losses from cultivated fields can be associated to shallow groundwater conditions despite their intrinsic high vulnerability. Biological denitrification stimulated by anaerobic conditions may, in fact, promote nitrates abatement in water but it is necessary to monitor nitrous oxide (N2O) emissions during the denitrification process. Aquifers can directly influence the gas balance by acting as a sink of N2O, which can accumulate in groundwater due to its high water solubility (Minamikava et al., 2013). Considering the higher N input levels allowed by the recent derogation of the Nitrates Directive obtained by the Veneto Region (NE Italy), this study assesses the effects of a) groundwater level and b) N input (mineral+organic) on soil N2O emissions and dissolved N2O in shallow groundwater.

Materials and Methods

A two-year experiment (2011-2012) on continuous maize (Zea mays L.) was conducted in 12 loamy-soil lysimeters (1 x 1 m2 width x 1.5 m depth) in Veneto Region (NE Italy). The factorial combination of three water table levels (free drainage – FD, water table at 60 cm – WT60 and water table at 120 cm – WT120 depths) with two N input levels (no derogation – 'Low' and derogation -'High') were evaluated. A mix of bovine manure and poultry litter was incorporated at sowing at 170 kg N ha-1 y-1 in Low and 250 kg N ha-1 y-1 in High. Complementary mineral N as urea was distributed 40% in base-dressing and 60% in top-dressing at 80 kg N ha-1 y-1 and 118 kg N ha-1 y-1 in Low and High, respectively. The experimental design was completely randomised with two replicates. Water input, exclusively supplied through irrigation, was 900 mm y-1, including 460 mm y-1 between sowing and harvest. Nitrous oxide flux rates from soil were measured using an automatic close dynamic chamber system (12 chambers) at chamber closure, at 25 and 50 minutes after closure. The samples were collected on a daily basis for the first seven days after fertilization and then every two weeks until the harvest. During the winter, on bare soil, only one sample per month was collected. Groundwater samples were collected in evacuated-vials and dissolved N2O concentrations were monitored every two weeks during the growing season. Differences in yearly N2O losses were tested applying a 3-way (fertilisation x water table x year) ANOVA.

Results and Discussion

Nitrous oxide emissions were affected by N fertilization at sowing. Emission promptly started the day after fertilization, peaking after about three days. The peak reached a maximum of 200 g N2O-N ha-1 d-1 in Low-FD in 2011 and 420 g N2O-N ha-1 d-1 in High-WT120 in 2012. A large portion of N2O (96% and 91% in 2011 and 2012, respectively) was emitted within 15 days after sowing. Top-dressed urea contributed to the residual 4% and 9% emissions in 2011 and 2012, respectively. Yearly cumulative emissions were higher in 2012 (2.29 kg N2O-N?ha-1?y-1) than 2011 (1.44 kg N2O-N?ha-1?y-1) (P< 0.01). N input affected N2O emission only in WT60 with higher values in High (2.37 kg N2O-N?ha-1?y-1) than Low (0.98 kg N2O-N?ha-1?y-1) (p< 0.05) (fig.1). Moreover, in Low treatments a negative trend was evident according to the water table depth as follows: FD (2.85 kg N2O-N?ha-1?y-1) >WT120 (2.03 kg N2O-N?ha-1?y-1) >WT60 (0.98 kg N2O-N?ha-1?y-1), also corresponding to a reverse trend in soil water content. These data could support the hypothesis that other biological processes not strictly requiring anaerobic conditions (e.g. mineralisation, nitrification) could have influenced the initial nitrate availability and N2O emissions. Moreover, the potential role of autotrophic nitrification (Bateman and Baggs, 2005) should be investigated. Annual direct N2O-N emissions as percentage of total N inputs did not exceed 1.3%, with an average of 0.6%. Dissolved N2O-N occurred in groundwater at very low concentrations ranging from 0.45 to 1.21 μg N2O-N l-1 H2O. No significant differences were observed according to the treatments. However, a significant positive trend in the concentration over the two years is worth noting, which increased from 0.6 to 1.1 between July 2011 and August 2012. Nonetheless, in term of mass balance the contribution of dissolved N2O was negligible, ranging from < 1.1 g ha-1N2O-N in WT120 to < 4.3 g ha-1N2O-N in WT60.

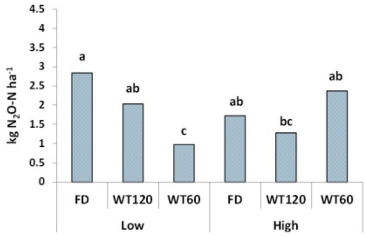

Figure 1. Mean (2011-2012) cumulative N_2O-N yearly emissions (kg $ha^{-1}y^{-1}$)

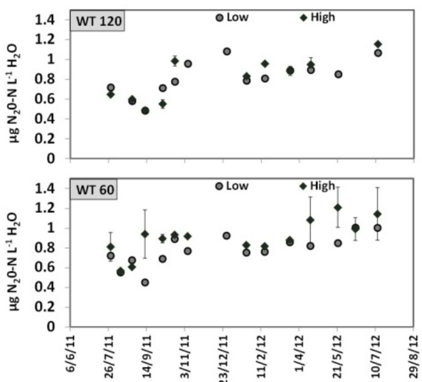

Figure 2. N_2O-N dissolved in groundwater.

LEGUMINOUS COVER-CROPS EFFECTS COMPARED TO NON-LEGUMINOUS ON NITRATE LEACHING AND NITROGEN SUPPLYING TO THE SUCCEEDING CORN AND SPRING BARLEY

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Nowadays in Europe and then in France, it becomes necessary to elaborate agricultural systems less reliant on synthetic fertilizer-N due to 1) the progressive rising of fertilizer prices, 2) the need of decreasing GHG (Green House Gaz) and ammonia emissions. Nevertheless, these systems should provide high yields and grain protein contents to satisfy the food security and market demands. So, the reduction of synthetic fertilizer-N must be counterbalanced by other nitrogen sources. Among them, leguminous cover-crops (LCC) have the advantage of introducing "free" atmospheric nitrogen without leading to a too strong change in the current agricultural systems. Since 1992 and the first Nitrate Directive program, agronomists have acquired wide knowledge about non-leguminous cover-crops (NLCC) but not on LCC due to their regulatory ban. Nitrate Directive update in 2009, allowing leguminous cover-crop under specific conditions, changed this situation and triggered a wide research program supported by several organizations in France with three questions to answer: 1) What is the effect of LCC on nitrate leaching? 2) What is their effect on nitrogen supplying of succeeding spring crops, in order to evaluate their potential to improve agricultural systems independence from synthetic fertilizer-N? 3) What are their technical specificities to obtain enough biomass, to provide the first two effects? This paper will put forward the main conclusions of points 1 and 2.

Materials and Methods

27 annual and 4 long-term experiments were carried out from 1991 to 2013 in several French regions and on the main soil types encountered in the country. All plots were statistically randomized with at least 3 replicates. LCC species studied were: pea, faba bean, vetch, several clover species and lentil. NLCC controls were: white mustard, fodder radish, rye-grass, spring oat, rye and phacelia. We also tested some mixtures of LCC and NLCC (MCC). Cover-crops termination dates ranged from November to February. Sowing dates ranged from mid-February to the beginning of April for spring barley, and from mid-March to mid-May for corn. Nitrate leaching was evaluated considering reduction of soil mineral-N content at the beginning of leaching period (compared to bare soil) in all experiments and with porous cup/lysimeters monitoring in two long term trials. N supplying to succeeding spring crop was calculated using soil nitrogen balance based on soil and plant N content measurements at the end of winter and at harvest. In 6 experiments, we also simulated N mineralization kinetics after cover-crop incorporation with INRA Lixim software (Mary et al. 1999) using regular soil mineral-N content measurements.

Results and Discussion

LCC led to a twofold less reduction of soil mineral-N content at the beginning of the leaching period than NLCC and MCC (figure 1). This result was confirmed by porous cups/lysimeters monitoring. Nevertheless, LCC effects on nitrate leaching was different from zero, allowing them to be used as nitrate catch crop in situations where leaching risk is low to medium (deep soils, no slurry applications). MCC showed the same display than NLCC, allowing them to be used in all agricultural situations. LCC showed the highest N supplying effect to succeeding spring barley and corn (figure 2-a). NLCC effect was variable and could sometimes lead to N supplying depletion (figure 2-a). The difference between LCC and NLCC is linked to their effects on soil mineral-N content before leaching and to their mineralization kinetics which imply different N dynamics release (figure 2-b). More than the type of cover-crop, it seems to be the C/N ratio which determines the mineralization kinetics and explain some differences between experiments. It is consistent with results obtained under laboratory conditions (Justes et al. 2009) and in well-known published results obtained in field trials (Laurent et al. 1995). The acquisition of N from soil and atmosphere allows LCC (and MCC) to reach very low C/N ratio and so to provide larger amount of N than NLCC.

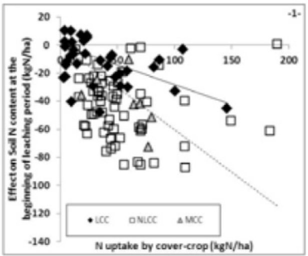

Figure 1: effect of LCC, NLCC and MCC on the reduction of soil mineral-N content at the beginning of the leaching period (compared to bare soil). Lines = linear models fitted with LCC (plain) and NLCC+MCC (dotted) data. Models are statically different (F test P-value < 0.0000).

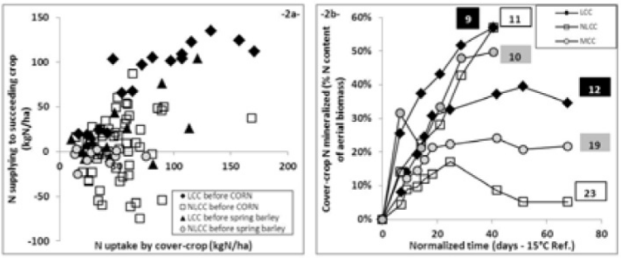

Figure 2: a- Nitrogen supplying to corn and spring barley of LCC (+MCC) and NLCC compared to nitrogen uptake / b- Nitrogen mineralization kinetics after cover-crops termination in 2 field trials. Labels = C/N ratio.

Conclusions

Our results showed that LCC and MCC could at once simultaneously reduce nitrate leaching and supply large amount of nitrogen to succeeding spring barley and corn. It is confirmed by a recent and large modelisation study conducted by INRA (Justes et al. 2012). The challenge is now to define technical specifications to grow LCC and MCC in most French agricultural situations.

Acknowledgement

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EFFECTS OF ORGANIC MATTER INPUT ON NITRATE LEACHING AND CROP YIELD IN ARABLE AND VEGETABLE CROPPING ON SANDY SOIL IN THE NETHERLANDS.

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Organic matter in soils fulfills vital functions in many ways for food production. However, organic matter can give uncontrolled release of nitrogen leading to high nitrogen leaching to groundwater in periods without crop uptake. In arable farming on sandy soils in the Southeast of the Netherlands, this is one of the reasons of exceeding the standard of 50 mg NO_3^-/l in groundwater of the EU Nitrate directive. Average nitrogen concentration in groundwater is 79 mg NO_3^-/l, while only 21% of the farms is below the EU-standard (Hooijboer & de Klijne, 2012). Other important reasons for high nitrate concentrations in groundwater are the intensive arable and vegetable crop rotations with a large share of leaching sensitive crops and the relative high nitrogen and animal manure inputs (mainly pig slurry). Two options are explored to reduce nitrogen leaching to groundwater: 1) No application of organic manure, causing less organic matter input and less nitrogen mineralization outside crop uptake periods and less nitrogen available for leaching to groundwater. 2) Organic farming with high organic matter input (cattle slurry and solid manure) and a lower total nitrogen input, causing more buffering of nitrogen in the soil. The question is how crop yields, nitrate concentrations in groundwater and soil quality are developing over time.

Materials and Methods

The options are tested on an experimental farm since 2001 in an intensive arable and vegetable six-year rotation. A comparison is made between three systems: 1) *system REG* with regular organic matter input from crop residues and animal manure, effective organic matter, (EOM) input about 1600 kg/ha, total N-input about 200 kg/ha; 2) *system MIN* with minimal organic matter input from crop residues only, EOM input about 900 kg/ha, total N-input about 170 kg/ha and 3) *system ORG* with an organic system with a higher organic matter input, EOM input about 2700 kg/ha, total N-input about 135 kg/ha. Since 2011, the crop rotation exists of the crops: potato, peas, grass (with clover in ORG), leek, barley, sugar beet (in ORG carrot) and maize. Nitrate concentrations in groundwater were monitored between 2005 and 2008 and in 2012. Crop yields were recorded every year. Chemical and biological soil quality parameters as nutrient contents, organic matter content, C/N-ratio, total N, available P, pH, CEC, fungal biomass, potential C and N mineralization, Hot Water Carbon and nematode groups were measured in 2011.

Results and Discussion

System REG has the highest nitrate content in groundwater, 120 mg NO_3^-/l on average in 2005-2008 and 66 mg NO_3^-/l in 2012. System MIN has a nitrate content in groundwater of 99 mg NO_3^-/l on average in 2005-2008 and 64 mg NO_3^-/l in 2012. The lower nitrate concentration in system MIN is caused by the lower EOM input in MIN. System ORG has the lowest nitrate content in groundwater, the only one below the EU threshold of 50 mg/l with 39 mg NO_3^-/l on average in 2005-2008 and 30 mg NO_3^-/l in

2012. The lower nitrate concentration in system ORG is caused by the lower total N-input, more use of cover crops and probably a somewhat higher water table than in system REG. Differences between 2005-2008 and 2012 are caused by different weather conditions and difference in crop rotation. 2012 had a wet summer and autumn compared to 2005-2008. In 2005-2008 lily was in the rotation, in 2011 lily was replaced by grass.

Since 2007, crop yields tend to be lower in system MIN compared to system REG. Differences are varying strongly from no difference in 2009 and 2010 up to 10% in 2012. There are indications that the magnitude of the difference is correlated with rainfall in summer period. Dry summers give larger differences. Crop yields in system ORG tend to increase to the level of system REG: crop yields of leek, maize and peas are in 2011 and 2012 comparable to system REG. Crop yields of barley and potato in system ORG are still much lower than system REG because of disease problems.

Chemical soil parameters as organic matter content, total N, available P and CEC were little lower for systems MIN and ORG compared to REG but still in range for good production. Potential N and C mineralization were higher in system ORG compared to MIN and REG, indicating higher microbial activity. The biological soil parameters have a large variation and a clear visible trend in differences between systems is lacking.

Conclusions

No input of organic matter with fertilization and organic agriculture are both measures to lower nitrate leaching. Only organic agriculture can reduce leaching below EU-standards. Reducing organic matter input with manure reduces crop yields after six years. Therefore it is not an economic viable option. Organic agriculture with high organic matter input increases crop yield. It is difficult to make the differences in strategy visible in measurement of soil parameters.
Uncertain is how the trends of leaching, yield levels and soil quality will develop on the long term. Therefore, the research is continued in the next years.

Acknowledgements

The writers of the article wants to thank Marc Kroonen and staff of the experimental farm Vredepeel for their work in the experiment and the Ministry of Economic Affairs and two regional divisions of the Dutch Federation of Agriculture and Horticulture (ZLTO and LLTB) for their financial contribution to the project.

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SIMULATION OF NITROGEN DYNAMICS FOLLOWING WINTER CEREALS UNDER FERTILIZED AND NON-FERTILIZED CATCH CROPS AND BARE FALLOWS

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In temperate humid climates, catch crops have proven to be a useful tool in the abatement of soil erosion, nutrient leaching and soil organic carbon losses. In Flanders (Belgium), the environmental policy allows farmers to apply manure after harvest of winter cereals at a rate of 60 kg N/ha, if they sow a catch crop before the 1st of September (on light textures) or before the 15th of October (on heavy textures). Farmers claim that fertilization stimulates catch crop growth and in that way increases benefits of catch crops. Nevertheless, the question was raised whether fertilizing catch crops would affect N losses during winter. A research project was initiated by the Flemish Land Agency (VLM) to investigate whether application of pig slurry to catch crops sown after harvest of winter cereals does not result in higher N losses from the soil between autumn and early spring compared to non-fertilized catch crops.

Materials and methods

Field experiments were installed during two consecutive years (2011-2013), each year on 4 locations with different soil textures. After harvest of winter cereals, pig slurry was applied at rates of 0, ±60 and ±120 kg total N/ha. On each location 3 to 4 common catch crop species were sown: white mustard (*Sinapsis alba*), Italian ryegrass (*Lolium multiflorum*), black oat (*Avena strigosa*) and a grass-clover mixture (*Lolium perenne, Trifolium repens, Trifolium pratense*). A bare fallow was included. Catch crops were sown on 2 different dates, 2-4 weeks apart. Soil mineral N content in the 0-90 cm layer (Nmin), aboveground dry matter yield (DMplant) and aboveground nitrogen yield (Nplant) were monitored. In addition, incubation experiments were carried out to investigate nitrogen mineralization from soil organic matter and from catch crops after incorporation. During winter (2011-2012), the upper soil layer (0-30 cm) was sampled for bare fallow (0 – 60 – 120N), white mustard (0 – 60 – 120N), Italian ryegrass (0N), black oat (0N) and grass-clover (0N) in Lemberge (sandy loam) and Rukkelingen-Loon (silt loam). Early sown catch crops were harvested on the 0N treatment in Lemberge and incubated according to their yields. All soils were maintained at a constant moisture content (50% water filled pore space), a temperature of 15°C and a relative air humidity of 70% during the incubation (3 months). The experiment was carried out in 3 replicates. On 7 sampling dates, Nmin content was determined with a continuous flow auto-analyzer. As in situ measurements of N losses by nitrate leaching and denitrification were practically unrealizable, N losses were simulated with the EU-rotate_N model (Rahn et al., 2010). Nmin, DMplant and Nplant measurements of field experiments and Nmin measurements of incubated non-fertilized bare fallows were used to calibrate unknown parameters in different modules of the model. Calibration was carried out in a stepwise manner, adding modules one by one. Calibrated parameters were selected based on a sensitivity analysis and calibrated using the parameter estimation program PEST. Parameter boundaries were defined for each parameter to prevent that calibration would lead to parameter values that deviate too much from the original model parameters. After calibration, the model was run to generate output on nitrate leaching, denitrification and nitrogen immobilization. So far, the model has only been applied on the field data of Lemberge (after winter barley on sandy loam, 2011-2012).

Results and discussion

Parameter calibration was successful: analysis of the residuals between simulated and measured values showed that only the highest values of Nmin were consistently underestimated by the model (Fig 1).

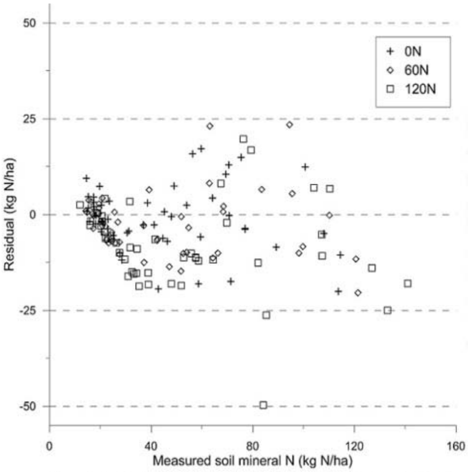

Figure 1: Residuals between simulated and measured soil mineral N for all treatments without (0N) and with fertilization (60 and 120 kg N/ha).

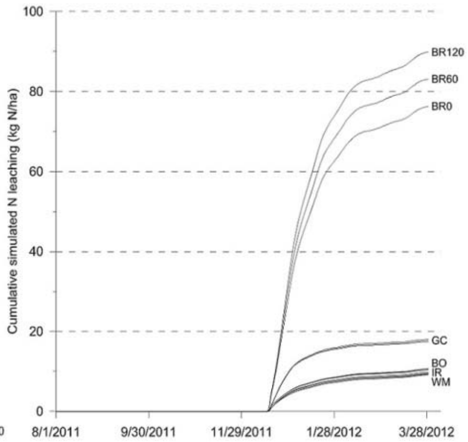

Figure 2: Simulated N leaching under bare fallow (BR) and under 4 early sown catch crops (GC: grass-clover, BO: black oat, IR: ryegrass, WM: mustard).

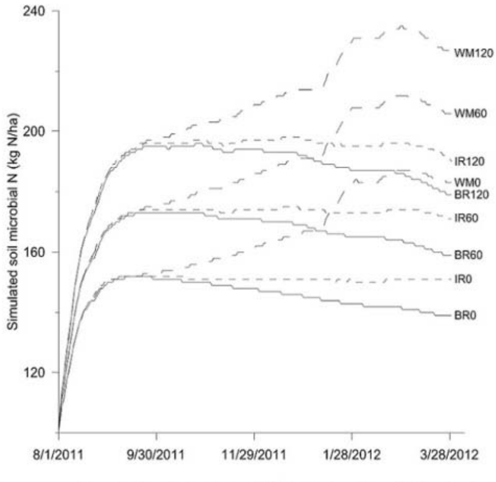

Figure 3: Simulated soil microbial N under bare fallow (BR) and under early sown white mustard (WM) and Italian ryegrass (IR).

Simulated nitrate leaching was lower under catch crops than under bare fallow (Fig 2). Nitrate leaching under grass-clover was higher than under the other 3 catch crops, but might have been slightly overestimated due to an overestimation of simulated N fixation by the clover. For both sowing dates no differences in nitrate leaching were found between fertilized and non-fertilized catch crops (second sowing date not shown). Nitrate leaching under bare fallow increased with fertilization, though differences were small due to considerable immobilization by the cereal stubble (Fig 3). From December onwards the immobilized N was released slowly. Under catch crops additional immobilization occurred from October onwards due to mineralization of dead leaves of the catch crops. This effect was most obvious under white mustard, especially after decay of the catch crop due to frost in January. Simulated N losses through denitrification were negligible (< 2 kg N/ha, results not shown) compared to the total N balance. In general denitrification was smaller under catch crops than under bare fallow and increased with fertilization. Non-frost resistant catch crops, especially those sown late, showed an increase in denitrification in spring.

Conclusions

Following a successful calibration of the EU-rotate_N model, N losses were simulated following winter barley on sandy loam (2011-2012). Compared to bare fallow, a strong reduction in nitrate leaching was observed under catch crops. No differences in nitrate leaching were shown between fertilized (60-120N) and non-fertilized catch crops. Simulated immobilization of the N released from the manure was considerably high. Losses due to denitrification were negligibly small.

Acknowledgements

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DIGESTION AND ACIDIFICATION OF CATTLE SLURRY TO IMPROVE N EFFICIENCY IN GRASS PRODUCTION

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Animal slurry is an important source of nitrogen (N) in crop production. However, the N fertilizer effect is limited due to problems with poor synchronisation of N availability and N losses through ammonia emissions. By digestion of the slurry in a biogas plant, the C/N ratio of the slurry is lowered and N becomes more readily available (Delin et al., 2012). Ammonia emissions are dependent on the pH of the slurry, and can therefore be reduced by adding sulphuric acid to the slurry in conjunction with spreading (Kai et al., 2008). The objective with this project was to investigate the effect on N recovery of acidification of cattle slurry and digested cattle slurry before application to grass ley.

Materials and Methods

Field experiments were conducted in grass ley (mixture of timothy, fescue and ryegrass) at two Swedish sites (Rådde and Bjertorp, 100 km east and north-east of Gothenburg, respectively). Thirteen treatments (Table 1) were replicated and randomised in four blocks. Slurry and digestate were applied with trailing hoses every 0.25 m across a width of 4 m. Mineral N fertilizer was applied as granulated ammonium nitrate. The first fertilization was delayed, so the grass was cut in late spring before the first fertilization. Fertilizer was then applied in late spring and after the next cut in summer. Just after application in treatments without acidification, sulphuric acid was poured into the tank to acidify the rest of the slurry or digestate, which was used in treatments with acidification. The acid was added step-wise during stirring to reach below pH 6, which meant 3 l m^{-3} for slurry and 6-9 l m^{-3} for digestate. Treatments without slurry or digestate were compensated with phosphorus and potassium fertilizer and treatments without sulphuric acid with sulphur fertilizer, in order to avoid deficiency of nutrients other than N. Dry matter yield and forage N content were measured for two cuts. Nitrogen fertilizer replacement value (NFRV) for total N in cattle slurry and digestate with and without acidification was calculated from their effect on N offtake in relation to that of mineral fertilizer.

Results and Discussion

Dry matter yield and N offtake were greater in acidified treatments than in the corresponding treatments without acidification at both sites and in all cuts (Table 1). At Bjertorp, the last cut yield was very low due to dry weather. In the other cuts, NFRV increased from 30 to 40-45% of total N in cattle slurry and from 30-50 to 70-80% of total N in digestate. This increase in NFRV after acidification corresponds to about 25-30% of the ammonium content in undigested slurry and 50-60% of that in digestate. According to Rodhe et al. (2006), ammonia emissions from cattle slurry applied to grassland represent about 40% of the ammonium content. Applying this figure to the slurry in our experiment, the increase in NFRV corresponded to 60-75% of the ammonia emissions from undigested slurry. That is very similar to the value reported by Kai et al. (2008), who observed a reduction in ammonia emission of 67%. In studies in Denmark, the effect on NFRV of acidification of slurry applied to grassland varied depending on clover density, but on average it increased by 9 %-units (Birkmose and Vestergaard, 2013). In our experiments the ley only contained grass, which may explain the somewhat higher increase obtained. The higher NFRV for digested slurry than undigested is most likely due to the lower C/N ratio in digestate (6-7) than in undigested slurry (8-10). However, the NFRV was not always higher for digestate than for undigested slurry (Table 1), which may be explained by higher potential for ammonia emissions due to higher pH before acidification (7.5-7.8 compared with 7.0-7.2) and higher ammonium content (60-65% compared with 55-60%). This suggestion is supported by the fact that NFRV for acidified digestate was always higher than for acidified undigested slurry.

Table 1. Nitrogen (N) offtake (kg N ha^{-1}) with harvested grass in different treatments with different amounts of N (kg N ha^{-1}) added as mineral fertilizer (MF), cattle slurry (CS) and digested slurry (DS) with and without sulphuric acid treatment (a) at first and second fertilization at the sites Bjertorp and Rådde.

First fertilization	Total N added	N offtake Bjertorp	N offtake Rådde	Second fertilization	Total N added	N offtake Bjertorp	N offtake Rådde
No N	0	14	14	No N	0	5	23
MF	60	49	56	MF	50	19	29
MF	100	71	95	MF	80	27	58
CS	90[1].93[2]	26	33	MF	50		
CS+a	90[1] 93[2]	35	46	MF	50		
MF	60			CS	83[1]/80[2]	7	24
MF	60			CS+a	83[1]/80[2]	10	32
DS	93[1],102[2]	40	48	MF	50		
DS+a	93[1],102[2]	59	67	MF	50		
MF	60			DS	83[1]/78[2]	12	22
MF	60			DS+a	83[1]/78[2]	22	41
CS+MF	90[1],93[2]+	49	60	CS+MF	83[1]/80[2]+3	18	41
CS+a+MF	90[1],93[2]+	63	77	CS+a+MF	83[1]/80[2]+3	22	49

Conclusions

Acidification can increase NFRV significantly for both digested and undigested cattle slurry. With both digestion and acidification, the NFRV of the slurry increased from about 30% to 70%.

Acknowledgements

Sparbanksstiftelsen Skaraborg, the Swedish Farmers' Association for Agricultural Research, the Swedish Rural Economy and Agricultural Society of Sjuhärad and the Bovine Foundation of Skaraborg. Staff at Lanna and Rådde experimental stations for taking care of the experiments. Special thanks to Anders Grandin and Tobias Karlsson for risking their lives while acidifying the slurry. Gruvaeus (Lantmännen SW seed) for providing cattle slurry, digestate and the grassland for the experiment at Bjertorp. Ola Hallin and Lennart Wikström (Swedish Rural Economy and Agricultural Society), Tavs Nyord (University of Aarhus), Lena Rodhe and Huibert Oostra (Swedish Institute of Agricultural and Environmental Engineering), Peter Hylmö (Hylmö industrikonsult) and Morten Toft (BioCover A/S) for good advice and engagement in the experiments.

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MITIGATION OF NITRATE LEACHING AND NITROUS OXIDE EMISSIONS FROM WINTER FORAGE GRAZING SYSTEMS

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In many parts of New Zealand, dairy cattle are taken off the milking platform to graze high yielding forage crops during winter. The high stocking rate of the winter forage grazing system means that a large proportion of the ground would be covered by animal urine patches, and the wet winter soil conditions would make winter grazing particularly prone to soil pugging (poaching) causing soil compaction. These conditions are highly conducive for nitrate (NO3-) leaching and nitrous oxide (N2O) emissions. There is an urgent need to develop effective management tools to reduce NO3- leaching to meet local regulatory standards on NO3- losses to ground- and surface-waters and to reduce greenhouse gas emissions from intensive farming systems. A nitrification inhibitor mitigation technology involving the use of dicyandiamide (DCD) to treat grazed pasture soil has been developed to mitigate both NO3- leaching and N2O emissions (Di and Cameron, 2002). However, the efficacy of this technology under winter forage grazing conditions where the soils are wet and potentially poached has not been well studied. This paper reports a series of laboratory, lysimeter and field studies to determine the efficacy of the DCD nitrification inhibitor in inhibiting ammonia oxidizers, reducing N2O emissions and reducing NO3- leaching under simulated winter forage grazing conditions.

Materials and Methods

In order to determine the impact of moisture status on the effectiveness of DCD in inhibiting ammonia oxidizers, a laboratory incubation study was conducted to determine the effect of three soil moisture conditions (60%, 100% and 130% field capacity) on the abundance of ammonia oxidizer communities, ammonia oxidizing bacteria (AOB) and ammonia oxidizing archaea (AOA), using a grassland soil (Horotiu soil: Typic Udivitrand). Two sets of incubations were set up, one set for soil sampling to determine the abundance of the microbial communities using real-time qPCR (Di et al., 2009) and the other set for determining N2O emissions using a static chamber method. A field experiment was conducted to determine the impact of animal treading on N2O emissions on a Wakanui sandy loam (Aquic Dystric Utrochrept). Field plots of 0.5 m diameter were established to simulate dairy cow urine patches. Dairy cow urine at the rate of 1000 kg N ha-1 was applied to the plots to simulate animal urine deposition. The nitrification inhibitor DCD was applied to some of the plots. Some plots were un-trampled, and some were trampled with a mechanical hoof delivering the same pressure as that of an adult cow hoof walking over the field. N2O emissions were determined using field static chamber methods. A lysimeter study was conducted to determine the effectiveness of DCD in reducing NO3- leaching under simulated winter grazing conditions. The lysimeters were 50 cm diameter and 70 cm deep, and the soil used was an Eyre stony silt loam (Dystrochrept) with annual ryegrass pasture. Urine was applied in mid-winter (June) at 1000 kg N ha-1 and DCD applied as a solution at 15 kg DCD ha-1. Nitrate concentration in the leachate was determined.

Results and Discussion

The laboratory incubation study showed that soil moisture content was a major driver affecting the growth of ammonia oxidizing bacteria (AOB) and N2O emissions in the soil that received animal urine. Total N2O emissions from the soil at 130% field capacity were 400 times higher than those from the soil at 60% field capacity. DCD was highly effective in inhibiting the growth of AOB in the wet soil conditions and total N2O emissions were significantly related to the abundance of AOB amoA gene copy numbers but not to that of AOA. The field plot study showed that animal treading of a wet soil resulted in a reduction in air permeability and air-filled pore space. Trampling increased average cumulative N2O emissions over a three month period from 15.9 kg N2O-N/ha to 45.0 kg N2O-N/ha in the urine treatments. DCD was highly effective in reducing N2O emissions, with N2O emissions being decreased by 58-63%. Trampling did not significantly affect the effectiveness of DCD in reducing N2O emissions. The lysimeter study showed that NO3- leaching losses from the animal urine applied in mid-winter to wet and heavily trampled soil were significantly decreased by 66% with a single application of DCD following simulated grazing in mid-winter.

Conclusions

These results suggest the soil moisture conditions and animal trampling found under winter forage grazing systems can have a major impact on N transformations and losses in the soil. The DCD nitrification inhibitor technology is an effective mitigation tool for NO3- leaching and N2O emissions in winter forage grazing systems. DCD can serve as a useful management tool to help local farmers to meet regulatory rules on NO3- discharges to waterways, while at the same time, reducing greenhouse gas N2O emissions.

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ESCAPADE TO QUANTIFY NITROGEN LOSSES IN TERRITORIES AND ASSESS MITIGATION STRATEGIES

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One of the agro-environmental and socio-economic challenges of agriculture is to maintain agricultural production while limiting the use of nitrogen inputs. The development of the Haber-Bosch process on an industrial scale in the XXth century to produce ammoniacal nitrogen from atmospheric dinitrogen allows the entry of large amounts of nitrogen in production systems. This feeds a cascade of processes within agroecosystems and losses of nitrogen to the environment at each stage of the cascade (Fig 1) with many environmental and societal impacts (degradation of air, water and soil quality, impacts on greenhouse gas balance, biodiversity and human health…). The overall costs of nitrogen losses in Europe are estimated between 70 and 320 million euros per year and thus exceed the direct economic benefits of the use of nitrogen in agriculture (Sutton et al., 2011). A major challenge is to better understand the processes involved in the nitrogen cascade and nitrogen losses to the environment, to integrate them by taking into account spatial and temporal interactions within landscape mosaics, to quantify them from modelling approaches combined with experiments and inventories, to assess agro-environmental scenarios of nitrogen management in territories, and to propose innovative mitigation strategies of nitrogen losses and/or adaptation strategies of production systems to global change. Since classical approaches at plot or farm scale do not make possible to control all impacts, levers must also be sought at larger scales (Cellier et al., 2011). In this context, the overall objective of the multidisciplinary project ESCAPADE (2013-2017) is to analyze the effect of agricultural activities and landscape mosaics on the nitrogen cascade in territories, with an approach that combines production of scenarios, modelling tools and observations of flows of different forms of reactive nitrogen (NO_3^-, NH_3, NO_x, N_2O…). The project mainly focuses on landscapes defined as areas from a few km^2 to a few tens of km^2 and also on larger areas from hundreds to thousands of km^2.

Materials and methods

The ESCAPADE project is organized into four main scientific tasks (Fig. 2). Under task 1, agro-environmental scenarios related to management of nitrogen and landscape mosaics are built at the classical scales of the plot (types, quantities and dates of nitrogen inputs...) and the farm (crop sequences, herd and waste management…) and also at the innovative scale of territories (structure of the landscape mosaics, spatial planning...). Integrated models dealing with the nitrogen cascade within landscapes (resp. larger territories) are developed and/or used under task 2 (resp. task 3) to quantify nitrogen flows and losses to the environment. Models developed under task 2 are calibrated and evaluated from data inventoried and measured on the experimental sites of the project (task 4). These sites are located in Brittany, Parisian Basin and southwest of France. They are characterized by a wide range and variability of nitrogen flows due to differences in agro-pedo-climatic conditions. Scenarios and models are then applied to the sites and territories to quantify nitrogen flows and losses within the sites and larger territories. The ESCAPADE project associates research actors (basic and applied) and actors of the agricultural development (technical institutes, cooperatives, local actors). Partners are from various disciplines (biophysics, biogeochemistry, agronomy, socio-economy, mathematics/statistics, computer science).

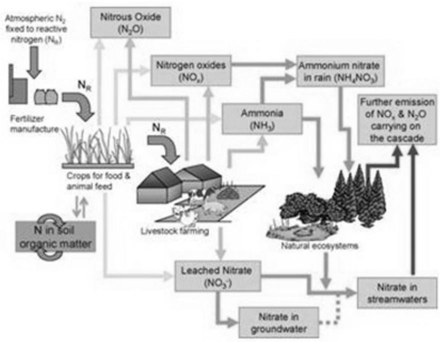

Figure 1. Simplified diagram of the nitrogen cascade (from Sutton et al., 2011).

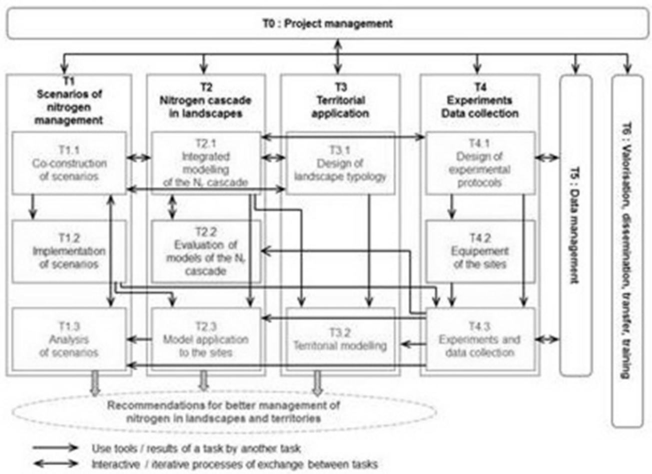

Figure 2. Structure of the ESCAPADE project.

Results and discussion

The expected results are: i) production, evaluation and interpretation of scenarios for nitrogen management in territories, ii) production of knowledge and reliable tools (models, databases) to quantify the nitrogen cascade in territories, iii) strengthening multidisciplinary partnerships between research and development, iv) co-construction of innovative solutions to reduce nitrogen losses in the environment or adapt production systems.

Conclusions, acknowledgements

The ESCAPADE project (2013-2017, http://www.n-escapade.fr/) is funded by the French National Research Agency (Contract No. ANR-12-AGRO-0003). It is supported by the GIS GC-HP2E (Scientific group on Crops – High Economic and Environmental Performance) and labelled by the VEGEPOLYS cluster.

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MODEL PREDICTIONS OF STREAMWATER NITRATE CONCENTRATIONS UNDER AGRICULTURE CHANGES: HOW GOOD ARE THEY?

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In the areas of intensive agriculture of Europe, achieving compliance with EU nitrate directive is often difficult. In 2007, French government was in litigation with European authorities concerning 9 catchments in Brittany (Western France) used for drinking water supply, whose nitrate concentrations were exceeding the 50 mg.L^{-1} NO_3 standard. a mitigation plan was applied to avoid penalties, consisting in mandatory decreases of nitrogen inputs by farmers (between 15 and 30%, depending on the farm type and initial situation). To evaluate ex ante the efficiency of the plan, a detailed agrohydrological model (TNT2) was applied to 5 of the catchments, and the future evolution of the streamwater nitrate concentrations was simulated until 2015. The effective implementation of the plan was controlled by the administration. Therefore, after 6 years, we are able to compare the predictions made and the actual evolution of the streamwater nitrate after a significant and documented evolution of the agricultural practices, which is a ideal situation to assess the prediction ability of the model and discuss the efficiency of the measures that have been taken.

Materials and methods

TNT2 (topography-based nitrogen transfer and transformations) is a spatially distributed agro-hydrological model based on the coupling of a hydrological model based on TOPMODEL assumptions and a crop model adapted from STICS. It has been developed to account for the soil-shallow groundwater interactions linked to topography and soil properties, which determine the fate of nitrogen according to the location of application and the existence of saturated areas in the valley bottom. The model is driven by daily climate data and agricultural management practices described at the field level. For catchments where exhaustive agriculture information is not available at the field levels, procedures based on farm classification and spatial modelling based on Markov chains provide realistic spatial and temporal distributions of management practices. The model was applied to five catchments located in Brittany, which is the most intensive livestock production area in France. Submitted to a humid temperate climate and underlain with impervious bedrock, the region is severely impacted by nitrate diffuse pollution, causing coastal eutrophication and exceedance of drinking water standards. In the five catchments are located drinking water supply facilities whose untreated water has not been complying the EU nitrate directive since the late 1970s. The five catchments differ in terms of agriculture production (some dominated by indoor pig and poultry productions, others by dairy production), climate (annual rainfall from 1200 to 700 mm), hydrological functioning and level of contamination (mean N-NO_3 concentration from 18 to 8 mg.L^{-1}). Data to apply and calibrate the model was supplied by local and national authorities, based on regulatory enquiries and surveys. The model was first applied on the 1998-2007 period, the whole period being used for calibration. Then two scenarios were applied, BAU (business as usual, keeping the agriculture at its current level) and MP (mitigation plan), for the 2008-2015 period. The MP scenario was constructed based on the regulations introduced in 2007 by the government, i.e. the limitation at 140 kg/ha of total nitrogen input calculated on the whole farms' surface, except for dairy farms (160 kg/ha) and vegetable farms (170 kg/ha).

Results and Discussion

The goodness of fit obtained in the calibration/validation procedure was good for the 5 catchments concerning daily discharge and daily nitrate fluxes (Nash efficiency from 0.7 to 0.8) and acceptable for nitrate concentrations (mean relative error from 11 to 22% of the mean observed concentrations). The modeled estimates of yields and nitrogen export from the main crops (wheat, silage and grain maize, pastures) were also close to the average values reported for each area. The two scenarios resulted in a decrease of the concentrations (from 7% to 19% and from 16% to 22% for BAU and MP, respectively). Surprisingly, the goodness of fit for the prediction period (from 2008 to 2013) was as good, or even better than for the calibration period (from 9 to 19%). In most cases, the observed concentrations were laying in between the BAU and MP scenarios. For one catchment, the observed concentrations decreased slightly more than the MP scenario, and for another, slightly less than the BAU scenario.

HIGH FERTIGATION FREQUENCY DECREASES POTENTIAL NITROGEN LEACHING IN PROCESSING TOMATO GROWN WITH HIGH NITROGEN AND WATER SUPPLY

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Fertigation is believed to minimise the risk of NO_3-N leaching through careful application of water and nitrogen (N) fertiliser according to crop requirements at any growth phase and fertiliser localization close to roots (Agostini et al., 2010). However, the impact of the fertigation technique on water and N dynamics in the soil may also depend on fertigation and irrigation frequency (Zotarelli et al., 2009). The aim of this experiment was to assess the effect of fertigation-irrigation frequency on crop N uptake and potential N leaching.

Material and methods

A field experiment was carried out in 2006 and 2007 in a clay loam soil in Central Italy on processing tomato (*Lycopersicon esculentum* Mill., cv. PS1296), grown with 3 N-fertiliser rates (0, 100 and 300 kg N ha^{-1}: N0, N100 and N300) applied with 3 different fertigation-irrigation weekly scheduling (F1: 1 fertigation; F1+I2: 1 fertigation + 2 irrigations; F3: 3 fertigations) in order to realize a total of 8 treatments (in a randomized complete blocks design with 4 replicates): F1N0, F3N0, F1N100, F1+I2N100, F3N100, F1N300, F1+I2N300, F3N300. All treatments received the same weekly water volume based on potential ETc estimated by the Penman-Monteith equation and Kc values reported by Tei et al. (2005). Above ground dry matter (DM) and total N accumulation and leaf area index (LAI) were determined by sampling plants fortnightly. Actual crop ETc for each treatment was then calculated *a posteriori* based on the dual crop coefficients method (Allen et al., 1998) by taking into account the different LAI and fraction of soil surface wetted by different irrigation frequency. Since no treatment revealed water stress symptoms at any growth phase, we assumed that the ETc of the treatments with highest LAI (F3N300 and F1+I2N300) was met by weekly water volumes and that the surplus in the other treatments generated drainage. The NO_3-N concentration in soil solution extracted weekly from suction lysimeters at 0.6 depth was measured by a portable ione-specific electrode (Cardy Spectrum Tech., IL USA). The amount of NO_3-N leached below 0.6 m was calculated as the product of weekly drainage volumes by NO_3-N concentrations in the soil solution for that week. The mineral N content (N_{min}) of the top 0.6 m soil layer was determined at final harvest.

Results and discussion

In both years LAI and DM and N accumulation increased with N rate and, only within N300, with fertigation frequency (Tab 1). As compared to F1N300, F3N300 increased DM by 21% and N uptake by 25%, (2-year average), while the increase of LAI was much higher in 2006 than in 2007. A similar increase for DM and N uptake was recorded in F1+I2N300 in 2007 and for LAI in both years. The N uptake of F3N300 in both years and of F1+I2N300 in 2007 was even higher than the N rate (Table 1), with an apparent recovery (Greenwood et al., 1989) equal to 0.70 in 2006 and over 0.80 in 2007, much higher than in the other N300 treatments (from 0.51 to 0.56 depending on year and treatment). Despite such a different N uptake efficiency, no remarkable differences within N300 were observed either for NO_3-N concentration in the soil solution at 0.6 m depth (Fig. 1) or for residual mineral N content in the soil (Table 1). This can be partly explained by assuming a different amount of N leached below the root zone due to different drainage volumes. Based on *a posteriori* actual ETc, the cumulated drainage of treatments other than F3N300 and F1+I2N300 ranged from 55 mm to 158 mm and was 77 mm for F1N300 (2-year average). Therefore, leached N in F1N300 was 36 kg N ha^{-1} (2-year average) which accounts for 50% of the difference in N uptake between F3N300 and F1N300 (Table 1). A further portion of this difference might be accounted for by the likely higher N evolution from the soil in F1N300, where the all at once supply of irrigation volume caused a more prolonged soil saturation.

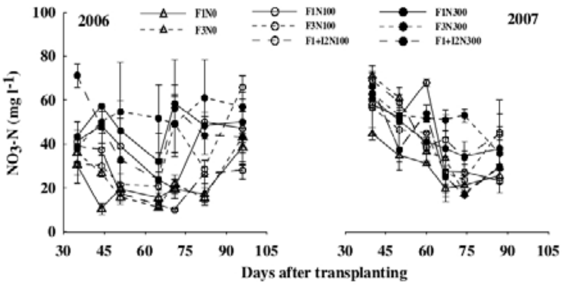

Figure 1. Time course of NO_3-N concentration in soil solution at 0.6 m depth during the crop cycle of processing tomato grown in 2006 and 2007 with different N rates and fertigation-irrigation frequencies. Bars indicate ± SEs.

Table 1. Above ground dry matter and total-N accumulation, maximum leaf area index, soil mineral N content at final harvest, cumulated drainage and leached N in processing tomato grown in 2006 and 2007 with different N rates and fertigation-irrigation Frequencies.

	DM ($Mg\ ha^{-1}$)		N uptake ($kg\ ha^{-1}$)		LAI_{max}		soil N_{min} ($kg\ ha^{-1}$)		Drainage (mm)		N leached ($kg\ ha^{-1}$)	
	2006	2007	2006	2007	2006	2007	2006	2007	2006	2007	2006	2007
F1N0	7,3	8,4	133	121	1,2	1,7	55	36	158	123	37	38
F3N0	7,9	7,9	137	128	1,6	2,1	45	40	105	126	21	53
F1N100	9,8	9,7	221	170	2,4	2,6	47	37	118	88	42	45
F1+I2N100	10,5	11,2	213	182	2,8	2,8	59	33	55	86	12	44
F3N100	9,7	11,1	179	182	2,9	2,9	55	37	64	98	19	47
F1N300	10,7	11,2	285	288	2,5	3,5	63	38	84	70	37	35
F1+I2N300	11,2	13,9	304	382	4,1	4,1	56	45	-	-	-	-
F3N300	12,9	13,6	349	369	4,5	4,1	65	34	-	-	-	-
sed	*0,90*		*28,5*		*0,38*		*2.1*		*12.1*		*3.8*	

Conclusions

Results indicate that high fertigation frequency may represent a tool to increase N uptake efficiency and limit the N loss from the soil in processing tomato fed with very high N and water supply, which is often the case in intensive processing tomato production. It is worth to underline, however, that an accurate estimate of critical crop N and water requirements is mandatory to prevent overfeeding and limit environmental pollution.

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PERMEABLE REACTIVE INTERCEPTORS – BLOCKING DIFFUSE NUTRIENT AND GREENHOUSE GASES LOSSES IN KEY AREAS OF THE FARMING LANDSCAPE

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Denitrifying bioreactors (DB) only target single contaminants along a nutrient transfer continuum, although mixed contaminant discharges to a water body are more common. The objective of the current paper is to present a framework to identify and prioritise contaminant leaks in DB systems.

Materials and Methods

The term 'permeable reactive interceptor' (PRI) is proposed when trying to achieve agricultural sustainability. A PRI is a modified DB with additional remediation cells for specific contaminants in the form of solutes, particles or gases. Researchers should work towards a '*mixed-contaminant blockade balance*' where both mixed-contaminant remediation and pollution swapping (PS) occur but at acceptable rates defined by specific environmental legislation to the area the bioreactor is installed in. The following procedure may be undertaken for any dissolved contaminants (e.g. NH_4^+, NO_3^-, dissolved reactive P, heavy metals), greenhouse gases (CO_2, N_2O and CH_4) and NH_3 gas. Consider, for example, N_2O: (Eq 1) where $F_{N2O(IN)}$ and $F_{N2O(OUT)}$ are the dissolved flux (F) of N_2O at the inlet (IN) and outlet (OUT) of a PRI, atmosphere (ATM), and B_{N2O} is the balance of N_2O between these three fluxes. If $B_{N2O}>0$, remediation within the PRI has occurred; if $B_{N2O}=0$, the compound is conserved during transport through the PRI (or remediation and production of the compound are equal), and if $B_{N2O}< 0$, then PS has occurred. Balances calculated using Eq (1) can be used to derive a sustainability index (x) and a weighting system can be applied, whereby the weighting of trade-offs between different loss pathways can be judged (Eq 2). An example here is where aeration is a problem in a system, resulting in a trade-off between CH_4 and N_2O: (Eq 2) where *a-d* and so on are weighting factors (WF) and *B* terms here are gathered from Eq (1). Other contaminants in gaseous (e.g. NH_3 and H_2S), dissolved (e.g. ammonium, metals) and particulate (e.g. P) forms may also be added. Working through Eq1-2 for all contaminants creates a balance, identifies contaminants of concern and remediation sequences. Case studies using the lab study of Healy et al., (2012) (see Table 1) were used where NO_3^- reduction and PS were tested in four C substrates (woodchips (L), cardboard (CB), lodgepole needles (LP) and barley straw (BS)) mixed with soil. NO_3-N spiked water (19.5-32.5 mg/l) was injected into columns at a hydraulic loading of 30 mm/d (460 days). Prominent environmental legislation or issues in the geographic areas where a PRI is installed were identified. In Ireland, PO_4^{3-}-P and NH_4^+ limits in rivers are set at 35 and 65 μg/l, respectively, while NO_3^- in groundwater should not >8.5 mg/l. Hence, WF for PO_4-P should be 1.86 times higher than for NH_4-N (65/35 = 1.86). The WF for PO_4-P should be 242.03 times higher than for NO_3-N (8471/35 = 242.03). Global warming potential

(GWP) is a measure of radiative forcing attributable to an individual GHG relative to that of CO_2, which has a GWP of 1. This variable is 25 and 296 on a 100-year basis for CH_4 and N_2O, respectively, could be used in addition to others to determine the WFs.

Results and Discussion

Case 1 (USA, legislative instruments (LI) focused on water quality): NO_3^- removal is the most important environmental concern, while GHGs emissions to the atmosphere and other contaminant losses to water are perceived as secondary. NO_3-N balances are attributed significantly highest WF in Eq (2) than other contaminants. Here WF for NO_3-N is set to 1, and all other WFs to < 1. Results of Eq (2) are then expressed in $g/m^2/d$ as NO_3-N. Here, the ranking of the different columns follows: 1-L; 2-LP; 3-BS; 4-CB. *Case 2* (Ireland, LI focused on water quality, GHG and transboundary pollutants. Contaminant losses to water are perceived as more important. When only accounting for NO_3^--N, NH_4^+-N and PO_4^{3-}-P, the WFs are set to 1, 0.538 (35/65) and 0.004 (35/8471), respectively. Results for Eq (2) are then expressed in g PO_4^{3-}-P-eq/m^2/d. The ranking of the different bioreactors is: 1-BS; 2-LP; 3-CB; 4-L. When accounting only for GHG and NH_3 emissions, one must refer to national legislation targets. In Ireland, the national target of GHG reduction is set to 20% in 2020, with no legislative limit set on individual farmers. In addition, while transboundary gases are limited to 100 Ktonnes NH_3, national emissions are 10% under this ceiling. Hence, potential WFs for GHG and NH_3 are 0.2 and 0.1, respectively. *Case 3*: (NZ, LI focused on GHG without NH_3) gaseous emissions to the atmosphere are perceived as a more important issue than contaminant losses to water. NZ is committed to reducing emissions by 5% by 2020 (10-20% in the case of a global agreement). In this case, CH_4, CO_2 and N_2O balances are attributed significantly higher WF than for N deposition or dissolved and particulate contaminants. For Case 3, where the WF for CO_2, CH_4 and N_2O are set at 1, 25 and 296, respectively, while all dissolved contaminants are given a WF of < 1. The results of Eq (2) are then expressed in g CO_2-eq/m^2/d. In this case, the ranking of the different bioreactors is: 1-L; 2-LP; 3-BS; 4-CB. One key point of the NZ approach is that agricultural emissions are quantified by ‘emissions intensity’ or emissions per unit product basis. Therefore in this case, the emissions generated from the PRI should be expressed in terms of the production activity (yearly milk or beef output for example) from the farm.

Conclusion

Mixed contamination mitigation needs co-location, new concepts and worked case studies to show pollution swapping and improve design criterion.

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AMMONIUM AND NITRATE IN SURFACE AND GROUND WATERS IN AN AGRICULTURAL AREA IN NW SPAIN

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Agriculture is one of the major sources of pollution of surface and ground waters, particularly ammonium and nitrate pollution. The district of A Limia, southeast of Galicia (NW Spain), is an area with intensive agricultural production, potatoes and wheat being the main crops. Potatoes, which are the main source of income for the region, are grown with heavy inputs of organic and mineral fertilisers. The agricultural area is a former lagoon dried in the twentieth century and the water table is near the soil surface. This fact, along with the coarse texture of the surface soil horizons, promotes the export of nutrients and other chemicals from agricultural soils to surface and ground waters. This paper examines the concentrations of ammonium and nitrate in surface and ground waters of A Limia over a year of sampling.

Materials and Methods

37 sampling points for surface water (drainage channels of the former lagoon, streams and River Limia) and 28 sampling points for groundwater (wells) were selected in the district of A Limia. Water was sampled over one year, from September to the end of July, making a total of six sampling dates.
Ammonium and nitrate were determined by steam distillation under alkaline conditions (with and without addition of Devarda alloy, respectively) and NH_3 titration with H_2SO_4 (Keeney and Nelson, 1982). Comparison of means and regression analysis were performed using SPSS 15.0 statistical software for Windows.

Results and Discussion

Ammonium concentrations ranged between 0.00 and 1.8 mg N/L in ground waters and between 0.01 and 8.31 mg N/L in surface waters. Concentrations were generally higher in ground than in surface waters (Figure), although the differences were not significant at $p< 0.05$ level. The high ammonium concentrations in ground waters may reflect a low capacity of soils to retain ammonium, related to their low cation exchange capacity, as well as conditions adverse for nitrification in groundwater. In both ground and surface waters, average ammonium concentration reached maximum values in October and July samplings. In October high concentrations are related to leaching of excess ammonium from soils by the first autumn rains. In July high concentrations are related to high evaporation. The value of 1 mg NH_4/L, established by the Spanish law as toxic to fishes (Royal Decree 927/1988) is reached or exceeded by 7% surface water samples in September, 18% in October, 8% in March, none in April and May, and 6% in July. The value of 0.5 mg NH_4/L established by the European Union as a maximum allowable concentration in drinking water (Directive 98/83/CE) was often exceeded by surface water and even more often by ground water (up to 70% of the surface waters and up to 90% of ground waters in the most unfavourable months).

Nitrate concentrations ranged between 0.17 and 20.6 mg N/L in ground waters and between 0.17 and 9.71 mg N/L in surface waters. Both the type of water (surface or groundwater) and the sampling date significantly ($p< 0.05$) influenced the nitrate concentration (Figure). The same as ammonium, concentrations were higher in groundwater. The differences between ground and surface waters reflect the high mobility of nitrate, which is not retained by the soil and easily reaches the water table. In surface waters, the seasonal variation is similar to that of ammonium, reaching maximum values in October and July, but differences were not significant. In groundwater, nitrate levels were significantly higher ($p< 0.05$) in April and July than in September. The high concentrations are related to nitrate leaching from agricultural fields. The strong demand of nutrients by the crop may explain the relative decrease of the nitrate concentration in May. In July, mineralisation of organic matter and nitrification, favoured by high temperatures and water supplied by irrigation, contribute to increased nitrate available and susceptible to be leached. The value of 50 mg NO_3/L, maximum allowable concentration in drinking water (Directive 98/83/CE), is exceeded by a maximum of 26% of groundwater samples (in July). The value of 25 mg NO_3/L is exceeded by a maximum of 67% ground waters (in July) and 8% surface waters (in March). The highest concentrations of ammonium and nitrate in surface waters occurred in the channels that drain the fields of the former lagoon of Antela, or in small streams. Concentrations were generally relatively low in River Limia, the largest river in the area, except for some points downstream of the village of Xinzo de Limia.

Conclusions

Concentrations of ammonium and nitrate in ground and surface waters in the district of A Limia (Galicia, Spain) were high in some sampling points and sampling dates. Concentrations were higher in ground waters, differences being significant ($p< 0.05$) for nitrate. Concentration peaks related either with nutrient leaching from agricultural fields by fall or spring rains, or with drought.

The high water table in the winter causes that ground water is loaded of soluble substances applied to the soil, so that macronutrient concentrations in shallow ground water are even higher than in surface waters. The soil does not seem to effectively play its usual role of filter, possibly due to the coarse texture of the surface horizons.

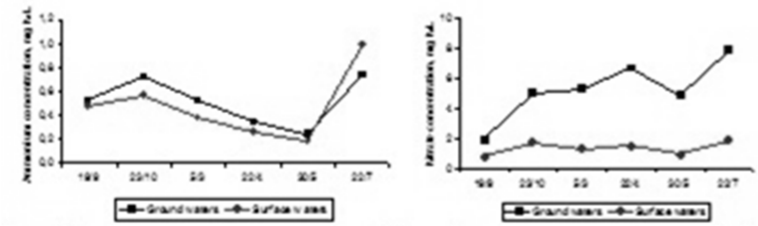

Figure. Seasonal variation of the average concentrations of ammonium and nitrate in ground and surface waters

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AMMONIA EMISSION FROM UREA SPREADING AND RELATIONSHIPS WITH METEOROLOGICAL VARIABLES

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Agricultural activities are responsible of over 95% of ammonia (NH_3) emissions in Italy (ISPRA, 2011) due to field application of organic and inorganic nitrogen (N) fertilisers. About 40% of N is applied as urea, even if a trend in reducing its application is recorded (ISTAT, 2011). Several factors affect N losses by NH_3 volatilization, among which weather conditions are important (e.g. Sommer et al., 2004). Thus, in order to mitigate NH_3 volatilization, the relationships between this phenomenon and its drivers has to be taken into account for addressing the agronomic management of cropping system.

Inverse dispersion models can be used in order to estimating NH_3 emissions (Carozzi et al., 2013) at field scale. In particular, WindTrax model (Flesch et al., 1995) has been applied to investigate NH_3 volatilization from urea spreading in two cropped fields subject to different climatic conditions in Italy: (i) sorghum in southern (RUT) and (ii) corn in northern Italy (LAND). The daily dynamics of NH_3 fluxes of these two summer crops and the relationships between the NH_3 volatilization and the meteorological conditions have been compared.

Materials and Methods

Table 1 summarises the details of the two trials performed during summer periods, where granular urea was surface broadcast.

The required inputs to the WindTrax model are the NH_3 concentrations measured from the emitting source and far from any other source (background) and the turbulent state of atmosphere. This latter can be characterized in terms of friction velocity and Monin-Obukhov length computable by data collected using three-dimensional ultrasonic anemometer: a Gill R2 (Gill Instruments Ltd, UK) and a METEK-USA-1 (Germany) were used during RUT and LAND trials, respectively. During RUT trial, NH_3 concentration was measured in the centre of the field with a fast NH_3 sensor, a QC-TILDAS developed by Aerodyne Research Inc. (ARI, USA) and averaged on 30 minutes. The background concentration was monitored by means of time integrated passive samplers ALPHA developed by Tang et al. (2001), the same employed for monitoring concentrations in LAND trial both in the fertilized field and as background. The ALPHAs were exposed in triple replicates for no more than 12 hours. Hourly NH_3 fluxes were estimated during the two trials. Moreover, standard meteorological stations were used in the two trials for measuring hourly temperature and humidity of the air, global solar radiation, rain and wind speed.

Results and Discussion

The meteorological conditions during the trials are summarised in Table 2. The most significant differences are in global radiation and wind speed, both higher during RUT than LAND trial, and in water availability as rain or irrigation, very small for RUT trial despite irrigation. Average daily flux patterns were calculated for the two experimental periods

(Figure 1). In both trials the trends are similar, with a period of rapid increase after sunrise: the peaks are in the hottest hours of the day, then there is a decrease in emission. However, the difference of magnitude between the fluxes estimated during LAND and RUT trials reaches one order of magnitude. The average hourly fluxes have been normalized with respect to the maximum value of each trial in order to compare the relationships between meteorological variables and NH_3 fluxes. The diurnal pattern of NH_3 fluxes was clearly affected by atmospheric turbulence, as shown by its relationship with friction velocity, with highest NH_3 fluxes measured during the most turbulent conditions: this response being larger for LAND than for RUT (Figure2a). The same consideration can be made on the effect of global radiation on NH_3 emissions (Figure2b), even if the difference between the two trials is not pronounced. About the water availability as rain or irrigation, the 27 mm of rain occurred in the 48 hours after the urea spreading in LAND could be responsible for more in depth penetration of the fertilizer and a consequent lower NH_3 fluxes with respect to RUT trial where NH_3 emissions started only after irrigation.

Table 1 Details relative to the two trials.

Acronym	*Site*	*Period*	*Irrigation and Crop*	*N applied*	*Field extension*	*Height crop (cm)*
RUT	Rutigliano (40°59′N, 17°54′E, 122 m a.s.l.)	17 – 29 July 2008	Irrigated sorghum	240 kg N/ha	2 ha	72÷125
LAND	Landriano (45°19′N, 9°16′E, 88 m a.s.l.)	14 June – 07 July 2010	Irrigated corn	106 kg N/ha	10 ha	60÷280

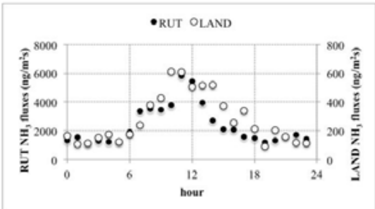

Figure 1 Averaged daily trend of NH_3 fluxes during the two trials.

Table 2 Statistics of the meteorological variables monitored. In brackets the standard deviation. Rain and irrigation are summed, while global radiation is the daily mean on the period of the trial.

Acronym	RUT	LAND
Air temperature (°C)	24.1 (±4.3)	23.3 (±5.4)
Relative humidity (%)	61.6 (±15.6)	71.0 (±18.4)
Rain+irrigation (mm)	23.6	70.3
Global radiation (W/m²)	13,997 (±1800)	6,279 (±2010)
Wind speed (m/s)	1.5 (±1.0)	0.9 (±0.8)

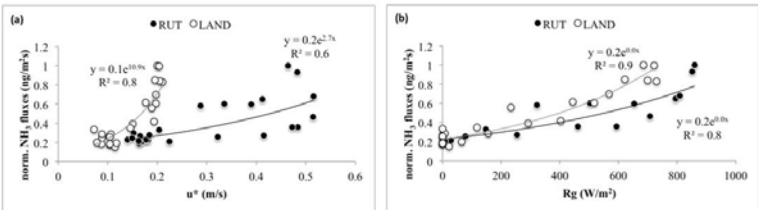

Figure 2 Averaged hourly NH_3 fluxes (normalized with respect to the maximum) in function of friction velocity (u^*) and global radiation (Rg).

Conclusion

Clear relationships between meteorological variables and NH_3 volatilization after urea spreading can be identify by estimating NH_3 fluxes using an inverse dispersion model, however the complete explanation on the difference in magnitude of the fluxes between the two trials has to be done, taking into account all the drivers of the phenomenon, especially the soil water content and the amount of fertilizer.

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MANAGING THE SOIL NITRATE POOL IN INTENSIVE GRASSLAND AND ARABLE SYSTEMS

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Managing nitrate (NO_3^-) in agricultural soil systems is a concern for agronomists, environmental scientists, and policy makers alike because it is vulnerable to leaching and denitrification loss pathways. The leaching loss problem has been in focus for many years because NO_3^- from agricultural is linked to elevated NO_3^- levels in surface and groundwater. Concerns regarding the climate change effects of greenhouse gas release, including nitrous oxide (N_2O), have increased in recent years, as evidenced by a dramatic increase in studies examining this issue. Soil systems with an abundance of NO_3^- are primed for leaching and gaseous N loss including N_2O loss by denitrification at microaerobic sites. Applying fertiliser N or managing the soil system to supply NO_3^- in synchrony with plant uptake is a basic strategy for managing both leaching and denitrification risk in intensive agricultural systems. Using fertiliser N formulations which meter out NO_3^- to plants is a practical solution for improving the synchrony between plant N uptake and the soil NO_3^- pool.

The objectives of the current study are to: (i) evaluate N source, calcium ammonium nitrate (C.A.N) and urea, as tools for managing the soil inorganic N pools.

(ii) evaluate the effect of using urea with and without the inhibitors N-(n-butyl) thiophosphoric triamide (Agrotain^{TM}) and dicyandiamide (DCD) either individually or in combination on the soil inorganic N pools.

Materials and Methods

Grassland

This experiment is being conducted at a loam and a sandy loam site. The experimental design is a randomized block with 3 replications. The annual N rate is 200 kg ha $^{-1}$ applied in five equal splits during the growing season. The experimental treatments are: (a) C.A.N., (b) Urea, (c) Urea + Agrotain, (d) Urea + DCD, (e) Urea + Agrotain + DCD, and (f) a control.

Arable

This experiment is being conducted on a poorly-draining clay loam and a free-draining loam site cropped with spring barley. The experimental design is a randomized block with 3 replications and the annual N rate is 150 kg ha^{-1} with 30 kg ha^{-1} applied at planting and 130 kg ha^{-1} applied during tillering (second split) as per standard agronomic practice. The experimental treatments are: (a) C.A.N., (b) Urea, (c) Urea + Agrotain, (d) Urea + DCD, (e) Urea + Agrotain + DCD, and (f) a control.

Soil sampling and processing

At each site soil samples from the surface 10 cm were collected once per week. The 10 cm zone was selected because denitrification potential is greatest in surface soil (Staley *et al.* 1990). Samples were wet sieved and soil inorganic N was extracted using 2 *M* KCl solution. Nitrate and ammonium in soil extracts was determined by colorimetric analysis using an Aquakem 600 discrete analyser.

Data analysis

The PROC REPEATED procedure of SAS (SAS Institute, Cary, NC, USA) was used to examine the relationship between treatments over time for soil inorganic N.

Results and Discussion

Data is presented from a grassland site (Fig. 1). A significant N source x day interaction was detected for soil NO_3-N (Table 1) which is consistent with effects of fertiliser N application and uptake by grass. Application of C.A.N. caused a rapid spike in NO_3-N levels in the soil system. This NO_3-N is available for plant uptake or loss through leaching and denitrification pathways. Urea-N must first undergo the microbially mediated processes of hydrolysis and nitrification before contributing to the soil NO_3-N pool.

Table 1. Analysis of variance table for soil NO_3-N ha^{-1} at a loam grassland site

Effect	Num DF	Den DF	F Value	Pr > F
Treatment	5	252	33.84	<.0001
Day	21	252	4.21	<.0001
N Source x Day	105	252	1.84	<.0001

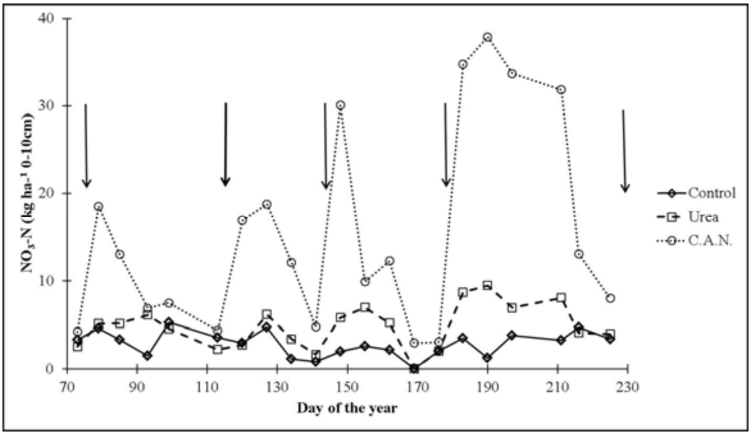

Figure 1. Soil NO_3-N pool in the surface 10 cm throughout the growing season affected by an annual application of 200 kg N ha^{-1} (applied in five 40 kg N ha^{-1}) splits (indicated by the arrows) at a loam grassland site.

Conclusions

The temporal profile of the soil NO_3-N pool during the sampling period was more stable for urea-N relative to C.A.N. This indicates the potential of fertiliser N source for managing the soil NO_3-N pool. Further details of the effects of N source and inhibitors will be presented.

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INFLUENCE OF TILLAGE AND LIMING ON N2O EMISSION FROM A RAINFED CROP

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Nitrous oxide (N_2O) is the main greenhouse gas (GHG) produced by agricultural soils due to microbial processes. The application of N fertilizers is associated with an increase of N_2O losses. However, it is possible to mitigate these emissions by the introduction of adequate management practices (Snyder et al., 2009).

Soil conservation practices (i.e.no tillage, NT) have recently become widespread because they promote several positive effects (increases in soil organic carbonand soil fertility, reduction of soil erosion, etc). In terms of GHG emissions, there is no consensus in the literature on the effects of tillage on N_2O. Several studies found that NT can produce greater (Baggs et al., 2003), lower (Malhi et al., 2006) or similar (Grandey et al., 2006) N_2O emissions compared to traditional tillage (TT). This large uncertainty is associated with the duration of tillage practices and climatic variability.

Liming is widely use to solve problems of soil acidity (Al toxicity, yield penalties, etc). Several studies show a decrease in N_2O emissions with liming (Barton et al., 2013) whereas no significant effects or increases were observed in others (Galbally et al., 2010). The aim of this work was to evaluate the effects of tillage (NT vs TT) and liming (application or not of Ca-amendment) on N_2O emissions from an acid soil during a rainfed crop.

Material and Methods

An eight months study was conducted in the Cañamero's Raña (SW Spain) measuring N_2O emissions. A total of 16 plots were established in the field following a split-plot design with four replicates. The main factor was tillage (NT vs TT) and the second factor was liming (i.e. application or not of a Ca-amendment), consisting of a mixture of sugar foam (rich in $CaCO_3$) and red gypsum. Therefore, the four treatments studied were: NT + amended, NT + not amended, TT + amended and TT + not amended. At the beginning of October (i) the Ca-amendment was appliedand incorporated into the 0-7 cm soil layer at a rate of 469 kg sugar foam ha^{-1} and 703 kg red gypsum ha^{-1}; (ii) 36 kg N ha^{-1}, 92 kg P_2O_5 ha^{-1} and 92 kg K_2O ha^{-1} were applied using $(NH_4)_2HPO_4$ and KCl; (iii) the NT plots were not ploughed and the TT plots were disturbed down to 20 cm using a cultivator (2 passes); and (iv) all plots were sown with hybrid rye at a rate of 140 kg ha^{-1}. At the end of January, a second top-dressing N fertilization was carried out applying 70 kg N ha^{-1}as $NH_4NO_3$to all plots. Nitrous oxide emissions were sampled periodically by using the closed static chamber technique and analyzed by gas chromatography using a HP-6890 gas chromatograph equipped with a Plot-Q capillary column and a ^{63}Ni micro electron-capture detector (Abalos et al., 2013).

Results and Discussion

Total cumulative N_2O emissions at the end of the experiment (May) are shown in Figure 1. Nitrous oxide cumulative fluxes increased due to N fertilizer application (control vs N-fertilized plots). This effect is well-documented. Regarding N-fertilized plots, N_2O losses from TT were significantly higher than NT *(p=0.003);* the influence of liming was significant only at *p=0.074* and, tillage x liming interaction was also significant at *p=0.034.*With respect to traditional tillage, the application of sugar foam+red gypsum as Ca-amendment decreased significantly *(p=0.010)* cumulative N_2O emissions. Liming had no significant effect on cumulative N_2O fluxes from NT plots. Denitrification rather than nitrification can be considered the main biotic process responsible for N_2O emission because soil WFPS exceeded 60%. The mechanisms by which liming influence N_2O fluxes are not fully understood and accordingly contrasting results have been found (Barton et al., 2013). Under the environmental conditions of our experiment, liming probably decreased N_2O emissions by promoting greater N_2O reductase activity, thus leading to a complete reduction to N_2. The higher crop productivity of NT compared to TT (data not shown) probably reduced N_2O emissions due to increased N uptake by vegetation and a consequent lower availability of soil mineral N.

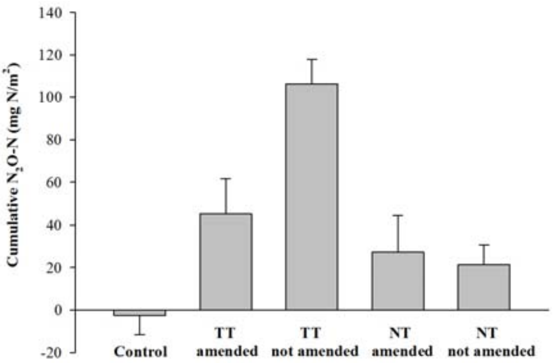

Figure 1. Total cumulative N_2O-N emissions.Error bars indicate standard errors.

Conclusion

Our results show that (1) NT may decrease N_2O losses compared to TT; and (2) liming may reduce these emissions when TT is used. Thus, this study underlines the key role ofno tillage and liming as effective N_2O mitigation strategies for rainfed crops of semiarid areas.

Acknowledgements

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RECONNECTING CROP FARMING AND CATTLE BREEDING FOR A REDUCTION OF NITROGEN LOSSES IN AN INTENSIVE AGRICULTURAL WATERSHED.

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Industrial agriculture and the use of nitrogen fertilisers have led to the massive introduction of reactive nitrogen into the biosphere, which subsequently cascades through a number of environmental compartments (Galloway et al 2003; Sutton et al 2011). Nitrate contamination of ground- and surface water as well as atmospheric pollution are among the major threats. To overcome this problem a drastic change must be initiated now in agriculture. Several experimental studies have been carried out to quantify the transfers and transformations of nitrogen of the Orgeval watershed allowing to implement a modeling approach able to simulate the current situation. Our objectives here are to explore scenarios to reduce nitrogen losses including one dealing with reconnection of crop production and cattle breeding, as was the rule in this area previously. Centered on a small watershed level, our analysis aims to extrapolate the result at the scale of the entire Seine basin (80000 km²) or at a pluriregional scales.

Materials and Methods

The Orgeval watershed is a small sub-catchment covering 104 km^2, located 70 km East of Paris (France) in the Seine basin (Fig. 1). It is highly homogenous in terms of climate (semi-oceanic) and topography (mean altitude, 148 m). Due to waterlogged soils in the winter, up to 90% of the arable soils have been artificially tile-drained. Land use is mostly agricultural land, dominated by cereal crops (wheat, maize and pea) with conventional practices, mainly based on mineral nitrogen fertilization (Fig. 1). On the basis of experimental data of i) NO_3, NH_4 and N_2O concentrations in surface and groundwater, and in the sub-root zone of agricultural soils, ii) N_2O emission from

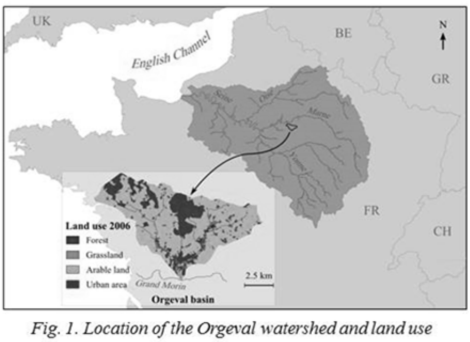

Fig. 1. Location of the Orgeval watershed and land use

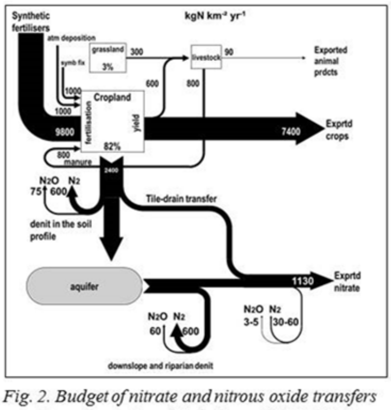

Fig. 2. Budget of nitrate and nitrous oxide transfers and transformations (Hydrology of 2006-2012).

cropped soils (Garnier et al in press), and of iii) discharge measurements at a number of hydrological stations, we have been able to quantify the budget of N transfer between the various compartments of the Orgeval watershed (Fig. 2). A GIS-based model of biogeochemical functioning of drainage network (Seneque-Riverstrahler) has been implemented, taking into account the main constraints (geomorphology, hydrology, point and diffuse sources) controlling the major processes governing the nitrogen fluxes.

Results and Discussion

Agriculture is responsible for a severe nitrate contamination of surface and groundwater resources, incompatible with the production of drinking water in the watershed. Despite directives, the increasing trend of water nitrate concentrations has not been inverted, but at best the concentrations have stabilized. The surplus of N brought to cropland soils as fertilizer with respect to the export of N with the harvested products is mostly leached. Among the 2400 kgN km^{-2} yr^{-1} leached, 47 % is exported at the outlet of the basin. The larger part of the remaining 53 % is eliminated by denitrification especially in the soils profile and in the riparian zones (Fig. 2). When conditions for denitrification are not optimum (e.g. lack of full anaerobiosis, substrate limitation, etc), N_2O is emitted, reaching ≈10 % of N_2. The contribution of agriculture managed land to N_2O has been estimated to 70 % of the land surface area (Garnier et al 2009) (Fig. 2). This current situation was modeled with the Seneque-Riverstrahler model, simulations being well in agreement with the observed fluxes and concentrations in the drainage network. A curative scenario involving the creation of drainage ponds showed a reduction in water nitrate concentration, but an increase in N_2O emission. A change of the agricultural system from conventional to organic farming (OF) both reduces nitrate concentrations and N_2O emission. OF requires a local supply in organic manure and an outlet for alfalfa which represents 2 or 3 years of the 8-years OF rotation in these areas. We have therefore explored the conditions required for the reintroduction of cattle breeding in this territory, traditionally producing the Brie cheese and a reconversion of some cropping land to grassland, especially in the footslope along the rivers.

Conclusions

The shift from manure-based to synthetic nitrogen fertilisation, has led to a strong land specialisation of agriculture in the whole Seine basin (Billen et al 2012). It exports 80% of its cereal production but has to import most of its animal proteins from the North and West of France, importing themselves about 30% of their feed from South America. The Orgeval watershed is well adapted to test such agriculture changes, allowing the agricultural landscape to diversify. This scenario will be explored at larger territorial scales.

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EFFECT OF A NOVEL NITRIFICATION INHIBITOR ON THE N2O EMISSION FROM A SOIL CROPPED WITH WINTER WHEAT

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Nitrous oxide (N_2O) is a climate relevant trace gas which contributes about 8% to the anthropogenic greenhouse effect (IPCC, 2007). N_2O production in agricultural used soils is affected mainly by N fertilization, which provides the substrates for the main microbial N_2O sources (i.e. nitrification and denitrification). A promising strategy to reduce N_2O emissions is the use of nitrification inhibitors (NI), such as 3,4 dimethylpyrazole phosphate (DMPP). Compared with a conventional N fertilizer application, NIs have been shown to reduce N_2O emissions by >30% (Akiyama et al., 2010). Due to the known N_2O reducing properties of DMPP on ammonium sulfate nitrate (ASN) we now tested the effect of a NI on calcium ammomium nitrate (CAN), a fertilizer with a lower ammonium (NH_4) concentration as compared to ASN. Here we present the first emission measurements with the novel NI Dimethylpyrazole succinic acid (DMPSA). The objective of this study was to quantify the effect of a NI application (DMPP and DMPSA) on the annual N_2O emissions from a loamy soil cropped with winter wheat.

Materials and methods

N_2O fluxes were measured with closed chambers between 15th March 2012 and 21st March 2013 in a fully randomized block experiment (n=4) on the experimental station “Heidfeldhof” of the University of Hohenheim, 13 km south of Stuttgart, Germany. Gas samples were taken weekly and measured with a gas chromatograph equipped with an electron capture detector (ECD). Additional samplings were conducted during expected high flux events, i.e. after fertilizer application or after tillage. 180 kg N ha^{-1} were applied as ASN, ASN+DMPP, CAN and CAN+DMPSA. N fertilizer amount was calculated according to the German Fertilization Ordinance (“good agricultural practice”).

Results and discussion

The N_2O emissions after fertilizer application were low (figure 1). Since the incorporation of straw increased the N_2O emissions after the harvest in all treatments, we speculate that the reason for the low fluxes after N-fertilization was the limited availability of easily degradable C, which serves as an electron donator for denitrifying microorganisms. As compared to the treatment with ASN, the share of the emissions during the vegetation period to the annual emission was higher when CAN was applied (table 1). The reason for this might be the higher nitrate content of CAN. The annual N_2O emission of both fertilizers does not differ, nevertheless. When applied together with the fertilizers, both NIs reduced the annual N_2O emission in the same order of magnitude (table 1). The N_2O mitigating effect of both NIs occurred only during the vegetation period. After the harvest no further N_2O reduction was observed in the treatments with NIs. Neither type of fertilizer nor use of a NI had an effect on yield and N content of wheat grain (not shown).

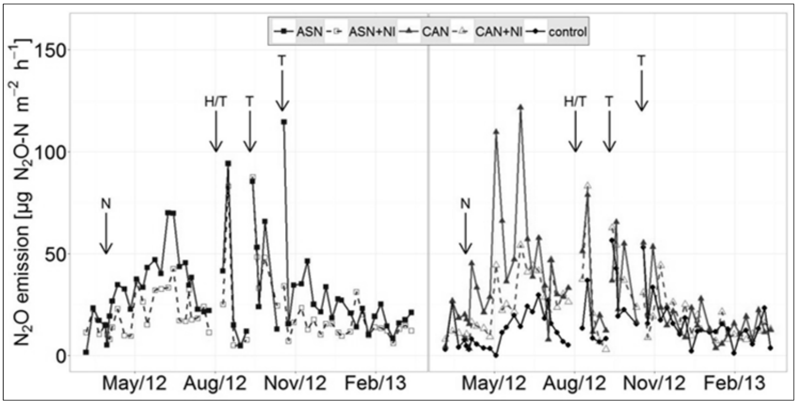

Fig. 1: Mean N_2O flux rates as affected by N fertilization and by addition of a NI. (N = N fertilization, H = harvest, T = tillage)

Table 1: N fertilization and mean N_2O emission during and after the growing season as affected by N-fertilizer and by application of a NI.

Treatment	Vegetation period		Winter		Total		Annual mitigation by NI
Period	15/03 – 25/07/12		26/07/12 – 21/03/13		15/03/12 – 21/03/13		
	(kg N_2O-N ha^{-1})						%
control	0.4	e	0.9	n.s.	1.3	c	
ASN	1.2	b	1.6	n.s.	2.8	a	33
ASN+NI	0.8	d	1.1	n.s.	1.9	b	
CAN	1.6	a	1.3	n.s.	2.9	a	26
CAN+NI	1.0	c	1.2	n.s.	2.1	b	

Mean values with different letters represent significant differences (Student-Newman-Keuls = 0.05).
ASN=ammonium sulfate nitrate; NI=Nitrification inhibitor; CAN=calcium ammonium nitrate.

Conclusions

The use of DMPSA as a NI reduced the annual N_2O emission from a loamy soil cropped with winter wheat. When compared to DMPP, no differences in the annual N_2O emission were found. DMPSA can be an effective N_2O mitigating tool in regions where CAN is often used as a fertilizer or when a higher nitrate concentration in the fertilizer is required.

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PRECISE PREDICTION OF AMMONIA EMISSION FROM MANURE: INCLUSION OF PH CHANGES AS AFFECTED BY EMISSION OF AMMONIA AND CARBON DIOXIDE

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The rate of ammonia (NH3) emission from a liquid is related to the pH at the surface, which may be different from the bulk pH. Here we describe models for emission of NH3 from animal manure that include this effect. Animal manure is a major source of NH3 emissions, which represent a loss of fixed nitrogen from farms, and contribute to poor air quality and degradation of terrestrial and aquatic ecosystems (Sutton et al. 2011). Rate of NH3 emission is sensitive to manure pH due to a shift in speciation toward free ammonia (NH3(aq)) as pH increases. Manure pH is mainly controlled by NH3 and carbon dioxide (CO2), which are volatile (Sommer and Husted 1995). Limited measurements suggest that CO2 emission increases surface pH and therefore NH3 emission. Results described below for ammonium bicarbonate solutions suggest that the surface pH of manure may be 1 unit higher than the bulk pH, resulting in a ca. 10-fold increase in NH3 emission (Fig. 1). A better understanding of this interaction is necessary to develop more accurate methods for estimating NH3 loss.

Models

We developed three models that include reactions among major manure solutes (NH4+, NH3(aq), CO2(aq), H2CO3, HCO3-, CO3-2, H+, and OH-), diffusive transport through manure, and volatilization of NH3 and CO2. With model E, all reactions are at equilibrium at all times, while model K includes kinetic reactions for H2CO3 (dehydration of H2CO3 to CO2(aq) is slow). Models E and K include activity corrections with the Debye-Hückel equation, but assume a single diffusivity for all species. Conversely, model D uses a distinct diffusivity for each species, but did not include kinetic reactions or activity corrections. Models E and K are described in more detail in Hafner et al. (2012) and D in Petersen et al. (in preparation). To evaluate the models, we measured pH gradients and emission of NH3 from manure and defined solutions. Solutions were made with NH4HCO3 or NH4Cl. Agarose or undisturbed conditions were used to restrict aqueous transport to diffusion. A software-controlled system was used to measure pH at a resolution of 100 μm.

Examples of findings

All three models predicted that rapid CO2 emission causes an increase in manure pH near the exposed surface, resulting in an increase in NH3 emission soon after exposure to air. The pH increase exceeds 1.0 unit in some cases. Measurements of NH3 emission and pH gradients in defined solutions were consistent with model predictions (Fig. 1). Slow H2CO3 dehydration was found to moderate the effect of CO2 emission on surface pH and NH3 emission. Using model K, we quantified the effect of environmental properties and manure characteristics on NH3 emission and pH. Results show that NH3 emission increases with temperature, and this effect is exacerbated by the kinetically-limited reactions. As another example, an increase in total inorganic carbon (TIC), at a constant initial pH, can substantially increase NH3 emission.

Beyond manure pH and total ammonia nitrogen (TAN), manure composition can have a significant effect on NH3 emission. Development of accurate models for estimating NH3 loss from field-applied manure will probably require recognition of these interactions. Quantification of the rate of H2CO3 dehydration in manure is needed. Both eukaryotes and prokaryotes produce carbonic anhydrases, which catalyze the reaction, but it is not clear if this enzyme is active in cattle manure. Our predictions may have implications for reducing NH3 emission. For example, both model predictions and emission measurements from manure show that an increase in mixing (e.g., stirring) can substantially reduce surface pH and actually reduce the rate of NH3 emission, despite increased transport of TAN to the surface.

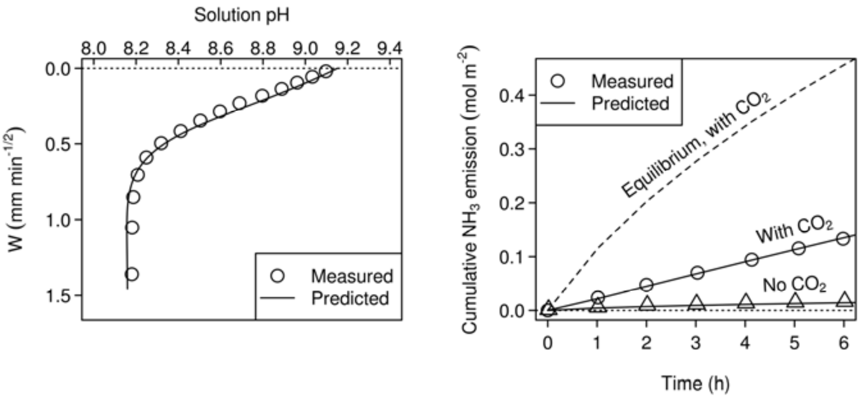

Figure 1. Left: Measured pH profile and profile estimated using model D. Time and distance is transformed to one dimension W. Right: Ammonia emission from defined solutions, measured and predicted with models E and K. Initial pH was 7.8 for both with CO2 (NH4HCO3) and without CO2 (NH4Cl).

Conclusions

Our models show that knowledge of manure TAN and bulk pH is not sufficient for predicting NH3 emission. Future work should include determination of carbonic anhydrase activity in cattle manure, measurement of sorbed and solid-phase ammonia, and application of these results to develop models that can accurately predict NH3 emission from field-applied manure.

Acknowledgements

USDA Agricultural Research Service and the Grønt udviklings-og demonstration program (Gylle – IT), Ministeriet for Fødevarer, Landbrug og Fiskeri – NaturErhvervstyrelsen. Lars B. Pedersen and Preben Sørensen (Aarhus University) for constructing the temperature sensors and pH electrodes (European Research Council, grant no. 267233)

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REASSESSMENT OF SWISS AMMONIA EMISSION FACTORS AFTER SLURRY APPLICATION

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The Swiss model Agrammon [1] which is used for the calculation of the national emissions of ammonia (NH_3) within the framework of the UN Convention on Long-range Transboundary Air Pollution [2] applies an average emission factor (EF) of 50% (cattle slurry) and 35% (pig slurry) of the total ammoniacal nitrogen (TAN) for splash plate (SP) application on grassland. For low emission application techniques such as trailing hose (TH) and trailing shoe (TS), specific factors for emission reduction of 30% and 50%, respectively, are used. The EF for SP application, which is considered as the reference technique, mainly bases upon the ALFAM model [3] and measurements in Switzerland from the 1990s [4]. Recent studies for Swiss conditions exhibited significantly lower emissions [5,6]. As a consequence, a new measurement campaign was launched to evaluate current EFs for slurry application used in the Swiss NH_3 emission inventory.

Materials and Methods

Over a period of two years, 16 field experiments were conducted at four different grassland sites in Switzerland. Experiments consisted in three to four plots of approx. 30 x 30 m^2, with one plot representing the reference system (cattle slurry, CS, applied by SP) and the other plots comparing different application techniques, slurry types, or application times. At the centre of each plot, an impinger based sampling device measured the NH_3 concentration at two heights (approx. 0.9 m and 1.6 m). Additionally, the background concentration was measured upwind. For each NH_3 sampling device, the emission rate was calculated using the backward Lagrangian stochastic Model WindTrax [7].

Results and Discussion

The average EF for the reference system SP/CS amounted to 22.8% of the applied TAN, with a range of 9.8 to 45.6% TAN. This is approximately half the level of what is used as reference EF in the Swiss model Agrammon. Even under high emission conditions (i.e. SP application of slurry with a relatively high dry matter content at noon on a hot and windy summer day), the determined EF did not exceed 50%. Comparing different emission reducing application techniques to the reference SP, the average reduction rates were 50.5% for TH, 53.1% for TS and 76.3% for Shallow Injection (SI). They were within the range given in the literature review by Webb et al. [8] and rather at the top end of the proposed reduction efficiencies given in the UNECE Guidance Document [2]. Investigations on the daytime of application revealed roughly 30% higher emissions at noon, than in the morning and in the evening. Figure 1 compares the measured NH_3 emissions to the predicted emissions from the ALFAM model. The trend of the measured emissions coincides with the ALFAM model, however, a systematic offset of roughly 23% TAN arises between the predicted and the measured emissions. Part of this systematic offset is likely due to the important fraction of Swiss data in the ALFAM database from the 90ies. A thorough reevaluation of these data showed a combination of errors producing an overestimation.

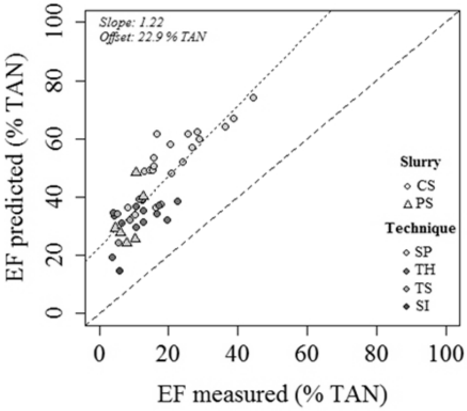

Figure 1: Measured EF compared to the predicted EF by the ALFAM model [3]. PS = Pig Slurry.

An extensive data analysis on the measured emissions from the reference system SP/CS revealed a strong dependency of the emission height on both, the air temperature (T) and dry matter content of the applied slurry (DM). The dependency was best described by the log-linear relationship given by:

$$EF/(\%\ TAN) = \exp(1.86+0.038*T/(°C)+0.0173*DM/(g/L))$$

Further emission driving parameters such as wind speed, application rate and others, did not add more information to the explanation of the variation of the EF.

Conclusion

This new measurement series of ammonia emissions after slurry application supports the earlier findings of Spirig et al. [5] and Sintermann et al. [6]. The results suggest a reduction of the emission factor related to the reference technique SP by 50% for Swiss conditions.

Acknowledgement

We would like to thank the Swiss Federal Office for the Environment (FOEN) and the Swiss Federal Office for Agriculture (FOAG) for financing this project.

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NITROUS OXIDE EMISSIONS FROM GRASSLAND AS IMPACTED BY FERTILISER NITROGEN FORMULATION

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Nitrogen (N) fertiliser is an important source of greenhouse gas emissions in Ireland accounting for almost 3% of the annual total (CO_2 equiv). Fertiliser is also the largest variable cost on Irish farms at over €400m annually. Increasing N use efficiency (NUE) is an important development needed to help achieve Food Harvest 2020 targets in a financially and environmentally sustainable way. Use of nitrate-N (NO_3-N) based fertilisers, under wet temperate conditions in Ireland, can result in fertiliser nitrous oxide (N_2O) emission factors which are significantly greater than the IPCC default of 1%. Switching from NO_3-N to urea based fertilisers could potentially reduce N_2O loss. Whilst increased use of urea based N fertilisers could increase national ammonia (NH_3) emissions, the use of urease inhibitors can manage NH_3 loss risk. The objective of this study is to evaluate the effect of switching from calcium ammonium nitrate (CAN) to urea or urea with a urease inhibitor (Agrotain™) on nitrous oxide emissions in temperate grassland.

Materials and Methods

The experiment took place at three permanent pasture sites: a silt loam, Johnstown Castle (JC), a sandy loam, Moorepark (MP) and a clay loam Hillsborough (HB). The experimental treatments were: 5 rates each of (a) CAN, (b) Urea and (c) Urea + Agrotain, at 100, 200, 300, 400 and 500 kg N ha $^{-1}$, one rate each of (d) Urea + DCD and (e) Urea + Agrotain + DCD (200 kg N ha $^{-1}$) and (f) a control. The experimental design was a randomized complete block with five replicates. The experiment simulated a grazing environment; the annual fertiliser N rate was applied in five equal splits through the growing season with grass harvested prior to each application. The N_2O emissions were measured throughout the year using the static chamber method and used to generate the N_2O emission factor.

Results and Discussion

There was a consistent trend for higher N_2O emissions from the CAN treatments compared with the urea treatments at all three sites: Figs 1, 2, 3. N_2O fluxes observed follow a trend clay loam > silt loam > sandy loam. Soil texture is closely related to both water holding and infiltration capacity thus these results are in line with other studies which linked soil drainage to N_2O emissions, with highest emissions produced on poorly and moderately drained soils and the lowest on well drained soils. The N_2O fluxes are closely related to fertiliser N application particularly at the clay loam HB site (Fig. 2). The reduction in emissions during the summer in the JC and MP sites can be linked to soil moisture conditions during the summer period in both sites. Each of the large peaks in the HB data can be linked to rainfall events. Minimal emissions were recorded during the drier conditions following the mid July peak at HB.

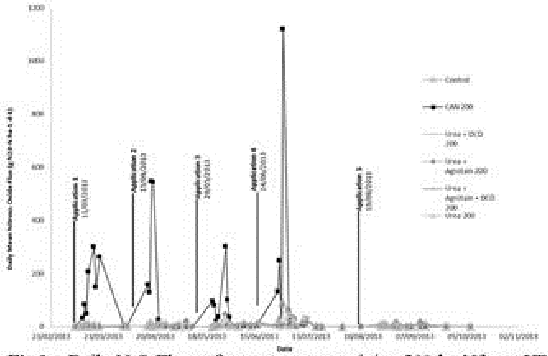

Fig 1 – Daily N_2O Fluxes for treatments receiving 200 kg N/ha at HB

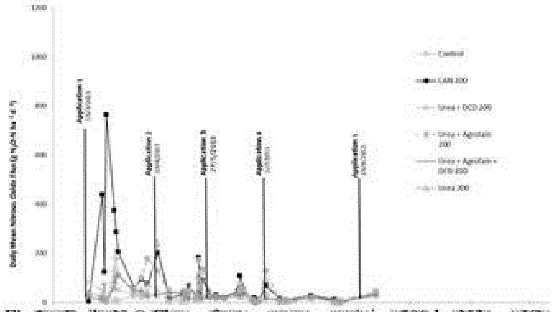

Fig 2 – Daily N_2O Fluxes for treatments receiving 200 kg N/ha at JC

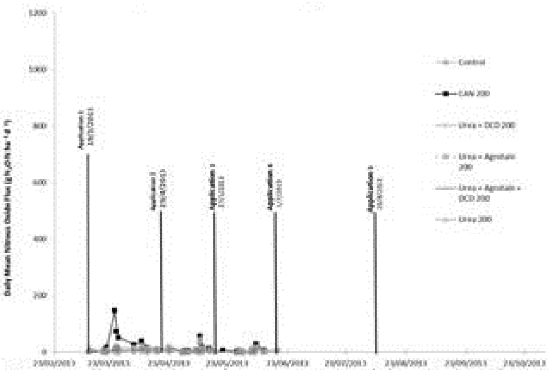

Fig 3 – Daily N_2O Fluxes for treatments receiving 200 kg N/ha at MP

Preliminary Conclusions

Use of urea based N fertilisers shows promise as a strategy for managing N_2O emission risk relative to CAN. N_2O emissions were closely related to fertiliser N application and to soil moisture. These results indicate that soil texture is a significant factor for evaluating N_2O emission risk.

Acknowledgements

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MECHANISMS OF INORGANIC NITROUS OXIDE PRODUCTION IN SOILS DURING NITRIFICATION AND THEIR DEPENDENCE ON SOIL PROPERTIES

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N_2O is an important greenhouse gas and today's single most ozone depleting substance. Soils have been identified as the major source of N_2O. Microbial processes nitrification and denitrification are considered the major N_2O sources. However, N_2O production in soils, especially during nitrification, is far from being completely understood. N_2O release during both processes has been described by Firestone and Davidson (1989) in their conceptual 'hole-in-the-pipe' model. The model attributes N_2O emissions from soils during nitrification and denitrification to leaks in the N transformation from ammonium to nitrate and the incomplete stepwise reduction of nitrate to N_2. Until now, the mechanisms behind this 'leaky' N_2O emission during nitrification have not been explained so it is unknown what drives these emissions. Several abiotic reactions involving nitrification intermediate hydroxylamine (NH_2OH) have been identified leading to N_2O emissions, but are neglected in most current studies. It is known that NH_2OH can be oxidized by several soil constituents to form N_2O (Bremner 1997). For better mitigation strategies it is mandatory to understand the underlying processes of N_2O production during nitrification and their controlling factors.

We studied N_2O emissions from different soils in laboratory incubations. Soils used covered a wide range of land use types from cropland to grassland and forest. Soil incubations were conducted at conditions favorable for nitrification. Soil samples were placed into vials, deionized water or NH_2OH solution was added, respectively, and water content was adjusted to the desired level. Vials were closed immediately for incubation. Experiments were conducted with non-sterile and sterile (autoclaved) soils. After 6 hours of incubation time the vials were sampled for N_2O mixing ratios. Additionally, CO_2 mixing ratios were analyzed to quantify microbial activity. For quantification of both gases a gas chromatograph (PerkinElmer, USA) was used. To get insight into the dynamics of N_2O formation, soil samples were placed in flow-through chambers, NH_2OH solution was added, and chambers were flushed continuously with pressurized air. N_2O production was quantified, at a temporal resolution of 1 Hz, using quantum cascade laser absorption spectroscopy (Aerodyne Research, USA). Furthermore, isotope ratio mass spectrometry (Isoprime, UK) was used to analyze the isotopic signature of the produced N_2O (i.e. $\delta^{15}N$, $\delta^{18}O$, and ^{15}N site preference).

We observed large differences in N_2O emissions between different soils upon NH_2OH addition. While a forest soil showed hardly any reaction to the addition of NH_2OH, a very high and immediate formation of N_2O was observed in a cropland soil. N_2O production after NH_2OH addition was also observed in autoclaved samples confirming an abiotic production. CO_2 data additionally suggested an abiotic N_2O production, as CO_2 production was not enhanced by NH_2OH addition to live soils. Laser spectrometry measurements revealed very fast reaction kinetics. An agricultural soil showed a turnover of about 50% in less than one hour. This proved that the fast reaction rate is also valid in soils. Further, isotopic signatures of N_2O were used to identify underlying processes. The site preference of ^{15}N in N_2O is believed to be a powerful tool to disentangle different N_2O production and consumption processes. We observed site preferences of about 33-36‰ in N_2O emissions caused by NH_2OH addition. This was in the same range as results found by Heil et al. (2014) for abiotic NH_2OH oxidation as well as by Sutka et al. (2003) for nitrification. To find a dependence on soil properties, we correlated N_2O emission rates after NH_2OH addition with soil chemical properties. We found three primarily controlling factors of NH_2OH induced N_2O production in the following order: soil pH, C/N ratio, and Mn content. The combination of those could explain up to 90% of the variability of the N_2O emissions caused by NH_2OH addition. Soil pH showed the strongest correlation with N_2O emission rates. This is explained by the higher stability of NH_2OH (pKs 5.8) at lower pH due to increasing protonation. C/N ratio had a strongly negative correlation with N_2O emission rates. Finally, Mn could oxidize free NH_2OH to N_2O. Although it was shown that NH_2OH can also react with Fe^{3+} to form N_2O (Bremner 1997), we could not find a correlation between Fe in soils and N_2O emission rates. This can be explained by the higher redox potential of the MnO_2/Mn^{2+} redox pair compared to the Fe^{3+}/Fe^{2+} redox pair.

Our results suggest a coupled biotic–abiotic production of N_2O during nitrification. We showed that some soils have a very high potential to oxidize NH_2OH so that abiotic N_2O emissions in those soils seem only to be limited by NH_2OH availability. We hypothesize that N_2O production is the result of a leakage of the nitrification intermediate NH_2OH, which acts as the substrate for abiotic oxidation to N_2O in the soil matrix. N_2O emissions during nitrification could then be explained as a function of nitrification rate and the combination of soil properties as mentioned above. However, further research is necessary to consolidate this relationship.

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NITRATE LEACHING FLUX IN DEEP VADOSE ZONE IN WHEAT-CORN DOUBLE CROPPING SYSTEMS IN NORTH CHINA PLAIN

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The North China Plain(NCP) is the most important wheat and maize production region in China, which produces over 20% of nation's food supply. High water and nitrogen (N) fertilizer inputs are critical for maintaining the high crop yield in the region (Chen et al., 2011; Zhang et al., 2011). However, excess fertilizer application and overdraft of aquifers for irrigation have caused contamination and depletion of groundwater in the NCP (Zhang et al., 1996; Hu et al., 2005; Ju et al., 2006; Hu et al., 2010; Qiu, 2010; Sun et al., 2012). Therefore, accurate estimation of nitrate leaching flux in deep vadose zone is essential for the efficient and sustainable groundwater protection in the North China Plain.

Materials and Methods

This study was conducted at the Luancheng Agro-ecosystem Experimental Station (37°53′ N, 114°41′ E, elevation 50 m) of the Chinese Academy of Sciences, Shijiazhuang, Hebei Province. Soil water transportation and nitrate leaching in deep vadose zone were measured in a twelve-meter deepth well.Soil water content and matric suction were measured by using 20 cm long TDR-probes and pressure transducers, respectively. TDR-probes were inserted horizontally at 9 and 11 m depth. The pressure transducers (Watermark probe, Campbell co., USA) were employed to determine soil matric suction at 8.75 m, 9.25 m, 10.75 m and 11.25 m depth, respectively, for the calculation of the corresponding water potential gradient. Soil water contents and matric suctions were recorded by TDR100 system (Campbell Co., USA) automatically every six hour from August 2009 to December 2012. Unsaturated hydraulic conductivity was determined by an evaporation measurement device (HYPROP, HYdraulic PROPerty analyzer, UMS co., Germany) in laboratory. Darcy's law was employed to estimate the water flux in the unsaturated zone. Soil water was collected by a portable vacuum pump from suction cups every 2 weeks, the suction cups were inserted 80cm into the soil from the well wall at 9 and 11 m depth. NO3--N in soil solution was measured by an automated flow IV injection analysis.

Results and Discussion

Precipitation is very important for water drainage. The precipitation in 2009-2012 were greatly varied, namely 557.3 mm, 330.3 mm, 413.1 mm and551.1 mm in 2009, 2010,2011 and 2012, respectively. In all four years, about 65-76% of the precipitation occurred during the summer (rainy season) between July and September. In addition, the precipitation in 2010 and 2011 was about 32.0% and 14.4% less than the long-term average, whereas in 2009 and 2012 they were about 14.8% and 13.5% greater than the long-term average.

The precipitation intensities and the consecutive days of precipitation events are important factor of water drainage. The consecutive precipitation (2009-8-26, 93.7mm and 2012-8-12, 96.2mm) could influence soil water content at 9 m and 11 m depth with a time lag. The time of water reaching to 11 m depth soil horizon lagged about 10 days behind 9 m depth. Three or four irrigation applications mainly supplied to crop growth, they had no effect on the water increase at the deep soil.

Table 1 lists the water flux at 9 m and 11 m depth estimated by Darcy's law during different period. The water drainage at 9m and 11m were 55.6mm and 14.8 mm of the whole measurement period (from August 2009 to December 2012), the nitrate leaching loss were 17.3 kg ha-1 and 5.9 kg ha-1.

Table 1. Water flux estimated by Darcy's law during different period (mm)

Soil depth	2009.8-2009.12	2010	2011	2012
9 m	19.6	17.8	7.7	10.6
11 m	13.7	0.2	0.0	0.8

Conclusions

In the North China Plain with a thick vadose zone, consecutive precipitation and irrigation could influence soil water content at 9 m and 11 m depth with a time lag. Water potential gradients were positive value at 9 m and 11 m, which showed that soil water always moved downward in the measurement period. The vertical water infiltration rate at 9m and 11m were 16.3mm yr-1 and 4.4mm yr-1. The nitrate leaching losses were 5.7kg ha-1 yr-1 and 1.1 kg ha-1 yr-1.

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THE NEW NITRIFICATION INHIBITOR DIMETHYLPYRAZOLE-SUCCINIC ACID (DMPSA) REDUCES N2O EMISSIONS FROM WHEAT UNDER HUMID MEDITERRANEAN CONDITIONS

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The intensive management of agricultural soils has led to high nitrogen inputs which have resulted in a significant increase of nitrous oxide emissions from soils. In order to mitigate N_2O emissions nitrification inhibitors have been developed (Menéndez et al., 2006; Weiske et al., 2001). The objective of this work was to evaluate the effect of the new nitrification inhibitor dimethylpyrazole-succinic acid (DMPSA) on N_2O emissions during a whole wheat crop cycle under humid Mediterranean conditions.

Materials and Methods

This work was conducted in a wheat crop in the Basque Country in 2012-2013 during a whole year, from wheat sowing in November 2012 to sowing of the next crop in November 2013. A randomized complete block factorial design with four replicates was established, with an individual plot size of 40 m^2. Three main treatments were applied: a control treatment without fertilizer, a second one with ammonium sulphate (AS) and a third one consisting in the combination of AS with dimethylpyrazole-succinic acid (DMPSA), a new nitrification inhibitor developed by EuroChem Agro. Fertilization was split into two amendments: 20^{th} February (tillering), when 60 kg N ha^{-1} were applied and 18^{th} April (stem elongation), when 120 kg N ha^{-1} were applied.

N_2O emissions measurements were started on November after sowing and finished after a whole cropping season one year later. Measurements were conducted three days per week after fertilizer applications and decreasing this frequency gradually to one or two times per week. Cumulative N_2O emission during the sampling period was estimated by averaging the rate of emission between two successive determinations, multiplying that average rate by the length of the period between the measurements, and adding that amount to the previous cumulative total. Nitrous oxide emission was measured using closed chambers (Menéndez et al., 2008). Samples were analysed by gas chromatography (GC) (Agilent, 7890A) equipped with an electron capture detector (ECD) for N_2O detection. N_2O standards were stored and analysed at the same time as the samples.

Results and Discussion

Ammonium sulphate application increased N_2O emissions with respect to the unfertilized treatment (Table 1), being the annual losses 0.36 % of the nitrogen applied. This emission factor was quite lower than that suggested by IPCC (1996) guidelines which assume an N_2O emission factor of 1% of fertilizer-N applied and similar to the emission factors described by Ranucci et al. (2011) under Mediterranean conditions.

Table 1. Cumulative N_2O losses during the whole year and emission factor. In brackets is shown the percentage of reduction induced by the nitrification inhibitor.

	Cumulative N_2O losses g N_2O-N ha^{-1} $year^{-1}$	Emission Factor
Control	473 b	-
60+120 kg AS	1121 a	0.36%
60+120 kg AS+DMPSA	493 b (56%)	0.01%

Different letters indicate significantly different rates using Duncan Test ($P< 0.05$; n=4).

The new nitrification inhibitor reduced N_2O emissions up to control treatment levels (Table 1). The percentage of reduction of N_2O losses in the AS+DMPSA treatment with respect to AS was 56 %. This percentage reduction is higher than those described by other authors (Pfab et al., 2009; Weiske et al., 2001) using DCD or DMPP in crops in areas with cold climates. This high efficiency resulted in an emission factor of only 0.01%.

Conclusion

The new nitrification inhibitor DMPSA results to be efficient reducing N_2O emissions up to levels of unfertilized treatment in humid Mediterranean conditions. Thus, the default emission factor for fertilizers with DMPSA should differ from the emission factor for fertilizers without nitrification inhibitor.

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EFFECT OF CROP MANAGEMENT AND CHANGES IN CLIMATIC FACTORS ON NITRATE LEACHING FROM A LONG TERM ORGANIC CEREAL CROPPING SYSTEMS IN DENMARK

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Nutrient pollution of the aquatic environment, especially nitrate pollution of surface and groundwater, is regarded as a significant environmental problem in Denmark. Organic farming has been included as one of the measures to reduce nitrate leaching from Danish agriculture. However, large uncertainties remain in the estimates of nitrate leaching from organic farming. The aim of the study presented here was to investigate the effect of (i) cropping management and (ii) air temperature and precipitation on N leaching from cultivation of winter and spring cereals grown in a long-term organic farming experiment in Denmark.

Materials and Methods

In 1997 an organic arable crop rotation experiment was initiated at three locations representing different soil types and climate in Denmark. The experiment included three consecutive four-year crop rotation cycles with three treatment factors in a factorial design (1) crop rotation with different proportion of grass-clover and pulses in the rotation, (2) with and without catch crop, and (3) with and without animal manure as slurry (Askegaard et al., 2005). Where present, catch crops were undersown in the cereals or pulse crops in spring and were incorporated into soil by ploughing prior to sowing the next crop in the rotation. All crop residues were incorporated or left on the soil. Soil nitrate-N concentration was measured using four suction cells per plot in selected plots installed at 100 cm depth. The water balance and hence water drained from the soil profile was calculated using the EVACROP model. Nitrate leaching was estimated using the trapezoidal rule. The estimated daily N leaching was aggregated to annual values for the period from 1 April to 31 March, covering the main leaching period during autumn and winter. The weather variables temperature and precipitation were calculated as averages and cumulated values, respectively on a seasonal timescale (winter (Dec–Feb), spring (Mar–May), summer (Jun–Aug) and autumn (Sep–Nov)). The effect of cropping management and seasonal temperature and precipitation on N leaching were investigated in a linear mixed model using the restricted maximum likelihood method. The model is:

$$Y_{ply} = \mu + \mathrm{c}_{1,\mathrm{a}}T_a + \mathrm{c}_{1,\mathrm{w}}T_w + \mathrm{c}_{1,\mathrm{sp}}T_{sp} + \mathrm{c}_{1,\mathrm{s}}T_s + \mathrm{c}_{2,\mathrm{a}}P_a + \mathrm{c}_{2,\mathrm{w}}P_w + \mathrm{c}_{2,\mathrm{sp}}P_{sp} + \mathrm{c}_{2,\mathrm{s}}P_s + \mathrm{Loc}_\mathrm{l} + \mathrm{Rot}_\mathrm{lpy} + \mathrm{PreCrop}_\mathrm{ly} + AWcover_{ly} + c_3H_l + C_{lb} + D_{ld} + E_{lp} + F_{ly}$$

where Y_{ply} is the square root of the measured N leaching from plot p at location l in year y; a, w, sp, s denote autumn, winter, spring and summer for year y; b and d are the level of block and sub-block; μ, *T, P, Loc, Rot, PreCrop, AWcover* and *H* are the fixed effects, main effects of air temperature (°C), precipitation (mm), location, crop rotation, previous crop, autumn and winter crop cover and depth of the A horizon (cm), respectively; c_1, c_2, c_3 are regression coefficients; C_{lb}, D_{lbf}, E_{lp} *and* F_{ly} are independent normally distributed random variables of block, sub-block, plot and year within location with zero mean and variances. The model described above was fitted to the square root transformed leaching data for winter and spring cereals separately. Only factors with significant effects were kept.

Results and Discussion

On average, N leaching for winter cereals varied considerably between years, from 17 kg N ha^{-1} in 2007/2008 to 63 kg N ha^{-1} in 1997/1998. This was due partly to differences in the amount of percolation. The same trend was also observed for spring cereals. On average, N leaching for both cropping systems was close to the mean N leaching of 34 kg N ha^{-1} y^{-1} measured in Danish arable systems at standard N fertilizer rate, although nitrate leaching was higher for spring cereals. The final model explained 68% and 77% of the variation in the square root transform of annual N leaching for winter and spring cereals, respectively. N leaching was shown to be site specific and driven by both climatic factors and crop management. The results showed that management of crop during autumn as the main determinant of N leaching. Nitrate leaching was lowest for a catch crop soil cover during autumn and winter and for a soil cover of weeds/volunteers when compared to bare soil conditions.

There were significant effects on N leaching of location, rotation, previous crop and crop cover during autumn and winter (Table 1). The relative effects of temperature and precipitation were found to differed between seasons and cropping systems; hence corroborating the findings of (Patil et al., 2012). Temperature was shown to positively affect N leaching with a difference between spring and winter cereals, though. The positive effect of temperature may be due to its positive effect on soil N mineralisation of soil organic matter and crop residues leading to higher soil mineral N. Precipitation also increased N leaching for the two cropping systems, because the flux of soil mineral N out of the root zone is largely influenced by the difference between precipitation and crop evapotranspiration. N leaching will therefore increase with increasing precipitation in autumn. Surprisingly, the effect of precipitation during winter was not significant for both cereal systems. This might be due partly to dilution effects since N concentration was low during winter and to the possible losses through denitrification which could be induced by temporary anaerobic conditions caused by saturated conditions in soil during winter.

Table 1. Results of the regression analysis relating square root transform of N leaching (kg N ha^{-1}) to the climatic and crop management parameters for winter (WC) and spring (SC) cereals.

	$\sqrt{Nitrate\ leaching}$				
	WC	SC		WC	SC
Intercept	-6.4350 *	-2.7131 ***	**Location**		
Temperature (°C)			Foulum	1.21 **	
Autumn		0.3322 ***	Flakkebjerg	0.04 NS	
Winter	0.3525 ***		**Rotation**		
Spring			R4	-0.90 **	
Summer	0.4082 **		**Horizon Depth (cm)**	0.03 ***	
Precipitation (mm)			**Pre-crop**		
Autumn	0.0160 ***	0.0089 ***	Grass-clover	-0.49 NS	2.69 ***
Winter			Potato	-2.09 ***	
Spring	0.0125 ***	0.022 ***	Pulses	-1.04 **	0.54 ***
Summer	0.0045 **	0.0074 ***	**Cover crop**		
			Weeds	-2.24 ***	-1.19 ***
			Catch Crop	-3.05 ***	-2.85 ***
			Grass-clover		-2.67 ***

Significance levels: *** $P < 0.001$, ** $P < 0.01$, * $P < 0.05$, NS not significant.

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REDUING NITROGEN LOSSES IN CHINESE CROP SYSTEMS

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The fate of fertilizer nitrogen (N) in crop systems is an integrated result of the crop N uptake, immobilization and residual in the soil, and losses to environments. It is the key step for evaluating the effects of fertilization practices on agronomy and the environment. There are close relationships among these three major fates of applied fertilizer N, which is influenced by many factors including crop feature, soil characters, climate conditions, and agricultural management practices. Many studies showed that there was a common negative correlation between N recovery rate and N losses rate (Ju et al., 2009). Therefore, reducing the losses rate is the key for improving N use efficiency and alleviating environmental stress.

Results and Discussion

Reducing Total Synthetic N Fertilizer Consumption in National or Provincial Scale

Chinese agriculture consumed more than 33 million tonnes of synthetic N fertilizer in 2008, accounting for about 30% of global N fertilizer consumption (Zhang et al., 2012). Studies across many sites and crops in recent years showed that reduction of at least 30% of current farmer's N application rates in cereal crops could maintain or even marginally increase the yields, while substantially reducing N losses to environment (Ju et al., 2009; Zhang et al., 2012). In cash crops, such as vegetable and fruit trees, this reduction even could reach to more than 50% of current farmer's practices (Zhang et al., 2012). With the potential reduction of N fertilizer use in whole country estimated conservatively at 30%, Chinese agriculture needs only 23 million tones of synthetic N fertilizer to support food security. Only by reducing total fertilizer N inputs can degraded environments be gradually restored and protected.

Controlling the Total N Inputs in Farmer's Plot Scale

Controlling the total N inputs in each farmer's plot to a rational level is the key step for reducing N losses. We first propose the concept of regional mean optimal N (RMON) application rate, calculated from the average of economically optimum N rates (i.e. the point of the N rate where the marginal grain production value is equal to the marginal N fertilization cost) based on large number of on-farm field experiments. In principle, RMON defines a regional rational N rate in combination with fine tuning according to specific field conditions, taking into account the relative uniformity of climate, soil N fertility, and absence of a satisfactory soil N test index.

Developing an approach for optimizing appropriate N application rates for the smallholder plots that constitute the main agricultural system in many developing countries is vital to control N misuse and reduce N losses. Recently we proposed the concept of theoretical N rate (TNR) to answer the important question about how much fertilizer N should be applied to intensive systems based on the N fluxes due to transformation processes in the soil-crop-environment continuum (Ju and Christie, 2011). We define TNR as the theoretically calculated fertilizer N rate with the quantitative relationships of the core N fluxes among fertilizer N, soil N and crop uptake N in the crop root zone to obtain high targeted yield, maintain soil N balance and minimize environmental risk. We concluded that N fertilizer rate could be roughly estimated as above ground crop total N uptake in these intensive managed cereal crops, such as maize and wheat. This simple N recommendation can be easily implemented in field and is a very cost-effective approach.

Counting N Supply By Organic Materials Into Total N Inputs

Application of solid manures and municipal waste composts to farmland is the primary method for nutrients recycling. It can reduce pollution caused by direct organic waste discharge into water bodies or NH_3 and NOx emitting into atmosphere. The available N in organic waste could substitute part of mineral N for crop demand, meanwhile improve soil fertility. Currently, another reason for over-use of chemical fertilizer N frequently occurring in Chinese farmer's practices is that the N supplied by organic manures is usually not taken into account (Zhang et al., 2012). In view of input-output relationship in a crop season, the balanced N management strategy provide a simple and useful tool for farmers to calculate mineral N input rate when using organic materials.

Improving Application Techniques Of N Fertilizer

Considering poor application techniques, such as surface broadcasting urea or ammonium bicarbonate before irrigation or rainfall, which were practiced by most farmers within their small scattered plot system, machinery deep application of fertilizer can substantially reduce N losses and further decrease N application rate. This approach could become more important soon if the size of farmers' plots increase as a result of land use reforms in China. Nevertheless, deep application can also be carried out with small simple machinery in current small plot, and greatly reduces ammonia volatilization.

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MODELLING OF AMMONIA VOLATILISATION IN FLOODED RICE FIELDS: THE MODIFIED JAYAWEERA-MIKKELSEN MODEL

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Since the 1980's, models describing soil and floodwater nitrogen dynamics for flooded rice fields have been extensively developed. In lumped-parameter models, ammonia volatilisation from the floodwater is typically described by either first order kinetics (Chowdary et al., 2004; Antonopoulos, 2010) or by logistic equations (Liao et al., 2012). In these ammonia volatilisation models, floodwater properties, such as floodwater pH and temperature, were not conceptualised. Meanwhile, in the model of Jayaweera and Mikkelsen (1990), the ammonia volatilisation is regulated by the observed floodwater pH, temperature, depth, and wind speed. However, the application of this model is limited for flooded soils without rice plants, and the effect of photosynthetic aquatic biomass activity on the floodwater pH was not taken into account. The objective of this study is to extend the Jayaweera-Mikkelsen model to predict ammonia volatilisation in flooded rice fields and to calibrate the model against secondary data.

Materials and methods

In this study, we adapted the Jayaweera-Mikkelsen model and made four major modifications. First, to estimate the two-hourly floodwater pH, we integrated a part of the floodwater routine from CERES-Rice (Godwin and Singh, 1998) and APSIM-Oryza (Gaydon et al. 2012) into the Jayaweera-Mikkelsen model. The floodwater pH is described by an equation which follows an absolute sinusoidal trend as function of photosynthetic aquatic biomass activity. Second, instead of assuming an ammonia concentration of zero at steady state, we conceptualised the pH and temperature dependent ratio of ammonia and ammonium in the model (Fig. 1a). The equation is adopted from Jayaweera and Mikkelsen (1990). Third, we included the rice plant nitrogen uptake by conceptualising the uptake directly from the floodwater. This is based on the study by Safeena et al. (1999), who demonstrated that rice plants are able to directly absorb nitrogen from the floodwater when roots are well developed. Fourth, the ammonia volatilisation from the floodwater is described by first order kinetic and independent of wind speed. As yet, we also assumed constant floodwater depth. The simulated ammonia volatilisation, using estimates of the unknown nitrogen-gift, is compared with secondary data obtained from Tian et al. (2001).

Results and discussion

In the simulation, the floodwater pH is generated by assuming that the photosynthetic aquatic biomass activity is only active between 12.00 to 18.00 hours, and reaches the maximum at 12.00 hours. The floodwater temperature is assumed to range between 25 and 38 °C, and reaches the maximum at 12.00 hours. Fig. 1b shows that the cumulative ammonia volatilisation predicted by the modified Jayaweera-Mikkelsen model reasonably approximates the data reported in Tian et al. (2001). The corresponding ammoniacal-nitrogen concentration in the floodwater is shown in Fig 1c. Previously, observed ammonia volatilisation from basins filled with ammonium sulphate solutions (ammonical-nitrogen concentration of 50 mg/L) and placed in a flooded rice fields, was used to validate the Jayaweera-Mikkelsen model (Jayaweera et al., 1990). Applying this model under planted conditions might result in overestimation of the cumulative ammonia volatilisation as time approaches infinity. By including the rice plant nitrogen uptake, the modified Jayaweera-Mikkelsen model can be used to simulate ammonia volatilisation in flooded rice fields.

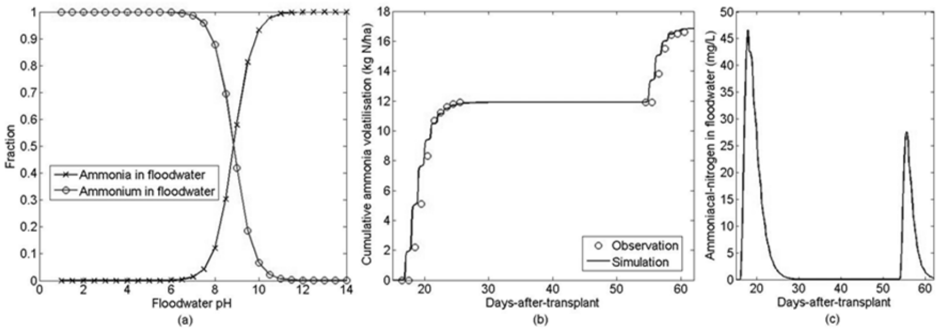

Fig. 1 a) pH-dependent-ratio free ammonia and ammonium in floodwater at 38 degrees Celsius, b) cumulative ammonia volatilisation and, c) the corresponding ammoniacal-nitrogen in the floodwater (data from Tian et al. 2001)

Conclusion

The modified Jayaweera-Mikkelsen reasonably approximates the data reported in Tian et al. (2001). Future work includes calibrating and validating this model for mid-season ammonia volatilisation.

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MODEL BASED ANALYSIS OF FACTORS GOVERNING N2O EMISSION FROM BIOENERGY CROPPING SYSTEMS

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There is growing concern that greenhouse gas (GHG) emissions from cropping systems for biomethane production might mitigate intended GHG emission savings. Biomethane as the major bioenergy source in Germany is based on intensive and highly productive cropping systems, mainly dominated by maize, while alternative crops are investigated. N_2O emission is the major GHG source in all intensive cropping systems because of high fertilizer N inputs. The influence of different cropping systems on N_2O emissions in interaction with temporal and spatial variations in weather and soil properties is still largely unknown. The aim of the presented study was to adapt a process oriented model approach for the analysis of factors governing N_2O emissions under different environmental and management conditions.

Materials and Methods

A dynamic simulation model was developed as a combination and adaptation of existing approaches. It calculates N_2O emissions from nitrification and denitrification in soil. Nitrification is calculated by first order kinetics with the influence of water content and soil temperature (Zhou et al. 2010, Hansen et al. 1990). Denitrification is calculated by Michaelis-Menten kinetics with influence of soil water, soil temperature, and C mineralisation rate (Zhou et al. 2010, Del Grosso et al. 2000, Hansen et al. 1990). The boundary conditions (nitrate concentration, water content in soil) for these processes are calculated with already existing modules for crop growth, water balance and mineralisation. A field trial was conducted at two sites in Northern Germany (Wienforth 2011). N_2O fluxes were measured with closed-chamber technique on average once a week, March 2007 – April 2009 (Senbayram 2009). Cumulative N_2O emissions were calculated by linear interpolation between measured daily fluxes. The present work is based on data of a maize monoculture at a sandy site and a crop rotation of silage maize, wheat (for whole crop silage use) and Italian ryegrass (catch crop) at a loamy site. We evaluated an unfertilised treatment (N1) and two levels of mineral N fertilization with 120 kg N ha^{-1} for maize and wheat and 160 kg N ha^{-1} for grass (N2) and 360 kg N ha^{-1} for maize and wheat and 160 kg N ha^{-1} for grass (N4).

Table 1: Measured and simulated annual N_2O emissions and emission factors for the period 1.4.2007- 15.3.2009

Crop rotation	Mineral Fertilizer [kg N ha^{-1} a^{-1}]	N_2O emission measured [kg N ha^{-1} a^{-1}]	N_2O emission simulated [kg N ha^{-1} a^{-1}]	Emission factor measured [% Fertilizer]	Emission factor simulated [% Fertilizer]
Maize – Wheat – Grass (loamy site)	0	1.0	1.6		
	200	4.3	3.2	2.1	1.6
	440	6.7	5.9	1.5	1.3
Maize – Maize (sandy site)	0	0.8	0.5		
	120	0.8	0.7	0.7	0.6
	360	3.3	1.7	0.9	0.5

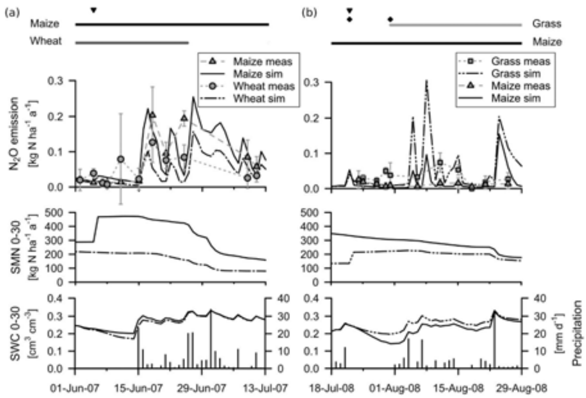

Figure 1: Daily measured (meas) and simulated (sim) N_2O emissions of the N4 treatment. Corresponding simulated SMN in 0-30 cm soil depth, SWC in 0-30 cm soil depth and daily precipitation at the loamy site for (a) Maize and Wheat 1.6. – 13.7.2007, (b) Ryegrass and Maize 18.7. – 29.8.2008. Triangles: fertilization dates, rhombs: tillage dates, horizontal lines: growing periods of maize, wheat or grass. Error bars for standard error of the mean of each treatment (n=3).

Results and Discussion

As a prerequisite for the simulation of nitrification and denitrification the boundary conditions were described as close to measured data as possible using dynamic modules and linear interpolation of measured data (plant N uptake). Then two parameters were fitted to N_2O emission data, all other parameters were taken from literature. The model reproduced the annual variation between crop rotation and fertilizer levels of the parameterization data set (Table 1). The simulated time course of daily N_2O fluxes is generally plausible (Figure 1). Fertilization events result in increasing soil mineral nitrogen (SMN). Higher SMN in soil results in higher N_2O fluxes in maize in comparison to wheat, under conditions of similar soil water content (SWC) in both crops (Figure 1a). On the other hand higher SWC under grass results in higher N_2O fluxes, even when SMN is lower than in maize (Figure 1b). During dry periods there are generally no N_2O fluxes (early June 2007). N_2O emissions start with increasing SWC after the dry period (Figure 1a). The model was not able to simulate increasing N_2O fluxes after tillage events properly (Figure 1b). Precipitation events mostly resulted in N_2O emission peaks. Sometimes simulated N_2O emission peaks were not captured by the measurement scheme but seem plausible to have happened (e.g. end of August 2008).

Conclusions

The model calculated reasonable annual N_2O emissions after site specific calibration. The variation due to differences in weather, fertilization, location and crop rotation was reproduced. In some cases the dynamics of simulated data is more plausible than linear interpolation of measured data. Accuracy of the results clearly depended on adaptation of boundary conditions to measured data.

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NEW STRATEGIES FOR NITROUS OXIDE MITIGATION AFTER GRASSLAND CULTIVATION

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Nitrification inhibitors (NI) are commonly applied together with mineral fertilizers to improve N use efficiency and reduce leaching losses. In recent years a consistent reduction of N2O emissions has also been shown (Akiyama et al., 2010), making use of NI a potential N2O mitigation option.

Traditionally, grasslands are ploughed, but field data indicated that shallow incorporation by rotovation two weeks prior to ploughing can reduce N2O emissions compared to ploughing directly. The time course of N2O emissions (see Baral et al., this volume) shows that nitrification of residue-N is at least partly responsible for N2O emissions, and we hypothesized that inhibition of nitrification of N from decomposing grass-clover can reduce N2O emissions. In this study, a novel use of NI is investigated, in which the inhibitor is sprayed onto the plant residues just before cultivation. DMPP (3, 4-Dimethylpyrazol-phosphate) appears to be more suitable for this application than other NI with respect to mobility, vapour pressure and persistence (Subbarao et al., 2006). A precondition for this novel application is that there are no ecotoxicological effects on non-target soil organisms or key functions. Thus, the objective of this study was to investigate new strategies for reducing gaseous nitrogen loss from N2O emission following grassland cultivation.

Materials and Methods

The soil used in the first experiment to identify ecotoxicological effects was collected from 0-20 cm depth within a grass-clover pasture and sieved (4mm). DMPP was added at 0x, 1x and 10x the recommended rate (0, 1 and 10 kg ha-1, respectively), and incubated for 1, 7 or 14 days (n = 3) at 20oC. The soil samples were packed to a dry bulk density of 1.3 g cm-3 in 100 cm3 steel rings with a final water-filled pore space (WFPS) of 50%. Potential ammonium oxidation (PAO) and PLFA were determined according to Belser and Mays (1980) and Petersen et al. (2002); Soil mineral N dynamics, dehydrogenase activity and fluorescein diacetate hydrolysis were also analyzed, but are not presented here. Statistical analyses were carried out by SPSS 18.0. The evaluation of ecotoxicological effects will be followed by incubation of DMPP amended grass-clover residues collected at the time of spring tillage, simulating cultivation patterns. Nitrous oxide emissions and soil mineral N dynamics, and the microbial response to the crop residues, will be monitored.

Results and Discussion

The PAO rate describes the ability of a soil to nitrify under suitable soil conditions. With 1x DMPP, PAO rates were slightly, but significantly reduced after 1 and 7 d, and still marginally reduced after 14 d. With 10x DMPP, the reduction relative to control soil was highly significant at all three samplings. These results indicated that DMPP, when applied at the recommended rate of 1 kg ha-1, has only minor effects on ammonium oxidizing populations, whereas a significant inhibition of their activity is expected. The negative effect on nitrifiers occurred already one day after DMPP addition, in accordance with many previous studies showing that AOB growth is significantly inhibited by DMPP application (e.g., Di et al., 2011). Substrate availability may influence the stress imposed by DMPP, and the experiment will be repeated with amendment of an NH4+ source.

PLFA concentration is considered to be a sensitive and reliable indicator of soil microbial biomass. In the present study, no significant differences were found in PLFA concentrations among treatments (Fig. 1b), even with the 10x DMPP application rate after 14 d. In the present study, no negative effects of 1x and 10x recommended application rates were also observed for dehydrogenase activity and fluorescein diacetate hydrolysis (data not shown). Thus, even if DMPP can be detected in the soil for up to 4–5 months, it appears to be safe for non-target soil organisms, in accordance with (Tindaon et al., 2012).

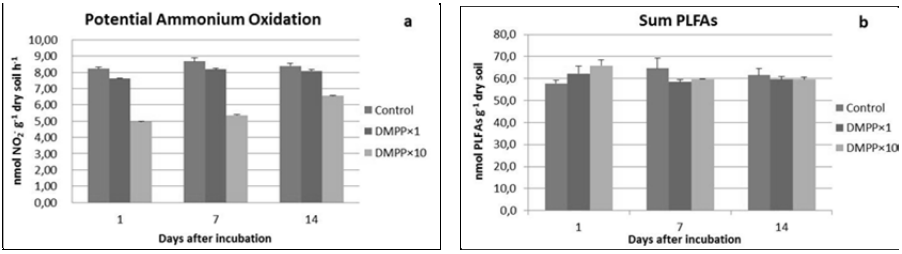

Fig. 1 Potential ammonium oxidation rate (a) and PLFA concentrations (b); DMPP ×1 and DMPP ×10 correspond to 1 and 10 kg DMPP ha-1; Vertical bars represent standard errors (n =3).

Conclusion

Ecotoxicological tests indicated that DMPP has little impact on non-target soil organisms or soil functions, and that it may therefore be safely introduced to soil. In the next phase of this study, DMPP will be applied to grass-clover prior to cultivation; effects on N2O emissions will be presented at the conference.

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AMMONIA LOSS RATES FROM UREA AND CALCIUM AMMONIUM NITRATE APPLIED TO WINTER WHEAT ON THREE DIFFERENT SITES IN GERMANY

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Granular urea is one of the dominant N fertilizers in crop production. However, its use is characterized by a relatively high potential of ammonia (NH_3) losses (Sommer et al. 2004). Appropriate quantification of NH_3 emissions from urea and other N fertilizers is a matter of increasing relevance and is governed by international protocols (EMEP/CORINAIR 2007). Assessments and evaluations should be based on emission data from field measurements with application conditions as in agricultural practice. However, the respective data base in central Europe is very limited. Thus, the principal objective of the present paper is to evaluate new datasets demonstrating NH_3 losses from urea and calcium ammonium nitrate applied to winter wheat under field conditions at 3 sites in Germany and to compare these emissions with EMEP emission factors.

Materials and Method

The field experiments were conducted in winter wheat at 3 Agricultural Research Stations in Northern (site N), Southern (site S), and Central Germany (site C): Site N (54°18'N, 9°58'E); altitude 30 m a.s.l; annual mean temperature 8.3°C; annual precipitation 750 mm; sandy loam, pH 6.5, total organic C 1.5%, total N 0.1%. Site S (48°24′N, 11°41′E): 480 m a.s.l.; 7.7°C; 800 mm; silty loam, pH 6.5, C 1.2%, N 0.13%. Site C (51°22′N, 12°33′E): 130 m a.s.l.; 9.1°C; 620 mm; sandy loam; pH 6.5; C 1.1%, N 0.12. At site N and site S, field measurements were conducted from 2011 to 2013, at site C in 2013. Three different N fertilizers were tested in 3 or 2 split applications per year (total N 180-220 kg/ha, growth stages EC 21, EC 32, EC 47-51, EC 37-51): calcium ammonium nitrate (CAN), granular urea (U), granular urea with the nitrification inhibitor DCD/TZ (U+NI). Fertilizers were applied simultaneously (n = 4, plot size 9 m x 9 m with 9 m x 9 m interspaces) and ammonia emissions were measured by the calibrated passive sampling method (Gericke et al. 2011) which has been validated as giving accurate absolute NH_3 loss values (Quakernack et al. 2012). The relative NH_3 losses (% of applied N) of fertilizers were compared by ANOVA and analyzed with the tukey test (HSD)

Results and Discussion

Relative ammonia emissions from field applied urea in single applications ranged between 1 and 30% (data not shown), similar as in many earlier studies. The weighted average N loss by NH_3 emission over all sites and years was 6.9% from U, 7.8% from U+NI, and 2.0% from CAN (Table 1). The different NH_3 emission levels were due to the different chemical nature of U and CAN in agreement with Sommer et al. (2004). The respective NH_3 emission factors for Germany in the EMEP guideline

(EMEP/CORINAIR 2007) are 11.5% for U and 0.9% for CAN. Thus, the measured NH_3 loss from U was only 3 to 4 times higher than from CAN instead of 12 times higher as provided by EMEP. This deviation can be explained by the EMEP factors being based on merely a few records mainly selected from Werden & Jarvis (1997). Several earlier studies in Southern Germany also indicated lower NH_3 losses following U treatments (Weber et al. 2002, Schraml et al. 2009). The use of the nitrification inhibitor DCD/TZ in the treatment U+NI showed no consistent effect on the cumulative NH_3 losses. Quantity and dynamics of NH_3 emission were affected by soil moisture, temperature and rainfall. Consequently, the application date strongly influenced the NH_3 loss rate from all fertilizers studied. Thus, differentiated emission factors for different application periods could be an alternative approach. Our calculations imply average relative emissions for U of about 4% for the earlier applications (EC 21-32) and 13% for the latest application (EC 47-51). The respective values for U+NI treatments were 3.4% (EC 21) and 12% (EC 37-51) (Table 2). The observation of low NH_3 losses following early U applications is relevant with respect to the common fertilization practice in Germany. Cereals with lower N demand and oil seed rape get nearly the whole amount of fertilizer N in the early spring.

Conclusions

In conclusion, similar to organic fertilization the consideration of appropriate application conditions (low temperatures, rainfall, early application dates) can be an effective measure to mitigate N losses from urea applied to arable crops apart of incorporation in soil by banding or irrigation. For German conditions, the NH_3 emission rates of U and CAN considerably differ from the corresponding EMEP factors. This questions the current European and German methodology for assessing NH_3 emissions from field applied CAN and U. Fixed NH_3 emission rates for climatic agro-regions for U and CAN seem to be insufficient. Differentiated NH_3 loss factors for different application periods could be a practicable approach. However, a broader data base, an agreed measurement methodology, and a more refined model analysis are the best way for deriving robust emission factors in future

Table 1: Relative cumulative NH_3 losses (% of applied N) from U, U+NI and CAN treatments per year and over 3 years (Sites: N = Northern, S = Southern, C = Central Germany).

treatment:	**U**		**U + NI**		**CAN**	
	% N loss	ANOVA *	% N loss	ANOVA*	% N loss	ANOVA *
site N, 2011:	9.8	b	14.1	a	3.3	c
site N, 2012:	10.5	b	16.1	a	1.9	c
site N, 2013:	4.9	a	3.0	b	1.0	c
site N, 2011-2013:	***8.4***		***11.1***		***2.0***	
site S, 2011:	11.5	a	14.1	a	7.0	b
site S, 2012:	< 1**		< 1**		< 1**	
site S, 2013:	3.9	b	5.9	a	2.0	b
site S, 2011-2013:	***4.2***		***5.6***		***2.4***	
site C, 2013:	8.4	a	3.8	b	1.3	b
sites N+S+C, 2011-2013:	***6.9***		***7.8***		***2.0***	

* Different letters represent a significant statistical difference per site and year at P<0.05 (tukey test).

** Calculation of NH_3 emission by CPS method was not possible because of small differences between fertilized plots and plots without N fertilization.

Table 2: Relative cumulative NH_3 losses (% of applied N) from U and U+NI treatments in different fertilization periods (characterized by plant growth stages).

treatment:	U			U + NI	
plant growth stage:	EC 21 to 23	EC 32	EC 47 to 51	EC 21	EC 37 to 51
	% N loss	% N loss	% N loss	% N loss	% N loss
site N, 2011-13:	5.3	3.3	16.8	4.6	19.0
site S, 2011-13:	0.9	4.9	4.7	0.7	9.3
site C, 2013:	4.9	4.6	16.1	4.9	2.7
sites N+S+C, 2011-2013:	***3.7***	***4.1***	***12.7***	***3.4***	***12.2***

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POTENTIAL OF HIPPURIC ACID AND BENZOIC ACID TO MITIGATE NITROUS OXIDE EMISSIONS FROM URINE PATCHES

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On average Irish ruminants deposit approximately 140 million litres of urine to grazed grasslands on a daily basis. This represents a nitrogen (N) load to the soil of approximately 1.2 million tonnes. The majority of this N load is in the form of urea which rapidly hydrolyses to ammonium, and is then nitrified and may be subsequently denitrified. As a result of these enzyme-catalysed, microbial processes a potent greenhouse gas, nitrous oxide (N_2O), is produced. According to the IPCC 2% of N applied to the soils in urine will be lost N_2O emission (IPCC, 2006). A rise in ruminant numbers to achieve national production goals could therefore lead to elevated fluxes of urine-derived N_2O. Urine composition varies with diet and its constituents hippuric acid (HA) and one of its breakdown products, benzoic acid (BA), have previously both been linked to mitigation of N_2O emissions from urine patches in laboratory studies (Bertram et al., 2009, van Groeningen et al., 2006, Kool et al., 2006). However, the sole field study to date found no effect of HA and BA concentration on N_2O emissions possibly due to unfavourable environmental conditions (Clough et al., 2009). Therefore the aim of this study was to investigate the *in situ* effect of urine and its constituents HA and BA on N_2O emissions as well as on soil mineral N (ammonium NH_4^+ and nitrate NO_3^-) from urine patches applied to an Irish soil. The specific objectives were: 1) to quantify the impact of urine patches on N_2O emissions, 2) to evaluate the effect of various HA and BA concentrations in urine on the N_2O emitted and soil mineral N levels, 2) to quantify the reduction in N_2O emissions from various urine compositions, 3) and to assess the differences between HA and BA amended urine on N_2O emissions and soil mineral N.

Materials and Methods

This experiment was conducted at a loam site in the South-East of Ireland. The experimental design was a randomized block with six replicates. Urine amended with urea, HA, and BA was applied to 0.16 m^2 plots of perennial ryegrass (*Lolium perenne*) pasture at a rate of 1012.5 kg N ha^{-1} in October 2013. Concentrations of urine constituents in various treatments are summarised in Table 1. Nitrous oxide was sampled from static chambers and analysed by gas chromatography. Nitrate and ammonium in soil extracts was determined by colorimetric analysis using an Aquakem 600 discrete analyser.

Results and Discussion

Urine application to the soil significantly influenced subsequent N_2O emissions. Nitrous oxide fluxes from the control treatments ranged from 0.49 to 42.2 g N_2O-N ha^{-1} d^{-1} and were generally significantly lower than from urine treatments. Highest daily flux of N_2O reaching 731.9 g N_2O-N ha^{-1} d^{-1} was seen from the urine + HA2 treatment 11 days after application (Fig 1 a). No significant differences between urine treatments were found. Cumulative N_2O losses were significantly higher ($p< 0.0004$) from urine treated soils than from the control soils (Fig 1 b) however no significant differences were distinguished between different urines. Controls yielded approximately 0.3 kg N ha^{-1}, while urine treatments varied between 9.2 (urine + HA1) to over 11.5 kg N ha^{-1} (urine + HA2). However, total N_2O-N losses expressed as a percentage of the applied N were significantly lower to the IPCC default emission factor of 2% and were: 0.88% for urine + HA1, 0.95% for urine + BA, 1.02% for urine, and 1.12% for urine + HA2.

Conclusions

Urine application resulted in elevated N_2O fluxes which had diminished after five weeks. Addition of HA or BA to urine did not reduce N_2O fluxes therefore manipulation of these urine constituents by diet manipulation shows little promise as a mitigation tool for N_2O emission. However, the proportion of urine-N lost as N_2O was significantly lower than the IPCC Tier 1 default value of 2%. This observation points to the importance of adopting the Tier 2 methodology as N_2O calculations to more accurately reflect the true emission.

Table 1. Hippuric and benzoic acid concentrations (mM) of urine treatments and the hippuric acid N content as a percentage of total urinary N.

Treatment	Hippuric acid		Benzoic acid
	(mM)	(% of urine-N)	(mM)
urine only	0	0.0	21
urine + HA1	8	1.2	52
urine + HA2	82	12.8	17
urine + BA	24	3.7	96

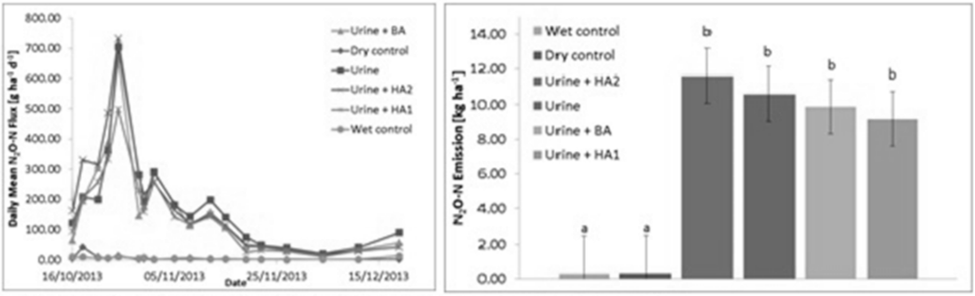

Fig. 1.a and b. Temporal (a) and cumulative (b) flux of N_2O-N from urine affected soil.

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NITROUS OXIDE EMISSIONS ASSOCIATED WITH LAND-USE CHANGE OF GRASSLAND TO ENERGY CROP PRODUCTION

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Bioenergy crops are increasingly popular alternative to fossil fuels used for electricity and heat generation due to their substantial yields combined with low inputs. As these crops are characterised by low nutrient requirements, they can facilitate mitigation of fertiliser-induced gaseous emissions and consequently aid meeting the greenhouse gas (GHG) reduction targets posed by various national and international policies. However very little research on GHG emissions associated with the cultivation of the energy crops is available. This study investigated the impact of land-use change (LUC) from pasture to two different energy grasses (Miscanthus x. giganteus and Phalaris arundinacea) on N2O emissions. A field trial investigated the effects of ploughing technique, fertiliser management, soil type and crop type on nitrogen (N) cycling within Miscanthus and Phalaris (Reed Canary Grass) over a two year period in comparison with pasture systems.

Materials and Methods

Five hectares of grassland was ploughed and converted to Miscanthus or Reed Canary Grass (RCG). These consisted of four large one hectare plots (two Miscanthus and two RCG) and a set of 18 small (3m x 10 m) plots. The effects of land-use change were examined by weekly measurements of N2O emissions from the large plots by a static chamber method over a two year period. The small plots were fertilised in year 2 with 80 kg N ha-1 and emissions measured. Soil temperature and water filled pore space were measured ancillary to N2O, additionally measurements of soil available nitrogen: ammonium (NH4+) and nitrate (NO3-), soil physical characteristics and grass yields were performed.

Results

Following the field-scale conversion to biomass crops the first year post-LUC exhibited high N2O fluxes with the highest losses from well-drained Miscanthus plot (9.3 ± 2.7 kg N ha-1 yr-1); indeed emissions were comparable to those of managed pasture systems (10.1 ± 3.4 kg N ha-1 yr-1). Emissions in the subsequent year were lower due to increases in both shoot and rhizome density. By contrast, emissions from newly established Reed canarygrass plots (2.90 ± 0.33 and 2.53 ± 0.09 kg N ha-1 yr-1 for well and poorly-drained plots respectively) did not differ significantly compared to fallow grassland plots (5.45 ± 1.17 and 3.83 ± 0.35 kg N ha-1 yr-1 for well and poorly-drained plots respectively). Upon fertilisation, the emission factor for Miscanthus and RCG were 0.9 and 0.7% respectively. This was significantly lower than the emission factor (2.8%) for fertilised pasture.

Preliminary Conclusions

- Ploughing of grassland resulted in considerable N2O release.
- N2O emission factor was lower for bioenergy crops compared to grasslands

Acknowledgements

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QUANTIFYING THE EFFECTS OF DISSOLVED ORGANIC MATTER CARBON ON DENITRIFICATION

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Sustainable soil organic matter (SOM) management of arable soils involves high annual returns of crop residues. This leads to temporarily and spatially increased abundance of dissolved organic matter (DOM) containing carbon (C) which is available for microbial utilization. Availability of C is an important driver for the production and reduction of the potent greenhouse gas nitrous oxide (N_2O) during denitrification. Specific carbon compounds have been shown to influence the rates of denitrifier N_2O and N_2 production, therefore affecting N_2O loss to the atmosphere. However, the interaction between SOM quality and quantity derived from residue decomposition with NO_3^- reduction pathways under anaerobic conditions remains poorly understood. Stimulating the denitrifier community abundance and activity higher abundance of DOM-C is hypothesized to increase denitrification with changes in timing, magnitude and duration of N_2O and N_2 emission peaks. The objectives of this study were to investigate the role native DOM abundance plays for nitrate reduction in order to identify opportunities of N_2O mitigation through SOM management in arable farming.

Materials and Methods

Denitrification rates were investigated using $^{14}NH_4{}^{15}NO_3$ fertilizer against a background of five DOM concentrations with four replicates in a 2-phase laboratory incubation experiment. In the first phase an arable loamy sand soil of inherently high OM content (5.6% total organic C) was supplemented with DOM obtained by hot-water extraction (HWE) of the same soil. After concentration by slow evaporation at 45°C for 72 h the HWE was re-applied to 50 g of soil at 80% water-filled pore space. The soils received rates of 0.15, 0.30, 0.45 and 0.60 mg DOM-C g^{-1} dry soil on days eight, six, four and two prior to fertilizer application to establish a gradient of labile OM background concentrations. Control soils received demineralised H_2O, respectively. Carbon dioxide and $^{14+15}N$-N_2O flux rates were measured daily. In a second phase, $^{14}NH_4{}^{15}NO_3$ fertilizer was applied at a rate of 120 kg N ha^{-1} to all experimental units. In addition to the aforementioned measurements, ^{15}N-N_2O and ^{15}N-N_2 samples were taken on a daily basis between days 0 (1h after application) and 7 as well as days 10, 14 and 21 to assess the effect of DOM quantity on denitrification rates and the N_2O-to-N_2 product ratios. Dissolved organic C, microbial biomass carbon and mineral ^{15}N pools were determined by destructive sampling on days 0, 1, 4, 7, 14 and 21.

Results and Discussion

Cumulative soil CO_2 emissions linearly increased with DOM-C applications (Fig. 1) indicating a positive response of the microbial biomass and activity to substrate quantity. The DOM application rates resulted in different background levels of DOC, microbial biomass C and activity throughout the incubation period. Within the first two days after fertilizer application ^{15}N-N_2O emissions peaked for all DOM treatments with a maximum flux of 7.8 µg ^{15}N-N_2O m^{-2} h^{-1} from the lowest DOM-C treatment (0.15 mg g^{-1} soil). This highlights the importance of the soil solution C-to-N ratio in driving N_2O emissions from soils under anaerobic conditions by either facilitating NO_3^- immobilization into microbial biomass or enhancing its reduction. Overall, total ^{15}N-N_2O emissions were found to decrease as DOM-C applications increased (p=0.009, n=4) (Fig. 2) which was due to greater reduction of N_2O to N_2. Thus, the abundance of DOM-C is contributing greatly to changes in denitrification product ratios.

Conclusions

Using a stable isotope approach, this experiment demonstrated the importance of native DOM quantity for driving N_2O losses from arable fields as well as its function in regulating denitrification product ratios. These results highlight the need to consider the soil's organic matter status when predicting greenhouse gas emissions from arable soils.

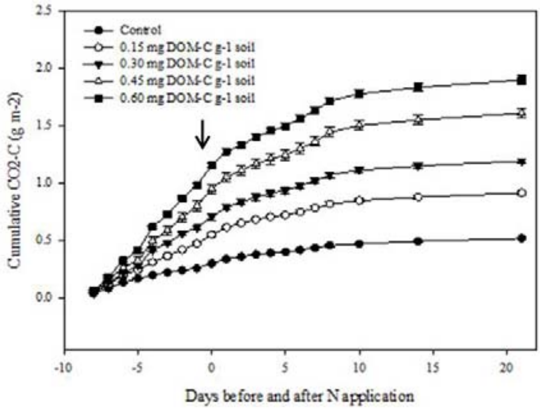

Fig 1. Cumulative CO_2-C emissions (g CO_2-C m^{-2}) for the control soil and dissolved OM (DOM-C) treatments throughout the incubation period. The arrow indicates N fertilizer application on day 0 prior to which DOM-C was applied on days -8, -6, -4 and -2. Error bars represent the standard error of the means (n=4).

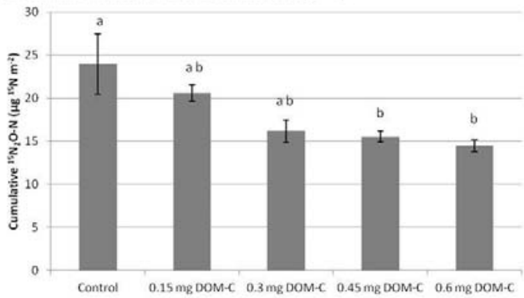

Fig 2. Mean total $^{15}N_2O$-N emissions (µg ^{15}N m^{-2}) for the control soil and dissolved OM treatments (mg DOM-C g^{-1} soil). Error bars represent the standard error of the means (n=4), bars not sharing a letter differ significantly (p<0.05).

SOIL NITROGEN IN SHRUB ENCROACHED MEDITERRANEAN OAK ECOSYSTEMS: EFFECT AFTER SHRUB CLEAR-CUT

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Shrub encroachment into grass-dominated ecosystems is occurring globally and particularly in arid and semi-arid ecosystems (Van Auken 2000). Anthropogenic activities, such as changes in land use (eg land abandonment), are key determinants in the encroachment process (Bugalho et al 2011). These changes are particularly important in the Mediterranean region which is a hotspot for global changes (Giorgi and Lionello 2008). Shrubs, in particular Cistus ladanifer, are expanding in the region due to their stress-tolerance ability (Simões et al 2008). Shrubs have different traits from herbs such as longer life-spans, higher above-ground and root biomass production, and higher tissue C:N ratios (McCulley et al. 2004), all of which can affect processes such as N cycling, decomposition and microbial activity. Our aim was to study the effect of shrub clear-cut on nitrogen availability and microbial activity

Material and Methods

This study was conducted in a cork oak (*Quercus suber*) open woodland invaded by *Cistus ladanifer*, in south-east Portugal ($38^0$47´N, $7^0$22´W). We established six paired plots of 20 by 20 m and in half of them all the shrubs were cut above-ground and all the biomass was removed from the plots. Total nitrogen given by the sum of N-NH4+ and N-NO3-, was determined in autumn, winter, spring in soil cores taken 5 cm in depth under tree canopies and in the open. Potential mineralization and nitrification and microbial activity were also determined after 10 days incubation under controlled conditions of soil moisture (50% of the water holding capacity) and temperature (25ºC). Shrub harvesting was done in September 2011 and the measurements were conducted until November 2012.

Results and discussion

Total nitrogen concentration was significantly higher in the clear-cut than in the shrub plots (GLM, repeated measures, $F= 107.15$, $P< 0.0001$), as expected because of the lack of N-uptake by shrubs. However, the potential mineralization, nitrification rates and the microbial activity were higher in the shrub than in the clear-cut plots. The maximum difference in total N between clear-cut and shrub plots was observed in January 2012 with 32.22±4.15 vs 5.69±1.14 in the open and 34.42±3.67 vs 6.73±1.10 mg N kg^{-1} dry soil under the canopies. Maximum potential mineralization rates were also found in January in both treatments, being 79% higher in the open and 70% higher under canopy in clear-cut plot. For both treatments, the potential mineralization rate was significantly higher under the tree canopies. In November 2012, mineralization rates were similar in the open in both treatments; but under canopy, the mineralization rates were higher in the clear-cut plots than in shrub plots. This suggests that clear-cut effects may disappear after one year of cutting. In the open, the potential nitrification rates were higher in shrub plots. In clear-cut plots, the potential nitrification rates remained inferior to 0.3 N $kg^{-1}.d^{-1}$ until May 2012 and increase drastically in November 2012 reaching 2.65±0.21 N kg-1.d-1 (significantly higher to 0.67±0.15 N kg-1.d-1 in shrub plot). Under the canopy, the potential nitrification rate was higher in shrub plots in November 2011 and May 2012 and similar in January 2012. In November 2012, the rates of potential nitrification in clear-cut plots also increased drastically to 3.65±0.95 N kg-1.d-1 (vs 1.37±0.49 N kg-1.d-1 in shrub plot). These values of potential nitrification rate in November 2012 approximate rates found in grassland plots with no shrubs (2.23±0.55 and 3.79±0.90 N kg-1.d-1 in the open and under canopy respectively) and tends to suggest that clear-cut effects may disappear after one year. Microbial activity was higher in the shrub than in the clear-cut plots (GLM, repeated measures, F= 293.013, P< 0.0001).

Clear effects of shrub clear-cut were found with a decrease in the potential mineralization, nitrification and microbial activity whereas total nitrogen concentrations were found higher. The results could point on effects on micro-environmental change such as humidity and temperature between treatments after the clear-cut. These changes could affect the microbial communities and therefore affect the nitrogen cycling. The 2011/2012 winter was especially dry which could have reduced N leaching from the clear-cutted forest (Holmes and Zac, 1999) that combined with a lack of N-uptake by shrubs, could cause N accumulation in the clear-cut plots. In the other hand, exudates such as phenolic compounds or terpenoids, released during the decomposition of *C. ladanifer* shrub roots (Dias, 2005) may constrain the nitrogen cycling by nitrogen immobilisation or nitrification inhibition (Smolander, 2012, White, 1994) and may also have effect on cellular level affecting the microbial population (White, 1994). It appears in our study that these possible effects may disappear after one year following shrub clear-cut. Further investigation on these processes will be carried on to complete and confirm this data.

SALT AS A DIURETIC FOR DECREASING NITROGEN LEACHING LOSSES FROM GRAZED PASTURES

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Nitrogen (N) leaching from grazed pastures represents a significant source of N loss and the main source of this is from urine excreted by the grazing animals (e.g. Ledgard 2001). Most leaching is associated with drainage over the late-autumn to early-spring period and mitigation practices targeted at reducing N leaching generally focus on practices in late-autumn and winter.

The large N leaching loss from animal urine patches occurs because of the high rate of N deposition in the patches (e.g. 400-1200 kg/ha equivalent). Thus, any practices that can result in a greater spread of the urine so that the rate of N deposition is reduced can potentially decrease overall N leaching losses. One potential mitigation option is the use of a diuretic such as salt.

This paper reports on three research components aimed at understanding the potential reduction in N leaching from administration of salt to cattle. The first is a cow metabolism stall study where cows were fed different levels of salt and the N excretion in urine and dung were determined. A grazing system trial with beef cattle had a treatment where the cattle were drenched daily with salt solution and a control treatment and the water consumption and urine N excretion patterns were measured. At this site, a lysimeter study was used to determine N leaching from urine deposited and different N rates to assess the potential effects of salt supplementation.

Materials and Methods

1. Metabolism stall study: Fifteen non-lactating Friesian dairy cows were placed in individual stalls with urine and dung collection devices attached to each cow. Five cows were allocated to one of three treatments; control or salt supplementation at 200 or 400 g/cow/day. After a 3-day lead in period, the urine and dung excretion from each cow was collected daily over 6 days and analysed for volume, weight and N concentration. Cows were fed silage at the equivalent of approximately 10 kg DM/cow/day, with the actual level of feed and water intake being recorded each day. Animal Ethics approval was obtained for the research and cow health was monitored.

2. Grazing trial: A beef cattle farm in the catchment of Lake Taupo, New Zealand had areas fenced up into 5 blocks with treatments randomly assigned to paddocks (0.4 ha) within each block. Treatment mobs of beef cattle (20/mob) were rotationally grazed through the treatment paddocks. Treatments included a control and daily drenching with salt solution at 150 g/cow/day during grazing periods in autumn and winter over a four year period (2007 to 2010). Water intake by cows from water troughs was recorded daily and cows were fitted with urine sensors (Betteridge et al. 2010) so that the location and volume of urine excretion was recorded via a GPS system.

3. Lysimeter study: Large intact soil cores (400 mm diameter x 600 mm depth) were collected from the pumice soil in an area near the above grazing trial that had been excluded from grazing for at least 6 months to avoid effects of residual animal excreta. These were placed in a trench and surrounded by soil. The lysimeter trial included treatments of a control or cow urine applied at 300 or 600 kg N/ha equivalent

in late-autumn. Climate data was recorded and leachate from the lysimeters was collected regularly and the volume and N concentrations measured over a 12 month drainage period.

Results and Discussion

The Metabolism stall study showed that salt supplementation led to increased water intake (by up to 80%) and increased daily volume of urination, with the urine having a lower N concentration (Figure 1). The grazing trial with beef cattle showed that salt supplementation resulted in an increase in water consumption from water troughs and a 13% increase in the average number of urinations per day from the salt-treated cows.

The lysimeter trial showed that decreasing the N concentration in urine from 600 to 300 kg N/ha reduced the average annual N leaching from 267 to 100 kg N/ha. Accounting for the 5 kg N/ha leaching from the control resulted in an average decrease (allowing for increased spread) of 25%.

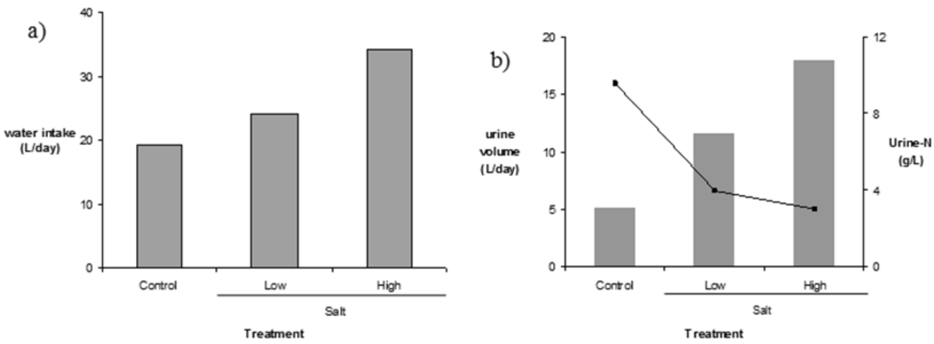

Figure 1: Effect of low and high daily salt supplementation to non-lactating dairy cows on a) daily water intake, and b) daily volume of urination (boxes) and N concentration of the urine (line).

Conclusions

These studies demonstrated that salt is an effective diuretic resulting in increased water consumption and increased volume and number of urinations per day. The latter is associated with a lower N concentration so that the rate of N excreted in urine is significantly decreased. Associated modelling work using a dynamic N cycling model showed that the use of salt supplementation has potential to reduce per-hectare N leaching by up to 25%. Thus, targeted administration of salt to cattle in late-autumn and winter can be an effective mitigation practice for decreasing N leaching in grazed pasture systems.

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EFFECT OF NITROGEN SOURCE ON NITRIC OXIDE AND NITROUS OXIDE EMISSIONS FROM A UK GRASSLAND SOIL

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To date only a few studies measured NO, N_2O, N_2 and CO_2 simultaneously, especially as N_2 emissions are very difficult to measure. Our denitrification incubation system (DENIS, Cardenas et al. 2003) enables us to incubate soil cores under an N-free atmosphere, by using a He:O_2 (80:20) mixture, allowing direct measurements of emitted N_2. Labelling of substrate N with ^{15}N is used to trace and link N-emissions to their source and provide insight in production and consumption pathways and temporal dynamics of the greenhouse gas N_2O. It has been shown that nitrifier- and denitrifier-derived N_2O differ in their ^{15}N to ^{14}N ratio and that denitrifying bacteria prefer the lighter N_2O over the heavier one, resulting in faster reduction rates to N_2 for the light N_2O and thereby enriching the remaining N_2O in ^{15}N. Conclusions about the underlying processes can be drawn from the isotopic composition of emitted N_2O. While using labelled N is a valuable and essential method to investigate N transformation processes, there is a potential that the addition of a higher amount of ^{15}N and thereby increasing the ratio of ^{15}N to ^{14}N could influence the processes occurring. In two different experiments the present study is using the DENIS to investigate the effects of N-source (KNO_3 and NH_4NO_3) and carbon (C) addition as well as labelling of the NO_3^- source on NO, N_2O and CO_2 emissions.

Materials and Methods

Soil samples were taken from a typical grassland soil from SW England. The soil was air dried to ~30-40% H_2O, sieved to < 2 mm and stored at 4°C until packing of cores. For each of the two experiments (Exp1; Exp2), 12 soil cores (140 mm diameter x 75 mm height) were packed to a bulk density of 0.8 g cm^{-3}. The moisture content was adjusted to final WFPS of 85% (Exp1) and 80% (Exp2), taking the later addition of 50 ml amendment into account. Cores were left over night and after final moisture adjustment placed into the DENIS, where they were kept at 20°C. To remove native N_2 from the soil, cores were flushed from the bottom with a He:O_2 (80:20) mixture at 30 ml min^{-1} over night. Flow rates were then decreased to 12 ml min^{-1} and the flow re-directed to flow over the surface of the soil core. Exp1 investigated the effects of NH_4NO_3 vs. KNO_3 addition and the effects of adding those N-sources with and without additional C in the form of glucose. It consisted of treatments: NH4NO3, KNO3, NH4NO3+C, KNO3+C. Exp2 investigated possible effects of using labelled NO_3^- and consisted of treatments: ^{15}N labelled KNO3+C (at 5 at%), unlabelled KNO3+C, control. For all treatments N was added at 75 kg N ha^{-1} and C as glucose at 400 kg C ha^{-1} in water. The control treatment consisted of water only. In both experiments 50 ml amendment were added to a sealed amendment chamber. The amendment chamber was flushed with He for 30 min to maintain an N_2-free atmosphere before releasing the amendment onto the soil cores. Measurements were taken "continuously" with one sample being analysed every 10 minutes resulting in bihourly values for each vessel. Emissions of N_2O and CO_2 were measured by GC with an electron capture detector (ECD), while NO concentrations were determined by chemiluminescence. All gas concentrations were corrected by the flow rate through the vessel and fluxes were calculated on a kg N or C per ha basis. Cumulative emissions were calculated by linear interpolation between sampling points. Soil samples were taken at the beginning and end of the experimental period to determine initial and final moisture contents and NH_4^+ and NO_3^- concentrations.

Results and Discussion

Figure 1 shows the cumulative emissions of the two experiments. Results of Exp1 show that higher N_2O emissions occurred when C was added to the amendment. NO emissions, however do not show a significant difference between treatments. CO_2 emissions were also higher with additional C indicating increased biological activity in those treatments corresponding with the increased N_2O emissions. With C addition N_2O appears instantly after amendment while without C addition the peak is about 24 h delayed (data shown on poster). This is probably due to the microbial community being stimulated by the additional C, therefore growing quicker and utilising more of the supplied N sources. Comparing the two N-sources of KNO_3 and NH_4NO_3, no significant differences in NO or N_2O emissions were detected between the respective amendments while with additional C, CO_2 emissions were slightly higher with KNO_3 than NH_4NO_3. Exp2 was looking at the effects of labelling the N-source (KNO_3). No difference was detected between the labelled or unlabelled treatments in any of the measured gases, but both treatments showed significantly higher emissions than the control. A KNO_3+C amendment was used in both experiments. Comparing emissions from KNO_3+C addition under 85% WFPS from Exp2 to those under 80% WFPS from Exp1 indicate higher NO and possibly N_2O, as well as CO_2 emissions under the lower WFPS.

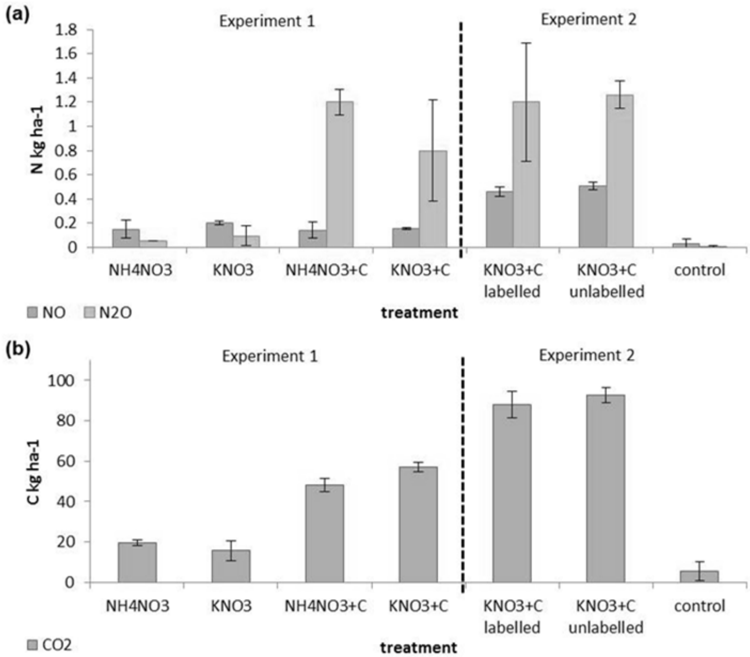

Figure 1: Cumulative emissions of (a) NO and N_2O in kg N ha^{-1} and (b) CO_2 in kg C ha^{-1}; error bars are standard deviation, n=2 for NH4NO3, NH4NO3+C and unlabelled; n=3 for KNO3, KNO3+C and labelled, n=4 for control.

Conclusions

This is the first time that NO was measured in the DENIS together with N_2O and CO_2 resulting in data collection at a relatively high frequency. Results show that NO emissions peak at around 6 hours after amendment application, followed by a CO_2 and N_2O peak. This study highlights the effects of adding a NO_3^- source together with an easily accessible carbon source on the transformation of the N in soil. Higher emissions were generally associated with additional C input. Emissions from amendments with labelled or unlabelled KNO_3 were not significantly different, and any possible effects on gaseous emissions are negligible.

Acknowledgement

The authors are grateful to the BBSRC for financial support.

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STUDY OF NITROUS OXIDE (N2O) LOSSES IN A DRIP IRRIGATED PEACH CROP SYSTEM

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Greenhouse gases (GHG) emissions are well studied in agricultural systems. Even though, little is known about the role of nitrogen (N) fertigation and irrigation systems on nitrous oxide (N_2O) emissions. Nitrogen fertigation management is important to maintain both productivity and fruit quality although it should be necessary to quantify N losses in the crop system. This paper studies the nitrogen losses in a peach crop due to N_2O losses as affected by irrigation system (surface vs. subsurface drip irrigation) and fertigation management (full vs. reduced fertigation).

Materials and Methods

The trial was conducted in 2012 in a commercial orchard of peach (*Prunus persica* (L.) Batsch. cv. Saturn) grafted on GF-677 and planted in 2011 in ridge planting system, spaced 5 x 2.5 m, in Los Monegros region (Huesca, Spain). The experiment was organized in a randomized complete block design with three replications. Four treatments about soil and water management were evaluated: drip irrigation at 100% of the calculated irrigation dose (R-DI-100); drip irrigation at 70% dose (R-DI-70); subsurface drip irrigation at 100% dose (R-SDI-100); and subsurface drip irrigation at 70% dose (R-SDI-70). Nutrients were provided through continuous fertigation by injecting a modified Hoagland solution (Hoagland and Arnon, 1950) with the following nutrients concentration: N 145, P 9.9, K 172.2, Ca 1.4, Mg 17.9, all of them in ppm, through the full irrigation cycle from 15th May to 30th July. In subsurface irrigation treatments, the dripperline was buried 0.25 m deep. Tree growth was assessed in 5 control trees per plot using two methods: 1) Trunk diameter measurement at the beginning and at the end of the season, and 2) canopy area measurement at the beginning and at the end of the season using a digital single-lens reflex camera (Nikon 300s with Zoom lens Nikkor 18-105 mm f/3.5-5.6G, Nikon, Japan) and specific software (Photoshop CS5, Adobe Systems, USA; ImageJ, NIH, USA) following the methodology proposed in Lordan et al., 2013. Soil gases emission (CO2, CH4 & N2O) was measured by using the GC method (Holland et al., 1999). Measurements were carried out in 2 trees per plot. A cylinder of polypropylene, 0.25 m in diameter and 0.1 m in height, was placed on ridge surface, placed 0.2 m aside the tree trunk and carefully driven 0.05 m into the soil one week before the measurement. Air samples were collected at 0, 30 and 60 minutes after chamber insertion with a syringe through the stopper. Gas analysis was performed by on-line gas chromatography using a 7890A GC system (Agilent Technologies, USA), equipped with a thermal conductivity detector and an electron capture detector. Peak areas were integrated by means of a computer running GC-assist software (Agilent Technologies, USA). GHG emissions are expressed as gas fluxes as mass m^{-2} d^{-1} (e.g. g CO_2 m^{-2} d^{-1}).

Results and Discussion

Trees under DI-70 treatment achieved the lowest growth results suggesting that N removal was lower than other treatments (Table 1). Regarding growth parameters, there were no differences between the other three treatments even at a low fertigation dose when applied by a subsurface system (SDI-70). Thus, it can be stated that it was possible to reduce water and nutrition dose when it was applied under a subsurface drip irrigation system since tree growth was not affected by its reduction. Concerning soil gas emissions, there were differences among treatments (Table 1). The results show that the highest CO_2 emissions were under the DI-70 treatment. This treatment actually registered the lowest growth parameters suggesting that it should have a low root activity and thus a low CO_2 emission (Kuzyakov and Larionova, 2006), however it may appear just the opposite. This phenomenon might be explained by the presence of active roots close to the soil surface, due to a limited wetted area (30% irrigation reduction). Root activity nearby soil surface might triggered soil root respiration as show provided results. Moving forward, results provide evidence that N_2O emission was lower for subsurface drip irrigation (SDI) treatments, regardless irrigation dose (Figure 1). This fact suggests that N_2O losses are reduced when fertigation is applied through a subsurface system. In addition, the CO_2:N_2O ratios for SDI treatments are significantly higher as compared to other treatments, suggesting that SDI was by far a much more effective technique since it was possible to reduce N_2O emissions while maintaining the crop growth and root activity. Actually, other studies (Lordan *et al.,* 2013) have shown higher crop performance when SDI systems where applied under soil limiting conditions.

Table 1. Effects of irrigation and fertigation treatments on trunk diameter increase (ΔTD), canopy area increase (ΔCA) and GHG emissions. Values followed by different letters indicate significant differences according to Tukey HSD test (P <0.05).

Concept	Units	DI-100	DI-70	SDI-100	SDI-70	Prob>F (model)
ΔTD	mm	34.08 a	24.82 b	34.79 a	35.58 a	< 0.0001
ΔCA	m^2	2.28 a	1.48 b	2.62 a	2.38 a	0.0005
CO_2	g m^{-2} d^{-1}	9.88 ab	14.44 a	5.40 b	8.61 ab	0.0249
N_2O	mg m^{-2} d^{-1}	45.53 a	38.26 a	15.08 b	10.24 b	0.0003
CO_2:N_2O	-	216.00 b	377.42 b	358.09 b	840.82 a	0.0252

Conclusions

Subsurface drip irrigation (SDI) reduces significantly N_2O emission regardless the fertigation dose. Both the growth parameters and the CO_2:N_2O ratio were high under this irrigation technique, suggesting a good crop performance and a high nutrient uptake efficiency under a SDI system.

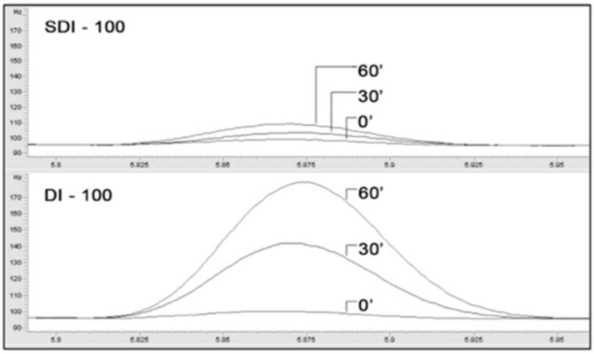

Figure 1. N_2O peak areas detected by the electron capture detector for SDI-100 (upper) and DI-100 (lower) treatments at 0, 30 and 60 minutes.

Acknowledgements

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TEMPORAL AND SPATIAL VARIATION OF N2O FLUXES FROM MANAGED PRE-ALPINE GRASSLAND

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Long term observations are indispensable to improve our knowledge of the complex biosphere-hydrosphere-atmosphere interactions and to detect and analyze impacts of climate change as well as to develop, improve and validate process model systems. Embedded into the German Helmholtz Society funded infrastructure project TERENO (Terrestrial Environmental Observatories) KIT IMK-IFU tnstalled 3 lysimeters with undisturbed intact grassland soil cores (area 1m^2, depth 1.5 m, 2-3 t of soil) in Garmisch-Partenkirchen in South-Bavaria, Germany, to monitor long term greenhouse gas emissions. An intensive field campaign was conducted at Garmisch-Partenkirchen station addressing the spatial variability in N_2O fluxes and elucidating the effect of snow cover on N_2O emission. The objectives of this study are characterizing the long term N_2O flux pattern, quantifying the annual green-house gases budgets, and investigating the spatial variability and magnitude of N_2O emissions with and without snow cover.

Materials and Method

The Garmisch-Partenkirchen station is situated at 734 m a.s.l. (47° 28' 32.12' N 11° 3' 44.47' E), with a mean annual temperature of 6.8 °C and 1371 mm of precipitation. N_2O fluxes were measured manually by static chamber method. For long term measurements lasting from November 2011 to November 2013, the entire top area of the lysimeters was closed with dark chambers for duration of measurement. In total over 100 single fluxes were obtained during the entire 2 year period. The intensive field campaign was carried out from 27.02.2013 to 04.03.2013, during days in which temperature was low enough to get the soil area not covered by snow frozen but high enough to have the same soil area unfrozen around midday. During the intensive field campaign, the snow cover was partly artificially removed inside the plot, resulting in 25 spots without snow cover and 5 spots with snow cover. N_2O fluxes were measured by using of dark chambers. In total 6 repeated measurements were performed to capture spatial variability in N_2O fluxes as well as elucidating the effect of frost-thaw event on magnitude of N_2O fluxes. All gas samples collected during long term measurements as well as intensive field campaign were measured by GC in the lab. Flux was calculated from the increase/decrease in the chamber GHGs concentration over time, and corrected by air pressure and temperature (Wolf et al., 2010). Soil water content and soil temperature were measured by TDR and SIS probes in 10 cm soil depth, respectively.

Results and discussion

From November 2011 to November 2013, N_2O fluxes ranged from -9.61 to 80.43 μg N m^{-2} h^{-1} with a mean value of 8.93 μg N m^{-2} h^{-1} (Fig. 1). There was no clear seasonal or diurnal pattern found. Even though correlation analysis proved significant positive relationship between N_2O flux and environmental factors such as soil temperature and soil moisture, the coefficient of determination was low (Fig. 2, Fig. 3). On the other hand, long term measurement revealed peak emissions directly after fertilizer application events and during frost-thaw events (Fig. 1). Long term measurements indicate that the source strength of these pre-alpine grassland ecosystems for atmospheric N_2O is dominated by short term events like precipitation, frost-thaw events as well as fertilizer application rather than by soil physical parameters (Imer et al., 2013). The N_2O emissions from the 5 replications with snow cover ranged between 0.0 and 2.3 µg N m^{-2} $h^{-1}$with a CV of 183%, while those from the 25 replications without snow cover varied between 3.0 and 44.1 µg N h^{-1} m^{-2} with a CV of 243% (Fig. 4). Mean N_2O flux rate for soil area with snow cover was with 1.26 µg N m^{-2} h^{-1} significantly lower than N_2O flux for soil area with no snow cover which was on average 16.91 µg N m^{-2} h^{-1}. Significant difference between both treatments proved the strong positive response of N_2O emission to frost-thaw events. The extremely higher N_2O fluxes from the intensive campaign indicated that there were several hot spot in the plot, most likely due to the high small scale heterogeneity in distribution of soil nutrients formed during the repeated frost-thaw conditions.

Fig 1

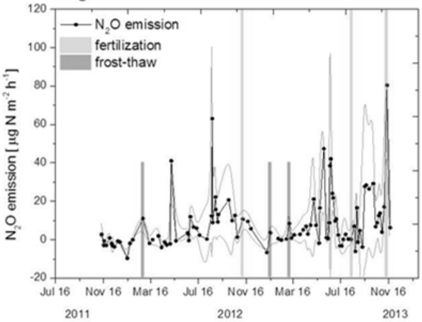

Fig 2

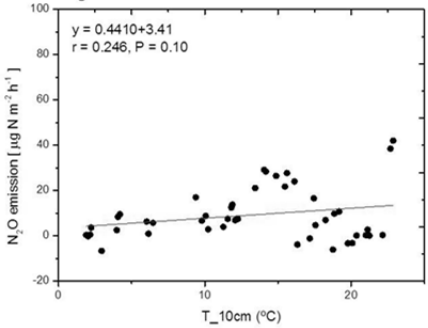

Fig 3

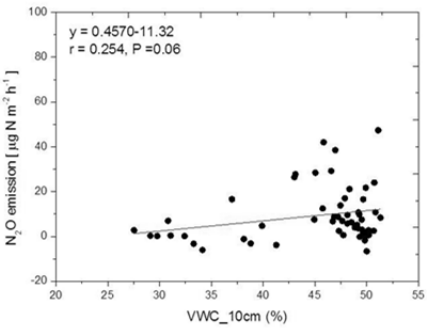

Fig 4

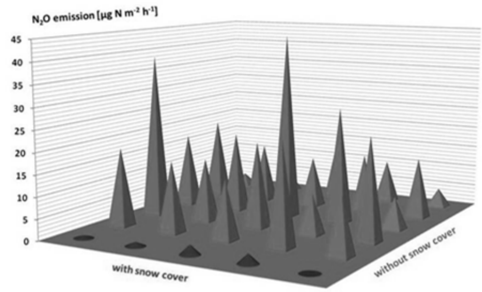

Fig. 1: Long term dynamics (Nov. 2011-Nov. 2013) of N_2O fluxes

Fig. 2: Relationship between N_2O emissions and soil temperature in 10 cm soil depth

Fig. 3: Relationship between N_2O emissions and volumetric water content in 10 cm soil depth

Fig. 4: Spatial variability of N_2O emissions during intensive field campaign

Conclusions

Managed grassland ecosystems in the pre-alpine region in Germany are mainly a small source for atmospheric N_2O (0.63 kg N ha^{-1} yr^{-1}), and management practice like e.g. fertilizer application increases the source strength of these sensitive ecosystems significantly. Not revealing an annual pattern, N_2O emission showed an event based pattern, and frost-thaw driven N_2O emissions contributed significantly to the annual budget. Spatial as well as temporal variation of N_2O emission was huge. To provide a reliable estimate of the annual N_2O budget, and to develop future mitigation strategies it is indispensable on the one hand to perform long term monitoring but on the other hand to capture single event driven peak emissions and to be able to correct for potential biases.

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EFFECT OF IRRIGATION AND FERTILIZER ON N_2O EMISSIONS FROM PADDY SOIL DURING THE SEEDLING STAGE IN NE SPAIN

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Agricultural soils are the most important source of N2O, accounting for 24% of total global emissions (IPCC 2007). The production of N2O in soils occurs as a consequence of nitrification and denitrification and it is affected by fertilization (Zou et al. 2009). Agricultural practices influence N2O emissions from rice paddies. Though, many measurements and models have focused on N2O emissions from rice paddies (Toshiaki et al. 2007; Zou et al. 2009; Sampanpanish 2012), to our knowledge, none has been done in Spain. The rice seedling stage (the first 35 days after sowing) significantly contributes to N2O emissions from rice paddies (Sampanpanish 2012). Sowing and cropping technique in Spain is different than in China where rice seedlings are cultivated in a separate paddy nursery under intensive management for approximately 1 month and transplanted to a paddy field while in Spain rice is sown directly on site and flooded. Total N2O emissions from rice seedling stage depend on: type of irrigation and fertilizer, soil texture, plant density and local weather (Zou et al. 2009). The main objectives of this study are to evaluate the effect of different rates of mineral and organic fertilizer on N2O-N and (N2O+N2)-N emission during the seedling stage.

Materials and methods

Field trials were carried out in 2012 on 2 experimental fields at 2 different sites. Treatments were (Table 1): (I) 2 doses of chicken manure (15.2, and 28.7 t ha^{-1}), urea (150 kg N ha^{-1}) and control at Amposta; (II) 2 doses of pig slurry (30 and 50 t ha^{-1}), ammonium sulphate (120 kg N ha^{-1}) and control at Alcolea de Cinca. Rice (*Oryza sativa* L.*)*, cv. *Gleva* at Amposta (150 kg seed ha^{-1}) and cv. *Guadiamar* at Alcolea (170 kg seed ha^{-1}) was sown directly on site. The fluxes of (N_2O+N_2)-N and N_2O-N were sampled weekly from sowing to harvest using the semi-static closed chambers method and the acetylene inhibition method. Samples of air inside the chambers were withdrawn in duplicate by adapted plastic 100 ml syringes. N_2O was quantified using the photoacoustic technique (Innova Multi-gas Photoacoustic Monitor 1412). Soil temperature and soil Eh were recorded on site.

Results and discussion

At Amposta during seedling the fertilizer had a significant effect on N_2O-N and (N_2O+N_2)-N emissions ($p<0.005$; Table 1). The highest flux of (N_2O+N_2)-N was emitted from the plots with urea. Emission fluxes were the lowest from the control plots, followed by the plots with chicken manure (Table 1). The higher dose did not increase the (N_2O+N_2)-N emission factor (percentage of the N applied). When applying urea, the dominant process is denitrification which starts very rapidly after application (Table 1). While when applying chicken manure containing a high amount of organic-N (some 460 kg in 28.7 t c.m.) the processes taking place are mineralization in the first place, followed by nitrification and most probably denitrification (due to waterlogged conditions) though not complete (not to N_2; Table 1). Cayuela et al. (2010) explained that the higher total N_2O emission from mineral fertilizer than from chicken manure during its initial decomposition may be due to that part of the organic N applied with the animal manure is not rapidly mineralized and may be released as mineral N subsequently. Results show that (N_2O+N_2)-N emission is higher from mineral N fertilizer than from chicken manure when applied to soils with a relatively high o.m. content, such as paddy soils. Usually, flooded rice soils have a high o.m. content and high denitrification potentials. Egginton and Smith (1986) and Velthof et al. (1997) found that the emissions from NO^-_3 fertilizer were much higher than from animal manure in paddy soils. At Alcolea, fertilizer did not affect the cumulative (N_2O+N_2)-N seedling emissions (Table 1). The (N_2O+N_2)-N flux during the 1st day after fertilization was up to between 10 to 18 times higher for 50 t pig slurry ha^{-1} than for 30 t pig slurry ha^{-1}, ammonium sulphate and control likely due to enhanced denitrification of soil NO_3^- by the addition of easily degradable organic substrates. The measured (N_2O+N_2)-N fluxes from pig slurry (170 kg N ha^{-1}) decreased from day one, and were low in all the treatments. The difference between the total (N_2O+N_2)-N emission during the seedling stage from ammonium sulphate and pig slurry (120 kg N ha^{-1}) was not significant and emissions were higher from pig slurry (170 kg N ha^{-1}) (Table 1). The cumulative N_2O-N emission measured is much higher than that reported by Liu et al. (2012) in China (from 0.07 to 0.98 kg N ha^{-1} under continuous flooding and moist irrigation regimes). Though as percentage of the applied N it is similar (Table 1) to that reported by Liu et al. (2012) (0.20-0.57%).

Table 1 Average cumulative N_2O-N and (N_2O+N_2)-N emissions from paddy soil during the seedling stage per site and year, plus minus standard error. Percentage of the applied N and of the cumulative (N_2O+N_2)-N emitted during the whole rice growth that the cumulative losses of (N_2O+N_2)-N during the seedling stage represent. Different letters in column mean significant differences

Site and year	N applied (kg ha^{-1})	N_2O-N (kg ha^{-1})	(N_2O+N_2)-N (kg ha^{-1})	% that (N_2O+N_2)-N represents of the applied N	% that (N_2O+N_2)-N represents of the whole crop emission
Amposta	0 kg N ha^{-1}	0.15±0.15b	0.87±0.52b	-	4.40
	150 kg N ha^{-1} (urea)	0.81±0.64b	16.64±3.89a	11.00	53.12
	170 kg N-NH_4^+ ha^{-1} (15.2 t c.m)	3.71±0.52a	3.21±0.85b	1.80	9.50
	340 kg N-NH_4^+ ha^{-1} (28.7 t c.m.)	2.85±0.96ab	2.67±0.36b	0.27	5.78
Alcolea	0 kg N ha^{-1}	0.43±0.06b	1.01±0.23a	-	12.24
	120 kg $(NH_4)_2SO_4$-N ha^{-1}	0.52±0.14ab	0.89±0.40a	0.74	8.08
	120 kg N-total ha^{-1} (30 t p.s.)	0.25±0.45b	0.84±0.28a	0.70	9.57
	170 kg N-total ha^{-1} (50 t p.s.)	0.87±0.04b	1.91±0.88a	1.12	25.95

c.m.: chicken manure; p.s.: pig slurry

Conclusions

Urea significantly increases cumulative (N2O+N2)-N emissions. Pig slurry significantly increases (N2O+N2)-N emission, while N2O-N emission in all treatments is very low, close to zero. The rice seedling stage significantly contributes to the (N2O+N2)-N emissions from rice paddies.

Acknowledgements

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USING 1H NMR PROFILING TO IDENTIFY SOIL METABOLITES RELATED TO NITROGEN MINERALISATION

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Metabolic profiling using the technique of nuclear magnetic resonance (NMR) is an emerging technology for investigating different constituents of soils. To date the application of NMR in soil science has mainly focused on the molecular nature and characterization of soil organic matter (Olk, 2008). However, ^{1}H NMR has shown to be an effective tool for simultaneously profiling multiple metabolites of a biological sample, which effectively supplies information about the biochemical properties of a sample, and relate differences or changes of identified metabolites to particular functional properties. Nitrogen mineralisation is a process that is driven by the enzymatic reactions of heterotrophic bacteria and fungi, which obtain their nutrition from organic substances such as carbohydrates, amino sugars, organic acids and amino acids within the soil (Liebeke et al., 2009).The objective of this study was to investigate if ^{1}H NMR profiling can isolate nitrogenous and carbon containing metabolites linked with the process of N mineralisation in 35 different temperate mineral soils.

Materials and Methods

In 2010, 35 mineral soils were collected at a depth of 10cm from a range of grassland sites across the Island of Ireland. Samples were dried at 40^{o}C and sieved to 2mm. Soils were analysed for a range of physicochemical properties and by variety of chemical N indices as outlined by McDonald et al. (in review). The mineralizable N (MN) pool of each soil was determined using the standard 7 d anaerobic incubation method (Waring & Bremner, 1964).^{1}H NMR data was obtained by extracting each soil sample in 40% methonal:water solution (40:60 v/v) at a ratio of 1:2 (w/v), lyophilizing the extract and reconstituting the sample in 750µl of 0.1M phosphate buffer (pH 7.0), in deuterium oxide (D_2O), containing 1mM of internal standard solution of sodium trimethylsiyl-2, 2, 3, 3-tetradeuteropropionate (Graham et al., 2010). Within a 5mm diameter NMR tube, 600 µl of the supernatant D_2O solution was analysed and the ^{1}H spectra recorded on a 400 MHz Bruker AVANCE III spectrometer. Baseline correction, spectral binning and measurement of the integral bins were carried out manually using the ACDlabs NMR processor version 12. The NMR integral data was then normalised to total spectral intensity and together with soil properties and MN data was analysed by the stepwise regression and multivariate statistical techniques of PCA and discriminate analysis using Genstat version 11. Identified metabolites were quantified using the equation as outlined by Graham et al. (2010).

Results and Discussion

Both the discriminant analysis and PCA isolated identified metabolites (e.g. glucose and trimethylamine) which grouped the 35 soils into two classes; A (n=15) soils with low N mineralisation potential and B (n=10) soils with high N mineralisation potential. In addition five other metabolites: glutamic acid, citric acid, aspartic acid, serine and 4-aminohippuric acid were identified as influencing MN. Soils associated high levels of MN, mainly within group B, were strongly associated with high labile carbon sources. The carbohydrate metabolite glucose significantly ($p < 0.0001$) explained 68% of MN variability across the 35 soils (Figure 1) and was more associated with soils that contained high levels of N, e.g. MN and total N, such as group B. This was expected as this metabolite is a highly utilized substrate by heterotrophic microbes in the processes of C and N breakdown. Glutamic acid, 4-aminohippuric acid (Figure 1), serine and trimethylamine also had positive relationships with MN (R^2 = 0.27, 0.34, 0.18 and 0.17, $p < 0.05$, respectively) as these metabolites provide sources of C and/or N for microbial activity.

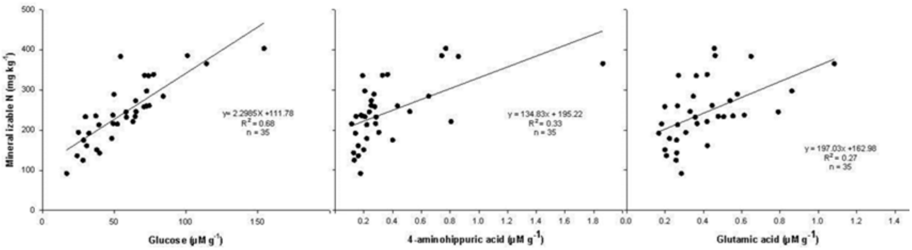

Figure 1. Identified ^{1}H NMR metabolites; glucose, 4-aminohippuric acid and glutamic acid versus mineralizable N (MN) for 35 soil types.

Conclusions

The availability of labile carbon (i.e. glucose) had the greatest positive influence of MN. The concentration of glucose in these grassland soils had the most significant influence on MN. Other soil metabolites that had significant relationships with MN were glutamic acid, 4-aminohippuric acid, trimethylamine and serine. ^{1}H NMR successfully identified metabolites in the soil related to N mineralisation and shows potential for increasing our knowledge of this important soil process. Further investigation of the ^{1}H NMR spectral profiles for these soils and results is being conducted and will be discussed in greater detail at the 18th Nitrogen Workshop.

Acknowledgements

Farmers who provided the soils for this work and technical support provided by the lab staff at Teagasc, Johnstown Castle, Queens University Belfast and Agri Food and Bioscience Institute Newforge Lane Belfast. Irish DAFM.

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COMPARISON OF AN AUTOMATED GAS SAMPLING SYSTEM (AGPS), WITH A STATIC CHAMBER METHOD FOR MEASUREMENT OF N2O EMISSIONS FROM A PERMANANT GRASSLAD SITE.

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The closed, static chamber method is the commonly adopted method to measure emissions of N_2O from soil. The static enclosure base is inserted into the soil to a depth > 5 cms. For measurements of fluxes, a lid is placed on top of the base for a specified time (40 mins), enclosing the atmosphere above the soil within the chamber. The accumulation of N_2O within the chamber is measured by manually taking a gas sample from the chamber, transferring to an evacuated glass vial and analysing by gas chromatography. Only one sample is taken per day and expressed as a daily flux. An automatic gas sampling system consists of an enclosure base fitted with a mechanical cover for automatic opening and closing. A fraction control unit fitted with a membrane pump and double needle assembly transfers headspace gas to 20ml evacuated vials. A programmable logic controller (PLC) interface is used to manage sample collection intervals allowing for multiple daily sampling events. The aim of this investigation was to compare results from the static chamber technique with an automatic greenhouse gas sampling system (AGPS) installed in a grassland field trial, measuring N_2O emissions from cattle slurry.

Materials and Methods

The experiment was conducted on a permanent grassland site at AFBI, Hillsborough, Northern Ireland. Cattle slurry was surface broadcast applied on 10thApril 2013 at a rate of 30m^3 ha^{-1}. The AGPS was set to sample on four occasions over a 24 hour period (04:00, 10:00, 16:00 and 22:00) for 21 days post application. Sampling timings were then reduced to two times per day (10:00 & 22:00) for the remainder of the experiment. At each sampling event, the AGPS was programmed to extract 3 gas samples at time zero, after 20 mins and after 40 mins (T0, T20 and T40). Fifteen static chamber measurements were taken between 10:00 and 12:00 on 32 scheduled sampling dates during 2013. The static chamber measurements were timetabled, initially for 10 occasions in the first two weeks post application, 4 occasions from weeks 2-4 and further reduced to bi-weekly or monthly for the remainder of the experimental period. The daily fluxes from the 15 chambers were averaged.

Results and Discussion

N_2O emissions for the first 12 days post application showed a close relationship between fluxes derived from the static chambers and autochamber. Heavy rainfall on the 17th and 18th (April 2013) increased the soil moisture to its highest level (73.6%) producing a subsequent offset in alignment between the autochamber and static chambers, which persisted for approximately 3 weeks. During this period, excess water was observed in some of the static chambers but not in the autochamber. This may have created conditions that were more conducive to N_2O emissions. It is possible that the excess water formed within the static chambers occurred as a result of impeded drainage which could be attributed to intensive sampling activity around the chamber perimeters during the early stages of the experiment. N_2O peaks continued to be observed during the remainder of the experiment after heavy rainfall events. High emissions observed in November 2013 were most likely due to soil N mineralisation and heavy rainfall.

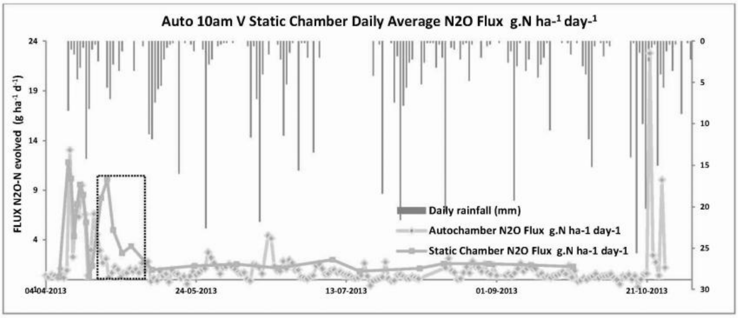

Figure 1. Comparison of N_2O fluxes from autochamber (at 10am) and static chambers (daily average N_2O flux g N ha-^{1} day-^{1}).

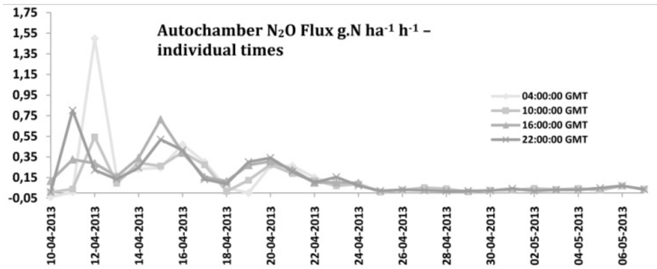

Figure 2. Autochamber N_2O Fluxes at four daily time intervals.

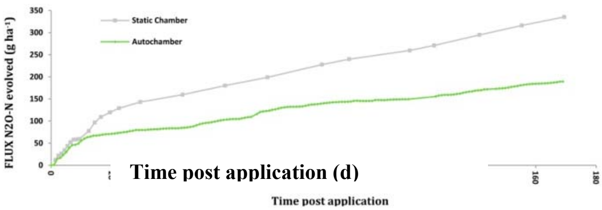

Figure 3. Cumulative N_2O fluxes (g ha^{-1}). Comparison between autochamber and static chambers.

Preliminary Conclusions

The diurnal N_2O fluxes were not significantly different, verifying that the adopted chamber sampling time of 10:00 to 12:00 is a good representation of the flux on any one day (Fig. 2). Comparison of the cumulative daily N_2O fluxes throughout the experimental period suggest, that the static chambers may be overestimating total evolved N_2O-N, under wet conditions (Fig 3).

Acknowledgement

The authors are grateful to the UK Department for Environment, Food and Rural Affairs (DEFRA) and the Devolved Administrations for financial support.

SUBSTRATE INDUCED DENITRIFICATION OVER OR UNDER ESTIMATES SHIFTS IN SOIL DI-NITROGEN/NITROUS OXIDE RATIOS

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Globally soils are the largest anthropogenic source of the greenhouse gas nitrous oxide (N_2O), and the increases in emissions present serious concerns for future global climate and environmental changes. Denitrification is a microbial process which sequentially reduces nitrates (NO_3^-) to dinitrogen (N_2) via nitrite (NO_2^-), nitric oxide (NO) and N_2O utilizing organic carbon (C) as the electron donor; a form of respiration in the absence of oxygen. Understanding the regulation of denitrification and the role of organic C in the production of N_2O and its reduction to N_2 is of global importance and offers the opportunity for managing soils to lower net emission of N_2O. Product stoichiometry of N_2 and N_2O is an important parameter for denitrification as it controls whether N_2O or the benign N_2 is released to the atmosphere. Increasing the N_2/N_2O ratio essentially mitigates the harmful effects of N_2O. The form and quantity of C utilised by soil denitrifiers can control the rate or efficiency of the N_2O reductase enzyme altering product ratios of the emitted gases. The regulation of denitrification by C is poorly understood compared to other soil parameters such as pH, N availability, redox status and water content. Many studies have investigated N_2O emissions and/or denitrification in the field and in laboratory experiments. A common practice in these investigations is the measurement of process rates (denitrification potential, denitrification enzyme activity and N_2/N_2O emissions) with the application of an exogenous organic C substrate to stimulate heterotrophic denitrification, often referred to as “substrate induced”. The C substrate of choice appears to be glucose, and current understanding of the regulation of N_2O reduction by C is principally based on glucose amendment to soil. Yet, the complexities of low molecular C compounds in soils, especially within the rhizosphere, that are available to the microbial population go beyond the model compound glucose. In this study we assessed how addition of individual organic C substrates (glucose, sucrose, acetate, malic acid, butyrate, succinate and cysteine) and no external C, here termed SOM-C, affected soil N_2 and N_2O production kinetics, using ^{15}N-labelled NO_3^- tracer techniques for quantification of denitrification stoichiometries. Specifically, we were interested in the extent to which different substrates deviated in denitrification stoichiometries from the model glucose.

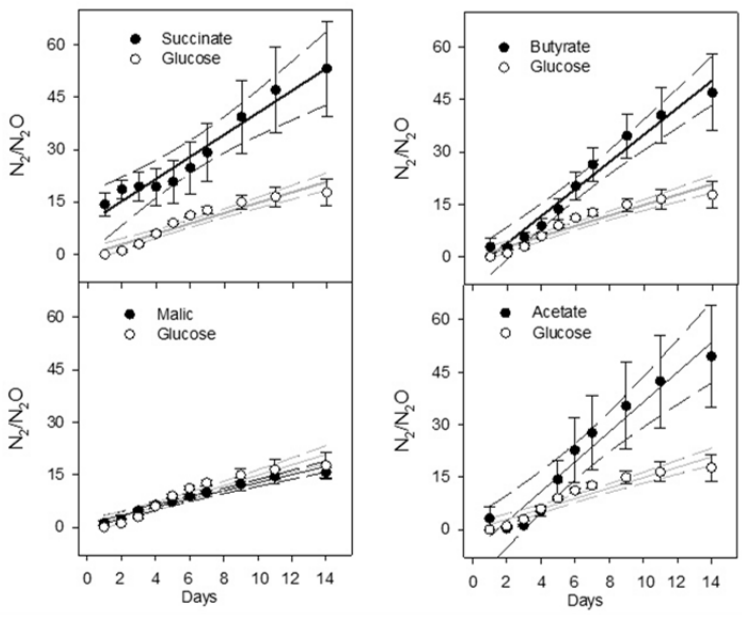
Materials and Methods

Soil was re-packed into PVC cores (5 cm diameter) to a 10 cm depth, re-wetted with water to 60% water filled pore space and allowed to stabilise for 7 days. Organic carbon substrates (glucose, sucrose, acetate, malic acid, butyrate, succinate and cysteine) or water were continuously supplied to soils along the whole vertical profile by means of a peristaltic pump and Micro-Rhizon tubes inserted into the centre of each core. ^{15}N-labelled NO_3^- was applied to each core so that soil denitrifier products could be quantified. This was achieved by closing each core and sampling soil headspace gas, which was analysed by continuous flow isotope ratio mass spectrometry for $^{15}N_2O$

and $^{15}N_2$. The experiment lasted for 14 days and each treatment was replicated 4 times. Additional cores were set up for destructive soil sampling for the measurement of soil nitrate concentrations.

Results and Discussion

To make comparisons in the efficiency of denitrification in terms of N_2O reduction to N_2, we examined N_2/N_2O ratios over time (example shown in figure), and applied linear regression models to the data. We were specifically interested in ratio differences between substrates when compared to glucose in a pair wise manner, as glucose is the most commonly applied compound for determining substrate induced soil processes. To show differences between treatments we used ANCOVA (C treatment as main effect and time as a covariate) to compare the slope coefficients from linear regression models. The result from the ANCOVA revealed that N_2/N_2O slope coefficients were not homogenous between the different treatments (assessed by the interaction term C-treatment*Time; F=41.5, df =7 $P< 0.01$), implying that the efficiencies of denitrification are not constant between the different organic substrate amendments. Following this we pair wise compared regression slopes to the glucose treatment using a Tukey-Kramer multi comparison procedure. The results revealed that acetate, succinate, butyrate and the SOM-C N_2/N_2O slope coefficients were significantly different from glucose at $P< 0.01$, and the cysteine treatment was different from glucose at $P< 0.05$. The regression slopes from sucrose and malic acid were not significantly different from that of glucose. These results suggest that soil microcosm studies, in which glucose has been applied to measure denitrifier stoichiometry observed an artefact of the substrate amendment, shedding little actual light on the the regulation of the N_2O reductase. Glucose may not be the most appropriate substrate for understanding the regulation of N_2O reductase and the regulation of N_2O reduction to N_2 is not possible to predict from glucose or sugar group of C compounds acting as the electron donor



ESTIMATES OF GREENHOUSE GASES EMISSIONS FROM THE USE OF SEWAGE SLUDGE AS FERTILIZER IN AGRICULTURAL SYSTEMS

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Agriculture accounted for an estimated emission of 10-12% of total global anthropogenic emissions of greenhouse gases (GHGs) (Smith et al, 2007). The use of sewage sludge must increase pasture production to be a profitable option from the farmers' point of view; and on the other hand, it should be contribute to mitigation climate change. This study aims to estimate carbon sequestration from the pasture production obtained with the use of different sewage sludge (anaerobically, pelletized and composted sewage sludge) in agricultural systems established in NW Spain.

Materials and Methods

The experiment was conducted in Pol (Lugo-NW Spain) in 2003. The experimental design was a randomized complete block with three replicates and four treatments distributed in experimental units (plot) of 8 m^2: *i)* fertilisation with anaerobically digested sludge with an input of 160 kg total N ha^{-1} (AN); ii) fertilisation with pelletized sewage sludge (PE) (160 kg total N ha^{-1}); iii) fertilization with composted sewage sludge (CO) (160 kg total N ha^{-1}); iv) and no fertilization (NF). The calculation of the required amounts of sludge was conducted according to the percentage of the total nitrogen and dry-matter contents (EPA, 1994), taking into account the European Union Directive 86/278/CEE (DOCE, 1986) and Spanish regulation RD 1310/1990 (BOE, 1990) regarding heavy metal concentrations for the application of sewage sludge on soil. Pasture production from each plot was harvest in June and December 2003. During each harvest, the ground surface area was cleared, with the pasture weighed in situ and a representative subsample taken to the laboratory, dried (48 h- 60ºC) and weighed to estimate annual pasture production. With the goal of quantifying the potential GHG emission from animals, we determined an average annual pasture carrying capacity (CG) that the system could support based on annual pasture production in each treatment (Steinfeld et al., 2006). The CH_4 emissions (enteric fermentation and manure management emissions) and N_2O emissions (stabling period) resulting from sheep livestock management were estimated (Figure 1). On the other hand, the direct N_2O soil emissions (stabling period and grazing period) and indirect N_2O soil emissions (atmospheric deposition and leaching) were also calculated (IPCC 2006). The method used to estimate livestock and soil emissions is described by the IPCC (2006). Finally, the equivalents in terms of CO_2 were estimated (Fig 1). The data were analysed using ANOVA and the differences between the averages were determined using the LSD test ($\alpha = 0.05$) using the statistical software package SAS (SAS, 2001).

Results and Discussion

The European Union promotes the use of sewage sludge as a fertilizer because of its specific organic matter and content of macronutrients, particularly nitrogen (MMA, 2006). The results showed that AN significantly increased pasture production and CG compared with the other treatments applied (PE, CO and NF) (Fig 1). As a result, GHG emissions from livestock and soil were also significantly increased by AN treatment. However, taking into account the carbon sequester (pasture aboveground + belowground) and the total emissions (livestock + soil), the results showed that AN treatment significantly increased the carbon sequestration compared with the other treatments (PE, CO and NF) (Fig.1).

Conclusions

The use of anaerobically sewage sludge, previously seen as a burden to society, can be viewed as a value resource to maximise the systems profitability (stocking rate) and carbon sequestration.

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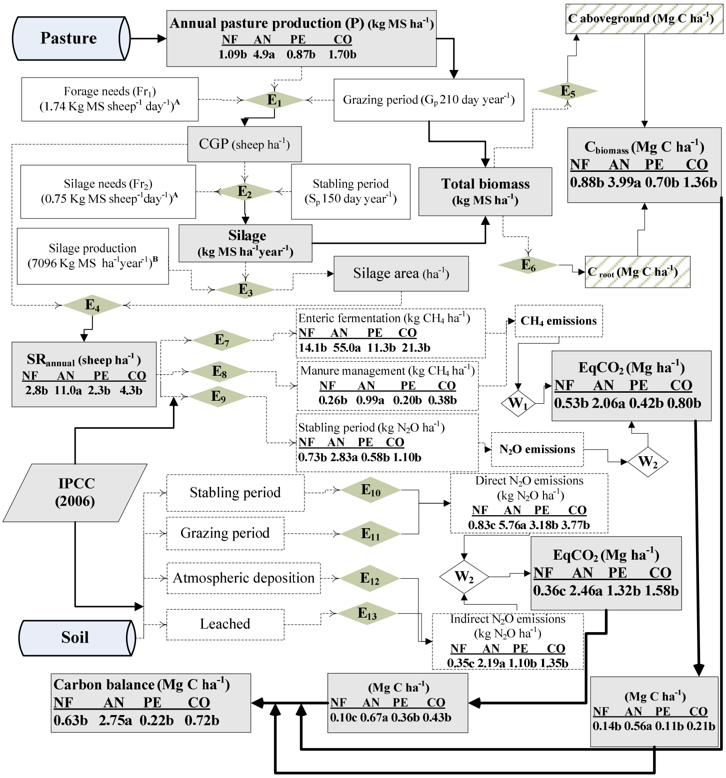

SR: stocking rate (sheep ha^{-1})
$E_1 = P/(Fr_1 \times Gp)$; $E_2 = Fr_2 \times SR_P \times S_p$; E_3 = silage needs / silage production ha^{-1}; $E_4 = SR_P$ / (1ha + Silage area)
$E_5 = Biomass \times M_1 \times N_1$; $E_6 = Biomass \times M_2 \times N_2$; $E_7 = SR_{annual} \times F_1$; $E_8 = SR_{annual} \times F_2 \times Sp$; $E_9 = SR_{annual} \times n_{ex} \times S_p \times F_3 \times 44/28$
$E_{10} = SR_{annual} \times n_{ex} \times [1-(Frac_{past} + Frac_{GASM})] \times FE_1 \times 44/28$; $E_{11} = SR_{annual} \times n_{ex} \times 0.02 \times 44/28$
$E_{12} = ((N_{FER} \times Frac_{GASFS}) + (SR_{annual} \times n_{ex} \times Frac_{GASM})) \times FE_2 \times 44/28$; $E_{13} = ((N_{FER} + SR_{annual} \times n_{ex})) \times Frac_{Lix} \times FE_3) \times 44/28$
F_1: 5 kg CH_4 $sheep^{-1}$ $year^{-1}$ F_2: 0.22 kg CH_4 $sheep^{-1}$ $year^{-1}$ F_3: stabling factor emission (0.02 kg N_2O-N/kg N)
nex: N_{manure} (20 kg $sheep^{-1}$ $year^{-1}$); W_1: warming potential of CH_4 (21); W_2: warming potential of N_2O (298);
$Frac_{PAST}$ N_{ex} during grazing (0.02 kg N/kg N_{ex}); $Frac_{GASM}$: Nex that is volatilized (0.02 kg NH_3-N + NO_x/kg Nex)
FE_1: emission factor (0.0125 kg N_2O-N/kg N input); N_{FER}: N applied in fertilisation;
FE_2: emission factor (0.01 kg N_2O-N per kg NH_3-N and NOx-N);
FE_3: emission factor (0.025 kg N_2O-N per kg N leaching of runoff);
M_1: 90.36 % (organic matter found in the Galicia pasture (A: Flores et al. 1992); N1: 50% (pasture C content (Montero et al 2005)
M_2: Root/aboveground biomass 32.37%; N_2: root C content 49.67% (Gordon et al 2005).

Figure 1. Methodology used to estimate carbon balance (Mg C ha^{-1}) and results obtained in the different systems studied. NF: no fertilization, AN: anaerobically digested sludge, PE: pelletized sewage sludge y CO: composted sewage sludge. Different letters indicate significant differences between treatments ($p < 0.05$).

N2O EMISSIONS FROM CROPPING SYSTEMS WITH INTEGRATED WEED MANAGEMENT

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Integrated weed management (IWM) in cropping systems aims to lower the reliance on herbicides of the crops, by introducing new combinations of agricultural practices in the system development (Munier-Jolain et al, 2009). These combinations may greatly change from a system to another and include a large variety of practices, such as false seed beds, late sowing, mechanical weeding, reduced tillage, specific crop rotations that alternate spring and winter crops, the choice of crop varieties and the use of pesticides with low ecotoxic impacts. Several implemented agricultural practices are likely to alter soil biogeochemical cycles and different components of the greenhouse gas budget (balance between the carbon sequestration and the greenhouse gas emission) of the system: e.g. crop rotation, dates and level of nitrogen fertilization and tillage. The main objectives of our study were to evaluate i) the N2O fluxes emitted from soil during one year for 4 cropping systems (i.e. 3 IWM systems and a local reference of conventional system), and ii) to investigate the relationship between the measured fluxes and soil parameters and the agricultural practices of each system.

Materials and methods

One reference cropping system (S1) and 3 IWM cropping systems (S2, S3 and S5) were studied (Table 1). Nitrous oxide (N2O) emissions were measured continuously using the automated chamber method (Vermue et al., 2013) from March 2012 to March 2013. Soil temperature and moisture were continuously recorded for the different systems and soil bulk density and inorganic N periodically measured.

Results and discussion

The intensity of the N2O emissions was highly variable, both over time and between systems. N2O emissions and water filled pore space (WFPS) were significantly correlated for all systems but no significant correlation could be established between N2O emissions and soil inorganic N dynamics. Over the year of experimentation, the IWM system S2 emitted significantly more N2O (5226 ± 670 g N-N2O ha-1) than the IWM system S5 (777 ± 177 g N-N2O ha-1), which also emitted significantly more N2O than the IWM system S3 (177 ± 172 g N-N2O ha-1) and the reference system S1 (326 ± 168 g N-N2O ha-1). In no-till system S2, WFPS values were significantly higher, and may have greatly enhanced the denitrification activity and the N2O fluxes emissions. The different crop rotations between systems may also have impacted N2O emissions, particularly the introduction of alfalfa, in the crop rotation of S5 system which may have enhanced N2O emissions.

Conclusion

Over the year, the continuous monitoring of the four cropping systems allowed to identify N2O fluxes as mostly resulting from short periods of favorable soil conditions for N2O production by denitrification in soils i.e. high WFPS and temperature. Overall the intensity of N2O fluxes significantly differed between systems, suggesting a strong impact of agricultural practices on N2O emissions. Some very high emissions, exceeding 5 kg N-N2O over the measurement period was observed on the S2 system characterized by the absence of tillage. Despite some limit of the experimental device, the results strongly suggest that a no-tilled integrated weed management system can promote N2O emissions in comparison to tilled systems either conventional or integrated. As an equivalent emission of 0.7 t C-CO2 ha-1 was observed to be emitted by the S2 plot, the probable carbon sequestration in this system has probably been canceled during the very rainy year of measurement. However, efficiency of no-till systems for mitigating global warming are known to increase with years, this study need to continue since the literature suggests that the observed effect of IWM on N2O emission could change overtime.

Table 1: Main characteristics of the 4 studied cropping systems (from Chikowo et al., 2009).

Crop system	Acronym	Description
Reference system	S1	Designed to maximize financial returns. Use of chemical herbicides to control weeds. Moldboard plowing each year. Choice of herbicides according to recommendations of extension services. Crop rotation: winter wheat/winter barley/oilseed rape.
IWM	S2	Minimum tillage between 2000 and 2007. No tillage since 2008. Time-consuming operations such as plowing, harrowing and mechanical weeding excluded. Treatment frequency index[1] reduced by 25 %. Diversified crop rotation[2].
IWM	S3	Plowing and other tillage operations allowed when necessary for weed seedbed management but mechanical weeding is excluded. Treatment frequency index reduced by 50 %. Diversified crop rotation[2] + leguminous.
IWM	S5	Use of any herbicides excluded. Physical and cultural means are allowed to contain weed infestation. Diversified crop rotation[2].

[1] amount of pesticides spread per ha expressed in % of the standard approved dosages of pesticides per ha.
[2] oilseed rape/winter cereal/spring crop/winter cereal/summer crop/winter cereal.

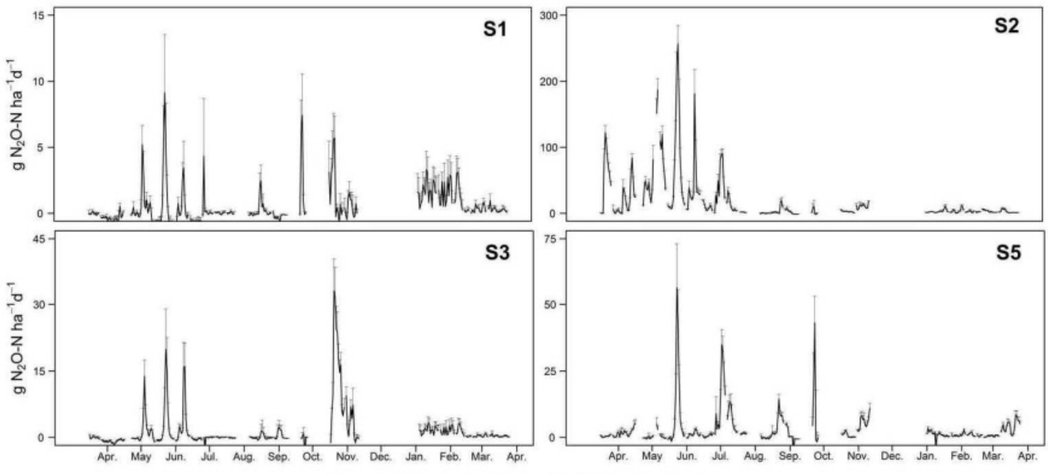

Figure 1: N_2O emissions for the 4 studied cropping systems.

Acknowledgments

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THE CONTRIBUTION OF CROP RESIDUES TO NITROUS OXIDE EMISSIONS FROM ARABLE CROPS

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The current UK Greenhouse gas (GHG) Inventory (2011) identifies crop residues as the third largest source of direct nitrous oxide (N_2O) emissions from agriculture after fertiliser and grazing returns. There is particular interest in the contribution of leguminous crop residues to N2O emissions given their importance in agriculture generally and low input farming in particular. Legumes can provide an important input of nitrogen to agricultural systems, with biological nitrogen fixation adding as much as 400 kg N/ha/y. The IPCC has recently reduced the emission factor associated with nitrogen fixation from 1% to 0% reflecting evidence that the fixation process itself contributes relatively little to N_2O emissions (Rochette and Janzen, 2005). However, the crop residues left by leguminous crops often contain high concentrations of nitrogen and their decomposition in the soil can lead to N_2O release (Baggs et al., 2002). It is assumed by the IPCC that an emission factor (EF1) of 1% can be applied (IPCC, 2006), but there are relatively few studies of emissions from this source, and large uncertainties remain. The objectives, of this study were therefore to undertake a rigorous assessment of emissions of N_2O from crop residues in the UK, to relate emissions to the amount of nitrogen incorporated into the soil in the autumn, and to the C:N content of the residue.

Materials and Methods

Across 2 continuous experimental years, 3 experimental sites were chosen to represent important arable cropping regions of the UK: i) South East England (clay loam, mean annual rainfall of 600 mm), ii) Central England (loamy sand, mean annual rainfall of 600 mm) and iii) East Central Scotland (sandy loam, mean average annual rainfall of 870 mm). All sites had a low mineral N status (generally < 80 kg/ha N to 90 cm depth) and followed a cereal crop. There was no history of long term organic manure applications and no manure applications in the previous 2 years. Three replicated plots (12 m x 12 m) per treatment were established in a randomised block design. Four pulses (winter beans, spring beans, combinable peas & vining peas) and a cereal (winter wheat (with no nitrogen applied) as the control) were sown (Table 1). Following harvest, plots were split, one half of the plot had the above ground crop residue incorporated and the other half had the above ground crop residue removed. Winter wheat was sown on all plots after harvest. No nitrogen fertiliser was applied to any plot both years. Nitrous oxide emissions (5 static chambers/ plot) were monitored from sowing of the winter legumes until the sowing of the wheat (12 months) and from sowing of the wheat for 12 months following the incorporation/removal of the crop residue (24 months in total) giving a total of 40-50 individual measurement events. On each sampling date soil mineral nitrogen (SMN) and gravimetric moisture were measured (0-10 cm) on every plot.

Results and Discussion

The results of this research programme are currently being analysed; these will be presented using different approaches to emission calculations (for example variations in the period over which emissions are integrated) and testing how differences in site and climate influenced absolute emissions and emission factors. This work will provide valuable new information on the importance of crop residues in contributing to N_2O emissions and on opportunities for greenhouse gas mitigation in the arable sector.

Table 1: Sowing dates for the crops at each of the 3 experimental sites

	South East England	Central England	East Central Scotland
Winter beans	28/10/2010	20/10/2011	30/09/2010
Spring beans	08/03/2011	20/03/2012	07/03/2011
Peas, combinable	08/03/2011	20/03/2012	07/03/2011
Peas, vining	08/03/2011	20/03/2012	07/03/2011
Winter wheat nil N	21/09/2010	21/09/2012	28/09/2010
Follow on crop winter wheat	10/11/2011	18/11/2012	04/10/2011

Figure 1: The allocation of plots and chambers after the complete sowing of the crops in spring 2011 (central photo) including pulses such as peas and beans (top right photo) followed by residue incorporation (bottom right photo) or not in autumn 2011 and immediate sowing of winter wheat.

Acknowledgements

UK Department for Environment, Food and Rural Affairs and Scottish Government through Sustainable Arable LINK Project LK09128. ADAS, Agricultural Industries Confederation, Bayer CropScience, British Sugar, Country Land and Business Association, The Co-operative Group, Frontier Agriculture, GrowHow UK, AHDB-HGCA, Hill Court Farm Research, NFU, North Energy Associates, PGRO, Renewable Energy Association, Rothamsted Research (North Wyke), Scotch Whisky Research Institute, SoilEssentials, SRUC, Vivergo Fuels, Warburtons and Yara UK

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NITROUS OXIDE INTENSITIES OF LEGUME BASED AGRICULTURAL SYSTEMS IN EUROPE

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Nitrogen (N) availability remains one of the key drivers of crop productivity from arable farms in Europe. Increased profitability and environmental performance across the arable sector requires systems with improved nitrogen (N) use efficiency and resilience to climatic fluctuations. Most crops receive applications of synthetic N fertilisers but these can contribute to significant losses of the greenhouse gas (GHG) nitrous oxide (N_2O). Legumes can potentially offer an opportunity to reduce GHG emissions from the agricultural sector, partly due to the avoidance of emissions associated with fertiliser manufacture, and partly due to lower field emissions associated with legume production. It is argued that agricultural systems need to increase production whilst reducing environmental impacts (Godfray et al., 2010). Currently about 10% of the agricultural land area is used for letting production on a worldwide basis (Sprent, 2009). Production in Europe is decreased in recent decades and it is estimated that there is the potential an increase of 15% to 25% crop cover. However, only around 0.5% to 6.5% of land is sown with grain legumes (Nemecek, 2008). There have been few recent studies of N_2O emissions from legumes and the their effect on subsequent crops. The main objective of this study was therefore to explore the extent to which different legume based agricultural systems across Europe could influence N_2O emissions per unit of grain production (emission intensity: kg N_2O-N per kg product) in the year of growth and the following year (subsequent cereal crop).

Materials and Methods

A network of sites across Europe were selected that represented different climatic regions. These were the Mediterranean (Spain, Greece and Italy), Continental (Romania) and Atlantic (United Kingdom) regions. In Spain, chickpeas, fababeans and wheat were growing as monocrops in a conventional system; in Greece, phaseolus in organic and conventional systems with low or high salinity; in Italy, faba beans and peas as monocrops or intercrops with barley in 50:50 replacement; in Romania, peas,lentils, soybean, mugabeans and phaseolus as monocrops in an organic system and in the UK, peas and beans as monocrops in a conventional system (Figure 1). Greenhouse gas emissions and extractable N were measured every 15 days and at key growth stages, from sowing to harvesting of legumes followed by the subsequent cereal crop at all sites. Grain yields were also recorded for the calculation of N_2O intensities. Direct emissions of N_2O were measured using the static chambers which were sealed for 40-60 minutes with an aluminium lid. A small open sampling point was used to collect two samples using a syringe. Gas samples were transferred to a portable evacuated glass vial and analysed for N_2O, CO_2 and CH_4 by gas chromatography using high purity standards. For consistency, gas sampling was carried out between 10:00 and 12:00 hrs.

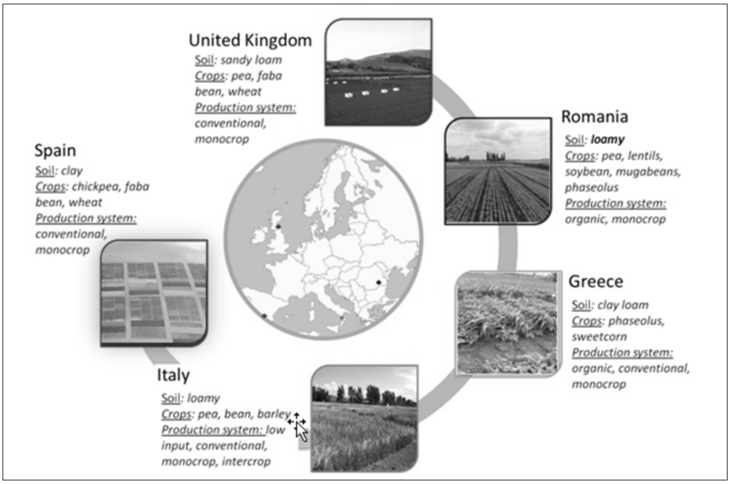

Figure 1: Crop and soil characteristics for the five sites during the experimental/growing periods.

Results

N_2O intensities in the year of legume production were as follows: the winter peas and soybeans (120 g N_2O-N t grain yield for both) were lower than other legumes and the unfertilised winter wheat (132 g N_2O-N t grain yield) in Romania. In Greece, Phaseolus plants growing after sweetcorn with under low salinity were significantly different from crops grown under high salinity, where the organic system treatments had the lowest emission intensity (75 g N_2O-N t grain yield). In Italy, the legume monocrops had higher intensities when compared with the cereal (barley) monocrop, but when the peas or beans were sown at 100:50 (legume:barley) (additive design), the intensities were reduced by 75% and 50% of the beans and peas monocrop, respectively. Finally in UK, the vining peas had the lowest N_2O intensity (26 g N_2O-N t grain yield) and all the legumes had lower intensities than the unfertilised winter wheat (298 g N_2O-N t grain yield) under the same growing conditions.

Conclusions

A peer review paper is under preparation at the moment including two years of each site and further results will be available for the revised abstract in February. We will illustrate how different approaches to emission calculations and differences in soil, site and climate influence emission intensities. This work will provide valuable new information on the importance of legume species/varietal combinations in agro-ecosystems and on developing management recommendations for optimising the productivity and environmental impacts of legumes in practice with similar climatic zones.

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LOWLAND SPRINGS: MODEL ESTIMATION OF NITROGEN LEACHING UNDER CURRENT AND FUTURE CLIMATE SCENARIOS IN A CASE STUDY IN NORTHERN ITALY

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The lowland springs in northern Italy are fed by waters which originate from an unconfined charged by the infiltrated precipitation and the irrigation water applied in the neighbourhood. The most prevalent agricultural land use is maize cropping and grassland, typically grown with high nitrogen (N) input. The protection of the groundwater quality is a goal of the European and Italian policies so that an accurate analysis has to be performed to estimate the actual risk of nitrate (NO_3-N) contamination from agricultural land use; moreover the lowlands spring represent a particular environment that needs to be preserve. The objective of this paper was to evaluate NO_3-N leaching under current and future climate scenarios of the two prevalent cropping systems in a lowland spring in Lombardy (northern Italy) by applying FOCUS PEARL 4.4.4 (Tiktak et al., 2000) and ARMOSA simulation model (Perego et al., 2013) to predict respectively the soil water and the N dynamics in the soil-plant system.

Materials and Methods

The case study was identified in two lowland springs in Lombardy of 1500 ha each: one is representative of the northern part of the belt that has constant groundwater recharge during the year (Isso, 45° 28' 07,01" N 9° 45' 25.50"E), the other is representative of the southern part that is characterised by a water flow dependent from land irrigation (Selva, 45° 25' 23,83" N 9° 51' 31.16"E). The conservative recharge area for the solutes has a radius of 2.5 km (Laini et al., 2012). The data of land use, organic N load, calculated on the basis of livestock density, soil characterization, which was extracted from the regional soil map at the scale 1:50'000, and meteorological condition were available from regional database of 2012. In particular, on the basis of the meteorological data observed by the weather station located in Capralba (45° 30' 35"N; 9° 34' 32"E) three dataset of 50 km grid were created: the first replicated the original dataset up to 66 years, while the other two (period of 2000-2100) were elaborated by SMHI from the SRS A1B greenhouse gas emission scenario, using the Ross by Centre Regional Atmospheric model RCA3.0 (RCA3 ECHAM5-r2 and r-3) and the CNRM regional scenarios. The r2 and r3 differed by the baseline period by respectively +2°C and +4°C from the average temperature and by -10% and -15% of annual rainfall. The 5-years rotations were: A (monoculture of FAO 600 maize) and B (grain maize-grass); crop parameters were previously calibrated and validated by Perego et al. (2013). Two type of irrigation with high and low efficiency were simulated, sprinkler(sp) and border (bo), and four events were set per year according the local practices. The both organic and mineral N-fertilization were simulated for maize cropping; the organic load (dairy slurry) was

300 kg N ha^{-1}, while the mineral N-fertilizer (100 kg ha^{-1}) was applied at side-dress, in agreement with the common practices. In the case of grass crop only the organic load spreading was simulated. Numerical simulations for predicting the soil water content, percolation and watertable fluctuation were performed using the model FOCUS PEARL 4.4.4. Once forced for the hydrological variables, the ARMOSA model was run to simulate crop development and growth and N dynamics. Both FOCUS PEARL and ARMOSA were applied in the 23 soil units which were identified from the regional map. The mean annual nitrate leaching was then calculated for the lowland springs of Isso and Selva as weighted mean on the basis of the % of UAA (Utilized Agricultural Area) of each soil type.

Results and Discussion

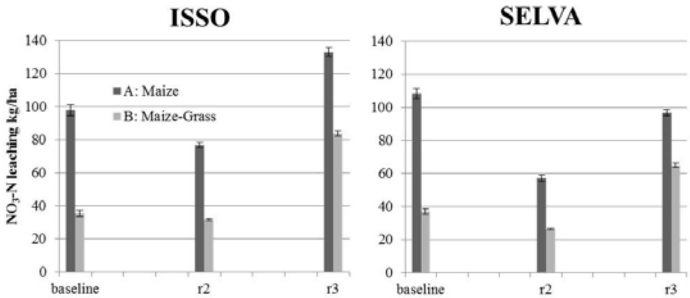

Figure 1. Mean annual leaching simulated under the three scenarios in the two lowland springs of Isso and Selva. The displayed data were calculated as weighted mean of the leaching simulated in the 23 soil units. SE ±

Mean N leaching amount were 70, 48 and 95 kg N $ha^{-1}y^{-1}$ under baseline, r2 and r3 scenarios. ANOVA test confirmed the statistically significance of scenario factor in affecting N leaching ($p< 0.01$). The A system led to higher leaching than B system ($p< 0.01$, +50%) due to the cycle length (Figure 1). Higher temperature under the 3 scenario involved higher rate of mineralization of the organic matter and therefore higher leaching. The interaction between A system and *bo* irrigation guaranteed a considerable recharge of the ground water body although involved more leaching ($p< 0.05$, +20% of percolation to groundwater). In comparison with the baseline scenario, the r2 involved higher crop biomass and N-offtake ($p< 0.05$) because of the water availability of lowland spring in addition to higher temperature (+2°C), whereas a relevant stress occurred under the r2 scenario, leading to decrease of yield up to the 35% ($p< 0.01$).

Conclusion

In the future it will beneficial to adopt a two-crops system, irrigated with high efficiency, to minimize the loss of NO_3-N to groundwater. As compromise, it will be fundamental to monitor the fluctuation of water table to evaluate whether it will require a water recharge. For the future, breeding and use of maize crop characterized by lower evapotranspiration could not affect the water table level.

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NITROUS OXIDE ANALYSIS BY LASER SPECTROSCOPY: NEW APPLICATIONS

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Recent developments in instrumentation for nitrous oxide (N_2O) analysis present new opportunities for investigating sources of N_2O. The N_2O Isotope Analyzer from Los Gatos Research (Mountain View, CA) enables analysis of N_2O concentrations and isotopic composition at ambient levels and after ^{15}N enrichment, and in both continuous mode and after batch injection. Nitrous oxide is an asymmetrical molecule ($N^{\beta}N^{\alpha}O$), and a $^{15}N/^{14}N$ isotope ratio, R, can be determined for each position. In delta notation, isotope ratios are defined as, e.g., $\delta^{15}N^{\alpha} = \{^{15}R^{\alpha}/^{15}R^{\alpha}_{ref} - 1\} \times 1000$. Pure culture studies have indicated that the site preference (SP) of ^{15}N, calculated as $\delta^{15}N^{\alpha}$ - $\delta^{15}N^{\beta}$, is different when N_2O is produced from NH_2OH during nitrification as compared to when N_2O is produced from NO_2^- via denitrification processes. If this distinction applies also at the community level, then possibly the contributions of nitrification and denitrification to N_2O in natural environments may be determined (Clough et al., 2013).

Among potential applications are analyses of gases with static or dynamic enclosures, and analyses of soil N_2O concentration profiles. Typically, the sample matrix will vary in composition, and the performance of the instrument therefore needs to be evaluated with respect to response and possible interferences from other gases or environmental variables. This presentation shows selected results from experiments to evaluate a N_2O Isotope Analyzer for use in N_2O emissions research. Results are also shown from laboratory incubation experiment.

Materials and methods

The instrument evaluated was a Model 914-0022 N_2O /CO Isotopic Analyzer based on off-axis integrated cavity output spectroscopy (OA-ICOS). The instrument has a port for injection of gas samples by syringe without (>80 mL) or with (>10 mL) dilution with "Zero air" containing < 1 ppb N_2O. Furthermore, a multi-port inlet unit (MIU) is used for continuous-mode applications, allowing intermittent analysis of calibration gases and ambient air. Reference gases used were synthetic air, and *c.* 500 and 2000 ppb N_2O calibration gases in a 20/80 mixture of O_2 and N_2.

In one test, sensitivity to relative humidity was tested by purging solutions of glycerol corresponding to 50 and 75% RH (Forney and Brandl, 1992). Concentrations of N_2O and H_2O, and $\delta^{15}N^{\alpha}$ and $\delta^{15}N^{b}$, were analyzed in ambient air and purged air across several cycles. A second test examined the detector response when the room temperature was lowered from 25 to 15°C. For soil gas sampling, any interference with O_2 concentration is critical, and this was examined by preparing N_2O reference gases in O_2/N_2 mixtures ranging from 0 to 20% oxygen. Storage of gas samples in Tedlar and aluminum foil gas sampling bags was compared.

An incubation experiment was set up with a sandy loam soil to examine the interaction between soil moisture (50, 65 and 80% final WFPS) and cattle slurry application (0, 1, 2 and 4 times recommended rate) with respect to short term N_2O emissions. In order to eliminate potential N limitation, the soil was amended, prior to slurry application, with 200 mg kg^{-1} N in NH_4NO_3 in which the NH_4^+ component contained 12 atom% ^{15}N. Then 100-cm^3 samples were incubated, and after 1, 2, 4 and

8 d samples were transferred to 1-L jars for 2 h. Using a glass syringe, 80 mL synthetic air was mixed with the headspace gas and an 80-mL sample withdrawn for analysis.

Results and Discussion

Both N_2O concentration and isotope ratios were robust over the range of RH examined. Also, variations in room temperature between 25 and 15°C changed N_2O concentration by < 1 ppb and had no effect on isotope ratios. Changing the O_2 dry mixing ratio from 20 to 5% reduced the N_2O concentration measured by 1.5%, with no additional changes thereafter. Both isotope ratios were affected by O_2 availability, while SP values were more stable, declining by 2 and 0.5 units in two separate tests. The moisture content of gas samples in Tedlar bags, but not aluminum foil bags, changed during the first few hours of storage.

The panel below shows N_2O concentrations (A) and ^{15}N enrichment (B) as influenced by WFPS and slurry application rate after 1 d of incubation. The importance of both N availability and O_2 depletion is evident. Patterns changed dramatically in the following days, with evidence for NH_4^+ derived N_2O production, especially at 80% WFPS.

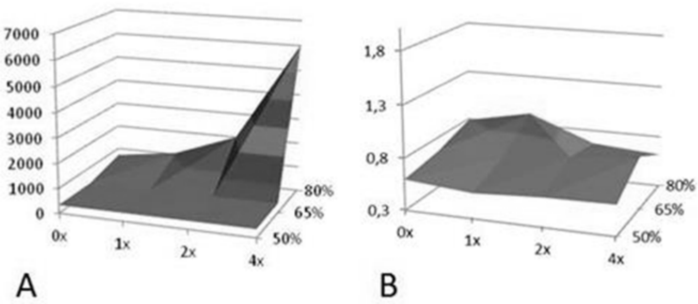

Figure 1. A. Nitrous oxide concentrations (ppb) and B. 15N (atom%) in N2O after 1-d incubation (see text).

Conclusions

The new possibilities for analysis of N_2O at ambient and elevated levels are promising. Test results indicate that robust information about dry mixing ratios and isotope composition of N_2O can be obtained across a wide range of conditions. In combination with labeled substrates, new insights may be obtained about the regulation of N_2O emissions in natural environments.

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SIMULATION OF NITROUS OXIDE EMISSIONS USING THE APSIM PLATFORM: AN EXPLORATORY STUDY ABOUT DENITRIFICATION´S CONSTANT FOR A BRAZILIAN SUGAR CANE SOIL

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The APSIM (Agricultural Production System Simulator- modified open code licensing) is a complete farm simulator (KEATING et al., 2003). The option of predict the soil emissions of nitrous oxide flux in a daily basis was recently added to the SOILN module (THORBURN et al., 2010) using a already existing mathematical model (DELGROSSO et al., 2000). The APSIM based forecast of nitrous oxide flux from Australians sugar cane soils were described by Thorburn et al (2010) that concluded that the SOILN denitrification constant should be fixed higher in comparison to the default one to produce a better fit about in field N2O measures. The application of APSIM to simulate farm´s process in Brazil is in its early stages, principally, with respect to sugar cane soils GHG emissions and its related parameters. The objective of this work was to explore the optimal values of denitrification constant (Kdenit) considering nitrous oxide emissions measures on farm related to different amount of trash above soil.

Materials and Methods

The N_2O data used for simulations were collected from sugar cane soils in Piracicaba (São Paulo state, Brazil) and described by Carmo et al., 2013, this data were produced in typical Brazilians sugar cane soil conditions and are related to an data production effort for feeding realistic estimative of nitrous oxide emission factor. For math modeling purpose,some assumptions were done: 1) virtual varietal qualities like Brazilian RB-72454 varietal 2) soil hydraulic properties based on pedotransfer function (TOMASELLA; HODNETT; ROSSATO, 2000); 3)Drained upper level at -100KPa matric potential (BATTIE LACLAU; LACLAU, 2009); 4) diffusivity (k1) in SOILN = 12,38; 5) Nitrification pH range started in 3 and finished in pH 9; 6) k2 (nitrification parameter) was assigned to 0,002 (THORBURN et al., 2010).

In a total accounting, fifteen simulations were done. For each treatment: T1- 7t trash/ha, T2- 14t trash/ha and T3- 21t trash/ha, five kdenit values were used to set up the performed simulations: P5-0.0002,P4-0.0004,P3-0.0006,P2-0.0008 andP1-0.001379. The results were summarized by the RMSE index.

Results and Discussion

Based on the simulations evaluated, in general the best fit was achieved with RMSE (0.044 Kg N-N2O/ha) (figure 1) were produced by the T2-P3 simulation; By the T1 the lower RMSE were related to T1-P2 (0.049 Kg N-N2O/ha) and by T3 the lower RMSE were related to T3-P1 (0.16 Kg N-N2O/ha). The results suggested the necessity of adjusting kdenit for each treatment, although T1 and T2 presented quite similar pattern and a relative agreement to the default kdenit value. However, in contrast to T1 and T2 the best fit for T3 is in agreement to the modified k_{denit} by Thorburn et al (2010).

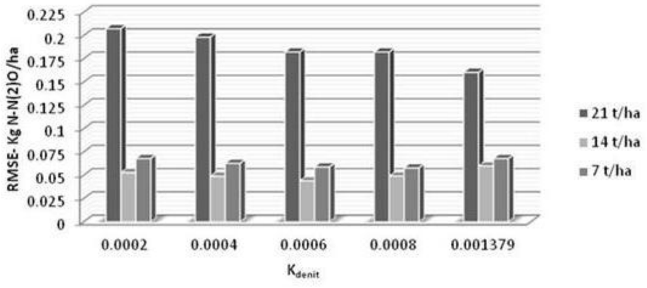

Figure 1 – RMSE of the all performed simulations for varying k_{denit} and dose of trash above soil.

Conclusions

The results reinforced that kdenit probably is site specific. Notably, in the fit of T2-P3 about forecasted values (with low RMSE) and related dynamics of simulation there are considerable indicators of the utility of APSIM as tool for management of the nitrogen as fertilizer in typical brazilian sugar cane farms of São Paulo state

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POTENTIAL EMISSIONS OF NITROUS OXIDE FROM VINEYARD SOIL: SIMULATION OF RAIN AND FERTILIZATION EVENTS UNDER CONTROLLED CONDITIONS

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Of global anthropogenic emissions of greenhouse gases, agriculture accounts for about 60% of nitrous oxide (N_2O) (Smith *et al.*, 2007). Globally, agricultural N_2O emissions have increased by nearly 17% from 1990 to 2005. N_2O emissions are linked to management practices and biogeochemical soil properties. Agricultural practices influence in different ways the soil properties and the biological processes occurring in soil as nitrogen (N) mineralization. The objectives of the study were to determine the potential emissions of N_2O in Champagne vineyard soil under different cultural practices and to provide a better understanding of the mechanisms behind N_2O emissions to improve mitigation strategies.

Materials and Methods

This study was conducted in a vineyard of 0.5 ha in Champagne Ardennes region in Northeast France (Montbré, 49°11'N, 4°02'E). The soil is of brown limestone and comprised of 20.8% clay, 46.0% silt and 33.2% sand with a pH_{H2O} of 7.6. In 1988, it was planted in vines and a field trial started from 1991 with four different managements in the alleys between grapevine rows. These were (1) control treatment with bare soil inter-rows, (2) organic N fertilizer with inter-rows receiving 51 kg N/ha/yr of dry-pellet form, (3) conifers bark applied every three years at rate 150 m^3/ha and (4) grass-covering treatment with bluegrass inter-rows and bare soil and 50 kg N/ha/yr (ammonitrate 33%) under rows. During the spring of 2012, soil samples were collected for the 0-5 cm and the 5-25 cm layers, sieved through a 4-mm mesh-size sieve and stored at 4°C. At the beginning of the experimentation, soil was repacked into PVC columns of 15.4 cm in diameter and 30 cm of height, with a bulk density close to field density. The experiment (with 3 replicates per treatment) was conducted in a climate chamber at 20°C. Rains were applied with deionized water with rain simulators, which consisted of capillary tubes (inner diameter 0.5 mm), equally distributed over the surface of a column (186 cm²). Columns received a total of 5 rains with 4 to 19 mm of water applied at 12mm/h intensity (Fig. 1). "Fertilization" was realized by the application of KNO_3 solution on the column surface at a rate equivalent to 15 kgN/ha. We used quantum cascade laser absorption spectroscopy (QCLAS) for the measurement of soil-derived N_2O emissions (Mappé-Fogaing *et al.*, 2012; Köster *et al.*, 2013). Some biogeochemical soil variables were measured at the beginning and the end of the experiment.

Results and Discussion

Soil moisture, total N and carbon (C) and microbial biomass C distinguished clearly the 4 treatments after 21 years of different agronomic practices (Table 1). Soils of control and organic N fertilizer treatments were characterized by low water holding capacity and low N and C organic content. Conversely, grass and bark treatments enhanced water retention properties and soil organic matter content with high contents of total N, C and microbial biomass, especially in the 0-5 cm soil layer. N_2O emissions followed the same hierarchy than the soil variables with soil-induced N_2O emissions ranking as control2O emissions. In spite of high content of C and N in soil of the grass treatment, soil did not emitted N_2O in initial conditions or after water application but responded in N_2O emissions only after the addition of water and nitrate together. Soil of conifers bark treatment was the only treatment with high N_2O emissions, up to 0.11 µgN/m²/s in response to the addition of water and nitrate together. N_2O emissions triggered around four hours after simulation events and lasted about 48 hours with a peak emission after about 9 hours.

Conclusions

Potential of N_2O emission was high in vineyards soil especially if conditions of accumulation of organic matter, high availability of nitrate and saturation of water in soil were met. In the study, these conditions were found in the soil under conifers bark management during 21 years, after addition of water and nitrate simulating possible field conditions.

Table 1. Soil variables of the four treatments in the 0-5 and 5-25 cm layers at the beginning and end of the experiment: soil moisture for wilting point (WP) of 3, contents of organic carbon (C), total nitrogen (N), C/N ratio, microbial biomass C, dissolved organic C (DOC), nitrate (NO_3^-) and ammonium (NH_4^+).

		Beginning of the experiment					End of the experiment			
Treatment	Soil layer (cm)	Moisture (WP=3)	Organic C (g/kg)	Total N (g/kg)	C/N	Microbial biomass (mg C/kg)	DOC (mg C/kg)	NO_3^- (mg N/kg)	NH_4^+ (mg N/kg)	Microbial biomass (mg C/kg)
Control	0-5	16%	20	1.2	16	68	13	68	0.4	306
	5-25	18%	13	1.0	14	31	-	27	0.4	-
Organic N fertilizer	0-5	18%	24	1.5	16	92	15	61	0.5	373
	5-25	18%	17	1.1	16	31	-	32	0.4	-
Grass-covering	0-5	29%	44	3.1	14	363	88	85	12	1050
	5-25	20%	21	1.4	15	65	-	4	3.4	-
Conifers bark	0-5	41%	104	3.4	31	622	34	117	0.6	911
	5-25	21%	20	1.1	18	43	-	43	0.5	-

Fig. 1. Column of soil under rain simulator

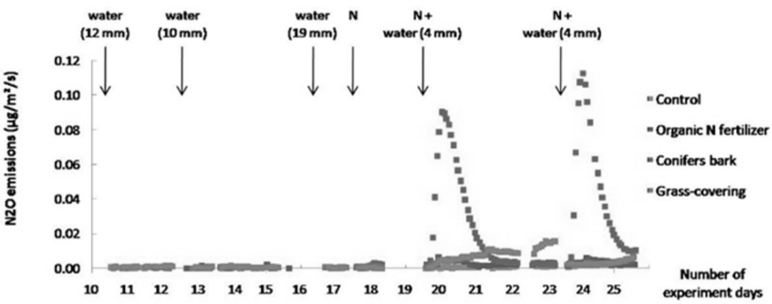

Fig. 2. Mean of the soil-derived N_2O emissions (n=3) from the four treatments of vineyard soil. Times of water and N additions are indicated with arrows.

REDUCING NITROUS OXIDE EMISSIONS FROM SPRING BARLEY IN IRELAND

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The main source of nitrous oxide (N_2O) emissions from tillage farms in Ireland is the use of nitrogen (N) fertilisers. Calcium ammonium nitrate (CAN) is the dominant N fertiliser used by Irish tillage farmers. When used under wet temperate climatic conditions, CAN has the potential to increase N_2O emissions by immediately contributing to the soil nitrate pool for denitrification processes. Switching from ammonium nitrate to urea based N fertilisers can reduce N_2O emissions by 73% in intensive grassland (Dobbie and Smith, 2003). On tillage soils Smith et al. (2012) reported that urea reduced emissions by 20% on a sandy clay-loam soil and increased emissions on two finer textured soils by 13% (silty clay-loam) and 70% (clay) compared to CAN. Urea could potentially increase the risk of N loss through ammonia volatilisation. Applying a urease inhibitor (N-(n-butyl) thiophosphoric triamide (nBTPT) or Agrotain ™) to delay the breakdown of urea to ammonia is one method of reducing these N loss risks. In addition, the application of a nitrification inhibitor (dicyandiamide (DCD)) would delay the conversion of ammonium to nitrate, potentially further reducing the denitrification N loss risks. In Ireland the tillage sector development plan (O Reilly, 2012) has projected a 64% increase in the area under crops by 2020. If this is achieved there will be an increase in N fertiliser use and potentially N_2O emissions. However, under the Kyoto Protocol, Ireland is required to limit greenhouse gas emissions to 13% above 1990 levels by 2020. In order for tillage farmers to meet these seemingly opposing agronomic and environmental objectives, mitigation techniques for N_2O emissions are needed. The objectives of this study are to (i) compare and quantify the effect of N fertiliser type (CAN vs. Urea) on N_2O emissions and (ii) quantify the effect of the urease inhibitor nBTPT and the nitrification inhibitor DCD on N_2O emissions and assess their potential to mitigate GHG emissions from spring cereal production systems in Ireland.

Materials and Methods

Two experimental field sites, both located in the southeast of Ireland, with contrasting soil types and drainage characteristics (poorly-draining clay loam and a free-draining loam) were used in this experiment. The experiment was initiated in 2013 and will be conducted over two successive field seasons to capture a range in climatic variability. The experiment was laid out in a randomised block design, with five replications of each treatment. A total of 20 treatments comprising of multiple combinations of N fertiliser formulations (CAN,Urea,Urea+Agrotain (UA), Urea+DCD (UD) and Urea+Agrotain +DCD (UAD)) at different rates including an untreated control were evaluated. Fertiliser was applied in two splits: 30 kg N ha^{-1} at sowing time (first split) and the remainder during tillering (second split) as per standard agronomic practice. Gas fluxes were measured using the closed chamber technique (Clayton et al. 1994) using stainless steel chambers. Samples were taken four times during the first two weeks after fertilisation, then twice per week for the following two weeks, then once per week until two months post-harvest where sampling has continued fortnightly. Nitrous oxide concentrations were analysed on a Varian CP-3800 Gas chromatograph with an electron capture detector (ECD). In this paper we present the preliminary N_2O emission results for the standard agronomic rate (~150 kg N ha^{-1}) following the second split N application. Further detail and discussion of results will be presented at the 18th Nitrogen Workshop.

Results and Discussion

Following a cold and dry period prior to the application of second split N fertiliser (day 0) the background N_2O (control) emissions were low (< 4.5 g N_2O-N ha^{-1}) at both sites (Figure 1). These background N_2O emissions remained relatively low for the first 9 days post application with all N treatments at the free drained site following a similar low trend over this period. The N_2O emissions for the CAN, Urea and UA treatments were higher compared to the control at the poorly drained site during this period. About day 15 peak emissions were measured at both sites. The N treatments with the lowest emissions were UD and UAD which were similar to background concentrations. Emissions declined to background concentrations from day 17 on the free draining site but a second emission peak occurred on day 30 at the poorly drained site before emissions returned to background levels for all N treatments.

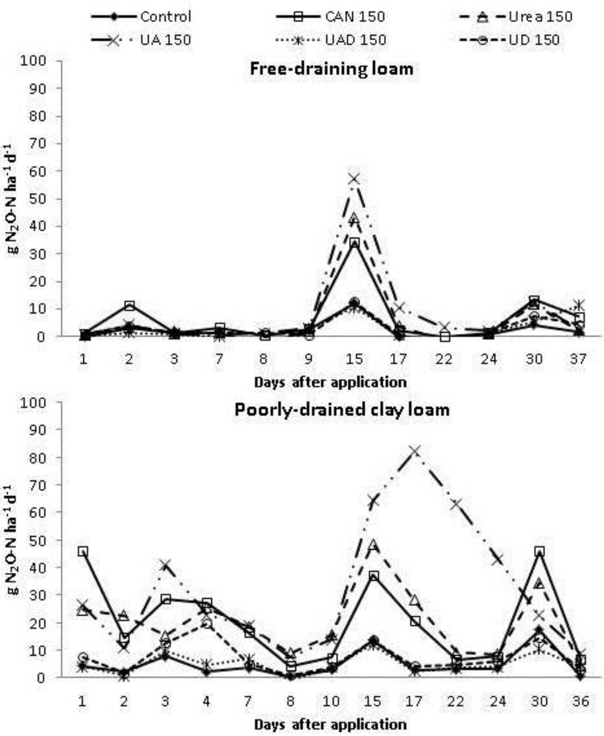

Figure 1.N_2O flux over a 37 day period following N application of 150 kg N ha^{-1}

Conclusions

These early results show a trend for lower N_2O emissions with Urea + DCD (significantly lower during the peak in N_2O emissions on day 15) in comparison to CAN, Urea and the Urea + Agrotain over the first 4 weeks after N fertiliser application. This indicates that DCD may be limiting nitrification, thus keeping the soil nitrate pool close to background levels for these N treatments.

Acknowledgements

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NITROGEN MINERALIZED DURING SORGHUM GROWTH AFTER SOIL INCORPORATION OF DIFFERENT WINTER COVER CROPS

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Growing catch crops during the autumn/winter period is a strategy of high ecological significance since it allows reducing the residual inorganic-N present in the soil after the summer season (Rodrigues et al., 2002). Thus, winter catch crops reduce the risk of denitrification and nitrate leaching associated to the excess of rain of the autumn/winter months. Incidentally, the evergreen systems confer several other additional benefits, including protection against soil erosion and increasing soil organic matter. In recent years, agronomists and soil scientists have studied the pros and cons of the introduction of cover/catch crops in different agro-ecological conditions and cropping systems. Some were focused in comparing the performance of different plant species when they were used as catch crops (Jensen, 1992). In addition, since winter catch crops precedes summer cash crops, it is important to know the effect of the catch crop in the performance of the cash crop. As a general rule, the catch crop should present good growth rate in winter and improve soil fertility to promote the growth of the summer crop. Theoretically, lupine (*Lupinus albus*) seems to have both features. It is a species of high biomass production in autumn/winter period (Rodrigues et al., 2013) and, being a legume species with tissues of low C/N ratio, net nitrogen mineralization should occur early in the growing season of the crop that follows lupine in the rotation. In this work, results are presented of the effect of several winter cover crops in nitrogen availability in a soil cultivated with sorghum in the following summer season.

Materials and methods

In the autumn of 2011 three different ground cover systems were established: natural vegetation (weeds); cereal (a mixture of small grains); and white lupine, in four plots (replications) each. In spring, the cover crops were mowed and buried during soil preparation for the installation of sorghum (*Sorghum bicolor* L. Moench). The lupine plots were divided into two equal parts each. In half the area of each plot, all the biomass of the lupine plants was incorporated in soil. In the other half of the area, the above-ground biomass of the lupine was removed, remaining in the soil the roots of the plants. Fifteen days after the sowing of sorghum, an in situ incubation has started, consisting of filling sharp PVC tubes (140 mm high and 32 mm in diameter), pushing them directly to the soil. The soil in PVC tubes was thereafter incubated within glass jars buried next to the field plots. By a sequential analysis of inorganic nitrogen in fresh and incubated soil samples it was possible to follow net nitrogen mineralization from the organic residues incorporated in soil. For more details on the incubation technique see Rodrigues (2004a). Soil extracts were prepared from fresh and incubated samples using a KCl (2*M*) solution as the extractant agent. The extracts were subsequently analysed for nitrate and ammonium concentrations by UV and visible spectrophotometry.

Results and discussion

Soil nitrate levels decreased during the growing season in all plots due to the increasing uptake capability of sorghum plants (Figure 1, a). Soil ammonium levels fluctuated between 3 and 9 mg kg^{-1} and represent the balance between net N mineralization and nitrification processes (Figure 1, b). Mineral N accumulates in aerated soils as nitrate. Nitrate accumulation during the growing season of sorghum was close to 13 mg kg^{-1} (~18 kg N ha^{-1}) in the plots of lupines (Figure 2, a). These values are unexpected low taking into account the biomass that was incorporated in soil, and reference values found in literature (Magdoff et al., 1984; Rodrigues, 2004b), but significantly higher than the values recorded in Weeds and Cereal plots. If total inorganic-N (NO_3^- + NH_4^+) was used to estimate net N mineralization (Figure 2, b) the values were lower due to the great dynamic of NH_4^+ in soil. However, the trend was similar to that found when NO_3^--N form was used alone. In the plots of lupine were recorded the higher values. Also unexpected was the fact of lupine (roots) has shown similar results of lupine (total) considering the great difference of biomass incorporated.

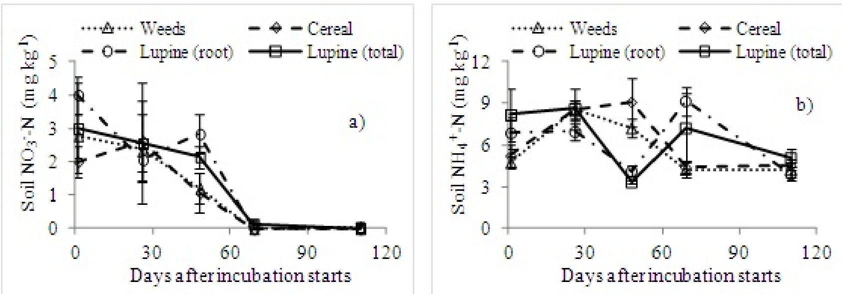

Figure 1. Nitrate-nitrogen (a) and ammonium-nitrogen (b) in soil during the growing season of sorghum in different cover cropped plots.

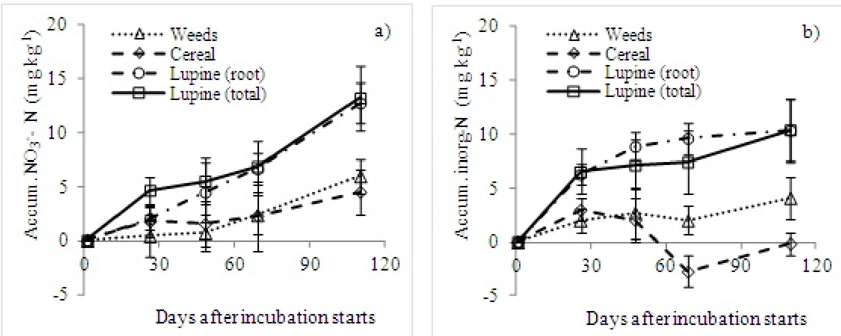

Figure 2. Total nitrate-nitrogen (a) and inorganic-nitrogen (b) accumulated in the successive periods of soil incubation during the growing season of sorghum and in the different cover cropped plots.

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THE NH3-ABATING EFFECT OF SLURRY INJECTION AS INFLUENCED BY SOIL MOISTURE IN A SEMIARID ARABLE SOIL

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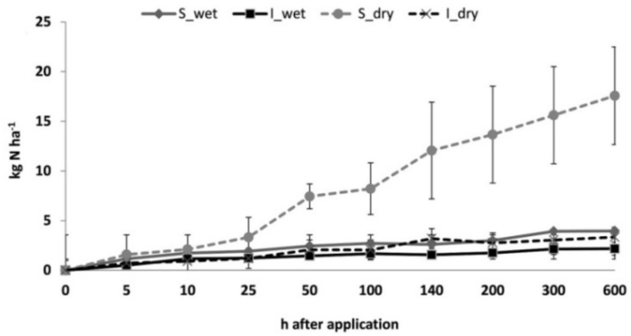
Accumulation of large volumes of dilute slurries is considered one of the major problems related to intensive farming (Sommer et al., 2004). In the EU-27, more than half of the total N excretion is applied to croplands due to technical advantages for farmers (e.g. reuse of nutrients). However, the N use efficiency of slurries produced by livestock is low, i.e. only 20-52% of the excreted N is recovered by crops. Much of the remainder can be lost into the atmosphere as ammonia (NH3), nitrous oxide (N2O), dinitrogen (N2) and nitrogen oxides (NOx). In the case of NH3, 80-90% of these emissions in the agricultural sector arise from the excreta produced by livestock (FAO, 2006). In Spain, slurries are, commonly, almost entirely surface applied to land via high-rate irrigation systems, which reduce the N fertiliser efficiency of animal slurries due to NH3 volatilization (Misselbrook et al., 2005). Research in several European countries has demonstrated that the injection of slurry, compared with surface broadcast application, may reduce NH3 emissions by as much as 90% (Misselbrook et al., 2005). Land spreading of slurries is therefore a serious concern, which also offers great potential for cost effective abatement through slurry application techniques. Some authors have reported on the influence of certain physicochemical factors such as temperature and soil moisture on the effectiveness of low emission slurry application techniques. Smith et al. (2000) suggest that soil conditions and, in particular, moisture status have important impact on NH3 emissions from applied slurries. Two field experiments were carried out on a fertilized barley (Hordeum vulgare) crop in central Spain with the main objectives of: (1) quantifying the NH3 emissions from an agricultural soil amended with pig slurry by means of a micrometeorological technique (IHF method) in a Mediterranean area; (2) comparing two application techniques (surface broadcast and shallow injection) to gain insight about the effectiveness of shallow injection as a mean of reducing NH3 volatilisation; and (3) the evaluation of the influence of soil moisture content on NH3 volatilisation and on the effectiveness of slurry injection as a mitigation method.

Materials and methods

The study was conducted in a Typic Xerofluvent in Madrid (40º 18´ 14´´ N; 3º 25´ 57´´ W) on a barley (Hordeum vulgare) crop. Some relevant characteristics of the top soil layer (0-20 cm) were: total organic matter, 1.4%; pHH2O, 8.1; bulk density, 1.47 Mg m-3; CaCO3, 3.4%; field capacity, 20.2% (w/w); porosity, 46%; sand, 37%; silt, 45%; and clay, 13%. The average annual temperature and rainfall (over the last 10 years) in this area were 13.5 ºC and 460 mm, respectively. The characteristics of the pig slurry were: total N, 3.1 g kg-1; total C, 18.02 g kg-1; DOC, 0.5 g kg-1; NH4+-N, 2.3 g kg-1; pH, 7.51 and moisture content, 98.4%. Four rectangular plots (16 m2) (i.e. two replicates per treatment) were delimited before the application of the slurry. Total volume of applied slurry was 8 m3 per plot adjusted to provide 85 kg N ha-1 as basal fertilization. Pig slurry was applied by both broadcast application and shallow injection on the 2nd November and 12nd December, for experiments 1 and 2, respectively. The micrometeorological mass balance method (Leuning, 1985; Misselbrook, 2005) was used for measuring NH3 emissions from treated plots.

Results and discussion

Ammonia volatilisation significantly peaked after slurry application in the two experiments. However, under conditions of high soil moisture (WFPS≥70%), resulting from intense rainfall (i.e. 44 mm, in the first week of November), total NH3 emitted was not higher than 4 kg N ha-1 (4.6% of the applied N). Under these conditions, shallow injection reduced NH3 volatilisation by 46% compared with surface broadcast. In contrast, results from the second experiment showed that surface application of slurry over a drier soil (13.5 mm rainfall, WFPS≤55%) enhanced N losses through NH3 volatilisation up to 17.6 kg N ha-1 (20.7% of applied N). Since soil temperature was not significantly different between the two experiments (i.e. 16.7 vs 15.8ºC, respectively), it could be concluded that the NH3-abating effect of injection was significantly affected by conditions of soil moisture, with an 81% decrease compared to surface application of the slurry. Under conditions of high soil moisture, NH4+ from the slurry would have been rapidly oxidized to NO3-, easily leached by the intense rainfall, thus decreasing NH3 losses. Additionally, as the soil was almost saturated, the effectiveness of shallow injection over the placement of the slurry in deeper zones of the soil was decreased. Under drier conditions, the injection of the slurry reduced the contact surface between the fertilizer and the atmosphere thus decreasing volatilization.



Acknowledgement

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N2O REDUCTION POTENTIAL OF 3,4-DIMETHYL-PYRAZOLE PHOSPHATE (DMPP) IN DIGESTATE INJECTION

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Due to high nitrogen losses, leaching and gaseous losses the N-use efficiency in crop farming is not optimizedis (Freney 1997). About 50% of the introduced Ntotal may be lost through NH3 emissions (Freney 1997). NH3 emissions can be reduced by rapid incorporation of manure or fertilizer injection by up to 80%. However, by injection anaerobic processes in the soil lead to a higher risk for nitrification and denitrification of the introduced nitrogen, resulting in potentially higher nitrogen losses in the form of N2O. By nitrification-inhibitors NO3- leaching and N losses in the form of N2O emissions can be reduced (Pfab et al. 2012). The aim of the study was to compare the digestate injection to the trailing hose application with immediate incorporation with respect to the emission release (N2O, CH4 and CO2) and find out how a nitrification inhibitor can reduce N2O emissions in addition to the application technique

Materials and Methods

In a microcosm study digestate injection (15 + 20 cm) was compared with digestate trailing hose application with immediate incorporation. 56 undisturbed soil columns [Gleysol (73 % sand, 13 % silt, 14 % clay, 1,7 % Corg, 5,9 mg/kg NO3- and 0,3 mg/kg NH4+) and a Plaggic Anthrosol (83 % sand, 8 % silt, 6 % clay, 2,4 % Corg, 5 mg/kg NO3- and 0,6 mg/kg NH4+)] were used. Soil columns were 40 cm high and have a diameter of 15 cm. The temperature in the microcosm system was adjusted to 12 ° C. In the experiment digestate from pig slurry (6,11 % DM, 0,51 % Ntotal, 0,25 % NH4+ and pH 7,5) was used.

For a comparison of variants each soil column was fertilized with 50 ml digestate ($30 m^2$ ha-1 = 150 kg N ha-1). Half of all variants 3,35 mg DMPP was added to the digestate (50 ml) (Dittert et al. 2001, Zerulla et al. 2001 and Weiske et al. 2001). 3,4-dimethyl-pyrazole-phosphate (DMPP) is blocking the Ammoniumoxygenase, so nitrification is delayed in the soil (Zerulla et al. 2001). The injection variants, 15 or 20 cm topsoil were pushed out and sieved to 5 mm. A furrow ($63 cm^3$ volume) was stabbed in the soil. The digestate was introduced with a syringe in the furrow. In the trailing hose variant 5 cm topsoil were sieved (5 mm) and mixed with the digestate. Digestate was mixed with the soil, because in Germany farmers have to incorporate digestate not later than four hours after application on soil. With shimadzu GC2014 N2O, CH4 und CO2 were measured.

Results and Discussion

Mean daily fluxes were cumulated for 52 days. The Gleysol shows tendencially but not significant higher N2O fluxes than the Plaggic Anthrosol. In both soils the use of DMPP shows a decrease in N2O emission. But the difference between variants treated with DMPP and don't treated with DMPP is not significant, because of high standard deviations. The emission reduction potential of DMPP at the Gleysol is 14.78 %, well below that of the Plaggic Anthrosol (57.74 %) (Figure 1).

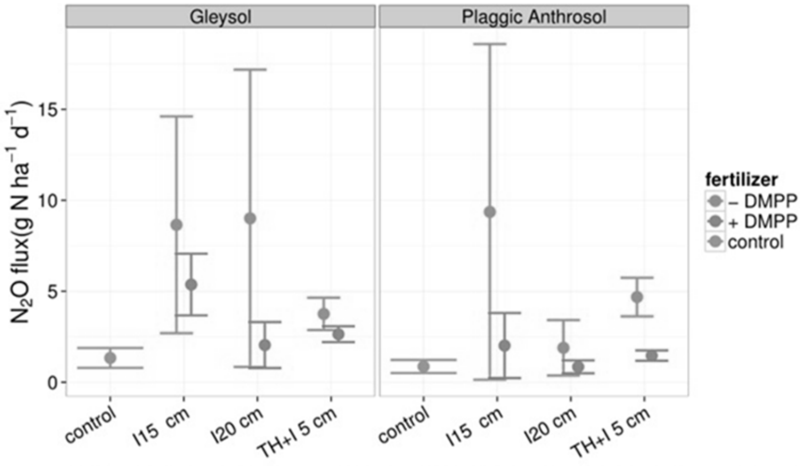

Figure 1: Averages of the daily N_2O emissions (mean and standard deviation) with an untreated control (control), injection (I15 and I20) and at the trail hose application (TH + I 5 cm) of digestate on Gleysol and Plaggic Anthrosol over a period of 52 days

In a field trial Weiske et al. (2001) could demonstrate a N2O reduction potential by DMPP addition of 49%. Also Liu et al. (2013) were able to demonstrate a N2O reduction potential by the addition of DMPP. The effect of DMPP is influenced by the type of soil and the soil structure. With increasing soil grain size, the effect of the nitrification inhibitor decreases (Akiyama et al. 2010). The analysis of NH4+ and NO3- indicates that the addition of DMPP delay the conversion of NH4+ to NO3- More NH4+ remains in variants treated with DMPP. In the untreated columns, a large part of the NH4 is converted into NO3. That is a sign of delayed ammonium oxidation (Zerulla et al. 2001)

Conclusions

A big part of released emissions after digestate injection depends on soil type. The application technique is less important for emission release. The investigated nitrification inhibitor DMPP can reduce nitrogen losses by 32.54 % on average. This small scale study can inform about the relations of emission release between application techniques and nitrification inhibitor effect, but it cannot give any values that are transferable to values from field trials. Field trials are needed to verify this results.

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AN EMPIRICAL APPROACH TO ESTIMATE THE EFFECTIVENESS OF DICYANDIAMIDE IN DECREASING NITRATE LEACHING FROM GRAZED PASTURE IN NEW ZEALAND

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Dicyandiamide (DCD) is a bacteriostat that inhibits the first stage of the nitrification process. DCD application over the ruminant urine patch has been suggested to reduce NO3-N leaching under grazed pasture (Monaghan et al., 2007). Its use needs to be correctly accounted for in any farm-scale assessment of NO3-N leaching risk. OVERSEER® Nutrient Budgets (Overseer) is a software tool that estimates farm-specific budgets for a range of nutrients including N; and includes an estimate for NO3-N leaching (www.overseer.org.nz). The aim of this work was to develop a DCD module for Overseer that would estimate DCD effectiveness in decreasing NO3-N leaching from urine patches at the block management level within the farm.

Materials and methods

In brief, Overseer estimates monthly N excretion from the grazing animals' energy requirements and feed consumption. The excreta are partitioned between urine and dung based on feed composition. A water balance model estimates soil drainage, and a transfer coefficient is applied (Cichota et al., 2012) to each month's urine to estimate the proportion leached. We hypothesised temperature (affecting degradation) and drainage (affecting mobility) would be two drivers altering DCD effectiveness, and these were used to develop an 'Effectiveness Index' (EI). When tested against a series of lysimeter experiments under NZ conditions, the EI explained 77% of the variance in DCD effectiveness ($P< 0.001$) in decreasing NO3-N leaching (Shepherd et al., 2012). The EI was then incorporated into a DCD module within Overseer in which up to three DCD applications could be made, with flexibility in timing. The model was tested against the few experiments where DCD effectiveness in reducing NO3-N leaching has been measured under grazing. The sensitivity of key component parameters on modelled DCD effectiveness was tested on an Overseer base file set up to represent a typical NZ dairy farm. These were: rainfall; temperature (by region); soil-type (2); number of DCD applications (0-3); and months of application (Feb-Aug). The component parameters were sampled at random from a priori uniform distributions to generate multiple (20,000) test files following a Monte Carlo based procedure. These were run through Overseer to assess the interaction between parameters and their combined influence on DCD effectiveness.

Results and discussion

DCD effectiveness was expressed as the percentage reduction in NO3-N leaching compared with a non-DCD control. When the DCD module in Overseer was compared with three grazing experiments, the agreement in decreased leaching was reasonable for two sites: 13% vs. 19% measured (Ledgard et al., 2008); 29% vs 21% measured (Ledgard, Pers. Comm.). Overseer appeared to underestimate effectiveness at a third site: 15% vs. 40% measured (Monaghan et al., 2009). However, the amount of N leached was only 13 kg N/ha, so a small difference in estimated leaching can be a large percentage value. Figure 1a shows the sensitivity plot for modelled DCD effectiveness in one region (Waikato, annual average temperature 13.7 °C) as affected by drainage, soil-type, and number and timing of DCD applications. Effectiveness declined with increased drainage. There were 3 broad bands, with 3 DCD applications generally better than 1 or 2 applications, but with some overlap. Variation in effectiveness within each band was due to interactions of soil-type and timing of application(s). The sensitivity analysis was repeated for three other regions (Northland, Canterbury and Otago) to cover a range of annual average temperatures. Figure 1b summarises the modelled DCD effectiveness for regions at 300 and 600 mm drainage to demonstrate responsiveness of the model. The sensitivity tests generally showed three DCD applications performed best and one application worst. Effectiveness increased as temperature decreased. and decreased as drainage increased. However, temperature was the more important driver. For the base dairy farm we modelled, best strategy was 3 DCD applications (March, May and July) to protect urine deposited during grazing in autumn and winter (all-year grazing on this farm). Effectiveness ranged between 15-30% for 2 DCD applications, with the best strategy to apply in April and July.

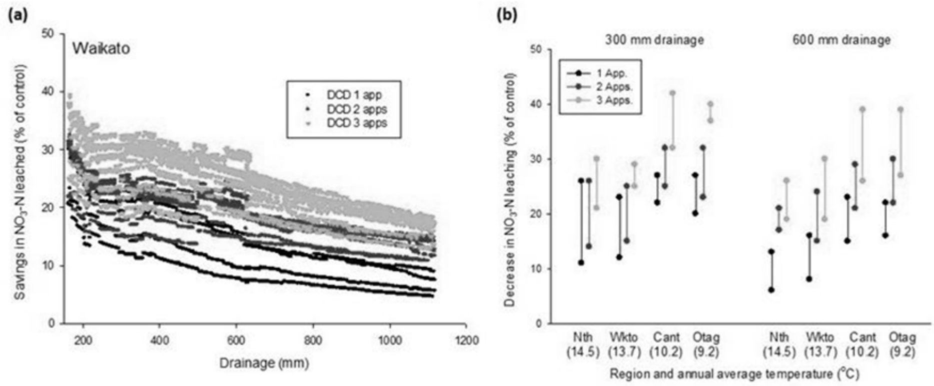

Figure 1. (a) An example of the range in sensitivity of modelled DCD effectiveness for the Waikato region. (b) The range of estimated DCD effectiveness at 300 and 600 mm drainage as affected by number of applications and annual average temperature (region).

Conclusions

One of the main challenges for the Overseer model is scaling to the paddock/farm level when most of the research has been done at the urine patch level. Applying to the Overseer model the empirical EI relationships based on drainage and temperature gave DCD effectiveness estimates similar to measurements in grazing experiments. Sensitivity analysis showed effectiveness varied with environmental conditions and number and timing of DCD applications. This suggests Overseer is a useful tool for identifying farm-specific strategies for DCD application to decrease NO3-N leaching.

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COMPARISON OF TWO DAIRY GRAZING SYSTEMS IN THE WAIKATO REGION OF NEW ZEALAND: EARLY RESULTS ON NITRATE LEACHING

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Sustainable production systems should be financially viable and minimise impacts on the environment. Dairy farming is central to the NZ economy, with the Waikato region responsible for c. 25% of the annual production (Anon., 2012). Our hypothesis was underpinned by farm system modelling (Vogeler et al., 2012). It is that production and farm profits can be increased and nitrate (NO_3-N) leaching decreased in Waikato dairy systems by using a combination of higher genetic merit cows stocked at a lower rate with reduced N fertiliser inputs and an improved dietary balance. This paper reports first year (2012) results for NO_3-N leaching from a study designed to test the hypothesis.

Materials and methods

Two dairy 'farmlets' were established during winter 2011 to replicate (1) a system typical to the Waikato region ('Current') and (2) a system modified to decrease NO_3-N leaching but maintain/improve profitability ('Future'). Each farmlet is 13 ha, made up of twenty-six 0.5 ha paddocks paired to take account of soil-types, which are a mix of mineral and organic/peaty silt loams. Pasture is perennial ryegrass/white clover (*Lolium/Trifolium repens*), with some tall fescue (*Festucca arundinacea*) also used. Further background information is provided by Chapman et al. (2012). The tactical management options for decreasing N leaching, summarised in Table 1, include: a lower stocking rate with more efficient cows (greater 'Breeding Worth', a measure of a cow's genetic potential); supplementing the diet with grain (lower N concentration than pasture); and removing cows from paddocks to a 'stand-off pad' for several hours per day during autumn/winter (thus reducing urine deposition to soil). Nitrification inhibitor (dicyandiamide) was applied in autumn/winter post-grazing because this can decrease NO_3-N leaching (Monaghan et al., 2009), but has since been withdrawn from the experiment. The use of these technologies in NZ dairy farming is described by Monaghan et al. (2007). Ten porous ceramic cup samplers were installed in each paddock, to a vertical depth of 60 cm, at an angle of 45 degrees. Soil solution was collected about every 60 mm of drainage. Samples were analysed for ammonium-N (NH_4-N) and NO_3-N using a segmented flow auto analyser (Skalar San^{++} System). Load of leached N was estimated from measured soil solution concentrations and drainage volumes (Lord and Shepherd, 1993) derived from a soil water balance model.

Results and discussion

Modelled drainage was 428 mm between May and September 2012, with a small additional amount in October. Porous cups were sampled seven times. There was a wide range of NO_3-N concentrations (Figure 1). The distribution was highly skewed and typical of grazed paddocks (Cuttle et al., 1992), but the variation was larger in the Current than in the Future treatment. This was interpreted as indicative of more urine patches being intercepted by samplers under the Current treatment. This fits with the hypothesis that standing animals off paddocks in autumn/winter to minimise urine deposition would decrease N leaching (Monaghan et al., 2007), although the effect will be due to a combination of practices in a systems study. The great mean NO_3-N concentration in soil solution (drainage) from the Current treatment produced a significant difference in NO_3-N leaching ($P< 0.01$ after log transformation for analysis of variance): Future 22 kg N/ha, Current 50 kg N/ha (untransformed means). This decrease of >50%, albeit for one year, is in line with expected reductions of c. 40% based on pre-experimental modelling of the system (Vogeler et al., 2012). Annual production was the same for both herds (1200 kg MS/ha), which was achieved by keeping the lower stocked herd in lactation for later into the autumn.

Table 1*. Treatment details for Current and Future farmlets.*

Treatment/Management options	Current	Future
Stocking rate (cows/ha) / cows per herd	3.2 / 42	2.6 / 34
N fertiliser (kg N/ha/year)	Up to 150	Up to 50
Cow Breeding Worth	90	170
Cow liveweight (kg)	500	480
Herd replacement rate (%)	21	18
Standoff/restrict grazing	None	Up to 16 hrs/day Mar-Jul
Grain purchased (kg DM/cow)	0	Up to 400
Nitrification inhibitor	No	Yes (only in year 1)

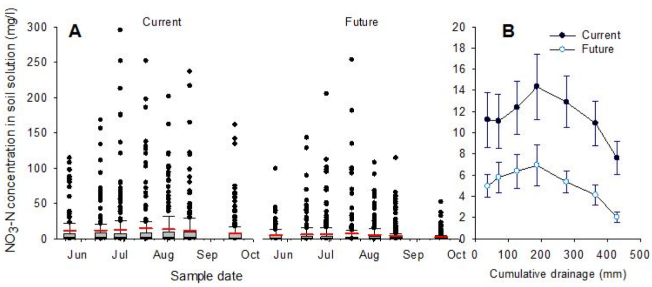

Figure 1. *A: Distributions of NO_3-N concentrations in individual soil solution samplers showing the mean (red line), median (black line), 75 and 25% percentiles (box), 90th and 10th percentiles (error bars) and outliers (closed circles). B: Mean NO_3-N concentrations against cumulative drainage.*

Conclusions

Modelling and measurements of NO_3-N leaching from this farmlet study suggests it is possible to modify management of the system to decrease N leaching losses from grazed dairy systems without compromising production. Measurements need to be repeated for several years and combined with economic data to assess rigorously the sustainability of the production system.

Acknowledgements

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INFLUENCE OF ANTHROPOGENIC LOADING ON NITROGEN LEACHING IN FLUVISOLS

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The liquid soil phase is very dynamic and sensitive soil component which is the first to respond to different influences and could be successfully used for early prediction of the possible changes in the soil. The vulnerability of Fluvisols (FAO) to anthropogenic loading and the influence of different fertilization rates on the losses of chemical elements and nitrogen with lysimeter water has been established in many studies (Stoicheva et al. 2009; Beaudoin et al. 2005). The application of unbalanced mineral fertilization and irrigation systems on soils with high permeability could cause the leaching of residual nitrogen outside the root zone and to establish a potential risk of groundwater pollution by nitrates. (Nitrate Directive 1991/676/EEC; Stoichev et al., 2001). The aim of the study was to evaluate the influence of fertilization on the nitrate nitrogen migration with lysimeter water on Fluvisols.

Materials and Methods

The field experiment, subject of this study, is located in South Bulgaria ($24^0$35'E; $42^0$14'N and 180 m above the sea-level), near the town of Plovdiv, Bulgaria. The region is characterized by the following mean annual climatic data: annual precipitation – 465 mm and the average annual air temperature is 12.5-12.8 ^{0}C. The experiment was carried out on Fluvisols, in a vulnerable area in terms of the risk of contamination by nitrates from agricultural sources. The arable horizon has the following mean characteristics: light texture, low clay content (18.6%), bulk density between 1.54 and 1.66 g.cm^{-3}, pH_{H2O} = 6.0-6.5, total nitrogen = 0.111%, humus content – 1.23%, and cation exchange capacity – 22.4 cmol.100 g^{-1}. During the experimental period the following crops were grown: 2008 – wheat (after effect); 2009, 2010 and 2011 – maize; and different rates of fertilization were applied: fallow; $B_1(N_0P_0K_0)$; $B_5(N_{100}P_{50}K_0)$; $B_3(N_{200}P_{150}K_0)$, kg.ha^{-1}. The field plots were equipped with Ebermeir lysimeters (Stoichev 1974) collecting water from a depth of 50 and 100 cm layers from the surface. The nitrogen content was measured by „Spectroquant Pharo 100".

Results and discussion

A characteristic feature of Fluvisols is the significant spatial heterogeneity and the great variety in the arrangement of the alluvial materials in the whole profile. These facts are the basis of the variation in the results for the quantities of the lysimeter waters during the considered period. Experimental results show that, lysmeter water volume for the year 2008 ranged from 11.5 to 77.5 l/m^2 for the layer 0-100 cm of the soil profile. For the year 2009 the received values are between 1.28 and 46.1 l/m^2, while during 2010 was obtained largest quantity of drainage flow with values from 14.7 to 92.9 l/m^2, which represents about 15,02 % of the total quantity of water received in the soil. Such significant volume is observed when large amounts of precipitation coincide with the applied irrigation. The analyses of data for nitrate nitrogen leaching shows that the exported quantity of fallow (control) in all the cases are lower than in the fertilized variants (fig.1). The obtained results show an increase in losses of nitrogen from variants of fertilization, especially observed at the highest fertilization rates B_3 (N_{200}). As it is visible at fig.1, when growing of wheat, the migration of nitrate nitrogen for the year 2008 is 1.0-8.1 $kg.ha^{-1}$ for the 0-50 cm soil layer and 0.9-15.1 $kg.ha^{-1}$ for 0-100 cm soil layer. Wheat growing without fertilization is a suitable way for an effective inclusion of residual nitrogen in the biological cycle of nutrients (fig.1).

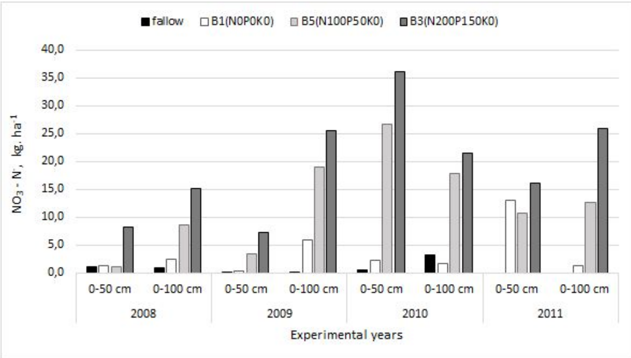

Figure 1. Nitrate nitrogen leaching (NO_3-N^-), $kg.ha^{-1}$ with lysimeter waters under 0-50 and 0-100 cm soil layer

The data obtained show that the values for 2009 are 0.8-7.3 $kg.ha^{-1}$ for 0-50 cm soil layer and higher 0.3-26.5 $kg.ha^{-1}$ for 0-100 cm soil layer. The values received for the leaching of nitrate nitrogen in 2010 are respectively 3.2-36.0 $kg.ha^{-1}$, which are the highest values measured. During 2011 the values ranged between 10.6-16.0 $kg.ha^{-1}$ for 0-50 cm soil layer and 1.3-25.8 $kg.ha^{-1}$ for 0-100 cm soil layer.

Conclusions

Long-term field experimental data show that nitrogen fertilization, especially with rates exceeding the uptake by the yield, leads to a significant leaching of nitrate nitrogen through the soil profile of Fluvisols. Under these conditions, the nitrate nitrogen in the lysimeter water increases and reach to 36.00 $kg.ha^{-1}$, which are the highest values measured at monoculture maize growing for the period of 40 years.

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ARTIFICIAL AMMONIA RELEASE AND LAGRANGIAN DISPERSION MODELLING

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Lagrangian Stochastic (LS) dispersion modelling, namely the WindTrax (WT) model (Flesch et al., 2004) is increasingly applied for the determination of agricultural emissions from confined plots (Wilson et al., 2013). WT has proven accurate under ideal Monin-Obukhov (MOST) conditions with CH_4 release experiments (Harper et al., 2011). While measurements of NH_3 are challenging due to the molecule's polarity and concentration dynamics (Sintermann, 2011), most studies addressing NH_3 emission levels from land-spread slurry take place over confined plots of roughly 30 to 50 m in diameter. Here, we report on a trial to check the WT model by release of known amounts of NH_3 from an artificial grid.

Materials and Methods

The gas release grid consisted of 36 critical orifices, spread over a circular area of 20m in diameter. Orifices were radially connected to the grid centre by 6mm OD PA tubes. Tube pressure was monitored and gas flow controlled. The grid was connected to a gas standard (2.005/5.04% NH_3 in N_2). Thus, the NH_3 release was equivalent to an area emission rate of 7/17µg/m^2/s. In order to avoid potential grass canopy interception of the sticky NH_3 molecules the grid was setup on an asphalt strip, located downwind of grassland with 15 cm canopy (Fig.1a). Up- and downwind 1min-averages of line-integrated concentrations were measured by open-path DOAS (Volten et al., 2012; modified). Impinger (64712-U Supelco) acid traps ('Loci') were distributed downwind of the grid, sampling with 1h time resolution. Concentration measurements were done at heights between 0.8 and 1.2m. Vertical profiles of wind-velocity () and turbulence were recorded by ultrasonic anemometers. NH_3 was released over 2h periods during daytime and morning. WT was run forward with the parameters u_*, L, z_0, σu, σv, σw, and wind direction measured at 1m height. Modelled 10min concentration to emission ratios were averaged over the respective measurement intervals in order to compare measured to expected concentrations (C/C_{WT}).

Results and Discussion

The vertical profile measured over the asphalt deviated from the MOST profile derived from 1m due to mixed internal boundary layers from grass and asphalt over the asphalt strip. MOST profile (used by WT) showed lower at heights below 1m. The WT input was modified to account for the altered profile by adjusting z_0 (minimising the differences in MOST and measured profile below 1m). Resulting MOST profiles underestimated near-ground less, but overestimated above 1m. z_0 decreased from about 1 to 0.5cm – a value more reasonable for the asphalt. Over a range of atmospheric conditions, DOAS derived emissions recovered 88 to 104% whereas the point sensor derived emissions recovered 73 to 105% of the released NH_3 (Tab.1). Loci3, located 10m downwind, showed 105 recovery vs. 74% of Loci1/2 since the emission plume was more homogeneously distributed with increasing distance (Flesch et al., 2004). The DOAS recoveries were better due to the line concentrations seeing a more representative spatial average over the plume. Underestimation could be explained by deviations of actual near ground profile from MOST profiles derived from measurements at 1m height. Deviations were most pronounced towards higher (Fig.1b). Generally, WT performance depends on (z-d)/L, measurement height, and u_* (Harper et al., 2011). Alternatively, vertical profile considerations can serve as quality indicator (Flesch et al., 2013). Here, we have applied WT in a situation with complex terrain and with one of the most challenging trace gases to measure. By considering vertical profile consistency the reported recovery ratios indicate an acceptable WT accuracy for NH_3 emission measurements.

Tab. 1: Meteorological conditions and concentration recovery ratios (C/C_{WT} = 1 means 100% recovery).

ID	NH_3 mix. ratio (%)	$\bar{u}$ (m/s)	(z-d)/L	DOAS avg. ± sd C/C_{WT}	Loci1 avg. ± sd C/C_{WT}	Loci2 avg. ± sd C/C_{WT}	Loci3 avg. ± sd C/CWT
1	5.04	3.7	-0.04	0.88 ± 0.05	0.75 ± 0.03	0.73 ± 0.02	1.05 ± 0.18
2a	5.04	3.2	0.01	0.88 ± 0.05	0.78 ± 0.03	0.75 ± 0.01	1.05 ± 0.27
2b	2.005	1.4	0.12	1.04 ± 0.09			
3	2.005	0.3	-2.44				
4	2.005	1.8	-0.29	1.02 ± 0.11	0.88 ± 0.11	0.78 ± 0.24	

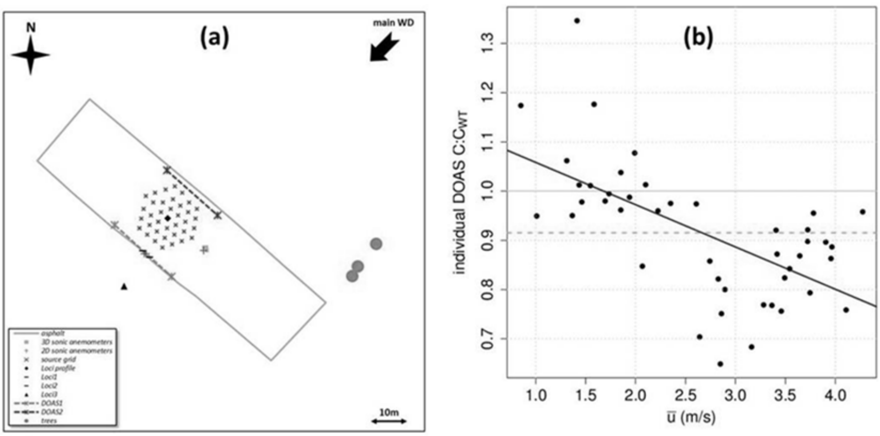

Fig. 1: (a): Experimental setup of the release experiment; (b): DOAS C/CWT vs. $\bar{u}$.

Conclusions

Conducting NH_3 release experiments downwind a 20m diameter release grid under non-ideal conditions, we show that WT dispersion modelling performs well (within approx. 75 to 105% recovery) for such a setup, given a reasonable MOST representation of vertical profile and considering actual turbulence.

Acknowledgement

We thank the staff from Payerne airport for their help and the Swiss Federal Office for the Environment for supporting this study.

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LINEAR AND NON-LINEAR RESPONSES OF NITROUS OXIDE EMISSIONS TO FERTILISER NITROGEN RATE AND THEIR IMPACT ON EMISSION INTENSITIES OF ARABLE CROPS

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Nitrogen (N) availability remains one of the key drivers of crop productivity from arable farms of Northern Europe. Most crops receive significant amounts of inorganic N fertilisers but these are also associated with significant losses of the greenhouse gas (GHG) nitrous oxide (N2O). National GHG inventories and current commercial GHG accounting procedures assume a direct proportionality between N fertiliser use and N2O emissions from land (Fig.1a). This assumption implies that drastic reductions in N fertiliser use and crop productivity are required to minimise N2O intensities of crop products (kg N2O-N per kg product). However, we hypothesise that the response of annual N2O emissions to N supply is, to some extent, related to the surplus of N supply over crop N uptake (Fig. 1a). If so, fertiliser N application strategies to minimise N2O emission intensities of crop products may have much less severe implications for crop productivity (Fig. 1b). If N_2O emissions are entirely N-balance related, N amounts that minimise N2O intensities would be similar to current use, with little effect on crop productivity. The ongoing research described here is assessing the shapes of the responses in annual N2O emissions to increasing amounts of applied N for UK arable crops.

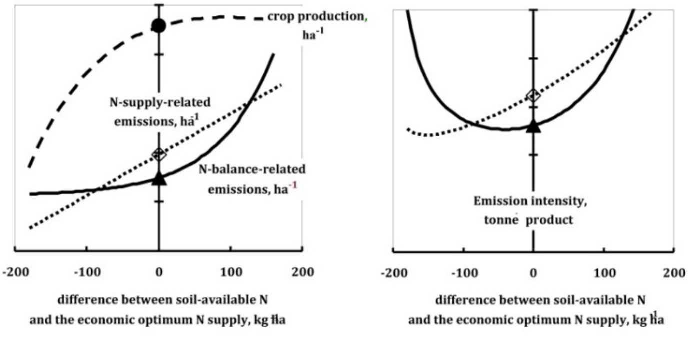


Figure. 1. Modelled effects of N supply on (a) crop production (circle), and on N_2O emissions if related directly to N supply (diamond; as estimated by IPCC Tier 1 approach) or to the balance between N supply and crop N uptake (triangle; as hypothesised here), and (b) consequent contrasting effects of these scenarios on N_2O emission-intensities of crop products.

Materials and Methods

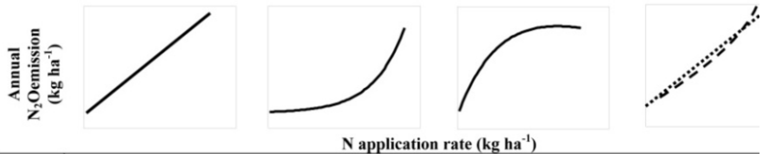
Across 3 years (2010, 2011 and 2012), 24 field experiments have been conducted across four UK sites: 1 & 2) South East England 3) Central England and 4) Central Scotland, covering a range of soil textural classes: from sand to clay. Nitrous oxide emissions (5 static chambers/plot) were monitored from replicated (x3) plots for 12 months, following spring ammonium nitrate fertiliser applications in the range nil to 240% of recommended

N. Up to three split fertiliser applications were made in order to achieve target rates. Nitrous oxide was intensively sampled in the first 2 weeks after each fertiliser application, decreasing in frequency to monthly sampling towards the end of the 12 months. On each sampling date soil mineral nitrogen (SMN) and gravimetric moisture was measured (0-10 cm) from every plot. The crops studied were winter wheat (all sites), winter oilseed rape (sites 1, 3&4), spring barley (sites 3&4), sugar beet (sites 2&3) and winter barley (sites 2, 3&4). Regression analysis was used to assess, for each sampling date, when cumulative N_2O, daily N_2O and SMN had a significant ($P< 0.05$) linear or non-linear (quadratic) response to N rate.

Results and Discussion

The weather in spring contrasted between experimental years, being very dry in 2010 & 2011 and much wetter than average in April 2012; allowing our hypothesis to be tested against extremes in UK arable conditions. All data will be presented, but taking the example of experiments conducted in England, 2010 and 2011 showed mostly a linear response of annual N_2O emission to N application rate (Tab 1). The lack of non-linear responses is surprising given that the generally dry spring conditions inhibited emissions soon after N application so that most peak emissions occurred after the main phase of crop N uptake. For experiments conducted in England in 2012, 3 out of 5 had non-linear annual N_2O responses (Tab 1) and the largest daily N_2O emissions generally occurred within 3 weeks following N application after fertiliser was applied to soils with high moisture contents. Regression analysis on each sampling date identified whether cumulative and daily emissions had linear or non-linear responses to N rate, and hence the sampling events that determined the final shape of the annual N_2O responses.

Table. 1. The frequency of significant (P<0.05) linear and non-linear (quadratic) responses of annual N_2O to N-rate, examples from arable field experiments conducted in England for harvest years 2010, 2011 and 2012. Dotted lines represent where linear and quadratic equations explain an equal percent of variation in the data.



Year	Frequency of N_2O response			
2010	4	1	-	-
2011	3	1	1	-
2012	1	3	-	1

Conclusion

Responses of N2O emission to applied N were affected in both magnitude and timing by weather conditions around the time of N-application, which spanned the extremes of likely UK spring conditions. Response shapes of N_2O emission to N-rate did not support our 'N-balance' hypothesis; more non-linear responses occurred in 2012 when emissions peaked soon after fertiliser application (before or during the main phase of crop N uptake) whereas more linear responses occurred after dry springs (when N_2O emissions tended to be delayed until after crop N uptake).

Acknowledgements

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EFFECT OF PIG SLURRY ACIDIFICATION AND FORM SOIL APPLICATION IN N APPARENT LEACHING UNDER FIELD CONDITIONS

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The soil application of the slurry technique is, usually, accomplished by important gaseous losses of N, reducing the efficiency nutrient application. Acidification or manure soil incorporation are strategies used in reducing gaseous N losses. In order to make these strategies more environmental compatibles, the respective potential dimension effects on the processes such N soil lixiviation is an obligate task.

For this purpose, a field incubation experiment without plants carried out during a 140 days period. Five treatments, with an equivalent of 80 kg of N per hectare in form of pig manure, were considered: (i) slurry non-acidified (pH 8.2) and surface applied (T1); (ii) slurry acidified (pH 5.5) and surface applied (T2); (iii) slurry non-acidified (pH 8.2) and buried (T3); (iv) slurry liquid-fraction non-acidified (pH 8.2) and surface applied (T4); and (v) slurry liquid-fraction acidified (pH 5.5) and surface applied (T5). A sixth treatment was also considered, in which no manures applications were performed, serving as control (T6). For each treatment, three samples data was performed, relative to 56, 84 and 140 incubation days. For each sample date performed four repetitions reactors per treatment were collected, destroyed and the N retained by ion exchange resins analyzed. The apparent N leaching was determined based in N retained by ion exchange resins in treatment with organic manure minus the control, and the results expressed in mg per g of N added.

For all sample date performed, the results revealed significant differences ($p<0.05$) between all treatments studied.

The more significant higher apparent leaching values were register in treatments subject to acidification, as T5 (412.3 mg/g N) and T2 (393.9 mg N/g N), or soil incorporation, as T3 (195.8 mg N/g N). Apparently, these results reveals that treatments for more effective technique in reducing N gaseous losses increase, simultaneous, the mineral N soil content and subsequently, the N availability for plants and/or for N losses by leaching.

NITROUS OXIDE EMISSIONS FROM CLOVER RICH LEYS DURING THE LONG NORTHERN WINTER

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In boreal grasslands, a significant amount of the perennial above-ground biomass decays throughout winter. We found that white clover leaves lost between 57 and 74% of their foliar nitrogen (N) between autumn and following spring (Sturite et al., 2006). Recovery on N in seepage/melt water was found to be small (19-40%; Sturite et al., 2007) raising the question about the fate of the remainder. Mørkved et al. (2006) showed that the water soluble fraction of freeze-thaw treated white clover had a great potential to trigger N_2O emissions in laboratory incubations. Winter emissions are an important component of the annual N_2O budget (e.g. Flessa et al., 1995), but little is known about how the management of perennials affects these emissions. The objectives of the present study were to quantify N_2O emissions from grass-clover stands in two grasslands of coastal Norway. We hypothesized that off-season N_2O emissions would be higher from clover-rich than pure grass stands because of higher foliar N content in clover, and that removal of herbage at the end of the growing season would reduce winter emissions.

Materials and Methods

Field trials were established in spring 2010 on mineral soils at at Tjøtta (65°49'N, 12°25'E) and Fureneset (61°22'N, 5 °24'E). Both sites have a costal climate with mild winters and frequent freezing-thawing. Three seed mixtures with 0, 30, and 100% clover were established in a complete randomized split-plot design. The mixture contained both red clover (*T. pratense* L.), white clover (*T. repens* L.) and various grasses. All plots received 110 kg N ha^{-1} cattle slurry in spring and were harvested twice a year. Periodic N_2O flux measurements were carried out manually throughout two winters from Oct. 2011 to May 2012 (Period I) and from Oct. 2012 to end of April 2013 (Period II). Two subplots of 1 m^2 each were established within each treatment plot in autumn, one remained undisturbed (U), whereas above-ground herbage was removed (R) in the other. In each subplot one aluminum frame (52 cm x 52 cm) with a water lock was installed serving as a base for 20-cm high vented aluminium chambers. For each flux measurement, four 15 ml samples were withdrawn from the chamber (at 0, 15, 30, 45 min) and transferred to preevacuated 10 ml septum vials for GC-analysis. Sampling frequency varied depending on freeze-thaw cycles. Plot-wise cumulative N_2O emissions (kg N ha^{-1}) were calculated by linear interpolation between sampling dates and averaged for each treatment.

Results and Discussion

Winter was milder in Period I, with little snow cover and only a brief period of soil frost. In Period II, low air temperatures in December without snow cover resulted in deep soil frost particularly at the northern site "Tjøtta" (data is not shown). On average for both sites and periods, cumulative N_2O losses were significantly greater from pure clover stands and clover-grass mixtures than from pure grass stands (Fig. 1). This effect was more pronounced at the southern site, probably because of the lower soil pH (5.6) as compared with the northern site with soil overlaying shell sand deposits (pH 6.5). Soil pH has been shown to greatly affect N_2O emission potentials (Liu et al. 2010). Overall greater emissions were observed in Period II, confirming that the severity of winter freezing controls N_2O emissions (Teepe et al. 2000). Removal of herbage at the end of the growing season had no consistent effect on cumulative N_2O emission, but there was a trend of higher emissions in grass-clover stands with removed herbage (Fig. 1). This suggests that frost damage of roots and nodules may play a role for off-season N_2O emissions. N-leakage from roots under severe winter conditions has been previously reported by Tierney et al. (2001) and Sturite et al. (2006).

Conclusions

We present experimental evidence that clover in grasslands promotes off-season N_2O emissions under northern winter conditions regardless of inter-annual variation in winter climate. Removal of above-ground herbage did not reduce N_2O emissions.

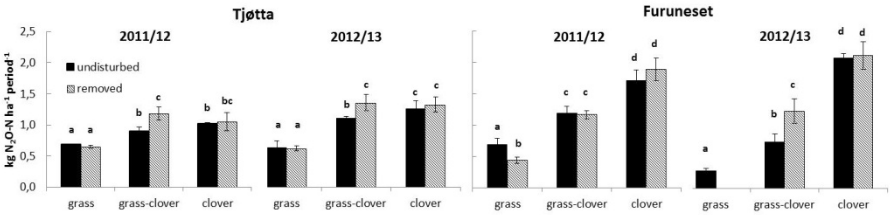

Figure 1. Mean cumulative N_2O emissions in grass, grass-clover, and pure clover stands at Tjøtta and Furuneset throughout the winter period (Oct. – April) 2011/12 and 2012/13. Error bars denote SEM. No treatments with removed grass were measured at Furuneset in 2012/13. n = 4 in 2011/12 and n = 3 in 2012/13. Different letters denote statistically significant differences in cumulative N_2O emission at each site and year ($p < 0.05$)

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EFFECT OF A NITRIFICATION INHIBITOR ON N2O EMISSIONS AFTER MINERAL FERTILIZER APPLICATION ON A SANDY LOAM SOIL

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Fluxes of greenhouse gas nitrous oxide (N2O) are variable according to the specific site conditions. Mineral fertilizer N input can increase the potential for N2O emissions of an agricultural area (Stehfest & Bouwman, 2006). According to IPCC (2006), losses of N2O from mineral N fertilizers were assessed by a uniform emission factor for all N fertilizer types. However, there is evidence that the use of a nitrification inhibitor (NI) can decrease the N2O emissions after mineral N fertilizer input (Akiyama et al., 2010). The aim of our study was to quantify the effect of different N types and the reduction potential of a NI on N2O emissions in the period between end of March up to harvest in 2011-2013 after mineral fertilizer input on a sandy loam soil cultivated with winter wheat.

Materials and Methods

In a fully randomized experiment N2O flux measurements, using closed chamber methode, were conducted in four treatments: control (without N), CAN, Urea (PIAGRAN® 46) and Urea+NI (nitrification inhibitor DCD/TZ, ALZON® 46). The experimental site is characterized as follows: 51° 21'N, 12°33'E, 140 m a.s.l., mean annual precipitation 614 mm, mean annual temperature 9.5 °C, soil: 3-6 dm sandy loam deposits overlying boulder clay, Stagnic Gleysols and Stagnic Podzoluvisols. These soil conditions represent 50 % of arable land of Saxony. N2O emissions were measured in a field experiment during the growing season of winter wheat and in a subsequent model experiment with constructed loss conditions each year (2011-2013). In the field experiment, fertilizer input (2011: 200 kg N ha-1; 2012, 2013: 220 kg N ha-1) was split into three (CAN, Urea) or two applications (Urea+NI), respectively. Measurements were conducted weekly and additionally event-oriented after fertilizer application or rainfall. After harvesting yield parameters were analyzed. In the following model experiment, loss conditions (100 kg fertilizer N ha-1, fresh organic matter input by stubble tillage, additional irrigation) were constructed to quantify the N2O potential of the CAN, Urea and Urea+NI (2011, 2013: DCD/TZ, 2012: P 70/05). Soil moisture (0-10 cm) was measured gravimetrically and water filled pore space (WFPS) was calculated. The N2O emissions were analyzed with the tukey test (HSD; $P < 0.05$)

Results and Discussion

During the growing season (from March up to harvest) very low N2O emissions occurred in the fertilized plots in 2011 und 2012, also immediately after N fertilization. In contrast, in 2013 relevant N2O peaks were measured because of wet conditions during the growing season (Figure 1). Only in 2013 a strong correlation of N2O emissions and WFPS in all fertilizer treatments was found. Differences between the fertilizers in the cumulative N2O emissions were not statistically significant in the field experiments of all three years. Nevertheless, the treatment Urea+NI tends to result in lower emissions compared to Urea in all three years and compared to CAN (2011, 2012). High variability of the N2O measurements (n=4) at the different measuring times indicate the strong impact of hot spots and hot events on N2O fluxes. Both, N2O emission losses during the main vegetation period and yields showed no significant difference between the tested fertilizer treatments. Before and after all measurement campaigns, equal mineral N contents were recorded in the 0-30 cm soil layer of all treatments. The N2O emissions in the model experiment were considerably higher than in the field experiment (Figure 1). Due to the prolonged nitrification, the NI caused a statistically significant reduction of N2O emissions compared to CAN in 2011 and 2012. Compared to Urea the treatment Urea+NI tends to result in lower cumulative N2O emissions in all three years. In 2013 the differences between all treatments were not significant.

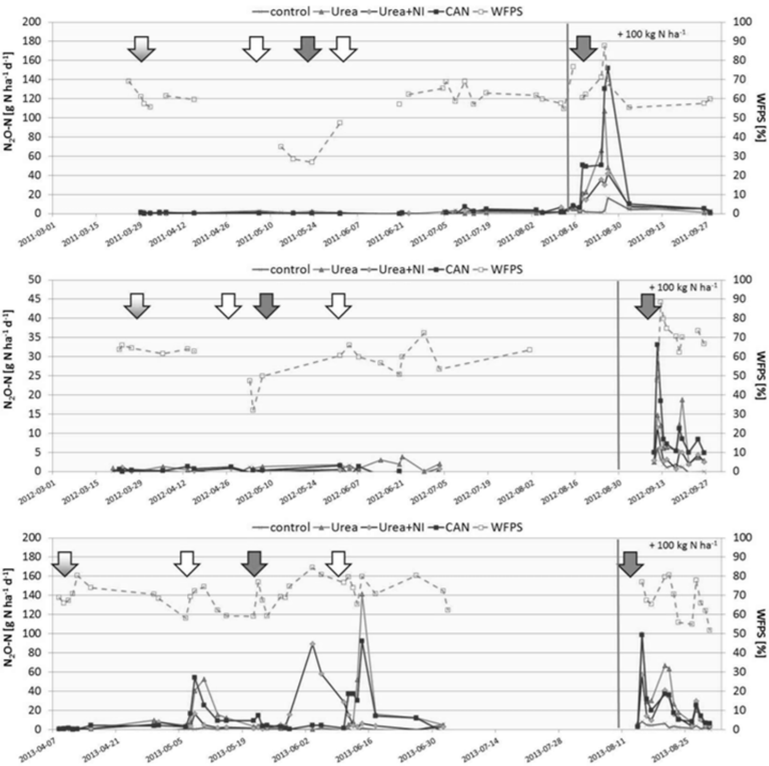

Figure 1. mean N2O emissions (n=4) during the field and model experiment in 2011, 2012 and 2013: white arrow indicates fertilizer input CAN/Urea; green arrow indicates fertilizer input Urea+NI; brown arrow indicates fertilizer input model experiment

Conclusions

The N2O emission level on the investigated site was very low in 2011 and 2012. Only in 2013 relevant N2O peaks were measured within the field experiment rather determined by weather and soil water content than by the provided N types. In order to analyze the N2O potential of different N fertilizers, limited measurement periods (growing season up to harvesting) seem to be sufficient whereas total N2O losses from the cropping system cannot be deduced. Within the model experiment the potential of NI treatments could be demonstrated. Under loss conditions Urea showed less N2O emissions compared to CAN in two of three years. This effect was significant when a NI was applied. The application of Urea+NI seems to be a promising approach to reduce N2O losses from inorganic fertilizer N.

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NITRIFICATION INHIBITOR AND NITROGEN FERTILISER APPLICATION TIMING STRATEGIES TO REDUCE NITROUS OXIDE EMISSIONS FROM WINTER WHEAT LAND

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Agriculture in the UK is expected to have an increasingly important role in contributing to feeding a growing global population, whilst minimising climate change and other environmental impacts. The use of new and modified nitrogen (N) fertiliser strategies are potential changes in agricultural management which could contribute to reductions in greenhouse gas (GHG) emission. Nitrification inhibitors (NIs) have the potential to reduce *direct* nitrous oxide (N_2O) emissions from soil following N application. Modifications to fertiliser N application timing strategies also have the potential to reduce *direct* N_2O emissions from soil; more frequent application of smaller amounts of fertiliser N in close synchrony with crop requirements would minimise soil mineral N levels at risk from elevated N_2O losses at any point in time, including those when soil conditions are conducive to high N_2O emissions e.g. when soils are 'warm and wet'. The use of NIs and changes to fertiliser N application timings may not only potentially reduce *direct* N_2O emissions, but they may also influence crop yields and N recovery, as well as ammonia (NH_3) and nitrate (NO_3) losses with consequential effects on *indirect* N_2O emissions.

Materials and Methods

On a commercial arable farm near Cambridge, eastern England (clay loam topsoil texture), *direct* N_2O-N and NH_3-N emissions were measured from replicated (x3) plots (24 x 11 m), following spring applications of manufactured N fertilisers to winter wheat. A control treatment was included where no N fertiliser was applied. Ammonium nitrate (AN) fertiliser (34.5% N), urea fertiliser (46% N) or an ammonium sulphate nitrate (ASN) fertiliser (26% N) were applied at a rate of 200 kg N/ha, in three split applications (mid-March, mid-April & early May). In separate treatments, two commercially available NIs were tested; dicyandiamide (DCD) was sprayed onto the AN and urea plots immediately after N fertiliser application, and a 3,4-dimethylpyrazole phosphate (DMPP) containing fertiliser (26% N) was also applied at a rate of 200 kg N/ha, in three split applications. The N supplied by the DCD was accounted for in the 200 kg N/ha application. Additionally, AN and urea fertiliser was also applied in five equal splits in mid-March, late March, mid-April, late April & early May i.e. 'little & often'. Following N fertiliser application, measurements of *direct* N_2O-N were made over *c*.12 months, using 5 static chambers (0.8 m^2 total surface area) per plot and analysed by gas chromatography. Ammonia-N emissions were measured for *c*.3 weeks after each split N fertiliser application, using a wind tunnel technique (one per plot). *Indirect* N_2O-N emissions were estimated from the measured NH_3-N losses and using the Intergovernmental Panel on Climate Change default emission factor, namely: 1% of volatilised N is lost as N_2O-N. As N fertiliser was applied in the spring and there were no large rainfall volumes in early spring, no *indirect* N_2O-N losses were assessed to occur via NO_3 leaching. Grain yields and total N offtakes were measured following harvest in August 2012.

Results and Discussion

The application of DCD reduced (P<0.05) *direct* N_2O-N losses from urea fertiliser by 93%, but had no effect (P>0.05) from AN fertiliser, compared with the respective unamended 3-split fertiliser application treatments (Fig. 1). There was also no effect (P>0.05) of the DMPP ASN containing fertiliser on *direct* N_2O-N emissions compared with unamended ASN. The difference between the inhibition efficiencies of the NIs is likely to be related to soil conditions, N fertiliser form and the resulting N_2O production processes. Ammonia emissions, total N offtakes and grain yields were not affected (P>0.05) by either the DCD or DMPP.

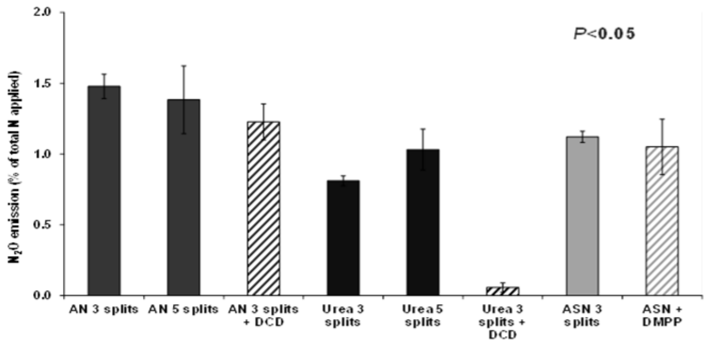

Figure 1. Annual nitrous oxide (N_2O-N) emissions following ammonium nitrate (AN) and urea (3- or 5- splits) with and without the nitrification inhibitor DCD, and ammonium sulphate nitrate (ASN) fertiliser application (3- splits) with and without the nitrification inhibitor DMPP. Error bars represent ± one standard error of the mean

There was no effect (P>0.05) on *direct* N_2O-N emissions of changing the N fertiliser timing application strategy from the conventional 3-split to the ‘little & often’ 5-split treatment (Fig. 1). As a result of differences in rainfall and temperature following the contrasting N application timings, there were greater (P<0.05) NH_3 losses and consequently greater (P<0.05) *indirect* N_2O-N emissions from the urea 3-split compared with the urea 5-split treatment. This difference in indirect emissions was not, however, large enough to affect *total* N_2O-N losses (P>0.05). Total N offtakes and grain yields were also not affected (P>0.05) by changes in N fertiliser timing. *Direct* N_2O-N losses were greater (P<0.05) from the conventional AN 3-split fertiliser treatment (1.48% total-N applied) than from the urea 3-split treatment (0.81% total-N applied), Fig. 1. This difference was probably due to a combination of NH_3 loss and soil conditions after application. There were much higher (P<0.05) NH_3-N emissions from the urea 3-split treatment (24% total-N applied) than from the AN 3-split treatment (<1% total-N applied), which would have depleted the soil mineral N pool available for N_2O production. When *direct* N_2O-N emissions were expressed as a percentage of total N applied remaining after NH_3 loss, there were greater (P<0.05) *direct* N_2O-N emissions from the AN 3-split treatment (1.58%) than from the urea 3-split treatment (1.06%); which suggests that it was not just the difference in NH_3 loss that led to greater *direct* N_2O losses from AN fertiliser compared with urea. Soil conditions were also a major factor in the difference in emissions between AN and urea; following 75 mm of rain over 14 days in late April, ‘wet’ soil conditions would have encouraged N_2O production via denitrification of the NO_3-N in the AN fertiliser. *Indirect* N_2O-N emissions from the urea 3-split treatment (0.24% total-N applied) were greater ($P<0.05$) than from the AN 3-split treatment (<0.01% total-N applied), however, this was not enough to offset the greater *direct* N_2O losses from AN, such that *total* N_2O-N emissions were greater ($P<0.05$) from the AN 3-split treatment than from the urea 3-split treatment. Total N offtakes and grain yields were also not affected ($P>0.05$) by N fertiliser type.

Conclusion

These data indicate that fertiliser choice and NI use has the potential to substantially reduce *direct* N_2O-N emissions whilst maintaining productivity, whereas the ‘little & often’ fertiliser application strategy may not. The efficiency of the NI is, however, likely to depend on the NI and fertiliser type, as well as soil conditions following fertiliser application.

Acknowledgements

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REDUCING EXTERNAL NITROGEN COSTS OF PIG PRODUCTION IN THE EUROPEAN UNION BY SPATIAL RELOCATION

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The average consumption of pork products in the European Union (EU27) has increased from 15 kg per head in 1960 to nearly 25 kg currently, and has become by far the most popular meat type. For various economic reasons, pig production is concentrated in distinct regions, close to urban consumers and to harbours to import the feed. However, the downside of this is a concentration of external effects related to emissions from farms, fields and transport causing diverse effects on health and nature, and nuisance for local residents related to odour and landscape. This paper explores the question whether relocation of pig production can contribute to lessening external cost of nitrogen pollution. Considered external costs are those from emissions of ammonia to air and of reactive nitrogen to soil and water related to manure production, storage and application, and from emissions of NO_x from transport. The resulting set of optimal regions for allocation additional pig production will be reviewed regarding the consequences for the production costs and nutrient use efficiencies.

Material and methods

We developed a procedure to calculate the external nitrogen costs of an increase of pork production as a function of production location in the EU27 at NUTS2 level using the MITERRA model (Velthof et al., 2009). The location where external costs are the least will be the location where from an environmental point of view it is most beneficial to allocate additional pig production. The external cost of emissions were monetized using unit costs per unit of emission per impact (Van Grinsven et al., 2013).

Results and Discussion (provisonal)

Although model calculations are still in progress, we already have some provisional results. As a first test of our line of thought we relocated 1 million pigs from the NUTS region Noord-Brabant in the southern part of the Netherlands to the NUTS regions Sachsen (region in former East Germany). Vest in the east of Romania and Toscane in northern Italy and calculated the total change in damage (Figure 1).

NUTS2 regions Noord-Brabant, Sachsen, Vest and Toscane are chosen because of the following reasons: *Noord-Brabant* developed the highest pig density in the EU27, because it is close to sea harbours and population concentrations in the Netherlands and Germany. Noord-Brabant is also a region with high exceedance of nitrogen of nutrient nitrogen critical loads, and high external cost due to emission of ammonia and nitrate. *Sachsen:* moderate exceedance, near population concentrations, sea harbours further away. *Vest:* no or low exceedance, low population density, high cereal production potential, harbours are far away. *Toscane:* low to moderate exceedance, close to large Italian cities and relatively close to harbours.

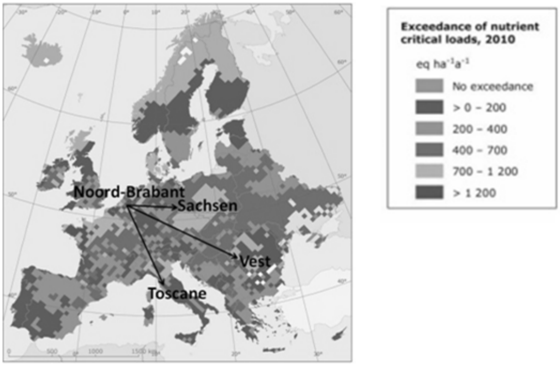

Figure 1 Exceedance of Nitrogen critical loads due to nutrient nitrogen in 2010 (Coordination Centre for Effects, 2009) with the regions Noord-Brabant, Sachsen, Vest and Toscane.

To calculate the net external costs of relocation three scenario's are distinguished□1 Current emission factors and identical cost per unit in all regions.□2 Improved emission factors in pig receiving regions at current values in Noord-Brabant (low emission housing and low emission manure application).□3 Differentiated unit cost per emission in receiving region to account for lower (GDP dependent) willingness-to-pay and lower base line emission levels and exceedance of critical loads. Relocation of pig production reduces costs in Noord-Brabant because of a reduction of ammonia emissions from housing. External costs in the rest of the Netherlands decrease because of reduced ammonia emission from application and because of reduced N surpluses. Currently about 45 million kg N in manure in Noord-Brabant is exported to other Dutch provinces. Relocation of 3 million pigs would bring an end to this export, saving pig farmers manure disposal costs a about 5 euro/ton (3 euro/kg N). Arable farmers previously receiving manure N would have to replace part of this N by synthetic fertilizer, at a considerable cost. In view of the lower fertilizer equivalency of manure N, this would reduce the N surplus and external cost related to N emission to groundwater and surface water in other Dutch provinces.

Preliminary results for scenario 2.

	Characteristics	Before relocation	After relocation		
	Agricultural Area (1000 ha)	Grassland Area (1000 ha)	Number pigs (x 1000)	Number pigs (x 1000)	Change external cost (million euro)
Noord-Brabant	219	49	5236	2236	-37
Rest of Netherlands	1912	794	12026	9026	-41
Germany, Sachsen	900	182	616	1616	34
Romania, Vest	1607	532	831	1831	28
Italy, Toscane	725	83	102	1102	36

Preliminary conclusion□In scenario 2 external costs in the Netherlands are reduced considerably, but overall the sum of external N costs in all regions involved increases. Relocation of pigs leads to a net reduction of external N costs only when taking into account the larger gap between projected loads and critical loads of ammonia and nitrate in the pig receiving regions as compared to the Netherlands. This would imply threshold values below which increase of emission would not create additional extern cost. Proper judgement of the benefits and desirability for the EU27 of relocation of pig production would also require insight in the direct economic implications at several levels, and external costs other than those related to nitrogen emissions.

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IMPACTS OF LAND-BASED NUTRIENT LOSSES TO THE MARINE ECOSYSTEM

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Human life depends on the use of reactive nitrogen for fertilization of agricultural land as approximately half of the earth population is fed due to mineral fertiliser. While about 130 Tg of nitrogen are introduced per year as synthetic fertiliser (artificially fixed in the Haber-Bosch process, (Fowler et al. 2013), only 30 Tg y^{-1} are consumed by humans in food (Billen et al., 2013). The rest travels through air, soil, water and has tremendously negative effects on the environment. Although a major percentage of the nitrogen traveling through watersheds is lost during the passage to the ocean, the amount of reactive nitrogen reaching the sea is estimated to be around 65 Tg y^{-1}. Coastal oceans are impacted by these nutrient sources. Pathways of transport to the ocean are the rivers and the atmosphere. As soon as the nutrients enter the coastal ocean they support plankton growth, reduce water transparency and oxygen concentration. The development of hypoxic zones may follow. In many European countries efforts to reduce the nutrient input were at least partly successful so the nutrient concentrations in surface waters decrease e.g. in the Baltic Sea (Voss et al., 2011). Nevertheless a severe eutrophication problem in coastal areas exists which is driven by nitrogen and also by phosphorous. Moreover, the decrease of silica input to the sea, as a result of human activities on land, plays a major role for the composition of phytoplankton populations. We are thus facing a multifactorial problem of nutrient concentrations and nutrient ratios which impact the oxygen concentrations and the biogeochemical processes in water and sediment of coastal seas.

Two aspects are of particular concern considering the current trend of nutrient loss to the marine ecosystem: the impact of oxygen stress on the biophysical processes and the consequence of unbalanced nutrient input on the oceanic cycles. These two aspects will be presented and discussed.The presentation is based upon studies of the authors and literature data. Estimates of the nitrogen and phosphorus export to European seas will be presented. The concept of marine nutrient stoichiometry will be explained as well as the effects of the deviation from an ideal nutrient ratio on the plankton community. Moreover, the role of oxygen for the nutrient cycling will be discussed.

Between 1985 and 2005, the nutrient input to European coastal zones has been 4.1-4.8 Tg N y^{-1} and 0.2-0.3Tg P y^{-1} (Grizzetti et al., 2012). Except for the North Sea and part of the Baltic Sea, the annual nutrient load have not changed significantly, but the N:P ratio has been steadily increasing, probably due to effective measures of phosphorus reduction combined with less successful strategies to reduce nitrogen. This may have a significant effect on the N-retention in estuaries. A study from the Elbe River Estuary could not find a significant decrease in nitrate concentration as expected, they rather report a high nitrification. This is in accordance to an experiment with water from a small river system in the US, where different N:P ratios were applied. Results suggest a convincing chain of processes supported by the high N:P ratios of the nutrients (Lunau et al., 2012). Accordingly, surplus nitrate is consumed by the phytoplankton community, however it is not used for biomass buildup but instead converted to dissolved organic material (DOM). The DOM is consumed by bacteria that generate ammonium which is immediately nitrified again. Nitrate thus remains in the system creating higher and higher N:P ratios. Removal processes play a minor role in such systems. Coastal sediments are known for their high capacity to reduce nutrient loads from land by denitrification. However, this process relies on oxygen in the overlying water where ammonium is nitrified and serves as a substrate. Under conditions of frequent or prolonged anoxia denitrification is reduced. Moreover, the sediments are no longer sites of denitrification but the process is shifted to the water column where denitrification may be expressed at lower rates. If conditions turn worse in the sense that oxygen lacks also in the bottom waters, then ammonium refluxes from sediments into the water column where they support plankton growth and in turn oxygen consumption. A vicious cycle develops and in consequence, most benthic animals die, fish habitats are lost and the recreational value of coastal areas is gone. In summary, unbalanced nutrient inputs lead to a faster recycling, and more land derived nutrients may arrive in the open ocean with unforeseen consequences for the oceanic element cycles.

This evidence suggests that land-based nutrients may have a lasting impact on the marine ecosystem, and also shows that policies affecting the river basin influence coastal and marine ecosystem conditions and services. For this reason nutrient reduction must remain high on the political agenda. Especially the reduction of nitrate in waters to prevent coastal eutrophication is of great importance.

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N MINERALISATION, DENITRIFICATION AND N2O FLUXES IN A HISTIC GLEYSOL FOLLOWING DIFFERENT GRASSLAND RENOVATION TECHNIQUES IN NORTHWEST GERMANY

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Grassland renovation by reseeding is a common practice to improve productivity, but knowledge on enhanced nitrate leaching and N_2O emission due to disturbance during associated soil tillage is scarce. Denitrification in hydromorphic soils under agricultural management is potentially extremely high due to the coincidence of high nitrate concentrations, labile organic carbon and oxygen depletion during extended periods of water saturation close to the soil surface (Well et al. 2003, Well et al. 2005). We investigate the impact of grassland renewal or conversion to arable land on greenhouse gas fluxes and N budget. One of our two sites is a hydromorphic soil rich in organic C, with groundwater level always within the rooting zone and close to the surface during winter. Assessment of the N budget to estimate enhanced N mineralization following grassland renewal as well as associated N leaching is complicated by potentially complete NO_3^- consumption via denitrification. Robust estimation on denitrification losses at this site is crucial to assess the impact of grassland renewal on its N dynamics and budget. One aim of this study is to determine denitrification in the surface and subsoil in order to close the N budget.

Materials and Methods

A field plot comparing different techniques of grassland renewal is conducted on a Histic Gleysol located near Oldenburg, Lower-Saxony, Germany. In addition to weekly N_2O and CH_4 flux measurements we apply five approaches to investigate spatial and temporal dynamics of denitrification:

(1) N balance approach: The N budget is obtained by weekly measurement of N_2O fluxes and mineral N in the top soil, mineral N twice a year at 0 to 90 cm depth, N uptake, N fertilization and modelling N leaching based on mineral N and hydrological model data. Unaccounted N is attributed to possible denitrification.

(2) Isotopologue approach: $d^{18}O$, average $d^{15}N$ and ^{15}N site preference of N_2O as well as $d^{15}N$ and $d^{18}O$ of NO_3^- are measured at times to estimate N_2O reduction to N_2 in the topsoil during periods of unsaturated conditions using the N_2O isotope fractionation approach (Lewicka-Szczebak et al., 2014).

(3) ^{15}N gas flux method: N_2 and N_2O fluxes from denitrification will be measured periodically by in situ ^{15}N-labelling of soil mesocosms and analysing ^{15}N enrichment of emitted denitrification products (Lewicka-Szczebak et al., 2013). This is to validate results of the isotope fractionation approach.

(4) Excess-N_2 method for groundwater: During winter, dissolved N_2 and Argon are analysed to determine excess-N_2 from denitrification (Blicher-Matthiesen et al., 1999; Weymann et al., 2008). Loss of dissolved N_2 by diffusion will be estimated by modelling (Well et al., 2001).

(5) Empirical functions: Denitrification during phases of water-saturation is modelled based on groundwater level data, organic C, C/N ratio, texture and pH using

regression functions for potential denitrification in hydromorphic soils (Well et al., 2003, Well et al, 2005).

Results and Discussion

During fall of 2013, we observed increased mineral N and N_2O in grassland renovation treatments after tillage and reseeding when compared to the control. High soil moisture and isotopic fingerprints of emitted N_2O suggest denitrification as the dominant N_2O producing process. In view of ongoing high water contents during winter with groundwater levels close to the surface we expect that NO_3^- accumulated during fall will decline due to denitrification while N_2O fluxes might also decline due to N limitation and because inhibited diffusive exchange in the high groundwater will favour N_2O reduction to N_2. Due to the latter, we expect that we will detect excess-N_2 in the water-saturated soil layers and can use these data to differentiate between NO_3^- lost by leaching and denitrification.

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BANDSPREAD SLURRY APPLICATIONS TO GRASSLAND: EFFECTS ON THE BALANCE OF AMMONIA AND NITROUS OXIDE EMISSIONS TO AIR AND SLURRY NITROGEN USE EFFICIENCY

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Reducing nitrogen (N) losses (ammonia and nitrous oxide to air and nitrate to water) following slurry application is important to minimise the environmental impact of livestock farming systems**.** On grassland, bandspread slurry application (e.g. using a trailing shoe) can be effective in reducing ammonia losses compared with surface broadcasting and increase the soil mineral N pool available for crop uptake, along with reducing the need for manufactured fertiliser N applications to meet crop demand. However, ammonia loss reductions may increase the risk of nitrous oxide emissions; an example of "pollution swapping". The effectiveness of trailing shoe slurry application in reducing ammonia emissions can be variable, with factors such as slurry application rate (which is the main factor that can be controlled by the farmer) retention of slurry in a band and slurry infiltration into the soil likely to be important in controlling ammonia emissions.

Materials and methods

Four experiments were carried out at three sites in England, with contrasting soil textures and climatic conditions (Table 1). Slurry was applied by trailing shoe and surface broadcasting at five rates, viz: 20, 35, 50, 65 and 80 m^3/ha, using a purpose built small-plot applicator. Ammonia emissions were measured from each treatment for 7 days after application using windtunnels. Nitrous oxide emissions were measured from the 35m^3/ha slurry rate for three months after application from both application techniques and an untreated control, using the static chamber technique (five chambers per plot). For each experiment, additional plots received manufactured fertiliser N applications at six incremental rates (0-300 kg/ha N) to quantify the fertiliser N replacement value of the different slurry applications.

Table 1. Site details and slurry analysis

Site	Topsoil texture	Slurry type	Dry matter (%)	Total N (kg/m^3)	NH_4-N (kg/m^3)	Application timing
North Wyke (Devon)	Sandy loam	Pig	6.7	8.1	4.7	November 2003
Burley Dam (Cheshire)	Clay loam	Cattle	8.9	4.5	2.3	March 2004
North Wyke (Devon)	Sandy loam	Cattle	4.3	1.7	0.7	November 2004
Gleadthorpe (Nottinghamshire)	Loamy sand	Pig	0.6	1.4	1.2	November 2005

Crop (fresh weight and dry matter) yields and N offtakes were measured at harvest. Slurry fertiliser N replacement values and N use efficiencies were determined by comparing grass yields and N offtakes on the slurry treatments with those on the (spring applied) manufactured fertiliser N treatments.

Results and discussion

Trailing shoe slurry application reduced ($P<0.05$) ammonia losses compared with surface broadcasting at North Wyke in November 2003, but not in any of the other three experiments. At North Wyke (2003) mean ammonia losses following the trailing shoe slurry application were 32% lower than losses following surface broadcasting (Figure 1). The reduction in ammonia losses occurred as the slurry stayed in a band (reducing the area of slurry exposed to the air) *and* rapidly infiltrated into the sandy loam textured soil. Band widths increased from 4 cm at the 20 m^3/ha application rate (19% ground coverage) to 7 cm at the 80 m^3/ha application rate (37% ground coverage). There was no effect ($P>0.05$) of slurry application rate on ammonia losses (expressed as a % of total N applied) on any of the trailing shoe treatments and on three out of the four broadcast treatments. However, at North Wyke (2003) there was an inverse relationship between broadcast slurry application rate and ammonia emissions.

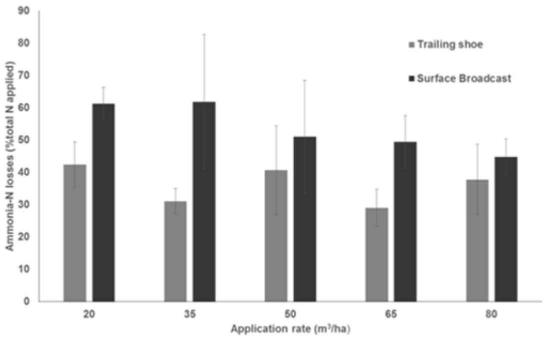

Figure 1. Ammonia emissions following trailing shoe and surface broadcast slurry applications at North Wyke (November 2003)

There was no effect ($P>0.05$) of trailing shoe compared with surface broadcast application on nitrous oxide emissions in any of the four experiments. At North Wyke (2003) and Burleydam nitrous oxide emissions were below the IPCC default value of 1 % of total N applied. At North Wyke (2004) and Gleadthorpe, nitrous oxide emissions following slurry application were equivalent to 1.21% and 1.78% of total N applied from the trailing shoe, and 1.57% and 1.67% of total N applied from the broadcast applications, respectively. The higher losses at these latter 2 sites were probably a reflection of soil and climate conditions at the time of application and low amounts of grass N uptake during the late autumn/winter period which would have enhanced the potential for nitrous oxide emissions. The mean slurry N efficiency (i.e. the manufactured fertiliser N replacement value expressed as a % of total slurry N applied) was 46% at Gleadthorpe and 25% at North Wyke (2004). The higher slurry N efficiency was probably a reflection of the lower over-winter drainage volumes (36 mm at Gleadthorpe and 216 mm at North Wyke) which would have reduced over-winter nitrate leaching losses and increased crop available N supply from the November slurry applications. There was no effect of application technique on N use efficiency at Gleadthorpe or North Wyke (2004) and no response to manufactured fertiliser N applications in the other two experiments.

Conclusions

Trailing shoe slurry application reduced ammonia emissions compared with surface broadcasting in only one out of these four experiments, where the slurry stayed in a band *and* rapidly infiltrated into the soil. Soil conditions at the time of application were important factors influencing the effectiveness of the trailing shoe application technique at reducing ammonia emissions. Trailing shoe applications did not increase nitrous oxide emissions or slurry N use efficiency in any of the four experiments compared with surface broadcasting.

Acknowledgements

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MODELLING N_2O EMISSIONS FROM AUSTRIAN SOILS: IMPACT OF SOIL TYPE, CROP MANAGEMENT AND CLIMATE

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Within the project FarmClim ("Farming for a better climate") we assess recent N_2O emissions from two selected regions in Austria. Our aim is to deepen the understanding of Austrian N_2O fluxes regarding region specific properties. Currently, N_2O emissions are estimated with the IPCC default emission factor which only considers the amount of N-input as an influencing factor for N_2O emissions. We evaluate the IPCC default emission factor for its validity under spatially distinct environmental conditions.

Material and Methods

Two regions for modeling with LandscapeDNDC have been identified in this project. The benefit of using LandscapeDNDC is the detailed illustration of microbial processes in the soil. Required input data to run the model included daily climate data, vegetation properties, soil characteristics and land management. The analysis of present agricultural practices is basis for assessing the hot spots and hot moments of nitrogen emissions on a regional scale.

Results and Discussion

Soil type determines N_2O emissions. During our work with LandscapeDNDC we were able to adapt specific model algorithms to Austrian agricultural conditions. The model revealed a strong dependency of N_2O emissions on soil type. We could estimate how strongly soil texture affects N_2O emissions. Based on detailed soil maps with high spatial resolution we calculated more precisely region specific contribution to N_2O emissions. Accordingly we differentiated regions with deviating gas fluxes compared to the predictions by the IPCC inventory methodology.

Different Crops display different emission factors. Taking region specific management practices into account (tillage, irrigation, residuals) calculation of crop rotation (fallow, catch crop, winter wheat, barley, winter barley, sugar beet, corn, potato, onion and rapeseed) resulted in N_2O emissions differing by a factor of 30 depending on preceding crop and climate. A maximum of 2% of N fertilizer input was emitted as N_2O. Residual N in the soil was a major factor stimulating N_2O emissions.

Climate results in interannual variability of N cycling with occasional "hot years". Interannual variability is affected by varying N-deposition even in case of constant management practices. High temporal resolution of model outputs enables to identify hot moments of N-turnover and total N_2O emissions according to extreme weather events. We analysed how strongly these event based emissions, which are not accounted for by classical inventories, affect emission factors.

Conclusion

The evaluation of the IPCC default emission factor for its validity under spatially distinct environmental conditions revealed which environmental conditions are responsible for major deviations of actual emissions from the theoretical values. Scrutinizing these conditions can help to improve climate reporting and greenhouse gas mitigation measures.

Acknowledgements:

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AGRONOMIC ASSESSMENT OF NITRATE LEACHING AND GREENHOUSE GASES EMISSIONS IN WHEAT AND MAIZE SYSTEMS

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Guaranteeing high crop yields while reducing environmental impacts of nitrogen fertilizer use due to associated losses of nitrate (NO_3^-) leaching and emissions of greenhouse gases (GHGs) in the context of sustainable intensification agriculture (*Zhou et al., 2013, 2014*). Previous study demonstrated that the lowest yield-scaled GWP (kg CO_2 eq Mg^{-1} grain) were achieved at 91% and 90% of maximal yields for wheat and maize systems, respectively, suggesting that yield-scaled approach is a valuable integrated metric for assessing GHGs emissions from cereal cropping (*Linquist et al., 2012*). However, it is still a gap in knowledge to assess NO_3^- leaching loss by linking crop yield for a given cereal cropping system.

Materials and Methods

We conducted a meta-analysis on 32 published studies reporting both NO3- leaching losses and crop yields to develop a new metric of yield-scaled NO_3^- leaching and assesse NO_3^- leaching loss on a crop-yield basis in cereal cropping systems. Besides the meta-analysis study a year-round large-scale field lysimeter (8×4 m^2) experiment under one control (NF) and three N fertilization treatments with same total rate of N application (280 kg N ha-1) were conducted in the Sichuan Basin, China from October 2009 to October 2010. NO_3^- leaching losses, CH_4 and N_2O emissions as well as crop yields were simultaneously measured throughout the experimental period.

Results and Discussion

Our meta-analysis study indicated that in average for agricultural soils worldwide production of maize or wheat is associated with significant NO_3^- leaching losses: maize: 5.40 kg N Mg^{-1}; wheat: 5.41 kg N Mg^{-1}. Lowest yield-scaled NO_3^- leaching losses (maize: 2.6 kg N Mg^{-1}; wheat: 2.4 kg N Mg^{-1}) were achieved at 93% and 96% of maximum crop yields. Thus, reductions of either GHG emission or NO_3^- leaching loss and guaranteeing high crop yield can be achieved at slightly suboptimal crop yields (GHG emission: 90-91% [Linquist et al., 2012]; NO_3^- leaching: 93-96%) in the wheat and maize systems. In the field study comparable average yield-scaled GWP were observed in the wheat (81.8 kg eq CO_2 Mg^{-1} grain) and maize (83.6 kg eq CO_2 Mg^{-1}) growing seasons (Fig.1). The yield-scaled NO_3^- leaching ranged from 6.4 to 10.7 kg N Mg^{-1} for maize growing season and were higher than the aggregately global mean value of 5.4 kg N Mg^{-1}. Because over 20% of applied N fertilizers were lost in the maize growing season most likely due to the subtropical climate (>65% of annual precipitation in maize season) and the relatively thin soil layer (20-80cm) of sloping cropland in the study region (*Zhou et al., 2012*).

Conclusions

Yield-scaled approach is valuable integrated metric for assessing either NO_3^- leaching loss or GHGs emission in cereal cropping systems. By applying the yield-scaled approach, our study found that combination of synthetic N and manure fertilizers (60%+40%) can reduce negative environmental impacts of NO_3^- leaching and emissions of N_2O and CH_4 without compromising crop productivity from the rain-fed wheat-maize rotation system in the Sichuan Basin, China.

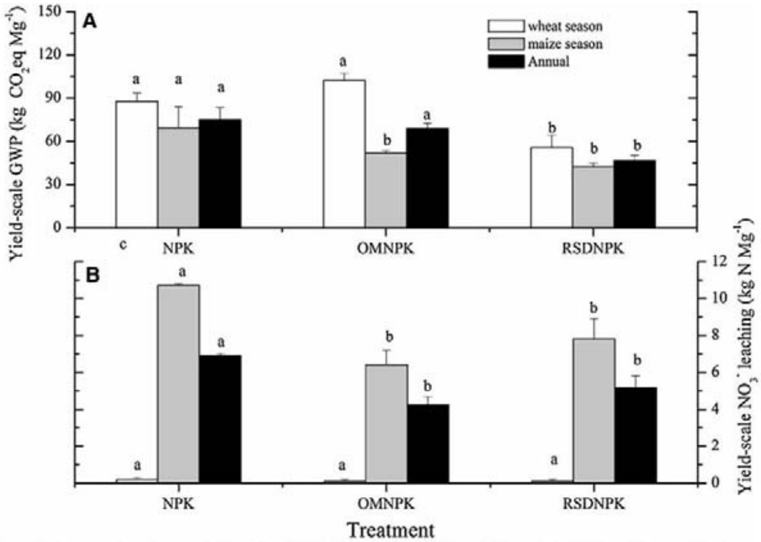

Fig.1 Seasonal and annual (October 2009-September 2010) yield-scaled GWP with considering soil CH_4, direct and indirect N_2O fluxes (**a**) and yield-scaled NO_3^- leaching (**b**) for the different fertilizer treatments. The different lowercase letters indicate significant differences ($P<0.05$, LSD) among the fertilized treatments.

Acknowledgement

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Topic 4

Food security, integrative and global nitrogen challenges

FEEDING THE WORLD IN 2050: ASSESSING THE REALM OF POSSIBILITIES

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Through a detailed analysis of the FAO database, we have proposed a generalised representation of the nitrogen transfers characterising the current agro-food system of 12 macro-regions of the world, defined on the basis of their pattern of international trade exchanges and level of self sufficiency with regard to their local needs of proteins for human and livestock feeding (Lassaletta et al, 2014; Billen et al., submitted). Figure 1 summarises this analysis, aggregated at the global scale.

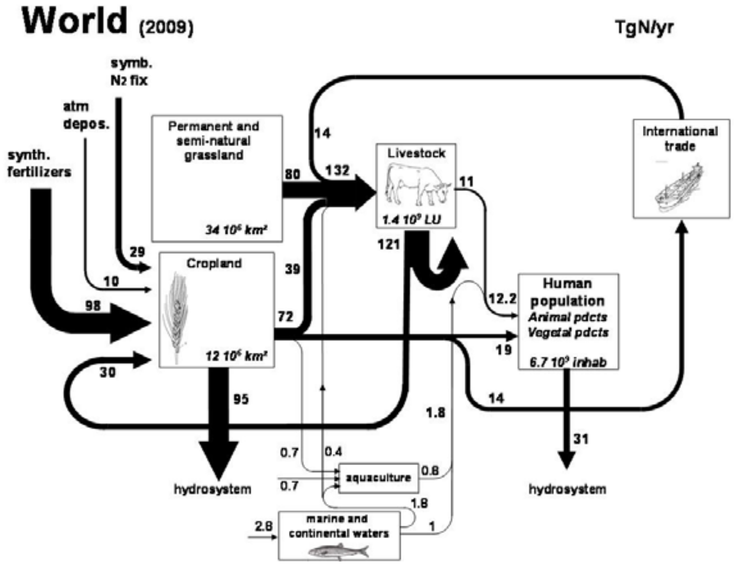

Figure 1. Fluxes of N characterising the global agro-food system in 2009. (Billen et al., submitted)

The analysis first highlights the inequality between the different regions in terms of human diet (total protein intake between 3.3 and 6.5 kgN/cap/yr with 15 to 58 % animal proteins), as well as considerable differences in the efficiency of vegetal to animal protein conversion by livestock systems (from 2.4 to 21%). For the cropping systems of each region, we established the relationship between total inputs of nitrogen to cropland and crop production expressed in nitrogen content and integrated over the whole rotation cycle (Billen et al, 2013; Lassaletta et al., subm.). This relationship characterizes both the agronomical and environmental performances of the agriculture of each region of the world. In terms of food sovereignty, the analysis reveals that a small number of net exporting countries such as Brazil, Argentina, the USA and Canada are closing the gap between production and demand of a large number of deficient, net importing countries. Based on this analysis, we wanted to assess the changes that would be required to meet the requirements of the estimated world population in 2050, according to various combinations of objectives in terms of human diet, food sovereignty and environmental contamination.

Method

A simple model has been establish to systematically explore the possibilities of meeting the requirements of human population of each of the 12 regions in 2050 (according to FAO predictions) for varying human diet patterns (in terms of total protein intake and the fraction of animal proteins), varying degrees of self-sufficiency (from an objective of complete self-sufficiency to complete openness to imports), and varying ceilings of environmental contamination (based on the value of the N surplus to cropland). In this exercise, the total cropland area, as well as the performances of the cropping and livestock systems of each region, is kept unchanged with respect to current values. The recourse to synthetic N fertiliser is calculated from the total fertilisation rate required in each scenario, taking into account the importance of other available sources of fertilisation (manure and symbiotic fixation).

Results and Discussion

The interest of this simplified approach based on a description of the global Agro-Food system in terms of nitrogen only, is that it allows a systematic exploration of a large range of future scenarios and to check their capacity to meet certain criteria. The results show that the realm of possibilities for feeding the world in 2050 is quite large, and does not necessarily depends on a considerable intensification of crop production, or an increase in the use of synthetic fertilisers. However, the hierarchy set in each region regarding the objectives of improved diet, self sovereignty and environment quality has considerable implication on the resulting global agro-food system. Particular attention is devoted to the benefits of reconnecting livestock and crop farming within each region of the world, in terms of optimisation of local resources exploitation and environmental quality (Lemaire et al., 2013).

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STUCK IN THE ANTHROPOCENE: THE CASE OF REACTIVE NITROGEN

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Reactive nitrogen (Nr) is an indispensable nutrient for agricultural production and human alimentation. At the same time, terrestrial, aquatic, air and atmospheric Nr pollution is estimated to cause damage in the magnitude of 0.3% to 3% of global GDP, mainly to human health and ecosystem services (Sutton et al. 2013). An influential study (Rockström et al. 2009) suggests to reduce anthropogenic Nr sources below a 'planetary boundary' of 35 Teragram (Tg) Nr in order to keep the earth system within the stable conditions of the Holocene, which is about one sixth of the current level (Fowler et al. 2013). As agriculture is responsible for three quarters of current anthropogenic Nr sources (Fowler et al. 2013), this sector is central to Nr mitigation. Key mitigation options to reduce agricultural requirements of Nr include the improvement of Nr efficiency in crop and animal production systems, the reduction of food waste in households, and lower consumption of Nr-intensive products like meat and milk (Sutton et al. 2013). However, the mitigation potential of these measures remains unclear, especially under the added pressure of population growth and changes in food consumption. This study quantifies the impact of these mitigation measures on Nr pollution and estimates the amount of Nr sources agriculture requires even under ambitious mitigation measures.

Materials and Methods

We create projections of the agricultural Nr cycle using the Model of Agricultural Production and its Impact on the Environment (MAgPIE)(Lotze-Campen et al. 2008; Popp, Lotze-Campen, and Bodirsky 2010; Bodirsky et al. 2012). For a given food demand (Bodirsky et al. in review) the model estimates cost-optimal production patterns under the limitations of scarce land and water. The model distinguishes 10 world regions, 18 crop groups and 5 livestock groups. The model includes a nitrogen-budget module (Bodirsky et al. 2012) that covers the agricultural Nr flows in cropland and livestock management, processing of agricultural products and food consumption. The closed budget approach guarantees that the sum of Nr fixation, Nr release and inflows from other sectors corresponds to the sum of Nr losses and Nr flows to other sectors. Similarly, intermediary closed budgets are also used on a regional level for cropland soils, distribution and processing of agricultural products, livestock feeding, manure management, and the household sector. Taking the example of cropland soils, the Nr withdrawn from the soil by crop biomass and lost to the environment has to equal Nr inputs like inorganic fertilizers, manure and atmospheric deposition.

Results and Discussion

We show that, with regard to Nr pollution, the return to Holocene conditions is out of reach for decades to come. If current trends persist, crop production alone will require Nr sources of 220 Tg Nr by the year 2050. Even if all key mitigation options were implemented (see Figure 1), crop production still requires 86 Tg Nr per year by the year 2050, more than twice the level of the 'planetary boundary'. Beyond the analysed mitigation measures, few further options remain. These include lowering material use of agricultural products, abandoning the burning of crop residues, or reducing and recycling processing waste. Our scenarios did also not consider morally problematic options like reducing population growth (O'Neill et al. 2010), profound behavioural changes like diets solely based on leguminous proteins (Pelletier and Tyedmers 2010), or technological breakthroughs until 2050 like cereals that fix nitrogen (Beatty and Good 2011).

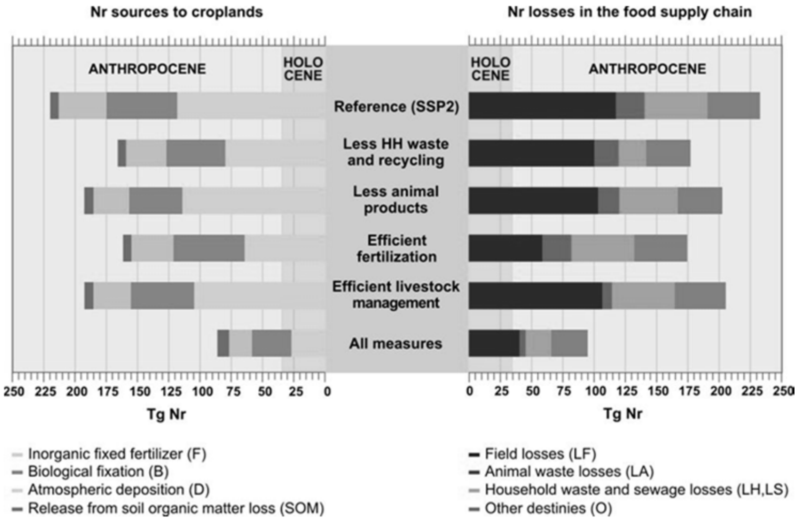

Figure 1: Reference and mitigation scenarios of Nr sources and the subsequent loss of Nr throughout the food supply chain in the year 2050.

Conclusions

Our findings stress the urgent need for ambitious mitigation policies, targeting both farmers and consumers. As Nr pollution will also persist under mitigation, a strategy is required that protects vulnerable systems from Nr pollution.

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A FIRST APPROACH TO THE CALCULATION OF N-FOOTPRINT IN PORTUGAL

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Nitrogen (N) is one of the most important elements on earth, about 78% of the atmosphere. N compounds can be grouped in nonreactive N (N_2) and reactive N (Nr). When reactive forms of N are in excess in the environment they threaten air, water, and soil quality and lead to changes in biodiversity and ecosystems as well as greenhouse gases balance (Galloway et al., 2008). Before the 18th century Nr did not accumulate in the environment, because biological nitrogen fixation and denitrification were mutually compensated. World population growth after the Industrial Revolution resulted in an increased energy and food production and consequently industrialization of agriculture, which led to uncontrollable waste production with negative consequences for the environment. Feeding the world's population was only possible due to the invention of the Haber-Bosh process in 1909, through synthesis of cheap mineral N fertilizers from N_2 and energy, which boosted the use of N in agriculture. This invention brought serious changes in the N cycle, thus making N one of the major pollutants from agriculture. Leach et al. (2012) developed a tool which will put together the N-PRINT system for the calculation of Nr losses into the environment. The first tool called N-calculator, is a personal footprint model to estimate the total amount of Nr lost into the environment due to individual consumption of food and energy. It accounts not only for the losses resulting from food and energy consumed, but also for losses resulting from all the activities upstream. Upstream losses include all Nr released into the environment from production of food and energy (housing, transportation, goods and services), and accounts for as virtual factors (Leach et al., 2012). The objective of this work was to calculate the average *per capita* N Footprint for Portugal and the average individual´s N Footprint for a specific region in Portugal, Lisbon, by adapting the survey proposed by Leach et al (2012) model according to Mediterranean consumption habits.

Material and Methods:

One thousand surveys were made randomly, to individuals of both sexes, and different ages, according to sex and age composition of the population in Lisbon region. The survey was performed in different days and in different places of the city and surroundings, including commercial areas, touristic places, office neighborhoods and residential areas. The survey consisted of questions about food consumption habits, and about energy consumption habits, such as the use of several types of transportation, and household equipment. Average per capita N footprint and individual´s food N footprint were calculated to poultry meat, pig meat, bovine meat, milk, butter and yogurts, cheese, fish and shellfish, animal fat, mutton and goat meat, cereals (wheat), rice, other cereals (including breakfast flakes), pasta, fruit, beans and grain, starchy roots, dry fruits, olive oil and olives, cooked vegetables, fresh vegetables, sugar and sweeteners, oil crops, spices, eggs, wine, alcoholic white spirits drunk, coffee and tea, beer and soft drinks.

The average per capita N footprint and individual´s energy N footprint were calculated to each type of housing energy (electricity, natural gas and gas cylinder) and transportation energy (plane, public transports, cars and motorcycles). As goods and services sector shows great complexity in quantification of specific emission factors, the Dutch values considered by Leach et al (2012) were used in this study.

Results and conclusions:

Portuguese average *per capita* N Footprint was 27.76 Kg N/capita/yr. Average N footprint from food consumption was 6.45 Kg N/capita/yr and the average N footprint from food production was 16.54 Kg N/capita/yr. Average N footprint from energy was 4.77 Kg N/capita/yr, consisting of 0.74 Kg from housing and 3.53 Kg from transportation. The average N footprint from goods and services was 0.5 Kg N/capita/yr (Figure 1)

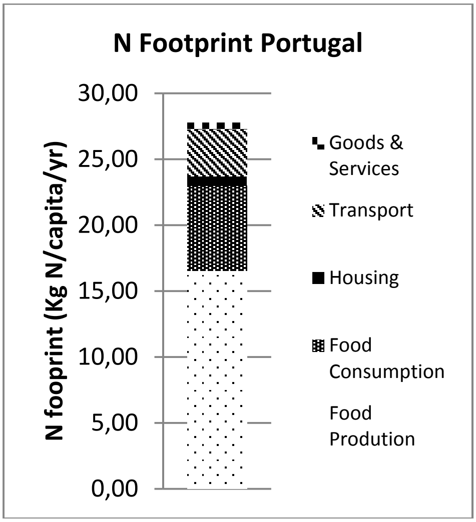

Figure 1. Nitrogen footprint for Portugal.

Average individual N footprint for Lisbon was 22.89 kg N/capita/yr. We observed that the average individual´s N footprint for men and women was similar. There was a slight difference between individual´s N footprints within the different age classes surveyed. Different age groups conditioned the values of the N footprint. The lowest N footprint was found for people with more than 65 years old (21.68 kg N/capita/yr) and was higher for people between 21-30 years old (23.49 kg N/capita/yr). Average *per capita* N footprint in Portugal was a value between USA and The Netherlands values, yet closer to The Netherlands. To increase reliability of the results, more surveys should be made in the North and South of Portugal. There are several ways of reducing individual N footprint with small changes in our daily choices. This message must be shared through communication and education. Several educational initiatives are being developed in Portugal, by us.

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THE RELATIONSHIP BETWEEN CROP YIELD AND NITROGEN INPUT TO CROPLAND IN 131 COUNTRIES: 50 YEARS TRENDS

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Feeding an increasing world population while reducing environmental impacts is one of the most important challenges for the next decades. The capacity of cropping systems to produce enough food, the composition of the human diet, the amount of food waste, the international trade and the global disconnection between crop and livestock are key drivers of the functioning of the regional and global agro-food system (Lassaletta et al., 2014; Billen et al sub). The availability of reactive nitrogen (N) is, with water, one of the factors most limiting crop production, however in many parts of the world nitrogen is being applied in excess producing very high environmental impacts with low yield increases at the same time (Mueller et al. 2012). In this work we study the evolution of the relationship between N inputs and crop production from 1961 to 2009 by 131 countries in order to explore at which extent increasing N input has really produced significant yield increases. This analysis also permits to characterize and compare the cropping systems of the different countries of the world in terms of both agronomical and environmental performances.

Methods

Principally based on FAOstat database, we have reconstructed the trajectory followed, in the past 50 years, by 131 countries in terms of crop yield (Y, expressed in kgN/ha/yr) and total nitrogen inputs to cropland (F, sum of nitrogen in manure, synthetic fertiliser, symbiotic fixation and atmospheric deposition, in kgN/ha/yr) (Lassaletta et al., submitted). In this work we expand the yield-fertilisation relationship conventionally used in agronomy for individual crops, by integrating it at the country level and over the whole crop rotation, and considering the mean total yield of a given territory and the total N inputs to the cropland soils. The ratio Y/F is a measure of its nitrogen use efficiency (N.U.E.) while the difference F-Y is the regional N surplus (or N balance) representing the potential for gaseous or hydrological losses of nitrogen to the environment.

Results and Discussion

We can classify the observed trajectories yield-fertilisation relationship during the last 50 years in 4 different types (Figure 1):

Type 1: Countries like China, Egypt or Chile are presenting a simple trajectory with regularly increasing fertilisation and progressive reductions in the responses of the crop yields (the trajectory approaches therefore to a yield plateau or Ymax). In these countries the N.U.E. has progressively dropped while surpluses disproportionately increased.

Type 2: In many countries, such as USA or Brazil, historical trajectory with first a regularly increasing fertilisation and yield similar to Type 1, then a turning point with a shift of the trajectory to a Y vs F relationship with a higher Ymax. After an initial period of reduction of drop in the country N.U.E. this efficiency remains constant of even rises.

Type 3: In most European countries, the trajectory also shows a bi-phasic pattern, describing a regular increase in both fertilisation and yield during the 1960-1975 period, followed by a shift toward improved yields without further increasing fertilisation and even decreasing fertilisation from the 1980's. Despite the undoubtedly positive aspect associated to the increase of the N.U.E. together to decreases in N surpluses, the excess of nitrogen emitted to the environment is nevertheless in many cases much higher than those of other countries belonging to Types 1-2.

Type 4: In some countries like many Sub-Saharan countries, the trajectory does not display any consistent Y vs F relationship. These countries have always very low yield. Very often, their trajectory in the Y vs F figure crosses the 1:1 line, indicating suggesting existence of soil N mining..

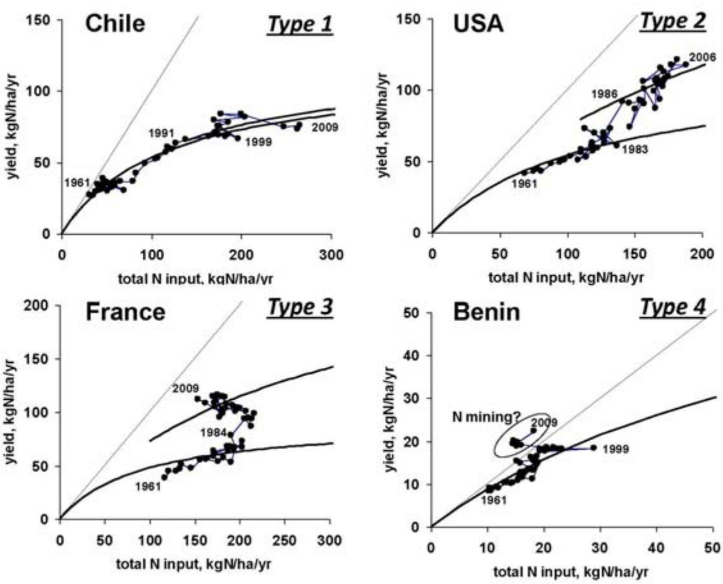

Figure 1. Examples of trajectories described by countries in the Y vs F diagram (Y: crop yield in protein harvested, kgN/ha/yr; F: total N inputs to cropland soils, kgN/ha/yr).

Conclusions

Our data suggest that in many countries further increase of nitrogen fertilization would only result in a disproportionately low increase of crop production. Only acting in other components of the cropping systems the proportion of the applied N that is finally uptaken by the crops, it could be optimize as has occurred in some of the studied countries. The case of European countries is a clear example of the improvements associated to the application of environmental policies. Despite these improvements the N surpluses of European countries still need to be reduced.

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LIFE CYCLE ASSESSMENT OF ENVIRONMENTAL EMISSIONS FROM EXPORTED NEW ZEALAND DAIRY AND LAMB PRODUCTS AND THE IMPLICATIONS FOR NITROGEN FOOTPRINTING

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Life Cycle Assessment (LCA) is a relatively recent tool for use in agriculture to evaluate the total resource use and environmental emissions associated with products throughout the supply chain from raw material extraction to consumer use and waste (e.g. ISO14044 2006). It has been used for eco-labelling of products in supermarkets, with most application for carbon footprinting. The carbon footprint of products accounts for all greenhouse gas (GHG) emissions, including nitrous oxide (N_2O) emissions. However, the environmental labelling of products in supermarkets is being extended to other environmental indicators including eutrophication due to nitrogen (N) and/or phosphorus emissions to waterways.

In recent years, the emissions of reactive N into the environment in general have been summed to produce a nitrogen footprint related to the life-style of people and recognising the contribution from the food we eat and our transport related N emissions (e.g. Leach et al. 2012). This uses partial LCA principles, although it mixes different environmental impacts into one single indicator.

The aim of this research was to examine the life cycle of lamb and dairy products using LCA covering the "cradle-to-grave" (i.e. from raw material extraction, farm production, processing, transportation, retailing, consumer use and waste) for a range of resource use and environmental indicators. This included fossil fuel use, carbon and N footprinting, and other impact categories including eutrophication. The implications for N efficiency, N losses and N footprinting were assessed

Materials and Methods

Two value chain systems were used for this research. The first was for lamb produced from hill country farms in New Zealand (NZ), processed in abattoirs, shipped to the United Kingdom, where it was purchased, cooked and eaten by consumers before contributing to the waste management system (Ledgard et al. 2011). The second was the production of milk and meat products from dairy farm systems in NZ, but was confined to the "cradle-to-farm-gate stage".

For both value chains, all resource inputs were defined based on survey data from a wide range of farms. For the sheep supply chain, input data was also collected from abattoirs, while secondary published data was used for transportation, retail, consumption and waste stages using LCA principles. This included data collection on the amount and types of energy sources and N inputs for all relevant stages of the life cycle. Validated models were used to calculate ammonia and N leaching (Ledgard et al. 2004) and N_2O emissions (NZ GHG Inventory methodology).

Results and Discussion

For sheep production on-farm, the "fossil-N' inputs from N fertiliser were low relative to that from atmospheric N_2 fixation by pasture legumes, i.e. 14 versus 57 kg N/ha/year. Emissions of N throughout the life cycle (i.e. raw material extraction through to consumption and waste) were 52, 37, 5 and 6 % as ammonia, nitrate, N_2O and NO_x, respectively. The relative contributions from the different life cycle stages were 94, 4, 1 and 1% for farm, transport, processing and consumer-use/waste stages, respectively. The carbon footprint was also dominated by the farm stage (82%), whereas the fossil fuel energy use was dominated by the retail/consumer use stage (56%).

For the average NZ dairy farm system (land for dairy+replacement animals; unallocated), the N inputs equated to 74, 34 and 80 kg N/ha/year for N fertiliser, brought-in feed and clover N_2 fixation. The total reactive-N outputs (from the dairy farm system plus all crops and system inputs) from N leaching, ammonia, N_2O and NOx emissions were calculated using validated models at 0.61, 0.53, 0.07 and 0.02 kg N/kg milk (fat-and-protein-corrected), respectively. The main contributor to NOx was fossil fuel use (from fertiliser production/use and brought-in feed), whereas animal excreta and N fertiliser use were the main contributors to N leaching, ammonia and N_2O emissions.

The relative reactive-N output per total hectares for farm production were 31.0 and 64.7 kg/ha/year for sheep and milk (after allocation for other products). The corresponding N footprints, as kg N per kg protein-N output, were 1.78 and 1.32, respectively. Similarly, sheep had a higher carbon footprint than milk at 251 and 172 kg CO_2equivalent/per kg protein-N, respectively. Results will also be compared with that of other resource use (e.g. fossil fuels) and environmental emissions (e.g. eutrophication potential) and the implications for pollution swapping will be discussed.

Conclusions

In the application of LCA for agricultural products, N emissions are major contributors to a number of different environmental impact categories including climate change (via GHGs), eutrophication, acidification, photochemical oxidation and human toxicity.

The implications of using an N footprint to sum reactive N emissions are discussed relative to the use of LCA to estimate multiple environmental impacts from N emissions.

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EFFECTS OF DIETARY PROTEASE ON N EMISSIONS FROM BROILER PRODUCTION: A HOLISTIC COMPARISON USING LIFE CYCLE ASSESSMENT

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The aim of the study was to quantify the effects of the use of a protease "RONOZYME ProAct" in broiler diets on the environmental impacts, including the nitrogen emissions, of standard indoor broiler production. This was done by using a Life Cycle Assessment (LCA) modelling approach for different scenarios of broiler production where diets either (a) without protease or (b) with protease combined with reduced protein content were applied. LCA evaluates the production chain systematically to account for all inputs and outputs that cross a specified system boundary and relates these to the useful outputs.

Materials and Methods

All experimental and other primary data were provided by the industry. The bird performance data came from seven separate trials where the effects of low-protein diets including protease on broiler performance were evaluated. Additional background data, such as energy use in production, breeding and transport were obtained from earlier studies on UK broiler production (Leinonen et al. 2012). The structural model for the broiler production system calculated all of the inputs required to produce the functional unit, allowing for breeding overheads, mortalities and productivity levels. A separate sub-model for arable production was used to quantify the environmental impacts of the main feed ingredients (Williams et al. 2010). A separate sub-model was also used for manure management and for the nutrient cycle (Williams et al. 2006). As an output of the LCA model, the nitrogen and other emissions were aggregated into environmentally functional groups as follows: **Global Warming Potential (GWP)** is a measure of the greenhouse gas emissions to the atmosphere. The main sources of GWP are CO_2 from fossil fuel and land use changes, N_2O and CH_4. **Eutrophication Potential (EP)**, where the main sources are NO_3^- and PO_4^{3-} leaching to water and NH_3 emissions to air. **Acidification Potential (AP)**, where the main source is NH_3 emissions, together with SO_2 from fossil fuel combustion. The analysis was carried out for two alternative systems boundaries: **1. Feed production chain** included growing of feed crops, production of additives, processing of ingredients and mixtures, transport of the ingredients, production of fertilizers etc. **2. Broiler production chain** quantified all impacts related to feed production (as above) and included also energy use in housing the broilers, emissions from broiler house and storage and land spreading of the manure, broiler breeder production, hatching etc.

Results and Discussion

The results for the feed production chain (Fig. 1A) show that there is a reduction in all environmental impact categories, per mass unit of feed, when protease is used in the diets. The main reason for this is the reduction of the amount of soya used in the diets. The biggest reduction occurs in the category of Global Warming Potential. This is mainly caused by decreased CO_2 emissions from land use changes related to soya production and lower emissions from the transport of the soya. With Eutrophication Potential, there were only small differences between the diets, while Acidification showed bigger reduction, again as a result of lower requirements of soya. In the results of the analysis of the broiler production chain (Fig. 1B), there were relatively bigger reductions in Eutrophication Potential and especially in Acidification Potential, compared to the feed production chain alone. The reason for this is that a major part of these impacts arises from the emissions from housing and manure management. When protease was used in the diets, the crude protein content of the feed was reduced, which reduced the emission of ammonia which affects both the Acidification and Eutrophication Potentials, and leaching of nitrate, affecting the Eutrophication Potential. A substantial benefit is the reduction of ammonia emissions *per se*, as these are problematic in poultry production and the subject of regulation from large units. A nutritional solution requires no changing in building design or use of retrofitted scrubbers. Reduced ammonia emissions should also improve air quality for both birds and workers. In broiler production, the main motivation for using protease is reduction of feeding costs, as protein is one of the most expensive components in diets. The results of this study suggest that with protease, this economic benefit can also be combined with environmental improvements.

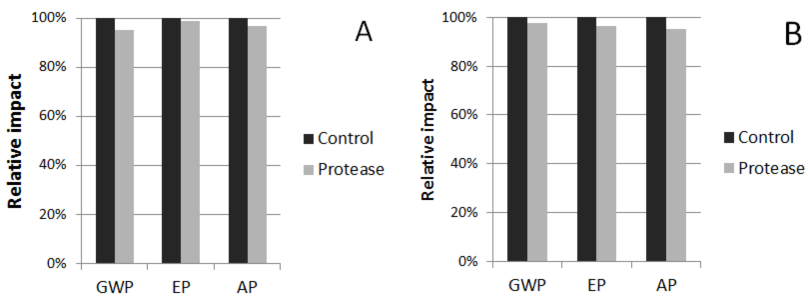

Figure 1. The average effect of different diets on the environmental impacts, including Global Warming Potential (GWP), Eutrophication Potential (EP) and Acidification Potential (AP), **per unit of feed** at feed mill gate (A) and **per unit of expected carcass weight** at farm gate (B).

Conclusions

The use of protease in the broiler diets and the resulting reduction of the use of soya reduced the environmental impacts of both the feed production (mainly Global Warming Potential) and broiler production (mainly Acidification Potential) chains. The latter is mainly through reduced ammonia emissions, which has a range of environmental benefits.

Acknowledgements

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HOW LONG IS THE NITROGEN CASCADE?

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Once a molecule of inert nitrogen has been unlocked by nitrogen fixation or burning of fossil burning and released as reactive nitrogen to the environment, it may undergo several transformations until it will be locked again by either long-term sedimentation or denitrification to molecular / inert nitrogen. During each stage of this transformation process, Nr contributes to an impact, some of which are intended, while others are unintended. The positive impacts include the Nr to serve as a source of protein for humans or livestock and the increase in soil fertility, while the negative impacts are manifold and include effects on human health, biodiversity, air and water pollution, soil acidification. The concept of such a cascading impact of Nr has first been introduced by Galloway et al. (2003), however, to our knowledge, analyses have so far been performed as static budgeting using the N-cascade as a conceptual underpinning only (cf. Galloway et al., 2008; Gruber and Galloway, 2008; Billen et al., 2013). Detailed full integrated nitrogen budgets (iNBs) as proposed by Leip et al. (2011) and later on defined by UNECE (2013) provide the means for a more quantitative analysis of the cascading effects of a new unit of Nr. The objective of this paper is to exploit this possibility and provide quantitative indicators for the length of the N-cascade and the dispersion of Nr throughout the environment as a function of the point of entry on the basis of published iNBs.

Materials and Method

Published iNBs are processed with the DynIB (Dynamic Integrated Budgets) tool (Leip and Heldstab, 2013) in order to derive quantitative indicators describing the N-cascade. The tool calculates the number of steps that is required to 'lock' new introduced Nr, considering as sink denitrification to N_2 or N_2O (which is non-reactive in the troposphere and will not contribute to further cascading) or accumulation in a pool (ie sedimentation in lakes, increased biomass stocks in forests, accumulation in the anthroposphere in materials, etc.). Flows that transport Nr outside the system boundaries are not considered as a permanent sink. Indicators that can be derived include the length of the N-cascade until a defined share (e.g. 50% or 99.9%) of a unit of nitrogen introduced into a *pool* are transferred to a sink; the dispersion of Nr over the final sinks, the overall N-input to the different pools indicating how often a unit of Nr passes at the various pools, and the total N availability. Here we present some results on the basis of the European Nitrogen Budget (ENB, Leip et al., 2011a). Assessment of other iNBs (e.g. Clair et al., nd; Leip et al., 2011b; Junker, 2013; Fowler et al., 2013) are ongoing.

Results and Discussion

Final sinks depend largely on the length of the cascade, and the number of times an accumulating pool is passed, such as landfills (solid waste, solw), the anthroposphere (consumer, cons), coastal sedimentation (coas), or in forests (fore). Those accumulations are not necessarily permanent and reflect a current status quo situation. The only clearly permanent sink is denitrification. It makes between 54% and 95% of the total sink for nitrogen entering the system by industrial N-fixation (indu) or biological fixation in the hydrosphere (hydr), respectively (see Figure 1). As an example, the total overall N-input to the 'crop' pool – for each unit of Nr – is 1.56, which means that more than every second molecule of Nr passes this pool a second time. 0.56 kg N/kg new N is supplied overall to the consumer, a total of 0.84 kg N/kg new N is emitted to the atmosphere. Over all pools, each unit of Nr newly created for the crop pool enters 4.43 times a pool, thus the overall NUE is 0.56/4.43=12%. This is achieved – for the crop pool – after 17 steps, whereby 50% of the Nr is inactivated already after 2.6 steps, and 75% after 4.4 steps (see Figure 2). Similar numbers are obtained for several entry-points, however, for forests and the hydrosphere smaller N-cascade-lengths are calculated.

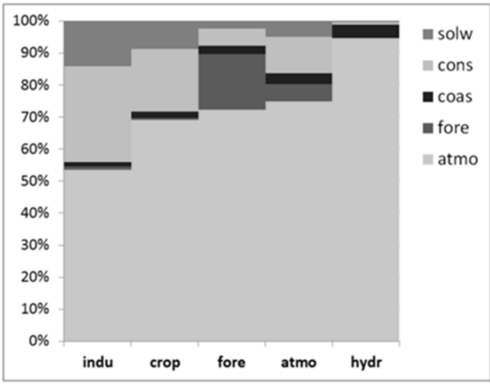

Figure 1: Dispersion of new Nr with different entry points (x-axis) over the final sinks considered

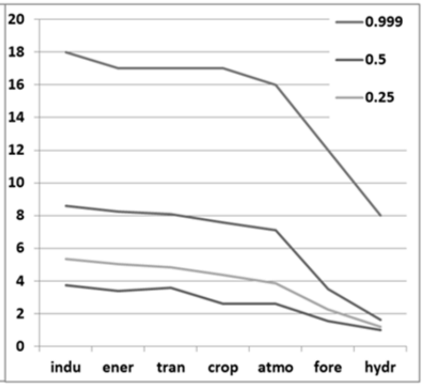

Figure 2: Length of N-cascade until inactivation of different shares of new Nr for different entry points

Acronyms: indu: Industry, ener: energy generation by combustion of fossil fuel; tran: transport; crop: agriculture, soil cultivation; cons: consumer, humans and settlements; fore: forest and semi-natural vegetation; atmo: atmosphere; hydr: hydropsphere, ground and inland surface waters; coas: coastal zones; solw: solid waste; sewa: sewage systems,

Conclusions

Integrated nitrogen budgets are a powerful tool for visualizing the complexity and interconnectedness of the N-cycle and highlighting possible handles. However, they can also be used to derive quantitative indicators describing the N-cascade at regional scale and providing a measure for whole-society NUE.

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ASSESSING THE ENVIRONMENTAL IMPACT FOR FOOD RECOMMENDATIONS IN EUROPE

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The assessment of the quality of diets is a multi-dimensional problem, with the healthiness of the dietary choices being one very important dimension and other dimensions including the environmental impact, such as the carbon, water, and nitrogen footprints. The composition of diets vary across Europe (Elmadfa et al., 2009; Freisling et al., 2010; EFSA, 2014) and the same is true also for both the health aspect and the environmental impact. For example, the Mediterranean diet has been associated with lower risk of developing chronic diseases (Sofi et al., 2008). However, regional dietary patterns may not be culturally acceptable or easy to follow in other parts of Europe (Poulsen et al., 2014). Ideally, food based dietary guidelines are developed based on the best available evidence to provide nutritionally adequate diets that promote good health and help preventing chronic disease development and are adapted to national or regional local and cultural contexts (see e.g. Switzerland, Hayer, 2011; Spain, France and Sweden, van Dooren and Kramer, 2012; Nordic countries, Norden, 2013). However, environmental aspects are not or only to a small extent taken into consideration when developing nutrition guidance. Regional variability of the environmental impact of food production is caused by different environmental conditions, agricultural systems and management practices, food processing and wastages (Weiss and Leip, 2012; FAO, 2013; Opio et al., 2013; Leip et al., 2013; Vanham et al., 2013). The objective of this paper is to evaluate the carbon and nitrogen footprints of current dietary customs and recommendations for major areas in Europe.

Materials and Method

In this abstract, we base the analysis on the data provided by Vanham et al. (2013) who provide data of a reference diet, a healthy diet and a vegetarian diet for four macro-regions in Europe (North, West, East, South). Quantification is done on the basis of 'emission intensities' (N-footprint and carbon footprint per kg of product) obtained from (Leip et al., 2013) and (Leip et al., 2014). Briefly, the emission intensities are calculated with the CAPRI model (Britz and Witzke, 2012) using a cradle-to-gate life cycle analysis (Weiss and Leip, 2012; Leip et al., 2013) with data representative for EU micro-regions (NUTS2).

Results and Discussion

Results of the total greenhouse gas (GHG) emissions and the total N-losses following cradle-to-gate life cycle analysis are shown in Figure 1. For the reference diet, total GHG emissions (carbon footprint) varies between ca. 1.3 and 1.8 t CO_2-eq (cap yr)$^{-1}$ for the regions East and South, respectively. This difference is linked to the lower consumption of high-protein food (all meats and pulses) partly compensated by higher consumption of cereals and potatoes. However, also overall food consumption was smaller by 13%. The effect is even larger when looking at total N-losses, where it is combined with lower emission intensities for major products (such as cereals and potatoes). Overall, the reduction in GHG and N-emissions when changing diet from reference to healthy is between 8% (north) and 45% (south) which increase to a range between 34% (east) and 60% (south) when changing to a vegetarian diet. Reductions are all in the animal product categories, while emissions from crop product consumption are stable or slightly increasing, with the exception of east due to increased consumption of fruits and vegetables.

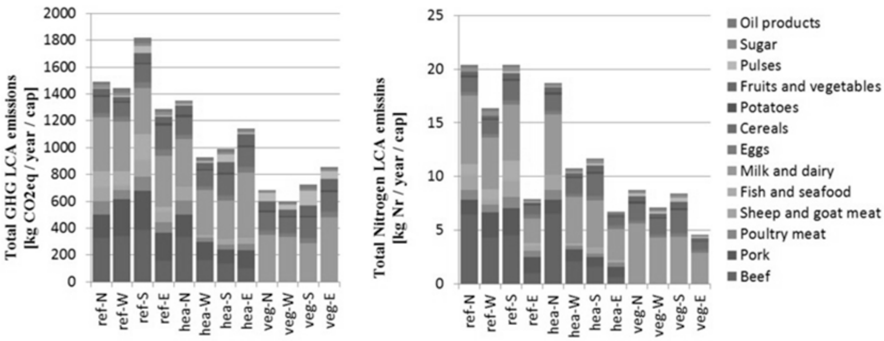

Figure 1. Total GHG emissions [kg CO_2-eq (cap yr)$^{-1}$] (left) and total reactive N-losses [kg N (cap yr)$^{-1}$] (right) for three diet scenarios (Reference, ref; healthy diet, hea; and vegetarian, veg) for four large regions in Europe (North, West, East, South). Regional grouping and definition of diets is done according to Vanham et al. (2013); emissions intensities [kg emissions (kg product)$^{-1}$] are done according to Leip et al. (2013, 2014). The data exclude emissions related to the consumption of alcoholic beverages, stimulants and spices.

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CROP COMBINATIONS, MANAGEMENT AND ECOMONIC BENEFITS OF INTERCROPS ACROSS EUROPE: A REVIEW

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There is an urgent global challenge in providing sufficient primary production to sustain a growing population, with increasing demands for foods, feeds and fuels, without exacerbating climate change and other environmental impacts of agriculture (Godfray et al., 2010). Nitrogen (N) availability remains one of the key drivers of crop productivity from arable farms of Europe. Compared with manufactured N fertilisers, the increased use of biologically fixed N from legumes is a more sustainable way of introducing N into agriculture (Fustec et al., 2010) whilst improving biological diversity, soil structure and protection from erosion (Jensen & Hauggaard-Nielsen, 2003). Symbiotic N_2 fixation by grain legumes is often the main N input into stockless organic cropping agro-ecosystems. Thus, the N input can be increased either by increasing the area on which such crops are growing or through management that can enhances the N_2 fixation ability. Modern European intercropping systems have been used for either organic agriculture and/ or conventional systems, where the tactical use of fertilisers and pesticides provide powerful additional management options.

Materials and Methods

In this review, we consider how legumes are used as mixtures with other legumes or in combination with other crop types as intercrops within practical arable European cropping systems taking into consideration the different climatic zones (North and South, East and West). In addition, a meta-analysis study included using all the peer-reviewed journal articles from January 2000 to January 2014, containing data on one or more of the following parameters: i) grain yield, ii) grain N, or iii) nitrous oxide emissions, and including independent variables such as: country, climatic zone, soil type, cereal species, legume species and fertiliser rates to examine the trends in each climatic zone. It is also conducted a short review on existing economic published studies on intercrops highlighting direct economic impacts.

Results

Grain yield benefits and sowing density; In the Mediterranean area, pea and cereals (wheat, barley, oat and triticale) were tested for grain yield and N content in grain, and shown a Land Equivalent Ratio (LER) higher than 1 in all cases, demonstrating a better resource use efficiency if compared with the respective sole crop systems. Similar results have been shown in Northern countries with pea barley to give high LER values above 1 distinct of all varietal combinations (Hauggaard-Nielsen and Jensen, 2001). And also clover with cereals had a great advantage in the cereal grain yields.More than 80% of the forage crop grown in the arable land is represented by a mixture of grasses and legume species sowing in the autumn period. However, the oat/common vetch (V. sativa) is the most popular intercropping system in the most southern Mediterranean areas, because this combination contributes to improve the hay quality than using oats in pure stand. In contarst, oat intercrops with the Vicia villlosa species grown in Mediterranean areas above 38° latitude (such as Northern Italy) due to the higher cold tolerance than V. sativa.

Economic benefits of intercropping; Studies on the productivity of cereal-legume intercropping and multispecies systems report very different and contrasting results depending on the crop, location and nitrogen fertilization compared to sole cropping. Prices for grain produced in intercropping systems are generally equal to grain produced in sole cropping except if the grain quality is influenced e.g. higher protein concentrations. However, this is only the case if the legume crop can also be marketed effectively. Economic analysis by Pelzer et al. (2012) showed higher average gross margins for pea-wheat intercropping compared to sole pea and sole wheat cultivation in unfertilised conditions and slightly lower gross margins under fertilised conditions. In contrast, Hauggaard-Nielsen and Jensen (2001) report higher economic benefits of pea sole cropping compared to pea-barley intercropping and barley sole cropping. Pelzer et al. (2012) also found, that the average gross margin of pea-wheat intercropping was in 70% of the cases higher than the average gross margin of pea and wheat sole crops together. As a result, pea-wheat intercropping leads to higher profitability if it replaces pea or if both crops are cultivated and replaced by the intercropping system e.g. 2 ha pea-wheat is more profitable than 1 ha pea and 1 ha wheat.

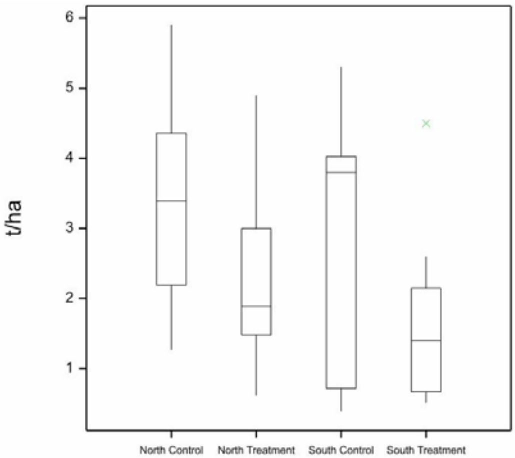

Figure 1. Statistical analysis based on existing published data according to climatic zones and crop treatment for intercropping field based experiments across Europe.

Conclusions

Improved understanding of different species/varietal combinations and seeding ratios will help to develop management recommendations for optimising the productivity and environmental impacts of intercropping systems in practice. Also using modelling approaches can help to simulate grain legume: cereal yield and rotational benefits in EU and worldwide. There is luck of information available to conclude whether intercropping leads to significant direct economic impact. However, such systems have the potential to use less agrochemical and fertiliser inputs, have an increased yield stability compared to sole cropping especially of sole legumes and are valid option for low-input and systems dominated by cereals.

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AN ANALYSIS OF CONSUMER-RELATED N FLOWS: FOOD AND MATERIAL USE.

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Facing potential negative effects of excess nitrogen (N) in the environment, ways to achieve more nitrogen-efficient agricultural production have been discussed extensively. We argue that consumer demand considerably shapes N emissions. Thus consideration of demand-side emission mitigation seems worthwhile, in addition to the technical options implementable at the production side. Many authors suggest a shift in diets as well as the substantial reduction of food waste (in the developed world) to reduce excess release of nitrogen compounds to the environment (Smil 2002; Zessner et al. 2010). At the same time, these measures would be beneficial to other environmentally relevant issues – such as energy consumption, land use or GHG emissions (Stehfest et al. 2013; Steinfeld et al. 2006).

While food contributes the largest share to N flows related to consumers, there are also other aspects that have largely been neglected so far, apparently due to the lack of easily accessible data and other research priorities. This includes mainly non-food industrial products and forest products, as well non-agricultural animals (pets). Leip et al. (2011) assume that the quantity on non-food N compounds can be quite significant. It thus seems highly appropriate to extend research in this field. In this work we aim to systematically identify relevant N flows caused and influenced by consumers, and provide quantitative estimates on their magnitude, using Austria as an example.

Materials and Methods

From a nitrogen budget perspective, we identify relevant inflows and outflows of nitrogen and estimate their order of magnitude using a range of national and international data sources (including the databases of the FAO, EUROSTAT, and STATISTICS AUSTRIA). Instead of providing a complete nitrogen budget with all its interrelations, we zoom in and focus on the consumer part that is in fact the central reason for all the surrounding activities.

This is done here on a national basis, using Austria as an example. In order to trace relevant changes and developments from the past, we cover the period from the 1960's until today. This helps to identify the effects of possible changes in diet and general consumption patterns (e.g. shift from synthetic polymers to renewable materials) in the future, which are assessed in a range of scenarios.

Results and Discussion

N flows relevant to consumers include mainly food products (primarily based on their protein content), but also a range of non-food products. This can be natural and synthetic polymers (polyurethanes, polyamides, or melamine), as well as surfactants, pet food, wood & paper products, and N products related to private gardening. Further N flows, such as those from pharmaceuticals & dietary supplements, products from semi-natural vegetation (cut flowers etc.) or nitrate in drinking water and food need to be scrutinized for their relevance. Quantitative estimates on the magnitude of these flows are provided. Furthermore, the share of imported N gives valuable insights into the extent of N leakage across national borders. Products that contribute the most to N leakage are identified.

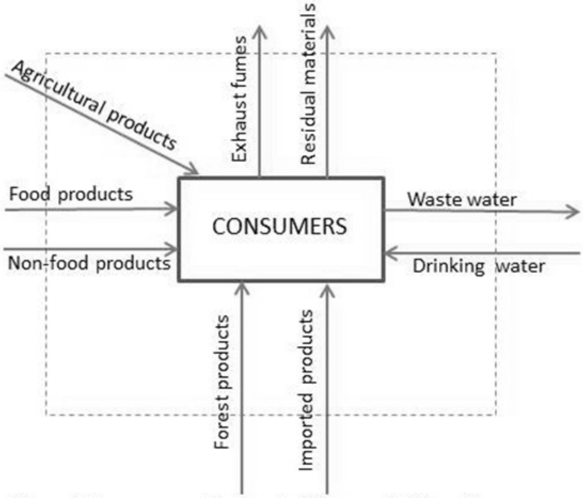

Figure 1: Consumer-related product flows containing nitrogen

Conclusion and Acknowledgement

Besides contributing to a (prospective) comprehensive national nitrogen budget, the results we present do also supplement the on-going discussion on N footprints of products as well as consumption patterns (Leach et al. 2012). The national budget (top-down) approach chosen allows for comparison with other (bottom-up) studies, and thus the validation of our results.

This paper is related to the work in progress on the annexes to the UNECE "Guidance document on National Nitrogen Budgets", that support compiling coherent and comparable national nitrogen budgets. Consequently, this work needs to be seen embedded in the broader overall context.

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WATER AND NITROGEN FOOTPRINT IN AN IRRIGATED CROP UNDER MINERAL AND ORGANIC FERTILIZATION.

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In order to establish rational nitrogen (N) application and reduce groundwater contamination, a clearer understanding of the N distribution through the growing season and its balance is crucial. Excessive doses of N and/or water applied to fertigated crops involve a substantial risk of aquifer contamination by nitrate; but knowledge of N cycling and availability within the soil could assist in avoiding this excess. In central Spain, the main horticultural fertigated crop is the melon type 'piel de sapo´ and it is cultivated in vulnerable zones to nitrate pollution (Directive 91/676/CEE). However, until few years ago there were not antecedents related to the optimization of nitrogen fertilization together with irrigation. Water and N footprint are indicators that allow assessing the impact generated by different agricultural practices, so they can be used to improve the management strategies in fertigated crop systems. The water footprint distinguishes between blue water (sources of water applied to the crop, like irrigation and precipitation), green water (water used by the crop and stored in the soil), and it is furthermore possible to quantify the impact of pollution by calculating the grey water, which is defined as the volume of polluted water created from the growing and production of crops. On the other hand, the N footprint considers green N (nitrogen consumed by the crops and stored in the soil), blue N (N available for crop, like N applied with mineral and/or organic fertilizers, N applied with irrigation water and N mineralized during the crop period), whereas grey N is the amount of N-NO3- washed from the soil to the aquifer. All these components are expressed as the ratio between the components of water or N footprint and the yield (m3 t-1 or kg N t-1 respectively). The objetives of this work were to evaluate the impact derivated from the use of different fertilizer practices in a melon crop using water and N footprint.

Materials and Methods

During successive years, a melon crop (Cucumis melo L.) was grown under field conditions applying mineral and organic fertilizers under drip irrigation. Different doses of ammonium nitrate were used as well as compost derived from the wine-distillery industry which is relevant in this area. The water needs were calculated using FAO method as $ETc = Kc \times ETo$ (Doorenbos and Pruitt, 1977); Kc, the crop coefficient used (Ribas et al., 1995) and ETo is the reference evapotranspiration calculated by the FAO Penman–Monteith method (Allen et al., 2002). Each growing season, N uptake by the crop was determined. To quantify the N leached to groundwater the drainage was calculated weekly using a water balance ($D = Irr + P - ETc - Rf \pm \Delta\theta v$ (Doorenbos and Pruitt, 1977), where D is drainage, Irr is the irrigation applied, P is the precipitation, Rf is runoff that is considered negligible and $\Delta\theta v$ is the variation of the volumetric soil water content between two consecutive week; as well as the concentration of nitrate in the soil solution. In adition, N in soil at the begining and at the end of the crop cycle was measured. With all these parameters, N mineralized during the crop period was estimated.

Results and Discussion

The variability observed on the water footprint between years pointed out that this indicator is closely related to the climatic conditions found in each growing season. As all the treatments were watered based on the crop requirements, differences in the water footprint were related to the yield. The highest doses of inorganic fertilizer (MF243 and MF393) resulted in higher components of the N footprint, because substantial amounts of available N forms were applied through fertigation without increasing the yield. This fact resulted in an increase of the grey N footprint, because this excess of nitrates was transported with the drainage water and poses a risk of groundwater contamination. However, the most part of N applied with the organic fertilizer is in organic form and need to be mineralized during the crop cycle to be available for the crop or washed through the soil profile. For this reason, although the highest amounts of N were applied with compost, the components of the N footprint remained lower with respect to the highest doses of inorganic fertilization due to a slow rate of N mineralization.

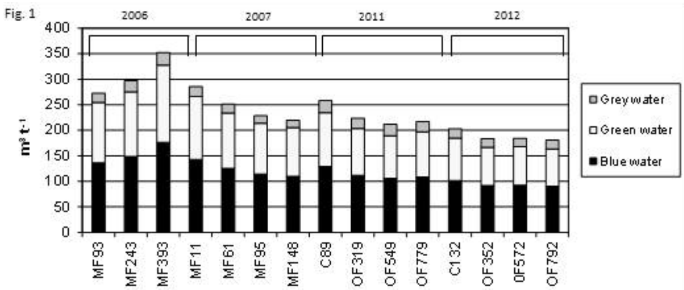

Figure 1. Water footprint for all the treatments (MF=mineral fertilizer (followed by kg N ha-1 applied with mineral fertilizer and irrigation water); C= control (followed by kg N ha-1 applied with irrigation water); OF=organic fertilizer (followed by kg N ha-1 applied with compost, considering N organic and N mineral, and irrigation water)).

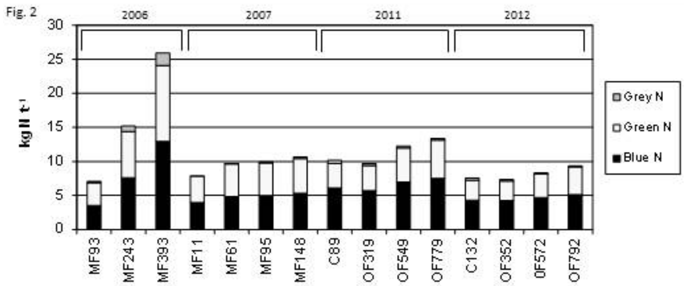

Figure 2. Nitrogen footprint for all the treatments (MF=mineral fertilizer (followed by kg N ha-1 applied with mineral fertilizer and irrigation water); C= control (followed by kg N ha-1 applied with irrigation water); OF=organic fertilizer (followed by kg N ha-1 applied with compost, considering N organic and N mineral, and irrigation water)).

Conclusions

Water footprint is dependent on the climate conditions of each growing season and the crop yield explains the differences obtained between treatments. The type of fertilizer as well as the application dose had an effect on the amount of the different components of the N footprint.

Acknowledgements

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WILL A CHANGING CLIMATE LEAD TO INCREASED NITRATE INDUCED EUTROPHICATION?

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Nitrate loss to water via leaching and overland flow contributes to eutrophication of freshwaters, transitional and near coastal waters (Stark and Richards, 2008). The Water Framework Directive requires member states to improve all waters to good status by 2015 and to prevent deterioration of water status. Agriculture contributes significantly to nitrogen (N) loading to waters and reducing N loss to water has been an important policy at both member state and EU levels for many years. Implementation of the Nitrates Directive has dramatically increased regulations and constraints on farmers to improve N management in regions at risk of nitrate loss to water. In addition to these legislative constraints, farmers also have to manage their systems within a changing climate that is resulting in more frequent extreme events such as floods and droughts. Previous research has linked changing climate to increases in riverine nitrate concentrations which are strongly linked with eutrophication (Justic et al., 2003). The objective of this study was to investigate the link between climate and the concentration of leached nitrate as an indicator of eutrophication potential.

Materials and Methods

Nitrate leaching was quantified under spring barley grown on a well-drained, gravelly sandy soil, under a temperate maritime climate, Co. Carlow, Ireland. The study design has been previously described (Premrov et al. 2014). Two published studies were conducted on the same long term trial and nitrate leaching was quantified over 6 drainage years under the same farming practices and treatments. In this paper the soil solution nitrate concentrations under spring barley grown by conventional inversion ploughing (CP) and reduced tillage (RT) with no over winter cover crop were compared to weather parameters over the 6 monitoring years. Soil solution nitrate (NO_3^--N) samples were collected using ceramic cups installed at 0.9m, with 4 cups per plot and 4 plots per treatment. In the CP treatment soil cultivation was by inversion tillage (mouldboard ploughing) to a depth of 20 cm in March each year, followed by tine harrowing and sowing. In the RT treatment the soil was shallow tine cultivated (10-15cm) in September using a Horsch Simba Terrano 3FX (Horsch Maschinen GmbH, Schwandorf, Germany) followed by rolling with some further cultivation, using the discs of the sowing machine during sowing of the subsequent spring barley crop in March. Barley was sown in both treatments using a Vaderstad Super Rapid S 300 drill. Weather was recorded at a national Met Eireann weather station on site and the data collected was used to estimate soil moisture deficit (SMD) and effective rainfall using an SMD-crop model (Premrov et al., 2014).

Results and Discussion

Soil solution NO_3^--N varied significantly between years. The average soil solution concentrations observed across the 6 winter drainage periods can be seen in Fig. 1. Within individual years NO_3^--N concentrations varied over the drainage season, with peak concentrations generally observed in the autumn time, decreasing thereafter. Overall, there was no significant effect of tillage practice (RT v. CP) on soil solution NO_3^--N concentration. Within the RT treatment the overall mean annual NO_3^--N was 36.2 mg/L (range: 21.0 to 65.6 mg/L) and within the CP treatment mean was 34.5 mg/L (range: 20.8 to 60.8 mg/L). Under both treatments there was a three-fold difference in mean annual soil solution NO_3^--N concentration over the 6 years with no change in the agronomic practices (crop type, tillage type and fertiliser input). This indicates that there were other factors influencing the NO_3^--N concentrations beneath the treatments. Rainfall is one of the drivers that influences the rate and volume of leachate moving through soil, crop moisture availability and soil microbial N processes.

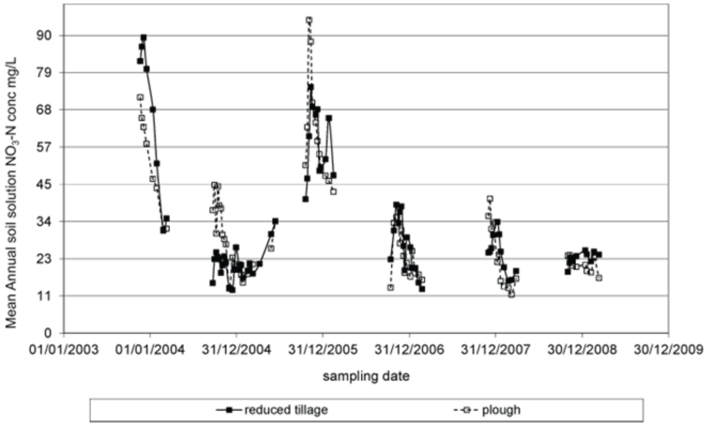

Figure 1. Mean soil solution NO_3^--N concentrations from spring barley grown under reduced tillage and conventional ploughing, over a 6 drainage year monitoring period.

Soil solution NO_3^--N concentrations were negatively correlated with total annual rainfall and total effective drainage and positively with mean and maximum SMD. The relationship between rainfall and NO_3^--N concentrations is expected as the soil nitrate pool would be diluted with higher drainage volumes transported through soil. The relationship with SMD is more complex as it influences crop growth which would become restricted at higher deficits, thus restricting soil nitrogen uptake. Soil microbial processes like mineralisation are also strongly influenced by soil moisture. The strong relationship between NO_3^--N concentrations and climatic drivers suggests that climate change may increase the impact on receiving waters and this requires new solutions for N management.

Conclusion

Soil solution NO_3^--N concentrations are strongly linked with climatic drivers and under climate change the impact of NO_3^--N loss to water on eutrophication is expected to increase. The increased impact on eutrophication of waters, related to climatic variability, requires new management solutions to help farmers maintain or improve the sustainability of their farming systems.

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NITROGEN FOOTPRINT: NITROGEN USE AND NITROGEN LOSS RELATED TO FOOD PRODUCTION IN CHINA

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Human activities substantially altered the global nitrogen (N) cycle, in particular in increasing reactive N (Nr, all species of N except N_2) release to the environment, resulting in a series of environmental and health problems (1). China has long been the world's most populous nation; its contribution to the global problems has garnered special attention (2). The increasing Chinese food demand propelled by an increase in population and greater per capita income anticipated through past decades may continue for future decades and require the substantial Nr creation and use. China has put great effort to maximize crop productivity in order to ensure food security. However, the intensive use of existing croplands and low N fertilizer use efficiency aggravated the phenomenon of N imbalance in China's agricultural development (3). In addition, changes in the role of animals in Chinese farming and transformation to more high-protein dietary systems imply a greater N demand and loss related to food production (4). N footprint (NF) is used as an environmental indicator to evaluate the burden of Nr. Because previous studies haven't connected the NF with the each individual food type; the tradeoffs of the alternative diet patterns during the rapid urbanization in China are still unclear. The assessment of NF at the product level is necessary to look forward a balanced food structure, which can meet both the demands of food security and friendly environment.

Materials and methods

Our NF is defined as an indicator of quantitative Nr losses of vegetable and animal food systems from the perspective of food production. A combination of top-down and bottom-up approach is used to determine China's NF from both crop and livestock farming systems and to allocate these NF to the supply of individual 8 vegetable or 6 animal food categories for the given year 2009. We further measured virtual N factor (VNF) for each food type as an indicator to estimate the Nr used in food production but not embedded in the final product. To investigate how changes in domestic diets affect NF in the future urbanization, we started with projections of the per capita consumption of a food crop or animal product and then determining the N uses and losses due to grow the product by 2020 and 2030 based on the assumption that all food availability is used for the consumption demand of China. Four scenarios on consumption pattern are further developed including a baseline scenario and three nutritional scenarios recommended by Chinese Nutrition Society (CNS).

Results and discussion

Total N use for food production was 55.6 Tg in China in 2009; only 13.2 Tg N accumulated in primary food products. As a result, NF of food production was 42.5 Tg N in China in 2009. Production of vegetable foods summed 29.6 Tg N, of which 5.0 Tg N was lost as the use of domestic crops feed for animal production; while animal foods summed 17.8 Tg N. Cereals were the single largest source of NF, accounting for

30% of total NF, followed by bovine meat and vegetables with 18% and 17%, respectively. Only 1.1 Tg NF from exported food. The larger fraction (about 60%) of NF embodied in exported animal food implied the potential Nr burdens experienced by China due to the unreasonable export structure. VNF varied substantially between different food types with the highest value for fruits and bovine meat in vegetable and animal products, respectively. For the delivery of 1 kg of food N to the consumption stage, food production processes of fruits and bovine meat lost 17.6 and 53.3 kg N to the environment. High N inputs of fertilizer and feed, and the low use and conversion efficiency resulted in the higher VNF of fruits and bovine meat. Under the baseline scenario, the production-based NF for domestic consumption will increase to 43.9 Tg in 2020 and 44.4 Tg in 2030. If adopting the median energy diets recommended by CNS, the corresponding NF will reduce about 13% in 2020 and 2030. Hence, the diet pattern under baseline scenario appears a higher Nr burden because the larger consumption of animal foods than recommended scenario.

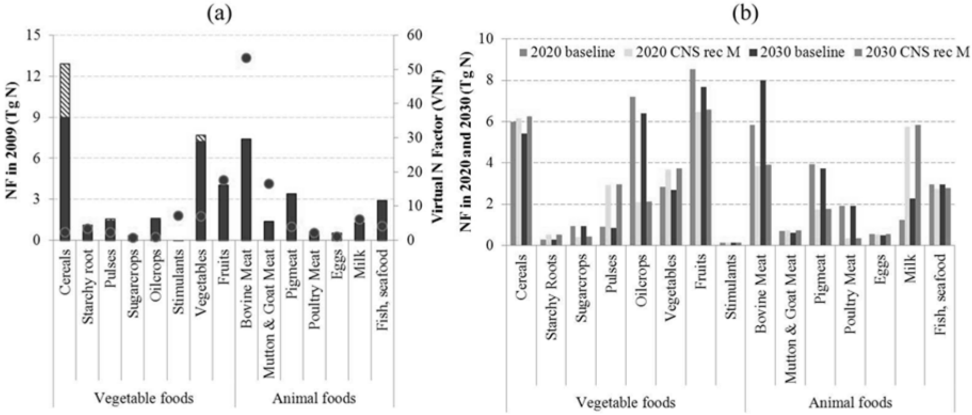

Fig. Nitrogen footprint (NF) by food type. (a) NF and VNF in 2009, the red circle represents the VNF, the column represents the NF, while column with stripe represents the NF of vegetable foods as feed; (b) projection and scenario analysis of NF in 2020 and 2030, CNS rec M represents the median energy diet recommended by China Nutrition Society.

Conclusions

NF is projected to increase due to the increasing food demand. Technical improvement is the essential measure to enhance N use efficiency and reduce Nr use and loss. Animal foods commonly have a larger impact than vegetable foods on the Nr loss in food production; the VNF of bovine meet is the highest, nearly up to 70 times of lowest that of sugarcrops. Diet shifts offer practicable opportunities to reduce environmental Nr impacts in the food production sector on a low-cost basis. Adjustments of food export structure also possess a certain potential to mitigate the Nr burden. An improved N management and a N balanced diet pattern should be both implemented to integrate agricultural, environmental and nutritional objectives.

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QUANTIFICATION OF NITROGEN IN THE N2013 FOOD CHAIN

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The human use of reactive nitrogen (N_r) in the environment has profound beneficial and detrimental impacts on all people. Its beneficial impacts result from food production and industrial application. The detrimental impacts occur because most of the N_r used in food production and the entire amount of N_r formed during fossil fuel combustion are lost to the environment where it causes a cascade of environmental changes that negatively impact both people and ecosystems Galloway et al., 2003; Sutton et al., 2011, 2013. It is a challenge in communicating to the public the complexities of nitrogen's interactions with the environment. One way to address this is through a nitrogen footprint model which is the total nitrogen released to the environment as a result of food production and consumption, energy use and other forms of consumption and helps consumers understand their role in nitrogen losses to the environment. Leach et al., 2012; Leip et al., 2013b. The objective was to quantify the N lost during food consumption for conference events held at the Munyonyo Resort Speke Hotel in Kampala City so as to estimate the event's impact on the Nr balance of a low-input agricultural system. This would inform Nr management interest groups on planning to minimize the negative impacts of such necessary activities on the environment.

A survey was carried out at the Speke Resort Hotel registering total supply, consumption, recycling and wastage for all meals served for two events: a pre 6th International Conference N2013 meeting of 200 participants that served to provide baseline information of normal hotel procedures, and the N2013 where food consumption was controlled, for comparison. In contrast to the baseline conference, chefs and participants of N2013 were aware of the N-impact related to their cooking and eating habits. Also, N2013 attendants participated to an N impact compensation scheme (Leip et al., 2013a).

We used a model of the farming N-loss factor that calculates all losses of nitrogen caused by cultivation of the food product, processing and transport to the point of resale for the N2013 conference. For animal food ingredients N-losses in feed cultivation and animal raising was quantified. At each stage of food production, only some N proceeds to the next step. The remainder is either recycled back into the system or lost permanently to the environment. The total N loss to the environment is summed over the journey from growing crop to prepared meal. The amount of N lost in producing that final food is divided by the N available for consumption. For the main ingredient/farming system combinations, the farming N-loss factor was quantified basing on what happens in their farming systems.

Data from the baseline event indicated that meat consumption was up to 24.3g/cap/day which is 3 times higher than the FAO recommended 8.3g/cap/day. Pre-informed food preparation and consumption during N2013 showed a reduction of all food staffs with a marked (5 times) reduction in the amount of meat consumed, which was compensated for by increase in consumption of vegetables from 7-11% and fruit from 10-17%, a positive step towards reducing the Nr footprint and its impacts on the environment. Food wastage was 16%. Results also showed a reduction in wastages of all food staffs with increase in wastages of fruits from 5-12%because chefs prepared more fruits than what was anticipated. The findings of the N2013 conference have demonstrated that besides savings in terms of money not spent to buy food in excess of what is actually required, it is possible to reduce the N footprint by conforming to acquisition, preparation, consumption and re-use/recycling of food based on the awareness about the N footprint. There is reduction of N-foot print of the N2013 event than the baseline event, mainly linked to the reduction of meat consumption foods from 24.3g/cap/day of baseline to 4.5g/cap/day for N2013 event. This is a useful indicator for environmental problems linked to N emissions and N depletion, hence an important tool in educating Nr management stakeholders. The reduction of N-foot print of the N2013 event indicates the potential contribution of future international nitrogen conferences to awareness-based reduction of the N footprint with net gains to both people and the environment.

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EUROPEAN DIETS, NITROGEN USE, EMISSIONS AND IMPACT ON HUMAN HEALTH

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Diets in the European Union are, compared to the global average, characterised by a relatively high consumption of animal products, although there are there differences between individuals and also between different countries (Westhoek et al., 2011). Current European diets lead to overconsumption of saturated fats and proteins compared to dietary guidelines, where especially the intake of saturated fats is related to the consumption meat and dairy. Recent research has highlighted the large differences in nitrogen footprints between different food products, with animal products usually having considerably larger foot prints than vegetable products as is demonstrated with life-cycle analyses (Leach et al., 2012; Leip et al., 2013). Given the environmental of meat and dairy production and the health impacts of consumption, we examined the effects of a considerable reduction (by 25 to 50%) of meat and dairy in the EU (Westhoek et al., 2014).

Method

The lower consumption of meat, dairy and eggs was assumed to be compensated on an energy basis by additional cereal consumption. The assumption was made that a reduction in the consumption of meat, dairy and eggs would have a proportional effect on livestock production within the EU. The lower demand for feed was quantified starting with data on current feed use from the CAPRI-model. We discriminated four main feed components (protein-rich feed, energy-rich feed cereals, roughage and forage maize). All calculations were done per animal category and per country. The effects on land use, nitrogen flows and GHG emissions within the EU were quantified with the model MITERRA-Europe. This is an environmental impact assessment model that calculates various types emissions of reactive nitrogen, greenhouse gas emissions (carbon dioxide from soils, methane and nitrous oxide) on a deterministic and annual basis using emission and leaching factors.

Results and Discussion

The results show that halving the consumption of meat, dairy products and eggs in the EU reduces nitrogen pollution from the food system by 40% and greenhouse gas emissions by 25-40%. The per capita use of cropland for food production is reduced by 23%. Due to the lower intake of saturated fats (up to 40% lower) human health risks are lowered. Ecological risk and to some extent human health risk, are also lowered by the reduced emission of ammonia to air and nitrate to ground and surface waters. The EU becomes a net exporter of cereals. The use of (mainly imported) soybean meal is reduced by 75%. The nitrogen use efficiency (NUE) of the food system increases from the current 18% to 41-47%, depending on choices made regarding land use. The results demonstrate the large share of livestock production in the total environmental impact of EU agriculture. Of course the environmental benefits within the EU would be less if livestock production within the EU would not follow the lower consumption, in which case the surplus meat and dairy could be exported.

As the general public is not really aware of issues around nitrogen use and emissions, other considerations as reduction of greenhouse gases, animal welfare and personal health will be more important drivers for change. As there is a clear correlation between nitrogen and greenhouse gas footprints of the various food commodities (Leip et al., 2013; Lesschen et al., 2011; Weiss and Leip, 2012), the direction of change to reduce emissions is the same. Increasing animal welfare will usually lead to higher nitrogen and greenhouse gas emissions, partly due to a higher feed demand (Westhoek et al., 2011) and partly due a more difficult recovery of manure in case of free range systems. Reducing food waste will be beneficial for most environmental impacts. Governments and food industry and retail are already working together to reduce food waste. Governments and private actors are currently quite reluctant to interfere with consumers choices, although especially retailers have a large effect on consumer's choices.

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NITROGEN FIXATION OF SELECTED FORAGE LEGUMES FOR SMALLHOLDER FARMERS IN UGANDA

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Access to reliable forage of sufficient quality, especially during the dry season, poses the main challenge to smallholder dairy producers in semi-arid areas of East Africa (Hall et al., 2007). In contrast to most other crops, many forage species can be grown on marginal lands and thus provide an opportunity for farmers to build a livelihood. Legumes offer the extra benefit of improving the nitrogen-poor soils. Therefore, in this study, five forage legumes were tested for their ability to provide biomass and fix nitrogen (N) in a field trial in Uganda. We hypothesized that the legumes would differ in their stable N isotopic signatures, used here as a proxy for N fixation, and that the N yield would differ depending on water availability.

Materials and Methods

Five forage legumes, namely *Lablab purpureus*, *Desmodium uncinatum* cv Silverleaf, *Desmanthus virgatus*, *Macroptilium bracteatum* cv Burgundy bean, and *Canavalia brasiliensis*, were grown in a completely randomized block design (plots of 3 m x 6 m, 1 m in between plots) with five replicates with or without additional irrigation (by hand, if no precipitation had fallen the previous day; over the growing season 23,100 l water added/plot) in field sites in Uganda (National Livestock Resources Research Institute, Tororo district; annual rainfall 1130-1720 mm; AATF, 2009). On average, the soil water content of the non-irrigated plots was about 14% lower than in the irrigated ones (measured at five occasions throughout the season). Planting was done in the rainy season (October 2012) at recommended rates and spacing. Biomass was harvested 10 cm above the ground five times in two-monthly intervals until June 2013. Samples of about 200 g were oven-dried (60°C for 48 hours) and weighed. Just before harvest, the youngest leaf of several plants was sampled for stable isotope analysis at the first four harvest occasions. Isotope and N content measurements were done on an isotope ratio mass spectrometer (Delta plus Finnigan MAT, Bremen, Germany) coupled to an elemental analyser (NA2500 CE lnstruments, Rodano, Milano, ltaly) via an interface (Conflo lll Thermo Electron Cooperation. Bremen, Germany). Isotopic values are given as $\delta^{15}N$ values (‰; standard: air). Statistical analyses (ANOVA, harvest as random factor, testing for normality and homogeneity of variances) were done with SPSS version 16.

Results and Discussion

The total dry matter production of the legumes was on average about 600 g m^{-2}, with small, albeit not significant differences among species and between irrigation treatment (Table 1). The nitrogen isotopic values were most depleted for *L. purpureus* and *D. uncinatum*, intermediate for *M. bracteatum* and *C. brasiliensis* and most enriched for *D. virgatus* (Table 1, $P < 0.001$), suggesting a potentially larger proportion of N derived from air for *L. purpureus* and *D. uncinatum*. However, so far no B values of the plants are available. The percentage of N in the plant material was smallest for *M. bracteatum*, intermediate for *D.*

uncinatum and *D. virgatus* and largest for *L. purpureus* and *C. brasiliensis* (data not shown, $P < 0.001$). Irrigation had no significant influence on $\delta^{15}N$ or N percentage ($P = 0.196$ and 0.961, respectively). This is not in line with our hypothesis and earlier studies (e.g. Ledgard and Steele, 1992). Least water was available at the third harvest, when the gravimetric soil water content fell to on average 17% in the irrigated plots, with that in the non-irrigated being around 72% of that value. Even at this harvest, no significant difference was observed in $\delta^{15}N$ values. Over time, the combination of larger biomass yields and changes in N content led to the observed increase in N yield (Fig. 1), with largest N yields from *L. purpureus* and *C. brasiliensis*.

Conclusions

L. purpureus and *C. brasiliensis* seem most promising in terms of biomass production and N yield under the conditions tested. The missing effect of drought stress on the N fixation has to be further tested.

Table 1. Total annual dry matter yields of five forage legumes over five harvests (g m-2) and δ15N signatures of their youngest leaves at the fourth harvest. Shown are means and standard deviations (n = 5)

	Lablab purpureus	*Desmodium uncinatum*	*Desmanthus virgatus*	*Macroptilium bracteatum*	*Canavalia brasiliensis*
	Dry matter yield [g m^{-2} a^{-1}]				
Irrigated	701 ± 114	530 ± 71	602 ± 143	542 ± 47	704 ± 105
Non-irrigated	635 ± 61	447 ± 44	625 ± 126	508 ± 67	634 ± 59
	$\delta^{15}N$ [‰]				
Irrigated	0.80 ± 1.49	0.43 ± 0.39	2.42 ± 0.67	1.09 ± 0.28	1.85 ± 0.69
Non-irrigated	1.78 ± 0.66	1.02 ± 0.59	2.72 ± 1.18	1.63 ± 0.80	1.86 ± 1.12

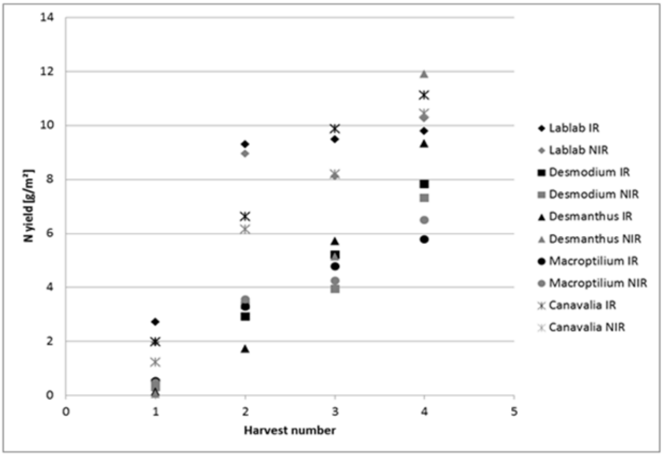

Figure 1: Nitrogen yield of five legume species grown with (IR) or without (NIR) irrigation. Shown are averages (n = 5) for four harvests from December 2012 until April 2013.

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EFFECT OF FERTILIZER APPLICATION ON NITRIFICATION ACTIVITY AT ELEVATED TEMPERATURE SUBSEQUENT TO SOIL SOLARIZATION

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Materials and Methods

Nitrification activity was defined as the net cumulative NO3-N released from the added NH4-N during incubation. In brief, 10 g dry weight equivalent of moist soil (60% of maximum water holding capacity) was mixed with NH4-N (350 mg kg–1) in 250-mL plastic bottles, the mouths of which were then covered with a plastic wrap and the bottles incubated in the dark. Incubation period was 2 weeks in the Experiment 1, and was 1 week in the experiment 2. Experiment 1. Effect of Soil Solarization, and Fertilizer Application on Nitrification Activities at 45°C Four subplots were established which received the following treatments over the two years (2010 and 2011) in an experimental field: (1) solarized and fertilized (SF); (2) not solarized but fertilized (UF); (3) solarized but not fertilized (SN); and (4) not solarized and not fertilized (UN). In the fertilized plots, organic fertilizer was applied (16.5 g N m–2 in 2010; 15 g N m–2 in 2011). The organic fertilizer used was Agret 6-6-6 (Asahi Industries, Co., Ltd, Tokyo, Japan). Soil samples for determination of nitrification activity were collected in July 2012. Experiment 2. Effect of Soil Solarization under Organic or Conventional Farming on Nitrification Activities at 45°C Field experiments were conducted during 2008-2013 in another experimental field. Eight subplots were established, and lettuce and carrot were grown every year. In the organic farming plots, Agret 6-6-6, poultry manure compost, and fish scrap were applied. For plots subjected to soil solarization, the treatment was conducted once in the year during 2008-2012. In the conventional farming plots, ammonium sulfate, magnesium multi-phosphate, and potassium sulfate were applied. For plots subjected to soil solarization, the treatment was conducted in 2012. In the conventional plots, chemical herbicides, insecticides, disinfectants are used.

Results and Discussion

In experiment 1, nitrification activity at 45°C tended to be higher in the solarized subplots (Figure 1). This result was consisted with our previous study. Nitrification activity at 45°C tended to be SF > SN, but UN > UF. This result supports a hypothesis that nitrifying microorganisms which are active at 30°C and which are more active at 45°C are compete against each other, and fertilizer application with the solarization stimulated a shift in the population toward nitrifying microorganisms that are more active at higher temperatures. In experiment 2, Nitrification activity at 45°C was significantly higher at solarized plots under organic farming management treatments, either (Table 1). In the conventional farming plots, nitrification activity at 45°C tented to be higher at solarized plots but not significant.

Conclusions

Change in nitrification activity subsequent to soil solarization successively detected by a new assay method, nitrification activity at 45°C, in two different field sites.

Acknowledgement

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Table 1 Residual effect of solarization and manure application under organic or conventional farming on nitrification activity at 45°C (mg kg-1) (Experiment 2) +S2008-2012: solarized in 2008–2012, +S2012: solarized in 2012, –S: had never solarized +M: cattle manure compost was applied in 2008-2012, –M: manure compost had never applied. †Means in a column followed by the same letter are not significantly different according to Tukey's HSD test at the 5% level.

Treatments			NO_3 production during incubation	Treatments			NO_3 production during incubation
Organic	$+S_{2008\text{-}2012}$	+M	105 ± 16 a	Conven	$+S_{2012}$	+M	24 ± 9 a
	$+S_{2008\text{-}2012}$	-M	105 ± 3 a	-tional	$+S_{2012}$	-M	9 ± 10 a
	-S	+M	19 ± 7 b		-S	+M	11 ± 5 a
	-S	-M	17 ± 1 b		-S	-M	1 ± 3 a

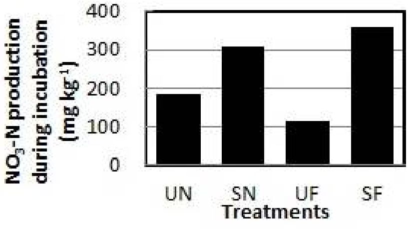

Figure 1 Nitrification activity at 45°C in an experimental field with plots of different management treatments (Experiment 1) UN, SN, UF, and SF are management treatments described in the text.

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