Recent Research in Science and Technology 2010, 2(1): 014–022
ISSN: 2076-5061
www.recent-science.com

PLANT BIOLOGY & AGRICULTURE

COMPARING MINERAL AND ORGANIC NITROGEN FERTILIZER IMPACT ON THE SOIL-PLANT-WATER SYSTEM IN A SUCCESSION OF THREE CROPS

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Abstract

In the belief that the investigation of the whole system helps to avoid the risk of incomplete or misleading responses resulting from the analysis of the single segments, a research was conducted in a succession of three crops (lettuce, red chicory and celery) to investigate in an integrated approach the different response of the soil-plant-water system to mineral and organic nitrogen fertilization.

The experimental plan included the application of two amounts of fertilizer, corresponding to 240 and 360 kg ha^{-1} N under mineral or organic form per crop cycle, plus a control, in three replications.

Mineral N resulted more promptly available to plants and increased the fresh and dry weight and protein content in leaves of the three crops while no significant difference in the tissue moisture content between the treatments was found. The inspection of combined data resulting from soil, plant and water analysis and from N budget demonstrates that altogether more mineral N was released in soil and water from the organic fertilizer while more N was uptaken by plants with the mineral fertilizer.

Nitrogen uptake efficiency and N use efficiency in fact were highest in the mineral fertilized plots while surplus N was only found with the organic fertilization.

Microbial population in the soil was unaffected by the type and amount of fertilizers; on the contrary, enzymatic activity responded positively to organic N while was depressed by the synthetic N form. The results suggest that the use of organic N integrated with mineral N at the appropriate crop stages is the solution to be recommended.

Key Words: Lettuce; Red Chicory; Celery; Sustainable Agriculture; Integrated Fertilization; Nitrogen Use Efficiency.

Introduction

Ecosystem pollution depending on agricultural practices has prompted a number of investigations which often yielded non consistent or even contradictory results. In the present research an “integrated” experimental approach was planned taking into consideration simultaneously the three segments soil-water-plant, in the belief that such an approach can reduce uncertainties resulting from the researches oriented to explore single segments.

The rules of organic farming ban the use of synthetic nitrogen fertilizers on the assumption that they impair soil fertility and enhance N leaching (e.g. Haas et al., 2002) while organic fertilizers are supposed to enrich the soils in organic matter and have less N losses.

While soil enrichment in organic matter is unquestionable, reduction of losses has been questioned, mainly on the basis of an assumed lack of synchronization between N release by organic matter and uptake by crops, which can lead not only to an insufficient availability to crops in their critical phenophases (e.g. Myers et al., 1997; Pang and Letey, 2000; Shepherd, 2002; Berry et al., 2002) but also -as a consequence- to the loss of unused untimely mineralized nitrogen (e.g. Bonde and Rosswall, 1987; Yadvinder-Singh et al., 1992; Kirchmann and Thorvaldsson, 2000; Bergström et al, 2005; David et al., 2007; Evanylo et al, 2008).

Further uncertainties regard organic fertilizers action on soil bacterial population: to quote Shannon et al.

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(2002) "*While a number of studies have indicated that organic farming practices have beneficial effects on soil microbial numbers, processes and activities, other research has concluded that organic management has zero or even negative impact on soil microorganisms; published data show no consistent trends that may be attributable to different management practices.*"

Furthermore it is generally accepted that the application of organic N fertilizers stimulates soil microbial activity while mineral forms depress it (e.g. Crecchio et al., 2001; Alloway and Jackson, 1991; Benckiser and Simarmata, 1994; Albiach et al., 2000; Perucci et al., 2000; Debosz et al., 2002; Bell et al., 2006). Although the determination of enzymatic activities, particularly of proteolitic and ammonia-oxidizing enzymes, is afflicted by uncertainties depending on the vast number of intervening factors, including variability in soil conditions in space (pedological properties) and time (moisture, aeration and temperature) and on differences in analytical methods, yet it can be of interest to assess their relative variation in response to different fertilizers.

In order to achieve a thorough view of the concurring factors it was believed of interest to investigate also the N budget in the soil-plant-water system and the variations in microbial population and activity. As a consequence a research was planned aiming at observing the influence of two high doses of organic and mineral N fertilizer applied to a succession of three different crop cycles on

1. plant production and their protein content
2. the different N amounts uptaken by plants
3. the different amounts of residual N in the soil and drained water
4. the variations in soil enzymatic activities and
5. the variations in soil microbial population

Also some indicators for assessing the different impact of organic and mineral fertilizers on nitrogen use efficiency and budget were applied, as illustrated below.

Materials and Methods

The experiments were conducted in Eastern Sicily, south of Catania (latitude 37°20' N, longitude 12° 30' E) at an elevation of 135 m above sea level.

Fifteen lysimetres were installed sized m 3.0 x 4.0 x 1.0, hydraulically insulated by means of a lining membrane; the soil is an alluvial sandy loam whose principal physical and chemical characteristics are reported in table 1. Lysimetres were irrigated by means of an automatic irrigation system and each of them was equipped with a drainage system permitting to sample drained water. Emitters were in-line, self compensating 2 L/h drippers spaced 30 cm along the laterals and the system had a high application uniformity, with a "Field Emission Uniformity Coefficient" as defined by Karmeli and Keller (1974) in excess of 90 %. Irrigation was not a variable since the same water amounts were applied to all the treatments with some excess water to obtain a sizable amount of drainage.

Three crop cycles were grown in succession: lettuce (*Lactuca sativa* L.: June – August 2005), red chicory (*Cichorium intybus* L.: October – December 2005) and celery (*Apium graveolens* L.: June – November 2006). In every lysimetre 66 plants were planted, with a population density corresponding approximately to 55.600 plants ha^{-1} considering the net surface of lysimetres.

While one treatment in three lysimetres was the unfertilized control, six lysimetres were fertilized with ammonium sulphate containing 21% N and six with a liquid organic fertilizer containing 8.5 % N ("Protamix", derived from hydrolyzed animal epithelium).

In a further subdivision, three lysimetres in each fertilized group received N amounts corresponding to 240 kg ha^{-1} and three lysimeters amounts corresponding to 360 kg ha^{-1} per crop cycle. It was decided to apply such relatively high quantities because the objective of the research was not to optimize fertilizer quantities but to assess and compare their polluting potential. Treatments were labeled as control, T1, T2, T3, T4 as illustrated in table 2. No chemical other than N fertilizers was applied to the crops and all the cultural practices, including weeding, were made by hand.

Nitrogen was simultaneously applied in solution to the twelve fertilized lysimetres through the irrigation system; irrigation and fertilization were managed through a central automatic control station.

Soil was sampled at irregular intervals, depending not only on the planned crop schedule but also on the rain events; at every sampling three soil samples from the rhizosphere were taken in each lysimetre. Fourteen samplings were made during the 18 months of the experiment, with a total of 630 soil samples (3 samples per lysimeter x 15 lysimeters x 14 times), from the depth of 10-30 centimeters.

Ninety plants, six per lysimetre, were sampled at the end of every cycle, 270 plants in total, and the aerial parts and roots were separately analyzed.

Drained water was sampled 14 times with a total of 210 samples (14 times x 15 lysimetres); parameters analyzed in soil, plants and water are reported in table 3.

Table 1. Selected soil characteristics

Sand %	58.7
Silt %	21.2
Clay %	20.1
Hydraulic conductivity (mm h^{-1})	28
pH (25°C)	7.54
CE (dS m^{-1}, 25°C)	2.06
N org (‰)	1.21
NO_3^- (mg kg^{-1})	11.49
NO_2^- (mg kg^{-1})	0.65
NH_4^+ (mg kg^{-1})	3.94
N Kjeldhal (‰)	1.23
Organic carbon (g kg^{-1})	11.81
Organic matter (‰)	20.36
C/N	9.60
P-available (mg kg^{-1})	1024
P-soluble (mg kg^{-1})	11
K^+-available(mg kg^{-1})	55
Ca^{2+}-available (mg kg^{-1})	209
Mg^{2+}-available(mg kg^{-1})	96

Table 2. Description of the treatments

Treatment	N amount (kg ha^{-1})	N source
control	--------	--------
T1	240	mineral
T2	360	mineral
T3	240	organic
T4	360	organic

Table 3 reports the list of analyses in the three segments of the system.

Segment	Parameter
Soil	Total n
	Organic n
	Mineral n (n-no_3^- , n-nh_4^+)
	Arginine-deaminase, protease activity
	Protease-producer eubacterial and actinomycetes population
	Ammonia-oxidizing bacterial population
Plant	Fresh weight
	Dry weight
	Total n
	Total proteins
	Mineral n (n-no_3^-, n-nh_4^+)
Water	Organic n
	Mineral n (n-no_3^-, n-no_2^-, n-nh_4^+)

Table 3. Parameters analyzed in soil, plants and drained water

The analytical procedures adopted were the following:

- Total N: Kjeldhal (SISS, 1985)
- N-NO_3^- : Cataldo et al. (1975)
- N-NO_2^- Bates (1973)
- N-NH_4^+: Berthelot (1959)
- Organic N: as the difference between its total and mineral forms
- Arginine-deaminase activity: Alef and Kleiner (1986)
- Protease activity: Ladd and Butler (1972)
- The protease-producer eubacterial and actinomycetes populations, and the ammonia-oxidizing populations were estimated according to the DGGE technologies (Denaturing Gradient Gel Electrophoresis). The DNA was extracted from the soil samples by means of the QIAamp DNA Stool Mini Kit and was subsequently amplified by means of the PCR (Polymerase Chain Reaction). The DGGE analysis of the eubacterial community was done according to the Heuer et al. (1997) procedure, whereas for actinomycetes the procedure of Weisburg et al. (1991) was followed.
- Proteins: Bradford (1976)

Soil temperature was monitored throughout the experiment by means of two geothermometres placed at the depth of 10 and 30 centimeters while rainfall height was recorded by means of a pluviometer. Available moisture in the soil was monitored through five batteries (one per treatment) of three tensiometres, placed at the depth of 10-30-60 cm.

The analysis of variance was conducted using the Statgraphics Centurion package; through the Multiple Range Test it was possible to appreciate which means were significantly different from the others ($P \leq 0,05$).

Results

Environmental data

Fig. 1. Soil temperature evolution at two depths

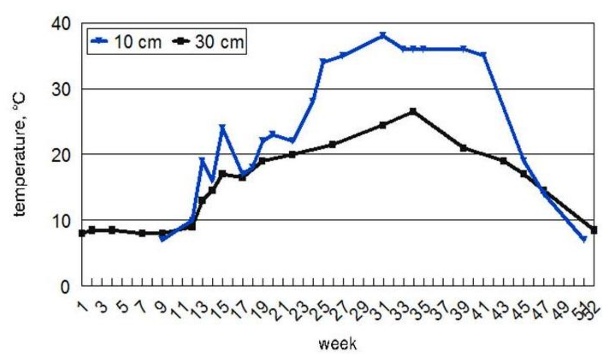


Table 4. Monthly precipitations

J	F	M	A	M	J	J	A	S	O	N	D
91	64	117	44	---	14	44	6	27	60	83	198

Figure 1 and table 4 show the evolution of soil temperature at the depth of 10 and 30 cm and monthly precipitations, respectively, both averaged over the two years 2005 and 2006; the average annual total precipitation of 748 mm exceeded by almost 50% the local long-term mean. Tensiometric data are not reported because not relevant: matric water potential in the soil was in fact always maintained above -30 kPa.

Nitrogen residues in soil and water

No statistically significant difference ($P \leq 0,05$) in the content of total and organic nitrogen in soil and drained water was in any case evidenced between the various treatments throughout the period of the experiment and as a consequence data are not reported. This result was expected, due to the well known limited reactivity of N in soil organic matter (SOM), which notoriously is by far prevalent on the mineral fraction.

On the contrary, differences were found among the treatments in the content of NO_3^- and NH_4^+ in the soil. When the entire sets of soil data throughout the three cycles were considered the Multiple Range Test analysis evidenced two homogeneous groups of mean values: one including the control and treatments T3 and T4 and a second one including treatments T1, T2, T3 and T4.

Although the mineral N content resulted always highest in treatment T2, statistically significant differences could only be found when T2 means were compared to those of the control whereas no significant differences could be detected among the four fertilized treatments, due to the rather high scatter of analytical results.

However when only the results of "snapshot" analyses referring to single sampling days rather than means were compared, almost constantly higher mineral N values in soil and water were found in treatments T1 and T2; not surprisingly the ranges of variation in T1 and T2 were much wider compared to T3 and T4.

In figures 2 and 3 the evolution in time of mineral N in the soil in its NH_4^+ and NO_3^- forms during the experiment is reported, showing the three peaks corresponding to N applications during the initial phases of the three crop cycles and the following decrease depending on plant uptaking and the leaching after the rainfall events. It is also possible to appreciate the smoother N variations in the organically fertilized treatments T3 and T4.

Figure 2. Evolution of N-NH4 in the soil

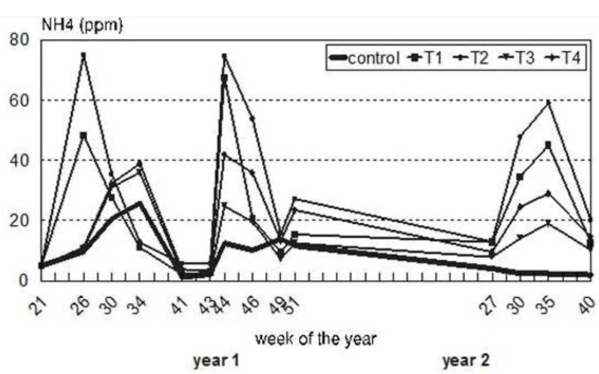


Figure 3. Evolution of N-NO3 in the soil

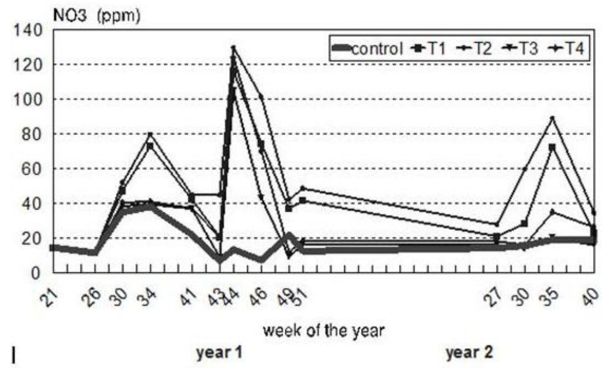
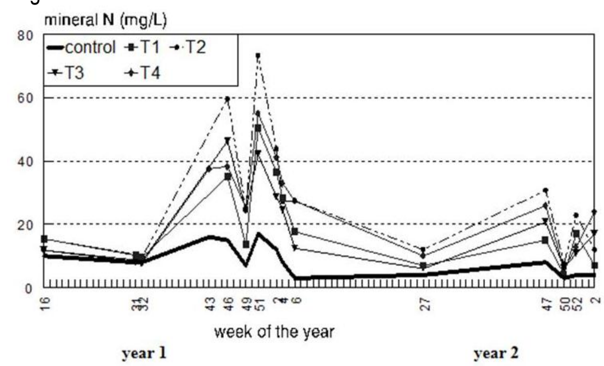


Figure 4. Evolution of mineral N in drained water



Interesting information is conveyed by the scrutiny of figure 4 showing the evolution of mineral N concentration in drained water, with the three peaks shifted a few weeks later compared to those in the soil. Throughout the experiment the N content in the drained water was constantly higher in the four fertilized treatments than in the control. A difference can be appreciated among the fertilized treatments, with mineral N content ranking almost always in the order T2>T4>T1>T3. However after the conclusion of the third crop cycle (celery was harvested at week 46 of the second year) a sharp decrease in N concentration was recorded, due to the heavy precipitations occurred, followed by a rise in all the treatments. Thereafter in the two subsequent weeks the N concentration decreased in treatments T1 and T2, steadily increasing in T3 and T4, so that the final ranking became T4>T3>T2>T1, thus giving evidence of a higher residual content of mineral N in the organically fertilized plots and confirming the results of plant analysis and N budget illustrated below.

Results in plants

Chicory crop was damaged by a frost in December 2005, which explains its remarkably low yield.

Fresh and dry matter content of the aerial part of plants as well as their content in moisture and total protein are reported in table 5 showing the significantly higher yields in mineral treatments T1 and T2, compared to the organic treatments T3 and T4.

It is also possible to see how the higher doses of N did not impact the moisture percentage in plant tissues, contrary to what is commonly believed: the moisture percentage in treatment T2 was in fact consistently lower than in the other treatments although the difference was not statistically significant. This is supported by the higher yield of chicory in treatments T1 and T2, in spite of the frost event notoriously affecting more the more aqueous tissues. Also the content in total proteins was higher in treatments with mineral fertilizer, but the difference achieved a statistically significant level only in the case of lettuce.

Table 5. Fresh and dry weight, moisture percentage and total protein content in plant aerial part

plant	treatment	fresh weight (g plant^{-1})	dry weight (g plant^{-1})	moisture content %	protein content (mg g^{-1} fresh matter)
lettuce	control	456.4^{c}	20.5^{c}	95.5^{ns}	98.0^{b}
	T1	799.2^{b}	55.9^{b}	92.9^{ns}	119.7^{ab}
	T2	955.8^{a}	75.5^{a}	92.1^{ns}	137.8^{a}
	T3	677.7^{b}	44.7^{b}	93.4^{ns}	102.2^{b}
	T4	785.3^{b}	56.5^{b}	92.8^{ns}	119.3^{ab}
red chicory*	control	90.4^{c}	5.8^{d}	93.6^{ns}	43.0^{ns}
	T1	175.8^{b}	17.1^{b}	90.2^{ns}	44.0^{ns}
	T2	225.8^{a}	25.5^{a}	88.7^{ns}	45.1^{ns}
	T3	155.3^{b}	8.7^{cd}	94.4^{ns}	43.9^{ns}
	T4	185.5^{b}	11.3^{c}	93.9^{ns}	44.3^{ns}
celery	control	565.0^{c}	79.8^{d}	85.9^{ns}	92.8^{ns}
	T1	899.2^{b}	146.4^{b}	83.7^{ns}	98.4^{ns}
	T2	1055.0^{a}	221.4^{a}	79.0^{ns}	108.8^{ns}
	T3	776.7^{b}	116.3^{c}	85.0^{ns}	96.7^{ns}
	T4	958.3^{ab}	176.2^{b}	81.6^{ns}	99.5^{ns}

** frost damaged*
different letters in the same column indicate statistically significant difference at $P \leq 0.05$

Table 6. Dry matter production and total nitrogen content in leaves and roots

plant	treatment	dry matter leaves (kg/ha)	dry matter roots (kg/ha)	N leaves %	N roots %
lettuce	control	1141	5530	4.26	2.60
	T1	3108	4084	4.48	7.14
	T2	4195	3664	5.77	9.95
	T3	2485	4399	3.56	3.04
	T4	3141	3727	4.76	4.83
red chicory*	control	323	1248	0.56	4.89
	T1	950	1058	1.12	7.66
	T2	1416	916	1.40	7.88
	T3	481	1032	0.84	6.10
	T4	628	974	1.12	6.73
celery	control	4435	4804	1.46	3.46
	T1	8133	4042	2.20	5.29
	T2	12302	3701	2.56	5.81
	T3	6459	4425	2.00	4.04
	T4	9790	4479	2.40	4.50

**frost damaged*

Table 6 shows the distribution of dry matter between roots and aerial parts and their N content in the various treatments. Data resulting from the single plots were elaborated and referred to one hectare in order to convey a clearer representation of results. It is of interest to remark that while the aerial part was constantly highest with the T2 treatment and lowest in the control, the opposite was true with roots; more in general the development of the aerial part resulted directly correlated and that of the roots inversely correlated to N availability. This matches well the long-denounced changes in the shoot/root ratio as a response to nutrient availability (e.g. Passioura, 1983; Shangguan et al., 2004).

Table 7. Nitrogen balance (kg ha^{-1})

parameter	plant	treatment				
		control	T1	T2	T3	T4
total applied N		---	**720**	**1080**	**720**	**1080**
N in leaves	lettuce	49	139	242	88	150
	red chicory	2	11	20	4	7
	celery	65	179	315	129	235
N in roots	lettuce	144	292	365	134	180
	red chicory	61	81	72	63	66
	celery	166	214	215	179	202
total absorbed N in leaves + roots		**487**	**916**	**1229**	**597**	**840**
N surplus at the end of the 3 cycles		**---**	**---**	**---**	**123**	**240**

In table 7 a nitrogen balance is elaborated starting from the total application in the three cycles and subtracting the quantities absorbed in leaves and roots: only in the "organic" treatments T3 and T4 plants resulted unable to uptake the entire N quantities applied to the soil. Presumably the lack of synchronization between N availability and plant requirements in such treatments determined such an appreciable unused fraction as confirmed by the considerable decrease in N use efficiency and uptake efficiency (table 8), and the higher final content in mineral N of drained water in those treatments. As Boldrini et al. put it *"The determination of nutrient surplus (N supply minus N off-take) at a field scale is often used as an indicator of the potential loss of N … and can give an indication of the risks that are associated with specific farming practices"* (Boldrini et al., 2007).

Table 8. Comparing two different "efficiencies"

plant	treatment	N supply (kg ha^{-1})	yield dry weight (kg ha^{-1})	total N in plants including roots (kg ha^{-1})	N uptake efficiency	N use efficiency
lettuce	control	---	1141	193	--	---
	T1	240	3108	430	1.79	12.95
	T2	360	4195	606	1.80	11.65
	T3	240	2485	222	.92	10.35
	T4	360	3141	329	.91	8.72
chicory	control	---	323	63	---	---
	T1	240	950	92	.38	3.96
	T2	360	1416	92	.25	3.93
	T3	240	481	67	.28	2.00
	T4	360	628	73	.20	1.74
celery	control	---	4435	231	---	---
	T1	240	8133	393	1.64	33.89
	T2	360	12302	530	1.47	34.17
	T3	240	6459	308	1.28	15.50
	T4	360	9790	436	1.21	27.19

Table 8 shows the values of two contrasting "efficiency" indicators as defined in the review of Dawson et al.(2008); we are aware that objections can be raised to the term "efficiency", but prefer to maintain it for the sake of uniformity and ease of comparison.

The indicators, elaborated broadly following their approach, with minor modifications mainly due to the will of considering also N content in roots, are defined as follows:

- N uptake efficiency = Nt (total N in plant) / Ns (N supply)
- N use efficiency = Yw (yield dry weight) / Ns (N supply)

By definition both indicators are a measure of plant response to available N, one in terms of ability in fertilizer absorption and the other in terms of fertilizer elaboration.

The higher N efficiency obtained with mineral rather than organic N form is in agreement with a number of former findings (e.g. Myers et al., 1997; Pang and Letey, 2000; Jenkinson, 2001; Kirchmann and Bergström, 2001).

Proteic efficiency, defined as the ratio of organic to total N in plants, was also determined but results were far from significant and as a consequence are not reported.

Enzymatic activity

Table 9- Evolution of deaminase activity (μg N-NH4+ g-1dm h $^{-1}$)

treatment	week of the year ← year 1							→	← year 2 →		
	26	30	34	43	44	46	49	51	30	35	40
control	2.5	7.2	7.5	1.1	2.4	1.2	2.5	1.2	3.2	2.0	1.7
T1	1.0	1.3	5.7	0.9	0.7	0.9	2.1	1.1	2.1	2.0	2.3
T2	0.2	0.7	5.1	0.8	0.3	1.3	1.6	0.6	1.6	1.7	2.1
T3	4.6	10.4	12.0	1.8	4.0	2.3	2.2	1.4	3.7	2.2	2.3
T4	5.9	13.1	16.3	2.1	9.6	8.6	3.3	2.7	4.0	2.4	2.7

Table 9- Evolution of protease activity (μg tyrosine g^{-1}dm 2h $^{-1}$)

treatment	week of the year ← year 1							→	← year 2 →		
	26	30	34	43	44	46	49	51	30	35	40
Control	52.4	82.0	89.6	55.5	80.0	61.6	82.2	104.0	104.0	82.4	77.0
T1	29.1	44.8	77.3	53.9	47.4	90.7	99.5	108.4	91.9	59.8	96.8
T2	8.5	26.7	67.4	39.2	37.7	42.2	57.4	82.6	82.6	51.2	88.4
T3	68.3	106.9	107.6	118.9	246.6	176.8	117.1	153.6	164.2	108.6	118.4
T4	90.2	137.5	141.5	124.2	363.4	245.7	177.3	211.0	221.0	117.4	146.6

The response of deaminase and protease activity was consistent, with mineral fertilizers bringing about an evident reduction in activities whereas organic fertilizers stimulated them. The level of the means in enzymatic activity in fact in both cases was ranked as follows: T2 < T1 < control < T3 < T4, and the Multiple Range Test confirmed the statistical significance of the differences. Apparently the application of ammonium sulphate shifted the balance of available N in the soil by promptly increasing N availability in the circulating solution and consequently reducing the need for enzymatic action aiming at making organic nitrogen available to plants. Opposite to it the organic fertilizer rich in proteins with low molecular weight supplied a substrate apt to stimulate the enzymatic activities.

It is interesting to remark the enhancement in enzymatic activities in the final sampling for all the fertilized treatments (as opposed to the control) particularly in the case of protease, probably as a response to the leaching of mineral N consequent to heavy rains occurred between the two final samplings and its reduced availability.

Tables 8 and 9 report in detail the evolution of the enzymatic activities in the various treatments.

Microbial population

No difference could be appreciated in bacterial population as a consequence of the different treatments.

The photos below, taken with a Polaroid Gel Cam, evidence in fact that all the bands resulting from the DGGE analysis are similar and have similar intensity in the four sampling periods, encompassing the whole duration of the experiment.

Figure 5 – DGGE analysis of the eubacterial population

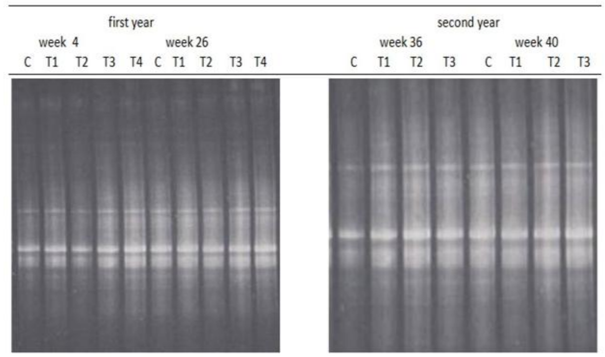
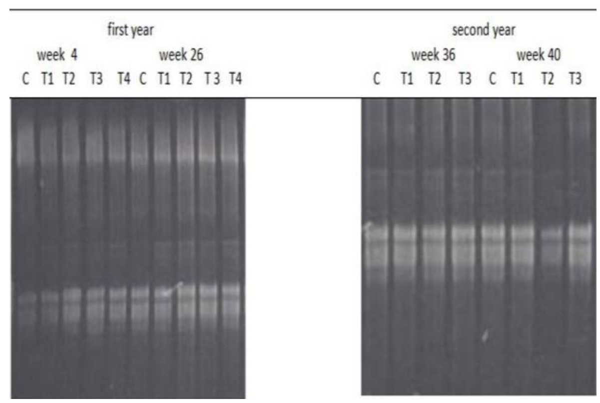


Figure 6 – DGGE analysis of the ammonia-oxidizing population



Such result can depend on the relatively short duration of the experiment, but demonstrates anyway that variations found in enzyme activity depended on factors linked to the soil environment, particularly N availability, rather than on the varying intensity of microorganism population, which was unaffected by the different types and amounts of fertilizers.

Discussion and Conclusion

The results show that same amounts of N when applied under mineral or organic form can determine different responses in the soil-water-plant system. Plants yielded a higher biomass production, richer in proteins and N content when treated with mineral N; soil showed a more intense enzymatic activity and higher residual N but not more bacterial population when treated with organic N; drained water had a more elevated final mineral N concentration in organically-treated plots.

Altogether better results were achieved through the application of N in its mineral form, although the differences were not always statistically significant. N uptake efficiency and N use efficiency resulted in all cases lower with organic fertilizer probably, it can be speculated, because of the often mentioned lower N availability in critical crop phenophases.

Such results confirm that higher skill and accuracy are required for the appropriate management of organic fertilizers to reduce their potential disadvantages in comparison to the mineral forms.

The results give support to the view of Manlay et al. (2007), who state "*One of the key features of sustainable soil practice, is the return to managing soil fertility through the combination of organic matter…and mineral nutrient inputs.*"

The N variations found in the unfertilized control plots can be explained as a response to the nutrient uptaking by the crops, the leaching action of the rains and the intensity of biocenosis activities varying in time with plant development, soil redox potential, moisture and temperature.

Similarly, the variations in time of the enzymatic activities in control plots can be seen as the result of the combined action of the same factors.

It is not easy to understand whether the reduction in N uptake efficiency differences between the mineral and the organic treatments, passing from lettuce to chicory and celery, depends only on the different plant ability to absorb N in its various forms or also on its different availability in time, perhaps as a consequence of increased N availability from decaying organic matter in treatments T3 and T4.

The following conclusions can be drawn at the end of the three crop cycles:

- mineral fertilizer released NH_4^+ and NO_3^- more promptly than organic fertilizer, and higher peaks of such ions were reached in the soil, but the differences between the means throughout the three cycles were not statistically significant; fluctuations were lower with organic fertilizers;
- fresh and dry matter, nitrogen and protein content in the plants was consistently higher in those plots fertilized with mineral N, which evidences the importance of the fertilizer availability in due time;
- the slowly released N from the organic fertilizer, being utilized by crops to a lesser extent, raised the risk of pollution at the end of the three crop cycles giving origin to a surplus in the soil and consequently to a higher N concentration in drained water;
- enzymatic activity was strongly influenced by the form of applied N: the presence of ammonium sulphate reduced the need for it while the organic fertilizer stimulated it;
- microbial population in the soil resulted unaffected by the source and amounts of fertilizers demonstrating a considerable resilience to the action of relatively high amounts of mineral and organic fertilizers;
- the results suggest that the best solution for nitrogen fertilization is probably the combined use of basic organic fertilizers with the addition of modest amounts of mineral N in the periods of crop peak demand.

Acknowledgement

The authors gratefully acknowledge the support of the Sicilian Region (Assessorato Agricoltura e Foreste) who contributed to finance the research.

References

Albiach, R., Canet, R., Pomares, F., Ingelmo, F.(2000): Microbial biomass content and enzymatic activities after the application of organic amendments to a horticultural soil - Bioresource Technology 75 43–48

Alef K., Kleiner D. (1986): Arginine ammonification, a simple method to estimate microbial activity potentials in soils- Soil Biol. Biochem. 18: 233 – 235

Alloway, B.J. and Jackson, A.P.(1991): The behavior of heavy-metals in sewage sludge-amended soils - Sci. Total Environ., 100: 151–176

Bates, L.S., Waldren, R.P., Teare, I.D. (1973): Rapid determination of free proline for water stress studies. *Plant and Soil* 39: 205-207

Bell, J.M., Robinson, C.A., Schwartz, R.C. (2006): Changes in soil properties and enzymatic activities following manure applications to a rangeland - Rangeland Ecology and Management,. 59, (3): 314-320.

Benckiser G. and Simarmata T. (1994): Environmental-impact of fertilizing soils by using sewage and animal wastes - Fert. Res. 37: 1–22

Benincasa P., Boldrini A., Tei F., Guiducci M. (2004): N release from several green manure crops.

Proceedings of the VIII ESA Congress - Addendum, Copenhagen, Denmark, 11-15 July 2004, 971-972

Bergström, L., Bowman, B.T., Sims, J.T.(2005): Definition of sustainable and unsustainable issues in nutrient management of modern agriculture- Soil Use and Management 21: 76-81

Berry, P.M., Sylvester-Bradley, R., Philipps, L., Hatch, D.J., Cuttle, S.P., Rayns, F.W., Gosling, P. (2002): Is the productivity of organic farms restricted by the supply of available nitrogen? - Soil Use and Management, 18: 248-255

Berthelot M. (1959): Violet d'alanine. Report Chim. Appl. 58: 284

Boldrini, P. Benincasa, G. Tosti, F. Tei, M. Guiducci (2007): Apparent N Balance in Organic and Conventional Low Input Cropping Systems - 3rd QLIF Congress, Hohenheim, Germany, March 20-23, 2007 Archived at http://orgprints.org/view/projects/int_conf_qlif2007.html

Bonde, T.A. and Rosswall, T. (1987): Seasonal variation of potentially mineralizable nitrogen in four cropping systems – SSSA Journal 51: 1508-1514

Bradford, M.M. (1976): A rapid sensitive method for quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Analytical Biochemistry 72*: 248-253

Cataldo D.A., Haaron M., Schrader L.E., Youngs V.L. (1975). Rapid colorimetric determination of nitrate in plant tissue by nitration of salicylic acid. *Commun Soil Plant Anal* 6: 71-80

Crecchio C., Curci, M., Mininni R., Ricciuti P., Ruggiero P. (2001): Short-term effects of municipal solid waste compost amendments on soil carbon and nitrogen content, some enzyme activities and genetic diversity .Biology and Fertility of Soils. 34: 311-318

David, C;. Jeuffroy, M.H; Valantin-Morison, M.; Herbain, C. (2007): Improvement of the soil-crop model AZODYN under conventional, low-input and organic conditions-3rd QLIF Congress, Hohenheim, Germany, March 20-23, 2007 Archived at http://orgprints.org/view/projects/int_conf_qlif2007.html

Dawson, J.C.; Huggins, D.R.; Jones, S.S. (2008): Characterizing nitrogen use efficiency in natural and agricultural ecosystems to improve the performance of cereal crops in low-input and organic agricultural systems - Field Crops Research 107: 89–101

Debosz, K., Petersen, S.O., Kure, L.K., Ambus, P.(2002): Evaluating effects of sewage sludge and household compost on soil physical, chemical and microbiological properties - Applied Soil Ecology 19: 237–248

Evanylo, G., Sherony, C., Spargo, J., Starner,D., Brosius,M., Haering,K. (2008): Soil and water environmental effects of fertilizer-, manure-, and compost-based fertility practices in an organic vegetable cropping system - Agriculture, Ecosystems and Environment,127: 50–55

Haas G., Berg M., Köpke U. (2002): Nitrate leaching: comparing conventional, integrated and organic agricultural production systems. In: Steenvorden, J., Claessen F., Willems J. (eds.): Agricultural effects on ground and surface waters, IAHS Publ. no. 273:131-136.

Heuer H., Krsek M., Baker P., Smalla K., Wellington E.M.H. (1997): Analysis of actinomycete communities by specific amplification of genes encoding 16S rRNA and gel-electrophoretics separation in denaturing gradients. Appl. Environ. Microbiol. 63: 3233-3241

Jenkinson D. S. (2001): The impact of humans on the nitrogen cycle, with focus on temperate arable agriculture. Plant and Soil 228: 3-15.

Karmeli, D. and Keller, J. (1974): *Trickle Irrigation Design,* Rain Bird Sprinkler Manufacturing Corp., Glendora, USA

Kirchmann H. and Bergström L. (2001): Do organic farming practices reduce nitrate leaching? Soil Science and Plant Analysis 32-7/8: 997-1028.

Kirchmann, H. and Thorvaldson, G. (2000): Challenging targets for future agriculture – European Journal of Agronomy 12: 145-161

Ladd J. N. and Butler J.H. A. (1972): Short – term assays of soil proteolytic enzyme activities using proteins and dipeptide derivatives as substrates. Soil Biology and Biochemistry 4: 19-30

Manlay,R.J., Feller, C., Swift, M.J. (2007): Historical evolution of soil organic matter concepts and their relationships with the fertility and sustainability of cropping systems - Agriculture, Ecosystem and Environment 119: 217-233

Mulvaney, R.L., Khan, S.A., Hoeft, R.G., Brown, H.M. (2001): A soil organic nitrogen fraction that reduces the need for nitrogen fertilization. Soil Sci. Soc. Am. J. 65:1164–1172

Myers, R.J.K., van Noordwijk, M.; Vityakon, P.(1997): Synchrony of nutrient release and plant demand: plant litter quality, soil environment and farmer management options – *Driven by Nature – Plant Litter Quality and Decomposition.* CAB international, Wallingford, UK

Pang, X.P. and Letey, J. (2000): Challenge of Timing Nitrogen Availability to Crop Nitrogen Requirements - Soil Science Society of America Journal 64:247-253

Passioura, J.B. (1983): Root and drought resistance - Agric. Water Managent **7:** 265-280

Perucci, P., Dumontet, S., Bufo, S.A., Mazzatura, A., Casucci, C. (2000): Effects of organic amendment and herbicide treatment on soil microbial biomass - Biology and Fertility of Soils 32: 17–23

Schnier, H.F., S.K. De Datta, Mengel, K. (1987): Dynamics of ^{15}N-labeled ammonium sulfate in various inorganic and organic soil fractions of wetland rice soils. Biol. Fertil. Soils 4:171–177 Shangguan,Z.P., M.A. Shao, M.A., Ren, S.J.,L.M. Zhang, L.M., Xue,Q. (2004): Effect of nitrogen on root and shoot relations and gas exchange in winter wheat - Bot. Bull. Acad. Sin. **45:** 49-54

Shannon, D., Sen, A.M. Johnson, D.B.(2002): A comparative study of the microbiology of soils

managed under organic and conventional regimes. Soil Use and Management 18: 274-283

Shepherd, M.A. (2002): Understanding soil fertility in organically farmed soils. Final project report OF0164. Defra Research Policy and International Division, Final Reports Unit & ADAS, Gleadthorpe Research Centre, Meden Vale, Mansfield, Notts NG20 9PF

S.I.S.S., Società Italiana della Scienza del Suolo (1985): *Metodi normalizzati di analisi del suolo*. Bologna, Edagricole

Stevenson, F.J. (1994): *Humus chemistry: Genesis, composition, reactions* - John Wiley & Sons New York

Weisburg W.G., Barns S.M., Pelletier D.A., Lane D.J.(1991): 16S ribosomal DNA amplification for phylogenetic study. J.Bacteriol. 173: 697-703

Yadvinder-Singh; Bijay-Singh; Khind, C.S.(1992): Nutrient Transformation in Soils Amended with Green Manures- *Advances in Soil Sciences,* Springer-Verlag, New York