



## Nitrogen uptake by crops, soil distribution and recovery of urea-N in a sorghum–wheat rotation in different soils under Mediterranean conditions

P. Nannipieri<sup>1,\*</sup>, L. Falchini<sup>1</sup>, L. Landi<sup>1</sup>, A. Benedetti<sup>2</sup>, S. Canali<sup>2</sup>, F. Tittarelli<sup>2</sup>, D. Ferri<sup>3</sup>, G. Convertini<sup>3</sup>, L. Badalucco<sup>4</sup>, S. Grego<sup>5</sup>, L. Vittori-Antisari<sup>6</sup>, M. Raglione<sup>7</sup> and D. Barraclough<sup>8</sup>  
<sup>1</sup>Dipartimento di Scienza del Suolo e Nutrizione della Pianta, Università di Firenze, P. le delle Cascine 28, I-50144 Firenze, Italy; <sup>2</sup>Istituto Sperimentale per la Nutrizione delle Piante, MIPA, Via della Navicella 2, I-00184 Roma; <sup>3</sup>Istituto Sperimentale Agronomico, Sezione di Bari, MIPA, Via C. Ulpiani 5, I-70125 Bari; <sup>4</sup>Dipartimento di Ingegneria e Tecnologie Agro-forestali, Viale delle Scienze, 13; I-90128 Palermo; <sup>5</sup>Dipartimento di Agrobiologia ed Agrochimica, Facoltà della Tuscia, Via S. C. DeLellis, I-0110 Viterbo; <sup>6</sup>Istituto di Chimica Agraria, Viale Berti Pichat 10, I-40127 Bologna; <sup>7</sup>Istituto Sperimentale per lo Studio e la Difesa del Suolo, Sezione di Rieti, MIPA, Via Casette 1, I-02100 Rieti; <sup>8</sup>Department of Soil Science, The University of Reading, Whiteknights, PO Box 233, Reading RG6 6DW, UK

Received 1 July 1998. Accepted in revised form 24 December 1998

**Key words:** bare soil, microbial biomass N, N balance, enriched urea, non-exchangeable  $\text{NH}_4^+$ , sorghum-wheat rotation

### Abstract

The N uptake by crops, soil distribution and recovery of  $^{15}\text{N}$  labelled urea-N ( $100 \text{ kg N ha}^{-1}$ ) were investigated in a sorghum-wheat rotation in two silty clay soils (Foggia and Rieti Casabianca) and one silt loam soil (Rieti Piedifiume) under different mediterranean conditions. Non-exchangeable labelled  $\text{NH}_4\text{-N}$  represented an important pool at both Rieti sites with higher values ( $p < 0.05$ ) under sorghum (14.0 and 24.6% of the urea N in the 0–20 cm layer at the end of the cropping season) than wheat whereas it was much less important in the Foggia soil (10.0% in the surface soil under sorghum). This is probably related to the clay minerals composition of the three soils; because vermiculite was present in both Rieti sites but not in the Foggia soil. At harvest from 4.4 to 5.3% of the urea N initially applied was present as microbial biomass N in the surface soil layer with no generally significant differences due to location and type of crops. Both sorghum and wheat N yields were higher in the driest site (Foggia) probably due to better light conditions, higher temperatures and irrigation during summer of the sorghum cropping period. The recovery of plant fertilizer N (about 21% for sorghum and 27% for wheat) and the percentage of N in the plant derived from the fertilizer (NDFF) were the lowest at Rieti-Casabianca probably as the result of the protection of immobilized fertilizer N against microbial mineralization by the swelling clays. The fertilizer N unaccounted for was nil or very low (10.8% at Rieti-Casabianca under wheat and 11.8 and 4.9% at Rieti-Piedifiume under sorghum and wheat, respectively). Urea-N losses occurred when Rieti Piedifiume and Rieti Casabianca soils were kept bare. In this case the urea N unaccounted for ranged from 12 to 56% of the urea N with higher losses in Rieti-Piedifiume than in Rieti-Casabianca. The higher recoveries in the latter soil were probably confirmed by the stabilizing effect of clays on the immobilized urea N.

### Introduction

The isotopic labelling method provides the most accurate measure of the relative contributions of soil N

and fertilizer N to plant uptake (Stevenson, 1986). Estimates of fertilizer N taken up by plants range from 30–70% of the fertilizer N applied to soil whereas a significant percentage (20–50%) is immobilized as organic N at the end of the first growing season (Hauck and Bremner, 1976; Legg and Meisinger,

\* Fax No: 39-55-333273. E-mail: nannip@iges.fi.cnr.it

1982; Stevenson, 1986). The N unaccounted for usually ranges from 20 to 40% and depends on N losses due to denitrification,  $\text{NH}_3$  volatilization and nitrate leaching as well as to accumulated errors of sampling and analysis. Sample variability is probably the greatest single source of methodological error in N-tracer studies (Hauck et al., 1994).

Most of the studies on the fate of  $^{15}\text{N}$  labelled fertilizers have been carried out in North America, Central and North Europe and in Australia. The relatively high cost of  $^{15}\text{N}$  enriched compounds and the sophisticated and expensive instrumentation for accurately determining the N-isotope ratio has limited the use of this decisive approach (Mulvaney, 1993). Results of fertilizer N uptake by crop and the distribution of fertilizer N in soil are markedly affected by environmental conditions and experimental design. Thus, it is wrong to extrapolate results to areas with different soils and environmental conditions. Only a few studies have been carried out in South Europe where Mediterranean climatic conditions generally prevail. In a grass-legume association 25% of the N-labelled urea was removed as forage whereas 59% of the initially applied N was accounted for in the 0 to 40-cm topsoil layer at the end of the cropping season (Nannipieri et al., 1985). Short-term samplings showed that ureolysis occurred within two days after application, with the highest  $\text{NH}_4^+$ -N concentration observed after 2 days; the rates of nitrification and labelling of microbial biomass N reached maximum values during the decrease in the concentration of labelled ammonium (Nannipieri et al., 1990). From 20 to 52% of the N applied as either  $(\text{NH}_4)_2\text{SO}_4$  or  $\text{NH}_4\text{NO}_3$  to wheat, on two calcareous soils near Tunis, was unaccounted for at the end of the growing season;  $\text{NH}_3$  volatilization was considered the main fertilizer N loss (Sanaa et al., 1992).

The winter sorghum-wheat rotation is quite common in Italian agriculture with a different growing season for the two crops: from late autumn to early summer for winter wheat; from April to September for sorghum. Cold temperature limits growth and development of sorghum (Duncan, 1996). No information is available on the fate of urea N, the most used fertilizer, in the sorghum-wheat rotation under Mediterranean conditions.

The aims of this study were to investigate how the fate of urea N was affected in a sorghum-wheat rotation by (1) climatic conditions, (2) texture and (3) type of clay minerals under Mediterranean conditions of Italy. In two of the three investigated soils the effect

of plant cover was also studied by considering the disposition of labelled N in N pools of the cropped and the corresponding bare soil.

## Materials and methods

### *Sites and soils*

The experimental sites were the Experimental Station of the 'Istituto Sperimentale Agronomico' at Foggia (South Italy) and the Experimental Station of the 'Istituto per lo Studio e la Difesa del Suolo' at Rieti (Central Italy). At the latter site two different soils (Rieti-Piedifume and Rieti-Casabianca) were selected. The main physico-chemical properties of soils are reported in Table 1. According to USDA soil classification, Foggia was a Typic Chromoxererts, Rieti-Piedifume a Fluventic Eutrochrepts and Rieti-Casabianca a Vertic Eutrochrepts. Illite (dominant mineral) and kaolinite were present in Foggia, which was a silty clay soil; smectite/vermiculite (dominant mineral), illite, kaolinite and smectite in Rieti-Casabianca soil, which was a silty clay soil; and smectite/vermiculite (dominant mineral), illite (dominant mineral), kaolinite and vermiculite in Rieti-Piedifume soil, which was a silt loam soil.

### *Experimental protocols*

In 1993  $^{15}\text{N}$  enriched urea (10% abundance) was applied to  $2 \times 2$  m plots cropped to sorghum (*Sorghum halepense*) as a powder at a rate of  $100 \text{ kg N ha}^{-1}$ . Fertilizer was applied in two equal doses before sowing (early April) and at the emergence of the 6–7th leaf (mid May). Sowing was carried out in May and harvests in mid September. In 1994  $2.0 \times 2.0$  m plots, cropped the previous year with sorghum and fertilized with unlabelled urea ( $100 \text{ kg N ha}^{-1}$ ), were sown in late autumn with winter wheat (*Triticum aestivum*) at both Rieti sites and with durum wheat (*Triticum durum*) at Foggia and fertilized with  $^{15}\text{N}$  enriched urea (10% abundance), applied in a single dose at a rate of  $100 \text{ kg N ha}^{-1}$  as an aqueous solution to allow urea penetration into soil.

In 1993 and 1994,  $2.0 \times 2.0$  m plots with bare soils were also established in both Rieti sites and annually fertilized with  $^{15}\text{N}$  enriched urea (10% abundance) at a rate of  $100 \text{ Kg N ha}^{-1}$ . The rare young shoots were hand-picked, dried at  $60^\circ\text{C}$  and stored at  $-40^\circ\text{C}$  prior to be analyzed for their N content and  $^{15}\text{N}$  enrichment.

Table 1. Soil properties

Property	Soils		
	Rieti-Casabianca	Rieti-Piedifiume	Foggia
Sand (%)	8.4	29.1	14.0
Silt (%)	41.6	55.3	40.4
Clay (%)	50.0	15.6	45.6
pH (H <sub>2</sub> O)	7.7	8.1	8.3
Organic C (%)	1.72	1.42	1.79
Total N (%)	0.21	0.15	0.16
CaCO <sub>3</sub> (%)	12.3	3.9	5.4

A randomized block experimental design with three replicates was used.

### Sampling

As suggested by Powlson and Barraclough (1993), the sampling area was limited to 1.0 × 1.0 m to avoid edge effects. Soil was sampled at two depths (0–20 and 20–40 cm), sieved (< 2 mm) with removal of visible plant remains by hand-picking and stored at –20 °C until analyses were run.

At harvest the total fresh material was dried at 60 °C for 48 h and then separated into straw (or stalks) and grain.

### Chemical, microbial and isotope analyses

Clay types were determined by X-ray diffraction.

Total N was determined by Kjeldahl method (Bremner and Mulvaney, 1982). Exchangeable NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N were determined on a KCl extract of moist soil (100 mL 1 M KCl: 20 g of oven-dry soil), after shaking for 1 h at room temperature followed by filtration through Whatman GF/A glass fiber filters under vacuum. The soil extract was divided in two parts: one analyzed for the <sup>15</sup>N enrichment and the other for NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N contents by a Flow Injection Analyzer (FIAS 300-Perkin Elmer) linked to a spectrophotometer Lambda 2 (Perkin Elmer) as reported by (Muller et al., 1992).

Microbial biomass N was determined by the chloroform fumigation-extraction method (Brookes et al., 1989) with slight modifications. After the KCl extraction of inorganic N (see above), the soil on the filter was washed twice with 1 M KCl (100 mL) to remove traces of labelled and unlabelled inorganic N, transferred to a glass beaker and placed into a desiccator containing CHCl<sub>3</sub>. After 24 h, CHCl<sub>3</sub> was removed and the soil extracted with 100 mL 0.5 M

K<sub>2</sub>SO<sub>4</sub> for 1 h at room temperature under shaking. The filtrate (50 mL) was digested with 10 mL 96% H<sub>2</sub>SO<sub>4</sub> and 1 mL 0.29 M CuSO<sub>4</sub> for 3 h at 360 °C with pre-heating at 120 °C for 2 h and predigestion at 180 °C for 1 h. Total N in the digest was determined by steam distillation and titration (Brookes et al., 1989). A blank with 0.5 M K<sub>2</sub>SO<sub>4</sub> was also analyzed.

Non-exchangeable NH<sub>4</sub><sup>+</sup> was determined by the method of Silva and Bremner (1966) modified in the following way: 2 g of soil were introduced in 150 mL Corex tubes and treated with 20 mL NaOBr (prepared by mixing 250 mL 2 M NaOH with 7.5 mL pure Br<sub>2</sub>); the soil mixture was incubated for 2 h at room temperature (Marzadori et al., 1994). Then, 40 mL of distilled H<sub>2</sub>O were added and soil organic matter completely oxidized in a microwaves oven (Mod. MDS-81D, CEM Corporation, Indian Trail) for 5 min at 90% and 2 min at 80% of the maximum oven energy (600 watt). After cooling, 20 mL of distilled H<sub>2</sub>O were added; the soil suspension was incubated at room temperature overnight and then centrifuged at 5000 g for 10 min. The soil-sediment was washed twice with 50 mL 0.5 M KCl, dried at 40 °C, pounded with a pestle in a mortar, introduced (1 g) into a 250 mL plastic bottle and treated with 20 mL of a mixture 1 M HCl–5 M HF. The soil suspension was shaken overnight before distillation and titration to determine NH<sub>4</sub><sup>+</sup>-N content (Marzadori et al., 1994).

The total N content of plant material was determined by an Automated Nitrogen Carbon Analyzer (ANCA) (Roboprep, Europa Scientific, Crew, UK).

For <sup>15</sup>N analysis, samples after titration (see determination of non-exchangeable NH<sub>4</sub><sup>+</sup>, total N and biomass N) or KCl soil extracts (see determination of exchangeable NH<sub>4</sub><sup>+</sup> and NO<sub>3</sub><sup>-</sup>-N) were introduced into 250 mL Erlenmeyer flasks and treated with 0.5 g MgO (treated at 700 °C for 2 h). Evolved NH<sub>3</sub> was adsorbed on a 5 mm glass fiber filter disc (Whatman,

GF/D) acidified with 10  $\mu\text{L}$  2.5 M  $\text{KHSO}_4$ , placed on a stainless steel (AISI 316, Goodfellow, UK) hook fixed to a rubber stopper covering the flask (Brookes et al., 1989). After 5 days at room temperature, the filter disc was removed, the flask left uncovered for 1 day and eventually 0.2 g of Devarda's alloy were added to reduce  $\text{NO}_3^-$  to  $\text{NH}_4^+$  in extracts containing  $\text{NO}_3^-$ . A new acidified disc was used to trap evolved  $\text{NH}_3$  as reported above. Then, the filters were dried in a desiccator containing  $\text{CaSO}_4$ , placed into tin cups and eventually their  $^{15}\text{N}$  enrichment analysed by the mass spectrometer (ANCA-MS system; Europa Scientific, Crew, UK).

#### *Calculations and statistical analysis*

The amount of fertilizer N, the percentage recovery of fertilizer N and the percent of N derived from the fertilizer (NDFP) were calculated as reported by Hauck and Bremner (1976).

Means and standard deviations values were calculated.

Experimental data recorded in three localities were jointly processed using analysis of variance by 'General Linear Models' procedure (GLM) and considering as sources of variation: (a) experimental sites; (b) years (or crops); (c) replications; (d) subsampling (SAS/STAT, 1988). The effect of location on yields and distribution of urea-N among N pools was also analyzed by the Pearson chi-square static and Fisher's exact test (SAS/STAT, 1988). Comparison among mean values was improved by the 'Student-Newman-Keuls' (SNK) multiple range test considering significance probabilities as determined by GLM procedure (SAS7STAT, 1988).

#### *Growing season conditions*

Monthly precipitations followed the typical behaviour of a Mediterranean climate with a summer minimum and autumn-spring maximum (Figure 1). Annual rainfall at Foggia was 455 mm in 1993 and 336 mm in 1994, at Rieti 902 mm in 1993 and 825 mm in 1994. Foggia was the driest site and it was irrigated ( $2600 \text{ m}^3 \text{ ha}^{-1}$ ) during summer to mitigate the negative effects of evapotranspiration on sorghum yield.

## **Results and discussion**

### *Presence of the plant cover*

*Yield and fertilizer N uptake.* The sorghum N yield was higher ( $p < 0.05$ ) at Foggia than at Rieti-Piedifume and Rieti-Casabianca (Tables 2, 3 and 4) probably due to the irrigation during summer. However, wheat N yield was also significantly ( $p < 0.05$ ) higher at Foggia than at both Rieti sites according to differences observed in the dry matter yields in the three sites ( $4.5 \text{ t ha}^{-1}$  for grain and  $7.0 \text{ ha}^{-1}$  for straw at Foggia;  $4.0 \text{ t ha}^{-1}$  for grain and  $3.4 \text{ ha}^{-1}$  for straw at both Rieti-sites). It is possible that higher temperatures, low precipitations (Figure 1) and better light conditions may be responsible for the higher wheat N yield at Foggia than Rieti sites. The higher fertilizer N recovery of sorghum stalks than grain at Foggia cannot be due to negative effects of water stress on N translocation in the xylem from roots to shoots because the crop was irrigated on summer. It may be hypothesized a N retranslocation in the phloem from shoots to roots to meet root demand of reduced nitrogen for growth (Marschner, 1995) but further research is needed to verify the validity of this hypothesis. The amount of straw N was higher at Foggia than in both Rieti sites probably because durum wheat showed an higher culm height than winter wheat.

Fertilizer N recoveries of sorghum grain and stalk could be ranked according to the following trend ( $p < 0.05$ ): Rieti-Piedifume=Foggia>Rieti-Casabianca and Foggia>Rieti-Piedifume>Rieti-Casabianca, respectively (Tables 2, 3 and 4). On the other hand, no significant differences were observed among the three sites in fertilizer N recoveries of wheat grain and straw (Table 2, 3 and 4). The overall wheat recoveries ranged from about 24 to 27% of the urea N initially applied and these data were comparable to values reported by Christensen and Meints (1982) and Sanaa et al. (1992) but lower than reported by Powlson et al. (1992) and Hamid and Ahmad (1994, 1995). Fertilizer N uptake was higher ( $p < 0.05$ ) in sorghum than wheat at Foggia and Rieti-Piedifume (Tables 2 and 3) but not at Rieti-Casabianca (Table 4).

The percentage of N in the plant derived from the fertilizer (NDFP) represents a yield-independent criterion since it can be calculated directly from the isotopic dilution. At Foggia about 15% of the total N in wheat straw and grain was derived from fertilizer N (Table 2). These values were in the range found by Sanaa et al. (1992) in Tunisian soils. Lower values

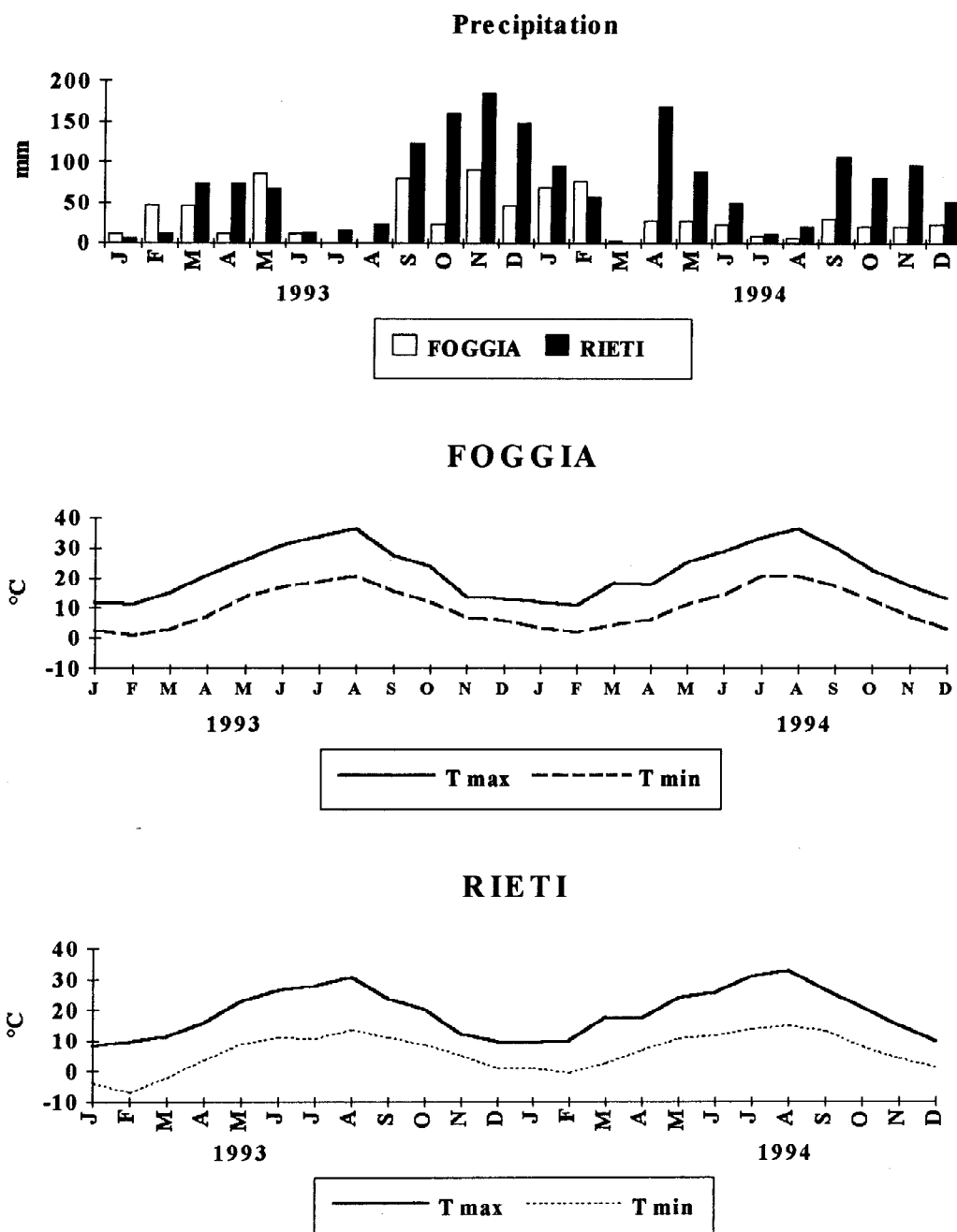


Figure 1. Monthly rainfall, and maximum and minimum temperatures at Foggia and Rieti.

were found for sorghum grains but not for sorghum stalks. The contribution of the labelled source to the wheat N needs was more important at both Rieti sites than at Foggia (Tables 2, 3 and 4). Differences in NDF values between the two Rieti sites were observed for sorghum with a greater contribution of

fertilizer N to the plant N needs at Rieti Piedifume (Tables 3 and 4).

Since the dry matter sorghum yields were higher in Rieti Casabianca (5.0 t ha<sup>-1</sup> for grain and 5.5 t ha<sup>-1</sup> for stalks) than in Rieti Piedifume (4.5 t ha<sup>-1</sup> for grain and 4.1 t ha<sup>-1</sup> for stalks) and equal for

Table 2. Total N and fertilizer N in the different plant and soil pools at Foggia

Pools	Soil depth (cm)	N content ( $\mu\text{g N g}^{-1}$ dry soil)	$^{15}\text{N}$ content ( $\mu\text{g N g}^{-1}$ dry soil)	Recovery of fertilizer N (%)	NDFP (%)
<b>Sorghum</b>					
<i>Soil</i>					
Total N	0–20	1668 $\pm$ 10	6.8 $\pm$ 2.5	25.9 $\pm$ 6.6	
	20–40	1629 $\pm$ 44	5.0 $\pm$ 1.7	12.0 $\pm$ 1.4	
Non-exch. $\text{NH}_4^+$ -N	0–20	84.3 $\pm$ 1.5	0.04 $\pm$ 0.0	0.09 $\pm$ 0.1	
	20–40	82.7 $\pm$ 0.6	0.02 $\pm$ 0.0	0.05 $\pm$ 0.0	
Exch. $\text{NH}_4^+$ -N	0–20	2.5 $\pm$ 0.5	0.02 $\pm$ 0.0	0.04 $\pm$ 0.0	
	20–40	1.6 $\pm$ 0.4	0.01 $\pm$ 0.0	0.01 $\pm$ 0.0	
$\text{NO}_3^-$ -N	0–20	15.3 $\pm$ 1.2	0.8 $\pm$ 0.2	2.0 $\pm$ 0.6	
	20–40	20.6 $\pm$ 1.2	0.7 $\pm$ 0.1	1.8 $\pm$ 0.2	
Biomass-N	0–20	55.8 $\pm$ 1.2	0.3 $\pm$ 0.0	0.8 $\pm$ 0.1	
	20–40	54.1 $\pm$ 6.7	0.3 $\pm$ 0.1	0.8 $\pm$ 0.2	
<i>Plant</i>					
Stalks		<b>57.1<math>\pm</math>3.3<sup>a</sup></b>	<b>9.3<math>\pm</math>0.3<sup>a</sup></b>	46.0 $\pm$ 4.0	16.2 $\pm$ 0.7
Grain		<b>33.5<math>\pm</math>2.6<sup>a</sup></b>	<b>4.1<math>\pm</math>0.3<sup>a</sup></b>	21.0 $\pm$ 1.6	12.3 $\pm$ 0.1
<b>Wheat</b>					
<i>Soil</i>					
Total N	0–20	1650 $\pm$ 10	18.7 $\pm$ 4.2	47.5 $\pm$ 6.2	
	20–40	1605 $\pm$ 20	17.8 $\pm$ 3.9	45.3 $\pm$ 4.1	
Non-exch. $\text{NH}_4^+$ -N	0–20	134.6 $\pm$ 19.5	0.8 $\pm$ 0.4	2.1 $\pm$ 1.1	
	20–40	142.6 $\pm$ 17.3	0.6 $\pm$ 0.1	1.5 $\pm$ 0.2	
Exch. $\text{NH}_4^+$ -N	0–20	1.4 $\pm$ 0.0	0.2 $\pm$ 0.0	0.4 $\pm$ 0.0	
	20–40	1.6 $\pm$ 0.0	0.2 $\pm$ 0.0	0.5 $\pm$ 0.0	
$\text{NO}_3^-$ -N	0–20	11.2 $\pm$ 0.2	4.1 $\pm$ 0.0	10.4 $\pm$ 0.0	
	20–40	10.1 $\pm$ 0.4	4.1 $\pm$ 0.6	10.4 $\pm$ 1.4	
Biomass-N	0–20	80.5 $\pm$ 8.6	2.1 $\pm$ 0.5	5.3 $\pm$ 1.3	
	20–40	75.2 $\pm$ 0.6	1.4 $\pm$ 0.5	3.5 $\pm$ 1.4	
<i>Plant</i>					
Straw		<b>9.7<math>\pm</math>0.3<sup>a</sup></b>	<b>1.5<math>\pm</math>0.3<sup>a</sup></b>	7.5 $\pm$ 1.6	15.3 $\pm$ 4.4
Grain		<b>22.4<math>\pm</math>2.8<sup>a</sup></b>	<b>3.3<math>\pm</math>0.3<sup>a</sup></b>	16.8 $\pm$ 1.4	14.9 $\pm$ 1.5

<sup>a</sup>In bold plant pools units were calculated by considering the yields per g of dry weight soil.

wheat as reported above, the lower fertilizer N recoveries and NDFP values of both sorghum pools at Rieti-Casabianca may be due to the protection of immobilized fertilizer N by swelling clays toward microbial mineralization. Swelling clays are not present in the Foggia clay soil. In Rieti-Piedifiume the protection effect is probably less due to the lower clay content than in Rieti-Casabianca soil (Table 1).

*Soil N pools.* The percentage of fertilizer N as  $\text{NO}_3^-$ -N was higher than the respective percentage as  $\text{NH}_4^+$ -

N in both layers of the investigated soils (Tables 2, 3 and 4). Rapid nitrification of labelled  $\text{NH}_4^+$ -N, low denitrification rates, and low microbial and plant nitrate uptake before harvest can be responsible for the observed behaviour. Under sorghum, percentages of fertilizer N present as  $\text{NO}_3^-$ -N in the 0–40 cm soil layer were higher at Rieti-Casabianca than at Foggia and Rieti-Piedifiume sites. At Foggia the cumulative percentage of urea N present as  $\text{NO}_3^-$ -N was higher under wheat than under sorghum, probably as the result of leaching due to sorghum irrigation.

Table 3. Total N and fertilizer N in the different plant and soil pools at Rieti Piedifiume

Pools	Soil depth (cm)	N content ( $\mu\text{g N g}^{-1}$ dry soil)	$^{15}\text{N}$ content ( $\mu\text{g N g}^{-1}$ dry soil)	Recovery of fertilizer N (%)	NDFP (%)
<b>Sorghum</b>					
<i>Soil</i>					
Total N	0–20	1347 $\pm$ 26	10.8 $\pm$ 0.9	33.6 $\pm$ 2.6	
	20–40	1406 $\pm$ 17	4.9 $\pm$ 1.1	15.1 $\pm$ 3.3	
Non-exch. $\text{NH}_4^+$ -N	0–20	184.3 $\pm$ 8.2	4.5 $\pm$ 1.0	14.0 $\pm$ 3.2	
	20–40	174.2 $\pm$ 1.3	1.3 $\pm$ 0.0	4.0 $\pm$ 0.3	
Exch. $\text{NH}_4^+$ -N	0–20	3.7 $\pm$ 0.1	0.02 $\pm$ 0.0	0.06 $\pm$ 0.0	
	20–40	0.9 $\pm$ 0.1	0.04 $\pm$ 0.0	0.01 $\pm$ 0.0	
$\text{NO}_3^-$ -N	0–20	4.3 $\pm$ 0.6	0.4 $\pm$ 0.1	1.2 $\pm$ 0.3	
	20–40	3.1 $\pm$ 0.3	0.1 $\pm$ 0.0	0.4 $\pm$ 0.0	
Biomass-N	0–20	179.3 $\pm$ 11.9	1.5 $\pm$ 0.1	4.7 $\pm$ 0.3	
	20–40	44.0 $\pm$ 2.0	0.2 $\pm$ 0.0	0.7 $\pm$ 0.0	
<i>Plant</i>					
Stalks		<b>11.9<math>\pm</math>0.4<sup>a</sup></b>	<b>2.6<math>\pm</math>0.4<sup>a</sup></b>	16.3 $\pm$ 2.3	22.2 $\pm$ 3.4
Grain		<b>13.7<math>\pm</math>1.6<sup>a</sup></b>	<b>3.8<math>\pm</math>0.3<sup>a</sup></b>	23.2 $\pm$ 2.0	27.8 $\pm$ 4.8
<b>Wheat</b>					
<i>Soil</i>					
Total N	0–20	1461 $\pm$ 5	19.5 $\pm$ 1.8	60.3 $\pm$ 5.6	
	20–40	1328 $\pm$ 58	2.5 $\pm$ 1.4	8.4 $\pm$ 4.4	
Non-exch. $\text{NH}_4^+$ -N	0–20	176.0 $\pm$ 18.7	1.7 $\pm$ 0.1	5.3 $\pm$ 0.4	
	20–40	175.3 $\pm$ 11.3	0.02 $\pm$ 0.0	0.1 $\pm$ 0.0	
Exch. $\text{NH}_4^+$ -N	0–20	4.5 $\pm$ 0.8	0.08 $\pm$ 0.0	0.3 $\pm$ 0.1	
	20–40	3.2 $\pm$ 0.8	0.01 $\pm$ 0.0	0.03 $\pm$ 0.0	
$\text{NO}_3^-$ -N	0–20	13.9 $\pm$ 2.2	1.5 $\pm$ 0.4	4.7 $\pm$ 1.4	
	20–40	2.1 $\pm$ 0.0	0.1 $\pm$ 0.0	0.3 $\pm$ 0.0	
Biomass-N	0–20	60.1 $\pm$ 8.2	1.5 $\pm$ 0.1	4.7 $\pm$ 0.4	
	20–40	44.9 $\pm$ 9.4	1.4 $\pm$ 0.5	1.1 $\pm$ 0.4	
<i>Plant</i>					
Straw		<b>3.8<math>\pm</math>1.0<sup>a</sup></b>	<b>1.2<math>\pm</math>0.2<sup>a</sup></b>	7.8 $\pm$ 1.5	33.9 $\pm$ 6.8
Grain		<b>10.3<math>\pm</math>1.4<sup>a</sup></b>	<b>3.0<math>\pm</math>0.4<sup>a</sup></b>	18.6 $\pm$ 2.7	30.2 $\pm$ 10.2

<sup>a</sup>In bold plant pools units were calculated by considering the yields per g of dry weight soil.

The concentration of non-exchangeable ammonium was higher ( $p < 0.05$ ) at Rieti-Casabianca than at Rieti-Piedifiume and Foggia (Tables 2, 3 and 4). No significant differences were observed at the end of the cropping season between sorghum and wheat at both Rieti sites whereas significantly higher values were observed at Foggia under wheat than sorghum. Non-exchangeable labelled  $\text{NH}_4^+$ -N represents an important pool at both Rieti sites with higher values ( $p < 0.05$ ) under sorghum than wheat (Tables 3 and 4). Vermiculite (present in both Rieti soils but not

in Foggia soil), degraded illite and, under particular conditions, montmorillonite can trap  $\text{NH}_4^+$  between the silica sheets (non-exchangeable  $\text{NH}_4^+$ ); the ion may be slowly released by cations which brings about an expansion of the lattice but not by cations which collapse it (Nommik and Vahtras, 1982). In the 0–20 cm soil layer 14.0 of the urea N was present as non-exchangeable  $\text{NH}_4^+$ -N under sorghum at Rieti-Piedifiume and 24.6% at Rieti-Casabianca, and these values were higher than under wheat. (Tables 3 and 4). It is probable that different crops have different effects

Table 4. Total N and fertilizer N in the different plant and soil pools at Rieti Casabianca

Pools	Soil depth (cm)	N content ( $\mu\text{g N g}^{-1}$ dry soil)	$^{15}\text{N}$ content ( $\mu\text{g N g}^{-1}$ dry soil)	Recovery of fertilizer N (%)	NDFP (%)
<b>Sorghum</b>					
<i>Soil</i>					
Total N	0–20	1803±18	21.5±1.3	63.5±2.7	
	20–40	1792±9	11.1±0.4	32.8±1.3	
Non-exch. $\text{NH}_4^+$ -N	0–20	209.3±2.8	7.0±0.5	24.6±1.6	
	20–40	194.6±9.4	2.3±0.2	6.8±0.6	
Exch. $\text{NH}_4^+$ -N	0–20	3.0±0.2	0.03±0.0	0.1±0.0	
	20–40	5.3±0.1	0.07±0.0	0.2±0.0	
$\text{NO}_3^-$ -N	0–20	4.4±0.2	0.7±0.1	2.0±0.4	
	20–40	9.4±0.5	2.7±0.0	7.9±0.0	
Biomass-N	0–20	121.8±1.2	1.7±0.1	5.0±0.3	
	20–40	151.1±0.0	0.04±0.0	0.1±0.0	
<i>Plant</i>					
Stalks		<b>12.0±2.0<sup>a</sup></b>	<b>1.2±0.0<sup>a</sup></b>	9.5±2.6	13.3±1.5
Grain		<b>10.9±1.1<sup>a</sup></b>	<b>1.9±0.3<sup>a</sup></b>	11.1±1.7	17.3±1.0
<b>Wheat</b>					
<i>Soil</i>					
Total N	0–20	1685±27	16.7±4.4	49.0±12.9	
	20–40	1460±55	4.6±0.3	13.4± 0.8	
Non-exch. $\text{NH}_4^+$ -N	0–20	190.0±14.9	5.3±1.4	14.9± 3.8	
	20–40	206.5±10.8	1.0±0.1	3.0± 0.2	
Exch. $\text{NH}_4^+$ -N	0–20	3.9±1.1	0.06±0.0	0.2± 0.0	
	20–40	3.9±0.1	0.03±0.0	0.1± 0.0	
$\text{NO}_3^-$ -N	0–20	7.3±5.0	0.9±0.1	2.5± 0.6	
	20–40	4.4±1.2	0.2±0.1	0.7± 0.2	
Biomass-N	0–20	58.1±3.0	1.5±0.4	4.4± 1.2	
	20–40	47.2±7.6	0.6±0.2	1.8± 0.7	
<i>Plant</i>					
Straw		<b>3.8±1.0<sup>a</sup></b>	<b>1.7±0.2<sup>a</sup></b>	9.9±1.0	27.7±6.2
Grain		<b>10.3±1.4<sup>a</sup></b>	<b>2.9±0.4<sup>a</sup></b>	16.9±2.8	26.5±5.8

<sup>a</sup>In bold plant pools units were calculated by considering the yields per g of dry weight soil.

on the non-exchangeable  $\text{NH}_4$  pool. Plants can affect the concentration of non-exchangeable  $\text{NH}_4^+$  both directly, by taking ammonium in the vicinity of clay minerals thus promoting diffusion of  $\text{NH}_4^+$  ions out of the interlayer, and indirectly, by supporting the activity of heterotrophic microorganisms through the release of exudates (Marschner, 1995). Indeed rapid release of non-exchangeable  $\text{NH}_4^+$  occurs in the presence of easily available C, such as glucose, promoting the activity of the heterotrophic microflora (Mengel and Scherer, 1981; Scherer and Werner, 1996). Different plant

species are characterized by different amounts and composition of root exudates which can differently affect both activity and composition of soil microflora (Lynch, 1990; Bolton et al., 1993; Marschner, 1995). In addition, the mobilization of non-exchangeable  $\text{NH}_4^+$  can also be favoured by autotrophic microbial processes such as nitrification (Green et al., 1994).

Microbial biomass N ranged from 3.3 to 4.9% of the soil N at Foggia with lower values under sorghum than wheat and no significant differences between the two soil layers (Table 2). Similar percentages were



Table 5. Ratio of native soil N to fertilizer N in the different pools of the cropped soils

Site	Depth (cm)	Exch. NH <sub>4</sub> -N	NO <sub>3</sub> -N	Nonexch. NH <sub>4</sub> -N	Biomass N	Organic N
Sorghum (1993)						
Foggia	0–20	148c	20c	2107c	187c	264b
	20–40	326d	29c	4352d	181c	363c
Rieti	0–20	184c	11b	38a	119b	210b
Piedifume	20–40	230cd	31c	134b	220c	350bc
Rieti	0–20	97b	6a	25a	72b	127a
Casabianca	20–40	75b	3a	84b	3777d	263b
Wheat (1994)						
Foggia	0–20	7a	3a	162b	38a	110a
	20–40	8a	3a	238b	54a	112a
Rieti	0–20	57b	9b	103b	40a	79a
Piedifume	20–40	406d	400d	7000e	112b	638c
Rieti	0–20	65b	8b	37a	39a	142a
Casabianca	20–40	130c	15b	205b	79ab	377b

Means in each column followed by the same letter do not differ significantly (Duncan's Multiple Range Test at  $p=0.05$ ).

observed at Rieti-Piedifume with the exception of the surface soil layer under sorghum where microbial biomass N accounted for 13% of the total N (Table 3). Sorghum also stimulated higher microbial biomass N values than wheat at Rieti-Casabianca (Table 4). The organic C released by roots is important for sustaining microbial biomass because most soil microorganisms are heterotrophic. Different climatic conditions may explain the opposite behaviour of the two crops in sustaining microbial biomass N content of soil at Foggia with respect to Rieti sites. More severe desiccation of the surface soil layer may have occurred at Foggia than Rieti in spite of sorghum irrigation and this may have caused a greater microbial death. Susceptibility of soil microorganisms to water stress has been related to their location in the soil structure (Hattori, 1988). Microorganisms located outside microaggregates respond more rapidly to soil drying than those of the inner parts. However, a different hypothesis has been suggested by Van Gestel et al. (1996); the microbial resistance to the desiccation is determined by properties of microorganisms with fast-growing, active cells more susceptible to drying than slow-growing microorganisms. Labelled microbial biomass N was the lowest under sorghum at Foggia where it accoun-

ted for 0.8% of the urea N initially applied in both soil layers; under wheat it was 5.3% in the surface layer and 3.5% in the deeper soil layer (Table 2). At both Rieti sites this percentage was significantly higher in the surface than in the deeper soil layer while no significant differences in labelled microbial biomass were observed between surface soils sampled under the two crops (Tables 3 and 4).

At Foggia 93% of the urea N was present in the 0 to 40 cm soil layer under wheat, mainly as organic N (about 67%); the fertilizer N immobilized as organic N (about 34% of the urea N initially applied) was lower under sorghum (Table 2). The lower recoveries of fertilizer N as microbial biomass or organic N in 1993 in the Foggia soil can be due to the high plant recovery of <sup>15</sup>N and may testify the poor competition of soil microflora with sorghum for urea-N. At Rieti-Piedifume the urea N immobilized as organic N was 28 and 58% under sorghum and wheat, respectively. This difference was not observed in the Rieti-Casabianca silty clay soil where fertilizer N immobilized as organic N was about 50% in the 0 – 40 cm soil layer under the two crops. It may be confirmed that mineralization of immobilized urea N occurred in Rieti-Piedifume during summer

Table 6. Total N and fertilizer N in the different soil pools at Rieti Piedifume in the absence of the plant cover

Pools	Soil depth (cm)	N content	<sup>15</sup> N content	% of the fertilizer N applied
		$\mu\text{g N g}^{-1}$ oven dry soil	$\mu\text{g }^{15}\text{N g}^{-1}$ oven dry soil	
1993				
<i>Soil</i>				
Total N	0–20	1456±3	9.9±2.6	30.9±13.6
	20–40	1133±34	4.0±1.1	12.3±3.4
Nonexchangeable NH <sub>4</sub> <sup>+</sup> -N	0–20	149.5±1.3	6.2±0.2	19.1±0.6
	20–40	175.2±3.0	2.0±0.1	6.2±0.5
Exchangeable NH <sub>4</sub> <sup>+</sup> -N	0–20	0.9±0.0	0.01±0.0	0.02±0.0
	20–40	4.1±0.3	0.01±0.0	0.03±0.0
NO <sub>3</sub> <sup>-</sup> -N	0–20	7.7±1.1	0.8±0.2	2.6±0.7
	20–40	6.3±2.2	0.7±0.3	2.1±0.9
Biomass N	0–20	56.0±1.5	0.3±0.0	1.1±0.0
	20–40	44.8±1.3	0.1±0.0	0.4±0.1
<i>Plant</i>				
Weeds		<b>6.5±0.1<sup>a</sup></b>	<b>0.9±0.01<sup>a</sup></b>	5.5±0.1
1994				
<i>Soil</i>				
Total N	0–20	1394±63	9.2±2.8	28.6±8.9
	20–40	1231±4	2.0±0.8	6.1±2.4
Nonexchangeable NH <sub>4</sub> <sup>+</sup> -N	0–20	172.3±9.1	5.1±0.2	16.0±0.8
	20–40	171.0±9.6	0.02±0.0	0.1±0.0
Exchangeable NH <sub>4</sub> <sup>+</sup> -N	0–20	3.9±0.7	0.1±0.0	0.3±0.0
	20–40	3.5±0.2	0.01±0.0	0.03±0.0
NO <sub>3</sub> <sup>-</sup> -N	0–20	6.2±1.2	0.4±0.0	1.4±0.1
	20–40	4.8±0.6	0.2±0.0	0.7±0.0
Biomass N	0–20	45.7±1.9	1.0±0.4	3.1±0.8
	20–40	41.5±7.7	0.3±0.1	0.9±0.4
<i>Plant</i>				
Weeds		<b>6.5±0.3<sup>a</sup></b>	<b>1.9±0.1<sup>a</sup></b>	11.7±0.7

<sup>a</sup>In bold plant pools units were calculated by considering the yields per g of dry weight soil.

(the sorghum growing period) at larger extent than at Rieti-Casabianca where the swelling clays probably protected incorporated microbial metabolites derived from microbial assimilation of urea N (see above). In all sites, in 1994, more fertilizer N was recovered in soil with respect to that found by Sanaa et al. (1992).

Data pertaining to the ratio of native soil N to fertilizer N in the different N pools may give further insights in the N dynamics of the studied agroecosystems by assuming that no pool substitution occurred. Nitrate usually showed the lowest ratios with

the exception of the value of the 20 – 40 cm soil layer at Rieti-Piedifume (Table 5). At Foggia, N ratios were lower under wheat than sorghum (Table 5). The same behaviour was observed for microbial biomass N of both soil layers at both Rieti sites. This may be indicative of a more rapid N turnover in soil under wheat than sorghum. Under sorghum non-exchangeable NH<sub>4</sub><sup>+</sup>-N showed higher ratios at Foggia than at both Piedifume sites. This is indicative of a slow turnover of non-exchangeable NH<sub>4</sub><sup>+</sup>-N in the Foggia soil and it confirms that this pool is less im-

Table 7. Total N and fertilizer N in the different soil pools at Rieti Casabianca in the absence of the plant cover

Pools	Soil depth (cm)	N content $\mu\text{g N g}^{-1}$ oven dry soil	$^{15}\text{N}$ content $\mu\text{g }^{15}\text{N g}^{-1}$ oven dry soil	% of the fertilizer N applied
1993				
<i>Soil</i>				
Total N	0–20	1821±61	21.5±1.8	61.6±5.2
	20–40	1753±71	9.3±1.4	27.4±4.0
Nonexchangeable	0–20	211.3±7.6	9.3±1.1	27.4±3.4
$\text{NH}_4^+$ -N	20–40	208.8±11.3	3.8±0.2	11.5±0.7
Exchangeable	0–20	5.7±0.5	0.1±0.0	0.2±0.0
$\text{NH}_4^+$ -N	20–40	4.6±0.3	0.1±0.0	0.2±0.1
$\text{NO}_3^-$ -N	0–20	6.8±0.0	1.3±0.6	1.9±0.2
	20–40	6.7±0.1	0.8±0.2	2.2±0.5
Biomass N	0–20	81.8±0.9	1.2±0.1	3.4±0.4
	20–40	76.1±12.2	0.7±0.1	2.0±0.1
1994				
<i>Soil</i>				
Total N	0–20	1610±19	16.4±4.6	57.9±13.5
	20–40	1372±11	1.2±0.0	3.8± 0.0
Nonexchangeable	0–20	194.6±7.2	6.2±0.5	18.3± 1.6
$\text{NH}_4^+$ -N	20–40	182.7±9.5	0.5±0.2	1.3± 0.7
Exchangeable	0–20	4.7±0.5	0.1±0.0	0.2±0.1
$\text{NH}_4^+$ -N	20–40	2.9±0.0	0.01±0.0	0.02±0.01
$\text{NO}_3^-$ -N	0–20	2.6±0.1	0.3±0.0	1.0± 0.1
	20–40	3.2±0.6	0.2±0.0	0.2± 0.1
Biomass N	0–20	64.8±2.3	1.9±0.2	5.6±0.7
	20–40	39.5±0.2	0.3±0.0	0.9± 0.7
<i>Plant</i>				
Weeds		<b>8.4±0.6<sup>a</sup></b>	<b>2.4±0.2<sup>a</sup></b>	7.2± 0.7

<sup>a</sup>In bold plant pools units were calculated by considering the yields per g of dry weight soil.

portant in the urea N dynamics in Foggia than in both Rieti soils. Evidence based on measurements at short time intervals after urea-N application are needed to verify this hypothesis.

#### *Absence of the plant cover*

The absence of the plant cover did not affect the percentage of urea N present as total N in the 0–40 cm soil layer of both Rieti soils with the exception of Rieti-Piedifiume in 1994, where about 69% of the fertilizer N initially applied were found under wheat and 35% in the bare soil (Tables 3, 4, 6 and 7). Total and labelled microbial biomass N of the surface soil layer

were lower in the bare soils at Rieti-Piedifiume in both years (Tables 3 and 6) while at Rieti-Casabianca the absence of the plant cover negatively affected microbial biomass N only in 1993 (Tables 4 and 7). In bare soil the absence of organic substrates released by roots is probably responsible for not maintaining the level of microbial biomass of the cropped soil (Ladd et al., 1996). As shown above, microbial biomass N values of the surface soil layer were generally lower under wheat than sorghum. In 1994 the absence of plant cover did not negatively affect microbial biomass N at Rieti Casabianca probably because the low stimulating effect of wheat on soil microflora was cancelled by the adsorbing properties of the swelling clays that

Table 8. Ratio of native soil N to fertilizer N in the different N pools of the uncropped soils at both Rieti sites

Sites	Depth (cm)	Exch. NH <sub>4</sub> -N	NO <sub>3</sub> -N	Nonexch. NH <sub>4</sub> -N	Biomass N	Organic N
1993						
Rieti	0–20	122b	8ab	23a	194c	375c
Piedifume	20–40	512d	10b	88b	382d	711d
Rieti	0–20	83a	4a	88a	69a	49a
Casabianca	20–40	64a	8ab	52b	109c	389c
1994						
Rieti	0–20	42a	13b	32a	45a	343c
Piedifume	20–40	386c	21b	8549d	144c	4289f
Rieti	0–20	66a	6a	30a	34a	106b
Casabianca	20–40	416c	50c	397c	146c	1714e

Means in each column followed by the same letter do not differ significantly (Duncan's Multiple Range Test at  $p=0.05$ ).

reduced the availability of root exudates to soil microorganisms. It is well established that interactions of metabolites and products with clay surfaces stabilizes the organic materials (Stotzky, 1986; Hattori, 1988; Van Veen and Kuikman, 1990; Robert and Chenu, 1992; Ladd et al., 1996).

In general non-exchangeable NH<sub>4</sub>-N concentrations were not significantly affected by the absence of plant cover while urea N recoveries in this N pool were generally higher in bare than cropped soils (Tables 3, 4, 6 and 7). By processing recoveries of non-exchangeable NH<sub>4</sub><sup>+</sup>-N together, values of bare soils were significantly higher ( $p < 0.05$ ) than in cropped soils. As mentioned above plants can directly and indirectly affect the concentration of non-exchangeable NH<sub>4</sub><sup>+</sup> in soil. Under the investigated field conditions the fertilizer but not the total non-exchangeable NH<sub>4</sub><sup>+</sup>-N is affected by both crops probably because the most recently fixed NH<sub>4</sub><sup>+</sup> is available to the plant (Dou and Steffens, 1995).

In the absence of the plant cover, higher fertilizer N losses occurred at Rieti-Piedifume than at Rieti-Casabianca (Tables 6 and 7). In 1993 and 1994 about 56 and 54% of the urea-N initially applied was unaccounted for at Rieti-Piedifume and could be attributed to accumulated errors of sampling and analysis, and to leaching and gaseous losses. At Rieti-Casabianca the percentages of the urea-N unaccounted for were 17% in 1993 and 31% in 1994 (Table 6). These data further

confirm that the presence of clay stabilize the immobilized fertilizer N, reducing N mineralization rates and thus the concentration of inorganic N.

By comparing ratios of native soil N to fertilizer N in the different N pools between cropped and bare soils (Tables 5 and 8), no consistent trend appears. This may indicate that the plant cover did not affect the dynamics of fertilizer.

## Conclusions

Important findings of this research were: (1) in the presence of the plant cover the urea N unaccounted for was nil or very low (10.8% at Rieti-Casabianca under wheat and at Rieti-Piedifume 11.8% under sorghum and 4.9% under wheat); (2) non-exchangeable labelled NH<sub>4</sub><sup>+</sup>-N was an important pool in both Rieti soils, characterized by the presence of vermiculite, an NH<sub>4</sub><sup>+</sup>-N fixing clay. The greater ratios of native soil N to fertilizer N indicated that the turnover of this N pool was less important in the Foggia silty clay soil where vermiculite was not present. The presence of the plant cover affected the urea-N present as non-exchangeable NH<sub>4</sub><sup>+</sup>-N in both Rieti soils because the concentration of labelled non-exchangeable NH<sub>4</sub><sup>+</sup>-N was greater in bare than cropped soils. The plant cover also affected the urea-N losses because the urea N unaccounted for in the bare soils ranged from 12 to 56% of the urea N. Urea-N losses were higher in both years in Rieti-

Piedifume than Rieti-Casabianca soil probably due to the stabilizing effect of clays on the immobilized urea N in the latter soil.

Both sorghum and wheat N yields were higher at Foggia than at both Rieti sites probably due to higher temperatures and better light conditions during the growing season. Fertilizer N uptake was higher in sorghum than wheat at Foggia and Rieti-Piedifume but not at Rieti-Casabianca. Swelling clays of Rieti-Casabianca soil may have protected immobilized fertilizer N from microbial mineralization during summer, when sorghum was active. Mineralization of immobilized N was probably greater in summer than in late spring and thus explaining the greater percentage of fertilizer N immobilized as organic N under wheat than sorghum in Foggia and Rieti-Piedifume soils. A better competition for urea N by soil microflora in late winter and spring than summer can not be excluded when the protection of the immobilized fertilizer N by swelling clays against microbial degradation is not important.

Further insights on the processes (N uptake by plants, microbial mineralization, nitrification, NH<sub>3</sub> volatilization, etc.) affecting the behaviour of urea-N in soil require to monitor changes in native and labelled N of the various pools immediately after the application of the fertilizer N. Determinations carried out on samples collected at harvest only permit to calculate the fertilizer N balance with scarce indications on the main N processes affecting the behaviour of urea-N in the soil-plant system.

## Acknowledgement

This work was supported by the finalized project PANDA, Subproject 3 of the Ministry of Agricultural Policies.

## References

- Bremner J M and Mulvaney C S 1982 Nitrogen-total. *In* Methods of Soil Analysis, Part 2. Eds. A L Page, R M Miller and D R Keeney. pp 594–624. American Society of Agronomy, Madison, WI.
- Brookes P C, Stark J M, McInteer B B and Preston T 1989. Diffusion method to prepare soil extracts for automated nitrogen-15 analysis. *Soil Sci. Soc. Am. J.* 53, 1707–1711.
- Bolton H. Jr., Fredrickson J. K. and Elliott L. F. 1993 Microbial ecology of the rhizosphere. *In* Soil Microbial Ecology. Applications in Agricultural and Environmental Management. Eds F. B. Metting Jr. pp 27–63. Marcel Dekker, New York.
- Christensen N W and Meints V W 1982 Evaluating N fertilizer sources and timing for winter wheat. *Agronomy J.* 74, 840–844.
- Dou H and Steffens D 1995 Recovery of <sup>15</sup>N labelled urea as affected by fixation of ammonium by clay minerals. *Z. Pflanzenernähr. Bodenk.* 158, 351–354.
- Duncan R R 1996 Breeding and improvement of forage sorghums for the tropics. *Adv. Agron.* 57, 161–185.
- Green C J, Blackmer A M and Yang N C 1994 Release of fixed ammonium during nitrification in soil. *Soil Sci. Soc. Am. J.* 58, 1411–1415.
- Hamid A and Ahmad M. 1994 Effects of <sup>15</sup>N-labelled ammonium nitrate and urea on soil nitrogen during growth of wheat (*Triticum aestivum* L.) under field conditions. *Biol. Fertil. Soils* 17, 232–236.
- Hamid A and Ahmad M. 1995 Interaction of <sup>15</sup>N-labelled ammonium nitrate, and urea and ammonium sulphate on soil N during growth of wheat (*Triticum aestivum* L.) under field conditions. *Biol. Fertil. Soils* 20, 185–189.
- Hauck R D and Bremner J M 1976 Use of tracers for soil and fertilizer nitrogen research. *Adv. Agron.* 28, 219–266.
- Hauck R D, Meisinger J J and Mulvaney R L 1994 Practical considerations in the use of nitrogen tracers in agricultural and environmental research. *In* Methods of Soil Analysis. Part 2 – Microbiological and Biochemical Properties. Eds. R W Weaver, S Angle, P Bottomley, D Bezdicek, S Smith, A Tabatabai and A Wollum. pp 907–951. American Society of Agronomy, Madison, WI
- Hattori T 1988 Soil aggregates as microhabitats of microorganisms. *Report Instit. Agricul. Res. Tohoku Univ.* 37, 23–36.
- Ladd J N, Foster R C, Nannipieri P and Oades J M 1996 Soil structure and biological activity. *In* Soil Biochemistry, vol. 9. Eds. G Stotzky and J-M Bollag. pp 23–78. Marcel Dekker, New York.
- Legg J O and Meisinger J J 1982. Soil nitrogen budgets. *In* Nitrogen in Agricultural Soils. Eds. F J Stevenson. pp 503–566. American Society of Agronomy, Madison, WI.
- Lynch J M 1990 Introduction: some consequences of microbial rhizosphere competence for plant and soil. *In* The Rhizosphere. Eds J M Lynch. pp 1–10. Academic Press, New York.
- Marschner H 1995 Mineral Nutrition of Higher Plants. Second Edition. Academic Press, London.
- Marzadori C, Vittori Antisari L, Gioacchini P and Sequi P 1994 Turnover of interlayer ammonium in soil cropped with sugar beet. *Biol. Fertil. Soils* 18, 27–31.
- Mengel K and Scherer H W 1981 Release of non-exchangeable (fixed) soil ammonium under field conditions during the growing season. *Soil Sci.* 131, 226–232.
- Muller H, Frey B and Schweizer B 1992 Techniques for Flow Injection Analysis in UV/Vis spectroscopy. Bodenseewerk Perkin-Elmer GmbH Ueberlingen (Germany). pp 118.
- Mulvaney R L 1993 Mass spectrometry. *In* Nitrogen Isotopes Techniques. Eds. R. Knowles and T. H. Blackburn. pp 11–57. Academic Press, San Diego.
- Nannipieri P, Ciardi C and Palazzi T 1985 Plant uptake, microbial immobilization and residual soil fertilizer of urea-nitrogen in a grass-legume association. *Soil Sci. Soc. Am. J.* 49, 452–457.
- Nannipieri P, Ciardi C, Palazzi T and Badalucco L 1990. Short-term nitrogen reactions following the addition of urea to a grass-legume association. *Soil Biol. Biochem.* 22, 549–553.
- Nommik K H and Vahtras K 1982 Retention and fixation of ammonium and ammonia in soils. *In* Nitrogen in Agricultural Soils. Ed. F J Stevenson. pp 123–172. 22 Agronomy, American Society of Agronomy, Madison, WI.
- Powelson D S and Barraclough D 1993 Mineralization and assimilation in Soil-Plant Systems. *In* Nitrogen Isotope Techniques. Eds.

- R Knowles and T H Blackburn. pp 209–242. Academic Press, San Diego, CA.
- Powlson D S, Hart P B S, Poulton P R, Johnston A E and Jenkinson D S 1992 Influence of soil type, crop management and weather on the recovery of  $^{15}\text{N}$ -labelled fertilizer applied to winter wheat in spring. *J. Agric. Sci.* 118, 83–100.
- Robert M and Chenu C 1992. Interactions between soil minerals and microorganisms. *In Soil Biochemistry*, vol. 7. Eds. G Stotzky and J-M Bollag. pp 307–404. Marcel Dekker, New York.
- Sanaa M, Van Cleemput O, Baert L and Mhiri A. (1992). Field study of the fate of labelled fertilizer nitrogen applied to wheat on calcareous tunisian soils. *Pedologie XLII-3*, 245–255.
- SAS/STAT User's Guide, Release 6.03 Edition. Cary, NC. SAS Institute Inc. 1988.
- Scherer H W and Werner W 1996 Significance of soil microorganisms for the immobilization of nonexchangeable ammonium. *Biol. Fertil. Soils* 22, 248–251.
- Silva J A and Bremner J M 1966 Determination and isotope-ratio analysis of different forms of nitrogen in soils: 5. Fixed ammonium. *Soil Sci. Soc. Am. Proc.* 30, 587–594.
- Stevenson F J 1986 *Cycles of Soil. Carbon, Nitrogen, Phosphorus, Sulfur, Micronutrients*. John Wiley & Sons, New York.
- Stotzky G. 1986 Influence of soil mineral colloids on metabolic processes, growth adhesion, and ecology of microbes and viruses. *In Interactions of Soil Minerals with Natural Organics and Microbes*. Eds. M Huang and M Schnitzer. pp 305–428. Special Publication No 17, Soil Science Society of America, Madison, WI.
- Van Gestel M, Merckx R and Vlassak K 1996 Spatial distribution of microbial biomass in microaggregates of a silty-loam soil and the relation with the resistance of microorganisms to soil drying. *Soil Biol. Biochem.* 28, 503–510.
- Van Veen J A and Kuikman P J 1990 Soil structural aspects of decomposition of organic matter by microorganisms. *Biogeochem.* 11, 213–233.

*Section editor: R J Haynes*