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**Journal Article****Author(s):**

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**Publication date:**

2008-03

**Permanent link:**

<https://doi.org/10.3929/ethz-b-000010836>

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**Originally published in:**

Nutrient Cycling in Agroecosystems 83(3), <https://doi.org/10.1007/s10705-008-9218-7>

# Nitrogen use efficiency of $^{15}\text{N}$ -labelled sheep manure and mineral fertiliser applied to microplots in long-term organic and conventional cropping systems

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Received: 15 January 2008 / Accepted: 1 October 2008 / Published online: 19 October 2008  
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**Abstract** Nitrogen (N) utilisation by crops has to be improved to minimize losses to the environment. We investigated N use efficiency of animal manure and mineral fertiliser and fate of fertiliser N not taken up by crops in a conventional (CONMIN) and a bio-organic (BIOORG) cropping system of a long-term field experiment over three vegetation periods (winter wheat–soybean–maize). Microplots planted with wheat received a single application of  $^{15}\text{N}$ -labelled slurries (either urine or faeces labelled) or mineral fertiliser. At the end of each vegetation period we tested whether higher microbial activity and larger microbial biomass in BIOORG than CONMIN soils, and lower long-term N input level in BIOORG, affected use efficiency and fate of fertiliser N not

taken up by crops. Recovery of  $^{15}\text{N}$  in wheat was 37%, 10% and 47% from urine, faeces and mineral fertiliser, respectively, and decreased strongly in the residual years. In total 41%, 15% and 50% of  $^{15}\text{N}$  applied as urine, faeces and mineral fertiliser was recovered by the three crops.  $^{15}\text{N}$  recovered from originally applied urine, faeces and mineral fertiliser in the topsoil (0–18 cm) at the end of the third vegetation period was 19%, 25% and 20%, respectively. Of urine-, faeces- and mineral fertiliser- $^{15}\text{N}$ , 40%, 61% and 29%, respectively, was not recovered by the three crops and in topsoil suggesting significant transport of  $^{15}\text{N}$ -labelled components to deeper soil layers. CONMIN and BIOORG differed neither in fertiliser N use efficiency by crops nor in  $^{15}\text{N}$  recovery in soil indicating insignificant difference in the turnover and utilization of the applied manure nitrogen in the conventional and the bio-organic cropping systems.

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**Keywords** Animal manure · Microplot ·  
Mineral fertiliser ·  $^{15}\text{N}$  · N use efficiency ·  
Organic and conventional cropping systems

## Introduction

Intensification of agricultural production has increased nitrogen (N) fluxes between the different compartments of the biosphere and the emission of N compounds from agroecosystems to the environment

(Tilman et al. 2001). Whilst between 50% and 80% of total N applied with mineral fertiliser is taken up by the first crops, N utilisation of organic fertilisers is usually lower and more variable (Matson et al. 1997; Gutser et al. 2005; Dobermann 2005). The N remaining, that is not immobilised and stabilised into soil organic matter (SOM) fractions, is susceptible to be lost from the soil–plant system. Hence N use efficiency of fertilisers by crops has to be improved to minimise losses and thus negative impacts to the environment (Matson et al. 1997).

Organic production systems are generally assumed to be environmental-friendlier than conventional systems (Pang and Letey 2000; Reganold et al. 2001) as production is based on lower inputs and emphasis is put on recycling of nutrients in order to conserve natural resources (IFOAM 2005). The use of synthetic fertilisers is prohibited in organic farming systems (FAO 2003). Therefore, organic fertilisers (e.g. animal manure, plant residues) are the most important N sources along with SOM and biological N<sub>2</sub>-fixation. Organic compounds present in organic fertilisers and in native SOM have first to be mineralised before becoming plant available, rendering their availability dependent on microbial mineralisation and immobilisation processes (Mary et al. 1996; Rasmussen et al. 1998). Greater soil microbial activity in organically than conventionally cropped soils has been frequently reported (Gunapala and Scow 1998; Mäder et al. 2002). Nitrogen inputs to organic systems are usually lower than for conventional systems (Kirchmann and Bergström 2001; Mäder et al. 2006) and a larger N retaining capacity in organically than conventionally cultivated soils has been suggested from long-term observations (Kramer

et al. 2002). However, detailed comparison of N use efficiency of freshly applied <sup>15</sup>N-labelled mineral and organic fertilisers by crops, and of the fate of fertiliser N not taken up by the crops in the field under identical climatic and pedological conditions in conventional and organic cropping systems are rare.

We carried out a microplot study using <sup>15</sup>N-labelled sheep manure and mineral fertiliser in cropping systems that have been conventionally (CONMIN) or bio-organically (BIOORG) managed since 1978. Because of higher microbial activity and biomass in BIOORG than CONMIN and lower amount of mineral N received with organic fertilisers in BIOORG, we hypothesised different N use efficiency of fertiliser N by crops and different N recovery in soils of the two cropping systems.

## Materials and methods

### Site and experimental design

Microplots were installed in December 2002 in plots (main plots) of a long-term field experiment located in Therwil (7°33' E, 47°30' N) near Basel (Switzerland) managed by Acroscope Reckenholz-Tänikon (ART) and the Research Institute of Organic Farming (FiBL). The soil is a loamy silt Typic Hapludalf (USDA 1999) developed on loess in a temperate climate. Selected soil properties are given in Table 1.

The conception and experimental design of the field experiment have been described in detail by Mäder et al. (2006). Briefly, two conventional and two organic cropping systems are being compared since 1978. All treatments are cultivated in four

**Table 1** Selected properties of the topsoil (0–18 cm) of the investigated soils in 2003 ( $n = 4$ )

Soil	pH (H <sub>2</sub> O)	Total N (g kg <sup>-1</sup> )	Total C (g kg <sup>-1</sup> )	Available nutrients (mg kg <sup>-1</sup> )		Microbial biomass <sup>c</sup> (mg kg <sup>-1</sup> )		Daily <sup>d</sup> respiration (mg kg <sup>-1</sup> day <sup>-1</sup> )
				P <sup>a</sup>	K <sup>b</sup>	N	C	
CONMIN	6.2 b	1.4 a	13 a	9.5 a	106 a	12.5 b	101 a	2.5 b
BIOORG	6.6 a	1.5 a	14 a	5.4 a	111 a	21.8 a	113 a	3.8 a

Within a soil property, column means followed by different letters differ significantly ( $P \leq 0.05$ ) (t-test)

<sup>a</sup> Quantity of isotopically exchangeable P within the first minute (Fardeau 1993)

<sup>b</sup> Extraction with ammonium acetate EDTA measured on samples collected in March 2003 (Cottenie et al. 1982)

<sup>c</sup> Microbial biomass N and C were determined by chloroform fumigation (Vance et al. 1987); no conversion factors applied

<sup>d</sup> Average over a 60 days incubation experiment (Alef and Nannipieri 1995)

replicates in the field with the same 7-year crop rotation in a Latin square split-split-plot design. For our study we selected the four replicate plots of the conventional (CONMIN) and the bio-organic (BIOORG) cropping system. The two cropping systems mainly differ in fertilisation and plant protection. CONMIN receives exclusively water-soluble mineral fertilisers and is managed according to the rules of integrated plant production (KIP 1999) (Table 2). BIOORG is managed according to bio-organic guidelines (VSBLO 2003) and gets exclusively organic fertilisers (Table 2) with an average organic carbon (C) input of 2,240 kg ha<sup>-1</sup> year<sup>-1</sup> (Fliessbach et al. 2007). Average total N inputs are slightly lower for BIOORG than CONMIN (1985–2003) while mineral N applied with manure is clearly less in BIOORG than CONMIN where total N equals mineral N (Table 2).

Crop rotation (duration, sequence), residue management (e.g. removal of winter wheat straw) and ploughing (frequency, depth) are the same in CONMIN and BIOORG, whereas mechanical weeding is conducted more frequently in BIOORG.

The microplot study started in 2003 (year of fertiliser application) and was continued in 2004 (first residual year) and 2005 (second residual year). Winter wheat (*Triticum aestivum* var. Titlis) was cultivated in 2002/2003 followed by soybean (*Glycine max* var. Mapple Arrow) in 2004 and maize (*Zea mays* var. Gavott) in 2005. Between winter wheat and soybean a green manure mixture mainly consisting of *Phacelia tanacetifolia* was sown. It froze to death during winter and was incorporated into soil in April 2004 before sowing of soybean. After harvest of soybean, a rye (*Secale cereale*) intercrop was sown, which was mulched before sowing of maize in May 2005. Yields of winter wheat, soybean and maize were assessed in selected areas of the CONMIN and BIOORG main plots defined here as harvest plots.

The microplots were defined by frames with a length of 33 cm, a width of 14 cm and a height of 23 cm, and were open at the top and the bottom. The size and location of the microplots within the main plots represented a compromise between making use of <sup>15</sup>N-labelled fertilisers and keeping disturbance of the long-term field experiment minimal. It allowed removal of all soil contained in the microplots (about 10 kg per microplot) without seriously affecting the main plots, e.g. without removing too much soil which

would have affected overall nutrient stocks, at the end of the three-year study. In December 2002 the microplots were driven into the soil of the four field replicates of CONMIN and BIOORG main plots to a depth of 18 cm. Within the main plots they were placed at the fifth and sixth row of winter wheat at a distance of 75 cm from the main plot borders to avoid border effects and with a distance of 50 cm between the microplots to avoid cross effects. Location of microplots was optimised with regard to the wheat crop, resulting in about 20 wheat plants per microplot. The microplot location also matched to soybean growing during the following year, since microplots also encompassed two rows, resulting in two soybean plants per microplot. During the third year of the study when maize was grown, the microplots (containing one maize plant) lay between the maize rows of the main plots because microplots could not be moved. Each fertiliser treatment was repeated twice within each main plot, with one microplot being destined for destructive plant and soil sampling (disturbed microplots) during the vegetation period of winter wheat while the other was exclusively used for plant and soil sampling at maturity of crops (undisturbed microplots) (Fig. 1). The disturbed microplots were further used for sampling of green manure in 2003, of soybean at flowering in 2004 to estimate N<sub>2</sub>-fixation (Oberson et al. 2007), and of rye intercrop in 2004.

To account for fertilisation practice in CONMIN and BIOORG we evaluated the fate of both mineral fertiliser and animal manure-N. To compare the effect of long term organic versus conventional cropping on N use efficiency, we applied both animal manure and mineral fertiliser treatments to microplots installed in BIOORG and CONMIN. Tested fertiliser treatments were two <sup>15</sup>N-labelled sheep manures and one <sup>15</sup>N-labelled mineral fertiliser which were applied at beginning of tillering of winter wheat in March 2003 as a one-time, nonrecurring application. The <sup>15</sup>N-labelled fertilisers were applied at one time because soybean as the following crop received no N fertiliser. The residual <sup>15</sup>N could be used to estimate symbiotic N<sub>2</sub>-fixation by the enriched <sup>15</sup>N dilution method (Oberson et al. 2007). In 2005 we deliberately added no fresh fertiliser to study residual fertiliser-N use by maize.

The sheep manure was faeces–urine mixtures (slurries) just differing in the labelled component. One contained <sup>15</sup>N-labelled sheep urine (Surine)

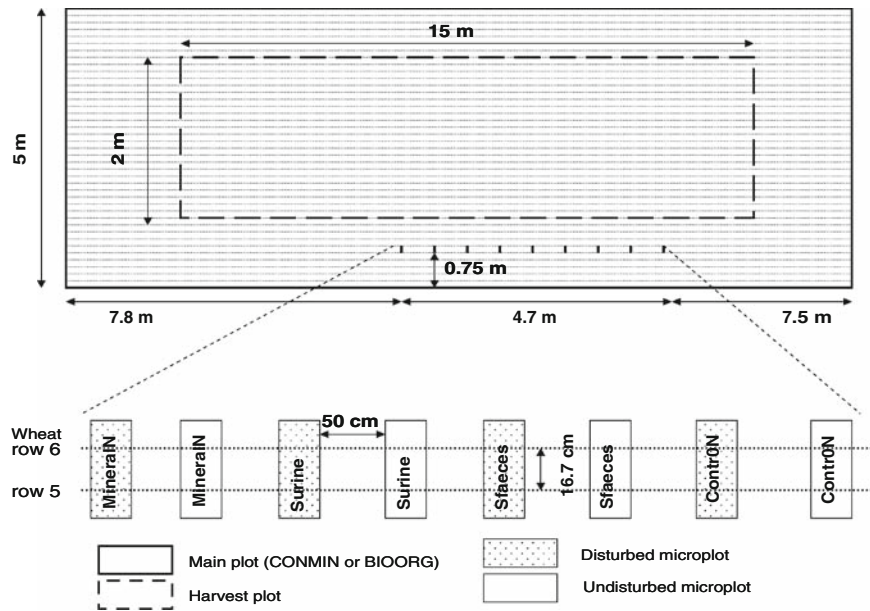
**Table 2** Selected management characteristics of the studied cropping systems

Nutrient management	CR period	Cropping system				Conventional (CONMIN)			
		Bio-organic (BIOORG)				Total N			
		Total N	Mineral N	P	K	Total N	P	K	K
Average nutrient input (kg ha <sup>-1</sup> year <sup>-1</sup> )	1978–1984	120	41	36	135	0	0	0	0
	1985–1991	100	32	31	116	101	46	225	225
	1992–1998	82	25	22	150	151	36	284	284
	1999–2003	149	59	22	315	136	37	260	260
Average budget <sup>a</sup> (kg ha <sup>-1</sup> year <sup>-1</sup> )	1978–1984	-68		4	-55	-172	-27	-114	-114
	1985–1991	-116		-1	-62	-133	11	-1	-1
	1992–1998	-89		-6	-7	-70	2	30	30
	1999–2003	-99		-13	81	-88	3	12	12
Type of manure/fertiliser		Slightly aerobically rotted farmyard-manure and slurry from 1.2 (1978–1991) or 1.4 (since 1992) livestock units ha <sup>-1</sup> year <sup>-1</sup>							
Plant protection		Unfertilized from 1978 until 1984; since 1985 exclusively water-soluble fertilisers according to official fertilization guidelines							
Weed control		Mechanical, herbicides							
Disease control		Fungicides							
Insect control		Insecticides							
Crop rotation	1978–1984	Mechanical Indirect methods Plant extracts, bio-control Winter barley ( <i>Hordeum vulgare</i> L.), 2 years grass-clover, potatoes ( <i>Solanum tuberosum</i> L.), (intercrop) <sup>b</sup> , winter wheat ( <i>Triticum aestivum</i> L.), (intercrop), white cabbage ( <i>Brassica vulgaris</i> L.), winter wheat							
	1985–1991	Winter barley, 2 years grass-clover, potatoes, (intercrop), winter wheat, (intercrop), beetroots ( <i>Beta vulgaris</i> L.), winter wheat							
	1992–1998	Three years grass-clover, potatoes, winter wheat, (intercrop), beetroots, winter wheat							
	1999–2005	Winter wheat, 2 years grass-clover, potatoes, winter wheat, (intercrop), soybean ( <i>Glycine max</i> L.), (intercrop), maize ( <i>Zea mays</i> L.)							

Average rate of N, P and K fertilisation, input–output budgets of the four crop rotation (CR) periods (1978–2003), crop rotation (1999–2005), types of fertilisers applied and methods of plant protection

<sup>a</sup> Difference between N input with fertiliser and N removed with harvested products

<sup>b</sup> Green manure or fodder intercrop



**Fig. 1** Arrangement of microplots in the main plots of the conventional (CONMIN) or the bio-organic (BIOORG) cropping system of the long-term field experiment. Treatments applied to microplots were Surine = sheep slurry ( $^{15}\text{N}$ -labelled urine + unlabelled faeces), Sfaeces = sheep slurry (unlabelled urine +  $^{15}\text{N}$ -labelled faeces), MineralN = mineral fertiliser

( $^{15}\text{NH}_4^{15}\text{NO}_3$ ) and Contr0N = unfertilised control. Disturbed microplots were used for plant and soil sampling during the growth period of winter wheat. Undisturbed microplots were exclusively used for plant and soil sampling at harvest of winter wheat, soybean and maize

while the other contained  $^{15}\text{N}$ -labelled sheep faeces (Sfaeces). The  $^{15}\text{N}$ -labelled urine and faeces were obtained by feeding a male sheep (101 kg live weight) with  $^{15}\text{N}$ -labelled ryegrass hay for 9 days and collecting urine and faeces separately. Production of the labelled sheep excrements is described in detail in Bosshard (2007). For the study, urine and faeces with highest enrichment excreted on the ninth day were applied. Using  $^{15}\text{N}$ -labelled animal manure in efficiency studies requires homogenous labelling (Powell et al. 2005; Sørensen et al. 1994a). Fraction of faeces (Langmeier et al. 2002) revealed that different faeces-N fractions (undigested dietary N, bacterial and endogenous N, water soluble N) had slightly different  $^{15}\text{N}$ -enrichments (Bosshard 2007). However, enrichment of mineralised N released from faeces during an incubation period of 112 days was not significantly different from  $^{15}\text{N}$ -enrichment of total faeces-N, showing that  $^{15}\text{N}$ -enrichment of total faeces-N could be used for N use efficiency calculation (Bosshard 2007). Mineral fertiliser (MineralN) was applied in form of  $^{15}\text{NH}_4^{15}\text{NO}_3$  as aqueous solution ( $109.7 \text{ mmol N l}^{-1}$ ). Microplots receiving no fertiliser at all served as control (Contr0N). The

applied rates and characteristics of the slurries and mineral fertiliser are shown in Table 3.

To reduce gaseous N losses and for easier application, both slurries were diluted 1:1 with water. To minimize disturbance of young winter wheat plants, slurries and mineral fertiliser were distributed into three narrow channels of about 5 cm depth and 14 cm length located between the wheat plants. The channels were covered with soil immediately after application of the fertilisers to minimise gaseous N losses, simulating direct injection. The same water volume was added to all microplots with fertilisation; otherwise microplots remained rain fed. Each cropping system-fertiliser treatment combination was repeated in four undisturbed and four disturbed microplots, respectively. The design of the study resulted in 32 undisturbed and 32 disturbed microplots, i.e. 64 microplots in total.

The original aim was to apply the same amount of  $^{15}\text{N}$ -labelled components, which would have resulted with total N applied with slurry being twice as much as with mineral fertiliser. This would have corresponded to the usual strategy in studies using  $^{15}\text{N}$ -labelled fertilisers (Thomsen et al. 1997; Langmeier



**Table 3** Selected characteristics and applied rates of the different fertilisers

Treatment	Fertiliser characteristics			Applied rates			
	Dry matter content (%)	Total N (% of DM)	<sup>15</sup> N abundance (%)	Total N (g m <sup>-2</sup> )	NH <sub>4</sub> + NO <sub>3</sub> -N (g m <sup>-2</sup> )	Total P (g m <sup>-2</sup> )	Total K (g m <sup>-2</sup> )
Surine <sup>a</sup>	7.4	6.4	5.9797	17.6	4.6	6.1	24.9
Urine	8.4	11.2	8.5127	12.7	4.5 <sup>b</sup>		
Faeces	38.1	3.2	0.3767	4.9	0.1		
Sfaeces <sup>a</sup>	5.9	6.9	3.6700	15.9	4.6	7.9	29.5
Urine	8.7	12.6	0.4487	11.1	4.4 <sup>c</sup>		
Faeces	34.3	3.5	11.2770	4.8	0.2		
MineralN <sup>a</sup>	–	–	9.8685	5.0	5.0	0	0
Contr0N	–	–	–	0	0	0	0

<sup>a</sup> Surine = <sup>15</sup>N-labelled urine + unlabelled faeces, Sfaeces = unlabelled urine + <sup>15</sup>N-labelled faeces, MineralN = <sup>15</sup>NH<sub>4</sub><sup>15</sup>NO<sub>3</sub> (applied as a solution), Contr0N = unfertilised control

<sup>b</sup> Difference between mineral N determined in Surine and faeces of Surine

<sup>c</sup> Difference between mineral N determined in Sfaeces and faeces of Sfaeces

et al. 2002). Because of an initial analytical problem more than twice as much urine-N than faeces-N or mineral fertiliser-N was applied. Still, N use efficiency of the different fertiliser components can be tested because the main aim of this study was to compare N use efficiency between CONMIN and BIOORG and not to compare the fertilisers among each other. Additionally none of the fertiliser treatments resulted in significant added N interaction (ANI) (see Discussion below). During fertilisation of the main plots, microplots were covered to avoid additional N input. All other crop cultivation measures (e.g. plant protection and application of plant growth regulator in CONMIN) conducted in the main plots of CONMIN and BIOORG were also carried out at the same time in the microplots. Because of the small area of the microplots (0.0462 m<sup>2</sup>) ploughing and mechanical weeding were done by manual work with hand-held tools.

#### Plant and soil sampling and sample preparation

Above-ground plant biomass of microplots was removed completely at physiological maturity of crops, i.e. of winter wheat in July 2003, of soybean in September 2004 and of maize in September 2005, when also the main plots were harvested. At harvests a part of root biomass of winter wheat and soybean was collected down to about 10 cm and for maize down to 18 cm using a small spade to loosen the soil

such that roots could be removed. Due to differences in absorption rate and translocation of N, <sup>15</sup>N-enrichment is known to differ in different plant parts, which may limit accurate determination of <sup>15</sup>N excess of the whole plant (Danso et al. 1993). Therefore, winter wheat was divided into stem, leaves, chaff, grains and roots, soybean into stem, leaves, pod, grains and roots, and maize into stem, leaves, tassel, silk, husk, cob, grains and roots. Additionally leaves from two wheat plants were collected from disturbed microplots 11, 47, 75, 97 and 112 days after fertiliser application. This corresponds to the following growth stages of winter wheat: tillering, stem elongation, flowering, dough stage and physiological maturity. All plant materials were dried at 45°C for 48 h.

Six randomly distributed soil cores were collected from undisturbed microplots with an auger (diameter 2.5 cm, Eijkelkamp, Netherlands) at maturity of winter wheat and soybean, respectively, down to 28 cm. The soil cores were then divided into the 0–18 and 18–28 cm soil layers and air-dried. After harvest of maize, all the topsoil in the microplots to a depth of 18 cm was removed from the main plots and soil was then air-dried, thoroughly mixed and prepared for analyses. The <sup>15</sup>N removed from microplots with soil sampled at harvest of winter wheat and soybean accounted for 1.1–1.4% and 1.5–3.7%, respectively, of initially added total <sup>15</sup>N and was taken into account for calculation of <sup>15</sup>N-recoveries. For nitrate-<sup>15</sup>N determination soil samples taken from

destructive microplots 11 and 112 days after fertiliser application were used.

## Analyses

### Total N, $^{15}\text{N}$ and mineral N in sheep manures

Faeces, urine and slurry samples were freeze-dried after acidification with concentrated  $\text{H}_2\text{SO}_4$  to minimize gaseous losses. Faeces and slurries were afterwards finely ground using a ball mill (Retsch, Haan, Germany). Total N and  $^{15}\text{N}$  abundance in freeze-dried faeces, urine and slurry samples were determined using a continuous flow Roboprep CN Biological Sample Converter coupled to a Tracermass Mass Spectrometer (Europa Scientific, Crewe, England). Mineral N ( $\text{NH}_4\text{-N}$  and  $\text{NO}_3\text{-N}$ ) in faeces and slurries was extracted as described in Langmeier et al. (2002) except for filtering the KCl solutions through Whatman No. 1 filters (Davidson et al. 1991).  $\text{NH}_4\text{-N}$  and  $\text{NO}_3\text{-N}$  were colorimetrically analysed on a  $\text{SAN}^{\text{plus}}$  Analyzer (Skalar, Netherlands).

### Total N and $^{15}\text{N}$ in plants

The dried plant parts from winter wheat, soybean and maize were homogenised separately by cutting the plant materials into small pieces with a centrifuge mill (Granomat, Fuchs Maschinen AG, Switzerland) and then finely ground using the same ball mill as for faeces and slurries. The dried fine roots, tassel and silk from maize were directly ground with the ball mill. Total N and  $^{15}\text{N}$  abundance analyses were carried out on the mass spectrometer previously mentioned.

### Total N, $^{15}\text{N}$ and mineral N in soil

Air-dried soil samples were finely ground with a ball mill prior to total N and  $^{15}\text{N}$  analyses.  $\text{NO}_3\text{-N}$  and soluble and exchangeable  $\text{NH}_4\text{-N}$  were determined using a 2 M KCl extraction as described in Davidson et al. (1991). Analysis for  $\text{NH}_4\text{-N}$  and  $\text{NO}_3\text{-N}$  was conducted colorimetrically using the  $\text{SAN}^{\text{plus}}$  Analyzer. Sequential diffusion of ammonium and nitrate from soil extracts to a polytetrafluoroethylene (PTFE) trap was used to determine  $\text{NH}_4\text{-}^{15}\text{N}$  and  $\text{NO}_3\text{-}^{15}\text{N}$  as described by Sørensen and Jensen (1991).  $^{15}\text{N}$  analyses were conducted on the mass spectrometer previously mentioned.

### Atom% $^{15}\text{N}$ excess

The atom%  $^{15}\text{N}$  excess of each sample denotes the  $^{15}\text{N}$  abundance of the sample minus the natural abundance of its reference sample. For reference, soil or plant material obtained under identical experimental conditions in the microplots of the ContrON treatment were used. Natural abundance of the reference plant and soil material was 0.37 atom%  $^{15}\text{N}$ .

## Calculations

The amount of N derived from the  $^{15}\text{N}$ -labelled component of the fertilisers in different plant parts ( $\text{Ndfc}_{\text{plp}}$ ) of winter wheat, soybean and maize, respectively, was calculated according to Eq. 1 using isotope dilution principles (Hauck and Bremner 1976):

$$\text{Ndfc}_{\text{plp}} (\text{g m}^{-2}) = N_{\text{plp}} ({}^{15}\text{Nex}_{\text{plp}} / {}^{15}\text{Nex}_{\text{lc}}) \quad (1)$$

where  $N_{\text{plp}}$  is the total N amount ( $\text{g m}^{-2}$ ) in the corresponding plant part,  ${}^{15}\text{Nex}_{\text{plp}}$  its atom%  $^{15}\text{N}$  excess (%), and  ${}^{15}\text{Nex}_{\text{lc}}$  the atom%  $^{15}\text{N}$  excess of the labelled component of the fertiliser (%). The quotient ( ${}^{15}\text{Nex}_{\text{plp}} / {}^{15}\text{Nex}_{\text{lc}}$ ) denotes the fraction of N derived from the labelled component. Expressed in percent it is:

$$\% \text{Ndfc}_{\text{plp}} = ({}^{15}\text{Nex}_{\text{plp}} / {}^{15}\text{Nex}_{\text{lc}}) \times 100 \quad (2)$$

If  $\text{Ndfc}_{\text{plp}}$  was obtained for  $n$  different plant parts then N derived from the labelled component of the fertiliser in the whole plant ( $\text{Ndfc}$ ) is:

$$\text{Ndfc} (\text{g m}^{-2}) = \sum_{i=1}^n \text{Ndfc}_{\text{plp} i} \quad (3)$$

The N derived from the fertilisers ( $\text{Ndff}$ ,  $\text{g m}^{-2}$ ) for slurry is the sum of N derived from urine and from faeces. Because of similar composition of the two slurries (Table 3) we assumed that the percentage of N derived from faeces in Surine was the same as in Sfaeces. This assumption is considered to be also valid for urine in Sfaeces. The  $\text{Ndff}$  was then assessed as follows: N derived from the unlabelled manure component in each plant part was calculated by multiplying the proportion of  $\text{Ndfc}_{\text{plp}}$  from the labelled treatment with the N amount of the plant part from the treatment with the unlabelled manure component. For MineralN  $\text{Ndff}$  corresponds to  $\text{Ndfc}$ .



For winter wheat and maize, N derived from soil (Ndfs) was calculated by:

$$\text{Ndfs}(\text{g m}^{-2}) = \text{N}_{\text{plant}} - \text{Ndff} \quad (4)$$

where  $\text{N}_{\text{plant}}$  ( $\text{g m}^{-2}$ ) denotes total N in the respective crop.

N derived from the atmosphere (Ndfa) in soybean grown in microplots was estimated by the  $^{15}\text{N}$  dilution method in a study evaluating  $\text{N}_2$ -fixation of soybean (Oberson et al. 2007). Ndfs for soybean could then be calculated as follows:

$$\text{Ndfs}(\text{g m}^{-2}) = \text{N}_{\text{plant}} - \text{Ndff} - \text{Ndfa} \quad (5)$$

The  $^{15}\text{N}$  recovered in each crop ( $\text{Rec}_{\text{crop}}$ ) was calculated using Eq. 6:

$$\text{Rec}_{\text{crop}}(\%) = \text{Ndflc}/\text{Nlc} \times 100 \quad (6)$$

where  $\text{Nlc}$  ( $\text{g m}^{-2}$ ) denotes total N applied with the labelled component and  $\text{Ndflc}$  ( $\text{g m}^{-2}$ ) is obtained from Eq. 3 for wheat, soybean and maize, respectively.

Recovery of manure- or mineral fertiliser-derived  $^{15}\text{N}$  remaining in the soil of the microplots was calculated according to Eq. (7):

$$\text{Rec}_{\text{soil}}(\%) = (\text{V}_{\text{mp}} \times \text{d} \times \text{N}_{\text{soil}} \times {}^{15}\text{Nex}_{\text{soil}}) / (\text{Nlc} \times {}^{15}\text{Nex}_{\text{lc}}) \times 100 \quad (7)$$

where  $\text{V}_{\text{mp}}$  ( $\text{dm}^3$ ) denotes microplot volume,  $\text{d}$  ( $\text{kg dm}^{-3}$ ) bulk density,  $\text{N}_{\text{soil}}$  ( $\text{g kg}^{-1}$ ) soil N concentration and  ${}^{15}\text{Nex}_{\text{soil}}$  atom%  $^{15}\text{N}$  excess of soil. Bulk density was for both CONMIN and BIOORG  $1.3 \text{ kg dm}^{-3}$  for the 0–18 cm soil layer and  $1.4 \text{ kg dm}^{-3}$  for the 18–28 cm soil layer (Oehl et al. 2002). The calculated content ( $\text{V}_{\text{mp}} \times \text{d}$ ) of  $10.8 \text{ kg soil}$  per microplot to a depth of 18 cm was confirmed by the effective soil weight determined after harvest of maize when microplots were removed from main plots. Average variation between effective and calculated weight was 2.9%. Because of technical reasons recovery of  $^{15}\text{N}$  in the 18–28 cm soil layer was only assessed after harvest of winter wheat and soybean. Thus, unaccounted  $^{15}\text{N}$  at the end of three vegetation periods was calculated as difference between the amount of  $^{15}\text{N}$  added with the labelled component of the fertiliser and the amount of  $^{15}\text{N}$  removed by the three crops plus the  $^{15}\text{N}$  recovered in the soil (0–18 cm) after harvest of maize.

## Statistical analysis

Analysis of variance was performed by using the GLM procedure of the statistical analyses package SYSTAT 11 (Systat Software Inc., USA). Effects of cropping system and fertiliser treatments on fertiliser N use efficiency and  $^{15}\text{N}$  recovery in soil as well as interactions between cropping systems and fertiliser treatments were tested using a two-way ANOVA. For analysis of variance percentage data was transformed using arcsin-transformation. In case of significant effects separation of means was conducted using Tukey's honestly significant difference (HSD) test with a significance level of  $P \leq 0.05$ .

## Results

### Soil characteristics

Soil microbial biomass and activity were higher in BIOORG than CONMIN in March 2003 (Table 1). This confirms several previous measurements on microbial biomass and activity in the same soils (Fließbach and Mäder 2000; Mäder et al. 2002). Soil pH was lower in CONMIN than BIOORG, probably due to acidifying effect of mineral fertilisers (Mäder et al. 2006). Available phosphorus (P) and potassium (K) levels of the CONMIN and BIOORG soils can be classified as moderate to sufficient (Gallet et al. 2003; Walther et al. 2001). In spite of long term organic fertilisation in BIOORG, total C and N concentrations in soils were not significantly different between CONMIN and BIOORG (Table 1). This agrees with Fließbach et al. (2007) who did an extended study on soil organic C in the same field experiment. Average N budgets of CONMIN and BIOORG, calculated as difference between N input by fertilisers and N removed with harvested products, are negative (Table 2).

### Weather conditions

In 2003, precipitation was lower than long-term (1864–2004) mean values, except in October. In March 2003 when the fertilisers were applied into microplots precipitation was very low. From April 2004 until September 2004 monthly rainfall was below the long-term mean values (Fig. 2a).

Exceptionally high air temperatures which resulted in high soil temperatures were measured from June 2003 until August 2003 (Fig. 2b). From September 2003 until September 2004 measured air temperatures agreed with the long-term mean air temperature curve (Fig. 2b).

## Grain yields and total N uptake

### Winter wheat

Grain dry matter (DM) yield of winter wheat grown in microplots in July 2003 did not differ between the two cropping systems and was comparable to yields from the harvest plots (460 g DM m<sup>-2</sup> for CONMIN and BIOORG) (Table 4). Grain yield was affected by the fertiliser treatment: Surine ≥ Sfaeces = MineralN ≥ Contr0N (Table 4) and was strongly linearly

correlated to total N input with the fertilisers ( $R^2 = 0.84$ ). Total N taken up by winter wheat at maturity ranged from 12.8 g N m<sup>-2</sup> up to 19.5 g N m<sup>-2</sup> (Table 5).

### Soybean

Grain yield (500–620 g DM m<sup>-2</sup>) of soybean harvested from microplots in September 2004 neither differed between the two cropping systems nor between the fertiliser treatments (Table 4) and was higher than in the harvest plots (280–330 g DM m<sup>-2</sup>). Total N contained in mature soybean ranged from 33.0 g N m<sup>-2</sup> up to 39.9 g N m<sup>-2</sup> (Table 5) and was not correlated with the amount of N applied with fertilisers in 2003.

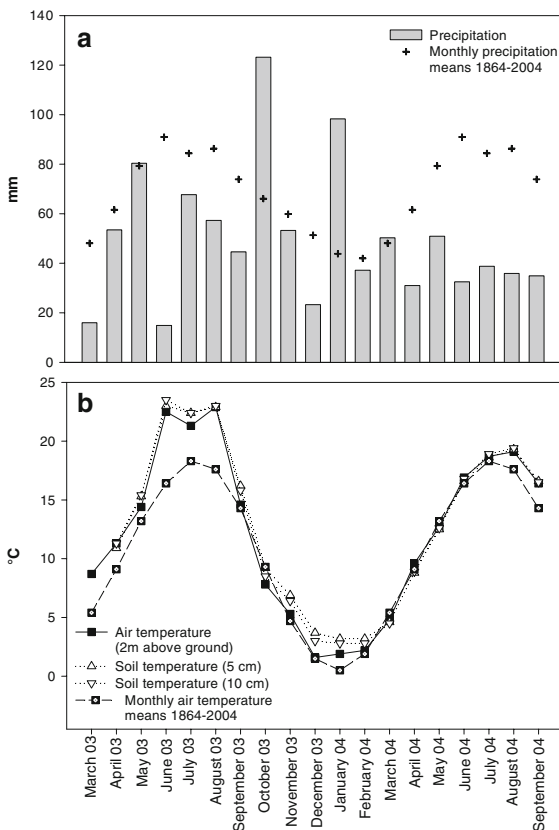
### Maize

Grain yield of maize harvested from microplots in September 2005 was not significantly different between the two cropping systems and the fertiliser treatments (Table 4) but, on average, was 40% lower than in the harvest plots (CONMIN: 1,900 g DM m<sup>-2</sup>, BIOORG: 1,500 g DM m<sup>-2</sup>). Total N uptake by maize at maturity ranged from 16.4 to 21.5 g N m<sup>-2</sup> and was not correlated with total N input with fertilisers in 2003 (Table 5).

N derived from the labelled component of the fertilisers (Ndfc) in winter wheat, soybean and maize

Neither %Ndfc<sub>plp</sub> (%) for winter wheat (Fig. 3) nor the absolute Ndfc (g m<sup>-2</sup>) for winter wheat, soybean and maize (Table 5) were significantly affected by the cropping systems. At harvest 24%, 3% and 16% of total N taken up by winter wheat was derived from urine, faeces and mineral fertiliser, respectively. The %Ndfc<sub>plp</sub> in leaves collected from disturbed microplots was greater for urine and mineral fertiliser than for faeces from the earliest sampling onwards (Fig. 3). However, while %Ndfc<sub>plp</sub> remained stable for urine-N (Surine), it slightly decreased for mineral fertiliser-N (MineralN) and slightly increased for faeces-N (Sfaeces).

There were only minor differences in N concentration (mg N g<sup>-1</sup> DM) in wheat during its growth (data not shown), which was at a level of



**Fig. 2** Precipitation and air and soil temperature measured by the meteorological station installed in the long-term field experiment and monthly long-term (1864–2004) precipitation and air temperature means measured at the meteorological station Basel-Binningen (7°34' E, 47°32' N)

**Table 4** Grain dry matter (DM) yields of winter wheat, soybean and maize of the fertiliser treatments

	2003 Winter wheat (g grain DM m <sup>-2</sup> )	2004 Soybean (g grain DM m <sup>-2</sup> )	2005 Maize (g grain DM m <sup>-2</sup> )
Fertiliser treatment <sup>a</sup>			
Surine	547 a	585 ns	1,211 ns
Sfaeces	457 ab	623 ns	1,015 ns
MineralN	421 ab	495 ns	996 ns
Contr0N	338 b	543 ns	909 ns
Standard error	46	130	257

Because of no significant differences between CONMIN and BIOORG only mean values are shown ( $n = 8$ )

Within columns, means followed by different letters are significantly different ( $P \leq 0.05$ ) by Tukey's multiple range test; *ns* not significant

<sup>a</sup> Surine = <sup>15</sup>N-labelled urine + unlabelled faeces, Sfaeces = unlabelled urine + <sup>15</sup>N-labelled faeces, MineralN = <sup>15</sup>NH<sub>4</sub><sup>15</sup>NO<sub>3</sub>, Contr0N = unfertilised control

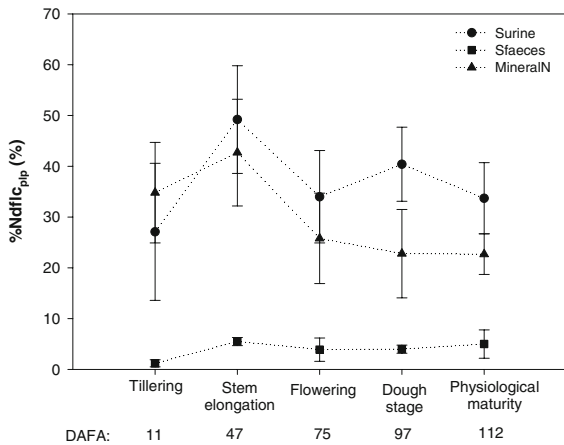
**Table 5** Total N uptake by winter wheat, soybean and maize, N derived from the labelled component of fertiliser (Ndfc), N derived from the complete fertiliser (Ndff), N derived from the soil (Ndfs) in crops and N derived from the atmosphere (Ndfa) in soybean of the fertiliser treatments at the year of fertiliser application (2003) and the residual years (2004 and 2005)

	N uptake (g m <sup>-2</sup> )	Ndfc (g m <sup>-2</sup> )	Ndff (g m <sup>-2</sup> )	Ndfa (g m <sup>-2</sup> )	Ndfs (g m <sup>-2</sup> )
<i>Winter wheat (July 2003)</i>					
Fertiliser treatment <sup>a</sup>					
Surine	19.5	4.64	5.2	–	14.4
Sfaeces	16.6	0.48	4.5	–	12.1
MineralN	15.1	2.36	2.4	–	12.8
Contr0N	12.8	–	–	–	12.8
Standard error	1.7	0.2	0.5	–	1.5
<i>Soybean (September 2004)</i>					
Fertiliser treatment <sup>a</sup>					
Surine	40.1	0.33	0.5	19.5	20.1
Sfaeces	39.9	0.16	0.5	18.0	21.3
MineralN	33.0	0.10	0.1	10.1	22.8
Contr0N	37.8	–	–	nd	nd
Standard error	9.1	0.03	0.06	7.2	7.0
<i>Maize (September 2005)</i>					
Fertiliser treatment <sup>a</sup>					
Surine	21.5	0.17	0.3	–	21.3
Sfaeces	19.9	0.07	0.2	–	19.6
MineralN	21.3	0.05	0.1	–	21.2
Contr0N	16.4	–	–	–	16.4
Standard error	3.8	0.02	0.03	–	3.8

Because of no significant differences between CONMIN and BIOORG only mean values are shown ( $n = 8$ )

*nd* not defined

<sup>a</sup> Surine = <sup>15</sup>N-labelled urine + unlabelled faeces, Sfaeces = unlabelled urine + <sup>15</sup>N-labelled faeces, MineralN = <sup>15</sup>NH<sub>4</sub><sup>15</sup>NO<sub>3</sub>, Contr0N = unfertilised control



**Fig. 3** Percentage of N derived from the labelled component of manure or mineral fertiliser in leaves of winter wheat (%Ndfc<sub>plp</sub>) at different growth stages. DAFA = days after fertiliser application. Because of no significant differences between CONMIN and BIOORG mean values only are shown (*n* = 8)

27 mg N g<sup>-1</sup> grain DM at physiological maturity suggesting that none of the fertiliser treatments resulted in N limiting concentrations (Walther et al. 2001). The high availability of urine-N and mineral fertiliser-N was also reflected in the soil nitrate N pool. At eleven days (tillering of winter wheat) after fertiliser application, nitrate-N deriving from urine, mineral fertiliser and faeces accounted for 76%, 65% and 8% of the soil nitrate, respectively, and decreased to 9%, 3% and 2%, respectively, at 112 days (harvest of winter wheat) (Table 6).

**Table 6** Total NO<sub>3</sub><sup>-</sup>-N and NO<sub>3</sub><sup>-</sup>-N derived from the labelled component of fertiliser (NO<sub>3</sub>dfc) in the topsoil (0–18 cm) of the fertiliser treatments 11 days (tillering of winter wheat) and

Fertiliser treatment <sup>a</sup>	11 days after fertiliser application (mg kg <sup>-1</sup> soil)		112 days after fertiliser application (mg kg <sup>-1</sup> soil)	
	Total NO <sub>3</sub> <sup>-</sup>	NO <sub>3</sub> <sup>-</sup> dfc	Total NO <sub>3</sub> <sup>-</sup>	NO <sub>3</sub> <sup>-</sup> dfc
Surine	70.9 (33.4) ns	55.2 (30.0) a	6.7 (2.0) ns	0.6 (0.3) a
Sfaeces	55.1 (16.8) ns	4.2 (2.2) b	5.7 (1.2) ns	0.1 (0.08) b
MineralN	40.2 (14.5) ns	26.6 (12.3) b	6.0 (2.0) ns	0.2 (0.1) b

Because of no significant differences between CONMIN and BIOORG only mean values are shown. Standard deviation is shown in brackets (*n* = 8)

Within columns, means followed by different letters are significantly different (*P* ≤ 0.05) by Tukey’s multiple range test; *ns* not significant

<sup>a</sup> Surine = <sup>15</sup>N-labelled urine + unlabelled faeces, Sfaeces = unlabelled urine + <sup>15</sup>N-labelled faeces, MineralN = <sup>15</sup>NH<sub>4</sub><sup>15</sup>NO<sub>3</sub>, ContrON = unfertilised control

Compared to the year of fertiliser application, Ndfc of slurries and mineral fertiliser decreased strongly in the residual years. Of total N taken up by soybean in the first residual year, 0.8% was derived from urine, 0.4% from faeces, and 0.3% from mineral fertiliser (Table 5). The Ndfc of all fertilisers remained on low level in the second residual year with 0.8% of total N taken up by maize deriving from residual urine-N, 0.4% from residual faeces-N and 0.2% from residual mineral fertiliser-N (Table 5).

The percentage of total N derived from the complete slurries (Ndf) taken up by winter wheat, by soybean and by maize, was 27%, 1.3% and around 1.2%, respectively. The remaining N taken up by the crops originated from the soil (Table 5). Furthermore, about 50% of total N taken up by soybean was derived from the atmosphere (Table 5).

### Fertiliser <sup>15</sup>N recovery

No significant differences in <sup>15</sup>N recovery were found between the two cropping systems. At harvest 37%, 10% and 47% of urine-, faeces- and mineral fertiliser-N was recovered in winter wheat (Table 7). At first residual year, <sup>15</sup>N recovered from originally applied labelled fertilisers by soybean decreased to 2.6%, 3.3% and 2.1% for urine, faeces and mineral fertiliser, respectively (Table 7). In second residual year, 1.3% of urine-<sup>15</sup>N, 1.5% of faeces-<sup>15</sup>N and 1.1% of mineral fertiliser-<sup>15</sup>N was recovered in maize (Table 7). Fertiliser-derived <sup>15</sup>N recovered in the topsoil (0–18 cm) was each year between 20% and 25%

112 days (physiological maturity of winter wheat) after fertiliser application

**Table 7** Fertiliser-<sup>15</sup>N recovery in winter wheat, soybean, maize and the soil of the fertiliser treatments at the year of fertiliser application and the residual years

	Fertiliser treatment <sup>a</sup>			
	Surine <sup>15</sup> N recovery (% of applied <sup>15</sup> N)	Sfaeces	Slurry (weighted) <sup>c</sup>	MineralN
<i>Year of fertiliser application 2003</i>				
Winter wheat	36.6 (3.7)	10.0 (2.0)	29.2 (3.1)	47.1 (4.2)
Soil (0–18 cm)	25.1 (10.9)	20.1 (6.3)	23.7 (8.7)	21.8 (6.5)
Soil (18–28 cm) <sup>b</sup>	2.0 (1.7)	2.4 (2.7)	2.2 (1.6)	2.5 (2.1)
<i>First residual year 2004</i>				
Soybean	2.6 (0.7)	3.3 (0.5)	2.8 (0.5)	2.1 (0.3)
Soil (0–18 cm)	25.2 (3.8)	47.1 (19.6)	31.2 (3.7)	22.4 (4.0)
Soil (18–28 cm)	7.1 (1.1)	8.6 (4.0)	7.5 (1.9)	7.2 (2.8)
<i>Second residual year 2005</i>				
Maize	1.3 (0.3)	1.5 (0.2)	1.4 (0.3)	1.1 (0.2)
Soil (0–18 cm)	19.4 (3.2)	24.5 (5.9)	20.8 (1.7)	20.3 (11.9)
Soil (18–28 cm)	nd	nd	nd	nd

Because of no significant differences between CONMIN and BIOORG only mean values are shown. Standard deviation is shown in brackets ( $n = 8$ )

nd not defined

<sup>a</sup> Surine = <sup>15</sup>N-labelled urine + unlabelled faeces, Sfaeces = unlabelled urine + <sup>15</sup>N-labelled faeces, MineralN = <sup>15</sup>NH<sub>4</sub><sup>15</sup>NO<sub>3</sub>, Contr0N = unfertilised control

<sup>b</sup> Significant effect of cropping system (BIOORG > CONMIN)

<sup>c</sup> Weighted recovery from Surine and Sfaeces

except for faeces-<sup>15</sup>N in 2004 where it was about twice as high, but at high variation (Table 7). At harvest of winter wheat between 2% and 3% and at harvest of soybean 7–9% of fertiliser-<sup>15</sup>N was recovered in the 18–28 cm soil layer (Table 7). Thirty months after application of the labelled fertilisers—at harvest of maize—unaccounted fertiliser-<sup>15</sup>N not taken up by the crops or recovered in the 0–18 cm soil layer amounted to 40%, 61% and 29% for urine-, faeces- and mineral fertiliser-N, respectively.

## Discussion

Limitations and advantages associated with use of <sup>15</sup>N-labelled fertilisers

The use of <sup>15</sup>N-labelled animal manure is restricted by high costs and high expenditure of time to produce homogeneously labelled manures (Powell et al. 2004; Sørensen et al. 1994b). This sets certain limits. We worked with <sup>15</sup>N-labelled faeces and urine that were excreted on the last day of feeding <sup>15</sup>N-labelled hay

obtained from one sheep. The amount of these excretions restricted the area of microplot that could be fertilised, and in turn, the number of plants that could be sampled. In addition, work with <sup>15</sup>N-enriched materials in a long-term field experiment where natural abundance work is also being carried out needs to be carefully rationalized. Spatial limitation by microplot frames is essential as any measure has to be taken to avoid contamination of the experimental side. As previously said, <sup>15</sup>N-labelled soil was to be removed at the end of our study. Therefore, the amount of labelled soil was kept small. In spite of these constraints, the stable isotope <sup>15</sup>N was highly useful to investigate N use efficiency by crops grown under the different cropping systems. The advantages of isotope techniques over the difference method, where crop N uptake from fertilised and non fertilised plots is being compared, is recognised (Hood 2001; Muñoz et al. 2004). Finally, only <sup>15</sup>N-labelled fertilisers allowed us to trace the fate of N not taken up by crops in BIOORG and CONMIN soil–plant systems over three vegetation periods.

## Dry matter production

Grain dry matter production of winter wheat grown in microplots at harvest in July 2003 was comparable to grain yield of winter wheat grown in harvest plots. The P and K concentrations in grains of wheat ranged from 4.1 to 4.3 g kg<sup>-1</sup> DM. Phosphorus and K concentrations in grains of wheat grown in microplots of treatments MineralN and Contr0N where no P and K was added in March 2003 were not significantly different from those of wheat grains obtained on microplots that received slurry. Also P and K concentrations in grains of wheat grown in microplots did not differ significantly from wheat grown in harvest plots of the field trial. Thus, growth of winter wheat plants of treatments MineralN and Contr0N seems not having been limited by P or by K. Lower DM production in Contr0N than fertilised treatments most probably was caused by N limitation.

Grain yield of soybean harvested from microplots in September 2004 was higher than in the harvest plots. This probably can be ascribed to the fact that grains of soybean in microplots were harvested manually and thus losses were reduced compared to harvest plots where soybean was harvested with a combine harvester. Furthermore, only two soybean plants could be grown per microplot hampering extrapolation of grain yield to larger areas. Total dry matter production of soybean in microplots was lower than of soybean sampled in areas of the main plots corresponding to microplot area, suggesting that growth of soybean was restricted by the microplot frames (Oberson et al. 2007).

The 40% lower grain yield of maize harvested in September 2005 from microplots compared to harvest plots could be explained by the following: (i) microplots remained unfertilized since March 2003 and, besides N (Walther et al. 2001), soil P- and K-availability might have limited plant growth and thus dry matter production as shown for soybean (Oberson et al. 2007), (ii) light conditions for maize in microplots were not optimal during the vegetation period as microplots were located between two rows of maize of main plots and maize in microplots was growing slower than in main plots due to no addition of fresh fertiliser and (iii) restriction of maize root growth by microplot frames down to a depth of 18 cm.

## N use efficiency and recovery in soils of animal manure and mineral fertiliser

### *Impact of cropping system*

Despite the differences in microbial activity, biomass and long-term fertilisation between CONMIN and BIOORG no differences in fertiliser N use efficiency were found. For a same N fertiliser (i.e. urine, faeces or mineral fertiliser), total <sup>15</sup>N recovery in crop and 0–18 cm soil layer did not differ significantly between CONMIN and BIOORG. Also, for a same fertiliser hardly any differences were found in the distribution of <sup>15</sup>N among different aggregate and size density fractions obtained from CONMIN and BIOORG soils (Bosshard et al. 2008). Thus, the fate of mineral or animal manure N seems not to be affected by activity and size of microbial biomass initially present in the soil. This finding is consistent with results reported by Langmeier et al. (2002) who conducted a pot experiment using the same soils with <sup>15</sup>N-labelled cattle manure and mineral fertiliser being applied to ryegrass. They reported a similar response of CONMIN and BIOORG to fertilisation. In contrast to our results, however, they found BIOORG to have a greater soil N mineralisation capacity than CONMIN which may be ascribed to the fact that several cuts of ryegrass were grown on a limited soil volume under N limiting conditions. Glendining et al. (1997) similarly found that the crop uptake of <sup>15</sup>N-labelled mineral fertiliser was unaffected by the previous management history of soil in a long-term experiment. Also net N and C mineralisation from cowpea leaves have been found to be unaffected by the level of the indigenous soil microbial biomass (Franzluebbers et al. 1995). In contrast to Langmeier et al. (2002) and to our study, where same fertilisers were applied to both soils from organic and conventional systems, Kramer et al. (2002) applied <sup>15</sup>N-labelled urea to a long-term conventionally and <sup>15</sup>N-labelled vetch plus unlabelled manure to a long-term organically managed cropping system. At higher recovery of urea-N (40%) than vetch-N (20%) in plants, total <sup>15</sup>N recoveries in plants plus the 0–15 cm soil layer were similar (organic: 73%, conventional: 63%) because recovery of N in soil from vetch residues (44%) was significantly higher than from urea (15%). Still, from this different approach they also concluded that



conventional and organic cropping systems do not differ in the capacity to retain N.

### *Impact of fertiliser type*

Direct comparison of N use efficiency of urine with faeces and mineral fertiliser might be hampered as a higher amount of total N and thus  $^{15}\text{N}$  was applied with urine in Surine compared to faeces in Sfaeces or mineral fertiliser in MineralN. Added N interaction (ANI) might also obscure the interpretation of results (Jenkinson et al. 1985; Kuzyakov et al. 2000), e.g. if the application of labelled N influences plant uptake of soil-derived N. In our study ANI can be neglected for several reasons. First, Ndfs was not significantly different between Contr0N and fertilised treatments, and Ndfs was only slightly and not significantly greater in Surine than the other fertilised treatments. Also, recovery of slurry-N and mineral fertiliser-N by winter wheat was similar for the  $^{15}\text{N}$  method or when calculated by the difference method (Muñoz et al. 2004) where the apparent recovery is defined as (N uptake in fertilised treatment – N uptake in Contr0N treatment)/total N applied  $\times$  100. The  $^{15}\text{N}$  method resulted in 29% and 47%  $^{15}\text{N}$  recovery for slurry and mineral fertiliser (Table 7), and the apparent N recoveries were 31% and 46%, respectively.

Recovery of mineral fertiliser-derived  $^{15}\text{N}$  in the crops declined strongly from 47% in the year of fertiliser application to below 2.5% in residual years. This low recovery in the second year might be due to an exhaustion of this N source as nearly half of originally applied mineral fertiliser-N was taken up by winter wheat in the year of fertiliser application and around 30% remained unaccounted.

Probably because of the same reasons as for mineral fertiliser, recovery of urine- $^{15}\text{N}$  in crops declined strongly from 37% in the year of fertiliser application to less than 3% in residual years. The N use efficiency of faeces-N in the year of fertiliser application was only 10%, which is in the range reported by Thomsen et al. (1997). Organic N forms accounted for the largest fraction of total faecal N and thereof about 30% were undigested dietary N (Bosshard 2007). Organic N compounds in faeces that remained undigested after having passed the animal are supposed to mineralise very slowly in soil (Muñoz et al. 2003; Sørensen and Jensen 1998). In the year of fertiliser application a small amount of organic faeces-N was mineralised at a

slow rate as the amount of faeces-derived N in winter wheat exceeded the amount of mineral-N applied with labelled faeces and as also suggested by the increasing proportion of %Ndfc<sub>plp</sub> during wheat growth. Low recovery in soybean (3.3%) and maize (1.5%) suggests that mineralisation of faeces-N was also low during residual years.

### *Fate of fertiliser N not taken up by crops*

Urine- and mineral fertiliser-derived  $^{15}\text{N}$  remaining in soil after each vegetation period were comparable to results found in other studies applying  $^{15}\text{N}$ -labelled animal manure to soils, whilst recovery of faeces- $^{15}\text{N}$  was about three times lower (Muñoz et al. 2003; Sørensen et al. 1998; Thomsen et al. 1997). Only the recovery of faeces- $^{15}\text{N}$  in soil in 2004 was comparable to results in other studies. Low recovery in 2003 might have been caused by sampling bias due to non homogenous distribution of  $^{15}\text{N}$ -labelled faeces in microplots. However, all soil contained in a microplot was thoroughly mixed in 2005 before sampling for  $^{15}\text{N}$  abundance analyses. These results confirmed low recovery of faeces- $^{15}\text{N}$  in soil. Because of recalcitrant N compounds in faeces, faeces-N is expected to accumulate in soil as shown by Sørensen et al. (1998) and Thomsen et al. (1997). The low recovery of faeces-N in soil is difficult to explain. Gaseous N losses, i.e. ammonia volatilisation, denitrification and nitrate leaching require mineralisation of applied N. However, as indicated by the low availability to crops, and as confirmed in incubation studies (Bosshard 2007), mineralisation of faeces N was low. Furthermore, slurry was applied to a depth of 5 cm and then covered with soil to minimise ammonia volatilisation. The low recovery of faeces-derived  $^{15}\text{N}$  suggests that non-mineralised faeces-N would have been lost from microplots for example by leaching of dissolved organic N or by redistribution of faeces-N outside the microplots by earthworms that were present numerously in microplots, e.g. into deeper soil layers by anecic earthworms. The recovery in the 18–28 cm soil layer suggests  $^{15}\text{N}$  displacement into deeper layers. It ranged from 2.0% to 2.5% in the year of fertiliser application and increased to 7.1–8.6% in the first residual year. Sørensen and Thomsen (2005) similarly found 10–13% of applied faecal  $^{15}\text{N}$  in the 20–40 cm soil layer 1 year after application and Muñoz et al. (2003)

found 4–8% in the 30–60 cm soil layer and another 1–4% in the 60–90 cm soil layer. These studies indicate that transport of labelled N below 18 cm depth could be around 20%, but low recovery in our study suggests an even higher transport of labelled faecal N to deeper soil layers.

Unaccounted  $^{15}\text{N}$  30 months after application of labelled fertilisers amounted to 40%, 61%, and 29% for urine, faeces and mineral fertiliser, respectively. Despite higher microbial activity in BIOORG than CONMIN soils the fate of fertiliser-N was the same in both systems for a same fertiliser applied at same rate as shown by similar fertiliser N use efficiency by crops,  $^{15}\text{N}$  recovery in the soil and unaccounted  $^{15}\text{N}$ . This suggests that the two cropping systems have the same potential to emit N compounds to the environment. Differences in the local emissions from cropping systems seem to be determined by form and level of inputs rather than by intrinsically different use efficiencies of inputs.

## Conclusions

Higher microbial activity and biomass in BIOORG than CONMIN soils was expected to affect the fate of added fertiliser-N through mineralisation-immobilisation processes. However, the cropping system had no significant impact on N use efficiency of animal manure or mineral fertiliser and on recovery of fertiliser-N in the soil over a 3-year period. N use efficiency was affected by the type of fertiliser. Recalcitrant N-compounds as contained in faeces were mineralised at a low rate in the year of fertiliser application which was expressed in low Ndfc and low recovery of faeces-N in winter wheat. The residual fertiliser N effect was very low for all tested fertilisers but comparable to other studies. Especially in residual years, the main N source for crops was the soil and for soybean additionally the atmosphere, again without significant effects of the cropping system. In spite of lower recovery of faeces-N in crops at the end of the third vegetation period,  $^{15}\text{N}$ -recovery in the 0–18 cm soil layer was similar for urine, faeces and mineral fertiliser. As recalcitrant faeces-derived compounds are expected to accumulate in soil,  $^{15}\text{N}$  recovered from faeces was unexpectedly low. The low  $^{15}\text{N}$ -recovery may be explained by the transport of faeces- $^{15}\text{N}$  to deeper

soil layers by earthworm activity and/or leaching of dissolved organic N.

**Acknowledgements** We gratefully acknowledge W. Jossi (ART) and R. Frei (FiBL) for their help in the field work as well as L. Gunst and U. Zihlmann (both ART) for providing data records on the field experiment. The authors thank H.-R. Roth (Seminar for Statistics, ETH) for advice in statistics, S. Recous (INRA, France) for helpful discussions and the anonymous reviewers for their helpful comments on the manuscript. We warmly thank M. Stocki (University of Saskatchewan, Saskatoon) for providing mass spectrometry analyses and R. Ruh, T. Flura, T. Rösch and S. Douxchamps (all from Group of Plant Nutrition, ETH) for technical assistance in the field and the laboratory.

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