

# Effects of green manure on soil nitrogen availability and crop productivity in a Mediterranean organic farming system

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## Abstract

European farmers have developed an increasing interest in organic agriculture, but often without including animal husbandry in their farming system. In this context, the use of green manure together with adequate residue management and crop rotation is crucial to conserve or increase soil organic matter and promote nutrient cycling for adequate crop uptake at farm scale. Five cover crops (*Vicia faba minor*, *Brassica juncea*, *Trifolium incarnatum*, *Avena sativa* + *Trifolium incarnatum*, *Trifolium pratense*) to be used as green manure and a no-cover crop control were compared in a field trial included in a larger farming system comparison between conventional and organic management for a 5-year arable crop rotation (sugar beet-common wheat-sunflower-pigeon bean-durum wheat). Green manure crops were included in the organic management system in-between wheat and the subsequent cash crops (sugar beet and sunflower). Experimental measurements, carried out in 2003, included: 1) cover crops biomass, nitrogen accumulation and weed control; 2) cash crop productivity and nitrogen recovery; 3) mineralisation of organic matter and subsequent N release after incorporation of green manure in soil. *Trifolium pratense* showed the highest biomass production and *Brassica juncea* the lowest. The two cash crops were influenced differently by green manure. Sunflower grain yield was higher after legumes, while sugar beet root yield did not differ among treatments. The concentration of NO<sub>3</sub> in soil after green manure incorporation showed significant differences among treatments, with higher values associated with legume presence compared to *Brassica juncea* and the control.

**Keywords:** organic agriculture, cover crops, green manure, stockless system, N supply.

## Introduction

Organic farms in Europe are often managed as stockless systems (Schmidt *et al.*, 1999). Davids *et al.* (1996), Stopes *et al.* (1996) and von Fragstein *et al.* (1996) pointed out the importance that stockless system acquired in France, the U.K. and Germany.

Italian farmers also have developed an increasing interest in organic agriculture but, as typical of Mediterranean areas, without including animal husbandry in their farming systems.

Even though the exclusion of animal husbandry turns into simplification of farm management, stockless farming systems often suffer from insufficient nitrogen availability to crops (Bulson *et al.*, 1996).

In this context, the use of green manure together with adequate residue management and crop rotation is crucial to conserve or increase soil organic matter content and promote nutrient cycling at farm scale to the benefit of crops.

In Mediterranean conditions, green manure are generally represented by winter cover crops. The problems that organic farmers had to face when choosing species to be used as green manure are: adaptability to agro-pedo-climatic conditions, biomass production in relation to sowing time of the following cash crops, nitrogen release and recovery after green manure incorporation in soil, control of weeds and soil borne pathogens.

To help clarify these points, a field trial was established in autumn 2002 comparing five cover crops included for green manure use in a 5-year arable crop rotation managed organically. The aim of the trial was to evaluate: 1) cover crop biomass, nitrogen supply and weed control; 2) productivity and nitrogen recovery of subsequent cash crops; 3) mineralisation of organic matter and subsequent N release following green manure incorporation in soil.

## Materials and methods

This research has been carried out in the 2002-03 growing season as a part of a long-term experiment established in 2001 at the Centro Interdipartimentale di Ricerche Agro-Ambientali E. Avanzi (CIRAA) of the University of Pisa (Italy) that compares a conventional and an organic management system for a 5-year stockless arable crop rotation (sugar beet-common wheat-sunflower-pigeon bean-durum wheat). In the organic system, red clover is interseeded into the wheat to serve as green manure for the subsequent cash crops (sugar beet and sunflower). Within this system, a specific trial was established to evaluate alternative cover cropping strategies. Five cover crops (*Vicia faba minor*, *Brassica juncea*, *Trifolium incarnatum*, *Avena sativa* + *Trifolium incarnatum*, *Trifolium pratense*) plus a no-cover crop control were then compared. Soil characteristics of the two fields used for the trial are reported in Table 1.

**Table 1.** Soil characterisation of trial fields (0-30 cm soil depth)

		<i>Sugar beet field</i>	<i>Sunflower field</i>
pH		8.45	8.46
Organic matter	(%)	1.54	1.48
Total N *	(g N kg <sup>-1</sup> )	1.14	1.09
Assimilatable P **	(mg P <sub>2</sub> O <sub>5</sub> kg <sup>-1</sup> )	15.11	12.56
Sand	(%)	43	44
Silt	(%)	36	35
Clay	(%)	21	21

\* Kjeldahl method, \*\* Olsen method

In each field, the experimental design was a randomised complete block with four replications, for a total of 24 plots field<sup>-1</sup>. Plot area was 100 m<sup>2</sup>. Details on cover crop varieties, seed rate and C/N ratio are reported in Table 2.

**Table 2.** Cover crop varieties, seed rate and C/N ratio

Species	Common name	Variety	Seed rate (kg ha <sup>-1</sup> )	C/N
<i>Trifolium pratense</i>	Red clover	Raja	35	27
<i>Vicia faba</i> var. <i>minor</i>	Pigeon bean	Vesuvio	120	14
<i>Trifolium incarnatum</i>	Crimson clover	Tombolo	30	13
<i>Avena sativa</i> / <i>Trifolium incarnatum</i>	Oats + crimson clover	Argentina + Tombolo	100 + 30	18
<i>Brassica juncea</i>	Brown mustard	ISCI 61	9	32

Cover crops were sown on 20 October 2002, except for red clover that was interseeded in February 2002 into the wheat crops that preceded sugar beet and sunflower .

Sowing of subsequent cash crops was delayed compared to the optimum period to obtain adequate cover crop biomass yield. On 8 April 2003 cover crops were incorporated by disc

harrowing followed by mouldboard ploughing. Aboveground cover crop biomass production was estimated in two 1 m<sup>2</sup> sampling areas in each plot. Samples were oven-dried at 75°C until constant weight and biomass was expressed on a dry matter basis.

Sugar beet (cv. *Puma*) and sunflower (cv. *Carlos*) were sown on 16 April 2003 at a seed rate of 18 and 8 seeds m<sup>-2</sup> respectively. Direct weed control consisted in mechanical hoeing + hand hoeing in sugar beet and in mechanical hoeing alone in sunflower. Cash crops yield was assessed at harvest time in one 2 m<sup>2</sup> sampling area plot<sup>-1</sup> and expressed as either fresh matter (for sugar beet) or dry matter biomass (for both crops). Cover crop and cash crop samples were then ground and analysed for total N concentration by the Kjeldahl method.

Soil NO<sub>3</sub><sup>-</sup> and total N concentration in the top 30 cm of the profile were determined at the following times: 8 April (immediately before cover crop incorporation), 23 April, 22 May, 19 June, 8 and 16 September 2003 (the latter two dates corresponded to sunflower and sugar beet harvest respectively). Soil total N was also measured on 20 October 2002, i.e. at sowing of winter cover crops.

Soil samples were ground, mineralised and analysed for total N (Kjeldahl method) and nitrate concentration (ionic chromatography) (Benedetti *et al.*, 2000).

Experimental data were subjected to analysis of variance (ANOVA). Soil total N values were square-root transformed prior to ANOVA because error variances were heterogeneous. Mean treatment effects were compared by a LSD test at the 0.05 level of probability (CoStat software).

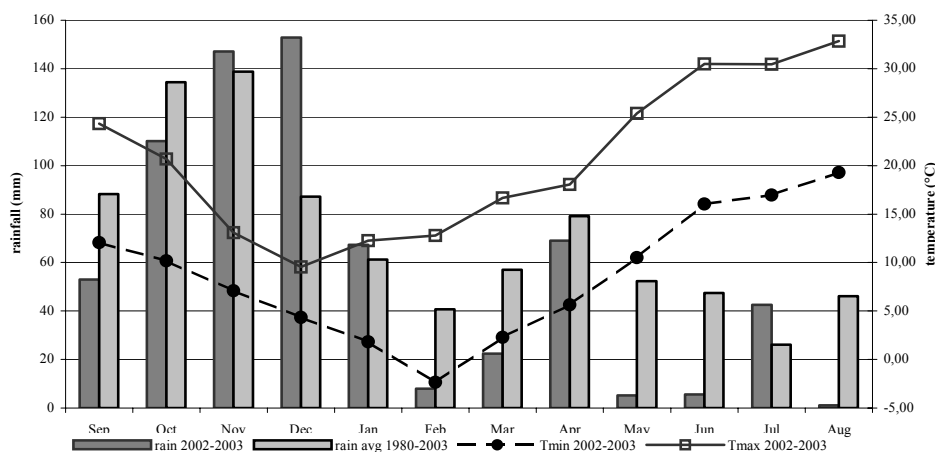
## Results and discussions

### *Climatic trend*

Values of precipitation and maximum/minimum air temperature of the period September 2002–August 2003 are shown in Figure 1.

The results obtained in 2003 may be affected by an abnormal climatic trend characterised by heavy rainfall during the 2002–03 winter and by absence of significant rainfall coupled with high temperature during the 2003 summer. High winter precipitation reduced cover crops biomass, particularly that of *Brassica juncea*, a species particularly sensitive to waterlogging. The hot and dry summer of 2003 influenced cash crops performance, that yielded much less than usual.

**Figure 1.** Climatic trend at CIRAA, University of Pisa, Italy (September 2002–August 2003).



## Cover crop and weed biomass and N supply

Biomass production and N supply in cover crops and weeds are shown in Tables 3 and 4.

**Table 3.** Cover crop and weed dry matter biomass. Sugar beet field.

Species	Cover crops (t ha <sup>-1</sup> )	Weeds (t ha <sup>-1</sup> )	Total (t ha <sup>-1</sup> )	N supply by cover crops (kg ha <sup>-1</sup> )	N supply by weeds (kg ha <sup>-1</sup> )	Total N supply (kg ha <sup>-1</sup> )
Red clover	4.82 a	0.12 b	4.94 a	90.80 a	2.88 bcd	93.68 a
Pigeon bean	2.48 b	0.49 b	2.97 b	76.46 ab	7.85 b	84.32 ab
Oats + crimson clover	2.14 bc	0.12 b	2.26 b	57.46 ab	1.44 d	58.90 b
Crimson clover	1.60 bc	0.53 ab	2.13 bc	48.96 bc	7.28 bc	56.24 b
Brown mustard	0.95 c	0.15 b	1.10 cd	13.13 c	1.96 cd	13.81 c
Control	- -	0.94 a	0.94 d	- -	13.31 a	13.31 c
Significance (F test)	**	**	**	*	**	**

In each column, means followed by the same letter are not significantly different at  $P \leq 0.05$  (LSD test). \*, \*\*, significant at  $P \leq 0.05$  and  $P \leq 0.01$  respectively.

**Table 4.** Cover crop and weed dry matter biomass. Sunflower field.

Species	Cover crops (t ha <sup>-1</sup> )	Weeds (t ha <sup>-1</sup> )	Total (t ha <sup>-1</sup> )	N supply by cover crops (kg ha <sup>-1</sup> )	N supply by weeds (kg ha <sup>-1</sup> )	Total N supply (kg ha <sup>-1</sup> )
Red clover	3.87 a	0.05 c	3.92 a	66.74 a	0.96 c	67.70 ab
Pigeon bean	2.33 b	0.55 ab	2.88 b	69.58 a	8.31 a	77.89 a
Oats + crimson clover	1.84 b	0.12 bc	1.96 c	48.45 a	1.41 bc	48.87 b
Crimson clover	1.96 b	0.44 abc	2.40 bc	61.62 a	6.86 ab	68.49 ab
Brown mustard	0.68 c	0.09 bc	0.77 d	11.18 b	1.44 bc	12.62 c
Control	- -	0.62 a	0.62 d	- -	8.58 a	8.58 c
Significance (F test)	**	(*)	**	*	*	**

In each column, means followed by the same letter are not significantly different at  $P \leq 0.05$  (LSD test). (\*), \*\*, significant at  $P \leq 0.10$ ,  $P \leq 0.05$  and  $P \leq 0.01$  respectively.

In both sugar beet and sunflower fields, *Trifolium pratense* showed the highest production of biomass (4.82 and 3.87 t ha<sup>-1</sup> respectively) and *Brassica juncea* the lowest (0.95 and 0.68 t ha<sup>-1</sup>).

Conversely, weed biomass was lowest in red clover plots (0.12 and 0.05 t ha<sup>-1</sup>) and highest in the control (0.94 and 0.62 t ha<sup>-1</sup>) and *Trifolium incarnatum* plots (0.53 and 0.44 t ha<sup>-1</sup>). Weed biomass in ISCI61 brown mustard was low despite reduced cover crop biomass, an effect likely due to the species allelopathic activity (Brown *et al.*, 2004).

The amount of nitrogen accumulated in cover crops biomass did not show any significant differences in the sunflower field except from brown mustard, that showed the lowest value (11.2 kg ha<sup>-1</sup>). In the sugar beet field, significant differences were found between red clover (90.8 kg ha<sup>-1</sup>) and crimson clover (49.0 kg ha<sup>-1</sup>), while brown mustard still showed the lowest value (13.1 kg ha<sup>-1</sup>).

Total (cover crop + weeds) N supply showed a similar trend in the two fields (red clover and pigeon bean > crimson clover with/without oats > brown mustard, as absolute values).

## Cash crops yield parameters and N uptake

The two cash crops included in this experiment were influenced differently by green manure crops. Sunflower appeared to be more influenced by green manure than sugar beet. Compared to the control, sunflower grain and total yield were positively influenced by all green manure crops except brown mustard. In particular, sunflower grain yield increased from 34% (crimson clover) to 73% (pigeon bean) as compared to the control (Table 5).

**Table 5.** Sunflower yield parameters following green manure.

Species	Grain (t ha <sup>-1</sup> )	Total (t ha <sup>-1</sup> )	Harvest Index (%)	Plants (n m <sup>-2</sup> )	1000 seeds weight (g)
Red clover	1.99 sab	5.88 a	34.0	6.2	42.01
Pigeon bean	2.13 a	6.14 a	35.0	6.1	39.53
Oats + crimson clover	1.83 bc	5.39 a	35.0	6.8	32.43
Crimson clover	1.65 c	5.22 ab	32.5	7.1	30.59
Brown mustard	1.23 d	4.16 b	30.0	6.6	28.14
Control	1.23 d	4.12 b	30.5	6.9	28.61
Significance (F test)	**	(*)	ns	ns	ns

In each column, means followed by the same letter are not significantly different at  $P \leq 0.05$  (LSD test). (\*), (\*\*), ns: significant at  $P \leq 0.10$ ,  $P \leq 0.01$  and not significant respectively.

**Table 6.** Sunflower N uptake.

Species	Grain N uptake (kg ha <sup>-1</sup> )	Total N uptake (kg ha <sup>-1</sup> )
Red clover	41.66 ab	60.65 ab
Pigeon bean	44.78 a	65.60 a
Oats + crimson clover	35.62 abc	51.72 b
Crimson clover	33.61 bc	50.38 bc
Brown mustard	25.47 cd	36.89 c
Control	24.81 d	37.21 c
Significance (F test)	*	*

In each column, means followed by the same letter are not significantly different at  $P \leq 0.05$  (LSD test). \*: significant at  $P \leq 0.05$ .

Total and grain nitrogen concentration in sunflower did not show any significant differences among treatments (data not shown), therefore sunflower nitrogen uptake showed the same trend previously observed for yield, with higher values for pigeon bean and red clover and lower for brown mustard and the control (Table 6). Sunflower nitrogen uptake and nitrogen supply from cover crops biomass were highly correlated ( $r^2 = 0.86^{**}$ ). Sugar beet production was negatively affected by climatic conditions and by late sowing (ca. two months later than usual), that was necessary to allow cover crops early spring growth and performance of the stale-seedbed technique. For these reasons, differences among treatments were less evident as compared to sunflower and not significant, an effect also probably due to the poorer sugar beet capacity of catching nitrogen from the soil (Table 7). No significant differences among treatments were observed in N uptake and no correlation between cash crops N uptake and cover crops N supply was found (data not shown).

**Table 7.** Sugar beet yield parameters following green manure.

Species	Roots FM (t ha <sup>-1</sup> )	Roots DM (t ha <sup>-1</sup> )	Total DM (t ha <sup>-1</sup> )	Roots (n m <sup>-2</sup> )	Sugar content (%)
Red clover	18.42	4.13	5.22	5.81	20.38
Pigeon bean	12.24	2.93	3.61	5.06	22.88
Oats + crimson clover	13.61	3.00	3.75	5.50	20.38
Crimson clover	13.71	3.30	4.11	5.38	21.88
Brown mustard	11.30	2.72	3.38	6.06	22.00
Control	15.59	3.63	4.43	6.63	21.13
Significance (F test)	ns	ns	ns	ns	ns

FM, fresh matter; DM, dry matter; ns, not significant.

### ***Green manure effects on soil organic matter mineralisation***

#### ***a) Total soil N content***

In both fields, total soil nitrogen content did not differ significantly among treatments throughout the whole sampling period. Values ranged between 1.08 and 1.17 g kg<sup>-1</sup> in the sugar beet field and between 1.05 and 1.15 g kg<sup>-1</sup> in the sunflower field.

#### ***b) Soil nitrate release***

In the sugar beet field (Table 8 and Figure 2) low values of soil nitrates were observed in the first period (8 and 23 April 2003). In both dates, higher values were found in the red clover plots than in the other treatments, probably due to the fact that red clover below-ground biomass was already at a more advanced mineralisation stage, since the biological cycle of the crop ended just before winter.

On 8 May 2003, i.e. one month after cover crops incorporation, soil nitrates increased, presumably due to the onset of the mineralisation process. At that date, red clover and crimson clover showed the highest NO<sub>3</sub><sup>-</sup> values (43.03 e 41.52 mg kg<sup>-1</sup> respectively).

**Table 8.** Soil NO<sub>3</sub><sup>-</sup> content (mg kg<sup>-1</sup>) in the sugar beet field.

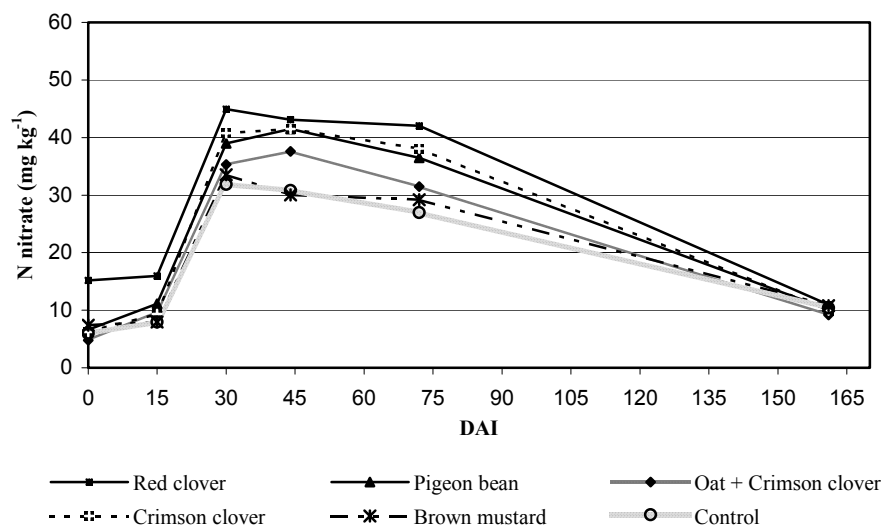
Species	Sugar beet field						
	Date	08.04.03	23.04.03	08.05.03	22.05.03	19.06.03	16.09.03
Days after incorporation		0	15	30	44	72	161
Red clover		15.18 a	15.97 a	44.92 a	43.08	42.06	10.97
Pigeon bean		6.63 b	11.10 b	38.98 bc	41.48	36.50	10.15
Oats + crimson clover		4.79 b	9.73 bc	35.28 bcd	37.57	31.46	9.16
Crimson clover		6.23 b	9.33 bc	40.76 ab	41.51	38.08	9.94
Brown mustard		7.41 b	7.93 c	33.55 cd	30.04	29.21	10.79
Control		5.93 b	7.95 c	31.84 d	30.79	26.99	10.40
Significance (F test)		**	**	**	ns	ns	ns

In each column, means followed by the same letter are not significantly different at P≤0.05 (LSD test). \*\*: significant at P≤0.01; ns, not significant.

In all treatments, soil nitrate content consequent to green manure mineralisation peaked between 30 and 44 days after incorporation (DAI).

No significant differences among treatments were observed 44, 72 and 161 DAI. From 44 DAI onwards, soil nitrates decreased in all treatments, likely because of the gradual extinction of the labile organic matter mineralisation process, a greater uptake by sugar beet and leaching consequent to late-summer rainfall events.

Figure 2. Soil NO<sub>3</sub><sup>-</sup> content in the sugar beet field.



Contrary to the sugar beet field, in the sunflower field (Table 9 and Figure 3) an overall increase in soil nitrate content was already observed 15 DAI. Furthermore, soil nitrate release lasted for a longer period compared to the sugar beet field, showing the highest concentrations 72 DAI, with the exception of red clover (44 DAI) and brown mustard (30 DAI). Differences in mineralisation peak periods are likely due to differential aging of residues at the time of incorporation in soil.

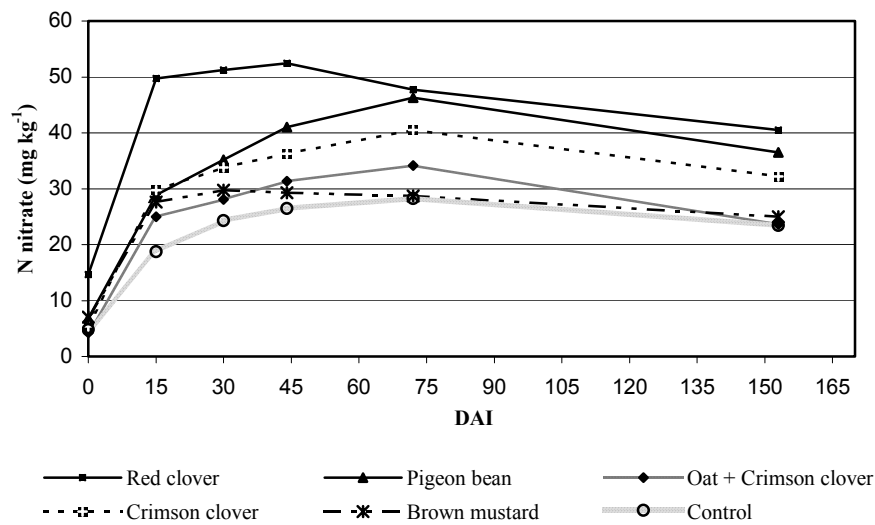
Soil nitrates following red clover incorporation were considerably higher than following all the other treatments from 0 to 30 DAI. From the fourth sampling date (44 DAI) onwards, the value for pigeon bean did not significantly differ from that of red clover. Soil nitrates released from crimson clover reached the same levels of red clover and pigeon bean 72 DAI and persisted until sunflower harvest.

Table 9. Soil NO<sub>3</sub><sup>-</sup> content (mg kg<sup>-1</sup>) in the sunflower field.

Treatment	Sunflower field						
	Date	08.04.03	23.04.03	08.05.03	22.05.03	19.06.03	8.09.03
Days after incorporation		0	15	30	44	72	153
Red clover		14.64 a	49.75 a	51.24 a	52.41 a	47.76 a	40.51 a
Pigeon bean		6.84 b	28.89 b	35.19 b	41.01 ab	46.29 ab	36.52 ab
Oats + crimson clover		4.27 b	24.97 b	28.06 b	31.32 bc	34.16 bc	23.60 c
Crimson clover		5.39 b	29.75 b	33.79 b	36.26 bc	40.53 abc	32.19 abc
Brown mustard		7.06 b	27.60 b	29.72 b	29.30 bc	28.73 c	24.96 bc
Control		4.79 b	18.76 b	24.32 b	26.48 c	28.28 c	23.46 c
Significance (F test)		**	*	**	**	*	*

In each column, means followed by the same letter are not significantly different at P≤0.05 (LSD test). \*, \*\*: significant at P≤0.05 and P≤0.01 respectively.

Figure 3. Soil NO<sub>3</sub><sup>-</sup> content in the sunflower field.



Significant correlations between maximum nitrate concentration during the summer period and total nitrogen released by cover crops were observed for both sugar beet ( $r^2 = 0.92^{**}$ ) and sunflower ( $r^2 = 0.86^{**}$ ) fields, hence levels of nitrates in soil are mostly due to cover crops nitrogen release.

## Conclusions

Despite the confounding effect of abnormal weather conditions in the 2002-03 season, some preliminary indications emerged from this research. Of the green manure crops tested, red clover and pigeon bean exerted the maximum beneficial effect on sunflower yield and N uptake, whereas green manure effects on sugar beet were less evident. Soil nitrate release was correlated with the amount of N supplied by green manure.

These results confirm that green manure are able to enhance N nutrition of spring-sown cash crops thanks to their N input in soil (Smith and Sharpley, 1993; Schimdt *et al.*, 1999; Stopes *et al.*, 1996).

However, the goal of maximising N supply can contrast with the optimum sowing date of subsequent cash crops thereby increasing the risk of reduced production.

It should be pointed out that accurate evaluation of the advantages and disadvantages of green manure crops and their management should be carried out not only taking into account following cash crop productivity (short-term vision) but also the effects on soil fertility dynamics (long-term vision).



## References

- Benedetti A., Trincherà A., Falchini L., Antisari L.V., (2000). Determinazione degli ioni nitrato per cromatografia ionica. *Metodi di analisi chimica del suolo*, Franco Angeli, sez. XIV, 32.
- Brown, J., Hamilton, M. & Brown, D.A. (2004). Using brassicaceae seed meal as an alternative to highly toxic soil fumigants in strawberry production. Proceedings of the 1<sup>st</sup> International Symposium "Biofumigation: a possible alternative to methyl bromide?". Firenze, Italy, 31 March - 1 April: Research Institute for Industrial Crops of the Italian Ministry of Agricultural and Forestry Policies, 14-15.
- Bulson, H.A.J., Welsh, J.P., Stopes, C.E. & Woodward, L. (1996). Agronomic viability and potential economic performance of three organic four year rotations without livestock, 1988-1995. *Aspect of Applied Biology*, **47**, 227-286.
- David, C., Fabre, B. & Gautronneau, Y. (1996). Toward modelling the conversion of the stockless farming to organic farming. On-farm research in South East of France. In *New Research in Organic Agriculture* (N.H. Kristensen H. Hogh-Jensen, eds.), 23-27. IFOAM; Okozentrum Imsbach, Tholey-Theley.
- Fragstein, P. von., (1996). Organic arable farming – a contradiction?. In *Fourth congress of the ESA – book of Abstract* (M.K. van Ittersum, G.E.G.T. Venner, S.C. van de Geijn & T.H. Jetten, eds.), Vol. 2, 438-439, European Society of Agronomy, Colmar Cedex, NL.
- Laureti, D., Pieri S., (2001). Sovescio di leguminose nella fertilizzazione del girasole. *L'Informatore Agrario* **30**, 52-54.
- Mazzoncini, M. & Bàrberi, P. (2002). La gestione della fertilità del suolo. *AZ Bio*, 10, 58-64.
- Schmidt, H., Philipps, L., Welsh, J.P., Fragstein, P.v. (1999). Legume breaks in Stockless Organic Farming Rotations: nitrogen accumulation and influence on the following crops. *Biological Agriculture and Horticulture* **17**, 159-170.
- Smith, S.J. & Sharpley, A.N. (1993). Nitrogen availability from the surface-applied and soil incorporated crop residues. *Agronomy Journal*, **85**, 776-778.
- Stopes, C., Bulson, H., Welsh, J. & Woodward, L. (1996). *Stockless Organic Farming – Research Review 1987-1995*. Elm Farm Research Centre; Hamstead Marshall, U.K.
- Stopes, C., Millington, S., Woodward, L. (1996). Dry matter and nitrogen accumulation by three leguminous green manure species and the yield of a following wheat crop in an organic production system. *Agriculture, Ecosystems and environment*, **57**, 189-196.