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Research article

Soil and vegetable crop response to addition of different levels of municipal waste compost under Mediterranean greenhouse conditions

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Abstract – In the soil thematic strategy of the European Union Commission, a soil organic carbon content of 2% is indicated as a threshold below which a reduction in soil chemical, biological and physical fertility, and increase in erosion can be observed. Composting of organic matter 'exogenous' to soil (such as from municipalities, industries and agriculture sources) is recommended as an effective way to ensure the return of biomass to soil and the return of the soil organic matter losses. The composting of municipal solid wastes is seen as a strategy to divert organic waste materials from landfills. A municipal source-separated solid waste compost was used in a study carried out during 2003-2006 in Southern Italy. An annual tomato-snap bean-lettuce rotation was planted on a sandy loam soil with 26 g kg^{-1} organic carbon under greenhouse conditions. Different rates of compost $(15-30-45 \text{ tha}^{-1} \text{ on a dry weight basis})$ and combinations of compost at a rate of 15 tha⁻¹ with reduced doses of mineral N fertilizer (1/2 or 1/4 of optimal supply) were compared with an untreated control and a N, P, K fertilized control. We found that: (1) increasing compost rates produced increasing positive soil organic carbon balances. The C conversion efficiency was 23 and 36% with 15 and 30 tha⁻¹, respectively, but declined to 28% with the highest rate of compost. Indeed, the higher the compost amounts applied, the higher the soil organic carbon losses. (2) Under tunnel-greenhouse conditions, all the fertilization strategies, except compost at a rate of 15 tha⁻¹, increased soil nitrate concentrations by up 100 to 400 mg kg⁻¹ dry weight of soil, particularly in the spring-summer seasons. In the same period, nitrate contents in the untreated control reached 100 mg kg⁻¹. (3) The average yield of marketable tomato for the four-year period was 114 tha⁻¹ and did not vary significantly among treatments. No differences in snap bean yields were detected among the fertilization treatments. In lettuce cultivation, however, 30 and 45 tha⁻¹ of compost yielded more than other treatments. In the tunnel-greenhouse environment, a high initial content of soil organic matter resulted in high vegetable yields over all four years, even without mineral or organic fertilizer supply. However, among the various fertilization strategies, the best solution able to restore annual soil carbon mineralization was the supply of 15 t ha⁻¹ of compost. In addition, this rate reduced the hazards linked to the high release of nitrates in soil caused by 30 and 45 tha⁻¹ rates of compost or mineral fertilization.

compost amendment / soil C balance / soil nitrates / vegetable crops / greenhouse / soil enzyme activity

1. INTRODUCTION

In Mediterranean areas, 74% of the land is covered by soils with organic carbon content below a threshold of 2% (corresponding to 3.4% organic matter) in the topsoil (0–30 cm) (Van Camp et al., 2004). Levels below this threshold often result in the loss of some soil characteristics, e.g. compactability, friability, water-holding capacity, source and storage of nutrients and energy, stability of aggregates, and infiltration of water in soil. According to Carter (2002), this threshold is soil-and site-specific. Loveland and Webb (2003) highlighted the lack of quantitative evidence in temperate regions for critical thresholds of soil organic carbon (SOC) below which potentially serious decline in soil quality will occur. They pointed

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out the need for identification of appropriate indicators that relate soil quality to soil functions. The composting of organic matter 'exogenous' to soil, such as from municipalities, industries and agriculture sources, is recommended in the soil thematic strategy promoted by the European Union Commission. It is an effective way to ensure that quality biomass is returned to soil, in order to counteract the loss of soil organic matter (Van Camp et al., 2004). The composting of municipal solid waste is seen as a strategy to divert organic waste materials from landfills by creating a product suitable for agricultural purposes at relatively low cost. Its safe use in agriculture depends on its quality, specifically referring to compost maturity, and low metal and salt contents (Hargreaves et al., 2008). An early source separation of municipal food wastes and the exclusion of sewage sludge are the keystones to improving the quality of municipal waste compost and to limiting its metal content (Sequi, 1996). In Italy, high quality compost is defined as a product whose chemical, biological and physical characteristics are in accordance with the thresholds fixed by the Italian decree of law n. 217/2006. Only compost complying with these criteria can be used in agriculture for soil amendment.

In most studies, compost supply has been investigated as mineral fertilizer replacement by assessing the ability to match the nutrient requirement of vegetable crops or by comparing mineral and organic fertilization on the basis of the supplying the same amounts of N, either mineral or total nitrogen. In the first case, high application rates (more than 50-60 t ha⁻¹ on a dry weight basis), often combined with mineral fertilizers, have been tested because only a fraction of compost N content is available to plants during the first year after application (Roe, 1998; Maynard, 2005; Hargreaves et al., 2008). In the second case, the different rates of N availability from a mineral fertilizer or a compost were not commonly taken into account (Zhang et al., 2006; Montemurro et al., 2007). When compost is applied to soil, several aspects should be jointly considered: first, the organic matter content and dynamic (Carter, 2001), and second, compost N content, its mineralization rate and crop N requirement to minimize nitrate leaching to groundwater (Jaber et al., 2005). However, stable benefits of compost use depend on C conversion efficiency, which varies with soil type, climate and cropping system.

Many soil functions such as decomposition of organic residues and their humification, nutrient cycling, stabilization of soil structure, degradation of pollutants, carbon accumulation, and release of CO_2 and other greenhouse gases depend on the metabolic capacity of the soil ecosystem (Van Camp et al., 2004). Several authors agreed that the simultaneous measurement of a set of soil biological parameters is a useful means to assess soil quality (Nannipieri et al., 1990). For the evaluation of data, however, reference systems defining values related to the quality of specific soil functions are needed.

In this article we report the results of a research carried out in a Mediterranean tunnel greenhouse in a four-year trial. We studied the effects of repeated compost amendments in a vegetable crop sequence. Our objectives were: to define, in an intensive cultivation system, the critical level of soil organic carbon below which soil functions and soil productivity decayed; to define the role of different rates of compost in maintaining and/or restoring the soil organic carbon (SOC) pool and the best conversion efficiency of compost C into soil organic C; to monitor the dynamics of nitrate release in soil as related to crop cycles and crop productivity, and to assess the risk of leaching along the profile; to study the changes in soil respiration and two soil enzyme activities as early indicators of soil quality changes.

2. MATERIAL AND METHODS

2.1. Study area and soil characteristics

The experiment was carried out in a Mediterranean area in Pontecagnano (Salerno) (lat. 40° 37' N, long. 14° 52' E), 48 m above sea level. The soil was a Sandy Loam Calcaric Cambisol (FAO, 1998) with 43% sand, 39% silt and 18% clay. Chemical characteristics were: pH 7.9, EC (1:2.5) 0.22 dS m⁻¹, cation exchange capacity 22.7 meq 100 g⁻¹, total and active CaCO₃ 59% and 16%, respectively, total organic carbon 26 g kg⁻¹, total N 0.23%, available P 52 mg kg⁻¹ and exchangeable K, Ca and Mg 528, 3900 and 20 mg kg⁻¹, respectively.

2.2. Treatments and experimental design

The experiment was carried out from 2003 to 2006 on 6×4 m plots in an unheated tunnel greenhouse (860 m² area) covered by Multisolar plastic film (0.18 mm). A completely randomized block design with three replications was laid out in order to compare seven treatments: unfertilized control, three levels of compost amendment at dry weight supply rates of 15, 30 and 45 t ha⁻¹ y⁻¹, mineral NPK fertilization and two treatments with compost at a dry weight rate of 15 tha⁻¹ combined with ^{1/2} or ^{1/4} of the mineral N dose of the NPK treatment.

2.3. Compost source and application

The compost derived from a mixture of source-separated municipal food waste and yard trimmings (1:1 ratio) treated by BIOE[®] Technology, which is useful in composting on small-scale farms due to its simple and cheap implementation. The active composting time was performed in aerated static piles covered by a Goretex film. The thermophylic phase lasted 30 days, while the curing phase lasted 60 days. Automatic monitoring of temperature, oxygen and biomass moisture during composting was provided by a software package. The main analytical characteristics of the final compost were compliant with Italian law (Tab. I), so that it could be applied as an amendment in agriculture. Every year during these trials, at the start of the annual crop rotation (March–April), soil was plowed and the fixed municipal food waste compost amounts were rototilled into the 0–20 cm layer of soil.

2.4. Crop rotation

An annual crop rotation schedule of tomato, snap bean and lettuce was adopted. Each of these crops received NPK fertilization according to its needs every year. Tomato for the fresh market was transplanted onto black plastic mulch between the middle of March and the beginning of April in 4 single rows at a plant density of 2.85 plants m⁻². Cultivars used were HF1 Lido in the first and third years, HF1 Cencara in the second year and HF1 Lacey in the fourth year. Plants were pruned over the eight flower cluster. Bumble bees were used as pollinators. For the mineral fertilization 120, 100 and 200 kg ha⁻¹ of N, P₂O₅ and K₂O, respectively, were supplied. The total N amount and part of K was distributed via fertigation as ammonium nitrate or potassium nitrate. The crop was drip irrigated, receiving an average water volume of 1100 m³ ha⁻¹ y⁻¹

Table I. Average contents of carbon, nitrogen, trace metals and inert material (on a dry matter basis) in the municipal food waste compost used and tolerance limits stated by Italian law (decree with force of law n. 217/2006).

Element	Min–Max	Tolerance limits
Total organic carbon (%)	24.0-33.7	<u>></u> 25
Total N (%)	1.74-2.10	
Organic N (%)	1.04 - 2.00	> 80% total N
C/N	13.3–16.4	<u><</u> 25
pH	7.5-8.5	6-8.5
$Cd (mg kg^{-1})$	0.55-1.36	<u><</u> 1.5
Cr^{6+} (mg kg ⁻¹)	< 0.5	<u><</u> 0.5
$Pb (mg kg^{-1})$	36.5-50.0	<u><</u> 140
Ni (mg kg ⁻¹)	3.9-12.5	<u><</u> 100
$Cu (mg kg^{-1})$	130–148	<u><</u> 230
$Zn (mg kg^{-1})$	256-297	<u><</u> 500
Inert materials (plastic, glass)		
Ø < 3.33 mm (%)	0.38-0.65	<u><</u> 0.45
$3.33 \text{ mm} < \emptyset < 10 \text{ mm} (\%)$	3.02-5.45	<u><</u> 0.05
$\emptyset > 10 \text{ mm} (\%)$	< 0.01	<u><</u> 0
Salmonella	Absent	Absent

of water. All measurements were carried out on plants harvested from the middle of June to the middle of July, on a central area of 12 m^2 of each plot. Marketable fruits were separated into three commercial categories (extra, I and II). An estimation of crop dry matter was made on three plants from the border rows by adding the dry matter of fruits measured during the harvesting phase to the dry matter of stems, leaves and roots measured at the end of the growing season.

Snap bean (cv Xera) was sown in single rows at the end of July on non-mulched soil. Seed density was 40 m⁻², but the actual average density at harvest was 28 plants per m². The crop received an average water volume of 1650 m³ ha⁻¹ y⁻¹. Before seeding, 40 kg ha⁻¹ of nitrogen as ammonium sulfate was applied to the mineral fertilized treatment. The harvest was performed in the first half of October on a central area of 12 m², picking all the plants at one time. Ten plants in the harvest area were sampled in order to estimate crop dry matter.

Lettuce (cv Coralis in the first two years, cv Arcadia in the third one, cv Cantambria in the fourth year) was transplanted in the second half of October, at a plant density of 8 m⁻² on unmulched soil. In the NPK treatment, the mineral fertilization rate was: 100, 60 and 90 kg ha⁻¹ of N, P₂O₅ and K₂O, respectively. The total amount of N was distributed via fertigation as ammonium nitrate while P and K were distributed as mineral perphosphate and potassium sulfate in pellets, respectively. The average water volume of irrigation was 1500 m³ ha⁻¹ y⁻¹. Harvest started in December or January, depending on the year. A maximum of 60 plants were picked from a central harvest area of 7.5 m². The crop dry matter was estimated from ten of the sixty plants after oven drying (60 °C). Nitrate content was measured in dry leaves of five marketable plants by ionic chromatography (Dionex DX 500). No soil sterilization was done during the research. The annual total rates of chemical fertilizer applied in treatments with mineral fertilizers were 260, 160 and 290 kg ha⁻¹ yr⁻¹ for N, P_2O_5 and K_2O , respectively. The total N applied in compost over the four years was 1080, 2160 and 3240 kg ha⁻¹ for C15, C30 and C45, respectively.

2.5. Soil organic C balance and soil nitrate release

All the analytical parameters were monitored by sampling each experimental unit before starting treatment and at the end of the annual crop rotation. The last samples were collected during February 2007. Soil samples were prepared by aggregating five subsamples per plot collected from the arable layer (0-30 cm).

Total organic carbon was determined by the Walkley-Black method. Taking into account the soil bulk densities for each treatment, which ranged from 0.95 to 0.89 tm⁻³ in Cnt and C45, respectively, soil carbon data were expressed as tons per hectare in the 0–30 cm layer. Crop residues were removed at the end of each crop cycle, so that soil carbon dynamics were strictly dependent on the initial C content and on C added in amendments. The efficiency of conversion of C in compost into soil organic C was calculated as the ratio of soil organic carbon change during 2003–2007 to the total input of C compost in the same period.

In order to monitor the magnitude of the soil nitrate release and its distribution during the year, soil samples were collected monthly from May to December in the years 2004– 2006, in all the experimental plots except those with compost rate 15, integrated with 1/4 mineral N. From each plot four soil cores from the 0–30 cm and 30–60 cm layers were sampled by a gouge auger for stepwise sampling (Eijkelkamp) in fixed points along the plant rows. Only the 2003–2006 average contents recorded, for each treatment in May, August, October and December, are presented in this paper.

Soil samples were frozen until they were analyzed; 40 g of soil was extracted in distilled water (1:5 ratio) by shaking for 30 min. Extraction was done with water and not 1 M KCl, in order to simulate the actual concentration of soil solution more accurately. Soil suspensions were centrifuged and filtered, and the nitrate content was measured by anionic chromatography (Dionex DX 500).

2.6. Soil respiration and enzymatic activities

Soil samples for biological activity measurements were collected from 2003 to 2005, three times per year, in spring (May–June), fall (September) and winter (January) from the 0-20 cm layer, along the plant row, where the soil microbial community is most strongly influenced by crop root systems. Five subsamples were collected from each plot and mixed to obtain a homogeneous sample. The soil was sieved through a 2 mm screen and stored at 4 °C for a few days. For soil respiration assessment, 3 g samples of soil were placed in 35 mL glass vials, sealed and incubated at 25 °C in darkness. After

48 hours, CO₂ concentration was measured in the headspace by gas chromatography. Acid phosphatase activity was assayed by measuring the hydrolysis rate of p-nitrophenyl phosphate (disodium salt) (0.115 M) as a substrate. The amount of p-nitrophenol released after 60 min of incubation at 37 °C in darkness was spectrophotometrically measured by detecting absorbance at 398 nm (Garcia et al., 1993).

The fluorescein diacetate hydrolysis (hydrolase) was assayed using fluorescein diacetate (3, 6 diacetylfluorescein) as a substrate and measuring the absorbance of released fluorescein at 490 nm (Schnürer and Rosswall, 1982).

2.7. Statistical analysis

In the experiment we performed repeated measurements on the same plots in different years. Data on yields and soil biological activities were analyzed with a mixed model in which fertilization treatments, years and their interaction were considered as fixed effects, whereas replication and plot were considered as random effects as well as times of sampling for biological activities (SSC, 2001). Mean values of yield data were separated by orthogonal contrasts.

Final soil organic carbon and its changes during the studied period were analyzed by one-way analysis of variance with regard to the factor fertilization. Mean values of soil organic carbon were compared with the confidence interval for P = 0.05.

Nitrate data were transformed logarithmically due to the skewness of their distribution (Webster, 2001). After ANOVA, means were re-scaled to natural numbers. Mean values were compared with the confidence interval P = 0.05.

3. RESULTS AND DISCUSSION

3.1. Dynamics of soil organic carbon

After four years of repeated mineral and organic fertilization treatments the soil organic carbon contents detected in the 45 t compost and 30 t compost treatments were higher than in all the other treatments (Fig. 1).

The two 15 t compost with N_{min} treatments showed higher but not significantly different C contents than 15 t compost alone, but this is related to the higher initial SOC content rather than to the effect of the compost added. Indeed, the soil organic carbon change, expressed in each treatment as the difference between the contents at the end and at the beginning of the experiment, showed that in 15 t compost, integrated with 1/2 or 1/4 of mineral N treatments, there was a decrease of 1.6 g kg⁻¹ and 1.7 g kg⁻¹, respectively; whereas in 15 t compost alone there was an increase of 1.3 g kg⁻¹. In addition, the 30 t and 45 t compost treatments showed a net increase of 5.0 g kg⁻¹ and 5.7 g kg⁻¹ soil organic carbon, respectively. NPK and the unfertilized control showed a net decrease of 4.5 g kg⁻¹ and



Figure 1. Soil organic carbon contents (SOC) at the end of the 2003–2006 period, as influenced by the repeated fertilization treatments. Vertical bars represent confidence interval of means at P = 0.05. Control = unfertilized control; NPK = mineral fertilization; 15 t, 30 t, 45 t comp = compost distributed at rates of 15, 30 and 45 t/ha, respectively; 15 t comp and 1/2 N = rate 15 t of compost integrated with 1/2 mineral nitrogen; 15 t comp and 1/4 N = rate 15 t of compost integrated with 1/4 mineral nitrogen.

2.1 g kg⁻¹ soil organic carbon, respectively. Mineral fertilization doubled the mineralization of the organic carbon.

The estimation of the efficiency of compost C conversion into soil organic C is shown in Table II.

The increasing compost application rates were related to decreasing marginal SOC gains, leading to higher SOC losses. In particular, the 45 t compost caused, after 4 years, the largest C losses (38 tha⁻¹), whereas the best conversion efficiency was found for C30. These data indicate that, as C input increases, the efficiency of conversion into stable SOC declines, as reported by Horwath et al. (2002) and Chang et al. (2007). Chang et al. (2007), in subtropical climatic conditions (Taiwan), in a greenhouse study, applied 8-16.5-25-33 t ha⁻¹ (dry weight) of compost annually and reported that the percentages of applied organic matter retained in soil were 21, 19, 11 and 12%, respectively. Campbell et al. (2002) summarized C conversion efficiency values from various long- and medium-term studies carried out in Canada. The authors found that the conversion efficiency increased with the number of crops per year, fertilization, manuring and no-tillage. Similar conclusions were reported by Giardini (2004), who summarized the results of long-term experiments (40 years) carried out in Northern Italy.

The combination of 15 t compost with reduced amounts of mineral N produced a negative carbon balance. According to Mamo et al. (1999) and Horwath et al. (2002), the addition of N fertilizer enhances compost mineralization. It has been suggested that added N is first incorporated into the soil microbial biomass, thus stimulating mineralization of the organic matter applied. In the open field, this strategy produced acceptable results, i.e. adequate yields, higher C stock in soil and minimal risks of NO₃-N leaching (Mamo et al., 1999; Cai and Qin, 2006). However, the greenhouse environment could have allowed for continuous microbial activity through the whole year, decreasing C sequestration while stimulating C losses.

Table II. Conversion efficiency of compost C into soil organic C (SOC) in the 0–30 cm layer after 4 years of compost application (2003–2006). Control = unfertilized control; NPK = mineral fertilization; 15 t, 30 t, 45 t comp = compost distributed at rates of 15, 30 and 45 t/ha, respectively; 15 t comp and 1/2 N = rate 15 t of compost integrated with 1/2 mineral nitrogen; 15 t comp and 1/4 N = rate 15 t of compost integrated with 1/4 mineral nitrogen.

Treatment	Compost C input (t ha ⁻¹)	SOC changes* (t ha ⁻¹)	SOC losses** (t ha ⁻¹)	Conversion efficiency*** (%)
Control	0	-5.5 (8.5)		
15 t comp	18	4.1 (6.4)	-13.6	23 (3.6)
30 t comp	36	13.2 (1.3)	-22.3	36 (3.7)
45 t comp	53	15.3 (1.7)	-38	28 (3.2)
NPK	0	-11.7 (9.3)		
15 t comp and 1/2 N	18	-4.8 (4.9)	-22.5	-27 (2.7)
15 t comp and 1/4 N	18	-3.1 (1.7)	-20.9	-17 (9.4)

* SOC 2007 minus SOC 2003; ** SOC 2007 minus (SOC 2003 + compost C input); *** SOC change divided by compost C input. In brackets is the standard error of mean.

Table III. Average effect of the fertilization treatments on tomato marketable yield and plant dry matter in the 2003–2006 period. Control = unfertilized control; NPK = mineral fertilization; 15 t, 30 t, 45 t comp = compost distributed at rates of 15, 30 and 45 t/ha, respectively; 15 t comp and 1/2 N = rate of 15 t of compost integrated with 1/2 mineral nitrogen; 15 t comp and 1/4 N = rate of 15 t of compost integrated with 1/2 mineral nitrogen; 15 t comp and 1/4 N = rate of 15 t of compost integrated with 1/2 mineral nitrogen.

Marketable yields					Fruit	Plant	
Treatments	total	extra	I cat.	II cat.	mean weight	dry matter	
	$(t ha^{-1})$				(g)	$(t ha^{-1})$	
Control	109.4	67.7	23.3	18.2	148	10.8	
15 t comp	111.0	73.1	24.0	13.8	151	10.2	
30 t comp	119.4	81.5	24.6	13.3	154	10.4	
45 t comp	116.6	79.3	23.6	13.5	154	_	
NPK	112.6	74.0	24.0	14.8	149	12.5	
15 t comp and 1/2 N	115.5	75.3	26.4	13.9	149	11.1	
15 t comp and 1/4 N	110.7	72.4	23.7	14.7	149	_	
Orthogonal contrasts and	probability						
Fertilization vs. Cnt	n.s.	n.s.	n.s.	0.001	n.s.	n.s.	

n.s.: not significant.

3.2. Crop yields

The planting of three vegetable crops per year was aimed at testing the ability of soil to support plant growth and yield under greenhouse conditions. With regard to the best compost supply scheduling, we chose the annual addition proposed by Mamo et al. (1999) and Hartl et al. (2003).

Tomato yield in the 2003–2006 period was 109 t ha⁻¹ in the unfertilized control (Tab. III). Although all fertilization treatments increased yields, these differences were not significant. When total yield was divided into commercial categories, the unfertilized control yielded lower amounts of extra fruits and higher amounts of category II fruits than the fertilization treatments. Total crop dry matter did not vary among different treatments.

Snap bean marketable yields and crop dry matter did not differ between the control and the fertilization treatments (data not shown). The yields ranged from 6.3 t ha⁻¹ for the unfertilized control to 7.9 t ha⁻¹ for 15 t compost combined with 1/4 mineral N. Crop dry matter ranged from 3.5 t ha⁻¹ for the unfertilized control to 4 t ha⁻¹ for 15 t compost integrated with 1/2 mineral N.

In unfertilized control plots, the level of fertility during the four-year period was sufficient to match the nutrient demand of tomato and snap bean. These results can be explained by taking into account that high native soil fertility hid a clear 'fertilization rate - yield response' link, as reported by Gallardo-Lara and Nogales (1987), who stated that yield response to compost is generally greater in soils with low fertility. Ozores-Hampton et al. (1998), in experiments carried out in Florida, reported no response of tomato to mature municipal solid waste compost rates of 0-145-224 tha⁻¹, or of snap bean to rates of 0-89 –134 tha⁻¹. Besides, crops with a low nutrient demand generally do not respond sensitively to mineral and/or compost inputs, as reported by Reider et al. (2000), in Pennsylvania, for pepper and Maynard (2005), in Connecticut, for carrot.

The last crop of the rotation was lettuce, which showed a different growth and yield behavior (Tab. IV). The average marketable yield, as well as the number of harvested plants and their mean weight, for all the fertilization treatments was significantly higher than the unfertilized control (+ 31%). No differences were found between the compost rates and Min treatment, except for the number of harvested plants. The higher rates of compost (30 t and 45 t) favored greater plant

Table IV. Effect of the fertilization treatments on lettuce yield, dry matter and nitrate content in leaves. Means of the 2003–2006 period. Control = unfertilized control; NPK = mineral fertilization; 15 t, 30 t, 45 t comp = compost applied at rates of 15, 30 and 45 t/ha; 15 t comp and 1/2 N = rate of 15 t of compost integrated with 1/2 mineral nitrogen; 15 t comp and 1/4 N = rate of 15 t of compost integrated with 1/4 mineral nitrogen.

Treatments	Marketable	Harvested	Mean weight	Crop	Nitrates	
	yields	plants	of plant	dry matter		
	$(t ha^{-1})$	$(N \times 0.000)$	(g)	$(t ha^{-1})$	(g kg ⁻¹ d.m.)	(mg kg ⁻¹ f.m.)
Control	16.7	59	271	0.83	19.2	1073
15 t comp	20.2	70	289	1.05	24.8	1161
30 t comp	24.7	77	324	1.11	30.8	1316
45 t comp	24.7	76	323	1.14	33.9	1559
NPK	20.6	65	310	0.91	34.8	1571
15 t comp and 1/2 N	21.2	68	303	0.94	35.1	1536
15 t comp and 1/4 N	20.6	68	303	0.87	35.4	1601
Orthogonal contrasts and probability						
Fertilization vs. control	< 0.0001	< 0.0001	0.001	0.002	< 0.0001	0.0007
Compost rates vs. NPK	n.s.	0.05	n.s.	0.06	n.s.	n.s.
15 t comp vs. (30 t, 45 t)	0.0005	0.04	0.008	n.s.	0.0008	0.02
30 t comp vs. 45 t comp	n.s.	n.s.	n.s.	n.s.	n.s.	0.08
15 t comp vs. $(15 t + N_{min})$	n.s.	n.s.	n.s.	0.06	< 0.0001	0.001
15 t comp and 1/2 N vs. 15 t comp and 1/4 N $$	n.s.	n.s.	n.s.	n.s.	n.s.	n.s.

production in comparison with 15 t compost integrated with 1/4 or 1/2 mineral N. These last did not increase yields compared with 15 t compost alone. Crop dry matter in fertilizer treatments was higher than in the unfertilized control, but the input of mineral nitrogen in mineral fertilization and 15 t compost integrated with 1/4 or 1/2 mineral N seemed to decrease crop dry matter in comparison with 15 t, 30 t and 45 t of compost, suggesting high water absorption induced by mineral N supply.

Table IV also shows some qualitative characteristics of lettuce. Nitrate content was lower in the unfertilized control. The reduced uptake of water and nitrate was consistent with the observed growth reduction of plants in the unfertilized control. However, average nitrate concentration in leaves was very low (1265 mg kg⁻¹ of fresh matter) with respect to the threshold of 4500 mg kg⁻¹ for fresh matter as stated by EU rules. We observed that nitrate increased either with increasing compost rate, or with mineral nitrogen added in mineral fertilization, or in combinations with 15 t compost.

Lettuce performance is linked to the influence of fall and winter on the reduction of nitrate release in soil, due to the lowering of soil temperature and of nitrifying bacteria activities (Fig. 2). In these seasons only, the soil nitrate contents in the unfertilized control decreased to values (30 and 20 mg kg⁻¹ in October and December, respectively) critical for matching crop nutrient needs, such that the differences in nitrate contents between fertilized and unfertilized plots were sufficient to determine significant increases in yields of fertilized treatments.

3.3. Soil nitrate release

Figure 2 shows the interaction of fertilization and time of sampling at 0–30 cm soil depth.

In May and August nitrate release ranged from 300 to 450 mg kg⁻¹ d.w., in plots treated with compost rates of 30 t and 45 t, as well as with 15 t compost integrated with 1/2 N and NPK fertilization. The lowest contents were detected in 15 t comp and Cnt and ranged from 130 to 190 mg kg⁻¹ d.w. of nitrate. In October and December, nitrate concentrations fell to low levels. However, in October, nitrate concentrations in compost-amended plots were 2-5 fold higher than in the unfertilized control, while in December mineral fertilization and 15 t compost integrated with 1/2 N exhibited nitrate concentrations 4-5 fold higher than the unfertilized control. At 30–60 cm soil depth, the interaction of fertilization \times time was not significant (data not shown). Nitrate contents were generally lower than in topsoil, but followed the same trend, e.g. in 45 t compost they ranged from 250 to 45 mg kg⁻¹ d.w. from August to December. The effect of the greenhouse environment has to be considered for the interpretation of data. For instance, Pepin et al. (2007) found nitrate contents ranging from 400 to 1200 mg kg⁻¹ in soils rich in organic matter, organically cropped under greenhouses of Canada.

Taking into account the crop yield data, the measured nitrate levels in unfertilized control soils (over 100 mg kg⁻¹ until August) were sufficient to meet the tomato and snap bean nitrogen demand (Hartz, 2006). On the contrary, during the lettuce cycle, nitrate contents in unfertilized control soil dropped to low values (30 and 20 mg kg⁻¹ in October and December), consistent with the observed reduced crop growth and production.

As a consequence, organic, mineral or combined fertilization only increased soil nitrate content above 100 mg kg⁻¹ soil d.w., but this level of nitrate availability did not result in higher yields and may be leached to groundwater. Only the rate of 15 t compost showed nitrate release close to that of the unfertilized control. The concentrations of soil nitrate detected at 30–60 cm may increase the risk of ground nitrate export,



Figure 2. Soil nitrate contents (mg kg⁻¹ d.w.) at 0–30 cm depth. The fertilization–time of sampling interaction was highly significant ($P \le 0.001$). Vertical bars represent confidence intervals of means at P = 0.05. Control = unfertilized control; NPK = mineral fertilization; 15 t, 30 t, 45 t comp = compost distributed at rates of 15, 30 and 45 t/ha; 15 t comp + 1/2 N = rate of 15 t of compost integrated with 1/2 mineral nitrogen.

which in a greenhouse is mainly dependent on irrigation volume. In the USA, Jaber et al. (2005) pointed out the scarcity of studies investigating the fate of N from organic amendments in soil and groundwater. They found that in sandy and calcareous soils of Florida the use of a municipal solid wastebiosolid compost as fertilizer (83 tha⁻¹ of compost, equivalent to 984 kg ha⁻¹ of N) did not increase groundwater N concentrations compared with standard inorganic fertilizer.

3.4. Soil respiration and enzymatic activities

Figure 3 shows the trends of soil respiration and enzymatic activities measured in the 2003–2005 period.

Soil respiration increased with compost application, consistently with the supplied rate. Soil respiration was not affected by mineral fertilization. The seasonal variation in soil temperature, which directly affects soil microbial activity, and the availability of labile organic compounds, which was higher after compost distribution and then decreased during the year, explain either the largest differences to unfertilized control and mineral fertilization found in spring-summer , or the lowest ones during fall-winter. An increase in soil respiration after organic amendments was found by several authors and indicates an increase in microbial activity (Hargreaves et al., 2008), which is responsible for nutrient release from the organic matter added to soil. In our study, the strong increase in soil respiration was consistent with increasing consumption of organic C added with rates of compost from 15 to 45 tha⁻¹ (see Tab. II).

Fluorescein diacetate hydrolysis (FDA) was directly affected by compost application and ranged from 90.6 to $141 \,\mu$ g FDA g⁻¹ d.w. 2h⁻¹. The activity also showed seasonal fluctuations, with the lowest values in winter. The magnitude of the activity increased with compost rate and with cumulative compost amendment, leading to the amplification of differences between compost-treated and untreated soils (unfertilized control and mineral fertilization). Nayak et al. (2007) suggested FDA hydrolysis as a sensitive indicator of soil microbial activity changes following long-term addition of compost.

Soil acid phosphatase activity was also increased by compost amendments. Unlike FDA hydrolysis, the increase in this enzymatic activity during the first year of compost addition was maintained at high levels and was not influenced by season. Phosphatase cycle is mainly performed by extracellular



Figure 3. Trend of soil respiration and two enzymatic activities measured from 2003 to 2005 in fertilized and unfertilized soils.

enzymes, protected against soil proteases by binding to soil organic matter and/or clay minerals, and only a small amount of the activity is attributed to proliferating microbial cells. It is, therefore, plausible to attribute the observed trend of phosphatase activity to organic matter increase.

Soil respiration, fluorescein diacetate hydrolysis and acid phosphatase were positively affected by compost supply, demonstrating shifts in microbial performances related to carbon, nitrogen and phosphorus cycles in soil (Iovieno et al., 2009). Mineral fertilization did not appear to influence these activities. Similarly, Chang et al. (2007) and Chu et al. (2007) found increases after three and 12 years, respectively, in dehydrogenase, phosphatase, β -glucosidase, arylsulfatase, protease and urease activities related to compost amendment repeated annually.

Yang et al. (2008) found a positive correlation (P < 0.01) between enzyme activities and cucumber yield, concluding that soil enzyme activities are indicative of soil fertility. In the soil tested here the relationship between compost amendment and soil enzyme activity and crop productivity was not clear. In fact, the increase in soil biological activities induced by the treatments did not correspond to an increase in crop yield, suggesting that baseline levels of enzyme activities observed in the unfertilized control do not represent a threshold below which soil functions are impaired.

4. CONCLUSION

We studied different soil fertility and plant nutrition management options under tunnels over a four-year period. Our experiment started in a soil with a high organic carbon content (2.6%), which should represent the final goal, rather than the beginning. This needs to be taken into consideration when analyzing our data. At the end of the experiment, the lowest soil organic C contents were approximately 2.4% in the unfertilized control and mineral fertilization treatments. At this level, we did not observe a decline in crop production and soil fertility. On the other hand, the increasing supply of municipal waste compost of 15, 30 and 45 t ha⁻¹ on a dry matter basis resulted in an increase in soil organic carbon, but the efficiency of conversion of C compost into soil C showed a parabolic trend. A rate of 15 t ha⁻¹ appeared to be the best for replacing the amounts of organic matter mineralized annually.

The supply of increasing amounts of compost under tunnel conditions activated soil respiration, hydrolase and acid phosphatase activities. The microclimate in tunnels, i.e. high soil and air temperature, and constant soil humidity, permitted almost continuous soil microbiological activity of mineralization and humification of organic matter. As a consequence, the flux of nitrates released in soil due to organic matter mineralization increased to 400 mg kg⁻¹ dry weight of soil, particularly in the spring-summer seasons. These high concentrations were recorded with the supply of N mineral fertilizers as well as with 30 and 45 tha⁻¹ of compost. In addition, the high contents of nitrate exceeded the needs of tomato or snap bean without significant yield increases.

In conclusion, the rate of 15 t of compost appeared the most adequate to maintain the level of soil organic C, sustain biological activity, minimize the risks of nitrate leaching to groundwater and to guarantee vegetable crop productivity, compared with the mineral fertilized control. The rate of 30 t of compost appeared to increase soil organic C, and showed the highest efficiency of conversion of C compost, but under tunnels the high soil microbial activity caused an exceedingly high and uncontrollable nitrate release. This fertilization management option should thus be tested in open fields to verify its ability to sustain an intensive crop system and to stock C in soil.

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