

# Effect of different management strategies on soil quality of citrus orchards in Southern Italy

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#### Abstract

Twenty-six soil samples were collected from 13 paired orchards (organically vs. conventionally managed) homogeneous for age, rootstock and cultivars, belonging to the Eastern Sicily Organic Citrus farm Network. The soil quality was evaluated by chemical and biochemical indicators. The total organic C, humification parameters and isoelectric focusing of extracted organic matter were measured to quantify the size of relevant soil C pools. In addition, C turnover was evaluated by determining microbial C mineralization, C microbial biomass and by calculating the mineralization and metabolic quotient (qCO<sub>2</sub>). The results obtained demonstrated that organic citrus soils were characterized by a general increase in all the organic matter pools, which means a greater C supply for soil metabolic processes. This observed trend did not directly influence the organic matter turnover, indicating that the organic approach could act as a soil C-sink. The soil microflora of organically managed soils showed an improved efficiency in use of energy and organic resources, corresponding to an increased ability of soils under organic management to sustain biological productivity in the long term.

Keywords: Soil quality, organic farming, citrus, qCO<sub>2</sub>, C mineralization, isoelectric focusing

# Introduction

In Italy the total land area organically managed or in conversion is 1,067,000 ha, which is about 25% of the European organically managed area, with forage, grassland and cereals representing 70% of the total, followed by olives, vines, citrus and other fruit (SINAB, 2007).

In 2006, the total area in citrus production was 170,000 ha (ISTAT, 2006). Citrus is grown in Southern Italy (Calabria and Sicily) which has a typically Mediterranean climate. The area of organic citrus (18,000 ha) represents 10% of the total citrus area and 2% of the total area of organic crops in Italy (SINAB, 2007).

Due to the presence of the organic citrus sector in Southern Italy and the peculiar characteristics of the Mediterranean environment, evaluation of the effects of organic management on soil quality is of interest. Agricultural practices can strongly affect the soil environment and are thus likely to modify soil processes in the long term (Paustian *et al.*, 2002).

Correspondence: S. Canali. E-mail: stefano.canali@entecra.it Received June 2008; accepted after revision November 2008 Studies on soil processes and their interactions are used to evaluate soil quality and can contribute to the definition of agro-ecosystem sustainability (Nannipieri *et al.*, 1990; Wander *et al.*, 1994; Watson *et al.*, 2002; Karlen *et al.*, 2003).

One of the aims of organic farming is the protection of environmental quality. It enhances beneficial biological interactions and processes (Reganold et al., 1993) and has positive effects on physical, biological and microbial properties of soils (Raupp et al., 2006). However, it must be noted that energy efficiency and nutrient conservation in organic systems are usually only reached after a long period of time, as demonstrated in long-term field experiments. No different or even unfavourable results, such as the decline of soil N availability and crop production, were obtained in the short term (USDA, 1980; Stark et al., 2007). Hepperly et al. (2006) verified that, after the 26th season of a long-term trial, the comparison of organic and conventional maize and soybean cropping systems showed a significant increase in C and N in soils under organic management, a higher crop yield and protein concentration, together with an increase in soil biomass. Such observations were confirmed by the longterm DOK experiment (comparison among bio-Dynamic, bio-Organic and Conventional 7-year rotation systems in Switzerland, from 1978), which recorded an increase in soil C, soil microbial biomass C and soil microbial biomass activity in biodynamic systems (Mäder *et al.*, 2002, 2006). Also in a 1986–2004 trial, Schjønning *et al.* (2004) reported significant effects of organic management on some soil biological (microbial biomass), chemical (total and hot-water extractable organic C) and physical (bulk soil strength and friability) parameters.

Since 2001, the MASCOT (Mediterranean Arable Systems Comparison Trial) experiment has been ongoing in Italy. This is the first long-term Mediterranean trial comparing conventional and organic management systems for arable agro-ecosystems of the Tuscany costal plain (Barberi & Mazzoncini, 2006). Results have shown the influence on crop yields of organic/conventional management but not the effects on soil quality.

There are limited soil quality data available for organically managed orchards, and most of them are not from semiarid regions (Reganold *et al.*, 2001). Probst *et al.* (2008) evaluated the effect of organic versus conventional management on soil microbial biomass and activity indices in vineyards. In Brazil, Franca *et al.* (2007) demonstrated that the soil of organically managed citrus orchards showed higher microbial activity and arbuscular mycorrizal fungal richness and diversity than that under conventional management.

Initial results of the introduction of organic farming on soil quality of organically managed citrus orchards in the Mediterranean region were reported by Intrigliolo *et al.* (2000). They reported that organic management induced only slight differences in the main physical and chemical characteristics of conventionally managed soil.

The aim of the present study was to compare the soil quality of 13 pairs of conventionally managed and organically-

 Table 1 Description of the main characteristics and practices in the 13 organic and 13 conventional citrus orchards in the survey. Yields are reported as fresh weight

managed citrus orchards converted at least 6 years earlier. Chemical and biochemical parameters were used as indicators of soil quality (Park & Seaton, 1996; Schloter *et al.*, 2005).

The same approach was recently used by van Diepeningen et al. (2006) in their study of the effects of organic versus conventional management on soil properties in Dutch commercial and experimental farms.

# Methods

#### Experimental site and design

The study was carried out in the Catania province, Eastern Sicily, Italy. The average rainfall and temperature of the area are 350 mm and 18.7  $^{\circ}$ C. The climate is classified as typical Mediterranean.

Soil samples (26) were collected from 13 pairs of orchards. Organic orchards were chosen only if they guaranteed the strict application of E.E.C. Regulation 91/2092 for at least 6 years at the moment of soil sampling (EEC, 1991). In accordance with the results of Karlen *et al.* (2001), conventional and organic citrus orchards were chosen on the basis of the homogeneity of inherent soil characteristics. The latter being those determined by parent material, climate and vegetation and which are meaningful in determining the capacity of a soil for a specific land use. A preliminary statistical analysis on soil inherent characteristics of the 13 pairs of orchards was carried out to verify the absence of significant differences.

## Orchard characteristics

A comprehensive description of the orchards and the agricultural practices adopted during the survey are provided in Table 1. The cover crops were communities of natural weeds,

Item	Organic		Conventional	
	Min	Max	Min	Max
Orchard area (ha)	0.40	6.00	0.17	27.00
Orchard age (years)	5	49	8	70
Number of trees (/ha)	400	625	334	625
Yield (t/ha)	10	20	8	35
C input from off-farm fertilizers (kg/ha)	423	1235	_	_
Nutrient input from off-farm fertilizers (kg/ha)				
Ν	47	190	29	300
$P_2O_5$	16	118	20	219
K <sub>2</sub> O	13	156	26	250
Tillage (no. operations)	1	8	0	4
Pruning material recycling (no. orchards)	11		8	
Cover cropping (no. orchards)	13		9	
Weed control	Mowing	Herbicides		

retained all the year and disturbed and partially ploughed only when fertilizers were incorporated into the soil.

Soil amendments (composts) and organic fertilizers (poultry manure, dairy manure, plant residues, etc.), allowed by Italian and European legislation, were applied on the organic farms. Mineral fertilizers were applied on conventional farms. Data shown are based on the information gathered from the owners of the citrus orchards and on estimates of the content of organic C and nutrients of input materials applied as fertilizers. In both organic and conventional farms the irrigation water was distributed by micro-sprinklers, at annual rates from 300 to 650 mm.

## Soils

The orchard soils located in the area surrounding the Etna volcano are Typic Xerorthents, Andic Xerochrepts and Ultic Haploxeralfs (Dazzi, 2005). Soil samples were taken 11-12 months after the last fertilizer application to minimize the effects of fresh nutrients on the soil properties to be measured. Representative soil samples from each of the 26 orchards were collected at 25 cm depth, according to a sampling procedure adapted to the size and shape of each orchard. The samples were then air-dried, crushed and passed through a 2 mm sieve (USDA-NRCS, 1996), and stored for subsequent analyses. Before carrying out biochemical assays, soil samples were rewetted and conditioned according to the procedures required for the methods used (see references under heading Biochemical determinations). Laboratory tests were carried out on three replicates in order to control intralaboratory variability and/or exclude non-systematic errors.

## Chemical determinations

Total organic carbon and humification parameters. Total organic carbon ( $C_{org}$ ) was determined by treatment with 2 N K<sub>2</sub>Cr<sub>2</sub>O<sub>7</sub> and 96% H<sub>2</sub>SO<sub>4</sub> solutions at 160 °C for 10 min according to Springer & Klee (1954).

Humification rate (HR) and the degree of humification (DH) were calculated according to Ciavatta *et al.* (1990) as follows:

$$HR(\%) = 100 \times C_{HA+FA}/C_{org} \quad DH(\%) = 100 \times C_{HA+FA}/C_{extr}.$$

where  $C_{HA+FA}$  is the C content of humic and fulvic fraction.

For determining total extractable organic carbon ( $C_{extr}$ ), 5 g of soil was extracted with 100 mL of 0.1 N NaOH/0.1 N Na<sub>4</sub>P<sub>2</sub>O<sub>7</sub> solution at 65 °C for 48 h, under N<sub>2</sub> atmosphere. C content was determined using 10 mL of the NaOH/Na<sub>4</sub>P<sub>2</sub>O<sub>7</sub> extracts.

The humic acids (HA) were precipitated from 25 mL of the extract by adding a solution of 50%  $H_2SO_4$ , drop by drop until pH < 2. After 10 min, the extract was centrifuged at 1048 g per 10 min and non-humified fraction was removed from the solution containing the fulvic acids (FA) by purification on a polyvinylpyrrolidone column (PVP) (Ciavatta *et al.*, 1990). The purified FA fraction, resolubilized after elution of the PVP column with 0.1 N NaOH, was added to the HA fraction, quantitatively transferred to a calibrated 25 mL flask, brought to volume with 0.5 N NaOH and stored under N<sub>2</sub> at 4 °C. Humic and fulvic acid carbon ( $C_{HA+FA}$ ) determination was carried out as above on 10 mL of 0.5 N NaOH solutions (Ciavatta & Govi, 1993).

*Isoelectric focusing of humic substances.* Humic substances to be analysed by isoelectric focusing (IEF) were obtained as described above. Ten millilitres of the solution were dialysed in 6000–8000 Dalton membranes (Spectra/Por<sup>®</sup>), then freeze dried and subjected to IEF.

The IEF separation was carried out in a Multiphore II, LKB electrophoretic cell, as described in Govi et al. (1994). About 10 mg of lyophilized HA, solubilized in 250  $\mu$ L of twice distilled water, was electrofocused in a pH range 3.5-8.0, on a 5.06% T (Temed) and 3.33% C (w/w N,N'methylenbis-acrilamide) polyacrylamide slab gel, using a mixture of carrier ampholytes (Pharmacia Biotech) constituted by 50% Ampholine pH 3.5-5.0, 25% Ampholine pH 5.0-7.0 and 25% Ampholine pH 6.0-8.0. A prerun (2 h; 1200 V; 1 °C) was performed and the pH gradient formed in the slab was checked by a specific surface electrode. The electrophoretic run (2 h 30 min; 1200 V; 1 °C) was carried out loading the concentrated HA (1 mg C/50  $\mu$ L sample). The bands obtained were stained with an aqueous solution of Basic Blue 3 (30%) and scanned by an Ultrascan-XL Densitometer. The typical IEF profile was obtained for each investigated soil. IEF peaks were then numbered, and the peak area was determined for each soil IEF profile, assuming as 100% the area under the entire IEF profiles. The sum of the areas of peaks focused at pH > 4.7 (corresponding to more humified organic matter) was calculated and expressed as  $A_{\rm s}\%$ .

#### Biochemical determinations

Soil C mineralization. Soil C mineralization was studied by measuring  $CO_2-C$  production in closed jars (Isermeyer, 1952). Each of the soil samples (25 g, oven dry-weight equivalent) were rewetted to their -33 kPa water tension and incubated at 30 °C. The  $CO_2$  released was determined at 1st ( $C_1$ ), 2nd, 4th, 7th, 10th, 14th, 17th and 21st ( $C_{21}$ ) day of the incubation period [mg( $CO_2-C$ )/kg<sub>soil</sub>/day]. The value of  $CO_2-C$  released [mg( $CO_2-C$ )/kg<sub>soil</sub>/day] on the 21st day of incubation was assumed as soil basal respiration ( $C_{basal}$ ). Cumulative  $CO_2-C$ released after 21 days ( $C_{21cum}$ ) was calculated for each soil.

*Microbial biomass C*. The carbon of the soil microbial biomass ( $C_{mic}$ ) was measured [mg C/kg<sub>soil</sub>] by the chloroform fumigation-extraction method of Vance *et al.* (1987) on airdried soils conditioned by an incubation for 21 days in open glass jars at -33 kPa water tension and 30 °C.

Metabolic quotient, mineralization quotient,  $C_{mic}/C_{org}$ ratio. The metabolic quotient q(CO<sub>2</sub>), defined as specific soil respiration of the microbial biomass, was calculated from basal respiration values (after the 21st day) by:

$$q(CO_2) = [(mg CO_2 - C/mg C_{mic}/h]]$$

(Anderson & Domsch, 1985a,b). The mineralization quotient  $C_{MIN}$ , defined as mineralized soil C at steady-state conditions (soil microflora basal respiration) with respect to the total amount of soil organic C, was calculated from soil basal C ( $C_{basal}$ ) by:

$$C_{MIN} = [(mg CO_2 - C/kg_{soil})/mg C_{org}/day]$$

(Dommergues, 1960). The ratio  $[C_{mic}/C_{org}]$ % was used as an index of microbial biomass contribution to soil organic C (Anderson & Domsch, 1989).

#### Graphical representation and statistical analyses

Box plots show the distribution of measured soil parameters. A statistical test for correlated patterns was used to compare differences between organic and conventional soils. Because the distribution of measured parameters did not meet the required conditions for the application of parametric statistical methods (i.e. ANOVA), the Wilcoxon signed rank test, a nonparametric test for paired data, was used (Soliani, 2004).

# Results

Figure 1 shows the box plots describing the frequency distribution of the main soil physical and chemical properties of the selected pairs of orchards. No significant differences were found for clay, sand, pH and cation exchange capacity (CEC), demonstrating the homogeneity of the inherent properties of each pair of soils.

#### Chemical determinations

The  $C_{org}$  values of soils under organic management were significantly larger than those of the conventionally managed orchards (Figure 2a). Both the extractable C in an alkaline environment ( $C_{extr}$ ) and the humified C ( $C_{HA+FA}$ ) were also larger in the organic systems (Figure 2b,c).

Four pairs of representative IEF profiles were chosen from the 13 pairs studied (Figure 3). These profiles showed



Figure 1 Frequency distribution of clay (a), sand (b), pH (c) and CEC (d) of the 13 pairs of orchard soils (organic vs. conventional). n.s., distributions not different according to the Wilcoxon signed rank test. Bars (or whiskers) represent the dispersion of the values below or above the lower quartile and the upper quartile, respectively. In (b) and (c) whiskers are coincident with upper/lower quartile.



**Figure 2** Frequency distribution of total organic carbon soil ( $C_{org}$ ) (a), extractable carbon ( $C_{extr}$ ) (b), humified carbon ( $C_{HA+FA}$ ) (c) and  $A_s$  (relative area of IEF peaks focused at pH > 4.5) (d) of the 13 pairs of orchard soils (organic vs. conventional). *P*-value, significance value of the difference between the distributions according to the Wilcoxon signed rank test. Bars (or whiskers) represent the dispersion of the values below or above the lower quartile and the upper quartile, respectively.

different patterns between organic and conventional soils in the 4.7–6.0 pH range. In particular, the IEF profiles of organically managed soils (10, 20, 30 and 40) were characterized by the appearance and/or the increase in some of the peaks focused at pH > 4.7, corresponding to more humified organic material.

These differences were quantified by calculating the  $A_s\%$  for each soil (Figure 2d). The  $A_s\%$  values of organically managed soils were significantly higher than those of conventionally managed ones.

On the other hand, the humification rate (HR%) and humification degree (DH%) were not significantly influenced by organic management (results not reported).

#### Biochemical parameters

Box plots for soil microbial biomass ( $C_{mic}$ ) values are reported in Figure 4a. The frequency distribution of the 13 pairs of soils showed significantly higher values in the organically managed soils.

Both microbial biomass and soil respiration are positively correlated with soil organic matter content and, often, with microflora metabolic activity (Alef, 1995). In our soils the values of mineralized C after the first day of incubation were higher in organic than in conventional soils, even though the frequency distributions were not statistically different (data not shown). However, the basal respiration C ( $C_{basal}$ ) was significantly higher under organic than conventional management (Figure 4b).

The metabolic quotient,  $qCO_2$ , links respiratory activity (basal respiration) to the quantity of soil microflora (Anderson & Domsch, 1985a,b). In our soils, the distribution of  $qCO_2$  values was significantly higher in conventional treatments (Figure 4c). However, the  $C_{mic}/C_{org}$  ratio (Anderson & Domsch, 1989) and  $C_{MIN}$  values, which link soil basal respiration ( $C_{basal}$ ) to total soil organic C ( $C_{org}$ ), were not statistically different in soil from conventional and organic orchards (data not shown).

## Discussion

The higher values of  $C_{org}$  in organically managed soils, shown in Figure 2a, suggested that the adoption of this farming system caused the increase in soil organic matter under these climatic conditions. Other authors found a trend, though not a significant difference, in the increase in organic C in organically managed soils with respect to the conventional ones (Mäder *et al.*, 2002; van Diepeningen



**Figure 3** IEF profiles of humic matter purified from four pairs of organically (O) and conventionally (C) managed soils. The dotted box identifies the profile region of peaks focused at pH > 4.7 selected for relative peak-area calculation ( $A_s$ %). Abs, absorbance at 622 nm.

*et al.*, 2006). According to Clark *et al.* (1998), our results can be explained by the larger input of C in citrus organic systems which, however, cannot be attributed uniquely to the different patterns of fertilizer use (organic vs. inorganic fertilizers) but, presumably, to a range of different agricultural practices (i.e. weed management, green manuring, depth of tillage, animal manures, fertilizer use) and their interactions.

In addition, the larger content of humified substances  $(C_{HA+FA})$  (Figure 2c) indicated that organic management resulted in not only more storage of organic C but also the subsequent formation of more humified organic compounds.

Because total organic C and  $C_{HA+FA}$  content are widely accepted as among the most robust and reliable indicators of soil quality, the significant recorded differences in these parameters attribute higher quality to these organically managed soils. However, the lack of differences of HR and DH



**Figure 4** Frequency distribution of soil microbial biomass ( $C_{mic}$ ) (a), soil basal respiration ( $C_{basal}$ ) (b) and soil metabolic quotient (qCO<sub>2</sub>) (c) of the 13 pairs of orchard soils (organic vs. conventional). *P*-value, significance value of the difference between the distributions according to the Wilcoxon signed rank test. Bars (or whiskers) represent the dispersion of the values below or above the lower quartile and the upper quartile, respectively.

between organic and conventional soils demonstrated that soil management did not influence the rate at which humified materials were formed in soil. In other words, it is important to emphasize that soil management positively influenced the size of each of the determined organic matter fractions (total, extractable and humified organic matter), without changing the relative importance of each C pool (Trinchera *et al.*, 1998, 1999). So, it can be hypothesized that, even when total soil organic matter increases, the transformation into humified/labile fractions follows a pattern similar to that observed in conventional systems.

As far as IEF is concerned, several authors (Ciavatta *et al.*, 1990; De Nobili *et al.*, 1990; Ciavatta & Govi, 1993; Alianiello & Fiorelli, 1998; Dell'Abate *et al.*, 2002; Canali *et al.*, 2004; Trinchera *et al.*, 2007) demonstrated that humic and humic-like substances purified from soils, peats, composts and organic wastes show a group of characteristic electrophoretic peaks focusing at pH higher than 4.7, used to calculate the  $A_s$ . The significant difference between the  $A_s$  value for organic and conventional soils (Figure 2d) showed a qualitative improvement in humic substances, characterized by lower acidity and increased molecular complexity, in organically managed soils (Dell'Abate *et al.*, 2002).

Soil microbial biomass represents the living organic matter fraction responsible for energy and nutrient cycling and for regulating organic matter transformation (Gregorich *et al.*, 1994). It is a potential source of plant nutrients (Brookes *et al.*, 1984) and a linear relationship has been found between soil C microflora and total organic C in agricultural soils (Anderson & Domsch, 1980). Results obtained in our work confirmed what has been reported above, because in organic management, both soil microflora C ( $C_{mic}$ ) and total organic carbon ( $C_{org}$ ) were higher than in conventional treatments. In addition, because microbial biomass content is generally considered as one of the indicators of soil fertility, the higher value observed in organically managed citrus soils could be interpreted as an indication of greater soil quality (Franca *et al.*, 2007).

It has been demonstrated that measurements of soil respiration can be used to discriminate between different soil management practices (Pankhurst *et al.*, 1995). This is also confirmed by our results which show that basal respiration was able to discriminate between organic and conventional managements (Figure 4b). Higher values for basal respiration in soils under organic management accord with the increase in soil organic matter and soil microbial C biomass. Because basal respiration represents the soil microbial energy requirement at a steady-state condition, the higher values found in organically managed soils indicate that its microbial community increased the energy needs.

The metabolic quotient  $(qCO_2)$  which links the basal respiration and soil C biomass has been widely used as an indicator of soil quality and soil C utilization efficiency at steady state. In particular, Anderson & Weigel (2003) demonstrated that this parameter is able to discriminate between soil management systems. In their study of soil microbial activity in organically and conventionally managed citrus orchards, Franca *et al.* (2007) found that  $qCO_2$  of organically managed soils was higher than that of the conventionally managed ones. This difference was explained by the greater availability of organic residues with a low C/N ratio in the organically managed soil causing increased respiration without an equivalent increment in microbial biomass.

Conversely, Anderson & Domsch (1990) showed that crop rotations favoured a lower qCO<sub>2</sub> than monocultures, whereas Fließbach & Mäder (2000) found a lower qCO<sub>2</sub> in organic rather than in conventional soils. Anderson (2003) showed that microbial communities in soil of long-term crop rotation systems are energetically more efficient, having a lower qCO<sub>2</sub> value and higher corresponding  $C_{mic}/C_{org}$  ratio (for increased microflora C content) compared with soil in monoculture systems. Mäder et al. (2006), in their long-term DOK experiment, found a lower  $qCO_2$  in biodynamic and organic systems compared with conventional systems. On the basis of this evidence these authors concluded that the lower values demonstrated that soil C utilization in conventional management is metabolically less efficient. Also, Probst et al. (2008) found a lower  $qCO_2$  in soil of organically managed vineyards compared to that in conventional vineyards, confirming that the increase in microbial substrate use efficiency probably cannot be attributed to specific agricultural practices but to the overall effect of the organic management of soil.

The same trend was confirmed by our data, because the organic citrus orchards were characterized by a significantly smaller  $qCO_2$  and an increase in  $C_{MIN}$  and  $C_{mic}/C_{org}$  (even though the latter parameters were not significantly different) compared with the conventional systems (Figure 4b). The metabolic quotient was more sensitive to soil management than either  $C_{MIN}$  or the ratio  $C_{min}/C_{org}$ .

### Conclusions

Our study demonstrated that the organic citrus orchard soils were characterized by a general increase in all the organic matter pools that we measured. Nevertheless it is notable that these increases did not directly influence the organic matter turnover, because the humification parameters (HR and DH) and the mineralization coefficient ( $C_{MIN}$ ) were not affected by the system of management.

What is remarkable was the effect of organic management on soil humified materials, as shown by higher  $A_s$ % values in soils from the organic orchards. This indicates that the organic approach can contribute to the build-up of stabilized organic matter. A final point concerns the role of organic management on soil microbial functioning. Lower q(CO<sub>2</sub>) values in organic systems supported the hypothesis of an improved efficiency of soil microflora in utilizing energy and organic resources, which indicates an increased tendency for organically managed soils to establish ecological systems able to sustain biological productivity in the long term.

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