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Microbial biomass in soils under different tillage and crop rotation systems

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Abstract A long-term study on the effect of different crop rotations [soybean/wheat, S/W; maize/wheat, M/W or cotton/wheat, C/W] and tillage regimes [no-tillage (NT) or conventional tillage (CT)] on microbial biomass and other soil properties is reported. The experiment was established in 1976 in southern Brazil as a split-plot experimental design in three replications. Soil samples were taken in 1997 and 1998 at 0- to 5-, 5- to 10- and 10- to 20-cm depths and evaluated for microbial biomass C, N, P and S by direct extraction methods. The NT system showed increases of 103%, 54%, 36%, and 44% for microbial biomass C, N, P, and C_{mic}:C_{org} percentage, respectively at the 0- to 5-cm depth. NT systems also increased the C to N:S:P ratios. These results provide evidence that tillage or crop rotation affect microbial immobilization of soil nutrients. The larger amount of C immobilized in microbial biomass suggests that soil organic matter under NT systems provides higher levels of more labile C than CT systems.

Keywords Microbial biomass · Tillage systems · Crop rotation

Introduction

Quality and degradation of soils is of particular concern in tropical regions where intensive management and year-round warm temperatures can result in high rates of organic matter decomposition. Agricultural practices that improve soil quality and agricultural sustainability have been receiving more attention from researchers and farmers.

Minimum tillage systems such as no-tillage (NT) or direct drill, which plant directly through the mulch with minimal soil disturbance, and diverse crop rotations are practices that could maintain and improve soil quality. Conventional tillage (CT), which uses plowing and disking to prepare the land, reduces soil organic matter, particularly in tropical/subtropical conditions. This can cause increased organic matter oxidation and reduction of soil structure by degrading soil aggregates. Macroaggregates in particular are susceptible to tillage degradation and represent an important mechanism for protection and maintenance of soil organic matter (SOM) that can be lost under CT (Beare et al. 1994). Similarly, manipulating the diversity of cropping sequences can also affect soils by affecting C levels, as can the chemical composition of organic residues that are added to soils.

These effects on soil physical and chemical properties of management affect the microbial biomass and important processes such as decomposition of organic matter and mediation of nutrient availability to plants. The microbial biomass drives nutrient mineralization and is a small but labile source of major plant nutrients (C, N, P and S) (Jenkinson and Ladd 1981; Dick 1992). Also, microbial biomass can be an early indicator of changes in soil management compared to total organic C and N, which are unresponsive over short periods (Powlson and Jenkinson 1981; Carter 1986; Powlson et al. 1987; Saffigna et al. 1989). Thus, microbial biomass can be used to determine the level of degradation of the soil (Smith and Paul 1990; Doran and Parkin 1994; Brookes 1995; Sparling 1997).

Although there has been considerable research on the effects of soil and crop management on soil microbiology in temperate regions (Carter 1986; Follet and Schimel 1989; Saffigna et al. 1989; Fauci and Dick 1994; Franzluebbers et al. 1995; Miller and Dick 1995; Mendes et al. 1999) there is relatively little information in tropical/subtropical environments, particularly for highly weathered soils (Alvarez et al. 1995; Guerra et al. 1995;

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Balota et al. 1998; Rheinheimer et al. 2000), which are typically warmer and have different annual rainfall patterns than temperate regions. The objectives of this study were to evaluate the microbial biomass and its activity in soil under long-term crop rotations and tillage systems in the subtropical soil of Brazil.

Material and methods

Study site and soil sampling

A study of crop rotations in combination with NT or CT systems was conducted on an experiment established in 1976 at the Experimental Station of the Agronomic Institute of Paraná (IAPAR), district of Londrina, State of Paraná, in southern of Brazil (23°40′S, 50°52′W and 576 m altitude) on an Oxisol (Typic Haplorthox) with 86% clay, 11% silt and 3% sand in the first 20-cm layer.

The experiment had a split-plot design (three replications) with tillage (NT or CT) as the main plot (65×25 m) and crop rotation [soybean/wheat (S/W), maize/wheat (M/W) and cotton/wheat (C/W)] as the subplot (8×25 m), which were separated by 2.0 m. No tillage consisted of planting crops into undisturbed soil by opening a narrow slot of sufficient width and depth to cover the seed. Conventional tillage consisted of one disk plowing and two diskings with a light harrow to level the soil and prepare the seedbed. Soil chemical analyses were done according to Pavan et al. (1992). The chemical properties at the beginning of the trial (1976) were 5.9 and 5.6 for pH, 12.8 and 11.3 g kg^{-1} for total C, 3.2 and 2.4 mg P kg^{-1}, and 9.7 and 9.0 cmol kg^{-1} for CEC, at the 0- to 10-cm and 10- to 20-cm depths, respectively. The fertilizers were added according to the soil analysis done before each cropping, which averaged 95 kg N, 55 kg P and 42 kg K ha^{-1} year^{-1} over 20 years. Nitrogen fertilizer was never applied to the soybean crop.

The soils were sampled in August at the end of the winter cropping period in 1997 and 1998. Five soil sub-samples were taken (+200 g with a small shovel for total of >1 kg soil) randomly from each subplot at 0- to 5-, 5- to 10- and 10- to 20-cm depths. The samples were homogenized and sieved through a 4-mm screen after removing large plant material.

All determinations were made in triplicate and are expressed on a dry weight basis. Data were analyzed using the SAS statistical package (SAS Institute 1998). Within each field, replication data was averaged over the 2 years before statistical analysis.

Analyses

The microbial biomass C (MBC) was determined by the fumigation-extraction method according to Vance et al. (1987) using a correction factor (k_c) of 0.33 (Sparling and West 1988). The microbial biomass N (MBN) was determined by the method of Brookes et al. (1985) using a 0.54 conversion factor (Brookes et al. 1985). The microbial biomass P (MBP) was determined by fumigation-extraction according to method of Brookes et al. (1982) and McLaughlin and Alston (1986) using a 0.40 conversion factor (k_p; Brookes et al. 1982). The microbial biomass S (MBS) was determined by the Chapman (1987) fumigation method using a conversion factor of 0.35 (k_s; Chapman 1987).

Results and discussion

Table 1 shows the chemical analyses where NT had 44%, 34% and 14% greater total C. Extractable P in the soil under NT increased by 398% and 96% for the 0- to 5- and

Table 1 Chemical properties of the soil after 22 years of cropping

Crop rotation	pH	Total C g kg^{-1}	Extractable P mg kg^{-1}	CEC	Exchangeable H + Al
				cmol kg^{-1}	
Conventional tillage (0- to 5-cm depth)					
S/W[a]	4.50	15.33	18.2	13.19	6.68
M/W	4.30	14.70	15.5	12.78	7.20
C/W	4.20	13.95	21.7	12.83	7.75
No-tillage (0- to 5-cm depth)					
S/W	4.80	20.58	79.1	15.93	6.68
M/W	4.60	22.35	73.6	13.61	7.20
C/W	4.70	20.60	122.8	16.74	7.20
Conventional tillage (5- to 10-cm depth)					
S/W	4.50	13.36	17.9	12.20	6.20
M/W	4.40	15.26	12.8	13.13	7.20
C/W	4.20	13.21	20.4	12.87	7.75
No-tillage (5- to 10-cm depth)					
S/W	4.50	17.27	27.0	14.13	7.20
M/W	4.60	18.96	29.0	15.51	7.20
C/W	4.80	19.67	44.3	15.77	6.68
Conventional tillage (10- to 20-cm depth)					
S/W	4.50	14.35	19.1	12.59	6.68
M/W	4.30	15.61	18.0	13.71	7.75
C/W	4.20	13.82	12.3	12.83	7.75
No-tillage (10- to 20-cm depth)					
S/W	4.60	16.32	10.8	13.88	6.68
M/W	4.60	17.19	9.7	13.89	6.68
C/W	4.80	16.24	14.9	14.56	5.75

[a] *S* Soybean, *W* wheat, *M* maize, *C* cotton;, pH: $CaCl_2$ 0.01 M; P: Mehlich; C: Walkley and Black

Table 2 Microbial biomass C, N, P and S as affected by different tillage and crop rotation systems

Crop rotation	Microbial biomass							
	C		N		P		S	
	CT	NT	CT	NT_plus	CT	NT	CT	NT
	μg g^{-1} soil							
0- to 5-cm depth								
S/W[a]	223 b[b]A[c]	369 aA	23.4 bA	33.6 aA	10.8 aA	12.1 aB	17.0 aA	8.9bB
M/W	181 bA	389 aA	17.5 bA	27.6 aA	9.0 bB	12.1 aB	16.8 aA	12.7aA
C/W	145 bA	372 aA	17.2 bA	28.3 aA	10.7 bA	18.3 aA	12.2 aA	12.0aAB
5- to 10-cm depth								
S/W	105 aB	154 aB	14.1 bB	23.7 aA	5.6 aB	5.6 aB	14.8 aB	16.5aA
M/W	195 aA	185 aAB	19.9 aA	19.3 aA	9.0 aA	8.2 aAB	19.7 aA	14.5bAB
C/W	105 bB	268 aA	8.4 bC	22.4 aA	8.6 aA	10.0 aA	16.3 aAB	10.1bB
10- to 20-cm depth								
S/W	220 aA	269 aA	25.4 aA	24.4 aA	8.8 aB	8.4 aA	9.5 bB	14.2aA
M/W	225 aA	195 aA	22.0 aA	16.7 aA	12.0 aA	7.3 bA	14.0 aA	13.1aA
C/W	111 bA	195 aA	11.5 aB	15.5 aA	8.0 aB	8.4 aA	7.2 aB	13.2aA

[a] *S* Soybean, *W* wheat, *M* maize, *C* cotton
[b] Values followed by the *same lower case letter* comparing tillage within crop rotation are not significantly different at $P \leq 0.05$
[c] Values followed by the *same upper case letter* comparing crop rotations within tillage regime are not significantly different at $P \leq 0.05$

5- to 10-cm depths, respectively, but decreased 39% in the 10- to 20-cm depth over CT. There was no effect of crop rotation on total C. Extractable P in the C/W rotation was about 60% higher than other crop rotations at the 0- to 5- and 5- to 10-cm depths.

Microbial biomass

Microbial biomass C, N and P had a higher range of values in NT over CT across all depths (Table 2), but MBS largely was unaffected or decreased with NT compared to CT. Averaging across crop rotations in the 0- to 5-cm depth, showed a 100%, 54%, and 39% increase of MBC, MBN, and MBP, respectively, for NT over CT. This same effect of NT was found down to 20 cm with 59% and 31% higher levels of MBC and MBN, respectively, in NT over CT. Conversely, MBP was only significantly affected by tillage in the 0- to 5-cm depth. These results are consistent with other studies of surface soils where NT caused an increase over CT of 57% and 181% (Alvarez et al. 1995) for MBC and 46% (Follet and Schimel 1989) and 65% (Franzluebbers et al. 1995) for MBN.

The general increase of microbial biomass under NT over CT, especially under tropical/subtropical conditions, could be attributed to several factors, such as a lower temperature, higher moisture content, greater soil aggregation and higher C content. The lack of a major disturbance event with NT likely provides a steady source of organic C to support the microbial community compared to CT where a temporary flush of microbial activity with each tillage event results in large losses of C as CO_2. Furthermore, less disturbance favors formation and stabilization of macroaggregates to improve and protect habitat for microbiota (Powlson and Jenkinson 1981; Alvarez et al. 1995; Sorensen et al. 1996; Hungria et al. 1997; Balota et al. 1998).

Although our results show a depth effect with NT it does not appear to be as striking compared to other long-term studies in temperate regions where there is a steady decrease of microbial biomass with depth. For example, inclusion of soybean in the rotation resulted in MBC decreasing from 369 μg C g^{-1} soil in the 0- to 5-cm depth to 154 μg C g^{-1} soil in the 5- to 10-cm depth and then increasing to 269 μg C g^{-1} soil in the 10- to 20-cm depth (Table 2). This trend follows that of another study on these same plots that showed enzyme activities staying fairly constant with depth for NT (Balota et al. 2002). It may be that the tropical environment with high soil temperatures has less potential than temperate regions to maintain high levels of organic matter at the surface to support the microbial populations compared to cooler soils in temperate regions.

There were few effects of crop rotations on microbial biomass. However, when maize was introduced in the rotation under CT, at the 5- to 10-cm depth, it had substantially higher MBC compared to the other rotations. This is likely due to the higher level of biomass input with maize (9.7 t ha^{-1} year^{-1} for maize vs 5.5 t ha^{-1} year^{-1} for soybean and 3.0 t ha^{-1} year^{-1} for cotton) and that this depth is where a significant amount of residue was placed upon incorporation with CT. Surprisingly, the crop rotation did not affect MBC or MBN in the 0- to 5-cm depth within each tillage regime. Lack of differences in microbial biomass (C and N) at this depth may reflect both long-term stability of substrate and higher immobilization of N in the maize than in soybean residues. The soybean biomass has a narrower C:N ratio, making it more labile for supporting microbial growth even though it has a lower residue input to soils than maize.

C_{mic}:C_{org} percentage

The C_{mic}:C_{org} percentage varied from 0.8% to 1.5% in the CT plots and from 0.9% to 1.8% in the NT plots (Table 3). The NT system significantly increased the C_{mic}:C_{org} percentage by 44%, 13% and 8% at the 0- to 5-, 5- to 10- and 10- to 20-cm depths, respectively. On average, NT resulted in an increase of 30% C_{mic}:C_{org} percentage over CT. There was a significant effect of crop rotation on C_{mic}:C_{org} percentage with greater effects from the maize rotation at the 5- to 10-cm depth and the soybean rotation at the 10- to 20-cm depth.

The C_{mic}:C_{org} percentage could be higher with NT because it has more labile organic substrates maintained in the soil, which allows a higher MBC per unit of soil C. The microbial biomass represents only from 1% to 3% (Powlson and Jenkinson 1981), or up to 5% (Sparling 1992) of total organic C. This ratio could be an indicator of conversion efficiency of organic C into microbial C and losses of soil C during decomposition (Sparling 1992). Consequently, Sparling (1997) proposed that the C_{mic}:C_{org} percentage could be useful as a soil quality indicator to allow comparisons across soils with different organic matter contents. Generally, if a soil is being degraded, the microbial C pools will decline at a faster rate than the organic matter, and the C_{mic}:C_{org} percentage will decrease as well. This might allow for a calibrated soil quality indicator to predict whether soils are accumulating or losing soil C.

Jenkinson and Ladd (1981) proposed a 2.2 C_{mic}:C_{org} percentage is an equilibrium threshold for cultivated soil. All of the treatments in our study were below this threshold with a tendency for NT to have a higher percentage. However, it must be emphasized that this property has varied widely in the literature from 0.27% to 7.0% across different soil management systems, sampling times, and analytical methods (Anderson and Domsch 1989).

Microbial biomass ratios

The microbial biomass C:N:S:P ratio was affected by tillage and crop rotation at all depths (Table 3.). On average the ratio under CT was 18:2:2:1 but under NT it was 27:2:2:1. The C:N microbial biomass ratio is an indication of the relative proportion of fungi to bacteria (Anderson and Domsch 1980; Wheatley et al. 1990; Fauci and Dick 1994). Consequently, the wider C:N biomass of NT would suggest that NT plots have a greater proportion of fungal compared to bacterial biomass than CT. This would follow the observation that reduced soil disturbance favors establishment and maintenance of fungal hyphal networks (Wardle 1995).

The range of microbial biomass C:P ratios are quite wide in the literature from 5 to 63 under different climates, soil management systems and analytical procedures (Perrot and Sarathchandra 1982; Brookes et al. 1984; Saffigna et al. 1989; Prasad et al. 1994; Guerra et al. 1995; Joergensen et al. 1995). The microbial biomass C:P ratio can provide an indication that of management conditions are affecting incorporation of P into microbial biomass (Joergensen et al. 1995). For example, Guerra et al. (1995) observed a narrower microbial biomass C:P ratio with addition of P in soils. The high values of MBP when compared with soil-available P would suggest that the microbial P pool is important in P fluxes in our soils. The microbial immobilization of P from P fertilizing is of particular importance in soils which have strong adsorption and occlusion of phosphate by Fe and Al oxides. Our data of microbial C:P ratios obtained in this work were slightly above the average of Anderson and Domsch (1980) who reported a C:P ratio of 17.6 for bacteria and 12.0 for fungi in pure cultures.

Another important measure for high P fixing soils is the MBP as a percentage of total microbial biomass because it reflects the potential of a soil to provide plant-available P via microbial mineralization (Table 4). From this P measurement, annual P flux can be calculated assuming a turnover of 2.5 times per year (Brookes et al. 1984; Rheinheimer et al. 2000). We obtained a P flux of 4.1 under CT and 5.5 mg dm^3 $year^{-1}$ under NT at the 0- to 5-cm depth compared to results obtained under subtropical conditions by Rheinheimer et al. (2000), who observed values from 8 to 22 mg dm^3 $year^{-1}$. Under temperate conditions, Brookes et al. (1984) observed values from about 7 kg P ha^{-1} $year^{-1}$ in arable soils to 23 kg P ha^{-1} $year^{-1}$ under grassland soils. Our results suggest that NT would have a moderate effect on P flux in our soils.

Table 3 Microbial biomass ratio (C:N:S:P) and C_{mic}/C_{org} percentage as affected by different tillage and crop rotations systems[a]

Crop rotation	C:N:S:P Ratios		C_{mic}/C_{org}	
	CT	NT	CT	NT
			%	
0–5 cm				
S/W	21:2:2:1	30:3:1:1	1.5 a[b]A[c]	1.8 aA
M/W	20:2:2:1	32:2:1:1	1.2 bA	1.7 aA
C/W	14:2:1:1	22:2:1:1	1.0 bA	1.8 aA
5–10 cm				
S/W	19:3:3:1	27:4:3:1	0.8 aB	0.9 aA
M/W	22:2:2:1	22:2:2:1	1.3 aA	1.0 aA
C/W	12:1:2:1	27:2:1:1	0.8 bB	1.4 aA
10–20 cm				
S/W	25:3:1:1	32:3:2:1	1.5 aA	1.7 aA
M/W	19:2:1:1	27:2:2:1	1.4 aA	1.1 aB
C/W	14:1:1:1	23:2:2:1	0.8 aA	1.2 aAB

[a] *S* Soybean, *W* wheat, *M* maize, *C* cotton
[b] Values followed by the *same lower case letter* comparing tillage within crop rotation are not significantly different at $P \leq 0.05$
[c] Values followed by the *same upper case letter* comparing crop rotations within tillage regime are not significantly different at $P \leq 0.05$

Table 4 Percentage of total biomass as N, P or S microbial biomass

Crop rotation	Percentage of total microbial biomass[b]						Annual biomass P flux	
	N		P		S			
	CT	NT	CT	NT	CT	NT	CT	NT
	%						mg P dm^3	
0–5 cm								
S/W[a]	6.1 a[c]A[d]	4.6 aA	2.7 aA	1.7 aB	4.1 aA	1.2 bA	4.3	4.8
M/W	5.0 aA	3.6 aA	2.6 aA	1.6 bB	4.7 aA	1.6 bA	3.6	4.9
C/W	6.1 aA	3.8 aA	3.9 aA	2.3 aA	4.3 aA	1.6 bA	4.3	6.9
5–10 cm								
S/W	6.8 aA	7.8 aA	2.7 aB	1.8 aA	8.2 aA	4.9 bA	2.2	2.3
M/W	5.1 aAB	5.5 aA	2.3 aB	2.3 aA	5.1 aA	4.0 aA	3.6	3.3
C/W	4.1 aB	5.5 aA	4.1 aA	2.3 bA	8.0 aA	1.9 bB	3.4	4.0
10–20 cm								
S/W	6.0 aA	4.0 aA	2.1 aA	1.6 aA	2.2 aA	2.7 aA	3.5	3.4
M/W	4.9 aA	4.3 aA	2.7 aA	1.9 bA	3.2 aA	3.4 aA	4.8	2.9
C/W	6.0 aA	4.0 aA	4.2 aA	2.2 aA	4.2 aA	3.5 aA	3.2	3.4

[a] *S* Soybean, *W* wheat, *M* maize, *C* cotton
[b] Assuming dry biomass contains 50%C
[c] Values followed by the *same lower case letter* comparing tillage within crop rotation are not significantly different at $P \leq 0.05$
[d] Values followed by the *same upper case letter* comparing crop rotation within tillage regime are not significantly different at $P \leq 0.05$

Conclusions

Reduction of tillage had a much bigger effect on the microbial biomass, particularly in the 0- to 5-cm depth than did crop rotations. This is somewhat surprising because inclusion of maize in the rotation resulted in more than twice as much biomass input compared to soybean or cotton. This may be due to the year round warm temperatures in southern Brazil that cause rapid decomposition of organic inputs. Thus, for the subtropical conditions on a highly weathered soil in our environment, soil disturbance is more important in maintaining organic C for a slow release of C to sustain the microbial biomass. The formation and stabilization of macroaggregates in NT soil is likely to be a key mechanism for the protection and maintenance of soil organic matter (Beare et al. 1994) and for microbial habitat (Dick 1992). Indeed, previous research in these treatment plots showed that mean weight diameter and geometric mean diameter of aggregates was significantly greater with NT than CT (Castro-Filho et al. 2002).

The results showed that the microbial biomass in no-till soils contains more nutrients than microbial biomass in CT systems. This suggests that there would be greater cycling and fluxes of nutrients through the microbial biomass in NT soils. There was evidence that there may be a somewhat greater P flux through the microbial biomass, which may improve P nutrition of plants on the highly weathered soils in this part of Brazil, where soils have high P fixation capabilities. Higher levels of biological P cycling could increase efficiency of P uptake by plants by reducing the potential for P sorption and precipitation reactions that dominate P availability in these soils.

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