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## Microbial populations and the activity of the soil under agricultural and agricultural-pastoral systems

Martha Regina Lucizano Garcia<sup>a</sup> & Ely Nahas<sup>b</sup>

<sup>a</sup> State Technical School of Ilha Solteira , Ilha Solteira , Brazil

<sup>b</sup> Universidade Estadual Paulista, Department of Crop Production, Jaboticabal, Brazil

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# Microbial populations and the activity of the soil under agricultural and agricultural-pastoral systems

Martha Regina Lucizano Garcia<sup>a</sup>\* and Ely Nahas<sup>b</sup>\*

<sup>a</sup>State Technical School of Ilha Solteira, Ilha Solteira, Brazil; <sup>b</sup>Universidade Estadual Paulista, Department of Crop Production, Jaboticabal, Brazil

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The effects of agricultural-pastoral and tillage practices on soil microbial populations and activities have not been systematically investigated. The effect of no-tillage (NT), no-tillage agricultural-pastoral integrated systems (NT-I) and conventional tillage (CT) at soil depths of 0-10, 10-20 and 20-30 cm on the microbial populations (bacteria and fungi), biomass-C, potential nitrification, urease and protease activities, total organic matter and total N contents were investigated. The crops used were soybean (in NT, NT-I and CT systems), corn (in NT and NT-I systems) and Tanner grass (Brachiaria sp.) (in NT-I system); a forest system was used as a control. Urease and protease activities, biomass-C and the content of organic matter and total N were higher (p < 0.05) in the forest soil than the other soils. Potential nitrification was significantly higher in the NT-I system in comparison with the other systems. Bacteria numbers were similar in all systems. Fungi counts were similar in the CT and forest, but both were higher than in NT. All of these variables were dependent on the organic matter content and decreased (p < 0.05) from the upper soil layer to the deeper soil layers. These results indicate that the no-tillage agricultural-pasture-integrated systems may be useful for soil conservation.

**Keywords:** urease activity; protease activity; potential nitrification; biomass-C; soil bacteria; soil fungal populations

#### Introduction

Soil management practices vary widely in Brazil. A conventional tillage approach for the cultivation of plants is most frequently used by farmers to reduce soil compaction, to improve aeration and to control weeds (Assis Júnior et al. 2003; Franchini et al. 2007; Crusciol et al. 2008; Carvalho et al. 2009). However, minimum tillage or no-till cropping are used to reduce erosion, to conserve moisture, to maintain and improve soil quality and to increase production (Doran et al. 1998; Allmaras et al. 2000; Nyakatawa et al. 2000; Kladivko 2001). Current efforts focus on alternative systems to optimize the productivity and sustainability of agroecosystems, as well as to reduce production costs and environmental impacts on Brazilian soils (Franchini et al. 2007). Systems that combine row agricultural crops and pasture in the same area, either successively or simultaneously, constitute

<sup>\*</sup>Corresponding author. Email: enahas@fcav.unesp.br

a method of sustainable use and management of natural resources (Assis Júnior et al. 2003).

One of these agro-silvo-pastoral systems began by planting eucalyptus (*Eucalyptus camaldulensis* and *E. urophylla*) and rice (*Oryza sativa*) together. The rice was subsequently replaced by soybean (*Glycine max*) and Tanner grass (*Brachiaria* sp.), and then cattle were brought out in the third year. This is done for 11 years until the *Eucalyptus* is cut (Ribeiro et al. 2007). Silvo-pastoral systems have high soil nutrients and high organic content, plus high levels of biological activity, not only in the grass, but also in the native thorn forest (Wick et al. 2000; Nair et al. 2007).

A system that integrates crops and livestock can eliminate or reduce physical, chemical and biological soil degradation. This system has agronomical, economical, ecological and social benefits for optimal agro-ecological sustainability (Kluthcouski and Aidar 2003). Grazing activities benefit from the application of limestone and residual fertilizers in the agricultural crop, which in turn benefits the physical characteristics of the soil and also the detrital material from the pasture (Mello et al. 2004). In an integrated system, in Brazil, it was possible to produce corn and use *Brachiaria ruziziensis* for animal grazing (Lara-Cabezas and Padua 2007). In Tanzania,  $\sim 50-70\%$  of cattle are kept in an agro-pastoral system consisting of various agricultural crops, and the raising of traditional livestock that graze on communal lands is also quite common (Kanuya et al. 2006). Under certain conditions, there is little competition between the plant consortia. For example, *Brachiaria brizantha* sown between the plant rows did not affect corn yield (Agnes et al. 2004).

In addition to reducing the erosion and the production costs, no-till soil management systems significantly impact the biota and the cycling of soil nutrients. No-till systems increase the concentration of nutrients, organic matter and pesticides in the surface soil (Dick 1992). Conventional tillage decreases the content of soil organic matter and the activities of  $\beta$ -glucosidase and the urease (Van Den Bossche et al. 2009). Biological activity has been found to be higher in soils under no-till management than under conventional tillage (Bolinder et al. 1999; Alvear et al. 2005).

Plowing buries most of the residues into deeper layers with conventional tillage, however, most residues are left at the soil surface in no tillage or reduced till systems (Burgess et al. 2002). Plowing significantly decreased the microbial biomass carbon, nitrogen and phosphorus contents, when compared with no-till or permanent pasture (Aslam et al. 1999). The maintenance of crop residues can benefit the community of organisms in the no-till system. Numbers of total bacteria, *Pseudomonas*, actinomycetes, total fungi and *Fusarium* spp. were higher in the no-till and pasture systems than in the residue-removal system of zero tillage and in conventional tillage (Govaerts et al. 2008). The amounts of microbial biomass carbon and nitrogen were 80 and 104% higher, respectively, in no-till than in conventional till systems (Franchini et al. 2007).

Soil quality is often related to the activity of soil microorganisms. Soil enzymes are derived primarily from microbial activity, and therefore, the determination of their activities can be used to obtain information about microbial populations and biochemical transformations in the soil. Management practices for plants and soil, which lead to an increase of organic matter, can create a favorable environment for the development of microorganisms and microbial activity. Several studies have indicated that no-till increases the activity of several enzymes. Dehydrogenase, phosphatase and arylsulfatase enzyme activities were lower in conventional management than in a no-till system (Bergstrom et al. 1998). However, significant results were not always obtained in these systems. Only readily mineralizable C and alkaline phosphatase activity clearly differed in no-till and tilled soils in South Dakota agricultural fields (Carpenter-Boggs et al. 2003). In addition, microbial biomass and enzyme activity are more sensitive indicators than the total carbon concentration to changes in soil management practices (Bergstrom et al. 1998). Enzymes in the soil can give information on the different biochemical reactions, which often reflect natural or anthropogenic processes (Kremer and Li 2003).

Changes in microorganism population and activities may potentially indicate the effect of agro-pastoral systems. However, there is a lack of consistent information on soil microbial populations and on their activities in this ecosystem type. The objective of this study was to evaluate the influence of no-tillage systems, no-tillage agricultural–pastoral integrated systems and conventional tillage management systems on the chemical and microbiological characteristics of soil at different depths.

#### Materials and methods

#### Experimental site

The experimental sites were located at two farms in Guairá, São Paulo, Brazil  $(20^{\circ} 20' 25'' \text{ S} \text{ and } 48^{\circ} 23' 90'' \text{ W})$ . The soil at both farms is classified as typic Eutrustox. The altitude is 490 m and the climate, according to Köppen classification, is subtropical type, with rainy summers and dry winters (humid subtropical climate, Cwa). The mean annual rainfall is 1300 mm, with average an annual temperature of 24°C. The main soil characteristics are found in Table 1.

Four systems were studied: no tilled (NT), no tilled integrated with livestock (NT-I), conventional tillage (CT) and natural forest (F). The NT system was

System	Depths	Р	$\mathbf{K}^+$	Ca <sup>2+</sup>	$Mg^{2+}$	$\mathrm{H^{+}} + \mathrm{Al^{3+}}$	BS	CEC	V
	(cm)	$(mg g^{-1})$	$(\text{mmol}_{c} \text{ g}^{-1})$						
NT	0-10	64	3.5	52	20	17	75	91	81
	10-20	47	1.3	48	19	14	69	83	82
	20-30	16	1.5	36	18	16	55	70	75
NT-I	0-10	15	2.0	23	13	75	38	113	33
	10-20	13	1.5	14	8	69	23	92	24
	20-30	10	1.1	11	7	59	19	78	23
СТ	0-10	66	2.4	50	14	21	66	87	76
	10-20	37	1.2	29	8	28	39	66	58
	20-30	19	1.4	26	8	26	35	61	57
Forest	0-10	79	4.9	60	15	26	79	105	75
	10-20	56	4.3	54	12	26	70	96	73
	20-30	22	3.7	44	10	22	58	80	72

Table 1. Chemical and physical properties of the soils under different tillage systems at three soil depths.

Note: No-tillage (NT), no-tillage agricultural-pastoral integrated system (NT-I), and conventional tillage (CT). CEC, cation exchange capacity; BS, sum of bases; V, degree of base saturation.

initiated in 2000, growing soybean with a rotation of corn every three years. Soybean CD 208 (Coodetec) was planted on 20 November 2005 at a row spacing of 0.50 m, and was fertilized with 225 kg ha<sup>-1</sup> of 02-20-25 (N-P-K) + micronutrients. An inoculant (*Bradyrhizobium japonicum* with  $1 \times 10^8$  cells g<sup>-1</sup> was added in the amount of 100 g per 50 kg seed) was used when the soybean was planted. Thirty days after emergence, the seedlings received foliar fertilization with Co + Mo. Two applications of herbicide and three applications of fungicide were used. In this system, the soil was only turned over in the line of seeding and thereafter vegetation was retained on the surface. In March and April, the soybean was rotated with winter corn or sorghum.

The NT-I system was established in 2000, in the same manner as the NT agricultural system. However, this system differed from the NT system due to the integration of livestock production, using the Santa Fé cattle management system that is utilized in the Goiás State (Brazil). In this system, Brachiaria sp. was used for the pasture and seeding during corn fertilization. The corn grew first, which impaired the growth of the Brachiaria sp. because of shading. With the maturation and drying of the corn, *Brachiaria* sp. then continued its development for pasture formation. In the NT-I system, corn was rotated with soybean every three years and between seasons (autumn-winter) with grazing. After pasture formation in mid-May/June, the cattle were pastured until the second week of October. The total area was 38 ha with 7 to 8.5 AU (animal units) per ha. The cattle breed used was Nellore. Soybean was planted on 3 December 2005, using the cultivar MG/BR 46, was spaced at 0.50 m, and fertilized with 225 kg  $ha^{-1}$ of 02-20-18 (N-P-K) + micronutrients. Thirty days after emergence, the seedlings were fertilized with 50 kg ha<sup>-1</sup> of KCl and with a foliar fertilizer with Co + Mo. Two applications of herbicides and four applications of fungicides were applied.

The CT system has been cultivated for more than 20 years and the soil was prepared with plowing the land followed by two gradings. The soybean MG/BR 48 (Garimpo RCH) was planted on the 15 November 2005 at a row spacing of 0.50 m and was then fertilized with 350 kg ha<sup>-1</sup> of 03-15-15 (N-P-K). Four applications of fungicides and three applications of insecticides were applied.

The natural forest system was used as a control (F), and had the typical characteristics of the Brazilian savanna (called 'Cerrado'), including closed woody vegetation (Oliveira-Filho and Ratter 2002). From the beginning of the crop growth period ( $\sim 20$  years), the natural forest has changed very little and has been kept in a preservation status.

#### Soil sampling

Soil samples were collected from the inter-rows in the summer, during January and February 2006, when the soybean grains were maturing. A completely randomized sampling approach was used to allocate six replicates per site each measuring  $\sim 100 \times 100$  m. For each field replicate, 10 subsamples of the soil at depths of 0–10, 10–20 and 20–30 cm were collected and pooled in the field, and transported to the laboratory. Soil samples were sieved in a 2 mm sieve and were then homogenized. Each sample was divided into two parts, one part was kept at 4°C until use, and the other part of the sample air dried and kept at room temperature.

#### Microbial count

The number of colony-forming units (CFU) of bacteria and fungi was determined using the culture media Bunt and Rovira (1955) and Martin (1950), respectively. Soil samples (10 g) were suspended in sodium pyrophosphate 0.1% (w/v), shaken for 30 min and diluted up to  $10^{-4}$ . The number of CFU was determined after incubation at 30°C for 24 h for the bacteria and 72 h for the fungi. After this period, the number of colonies was counted according to Vieira and Nahas (2005).

#### Microbial biomass and activity

The microbial biomass carbon (MBC) was determined using the fumigationextraction method proposed by Vance et al. (1987). The soil samples were extracted by shaking for 30 min with 50 mL of 0.5 M  $K_2SO_4$  and a correction factor (Kc) of 2.64 was used (Vance et al. 1987). The potential nitrification of the soil was measured by incubating the soil with 60% total water-filled pore space for 21 days at  $30^{\circ}$ C with or without the addition of 160  $\mu$ g NH<sub>4</sub><sup>+</sup>-N as (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>) g<sup>-1</sup> dry soil (Schmidt and Belser 1994).  $NO_3^-$  was extracted with 1 M KCl and determined using the method of Keeney and Nelson (1982). Urease activity was estimated by incubating the soil at  $37^{\circ}$ C for 3 h in a 0.1 M phosphate buffer pH 6.7 and solution of 10% (w/v) urea (McGarity and Myers 1967).  $NH_4^+$  was extracted and the content was determined using phenol-hypochlorite. Protease activity was determined based on the procedure proposed by Nannipieri et al. (1979), by incubating the soil up to  $52^{\circ}$ C for 1 h in 1% (w/v) casein in a buffer of 0.1 M Tris-HCl pH 8.1. After 17.5% (w/v) trichloroacetic acid was added, the samples were centrifuged and Folin reagent (1:4) was added to react with formed tyrosin. The absorbance of the solution was measured spectrophotometrically at 660 nm.

#### Physical and chemical analyses

The total N contents were determined using the Kjeldahl method after the digestion and distillation of the soil samples (Bremner and Mulvaney 1982). The moisture content was determined after drying the soil sample at 105°C for 24 h and the soil organic matter content was calculated after soil incineration at 550°C for 24 h. The soil pH was measured with glass electrode in a soil suspension of 0.01 M CaCl<sub>2</sub> 1:2.5. Available P and exchangeable cations (= sum of bases Ca<sup>2+</sup>, Mg<sup>2+</sup>, and K<sup>+</sup>) were extracted using ion exchange resins, cation exchange capacity (CEC) and degree of base saturation were calculated, all in accordance with Raij et al. (2001).

#### Data analyses

The experimental design consisted of two factors: land use (no tillage, no tillage integrated with livestock, conventional tillage and natural forest) and soil depth (0–10, 10–20 and 20–30 cm), with six replicates randomly selected in each area. The variation among the areas and the variation inside of the areas were compared using the *F*-test. The significance of the *F*-test was evaluated at the 5% and the 1% probability levels. The counts of the microorganisms were transformed to log (x + 1), where x equals the number of CFU. Statistical analysis was performed using the SAS Institute (1990); the means were compared by the Tukey's test at 5% of

probability. Pearson correlation coefficients between all pairs of variables were calculated.

#### Results

Tillage practices significantly influenced the organic matter, total N contents and soil pH when comparing the NT, NT-I, CT and F systems at the three depths of soil samples taken (Figure 1). The highest organic matter content and total nitrogen, and



Figure 1. (A) Organic matter and (B) total nitrogen content in the soil, (C) soil pH from different management systems and soil depths. NT, no-tillage; NT-I, no-tillage agricultural-pastoral integrated system; CT, conventional tillage; and Forest.  $\blacksquare$  0–10;  $\blacksquare$  10–20;  $\Box$  20–30. Bars followed by the same upper case letter (management systems) or lower case letter (depths in each system) show no significant differences (p < 0.05). Error bars correspond to standard error of mean.

the lowest pH value (p < 0.05, Figure 1A–C) were found in the forest soil. The decrease in organic matter content in forest soil was small, from 6 to 15% on average, but the decrease in total N content was high, 15–56%, compared with the others (Figure 1A,B). The pH ranged from 3.9 in the forest soil to 4.8–5.6 in the other soils, respectively (Figure 1C). pH was lower in the NT-I soil than in the NT and the CT soils. Soil pH and organic matter content showed a minimal decrease, 3 and 4–7%, respectively, with the soil depth (p < 0.05; Figure 1A,C). The total N content decreased gradually with soil depth, resulting in a significant decrease of 22 and 33%, on average, in the 10–20 cm and 20–30 cm layers, respectively, in relation to the upper soil layer (Figure 1B).

Total bacteria and fungi counts in all the management systems ranged from 5.76 to 6.87 and 3.33 to 5.18 log (CFU g<sup>-1</sup> dry soil), respectively (Figure 2). However, only the fungi count varied significantly (p < 0.05) between the systems. The highest number of fungi was found in surface soil (0–10 cm) of the CT system and this count decreased sharply from 81 to 94% in the other soils. However, in the 10–20 cm and 20–30 cm soil layers, the fungi community was predominate in the NT-I system and in the F system, respectively. There was a significant (p < 0.05) and severe reduction in the bacteria (85%) and fungi (84%) counts from the surface layer to the deeper layer soils (Figure 2A, B).



Figure 2. (A) Bacterial and (B) fungal populations in the soil from different management systems and soil depths. See Figure 1 for management system abbreviations. CFU, colony-forming units. Microorganisms counts were transformed to  $\log (x + 1)$ , x = CFU.  $\blacksquare 0-10$ ;  $\blacksquare 10-20$ ;  $\square 20-30$ . Bars followed by the same upper case letter (management systems) or lower case letter (depths in each system) show no significant differences (p < 0.05). Error bars correspond to standard error of mean.

MBC was higher in the forest soil than in the other soils, and declined on average by 2–3.7 times (Figure 3). Greater reductions in MBC (31–50% on average) were observed with increasing soil depth (p < 0.05, Figure 3).

Potential nitrification activity was also higher in the forest soil, however, it was similar to the values found in the NT-I soil (p < 0.05) (Figure 4 A). The activities of protease and urease were, on average, significantly higher in the forest soil than in the other soils (Figure 4B,C). The average activity of the urease in different depths of forest soil was 8.3–14.6 times greater than that found in other soils. Also, the potential nitrification and protease activities in different depths of forest soil were 1.2–3.9 and 1.6–2.1 times higher, respectively, than those found in other soils (Figure 4B, C). The activities of potential nitrification, urease and protease decreased significantly in lower soil layers compared with the superficial layer. Compared with the initial values measured in the superficial layers, the activities of potential nitrification, urease and protease decreased by a range of 47–57%, 2–13% and 24–48%, respectively (Figure 4A–C).

#### Discussion

Soil biological and biochemical properties such as the number and activity of the soil microbiota, as well as the activity of hydrolytic enzymes may change with land use. Several authors comparing land use have reported significant differences in soil quality parameters under forest soils compared with agricultural soils (Gallardo and Schlesinger 1994; Ralte et al. 2005; Trasar-Cepeda et al. 2008). Indeed, several of the soil properties from forest soil were reduced in agricultural soils. In this study, forest soil showed the lowest pH but the highest amount of organic matter and the highest total N. Similar results were found when forest soils were compared with conventional tillage soils (Cookson et al. 2007). The loss of organic matter in agricultural soils has been identified as one of the main factors that influences the



Figure 3. Microbial biomass carbon values in a soil under different tillage systems at three depths. See Figure 1 for management system abbreviations.  $\blacksquare 0-10$ ;  $\blacksquare 10-20$ ;  $\Box 20-30$ . Bars followed by the same upper case letter (management systems) or lower case letter (depths in each system) show no significant differences (p < 0.05). Error bars correspond to standard error of mean.



Figure 4. (A) Potential nitrification, (B) urease and (C) protease activities in different management systems. See Figure 1 for management system abbreviations.  $\blacksquare 0-10$ ;  $\square 10-20$ ;  $\square 20-30$ . Bars followed by the same upper case letter (management systems) or lower case letters (depths in each system) show no significant differences (p < 0.05). Error bars correspond to standard error of mean.

depletion of soil quality (Trasar-Cepeda et al. 2008). In the CT system, the residues that remain after harvest were incorporated into the soil through plowing and harrowing, which guarantees their distribution in the soil profile. Decomposition provided nutrients to ensure the future growth of crops. In the NT and NT-I systems, crop residues remained on the surface soil, resembling natural systems such

as forests where decomposition depends on interaction with the underlying layer of soil and decreases in the lower layers, due to the concentration of residues near the soil surface (Meyer et al. 1996). Total N content decreased with the soil depth (Schilling et al. 2009) but did not vary among the cultivated soils where the same plants were used.

Although the forest soil had the highest content of organic matter, the microbial count was not predominant in this soil. When agricultural systems were compared, fungi were predominant in the CT soil. Several authors have attributed the increase in microbial populations in no-till soil to the retention of residues on the soil surface (Govaerts et al. 2008; Helgason et al. 2009). In this study, the fungi populations predominated in the CT probably because they initiate the decomposition of the organic matter incorporated into the soil. By contrast, Simmons and Coleman (2008) showed that fungi populations decreased in the CT soil, because tillage disrupted the fungal mycelia. However, other reports confirm our results. In the conventional system without plant residues, fungi prevailed when compared with the conventional system using plant residues, which also had more bacteria. This was due to the high tolerance of the fungi to the more acidic environment and low nutrient content (Govaerts et al. 2008). In fact, both the sum of bases (BS; Table 1) and the soil pH (Figure 1C; p < 0.05) in the CT soils were lower than in the NT soils. Spedding et al. (2004) found significant seasonal effects on fungi, but no effect of tillage on the microbial community. Kirchner et al. (1993) showed that more fungi were found in soil receiving N fertilizer than in unfertilized soil. Accordingly, in this research, there was a positive correlation between total N and fungal populations ( $r = 0.24^*$ ) but not bacterial populations (Table 2).

NT-I soil had characteristics that should have stimulated the population of microorganisms, however, compared with other systems, only the fungal community was increased and not the bacterial community. These characteristics included the retention of plant residues in the soil, the accumulation of moisture, less soil disturbance (zero till) and the deposition of animal wastes. In addition, the cropping systems that introduce C through green manure, cropping sequences or animal wastes tend to increase the population of microorganisms (Kirchner et al. 1993). However, it may be assumed that a considerable amount of vegetation was taken by the animal grazing, which depleted the soil, reducing the fertility and the availability of the nutrients necessary for the bacterial growth. In fact, there was a significant decrease in the pH (4.8) in the NT-I soil when compared with NT (5.6) (Figure 1C) and the lower concentration of nutrients (Table 1) may have influenced these results.

As a consequence of these results, the MBC values found in the NT-I soil were the lowest among the different systems studied. The decrease of the MBC in the studied systems is in agreement with results showing a significant correlation between MBC and the organic matter content ( $r=0.77^{***}$ ) and bacterial populations ( $r=0.27^{*}$ ) (Table 2). The soil microbial biomass is both a source and a sink for biologically mediated nutrients and is responsible for decomposing organic matter and nutrients within the soil (Gregorich et al. 2000). Organic matter in natural systems may have more unstable carbon and may support more microbial biomass than in cultivated systems (Carpenter-Boggs et al. 2003). The organic matter in the natural systems, like in the forest soil, is physically and chemically protected from biological degradation (Doran et al. 1998; Franchini et al. 2007). The increase from 2 to 3.7 times of the MBC in the forest soil, in relation to other systems, is consistent with that observed in natural soils and pastures, which increased at least

	SOM	pН	Total N	MBC	Nitrif.	Urease	Protease	Bacteria
pН	-0.55***	_	_	_	_	_	_	_
Total N	0.58***	-0.49***	_	_	_	_	_	_
MBC	0.77***	$-0.53^{***}$	0.60***	_	_	_	_	_
Nitrif.	0.29*	-0.25*	0.54***	0.19NS	_	_	_	_
Urease	0.75***	$-0.82^{***}$	0.63***	0.76***	0.29*	_	_	_
Protease	0.49***	-0.38*	0.49***	0.65***	0.51***	0.54***	_	_
Bacteria	0.17NS	0.36*	0.08NS	0.27*	0.03NS	-0.07NS	0.22NS	_
Fungi	-0.02NS	0.09NS	0.24*	0.05NS	0.47***	-0.09NS	0.35*	0.21NS

Table 2. Correlation between enzyme activities and physical, chemical and biological properties of soil in different management systems.

Note: SOM, soil organic matter; MBC, microbial biomass carbon; Nitrif., potential nitrification. NS, not significant at p = 0.05; \*p < 0.05; \*\*\*p < 0.001. Calculated with the means of six replicates of each treatment. N = 72.

2.5 times at the two different depths (Acosta-Martínez et al. 2008). The decline in the MBC for the NT was 23% compared with the CT and thus lower than that observed in temperate climates, which was 32% between the different tillage practices (Aslam et al. 1999).

Several authors have indicated that enzyme activity decreases when virgin soils are cultivated (Islam and Weil 2000; Saviozzi et al. 2001; Dilly et al. 2003). The average potential nitrification as well as the urease and the protease activities were higher in the forest soil, which indicated that there was a decrease in activity with the soil use. Saviozzi et al. (2001) and Singh and Kumar (2008) also reported this decrease in urease activity in natural systems compared with agricultural systems. Similar results were obtained in this study, which showed that enzyme activities and potential nitrification decreased from forest to agricultural soils. Similarly, the influence of organic matter was relevant to these results, and there was a positive correlation ( $r = 0.97^{**}$ ) between urease activity and organic matter. The effect of organic matter on urease activity was expected because ureolytic microorganisms are dependent on C as a source for growth (Bezbaruah 1983). The correlation between the MBC and ureolytic activity ( $r = 0.76^{***}$ ; Table 2) shows this relationship.

According to Ball et al. (2008), the protease activity was significantly greater under no-tillage than under plowing. In this study, protease activity is related to the content of the total N (0.49\*\*\*), which predominated in the forest and CT soils. Nitrogen occurs in a wide range of compounds in organic matter, but most of the total N is in an organic form; 40% of total soil N is organic and found in proteins and peptides (Schulten and Schnitzer 1998; Sylvia et al. 2005), and the proteases catalyze the conversion of organic nitrogen into ammonia (NH<sub>3</sub>) or ammonium (NH<sub>4</sub><sup>+</sup>). Therefore, protease synthesis by soil microorganisms depends on the availability of N (Geisseler and William 2008). A significant correlation between the proteolytic activities determined for the microbial biomass C ( $r=0.65^{***}$ ) and the total N ( $r=0.49^{***}$ ) confirm this dependence (Table 2).

The stimulation of nitrification activity of the forest soil in relation to the NT and CT soils has been observed by various authors (Doran et al. 1998; Cookson et al. 2007). In this study, the nitrification activity was higher (p < 0.05) in the NT-I soil than in the other soils. Possibly, soil nitrification activity stimulation in the NT-I was due to animal excretions, which are rich in nitrogen compounds. It has been

estimated that, on average, one excretion is equivalent to 1200 g of manure and 200 mL of urine (Allen et al. 1996). According to Whitehead (2000), the relation C:N in cattle manure and urine is  $\sim 20:1$  and 3.9:1, respectively. A low C:N relationship can stimulate mineralization and affect nutrient availability in the soil. The nitrification activity can be considered as the final stage of the mineralization of organic nitrogen compounds by microorganisms, producing nitrate that is assimilated by plants (Schimel and Bennett 2004). Therefore, the influence of the organic matter content in nitrification activities was expected and was confirmed by a positive correlation between these parameters ( $r = 0.29^*$ ). There was a significant correlation between the potential nitrification and the urease  $(r = 0.29^*)$  and protease  $(r=0.51^{***})$  activities (Table 2), which suggests that part of the NH<sub>4</sub><sup>+</sup> produced in the mineralization of the organic matter came from the decomposition of urine by ureolytics, as well as proteins and peptides by proteolytic microorganisms. Significant positive correlations were found between the gross N mineralization rate and the soil microbial biomass, protease, deaminase and urease activities (Cameron 1999). In acidic soils, as in this study, the predominant form is  $NH_4^+$ , which is oxidized by autotrophic bacteria producing NO<sub>3</sub><sup>-</sup>. In addition, the subsequent nitrification of the NH4<sup>+</sup> produced by these reactions is necessary because the  $NH_4^+$  represses urease (McCarty et al. 1992).

#### Conclusions

The forest soil, which was used as a control, generally had the highest soil quality when compared with the other systems. However, the agricultural–pastoral system provided the best soil quality when compared with other tillage systems studied. The nitrification potential of the NT-I soil was similar to that of forest soil, and only urease activity was lower than in the forest soil. The organic matter content and proteolytic activity were similar in the NT and NT-I soils. The fungal count (0–10 cm layer) of the NT-I soil was only lower than in the forest soil. Therefore, the NT-I treatment may be useful in improving soil conservation and quality, and it may be more economical to have both agricultural production and livestock in the same area.

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