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DIVISÃO 2 - PROCESSOS E PROPRIEDADES DO SOLO

Comissão 2.1 - Biologia do solo

MICROBIAL CHARACTERISTICS OF SOILS UNDER AN INTEGRATED CROP-LIVESTOCK SYSTEM

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ABSTRACT

Integrated crop-livestock systems (ICLs) are a viable strategy for the recovery and maintenance of soil characteristics. In the present study, an ICL experiment was conducted by the Instituto Agronômico do Paraná in the municipality of Xambre, Parana (PR), Brazil, to evaluate the effects of various grazing intensities. The objective of the present study was to quantify the levels of microbial biomass carbon (MBC) and soil enzymatic activity in an ICL of soybean (summer) and *Brachiaria ruziziensis* (winter), with *B. ruziziensis* subjected to various grazing intensities. Treatments consisted of varying pasture heights and grazing intensities (GI): 10, 20, 30, and 40 cm (GI-10, GI-20, GI-30, and GI-40, respectively) and a no grazing (NG) control. The microbial characteristics analysed were MBC, microbial respiration (MR), metabolic quotient (qCO_2), the activities of acid phosphatase, β-glucosidase, arylsuphatase, and cellulase, and fluorescein diacetate (FDA) hydrolysis. Following the second grazing cycle, the GI-20 treatment (20-cm - moderate) grazing intensity) contained the highest MBC concentrations and lowest qCO_2 concentrations. Following the second soybean cycle, the treatment with the highest grazing intensity (GI-10) contained the lowest MBC concentration. Soil MBC concentrations in the pasture were favoured by the introduction of animals to the system. High grazing intensity (10-cm pasture height) during the pasture cycle may cause a decrease in soil MBC and have a negative effect on the microbial biomass during the succeeding crop. Of all the enzymes analyzed, only arylsuphatase and cellulase activities were altered by ICL management, with differences between the moderate grazing intensity (GI-20) and no grazing (NG) treatments.

Keywords: microbial carbon, microbial respiration, acid phosphatase, β-glucosidase, arylsuphatase, fluorescein diacetate hydrolysis.

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RESUMO: *ATRIBUTOS MICROBIOLÓGICOS DO SOLO EM SISTEMA DE INTEGRAÇÃO LAVOURA-PECUÁRIA*

O sistema de integração lavoura-pecuária (ILP) tem se evidenciado como alternativa viável para a recuperação e manutenção das características do solo. Este estudo foi desenvolvido em um experimento de ILP conduzido pelo Instituto Agronômico do Paraná no município de Xambrê, PR, com diferentes intensidades de pastejo. O objetivo deste trabalho foi avaliar o carbono da biomassa microbiana (CBM) e a atividade enzimática no solo, em sistema de integração lavoura-pecuária com soja cultivada no verão e Brachiaria ruziziensis no inverno, sendo esta submetida a diferentes intensidades de pastejo. Os tratamentos constaram de diferentes alturas de pasto e intensidades de pastejo: 10; 20; 30; e 40 cm (IP-10, IP-20, IP-30 e IP-40, respectivamente) e uma área sem pastejo (SP). Os atributos microbiológicos analisados foram CBM, respiração microbiana (RM), quociente metabólico (qCO_2), atividade das enzimas fosfatase ácida, β-glucosidase, arilsulfatase, celulase e hidrólise do diacetato de fluoresceína (FDA). Após o segundo ciclo da pastagem, o tratamento IP-20 (intensidade moderada de pastejo 20 cm) apresentou os maiores teores de CBM e os menores de qCO_2. Após o segundo ciclo da soja, o tratamento com maior intensidade de pastejo IP-10 demonstrou o menor teor de CBM. Os teores de CBM do solo na pastagem foram favorecidos pela inserção dos animais no sistema. A alta intensidade de pastejo (10 cm de altura da pastagem) durante o ciclo da pastagem pode provocar redução no C microbiano do solo, com efeito negativo sobre a mesma na cultura sucessora. Entre as enzimas avaliadas, somente a arilsulfatase e celulase foram sensíveis para avaliar o manejo ILP, com diferenças entre os tratamentos com intensidade moderada de pastejo (IP-20) e a área sem pastejo.

Palavras-chave: carbono microbiano, respiração microbiana, fosfatase ácida, β-glucosidase, arilsulfatase, hidrólise do diacetato de fluoresceína.

INTRODUCTION

Adequate management of agricultural soils is a primary concern for sustainable agriculture because the production system significantly affects the physical, chemical, and biological properties of a soil. Therefore, the current concept of soil quality advocates for a balance between these properties (Doran, 1980; Dick, 1994). Integrated crop-livestock systems (ICLs) alternately manage cultivated areas for pasture and annual crops, with the goal of producing meat and/or milk and grain within the same area (Anghinoni et al., 2011), and provide an alternative production system that allows for the maintenance and/or the recovery of the balance amongst soil characteristics, with good yields.

The basic principle of ICL is the promotion of nutrient cycling, i.e., the use of nutrients originating from the decomposition of plant or animal residues that are produced and remain in the area, for the production of pasture or grain (Assmann et al., 2008). Grazing stimulates a wide variety of soil organisms involved in organic matter decomposition, which release or mineralise N, P, S, and other nutrients originating from plant residues and animal faeces (Wakelin et al., 2009). Management of animals, pastures, and agricultural crops within a single area therefore affects the interactions amongst the chemical, physical, and biological soil components and the transformations involved in nutrient cycling (Assmann et al., 2008).

Soil microbial biomass represents a living and highly active portion of soil organic matter that is significantly affected by soil conditions (Kaschuk et al., 2009). However, although biomass is a measure of the living soil population and, consequently, a very dynamic soil characteristic, it provides little information when examined alone (Moreira and Siqueira, 2006). Soil enzymes also play a fundamental role in chemical reactions, acting as catalysts of several reactions, including those involved in the decomposition of organic wastes, nutrient cycling, the formation of organic matter, and the formation of soil structure; and the analysis of enzyme activities allows for the assessment of aspects of soil microbiology components and contribute to studies of the effects of various soil management practices (Balota et al., 2014).

The objective of the present study was to evaluate microbial biomass C and soil enzyme activity in an integrated crop-livestock system, with soybean grown in the summer and *Brachiaria ruziziensis* in the winter, with *B. ruziziensis* subjected to different grazing intensities.

MATERIAL AND METHODS

The present study was performed at the experimental station of the Instituto Agronômico do Paraná- IAPAR (Agronomy Institute of Parana), located in the municipality of Xambre, at 23º 44’ 10” S and 3º 29' 24" W, and altitude of 418 m. The soil at the experimental station is classified as a Latossolo Vermelho distrófico típico according to Embrapa (2013) [Oxisol], with 15 % clay, 5 % silt, and 80 % sand. Climate in the region is classified as Cfa, subtropical, according to the Köppen climate classification (Caviglione et al., 2000). The temperature and rainfall for the region during the study period, recorded at the IAPAR meteorological station, are shown in figure 1.

Before beginning the experiment, the area was cultivated for three crop seasons (2006/2007, 2007/2008, and 2008/2009), with soybean (*Glycine max*) grown in the summer and oat (*Avena sativa*) in the winter, without any animal grazing. In May 2010, the area was prepared, divided into 15 1.0-ha experimental plots, and *Braquiaria ruziziensis* pastures were established. Surface fertilisation with 60 kg ha^{-1} P_2O_5 as simple superphosphate and 50 kg ha^{-1} N as urea was performed at 25 and at 50 days following planting. Grazing animals were introduced in May 2010 when the forage reached a mean height of 30 cm. Animals were removed in September 2010; the pasture was then desiccated, and soybean was sown with no tillage in October 2010. Surface fertilisation with 60 kg ha^{-1} K_2O and 60 kg ha^{-1} P_2O_5 as simple superphosphate was performed at sowing. The same fertilisation program was performed for both pasture and soybean in 2011.

The treatments consisted of four grazing intensities (GI), GI-10, GI-20, GI-30, and GI-40, which corresponded to pasture heights of 10, 20, 30, and 40 cm after animal grazing, respectively, and an area with no grazing (NG). Treatments were distributed in a randomised block experimental design, with three replicates. The various pasture heights were achieved by varying stocking rates, adjusted weekly by the introduction or removal of regulating animals to maintain the pasture height as close as possible to the desired height (Aguinaga et al., 2006). The grazing method adopted was continuous grazing with a variable stocking rate, according to the put-and-take method (Mott and Lucas, 1952), with a fixed number of two experimental animals per plot and a variable number of regulating animals. The number of animals per plot for the various treatments during grazing are listed in table 1.

Soil was sampled from the 0-10 cm layer in October 2011, following the second cycle of pasture cultivation, and March 2012, following the second cycle of soybean cultivation, from 10 locations per plot to form a representative composite sample for each treatment. At the Soil Microbiology Laboratory of IAPAR, the samples were homogenised, sieved through a 4-mm mesh sieve, and stored at 7 ± 3 °C until analysis. Soil moisture was determined gravimetrically, by drying the samples in a laboratory oven at 100 °C for 24 h. Soil chemical characterisation (Pavan et al., 1992) was performed following the pasture and soybean cycles (Table 2).

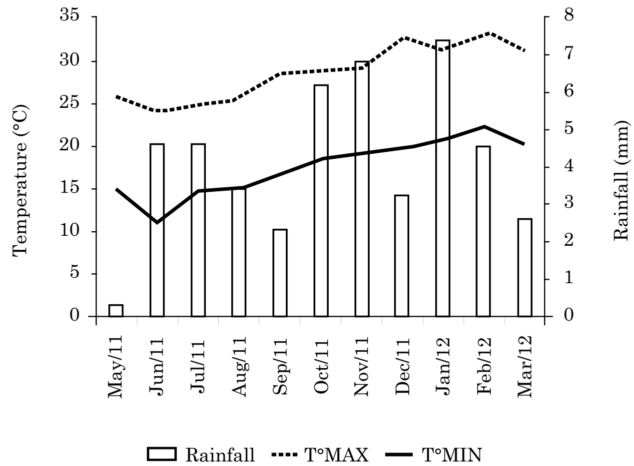

Figure 1. Rainfall and maximum and minimum temperatures recorded from May 2011 to March 2012 at the experimental station of the IAPAR at Umuarama, near Xambre, Parana, Brazil, where the crop-livestock integration experiment was conducted.

Microbial biomass C (MBC) was determined by the fumigation-extraction method (Vance et al., 1987). A conversion factor of Kc = 0.33 was used for the calculation (Sparling and West, 1988).

Microbial respiration (MR) was determined by incubating 50 g of soil in a hermetically sealed jar that also contained a beaker with 10 mL of 0.5 mol L^{-1} NaOH to trap the released CO_2 (Alef, 1995). After 7 days, the amount of remaining 0.5 mol L^{-1} NaOH was quantified by titration using 0.5 mol L^{-1} HCl and the indicator phenolphthalein. The metabolic quotient (qCO_2) was determined as the C-CO_2/MBC ratio, according to Anderson and Domsch (1990).

Arylsulfatase (EC 3.1.6.1), acid phosphatase (EC 3.1.3), and β-glucosidase (EC 3.2.1.21) activities, expressed in μg *p*-nitrophenol h^{-1} g^{-1}, were determined according to Tabatabai (1994). Cellulase (EC 3.2.1.4) activity, expressed in μg AR g^{-1}, was determined according to Schinner and von Mersi (1990). Microbial activity was determined by the fluorescein diacetate hydrolysis method and expressed in gram dry soil (μg F g^{-1}), as described by Schnurer and Rossawall (1982).

Results were subjected to analysis of variance, followed by the Tukey test ($p<0.05$). The analyses were performed using the Statistic Analytical System software (SAS, 1996).

Table 1. Stocking rates and number of cattle per plot for various grazing intensity (GI) treatments applied to obtain corresponding *Brachiaria ruziziensis* pasture heights during the grazing period (May/2011 to September/2011) in a crop-livestock integration experiment performed at the experimental station of the IAPAR

Mean stocking rate	Grazing intensity			
	GI-10	GI-20	GI-30	GI-40
Stocking rate (AU ha^{-1})(1)	2.6	2.4	1.9	1.7
Number of cows (unit ha^{-1})	5.7	5.3	4.0	4.0

(1) 450 kg live weight. GI was determined based on the height of the pasture remaining after grazing, corresponding to heights of 10, 20, 30, and 40 cm for the GI-10, 20, 30, and 40, treatments, respectively.

Table 2. Chemical characteristics of the 0-10 cm depth layer of a Latossolo Vermelho distrófico típico under an integrated crop-livestock system with soybean and pasture in succession, for various grazing intensities (GI) performed at the experimental station of the IAPAR

GI(1)	P	C	pH($CaCl_2$)	Al^{3+}	Ca^{2+}	K^+	Mg^{2+}	SB	CEC
	mg dm^{-3}	g dm^{-3}		cmol$_c$ dm^{-3}					
				After pasture cycle					
GI-10	31.63	7.63	4.63	0.14	1.08	0.20	0.52	1.80	5.97
GI-20	26.93	7.71	4.67	0.14	1.13	0.20	0.50	1.84	5.91
GI-30	26.27	7.74	4.63	0.17	0.99	0.21	0.49	1.69	5.76
GI-40	28.70	8.02	4.60	0.18	0.96	0.20	0.43	1.60	5.67
NG	30.73	8.02	4.67	0.17	0.92	0.20	0.46	1.59	5.66
				After soybean cycle					
GI-10	31.10	9.26	4.70	0.08	1.50	0.13	0.67	2.30	5.90
GI-20	33.33	9.54	4.63	0.09	1.38	0.13	0.57	2.08	5.76
GI-30	34.30	9.50	4.70	0.07	1.29	0.13	0.59	2.00	5.68
GI-40	34.70	9.22	4.70	0.08	1.37	0.13	0.57	2.07	5.76
NG	27.07	8.72	4.67	0.07	1.26	0.12	0.54	1.93	5.52

(1) Determined based on the height of the pasture remaining after grazing, corresponding to heights of 10, 20, 30, and 40 cm for the GI-10, -20, -30, and -40 treatments, respectively. NG - no grazing. P and K extracted by Mehlich-1; C extracted by Walkley-Black method; Ca, Mg and Al extracted by KCl 1 mol L^{-1}. SB: sum of bases and CEC: cation exchange capacity. Values are means of three replicates.

RESULTS AND DISCUSSION

Microbial biomass carbon (MBC) varied amongst treatments (GI-10, GI-20, GI-30 and GI-40) that had different pasture heights (10, 20, 30, and 40 cm, respectively) and the area without grazing (NG), following the pasture and the soybean cycles (Table 3). Following grazing, treatment GI-20 had a significantly greater ($p<0.05$) MBC (68.75 μg C g^{-1}) than the other treatments. The MBC for treatment GI-10 was 47.56 μg C g^{-1}, which was significantly greater than for treatment NG (26.51 μg C g^{-1}). Souza et al. (2010) also observed a higher MBC for a moderate grazing intensity (20 cm) compared to the other intensities studied (10, 30, and 40 cm, and an area without grazing) for a Latossolo Vermelho distroférrico (Oxisol) in southern Brazil after seven years of soybean/pasture rotation. At moderate grazing intensities (20 cm), there is a significant addition of shoot waste from Brachiaria ruziziensis pasture, which stimulates microbial biomass as a result of the release of organic substances such as exudates, mucilages, and secretions by plants in association with the constant renewal of the dense root system, increasing nutrient availability for the soil microbiota (Tisdall and Oades, 1982). The presence of grazing animals in the area also plays an important role in soil microbial ecology, through a series of specific factors associated with the presence of animals, such as the deposition of urine and faeces (Clegg, 2006).

ICLs with different grazing intensities deposit varying amounts of plant waste and animal wastes in various distribution patterns in the area. Under moderate grazing intensities (20 cm), the pasture offer is higher and animals remain at the same location for a longer period of time; animal waste thus becomes concentrated at certain locations

(Baggio, 2007). If the forage offer is low, such as in treatment GI-10, animals walk farther to acquire the feed that they require (Baggio, 2007), distributing waste amongst several locations throughout the area. This results in different distributions of labile organic material amongst treatments, which may affect microbial activity.

For pastures, the treatment with the highest grazing intensity (GI-10) contained a greater MBC than treatment NG, though the MBC of GI-10 did not differ significantly from the treatments with moderate grazing intensity (30 and 40 cm). However, it should be noted that temperatures were mild during pasture cultivation and grazing (Figure 1). Similar results were reported by Souza et al. (2010), who observed no differences amongst treatments GI-10, GI-30, and GI-40 in situations without stress. At high grazing intensities, there is a greater initial accumulation of C in the system because of the higher addition of animal waste and constant renewal of the pasture with intensive grazing (Souza et al., 2009). Because the MBC is affected by the availability of minerals and soil organic C, the accumulation of wastes in the soil increases the availability of organic matter and stimulates the microbial biomass. However, the period of establishment of the system may be a determining factor. Souza et al. (2009) observed C losses at a high grazing intensity (10 cm), and C increases at moderate grazing intensities (20 and 40 cm) or with no grazing following three years of ICL with no tillage.

Following the soybean cycle, the MBC of treatment GI-40 was greater than those of the GI-10 and GI-20 treatments, and did not differ from those of GI-30 and NG (Table 3). In contrast with the results observed following the pasture cycle, the treatment with the highest grazing intensity (GI-10) had significantly ($p<0.05$) lower MBC than the remaining treatments. In contrast with the pasture cycle, higher soil temperatures were observed during the soybean cycle (Figure 1). Coupled with the low level of soil protection caused by the high grazing intensity in treatment GI-10, this resulted in a further increase in soil temperature and decrease in soil moisture and, consequently, a decrease in microbial biomass. Thus, the treatment with the highest pasture height and lowest grazing intensity (GI-40) most likely resulted in a decreased water loss and a lower soil temperature, favouring a greater microbial biomass. This result is in accordance with Mercante et al. (2008), who evaluated various management systems involving varying cover intensities, and Souza et al. 2010, who examined conditions similar to the present study. A decrease in MBC under the highest grazing intensity was also observed by Souza et al. (2010) under water stress conditions.

High grazing intensities applied during pasture cycles decrease the amount of plant waste deposited at the soil surface for the succeeding

Table 3. Microbial biomass carbon in dry soil (MBC), microbial respiration (MR), and metabolic coefficient (qCO_2) of the 0-10 cm layer of a under an integrated crop-livestock system with soybean and pasture in succession, for various grazing intensities (GI)

GI(1)	MBC	MR	qCO_2
	μg C g^{-1}	μg C-CO_2 g^{-1} d^{-1}	μg C-CO_2 μg^{-1} MBC h^{-1}
		After pasture cycle	
GI-10	47.56 b	0.28 a	6.01 ab
GI-20	68.75 a	0.25 a	5.18 b
GI-30	40.51 bc	0.33 a	8.20 ab
GI-40	39.31 bc	0.31 a	7.85 ab
NG	26.51 c	0.26 a	9.88 a
CV (%)	13.97	20.46	25.35
		After soybean cycle	
GI-10	49.74 c	0.16 a	3.27 a
GI-20	72.60 b	0.21 a	2.92 a
GI-30	79.20 ab	0.20 a	2.51 a
GI-40	96.12 a	0.27 a	2.76 a
NG	80.95 ab	0.21 a	2.53 a
CV (%)	10.53	45.48	29.55

(1) Determined based on the height of the pasture remaining after grazing, corresponding to heights of 10, 20, 30, and 40 cm for the GI-10, -20, -30, and -40 treatments, respectively. NG - no grazing. Means followed by the same letter did not differ significantly according to the Tukey test ($p<0.05$).

crop (Chavéz et al., 2011). According to Assmann et al. (2008), 2,000 kg ha^{-1} dry mater, i.e., an approximately 15-20 cm height for oat and/or rye grass pastures, are required for good maintenance of soil organic matter as well as no tillage management to avoid damage to the system. For soils with low clay concentrations, such as the present study, 9,000 kg ha^{-1} dry matter are required to maintain the initial soil C stock (Vieira, 2007).

After the pasture cycle, the MBC of treatment GI-20 was 159 % greater than that of treatment NG. Treatments GI-10, GI-30, and GI-40 also contained 79, 53, and 48 % greater MBC, respectively, than treatment NG, although these differences were not significant (Figure 2). Following the soybean cycle, only the treatment with the highest grazing height (GI-40) had a greater MBC (18 %) than the NG treatment. The MBC was 38, 10, and 2 % lower for treatments GI-10, GI-20, and GI-30, respectively, compared to treatment NG (Figure 2).

There were no significant differences ($p<0.05$) amongst treatments in microbial respiration (MR) following the pasture or soybean cycles (Table 3). D'Andréa et al. (2002) also did not observe significant differences in microbial respiration for the 0-10 cm layer in a study evaluating various management systems, including Brachiaria decumbens pasture, under no tillage and conventional tillage, in southern Goias, Brazil. These authors attributed the absence of differences amongst treatments to the recent establishment of the no tillage system (between four and five years), during which time the microbial biomass could still be adapting to the new soil conditions. In the present study, the recent establishment of the ICL system (two years) may also explain the absence of differences amongst treatments. However, the differences in climate between the Cerrado (Brazilian tropical savanna) and southern Brazil should be considered because they directly influence the C dynamics and activity of soil microorganisms. Given that high MR values were observed for no tillage treatments in studies performed in the South of Brazil (Balota et al., 1998; Cervantes, 2012), higher MR values and, therefore, differences amongst treatments may occur over time.

A significant difference in qCO_2 ($p<0.05$) was observed between the GI-20 and NG treatments after the pasture cycle. For treatment GI-20, the qCO_2 was 1.64 times lower compared to the values in the for the area without grazing (Table 3). After soybean cultivation, there was no significant difference in the qCO_2 amongst treatments. Mercante et al. (2008) and Carneiro et al. (2009) reported that the amount of C that is lost as CO_2 through respiration decreases as the qCO_2 decreases, reflecting the more efficient use of organic compounds by the microbial biomass and incorporation of a greater proportion of C into microbial tissues. In this regard, soils with low qCO_2

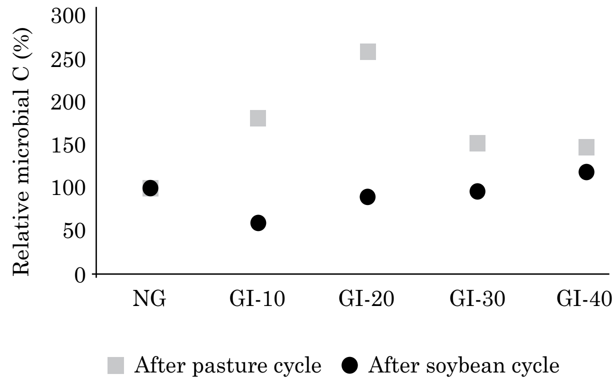

Figure 2. Relative microbial carbon (GI × 100/NG, for each grazing treatment and cultivation cycle) in the 0-10 cm soil layer of a Latossolo Vermelho distrófico típico under an integrated crop-livestock system at various grazing intensities, performed at the experimental station of IAPAR.

are close to equilibrium, given that higher values are found in stressed environments, where there is higher C consumption for maintenance of microbial biomass. Carneiro et al. (2009) observed lower qCO_2 values for integrated crop-livestock areas relative to a native Cerrado area. A similar result was reported by Souza et al. (2010) for a long-term experiment with various grazing intensities. In the GI-20 treatment, as previously mentioned, an abundance of easily decomposable material is created, resulting in a high efficiency of microorganisms for the conversion of the deposited organic wastes. In the area without grazing, the plant residues are more evenly distributed and less decomposed by the microorganisms relative to the grazing areas.

Following the pasture cycle, significant differences in enzyme activity amongst treatments ($p<0.05$) were only observed for cellulase activity (Table 4). The lowest activity was observed in the area without grazing (NG), which was 6.47 μg AR g^{-1} of the value of the remaining treatments (mean 11.85 μg AR g^{-1}). The higher cellulase activity after the pasture cycle observed in treatments GI-10, GI-20, GI-30, and GI-40 compared to treatment NG may be attributed to the grass species planted and the presence of animals, which increases recycling by depositing wastes, possibly increasing inputs of a cellulose-rich substrate into the agricultural system.

After the soybean cycle, only the arylsuphatase activities of the GI-30 and NG treatments were significantly different ($p<0.05$). Arylsulphatase activity was higher for GI-30 (4.74 μg PNP g^{-1} h^{-1}) and NG (4.47 μg PNP g^{-1} h^{-1}), and lower for GI-10 (2.51 μg PNP g^{-1} h^{-1}) (Table 4). Nogueira and Melo (2003) reported a positive correlation between arylsuphatase activity and total S concentrations in soil, which is mostly present in organic matter.

Thus, greater enzymatic activity is typically related to higher organic C concentrations, which reflect the higher availability of substrates upon which enzymes can act. The observed arylsuphatase activities were similar to those reported by Balota et al. (2014) for a Latossolo Vermelho distrófico (Oxisol), with concentrations ranging from 2, 3, and 15 μg PNP g^{-1} h^{-1} for clayey soils.

The acid phosphatase activities were considerably lower than previous reports (Cervantes, 2012; Balota et al., 2014), and ranged from 275.34 to 293.01 μg PNP g^{-1} h^{-1} following the pasture cycle and from 109.22 to 149.14 μg PNP g^{-1} h^{-1} following the soybean cycle. These results may be related to the application of P_2O_5 because acid phosphatase is produced when soluble P concentrations reach levels that limit the growth of plants and soil microorganisms (Trannin et al., 2007). Phosphate fertilisation increases soil P concentrations and may, therefore, decrease soil phosphatase activity (Tabatabai, 1994). Similar results were reported by Carneiro et al. (2004) for the Cerrado and Conte et al. (2002) for the South of Brazil. However, crop rotation studies in Ponta Grossa, Parana, Brazil, produced high acid phosphatase values, ranging from 799.1 to 489.9 μg PNP g^{-1} h^{-1}. This finding was attributed to the localised application of phosphate fertilisers in the planting furrows, as P is easily absorbed within colloids and Fe and Al oxides in the soil, which decreases its solubility and effect of inhibiting phosphatase activity (Cervantes, 2012). Furthermore, even with the addition of phosphate fertilisers to the soil, organic P originating from soil organic matter may become available to the plants through acid phosphatase activity. This process indicates that, even in cultivated areas, microorganisms and exoenzymes play a role in the availability of P to plants.

There were no significant differences ($p<0.05$) in ß-glucosidase or FDA activities amongst treatments for either sampling time (Table 4). In the case of ß-glucosidase, this observation may be related to the complexity of the wastes in integrated crop-livestock areas in which grass, legumes, and animals are present because this enzyme acts on less complex substrates (Matsuoka et al., 2003). Silveira (2007) studied 11 soils from cultivated and native areas in Rio Grande do Sul, Brazil, and did not observe differences in FDA activity either, indicating that FDA hydrolysis was not sensitive to differences amongst the soils studied.

After two pasture cycles of various grazing intensities and two soybean cycles, moderate grazing intensities (GI-20, GI-30, and GI-40) were observed to increase MBC concentrations with a higher efficiency by microorganisms in converting organic wastes into microbial biomass. Under the highest grazing intensity (GI-10), the MBC concentrations decreased. ICL systems with no tillage and varying pasture management intensities will, therefore, over time result in varying amounts of additions of plant

Table 4. Arylsulphatase, acid phosphatase, β-glucosidase, and cellulase and fluorescein diacetate hydrolysis activities in the 0-10 cm layer of a Latossolo Vermelho distrófico típico under an integrated crop-livestock system with soybean and pasture in succession, for various grazing intensities (GI)

GI(1)	Arylsulphatase	Acid phosphatase	β-glucosidase	Cellulase	FDA
		μg PNP g^{-1} h^{-1}		μg AR g^{-1}	μg F g^{-1}
			After pasture cycle		
GI-10	9.51 a	282.77 a	47.59 a	12.02 a	59.86 a
GI-20	8.81 a	275.34 a	41.26 a	11.40 ab	70.34 a
GI-30	10.49 a	287.30 a	46.36 a	11.68 a	71.34 a
GI-40	9.23 a	293.01 a	43.08 a	12.09 a	53.81 a
NG	9.70 a	289.35 a	47.63 a	6.47 b	53.09 a
CV (%)	18.87	9.31	17.12	17.03	16.27
			After soybean cycle		
GI-10	2.51 c	109.22 a	26.24 a	8.69 a	111.20 a
GI-20	3.41 bc	141.82 a	32.17 a	7.85 a	115.10 a
GI-30	4.74 a	138.18 a	31.12 a	8.94 a	122.81 a
GI-40	3.25 bc	149.14 a	32.13 a	6.73 a	118.19 a
NG	4.47 ab	142.02 a	33.16 a	8.96 a	121.52 a
CV (%)	12.28	14.37	12.27	28.43	6.37

(1) Determined based on the height of the pasture remaining after grazing, corresponding to heights of 10, 20, 30, and 40 cm for the GI-10, -20, -30, and -40 treatments, respectively. NG - no grazing. Means followed by the same letter did not differ significantly according to the Tukey test ($p<0.05$).

and animal wastes to the soil. This relationship allows for the selection of a set of variables related to nutrient cycling that indicate soil quality. Of the microbiological indicators examined, MBC, qCO_2, and arylsuphatase and cellulase activities were the most sensitive.

CONCLUSIONS

Soil MBC concentrations were favoured by the inclusion of animals in the pasture system.

A high grazing intensity (10 cm pasture height) during the pasture cycle may cause a decrease in soil microbial biomass C, with a negative effect on the succeeding crop.

Of the evaluated enzymes, only arylsuphatase and cellulase were sensitive to the ICL management. Differences were observed between treatments with a moderate grazing intensity (GI-20) and no grazing.

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