Vol.54, n. 2: pp. 221-228, March-April 2011 ISSN 1516-8913 Printed in Brazil

## **BRAZILIAN ARCHIVES OF BIOLOGY AND TECHNOLOGY**

# INTERNATIONAL JOURNAL

# Effect of Tillage Systems and Permanent Groundcover **Intercropped with Orange Trees on Soil Enzyme Activities**

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## **ABSTRACT**

The objective of this study was to evaluate the effect of different soil tillage systems and groundcover crops intercropped with orange trees on soil enzyme activities. The experiment was performed in an Ultisol soil in northwestern Paraná State. Two soil tillage systems were evaluated [conventional tillage (CT) across the entire area and strip tillage (ST) with a 2-m strip width] in combination with various groundcover vegetation management systems. Soil samples were collected after five years of experimental management at a depth of 0-15 cm under the tree canopy and in the inter-row space in the following treatments: (1) CT-Calopogonium mucunoides; (2) CT-Arachis pintoi; (3) CT-Bahiagrass; (4) CT-Brachiaria humidicola; and (5) ST-B. humidicola. The soil tillage systems and groundcover crops influenced the soil enzyme activities both under the tree canopy and in the inter-row space. The cultivation of B. humidicola provided higher amylase, arylsulfatase, acid phosphatase and alkaline phosphatase than other groundcover species. Strip tillage increased enzyme activities compared to the conventional tillage system.

**Key words:** arylsulfatase, acid phosphatase, tillage systems, vegetative cover

#### INTRODUCTION

Most Brazilian citrus orchards have been established using conventional soil tillage for incorporation with subsequent maintenance of a clean soil surface because the permanent intercropped vegetative cover could compete with citrus trees for soil water. However, this soil management practice promotes erosive processes, especially in the initial period of orchard establishment (Corá et al., 2005). The use of groundcover vegetation between the perennial crops is indispensable to minimize the soil erosion, especially because about 50% of citrus orchards in Paraná State have been planted in soil derived

from Caiuá sandstone, which has low clay content, low natural fertility and high water erosion potential (Fidalski, et al., 2007; Auler et al., 2008). The use of plants to cover the inter-row space between the perennial crops is one of the most efficient single practices for reducing soil erosion because this process acts upon the origin of the erosive process by reducing the energy of raindrops impacting against the soil surface, thus avoiding the disaggregation of soil particles and facilitating greater water infiltration. This process also improves soil chemical and physical properties (Chaves et al., 1997) and microbial activity (Balota et al., 2004; Rutigliano et al., 2004; Acosta-Martinez et al., 2010). Plant species

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used in such a management system must have the following characteristics: rapid development, high biomass production per unit area, high nutrient content, strong branching, a robust and deep root system and high capacity to mobilize soil nutrients and to fix N<sub>2</sub> biologically.

Results obtained previously have shown the efficiency of superficial limestone used in undisturbed soil (Fidalski and Tormena, 2005) and the use of minimum tillage (strip tillage) in orchard establishment with permanent groundcover between orange trees (Auler et al., 2008). These practices do not have negative effects on soil chemical properties (0-40 cm) or on orange production.

Soil microbial activity plays important roles in nutrient cycling and soil quality. Soil organic decomposition is mediated microorganisms, which produce enzymes that catalyze innumerable reactions necessary for their own metabolic processes, decomposition of organic residues, nutrient cycling, and formation of organic matter and soil structure. Most soil enzymes are produced by microorganisms and are able to accumulate, become inactivated and/or decompose in the soil, with considerable impact on nutrient recycling (Tabatabai, 1994; Dick, 1997). Soil enzyme activity can be used to indicate the intensity of certain biochemical processes because it is considered to be a major contributor to overall soil microbial activity (Badiane et al., 2001). Soil enzymes also have the potential to provide a unique integrative biological assessment of soils due to their relationship to soil biology and their rapid response to changes in soil management (Dick, 1997; Badiane et al., 2001; Ndiaye et al., 2000).

The objective of this study was to evaluate the effect of different species of groundcover crops cultivated between rows of orange trees and soil tillage systems on soil enzyme activities.

## MATERIALS AND METHODS

## **Experimental characteristics and soil sampling**

The experiment was started in 1993 at Alto Paraná, northwestern Paraná State, in a sandy Ultisol soil (90% sand) classified as a Typic Paleudult and derived from Caiuá sandstone, with pH = 3.9 (CaCl<sub>2</sub>), 1.9 mg kg<sup>-1</sup> of phoshorus (Mehlich), and 3.7 g kg<sup>-1</sup> of organic carbon (Walkley and Black) in the surface layer

(0-15 cm). It was installed in an area previously cultivated with pasture (*Brachiaria humidicola*) in a randomized complete block design with three replicates. Each parcel was composed of 15 orange plants arranged in three planting lines of five plants each.

Two soil tillage systems were evaluated, conventional tillage (CT) across the entire area and strip tillage (ST) with a 2-m strip width, in combination with various groundcover vegetation management systems. The citrus cultivar utilized was 'Pera' orange (Citrus sinensis) grafted onto 'Rangpur' lime rootstock. The conventional soil tillage (CT) system were prepared by one disc plowing at 20-cm depth and two light harrowings for the planting of orange seedlings, while the strip tillage (ST) system consisted of soil preparation only within a 2-m strip, while the remainder of the area (5 m) was not disturbed. Each line of orange trees was placed in the center of the trilled strip. In the ST system, only 30% of the soil was turned over, leaving 70% of the area undisturbed retaining its natural vegetative cover. Before soil preparation, two tonnes of dolomitic limestone per hectare was applied to the total area. From 1996 to 1998, 3.0 tonnes of a mix of dolomitic and calcitic (1:1) limestone per year was applied to the surface soil without incorporation under the tree canopy and 1.5 tonnes was applied similarly to the interrow space.

The following treatments were studied: (1) CT and an annual cover crop of the leguminous species *Calopogonium mucunoides*; (2) CT and a perennial cover crop of the leguminous peanut species *Arachis pintoi*; (3) CT and an evergreen cover crop of Bahiagrass *Paspalum notatum*; (4) CT and a cover crop of spontaneous *Brachiaria humidicola*; and (5) ST and maintenance of the remaining *Brachiaria humidicola* grass (pasture). The groundcover vegetation was controlled by mowing as needed. The experimental procedures followed as described by Fidalski et al. (2007) and Auler et al. (2008).

Five sub-samples of soil were taken at a depth of 0-15 cm within each replicate under the tree canopy and in the center of the inter-row space during two seasons, late summer (March) and spring (October), from 1997 to 1998, five years after the beginning of the experiment. Fresh soil samples were sieved through a 4-mm screen to remove the large plant material and stored at 4°C until analysis of microbial and chemical

characteristics. Chemical analyses were performed according to Pavan et al. (1992).

#### **Enzyme Activity Analyses**

Amylase (EC 3.2.1) and cellulase (EC 3.2.1.4) were determined according to Deng and Tabatabai (1994a). The activities of arylsulfatase and acid and alkaline phosphatase were measured according Tabatabai (1994). Arylsulfatase activity (arylsulfate sulfohydrolase, EC 3.1.6.1) was determined using colorimetric determination of pnitrophenol released when soil samples were incubated with *p*-nitrophenyl sulfate. phosphatase activity (EC 3.1.3) was analyzed with a modified universal buffer (MUB) (pH 6.5) using colorimetric determination of p-nitrophenol released when soil samples were incubated with pnitrophenyl phosphate. Activities of arylsulfatase and phosphatase are expressed as µg p-nitrophenol (PNP) g<sup>-1</sup> h<sup>-1</sup>. All enzyme activities were measured using a one-hour incubation period. All the determinations were made in triplicate and expressed on a dry weight basis.

## Statistical analyses

For each treatment, data were averaged over the four seasons before statistical analysis by ANOVA to determine the effects of groundcover cultivation and sample position on the soil properties. The treatment effects were statistically evaluated by the analysis of variance using Tukey's test for mean comparison at  $P \le 0.05$ . The relationships between the soil enzyme activities and chemical properties were analyzed by the Pearson

correlation coefficient (r). Soil enzyme activities for each sample position were also submitted to regression analysis. The significant equation was chosen by F test ( $P \le 0.05$ ) that presented higher correlation coefficient ( $R^2$ ). All statistical analyses were performed using the SAS statistical package version  $9^{th}$  (SAS, 2002).

#### RESULTS AND DISSCUSION

## **Chemical Properties**

The chemical properties of the soils after five years of experimentation due to different groundcover and soil tillage systems are shown in Table 1. The pH under the tree canopy was lower (from 3.8 to 4.0) than in the inter-row space (from 4.6 to 5.1). The acidification under the orange tree canopy was due to various factors, including the high quantity of fertilizer applied, nutrient leaching, and the high quantities of nutrients uptake by orange tree. The soil samples were taken near the edge of the tree canopy, at the site of the fertilizers application, where the acidification processes was higher. After five years, the groundcover cultivation and incorporation provided an increase in the pH of the soil. The organic matter addition to the soil contributed to the reduction of organic anion losses in the system and to an increase in H<sup>+</sup> consumption. The concentration of basic cations in the plant extract has been associated with the effects in alleviating soil acidity (Miyzawa et al., 2002).

**Table 1 -** Soil chemical properties (0-15 cm depth) after a five year period of cropping with permanent groundcover species between the orange trees. Average of twelve samples<sup>1</sup>.

Treatment	p	Н	P (mg	kg <sup>-1</sup> )	Al saturation (%)		
	TC	IRS	TC	IRS	TC	IRS	
CT-Calopogonium	3.9 aB	4.7 aA	28.6 bA	3.1 aB	36.5 bcA	2.7 abB	
CT-Arachis	3.9 aA	4.6 aA	35.2 bA	3.1 aB	46.0 aA	3.6 abB	
CT-Bahiagrass	3.8 aB	4.8 aA	67.7 aA	4.6 aB	39.2 bA	1.8 bB	
CT-Brachiaria	3.8 aB	4.8 aA	69.6 aA	2.6 aB	40.8 bA	3.9 aB	
ST-Brachiaria	4.0 aB	5.1 aA	66.6 aA	2.6 aB	34.0 cA	1.0 bB	
Average	3.9 B	4.8 A	53.5 A	3.2 B	39.3 A	2.6 B	

<sup>T</sup> pH: CaCl<sub>2</sub> 0.01 M; P: Mehlich; CT: Conventional tillage; ST: Strip tillage; TC: Tree Canopy; IRS: Inter-Row Space. Means followed by a different lower case letter within a column of the same sample position and upper case letter between sample position are significantly different by the Tukey test ( $P \le 0.05$ ).

The low Al saturation in the inter-row space can be attributed to the pH increase, which reduces the Al solubility through Al complexation with organic compounds. Several plants have the capacity to alleviate the soil acidity through water-soluble plant organic compounds (Meda et al., 2001). This chemical change is associated with the concentrations of basic cations in the plant extract, the higher the concentration the greater the effects in alleviating soil acidity.

The soil P concentration was higher under the tree canopy than in the inter-row space because P fertilizer was applied onto the soil surface without mechanical incorporation. For this reason, the P content accumulated with time. However, in the center of inter-row space, there was also an increase in the available P content. The CT-Bahiagrass increased about 142% the available P compared to initial content (1.9 mg kg<sup>-1</sup>). This increase might have occurred due to the P cycle through P redistribution into the undisturbed soil by different permanent groundcovers (Franchini et al., 2004).

#### **Enzyme Activity**

The average measurements obtained from the twelve samples (four seasons) indicated that the intercropping of different groundcover species influenced the soil enzyme activities, both under the tree canopy and in the inter-row space. In general, microbial activity in the inter-row space was higher than that observed under the tree canopy (Table 2). Amylase activity varied from 171 to 320 µg g<sup>-1</sup> h<sup>-1</sup> in the soil under the tree canopy and from 257 to 417 µg g<sup>-1</sup> h<sup>-1</sup> in the interrow space (Table 2). The CT-Brachiaria treatment showed higher activity than the other treatments, with an increase of up to 87% compared to CT-Arachis under the tree canopy, while in the interrow space the ST-Brachiaria treatment showed an increase of about 62% compared to CT-Arachis. Cellulase activity varied from 44 to 54 µg g<sup>-1</sup> h<sup>-1</sup>, but did not vary with different groundcover species and sample positions. Amylase and cellulase activities observed were within the ranges reported in the literature; however, they were lower than those observed in high fertility soil (from 350 to 830 µg g<sup>-1</sup> for amylase and from 67 to 220 µg g<sup>-1</sup> for cellulose) in Brazil (Balota et al., 2004). These enzymes play important roles in nutrient mineralization because they participate actively in residue decomposition, providing

available readily C for the growth microorganisms (Deng and Tabatabai, 1994b). Cellulose is the most abundant compound in the biosphere, making up almost half of the biomass synthesized by the photosynthetic fixation of  $CO_2$ . Mulching generally increases enzyme activities in the soils. With an increased supply of mulch, there would be an increase in the supply of readily available substrates, such as carbohydrates for the microorganisms. This leads to increase the glycosidase activity, because these enzymes play a major role in the degradation of carbohydrates in the soils (Deng and Tabatabai, 1996).

Arylsulfatase activity varied from 1.4 to 3.6  $\mu g$  PNP  $g^{-1}$   $h^{-1}$  in the soil under the tree canopy and from 6.3 to 11.6  $\mu g$  PNP  $g^{-1}$   $h^{-1}$  in the inter-row space (Table 2). Under the tree canopy, there were no differences among the groundcover species, while arylsulfatase activity in the inter-row space increased up to 84% in the CT-*Brachiaria* treatment compared to CT-*Calopogonium*.

Arylsulfatase activity levels reported in the literature vary widely (from 1.4 to 113  $\mu g$  g<sup>-1</sup> h<sup>-1</sup>) in many regions of the world (Acosta-Martinez et al., 2010; Silva et al., 2009). In Brazil, reported levels of arylsulfatase activity vary from 4.0 to 113 (Balota et al., 2004; Silva et al., 2009). However, the level of arylsulfatase activity obtained in this study was from 1.5 to 3.0 times lower than that observed in a nearby area of Paraná State under different soil tillage in clayey soil with high natural fertility (Balota et al., 2004).

The higher arylsulfatase activity obtained in the inter-row space compared to under the tree canopy, especially in the *Brachiaria* treatment, confirmed previous observations that mulching could significantly increase the arylsulfatase activity (Deng and Tabatabai, 1997). Large inputs of organic C via plant residues constitute a principal reservoir of sulfate esters, the substrate for this group of enzymes. Arylsulfatases are one of many types of sulfatases involved in the mineralization of ester sulfate. Most arylsulfatases are not constitutive enzymes, and their synthesis by the microorganisms may be controlled by the C and S content of the system (Tabatabai, 1994; Dick, 1997). Thus, arylsulfatase activity is dependent on soil sulfate content and nutrient content.

Acid phosphatase activity varied from 87 to 176 µg PNP g<sup>-1</sup> h<sup>-1</sup>, while alkaline phosphatase activity varied from 13.9 to 40.8 µg PNP g<sup>-1</sup> h<sup>-1</sup> (Table 2).

the inter-row space		•								py und m
	Enzyme Activity (µg g <sup>-1</sup> soil)									
Groundcover – Species –	Amy	lase	Cell	ulase	Arylsı	ılfatase	Acid	l Phos	Alkalin	e Phos
	TC	IRS	TC	IRS	TC	IRS	TC	IRS	TC	IRS

**Table 2 -** Amylase, cellulase, arylsulfatase and acid and alkaline phosphatase activities under the tree canopy and in

V V (NOB )											
Amylase		Cellulase		Arylsulfatase		Acid Phos		Alkaline Phos			
TC	IRS	TC	IRS	TC	IRS	TC	IRS	TC	IRS		
263 bA	293 bcA	51	52	3.4 aB	6.3 cA	156 aA	135 bA	25.8 bA	17.8 cB		
171 cB	257 cA	46	44	1.4 bB	7.4 bcA	92 cB	132 bA	13.9 cB	25.2 bA		
271 bA	303 bcA	46	47	3.2 aB	8.2 bA	96 cB	146 bA	23.2 bB	37.2 aA		
320 aA	343 bA	53	52	2.5 abB	10.7 aA	115 bcB	164 abA	37.2 aA	38.3 aA		
254 bB	417 aA	53	54	3.6 aB	11.6 aA	132 bB	176 aA	26.9 bB	40.8 aA		
256 B	323 A	50	50	2.8 B	8.8 A	118 B	150 A	25.4 B	31.9 A		
	TC  263 bA  171 cB  271 bA  320 aA  254 bB	TC IRS  263 bA 293 bcA  171 cB 257 cA  271 bA 303 bcA  320 aA 343 bA  254 bB 417 aA	TC IRS TC  263 bA 293 bcA 51  171 cB 257 cA 46  271 bA 303 bcA 46  320 aA 343 bA 53  254 bB 417 aA 53	TC         IRS         TC         IRS           263 bA         293 bcA         51         52           171 cB         257 cA         46         44           271 bA         303 bcA         46         47           320 aA         343 bA         53         52           254 bB         417 aA         53         54	TC         IRS         TC         IRS         TC           263 bA         293 bcA         51         52         3.4 aB           171 cB         257 cA         46         44         1.4 bB           271 bA         303 bcA         46         47         3.2 aB           320 aA         343 bA         53         52         2.5 abB           254 bB         417 aA         53         54         3.6 aB	Cellulase         Arylsulfatase           TC         IRS         TC         IRS           263 bA         293 bcA         51         52         3.4 aB         6.3 cA           171 cB         257 cA         46         44         1.4 bB         7.4 bcA           271 bA         303 bcA         46         47         3.2 aB         8.2 bA           320 aA         343 bA         53         52         2.5 abB         10.7 aA           254 bB         417 aA         53         54         3.6 aB         11.6 aA	Amylase         Cellulase         Arylsulfatase         Acid           TC         IRS         TC         IRS         TC           263 bA         293 bcA         51         52         3.4 aB         6.3 cA         156 aA           171 cB         257 cA         46         44         1.4 bB         7.4 bcA         92 cB           271 bA         303 bcA         46         47         3.2 aB         8.2 bA         96 cB           320 aA         343 bA         53         52         2.5 abB         10.7 aA         115 bcB           254 bB         417 aA         53         54         3.6 aB         11.6 aA         132 bB	Amylase         Cellulase         Arylsulfatase         Acid Phos           TC         IRS         TC         IRS         TC         IRS           263 bA         293 bcA         51         52         3.4 aB         6.3 cA         156 aA         135 bA           171 cB         257 cA         46         44         1.4 bB         7.4 bcA         92 cB         132 bA           271 bA         303 bcA         46         47         3.2 aB         8.2 bA         96 cB         146 bA           320 aA         343 bA         53         52         2.5 abB         10.7 aA         115 bcB         164 abA           254 bB         417 aA         53         54         3.6 aB         11.6 aA         132 bB         176 aA	Amylase         Cellulase         Arylsulfatase         Acid Phos         Alkalin           TC         IRS         TC         IRS         TC         IRS         TC           263 bA         293 bcA         51         52         3.4 aB         6.3 cA         156 aA         135 bA         25.8 bA           171 cB         257 cA         46         44         1.4 bB         7.4 bcA         92 cB         132 bA         13.9 cB           271 bA         303 bcA         46         47         3.2 aB         8.2 bA         96 cB         146 bA         23.2 bB           320 aA         343 bA         53         52         2.5 abB         10.7 aA         115 bcB         164 abA         37.2 aA           254 bB         417 aA         53         54         3.6 aB         11.6 aA         132 bB         176 aA         26.9 bB		

CT: Conventional tillage; ST: Strip tillage. TC: Tree Canopy; IRS: Inter-Row Space. Means followed by different lower case letters within columns and upper case letters between columns are significantly different according to the Tukey test (P≤0.05).

The CT-Calopogonium treatment showed higher acid phosphatase activity (70%) than the CT-Arachis treatment under the tree canopy, while in the inter-row space, *Brachiaria* treatments (both CT and ST) showed 21% higher activity than CT-Bahiagrass. The cultivation of *Brachiaria* also provided higher alkaline phosphatase activity than other groundcover species. However, the largest increases were shown by CT-Brachiaria compared to CT-Arachis (168%) under the tree canopy and by Brachiaria (both CT and ST) compared to CT-Calopogonium (119%) in the inter-row space.

Results for acid phosphatase activity were consistent with those reported in the literature, which varied widely from 77 to 1165 µg g<sup>-1</sup> h<sup>-1</sup> (Rutigliano et al., 2004; Carneiro et al. 2004). However, the phosphatase activity level obtained was from two to five times lower than that observed in another region of Paraná State under different soil tillage in clayey soil with high natural fertility (Balota et al., 2004). In the present study, acid phosphatase activity was about three times higher than alkaline phosphatase activity, which might be due to the acidity of the soil (pH from 3.8 to 5.1). This result confirmed previous findings that phosphatase activity was strongly influenced by the pH of the soil. Acid phosphatase prevails in acid soils and alkaline phosphatase prevails in alkaline soils (Tabatabai, 1994; Balota et al., 2004). According to Tabatabai (1994), either the rate of synthesis and release of phosphatases by the soil microorganisms or the stability of the enzymes are related to soil pH.

Phosphatase is the general name of a broad group of enzymes that catalyze the hydrolysis of both esters and anhydrides of H<sub>3</sub>PO<sub>4</sub>. Microorganisms are the most important sources of phosphatases in the soil due to their high metabolic activity and short lifespan. Acid phosphatases have been studied extensively because of their importance in soil organic P mineralization and plant nutrition and their optimum activity under acidic conditions (Tabatabai, 1994; Dick, 1997).

It is believed that phosphatases are produced when the available P content reaches the critical levels for plant and microorganismal growth (Tabatabai, 1994). For example, natural systems, such as forests, sustain growth without the addition of phosphate fertilizers, even with a low level of available P. In these systems, available P is controlled by the organic P cycling, in which microbial biomass is an essential component. On the other hand, in the agricultural systems, added phosphorus fertilizers may depress phosphatase activity.

The high variation in the soil enzyme activities due to different groundcover crops and soil tillage systems showed that these enzymes were sensitive to soil disturbance. The cultivation of grass increased the soil enzyme activities in the interrow space compared to the cultivation of leguminous species, by about 29% for amylase,

8% for cellulose, 49% for arylsulfatase, 21% for 80% phosphatase and for phosphatase. The large increase under Brachiaria cultivation was likely due to its high aboveground biomass production (up to 35 tonnes ha<sup>-1</sup> year<sup>-1</sup>), which was about three times higher than that of the other groundcover species. This plant residue becomes a substrate for the microbial growth and enzyme production. An increasing supply of mulch increases the supply of readily available substrates, such as carbohydrates, for the microorganisms that produce the majority of soil enzymes. Large additions of groundcover residues increase the soil organic matter. This alteration of the organic carbon pool can also protect the soil enzymes through the association with organic and inorganic colloids, contributing to enzyme stabilization in the soils. Plant roots also stimulate the enzyme activity by creating favorable microhabitats (increased porosity, water and diversity of compounds) for the microorganisms (Dick, 1997).

The increase in soil enzyme activities could be due to the large amount of plant residues produced by the groundcover species. It has been observed that *Arachis pintoi* produces an average of 10 tonnes ha<sup>-1</sup> year<sup>-1</sup> (Perin et al., 2003), while Bahiagrass can produce up to 7.0 tonnes ha<sup>-1</sup> year<sup>-1</sup> of shoot dry matter. *B. humidicola* can produce about 15 tonnes ha<sup>-1</sup> year<sup>-1</sup> of dry matter biomass; however, it has the capacity to produce more than 35 tonnes ha<sup>-1</sup> of dry matter biomass with the application of 120 kg ha<sup>-1</sup> of N (Orioli, 2008).

Another important factor is the large amount of roots produced and recycled annually. The total root biomass of *Arachis pintoi* in the 30-cm layer was more than 17 tonnes ha<sup>-1</sup> of dry matter (Valentin et al., 2001). For *B. humidicola*, about 60% of the root system was concentrated in the first 20 cm of soil depth, which contained 2.8 tonnes ha<sup>-1</sup> and 5.4 tonnes ha<sup>-1</sup> of root dry matter at the sites with one and seven years of cultivation, respectively (Santos et al., 2007).

In addition to the input of carbon to the soil from above- and belowground biomass, the soil carbon is contributed by the root exudates. In general, from 30 to 80% of net carbon fixed by the plants is transferred to the roots. Of this, from 23 to 80% is lost through the respiration and rhizodeposition in the soil (Whipps, 1990). Root exudates can contain more than 200 carbon compounds (Kumar et al., 2006), which are classified into various groups: water-soluble exudates (sugars, amino acids,

organic acids, hormones and vitamins), secretions (polymeric carbohydrates and enzymes), lysates (cell autolysis, cell walls) and gases (ethylene and CO<sub>2</sub>) (Whipps, 1990). These compounds can be utilized immediately by the microorganisms, significantly increasing the diversity, number and activity of microorganisms in the rhizosphere.

The ST-Brachiaria treatment presented increase of up to 22% in amylase activity compared to CT-Brachiaria in the inter-row space, demonstrating that the decreased soil disturbance (strip tillage) could improve the microbial activity. Conventional tillage (CT), which disturbs the soil to prepare the land, reduces the soil structure by degrading the soil aggregates. On the other hand, the lack of soil disturbance in strip tillage increases the microbial activity because it favors formation and stabilization macroaggregates, which provide the habitats for microbiota (Dick, 1997; Balota et al., 2004), increasing the microbial community.

#### **Correlations**

The enzyme activities were significantly inter-correlated, as observed in previous studies (Deng and Tabatabai, 1996; 1997; Balota et al., 2004). Arylsulfatase and acid phosphatase showed a strong relationship with the organic C (Table 3) and microbial biomass C (Figure 1). The significant correlations between the enzyme activities and organic C were likely due to the higher C levels supporting higher microbial biomass and thus more activity (Deng and Tabatabai, 1996; 1997). Furthermore, more organic matter provides better environmental conditions for stabilizing and protecting the extracellular enzymes.

There was a close relationship of arylsulfatase and acid phosphates with pH (r=0.93\* and r=0.72\*, respectively), as observed in some previous studies (Deng and Tabatabai, 1996; 1997; Balota et al., 2004; Mendes et al., 2003). However, some other authors have not found this relationship (Matsuoka et al., 2003; Carneiro et al., 2004), even though phosphatases are often closely correlated with soil pH, because acid phosphatases prevail in acid soils and alkaline phosphatases prevail in alkaline soils (Tabatabai, 1994).

There was a negative correlation between the acid phosphatase activity and extractable P (r=-0.65\*), confirming the observation that phosphatases were stimulated when soil phosphate levels were low (Mendes et al., 2003). However, this correlation

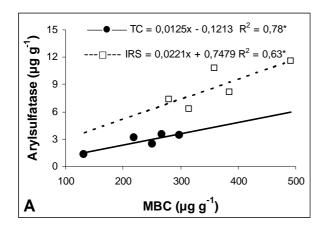
was not found in other studies (Balota et al., 2004; Carneiro et al., 2004).

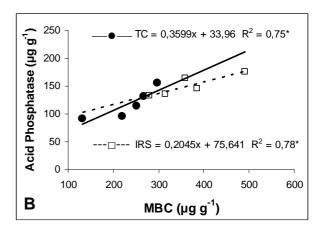
The results obtained in this study demonstrated the

responsiveness of soil enzyme activities to the changes in soil management practices.

**Table 3** - Simple correlations (r) between soil enzyme activities and chemical properties across all treatments and sampling positions.

Chemical	Amylase	Cellulase	Arylsufatase	Acid	Alkaline	
Properties				Phosphatase	Phosphatase	
Organic C	0.71*	0.44	0.75*	0.87*	0.68*	
pH	0.64*	0.20	0.93*	0.72*	0.45	
Ca	0.61	0.23	0.80*	0.64	0.24	
Mg	0.68*	0.22	0.92*	0.73	0.46	
CEC	- 0.29	0.33	- 0.70	- 0.40	- 0.35	
Base saturation	0.66*	0.18	0.93*	0.73	0.45	
Al saturation	- 0.55	- 0.09	- 0.91*	- 0.69	- 0.36	
Extractable P	- 0.34	- 0.02	- 0.78*	- 0.65	- 0.17	





**Figure 1** - Relationship of microbial biomass carbon (MBC) to arylsulfatase (A) and acid phosphatase (B) under the tree canopy (TC) and in the inter-row space (IRS). \* Significant at P≤ 0.05.

## **CONCLUSION**

- Various groundcover species intercropped with orange trees influenced soil enzyme activities both under the tree canopy and in the inter-row space.
- The cultivation of *B. humidicola* provided higher enzyme activities than the cultivation of Bahiagrass or leguminous species.
- Strip tillage increased the enzyme activities compared to conventional tillage.
- The cultivation of grass increased the soil enzyme activities compared to the cultivation of leguminous species in the inter-row space.

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Received: December 16, 2009; Revised: August 09, 2010; Accepted: December 17, 2010.