# Soil microbial biomass and enzyme activity in six Brazilian oxisols under cropland and native vegetation

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**ABSTRACT:** Oxisols are important soils that have been converted from native vegetation to croplands, and can affect soil biological properties such as microbial biomass and enzyme activity. Thus, the aim of this study was to evaluate the changes on soil microbial biomass and enzyme activity when native vegetation (NV) was converted to cropland (CL), such as maize or sugarcane in six oxisols from São Paulo state, Brazil. Soil microbial biomass C (MBC) and N (MBN), and the activity of arylsulphatase, dehydrogenase and fluorescein diacetate hydrolysis (FDA) were assessed in samples collected at 0-0.20 m. In general, MBC was higher under NV than CL (about + 40%), while MBN and FDA did not show a consistent pattern between NV and CL. All soils showed higher values of arylsulfatase (increased from 101 to 717%) and dehydrogenase (increased 15 to 220%) under NV than CL. In conclusion, soil microbial biomass C is usually higher under native vegetation than cropland. Arylsulphatase and dehydrogenase were the attributes that presented better differentiation between native and cropped soils. **Key words:** dehydrogenase, arylsulphatase, fluorescein diacetate hydrolysis, soil management.

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# INTRODUCTION

Oxisols are soils found almost exclusively in tropical areas from South America and Africa, being important to agriculture (Buol and Eswaran 1999). In Brazil, these soils cover about 100 million hectares, being described as highly weathered, and acidic, containing small amounts of plant nutrients (Gomes et al. 2019). However, liming and chemical fertilization can make these soils suitable for agriculture. Therefore, about 70 million hectares of Brazilian oxisols have been converted to cropland (Balota et al. 2015).

Although important for food production, the conversion from native soils to cropland decreases soil C storage as native vegetation is removed and replaced by crops which support lower soil C content and plant biomass (Fujisaki et al. 2015). Also, conventional tillage and mineral fertilization, applied in these soils, have promoted soil degradation (Dorneles et al. 2015). As consequence, there is a decrease in the soil biological processes (Gmach et al. 2020).

Soil biological processes, such as organic matter decomposition and nutrients cycling, are important to soil fertility and plant growth (Petter et al. 2019). Particularly, soil microbial biomass (SMB), the living part of soil organic matter (SOM), acts on the biological processes. Thus, soil degradation and C losses may alter negatively the size and activity of SMB, which affect soil biological and biochemical processes (Ferreira et al. 2016). Moreover, soil enzymes are indicators of biochemical functions and can provide quantitative changes on SOM. Some important enzymes, such as dehydrogenase, hydrolysis of fluorescein diacetate and arylsulphatase, are involved in the biogeochemical cycles (C, N and S) and consequently may



reflect changes in the soil metabolic processes (Notaro et al. 2018). In addition, these enzymes occur in all intact and viable microbial cells and may be with oxidative potential of SMB (Burns et al. 2013). Thus, soil microbial biomass and enzymes may be sensitive indicators of the effect of soil management on soil biological properties (Cardoso et al. 2013; Petter et al. 2019).

In this study, the hypothesis is that the conversion from native vegetation to cropland would change the status of soil biological properties. It could be expected since agricultural soils have different management and inputs which influence the soil properties. To address this hypothesis, this study assessed the changes on soil microbial biomass and enzyme activities in six different oxisols soils from São Paulo state, Brazil, that were converted from native vegetation to cropland (maize and sugarcane).

### MATERIAL AND METHODS

Soil samples were selected according a survey of soils from São Paulo state, Brazil: red-yellow oxisol (RYO), red oxisol (RO), acriferric red oxisol (ARO), yellow oxisol (YO), acricferri1 yellow oxisol (AYO), and dark red oxisol (DRO). The soil is classified as oxisol and ferralsol by USDA and FAO, respectively. These soils are present in all state of São Paulo, under different climatic conditions and use (native vegetation, sugarcane or maize) as shown in Table 1.

Red-yellow oxisol and DRO soils were cropped with maize over the past 5 years using the no-tillage cropping system, while RO, ARO, YO and AYO were cropped with sugarcane. For comparison between soils and their management, soil samples were collected in each soil under native sites and cropland. Therefore, each plot was codified as RYO<sub>1</sub>, RO<sub>1</sub>, ARO<sub>1</sub>, YO<sub>1</sub>, AYO<sub>1</sub> and DRO<sub>1</sub> for soil under native Cerrado; while RYO<sub>2</sub>, RO<sub>2</sub>, ARO<sub>2</sub>, YO<sub>2</sub>, AYO<sub>2</sub> and DRO<sub>2</sub> was codified for soil under cropland.

Each area under cropland or native forest was divided in four transects ( $100 \text{ m}^2$ ) where soil sampling was done. In each transect, ten subsamples were randomly collected in the 0–0.20 m layer to form a composite sample. For chemical and granulometric analyses, portions of soil samples (300 g) were air-dried, sieved (2 mm) and homogenized. The chemical (Table 2) and granulometric (Table 3) analyses were done according to the methods described by van Raij and Quaggio (2001) and Donagema et al. (2011), respectively. For biological analysis, samples were passed through a 2-mm sieve, and a 300-g aliquot of each sample was separated, placed in plastic bags, and stored in refrigerator at 4–8 °C for later determination of biological properties and enzymatic activity.

Soil microbial biomass C (MBC) and N (MBN) were determined according to Vance et al. (1987) with extraction of C and N from fumigated and unfumigated soils by  $0.5 \text{ mol.L}^{-1} \text{ K}_2 \text{SO}_4$ . Dehydrogenase (DHA), fluorescein diacetate hydrolysis (FDA) and arylsulphatase (ARYL) activities were analyzed as indicative measures of soil microbial activity. The FDA was determined according to the method of Schnürer and Rosswall (1982), DHA was determined using the method described in Casida Junior et al. (1964) and ARYL was determined according to Tabatabai and Bremner (1970).

Soil <sup>1</sup>	Cover	Management	Localization	Climate <sup>2</sup>
RYO <sub>1</sub>	NV	None	22°22'12"S, 47°54'16"W	CWa
RYO <sub>2</sub>	Maize	No-tillage, crop rotation with soybean, NPK fertilization	22°22'11"S, 47°55'09"W	CWa
RO <sub>1</sub>	NV	None	22°15'12"S, 47°50'38"W	CWa-AW
RO <sub>2</sub>	Sugarcane	Conventional tillage <sup>3</sup> , NPK fertilization	22°15'19"S, 47°50'37"W	CWa-AW
ARO <sub>1</sub>	NV	None	21º28'11"S, 47º53'38"W	AW
ARO <sub>2</sub>	Sugarcane	Conventional tillage <sup>3</sup> , NPK fertilization	21º28'10"S, 47º53'38"W	AW
YO <sub>1</sub>	NV	None	22°24'03"S, 47°52'52"W	CWa
YO <sub>2</sub>	Sugarcane	Conventional tillage <sup>3</sup> , NPK fertilization	22º24'12"S, 47º52'51"W	CWa
AYO <sub>1</sub>	NV	None	20°13'18"S, 48°01'40"W	AW
AYO <sub>2</sub>	Sugarcane	Conventional tillage <sup>3</sup> , NPK fertilization	20º13'18"S, 48º01'39"W	AW

Table 1. Climate characterization and information of the soils (classification, localization and management).

<sup>1</sup>Brazilian soil classification; <sup>2</sup>Koppen classification; <sup>3</sup>Tillage with plowing and harrowing. CWa = mean annual air temperature and average rainfall of 26 °C and 1500 mm·y<sup>-1</sup>; Cwa-Aw = transition between Cwa, and Aw, with a mean of annual air temperature and average rainfall of 27 °C and 1440 mm·y<sup>-1</sup>; Aw = mean annual air temperature and average rainfall of 22 °C and 1467 mm·y<sup>-1</sup>.

Sail	P1	OM <sup>2</sup>	рН³	K4	Ca⁵	Mg⁵	H+AI	SB	стс	V
3011	mg∙dm⁻³	g∙dm⁻³	CaCl <sub>2</sub>	mmol <sub>c</sub> ·dm³						%
$RYO_1$	7	35	5.3	1.5	36	4	20	42	62	67
RYO <sub>2</sub>	29	17	4.6	0.5	11	2	28	14	42	33
$RO_1$	14	27	4.3	0.6	12	3	52	16	68	23
RO <sub>2</sub>	27	15	4.5	0.6	6	1	60	8	68	12
ARO <sub>1</sub>	19	47	4.1	1.9	9	4	72	15	87	17
ARO <sub>2</sub>	69	36	5.5	2.4	41	25	28	68	96	71
YO <sub>1</sub>	15	36	4.5	5.2	14	9	47	28	75	38
YO <sub>2</sub>	69	36	5.5	2.4	41	25	28	68	96	71
aYO <sub>1</sub>	37	64	4.9	1.7	41	11	58	54	112	48
aYO <sub>2</sub>	41	40	5.3	4.7	28	9	38	42	80	52
DRO <sub>1</sub>	22	41	6.4	5.4	97	28	15	13	145	90
DRO <sub>2</sub>	25	28	5.6	3.7	31	17	25	52	77	67

#### Table 2. Chemical properties of evaluated oxisols.

<sup>1</sup>P extracted by ion exchange resin; <sup>2</sup>Organic matter extracted in K<sub>3</sub>Cr<sub>2</sub>O<sub>7</sub> + H<sub>3</sub>SO<sub>4</sub>; <sup>3</sup>PH measured in CaCl<sub>3</sub>; <sup>4</sup>K extracted by Mehlich-1; <sup>5</sup>Ca and Mg extracted in KCl. Red-yellow oxisol (RYO); red oxisol (RO); acriferric red oxisol (ARO); yellow oxisol (VO); acriferric yellow oxisol (AYO); dark red oxisol (DRO).

<b>ble 3.</b> Granulometry of different Oxisol soils under native vegetation or cropland.							
Soil	Sand	Silt	Clay				
	g⋅kg¹						
RYO <sub>1</sub>	759	20	221				
RYO <sub>2</sub>	738	60	202				
RO <sub>1</sub>	718	81	201				
RO <sub>2</sub>	368	102	530				
ARO <sub>1</sub>	141	143	716				
ARO <sub>2</sub>	159	139	702				
YO <sub>1</sub>	738	40	222				
YO <sub>2</sub>	597	60	342				
AYO,	407	123	470				

#### Та

Red-yellow oxisol (RYO); red oxisol (RO); acriferric red oxisol (ARO); yellow oxisol (YO); acriferric yellow oxisol (AYO); dark red oxisol (DRO).

430

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The data were evaluated for normality and subjected to analysis of variance (ANOVA) in a split plot design, being the land use as treatment 1 and the type of soil as treatment 2, under four replicates. To detect significant differences among treatments, when a significant p-value was detected, the means were compared using the Tukey's test (p < 0.05).

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207

198

#### **RESULTS AND DISCUSSION**

AYO,

DRO<sub>1</sub>

DRO,

The soils ARO and AYO showed highest values of MBC (+ 37% and + 45% in ARO and AYO, respectively) under native vegetation than cropland (Table 4). In contrast, the soil RYO under cropland showed highest values (+ 136%) of MBC than native vegetation. The soils RO, YO and DRO did not show differences in MBC between native vegetation and cropland.

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684

660

Land use affected MBN, but the values were not always higher always under native vegetation (Table 4). Thus, the soils RYO, RO and ARO showed highest values of MBN (125, 53, and 66% in RYO, RO and ARO, respectively) under cropland. In contrast, YO, AYO and DRO presented the highest values of MBN (85, 110, and 205% in YO, AYO and DRO, respectively) under native vegetation.

Except the soil YO, the SOM content was higher under native vegetation, while P content was higher in all cropped soils. Other soil chemical properties varied among the soil types and land use. On the other hand, the highest values of MBN in cropped soils may be due to the N fertilization. In addition, there could be an interaction between SOM, P and N on the responses of MBN as reported by Liu et al. (2013). Under cropland, the lowest value of MBN was found in RO cropped with sugarcane, while the highest value was found in RYO cropped with maize.

The arylsulfatase activity ranged from 12.28 to 229.42 mg PNP·kg<sup>-1</sup>·h<sup>-1</sup> in RO cropped with sugarcane and in DRO under native vegetation, respectively (Table 5). Similarly, the dehydrogenase activity ranged from 16.43 mg TTF·kg<sup>-1</sup> soil·h<sup>-1</sup> to 784.43 mg TTF·kg<sup>-1</sup>·h<sup>-1</sup> in RO cropped with sugarcane and in DRO under native vegetation, respectively. Interestingly, all oxisol soils showed higher arylsulfatase and dehydrogenase activities under native vegetation than cropland. Therefore, under native vegetation, arylsulfatase increased from 101 to 717%, while dehydrogenase increased from 15 to 220%, as compared to cropland.

The increase of diacetate fluorescein (FDA) hydrolysis ranged from 24.25 (YO; cropped with sugarcane) to 82.06 mg·kg<sup>-1</sup> soil·h<sup>-1</sup> (RO; under native vegetation). The FDA hydrolysis showed different pattern than those found for arylsulfatase and dehydrogenase activities. Therefore, in the soils RYO and DRO there were not differences between native vegetation and cropland. In the RO, the highest FDA hydrolysis was found under cropland (+ 33%), while in the ARO, YO and AYO the highest values

	M	BC	MBN				
Soil	mg C·k	⟨g⁻¹ soil	mg N·kg⁻¹ soil				
	Cropland	Native	Cropland	Native			
RYO	736.83 aBC	312.77 bC	207.22 aA	92.70 bD			
RO	577.39 aC	530.01 aC	72.73 aD	47.70 bE			
ARO	914.17 bB	1254.00 aAB	90.77 aCD	54.79 bE			
YO	796.66 aBC	979.35 aB	84.23 bCD	156.83 aC			
AYO	804.38 bBC	1167.24 aB	101.19 bC	212.45 aB			
DRO	1335.48 aA	1500.35 aA	135.42 bB	412.02 aA			

Table 4. Soil microbial biomass C (MBC) and N (MBN) from different oxisols soil under native vegetation or cropland.

Red-yellow oxisol (RYO); red oxisol (RO); acriferric red oxisol (ARO); yellow oxisol (YO); acriferric yellow oxisol (AYO); dark red oxisol (DRO). MBC = microbial biomass C; MBN = microbial biomass N. Means followed by the same lowercase letter in each line and capital letter in each column do not differ statistically from each other at p < 0.05 (Tukey's test).

Table 5. Arvisu	lfatase. deh	vdrogenase and fluorescei	n diacetate hvdrol	vsis (F	DA'	) from different oxisol	ls soil under	<sup>r</sup> native vec	etation or crou	bland
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	Arylsu	lfatase	Dehydro	ogenase	FDA mg FLU·kg <sup>-1</sup> soil·h <sup>-1</sup>		
Soil	mg PNP·k	g <sup>-1</sup> soil·h <sup>-1</sup>	mg TTF·k	g⁻¹ soil∙h⁻¹			
	Cropland	Native	Cropland	Native	Cropland	Native	
RYO	19.19 bBC	54.97 aD	43.04 bE	106.63 aE	24.84 aC	24.61 aE	
RO	12.28 bC	25.70 aE	16.43 bF	37.45 aF	40.58 aA	30.88 bD	
ARO	16.15 bBC	59.58 aD	289.74 bA	335.50 aC	44.34 bA	82.06 aA	
YO	32.56 bA	117.39 aB	237.09 bC	315.05 aD	24.25 bC	42.53 aC	
AYO	20.10 bBC	85.70 aC	212.28 bD	419.84 aB	33.55 bB	67.31 aB	
DRO	22.88 bAB	229.42 aA	245.29 bB	784.43 aA	27.70 aC	29.33 aDE	

Red-yellow oxisol (RYO); red oxisol (RO); acriferric red oxisol (ARO); yellow oxisol (YO); acriferric yellow oxisol (AYO); dark red oxisol (DRO). PNP = p-nitrophenol; TTF = triphenilformazan; FDA = fluorescein diacetate. FLU= fluorescein. Means followed by the same lowercase letter in each line and capital letter in each column do not differ statistically from each other at p < 0.05 (Tukey's test).

were found under native vegetation (+ 86, + 75, and + 103% in ARO, YO and AYO, respectively). Comparing the chemical properties of the RO under native vegetation and cropland, the main difference to explain this pattern is the P content, higher in RO under sugarcane. The content of SOM in ARO and AYO is higher under forest, which may explain why soil under native vegetation presented higher FDA hydrolysis than cropped soil.

In this study, soil microbial biomass was usually higher under native vegetation than cropped soils. Some reasons favor soil microbial biomass under native vegetation: a) higher plant diversity and lowest variation in temperature and moisture (Carvalho et al. 2018); b) higher organic inputs and better quality and quantity of plant litter (Lopes et al. 2010). These results are in agreement with previous studies, which have found higher soil microbial biomass under native vegetation than cropland (Viana et al. 2011; Novak et al. 2017; Freitas et al. 2017). D'Andréa et al. (2002) evaluated Brazilian oxisols under different land use and found a decrease of 49 and 73% in the MBC under pastures and cropland, respectively, as compared with native vegetation.

Interestingly, RYO soil cropped with maize presented higher MBC than native vegetation and it occurred probably due to the no-tillage system used in this soil. The no-tillage system may accumulate high amount of organic residue on the soil surface, favoring the soil microbial biomass (Choudhary et al. 2018). When the soil is not tilled, the organic matter accumulates seems and can support the increase in the soil microbial biomass (Holland 2004). Other important factor influencing the growth of soil microbial biomass is the mineral fertilization that supply nutrients to microorganisms, mainly P (Richardson and Simpson 2011). The highest values of MBN found in soils RYO, RO and ARO, under cropland, is due to the N fertilization previously used that increases the N pool and, consequently, its availability to soil microbial biomass. Similar results were found by Coser et al. (2007) which applied N to a Red-Yellow Oxisol cropped with wheat and found an increase in the MBN.

Activity of soil enzymes can be used as a sensitive indicator of soil microbial activity under native vegetation or cropland (Burns et al. 2013). The higher enzyme activities found under native vegetation may confirm the positive impact of the presence of diverse sources of substrates on soil microbial biomass. The increase in arylsulphatase and dehydrogenase activities were influenced by the increase in plant litter and soil microbial biomass. Arylsulfatase and dehydrogenase are oxidoreductases found in viable cells and their activities are correlated with soil microbial biomass and SOM (Madejón et al. 2007). Higher input of organic C, under native vegetation, promotes the increase of arylsulfatase and dehydrogenase activities. These results are in agreement with Acosta-Martínez et al. (2007), who found higher sulfatase activity in Oxisol soil under native vegetation than pastures and cropland. According to Schnürer and Rosswall (1982), FDA hydrolysis reflects the soil microbial activity and it is correlated with soil microbial biomass. However, the soil microbial biomass C and N did not differentiate FDA hydrolysis under native vegetation and cropped soils in this study.

# CONCLUSION

Arylsulphatase and dehydrogenase were the attributes that presented better differentiation between native and cropped soils. Soil microbial biomass C was usually higher under native vegetation. The exception was the soil under the no-tillage system that presented higher microbial biomass C. The interaction between N in the soil microbial biomass (an indicator soil N availability) and soil available P can promote higher biological activity in cropped than in native vegetation with higher organic matter content.

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