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CHEMICAL AND BIOCHEMICAL PROPERTIES OF *ARAUCARIA ANGUSTIFOLIA* (BERT.) KTZE. FOREST SOILS IN THE STATE OF SÃO PAULO⁽¹⁾

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SUMMARY

Araucaria angustifolia, commonly named *Araucaria*, is a Brazilian native species that is intensively exploited due to its timber quality. Therefore, *Araucaria* is on the list of species threatened by extinction. Despite the importance of soil for forest production, little is known about the soil properties of the highly fragmented *Araucaria* forests. This study was designed to investigate the use of chemical and biological properties as indicators of conservation and anthropogenic disturbance of *Araucaria* forests in different sampling periods. The research was carried out in two State parks of São Paulo: Parque Estadual Turístico do Alto do Ribeira and Parque Estadual de Campos de Jordão. The biochemical properties carbon and nitrogen in microbial biomass (MB-C and MB-N), basal respiration (BR), the metabolic quotient ($q\text{CO}_2$) and the following enzyme activities: β -glucosidase, urease, and fluorescein diacetate hydrolysis (FDA) were evaluated. The sampling period (dry or rainy season) influenced the results of mainly MB-C, MB-N, BR, and $q\text{CO}_2$. The chemical and biochemical properties, except K content, were sensitive indicators of differences in the conservation and anthropogenic disturbance stages of *Araucaria* forests. Although these forests differ in biochemical and chemical properties, they are efficient in energy use and conservation, which is shown by their low $q\text{CO}_2$, suggesting an advanced stage of succession.

Index terms: soil quality, microbial biomass, soil enzymes, soil respiration, microbial metabolic quotient ($q\text{CO}_2$).

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RESUMO: *PROPRIEDADES QUÍMICAS E BIOQUÍMICAS DO SOLO EM FLORESTAS DE ARAUCARIA ANGUSTIFOLIA (BERT.) KTZE NO ESTADO DE SÃO PAULO*

Araucaria angustifolia, comumente chamada de araucária, é uma espécie nativa brasileira considerada a mais explorada devido à qualidade de sua madeira. Por isso, encontra-se na atualidade ameaçada de extinção. Apesar da importância do solo na produção florestal, pouco se conhece sobre as propriedades edáficas das florestas de araucária, as quais estão altamente fragmentadas. O presente trabalho teve como objetivo investigar a utilização de propriedades químicas e bioquímicas do solo como indicadores de conservação e interferência antrópica em florestas de araucária, em diferentes períodos de amostragem. A pesquisa foi realizada em dois parques no Estado de São Paulo: Parque Estadual Turístico do Alto do Ribeira e Parque Estadual de Campos do Jordão. As propriedades bioquímicas do solo avaliadas foram: carbono e nitrogênio da biomassa microbiana (C-BM e N-BM), respiração basal (RB), quociente metabólico (qCO₂) e atividade das enzimas β-glicosidase, urease e hidrólise do diacetato de fluoresceína (FDA). A época de amostragem (estação seca/chuvosa) influenciou os valores encontrados, principalmente nas propriedades C-BM, N-BM, RB e qCO₂. As propriedades químicas e bioquímicas do solo avaliados, com exceção da variável K, foram indicadores sensíveis para detectar diferenças nos estádios de conservação e interferência antrópica das florestas de araucária. Embora diferentes com relação às propriedades químicas e biológicas do solo, as florestas envolvidas neste estudo foram eficientes no uso e na conservação da energia, já que apresentaram baixo qCO₂, sugerindo que elas se encontram em estágio mais avançado de sucessão.

Termos de indexação: qualidade do solo, biomassa microbiana, enzimas do solo, respiração do solo, quociente metabólico (qCO₂).

INTRODUCTION

Mixed ombrophilous forests, also known as *Araucaria* forests, represent a vegetation type of the Atlantic Forest biome. The main constituent of these forests is *Araucaria angustifolia* (Bert.) O. Ktze., a conifer endemic to South America (Whitmore, 1975). In Brazil, *Araucaria* is found on plateaus at 600-800 m asl and in some isolated cases above 1000 m, especially in the States of Rio Grande do Sul, Santa Catarina, Paraná, and São Paulo (Mattos, 1972). Due to its timber quality, this species has been heavily exploited for many years, resulting in the fragmentation of the existing forests, which caused the Brazilian Institute of Environment and Renewable Natural Resources (Instituto Brasileiro do Meio Ambiente e Recursos Naturais Renováveis) (IBAMA, 1992) to classify *Araucaria* as a Brazilian tree species at risk of extinction. Currently, it is estimated that the remaining *Araucaria* forests, either in early or advanced succession stages, cover no more than 0.7 % of the original area (Brasil, 2002), making this vegetation type one of the most threatened phytophysiognomies of the Atlantic Forest Biome (Medeiros et al., 2005).

Thus, the conservation of the remaining forests is essential for the survival of not only the *Araucaria angustifolia* species but also its entire ecosystem. Disturbances caused by anthropogenic interference in the remaining forests may lead to a loss of soil

quality, changing its fertility and associated microbial community, thus compromising forest sustainability. In this context, because heterotrophic microorganisms present in the soil are responsible for organic matter degradation and nutrient cycling and transformation, an evaluation of their activity may be useful to understand mineralization and the intensity of energy flows in the soil (Alef & Nannipieri, 1995; Choromanska & Deluca, 2001). Evaluations of microbial activity include analyses of biochemical properties of the soil, such as microbial biomass, enzyme activity, basal respiration rate, and metabolic quotient. These properties can serve as sensitive indicators of soil quality because microbial activity is highly sensitive to disturbances (Jenkinson & Ladd, 1981; Lynch, 1986; Kennedy, 1998).

Therefore, the purpose of this study was to evaluate the use of different soil chemical and biochemical properties, in different sampling seasons, as indicators of the conservation stage and anthropogenic influence on the remaining *Araucaria* forests in the State of São Paulo, with a view to their conservation.

MATERIALS AND METHODS

Site location and characteristics

For this study, six *Araucaria* forests were selected, three each from the Alto do Ribeira State Tourist Park

(Parque Estadual Turístico do Alto do Ribeira, PETAR) and from the State Park of Campos do Jordão (Parque Estadual de Campos do Jordão, PECJ), both in the state of São Paulo, Brazil.

PETAR is located in the southeast of the state in the municipalities of Apiaí and Iporanga (lat 24° 31' e 24° 34' S, long 48° 40' e 48° 44' W; 100 - 1000 m asl). The regional climate is described as humid mesothermal (Cfb) by the Köppen classification. Weather data of two years from the weather station of Capão Bonito (temperature) and the Apiaí rainfall station (precipitation) showed no abrupt temperature variations, with well-distributed precipitation over the years. A well-defined dry season could not be identified, but a dry period occurred between May and August (autumn/winter), and the rainiest period occurred between October and March (spring/summer) (Figure 1).

The following *Araucaria* forests were selected in PETAR (P):

1) Natural forest (NF) on a red-yellow latosol, with a great flora diversity, where *Araucaria* grows beside centennial trees, such as *Ocotea* spp., *Nectandra* spp., *Cedrella fissillis* Vell., *Ficus* spp., *Hymenaea courbaril* L., *Virola oleifera* (Schott) A. C. Smith, besides *Euterpe edulis* Mart.; epiphytes were also present (bromeliads, orchids, and lianas);

2) Replanted forest (RF), consisting of an approximately 18-year-old *Araucaria* plantation (at the first collection stage in 2002) with an understory containing grassy plants and many *Tibouchina granulosa* (Desr.) Cogn specimens, which are typical of secondary forests (Lorenzi, 1992). The soil of this forest was classified as Haplic Cambisol;

3) Anthropogenically influenced natural forest (ANF). This predominantly *Araucaria*-containing

forest overlying a haplic cambisol is affected by intense anthropogenic interference through subsistence agriculture in the forest surroundings and a high frequency of people and animals (mainly horses) transit. Epiphytes (bromeliads and orchids), *Syagrus romanzoffiana* (Cham.) Glassman, and *Baccharis trimera* (Less.) DC. were found and *Araucaria* regeneration was observed, in spite of the intensive kernel harvesting. At the third soil sampling (August 2003), a small-scale forest fire had evidently recently occurred in the area where the trees selected for this study were located, which affected the lower forest strata.

PECJ is situated in southeastern São Paulo (22° 45' S, 45° 30' W, average altitude of 1,500 m asl). By the Köppen classification, the regional climate was classified as humid mesothermal (Cfb). Climate data from the weather station of Campos do Jordão indicate a dry season with lower temperatures in the winter months (June/August). In the summer months (December/February) the rainfall volume is greatest and temperatures are highest (Figure 2). Although PECJ and PETAR belong to the same climate type (Cfb), the former has a dry season and lower mean temperatures in the winter months.

The following *Araucaria* forests were selected in PECJ:

1) Natural forest (NF): native forest in good state of conservation, over a red-yellow latosol. The floral diversity in this area was greater than in the other PECJ forests evaluated. In addition to *Araucaria*, the following species were found: *Clethra scabra* Pers., *Ocotea diospyrifolia* Meisn. Mez, *Persea pyrifolia* Ness Et Mart. ex Ness, *Podocarpus lambertii* Klotz., and others, as well as epiphytes.

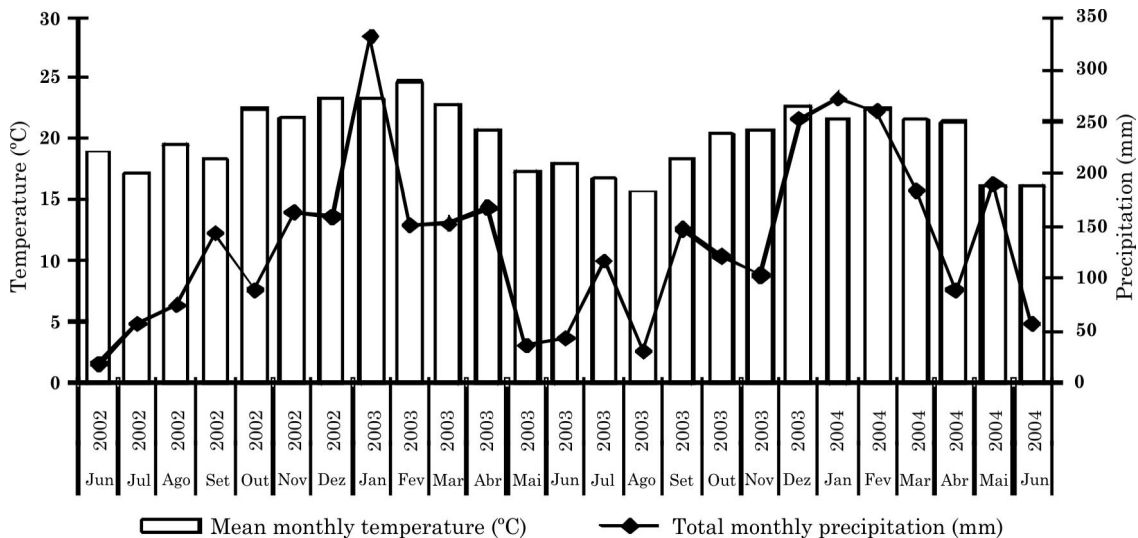


Figure 1. Climate diagram of the areas in the Parque Estadual Turístico do Alto do Ribeira (PETAR) selected for this study. The arrows indicate the sampling months (August 2002, January 2003, August 2003, and January 2004).

2) Replanted forest 1 (RF1). This forest was replanted in 1958 and overlies a haplic cambisol. The understory of this forest is mainly composed of *Aristida longiseta* Steud. and *Baccharis trimera* (Less.) DC. A forest fire occurred at this site in July 2001.

3) Replanted forest 2 (RF2). In this forest, replanted in 1959, the floristic diversity is high and *Araucaria* is regenerating. The soil underlying this forest was classified as yellow latosol. The same floral species as in the NF can be found here, although with reduced frequency.

Soil sampling and physical analyses

Soil collections were performed four times in two years (2002 - 2004), in contrasting seasons: twice in the rainy season and twice in the dry season (Figures 1 and 2).

For each forest under study, five soil samples were collected from the 0-20 cm layer. Each of the five soil samples was composed of three simple samples collected from under an *Araucaria angustifolia* tree canopy. The soil samples were sieved (2 mm mesh) and divided into two portions. One portion was air-dried and used to estimate chemical properties and to analyze the soil particle size. The other portion was maintained at field moisture to analyze the biochemical properties of the soil. The granulometric analysis was performed using the densitometer method proposed by Camargo et al. (1986) (Table 1).

Soil chemical properties

The soil was chemically characterized according to the methodology described by Raij et al. (2001). The pH was measured using a 5 mL of 0.01 mol L⁻¹ CaCl₂ solution. The total organic C and N of the soil were

estimated according to the method proposed by Raij et al. (2001). Calcium, phosphorus, magnesium, and potassium were extracted using an ion exchange resin (Raij et al., 2001). Phosphorus was quantified colorimetrically using ammonium-molybdenum. The Ca and Mg concentrations were determined using atomic absorption spectroscopy, and K by flame photometry (Raij et al., 2001). The cation exchange capacity (CEC) at pH 7.0 (defined as the amount of adsorbed cations at pH 7.0), as well as the CEC base saturation percentage (V) at pH 7.0 (Raij et al., 2001), was also evaluated.

Biochemical soil properties

The microbial biomass carbon (MB-C) content was determined using the fumigation-extraction method proposed by Vance et al. (1987); the microbial biomass nitrogen (MB-N) content was determined according to the method described by Joergensen & Brookes (1990) and Joergensen (1996).

The microbial basal respiration (BR) was estimated by measuring the amount of C-CO₂ released within 10 days (Alef & Nannipieri, 1995). The metabolic quotient (q_{CO_2}) was calculated from the ratio between BR and MB-C (Anderson & Domsch, 1993).

The enzymatic activity of urease was evaluated according to the method proposed by Tabatabai & Bremner (1972) and β -glucosidase activity by the method described by Eivazi & Tabatabai (1988). The method proposed by Diack (1997) was used to measure fluorescein diacetate (FDA) hydrolysis.

Statistical analyses

The results were subjected to analysis of variance (ANOVA) and means test (Scott-Knott, 5 %) using the statistical package Sisvar (Ferreira, 2003). Then

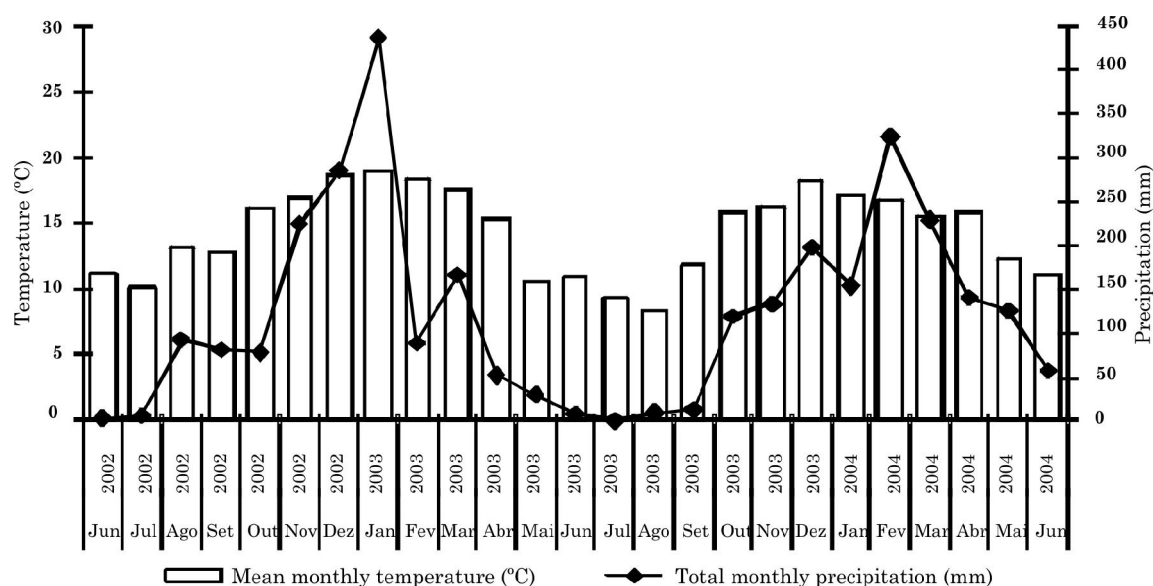


Figure 2. Climate diagram of the areas in the Parque Estadual de Campos do Jordão (PECJ) selected for this study. The arrows indicate the sampling months (September/2002, March 2003, August 2003, and February 2004).

the data were subjected to principal component analysis (PCA) using software Canoco 4.5, to summarize the multidimensional variation into one diagram, ranked by the degree of similarity of the components around the variables (Ter Braak, 1986). Both the forest and the soil properties data were transformed either into ordinates corresponding to their projection on the ordination axes or into eigenvalues representing the weight of each variable on each component (axis) and functioning as a correlation coefficient ranging from -1 to +1. PCA was chosen after performing a detrended correspondence analysis (DCA), which helped define the gradient length. Since the gradient was smaller than 3, PCA was preferred. In this study, eigenvalues ≥ 0.4 were considered to represent a high degree of association for principal component interpretation.

RESULTS

Chemical soil properties

The chemical analysis of the PETAR forest soils revealed that the only properties differing between the sampling periods were total organic carbon (TOC) and total nitrogen (TN) contents (Table 2). The TOC content was significantly greater in the rainy than in the dry season for all PETAR forests (Table 2). The highest TN contents were found in the dry season for all PETAR forests (Table 2). In the same period, the TN content in both NF and RF was significantly greater than in ANF (Table 2).

In the rainy season, no significant differences were found for any of the PETAR forests with regard to TN, P, and TOC contents (Table 2), and the P and TOC contents were considered high for all forests (Raij et al., 1997). The other chemical properties differed significantly among the forests selected for this study (Table 2).

The NF was the only soil with a pH value indicating low-level acidity, whereas all other PETAR forest soils were acidic, according to Bulletin 100 (Raij et al., 1997) (Table 2). The Ca content was significantly higher in NF and classified as "very good" (Raij et al., 1997) (Table 2). The other forests showed no significant difference in Ca content, which was considered "low" (Raij et al., 1997). The Mg content in NF was considered "very good" (Raij et al., 1997) and significantly higher than that in RF and ANF, where it was classified as "medium" (Raij et al., 1997) (Table 2). CEC (pH 7.0) was considered "very good" for both NF and RF (Raij et al., 1997), and the levels in both NF and RF were significantly higher than in ANF, for which the CEC (pH 7.0) was only considered "good" (Raij et al., 1997). In both RF and ANF, the V values were significantly lower than in NF (Table 2). The K content was considered low for all PETAR forests, although in the ANF forests, it was significantly higher in the rainy season when compared to the other forests (Table 2).

Similar differences in soil chemical properties were found in the dry season for all PETAR forests (Table 2), with the exception of TN and K, which did not differ significantly among the forests evaluated in this period.

The PECJ forests generally differed in terms of TOC and TN content between the sampling periods; the TOC content of NF was the only exception. In the dry season, the contents of both properties were significantly greater in replanted forests than in the rainy season (Table 3). All other soil chemical properties (pH, P, K, Ca, Mg, CEC, and V) did not differ among the sampling periods in all PECJ forests (Table 3).

In the rainy season, the pH, P, K, and Ca values did not differ significantly among the PECJ forests (Table 3). The pH was considered acidic (Raij et al., 1997) in all PECJ forests (Table 3). The P content was considered high in NF and medium in RF1 and

Table 1. Granulometric distribution of soils under different *Araucaria* forests located in the Campos de Jordão State Park (Parque Estadual de Campo do Jordão, PECJ) and Alto do Ribeira Tourist State Park (Parque Estadual Turístico do Alto do Ribeira, PETAR)

<i>Araucaria</i> Forests	Clay <0.002 mm	Silt 0.002-0.05 mm	Sand 0.05-2.0 mm	Soil texture
	dag kg ⁻¹			
PETAR				
NF*	53	15	32	Clayey
RF	51	41	8	Silty clay
ANF	56	19	25	Clayey
PECJ				
NF	44	46	10	Silty clay
RF1	39	41	20	Silty clay loam
RF2	34	43	23	Clay loam

*NF: Natural Forest; RF: Replanted Forest and ANF: Anthropogenically influenced natural forest.

Table 2. Chemical properties of soils under different PETAR *Araucaria* forests

PETAR Forests	Soil chemical properties								
	pH CaCl ₂	P	K	Ca	Mg	CEC	TOC	TN	V
	mg dm ⁻³		cmol _c dm ⁻³			g kg ⁻¹		%	
	Rainy season								
NF	5.5 a	14.4 a	0.2 b	8.0 a	2.1 a	18.6 a	55.7 aA	0.7 aB	71.4 a
RF	3.6 b	12.0 a	0.2 b	0.6 b	0.4 b	19.1 a	51.2 aA	0.7 aB	6.4 c
ANF	3.9 b	22.6 a	0.4 a	1.6 b	0.8 b	12.2 b	52.2 aA	0.5 aB	23.0 b
CV(%)	10.1	48.5	40.0	40.0	45.0	27.1	7.2	30.4	40.2
	Dry season								
NF	5.6 a	22.8 a	0.2 a	12.3 a	3.0 a	18.2 a	36.7 aB	1.9 aA	84.4 a
RF	3.7 c	12.8 a	0.2 a	0.8 b	0.5 b	18.0 a	42.7 aB	1.7 aA	8.4 c
ANF	4.1 b	15.2 a	0.2 a	1.5 b	0.6 b	8.92 b	36.6 aB	1.0 bA	25.0 b
CV(%)	3.9	48.0	32.9	44.9	40.1	18.2	38.7	32.5	35.5
	CV (%) referring to the sampling season								
NF	4.5	42.6	37.5	43.5	12.3	30.8	10.9	36.1	18.1
RF	3.7	17.3	28.5	35.8	27.8	14.0	37.8	22.0	7.4
ANF	6.7	38.4	39.9	37.9	40.1	21.1	34.8	20.8	35.8

(NF: natural forest. RF: replanted forest. and ANF: anthropogenically influenced natural forest)

Identical lowercase letters within the same season (dry or rainy) indicate no significant difference between the forests evaluated according to the Scott-Knott test (5 %). Absent uppercase letters within the same forest indicate no significant difference between the seasons evaluated (rainy and dry), according to the Scott-Knott test (5 %).

Table 3. Chemical properties of soils under different PECJ *Araucaria* forests

PECJ Forest	Soil chemical properties								
	pH CaCl ₂	P	K	Ca	Mg	CEC	TOC	TN	V
	mg dm ⁻³		cmol _c dm ⁻³			g kg ⁻¹		%	
	Rainy season								
NF	3.7 a	12.4 a	0.3 a	0.5 a	0.5 a	18.2 a	55.5 a	0.6 aB	7.0 a
RF1	3.7 a	8.4 a	0.2 a	0.3 a	0.2 b	11.6 b	29.4 cB	0.3 cB	6.0 a
RF2	3.6 a	8.0 a	0.2 a	0.3 a	0.1 b	19.1 a	45.2 bB	0.4 bB	2.8 b
CV(%)	2.3	31.6	26.3	34.2	36.2	15.6	11.7	17.2	33.2
	Dry season								
NF	3.6 a	11.2 a	0.2 a	0.5 a	0.3 a	23.3 a	61.2 a	1.7 aA	4.4 a
RF1	3.7 a	8.6 a	0.2 a	0.3 a	0.2 a	11.3 b	39.1 bA	0.9 bA	6.5 a
RF2	3.4 a	9.4 a	0.3 a	0.3 a	0.2 a	25.8 a	58.3 aA	1.5 aA	2.6 a
CV(%)	5.2	30.0	24.4	35.0	37.7	33.4	34.0	24.0	37.3
	CV (%) referring to the sampling season								
FN	4.3	31.2	22.9	39.9	41.7	28.6	8.5	25.1	34.3
RF1	2.9	31.8	37.2	42.2	40.5	21.5	13.7	33.1	31.4
RF2	4.7	26.0	20.7	36.2	32.2	26.8	9.7	22.3	18.5

(NF: natural forest; RF: replanted forest)

Identical lowercase letters within the same season (dry or rainy) indicate no significant difference between the forests evaluated, according to the Scott-Knott test (5 %). Absent uppercase letters within the same forest indicate no significant difference between the seasons evaluated (rainy and dry), according to the Scott-Knott test (5 %).

RF2; however, the differences were not significant (Raij et al., 1997) (Table 3). The Ca and K contents were considered low for all PECJ forests (Raij et al., 1997) (Table 3). The CEC (pH 7.0) differed among the PECJ forests in the rainy season, and NF and RF2 were highest. Despite the significant differences in CEC (pH 7.0) among the forests, the values were considered low for all PECJ forests (Raij et al., 1997) (Table 3). The properties V and Mg differed significantly among the PECJ forests (Table 3). However, all forest soils had low Mg contents and V was less than 50 %.

Similar differences as in the rainy season were found in the dry season samples for the variables pH, K, Ca, and CEC; however, CEC (pH 7.0) was classified as "good" in both NF and RF2 (Raij et al., 1997) (Table 3).

Biochemical soil properties

Microbial biomass carbon and nitrogen

Considering all forests evaluated, the MB-C and MB-N microbial biomass contents ranged from 545 to 738 $\mu\text{g C g}^{-1}$ soil and from 84 to 207 $\mu\text{g N g}^{-1}$ soil, respectively, in the natural forests and from 271 to 946 $\mu\text{g C g}^{-1}$ soil and 35 to 143 $\mu\text{g N g}^{-1}$ soil, respectively, in the replanted forests (Tables 4 and 5).

The MB-C and MB-N contents in the PETAR planted forests (Table 4) varied according to the sampling period (rainy or dry season); both values were significantly higher in the dry season for RF and ANF (Table 4). The natural forest showed no significant differences in MB-C and MB-N contents among the sampling periods (Table 4). The MB-C contents were not significantly different among the PETAR forests during the dry season (Table 4). However, in the same season, the MB-N content was significantly greater in the natural forest than in the planted forests (Table 4). In the rainy season, both MB-C and MB-N were significantly higher in the natural forest than in the planted forests (Table 4).

In the PECJ forests, MB-C contents varied according to the sampling period in RF1 and RF2, and the contents were highest in the rainy season. For the MB-N content, only in RF1 a similar variation according to sampling period was observed; however, the contents were highest in the dry season (Table 5). In the rainy season, the MB-C and MB-N contents did not differ significantly between NF and RF2; however, both had significantly greater contents than RF1 (Table 5). In the dry season, the MB-C content differed significantly among the forests, and NF had the greatest content, followed by RF2 and RF1,

Table 4. Biochemical properties of the soil: microbial biomass carbon (MB-C), microbial biomass nitrogen (MB-N), basal respiration (BR), metabolic quotient ($q\text{CO}_2$), fluorescein diacetate hydrolysis (FDA), β -glucosidase and urease of the following PETAR forests: natural forest (NF), replanted forest (RF), and anthropogenically influenced natural forest (ANF) in the rainy and dry seasons

PETAR Forest	Biochemical properties						
	MB-C	MB-N	BR	$q\text{CO}_2$	FDA	β -glucosidase	Urease
	$\mu\text{g C g}^{-1}$	$\mu\text{g N g}^{-1}$	$\text{mg CO}_2 \text{ g}^{-1} \text{ h}^{-1}$	$\mu\text{g CO}_2 \mu\text{g}^{-1} \text{ C g}^{-1} \text{ h}^{-1}$	$\text{mg fluorescein g}^{-1} \text{ day}^{-1}$	ρ nitrophenol $\text{g}^{-1} \text{ h}^{-1}$	$\mu\text{g N-NH}_4^+ \text{ g}^{-1} \text{ h}^{-1}$
Rainy season							
NF	679.5 a	184.2 a	0.04 a	0.07 a	449.8 a	500.6 a	40.8 a
RF	352.3 cB	58.8 bB	0.04 a	0.10 aA	381.5 a	348.4 a	7.1 b
ANF	490.9 bB	55.2 bB	0.05 a	0.10 aA	421.8 a	407.5 a	15.7 b
CV(%)	20.7	29.7	23.4	27.8	11.9	25.2	30.1
Dry season							
NF	737.6 a	207.3 a	0.06 a	0.08 a	441.5 a	417.2 a	48.7 a
RF	797.5 aA	131.5 bA	0.03 b	0.04 bB	446.8 a	369.9 a	6.1 b
ANF	946.3 aA	143.2 bA	0.04 b	0.05 bB	464.2 a	423.4 a	18.3 b
CV(%)	16.1	11.7	24.8	24.2	12.8	31.2	33.3
CV (%) for the sampling season							
NF	22.6	20.7	7.1	13.9	8.2	17.0	7.5
RF	11.9	15.4	18.5	23.4	19.7	8.4	9.6
ANF	13.1	18.6	23.3	20.9	13.3	14.9	20.9

Identical lowercase letters within the same season (dry or rainy) indicate no significant difference between the forests evaluated, according to the Scott-Knott test (5%). Absent uppercase letters within the same forest indicate no significant difference between the seasons evaluated (rainy and dry), according to the Scott-Knott test (5%).

Table 5. Biochemical properties of the soil: microbial biomass carbon (MB-C), microbial biomass nitrogen (MB-N), basal respiration (BR), metabolic quotient (qCO_2), fluorescein diacetate hydrolysis (FDA), β -glucosidase, and urease of the following PECJ forests: natural forest (NF), replanted forest 1 (RF1), and replanted forest 2 (RF2) in the rainy and dry seasons

PECJ Forest	Biochemical properties						
	MB-C	MB-N	BR	qCO_2	FDA	β -glucosidase	Urease
	$\mu\text{g C g}^{-1}$	$\mu\text{g N g}^{-1}$	$\text{mg CO}_2 \text{ g}^{-1} \text{ h}^{-1}$	$\mu\text{g CO}_2 \mu\text{g}^{-1} \text{ C g}^{-1} \text{ h}^{-1}$	$\text{mg fluorescein g}^{-1} \text{ day}^{-1}$	ρ -nitrophenol $\text{g}^{-1} \text{ h}^{-1}$	$\mu\text{g N-NH}_4^+ \text{ g}^{-1} \text{ h}^{-1}$
Rainy season							
NF	599.6 a	101.9 a	0.07 aA	0.12 aA	429.5 a	306.0 a	7.9 a
RF1	430.2 bA	34.6 bB	0.04 bA	0.9 aA	294.6 a	217.2 a	8.1 a
RF2	586.7 aA	105.9 a	0.06 aA	0.10 aA	435.2 a	353.6 a	12.1 a
CV(%)	13.3	16.4	19.0	17.1	17.4	25.9	24.1
Dry season							
NF	545.4 a	84.1 b	0.03 aB	0.05 aB	401.5 a	454.5 a	8.9 a
RF1	271.1 cB	72.7 bA	0.02 aB	0.08 aB	308.3 a	284.2 a	12.4 a
RF2	494.8 bB	118.2 a	0.03 aB	0.07 aB	423.5 a	383.1 a	11.7 a
CV(%)	6.3	12.4	15.3	15.5	17.2	30.1	29.5
CV (%) for the sampling season							
NF	7.5	13.3	14.2	10.7	11.6	24.9	25.9
RF1	4.9	11.7	18.1	15.0	23.4	20.7	26.8
RF2	12.9	17.4	15.6	10.7	9.3	16.3	18.9

Identical lowercase letters within the same season (dry or rainy) indicate no significant difference between the forests evaluated, according to the Scott-Knott test (5 %). Absent uppercase letters within the same forest indicate no significant difference between the seasons evaluated (rainy and dry), according to the Scott-Knott test (5 %).

respectively (Table 5). RF2 had a significantly greater MB-N content than both RF1 and NF (Table 5).

Basal respiration and metabolic quotient

BR did not differ among the sampling periods in the PETAR forests (Table 4). However, qCO_2 differed significantly among the sampling periods in the replanted forests, and the greatest qCO_2 was found in the rainy season (Table 4). Neither BR nor qCO_2 differed significantly among the forests in this study (Table 4). In the dry season, BR and qCO_2 were significantly higher in the natural forest than in the replanted forests (Table 4).

BR and qCO_2 were significantly different among the sampling periods for all PECJ forests, and the values were highest in the rainy season (Table 5). In the dry season, no statistically significant differences in BR or qCO_2 were found among the forests (Table 5). In the rainy season, the BR values were significantly higher both in NF and RF2 than in RF1 (Table 5). In contrast, qCO_2 did not differ among the PECJ forests in the rainy season (Table 5).

Enzymatic activity

The enzymatic activity of β -glucosidase, urease, and FDA hydrolysis did not differ significantly among

the sampling periods for all forests evaluated in this study (Tables 4 and 5). Neither β -glucosidase nor FDA differed significantly among the forests in this study, regardless of the season (Tables 4 and 5). The urease activity in NF of PETAR was significantly higher in both seasons (Table 4), while no PECJ forest was significantly different in the seasons (Table 5).

Principal component analysis

The PCA for the PETAR forests (Figure 3) showed that the first two components of the analysis accounted for 93.3 % of the total variance in the data, of which 63.3 % was explained by principal component 1 (PC1) and 30.3 % by principal component 2 (PC2) (Figure 3). PC1 was directly related to the variables pH (0.85), V (0.90), Ca (0.85), Mg (0.67), P (0.42), TOC (0.46), urease activity (0.77), β -glucosidase activity (0.52), and BR (0.60) (Figure 3). The variables MB-N (-0.78), MB-C (-0.81), TN (-0.89), and FDA (-0.40) were inversely correlated with PC1. CEC and K were not related to PC1 (eigenvalues < 0.4) (Figure 3).

PC2 was directly related to pH (0.56), TN (0.57), V (0.54), Ca (0.63), P (0.46), BR (0.86), MB-N (0.44), MB-C (0.67), and FDA (0.53). Both qCO_2 (-0.86) and TOC (-0.47) were inversely correlated with PC2. CEC and K were not related to PC2 (eigenvalues < 0.4) (Figure 3).

The segregation of points representing the ANF (Figure 3) revealed that certain soil chemical and biochemical properties changed according to the sampling period (dry or rainy season); ANF was mainly associated with TOC in the rainy season. The chemical and biochemical properties of the RF soil also differed according to the sampling period. In the dry season, RF was mainly associated with MB-N, TN, MB-C, and FDA and in the rainy season mainly with TOC. The segregation of points representing the NF, closer to the center of the figure, shows a better balance of the values obtained for the soil chemical and biochemical properties according to the sampling period and when compared to the other PETAR forests (Figure 3).

The PETAR forests were segregated, demonstrating that these forests could be ranked according to their degree of conservation and anthropogenic influence based on the soil chemical and biochemical properties (Figure 3).

Among the PECJ forests evaluated in this study, the first two PCA components explained 74.5 % of the total variance, of which 51.3 % was explained by PC1 and 24.1 % by PC2 (Figure 4). The properties directly correlated with PC1 were MB-N (0.50), β -glucosidase (0.46), FDA (0.56), TOC (0.87), and CEC (0.63). The variables pH (-0.47), V (-0.46), BR (-0.50), and MB-C

(-0.55) were inversely correlated with PC1. The variables K, P, Ca, Mg, urease activity, and qCO_2 were not correlated with PC1 (eigenvalues < 0.4).

For PC2, pH (0.55) and V (0.57) were directly correlated. The properties qCO_2 (-0.84), BR (-0.64), β -glucosidase (-0.57), Mg (-0.42), FDA (-0.43), MB-N (-0.45), TOC (-0.58), CEC (-0.50), and TN (-0.84) were inversely correlated with PC2 (Figure 4). The K, P, Ca, and urease activity variables were not correlated with PC2 (eigenvalues < 0.4) (Figure 4).

The segregation of points representing the PECJ forests evaluated in this study revealed the variability in the soil chemical and biochemical properties according to the sampling period for all forests studied. The segregation by PC1 showed an association of the rainy season with the properties MB-C, BR, qCO_2 , V, and pH; the dry season was correlated with the properties TN, CEC, MB-N, FDA, and TOC (Figure 4).

The PECJ forests were segregated, demonstrating that the soil chemical and biochemical properties allowed a ranking of these forests according to their degree of conservation and anthropogenic influence (Figure 4).

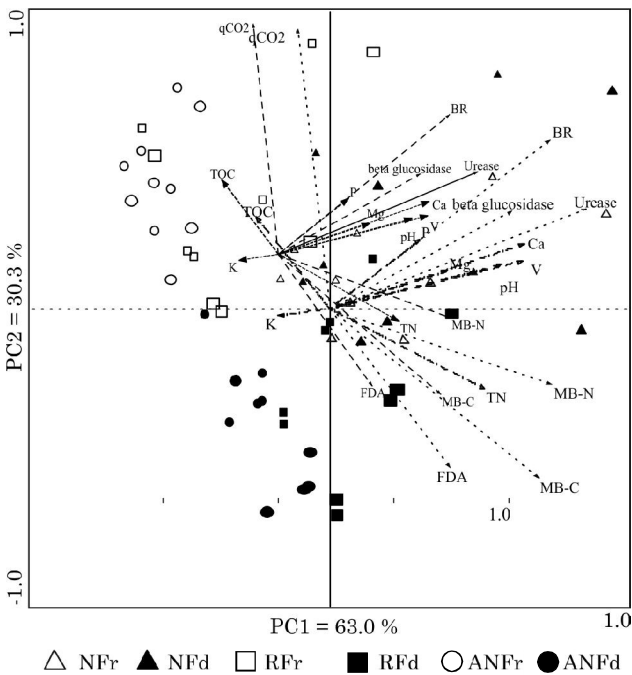


Figure 3. Principal component analysis (PCA) ordination diagram of the PETAR forests, NFr (natural forest, rainy season), NFd (natural forest, dry season), RFr (replanted forest, rainy season), RFd (replanted forest, dry season), ANFr (anthropogenically influenced natural forest, rainy season), and ANFd (anthropogenically influenced natural forest, dry season), as well as soil chemical and biochemical properties.

DISCUSSION

Microorganisms can be classified according to the optimal pH range for each species. Therefore, the pH value will have an indirect influence on the type of

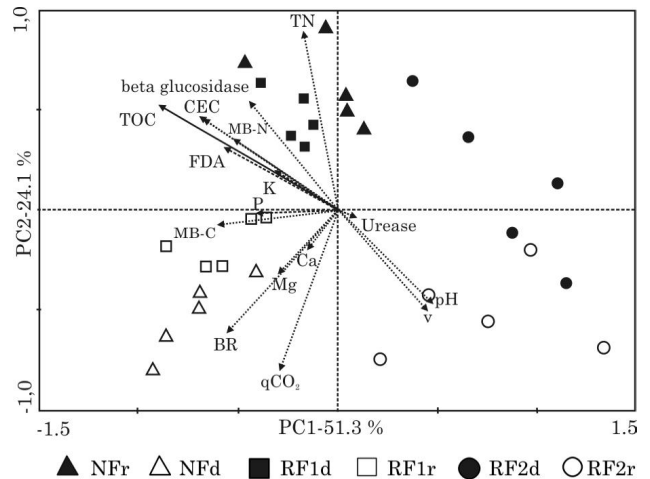


Figure 4. Principal component analysis (PCA) ordination diagram of the PECJ forests, NFr (natural forest, rainy season), NFd (natural forest, dry season), RF1r (replanted forest 1, rainy season), RF1d (replanted forest 1, dry season), RF2r (replanted forest 2, rainy season), and RF2d (replanted forest 2, dry season), as well as soil chemical and biochemical properties.

microbial community inhabiting the environment being studied by affecting biological processes, especially the activity of enzymes, most of which are pH-dependent.

The pH value influences the solubility and consequently the availability of several nutrients and elements, which can be toxic for microorganisms. In this study, chemical analyses suggest that the soils have a strong buffering capacity in view of the generally favorable properties, including the presence of a clay fraction, high organic material content, and high CEC, which prevent changes in pH-dependent properties and functions.

The only soil studied with weak acidity (pH = 5.6) was the PETAR natural forest. According to Lopes & Guilherme (1992), at this pH value, the Al saturation percentage of effective CEC should be near zero, which explains the high base saturation (V) in this soil of 71.4 and 84.4 % in the rainy and dry seasons, respectively. Furthermore, the nutrient content of this soil was greater, with Ca concentrations of 12.3 and 8.0 $\text{cmol}_c \text{dm}^{-3}$ and Mg concentrations of 2.1 and 3.0 $\text{cmol}_c \text{dm}^{-3}$ in the rainy and dry seasons, respectively. These concentrations can be considered high (Table 2). Calcium and magnesium are mainly provided by soil minerals, and the geologic formation of the PETAR area is mainly based on limestone and amphibolites. These elements can be combined with organic matter in the soil; organic and inorganic colloids can adsorb both Ca and Mg, thereby keeping them in exchangeable form and avoiding leaching losses.

The good CEC level found in the evaluated soils was expected because high TOC contents were obtained. However, in the PECJ RF1, TOC was lower than in the other forests, which led to lower CEC and may increase nutrient loss by leaching in these areas (Table 3).

Microbial biomass has been used to evaluate soil conditions for representing the fraction responsible for organic matter mineralization and nutrient cycling. It can be regarded as the key compartment of the carbon cycle, representing a considerable reservoir of nutrients that can act as a drain and source, depending on the amount of energy entering or leaving the system (Moreira & Siqueira, 2006).

The C contents of the microbial biomass obtained for the forests were within the range described in the literature for other Brazilian forest ecosystems, ranging from 109 to 1600 mg C g^{-1} soil (Moreira & Siqueira, 2006). This broad range of biomass C is related to the heterogeneity of the different ecosystems.

Biological properties can vary greatly with seasons, especially in the soil surface layers, where oscillations in temperature and moisture are greater (Campbell & Biederbeck, 1976; Martinez & Ramirez, 1978; Cattelan & Vidor, 1990). As shown in the PETAR climate diagram (Figure 1), rainy periods were well-distributed over the years evaluated, with no abrupt

temperature changes. Nevertheless, significant differences were observed in MB-N and MB-C contents among the sampling periods for all forests except the PETAR NF. This finding could indicate that the PETAR NF soil is less vulnerable to climate changes due to better soil protection by greater vegetation cover, decreasing the exposure to solar radiation and retaining moisture, which favors the stabilization of the microclimate and, consequently, of the ecosystem (Bragagnolo, 1986; Wardle & Parkinson, 1990). A similar finding was obtained for PECJ NF, since no significant differences in MB-N and MB-C were observed in this forest among the sampling periods, although the climate diagram (Figure 2) shows periods of dry weather and sudden temperature drops, reinforcing the important role of the vegetation cover in maintaining biological processes.

The MB-C and MB-N variables were influenced by the sampling period in the other forests evaluated. However, contrary to expectations, the MB-C content in the PETAR ANF was 49.8 % higher in the dry than in the rainy season. It is likely that the forest fire in this area induced rapid mineralization of accumulated organic matter, increasing the availability of essential nutrients for the growth of the microbial community. The PETAR RF also showed a 55.4 % greater MB-C in the dry season than in the rainy season. In this case, the most likely explanation is the high precipitation index of the summer months of this region because the vegetation cover in this forest, although present, is remarkably less dense than in the PETAR NF. This characteristic may make this soil more vulnerable to the effects of excessive precipitation, i.e., soil loss by erosion and greater nutrient loss by leaching.

The natural forests tended to have greater MB-C and MB-N contents. This finding could be explained by Odum's ecosystem development and evolution theory (Odum, 1983), which states that ecological succession involves changes in species structure and community processes over time. When comparing natural and replanted woods, it is evident that the changes within these ecosystems are different, serving as a reminder that the physical environment helps determine the rate of these changes.

Basal respiration has been used to assess the general activity of microbial biomass, and can be strongly influenced by a variety of soil factors, e.g., moisture, temperature, structure, and nutrient availability (Alef & Nannipieri, 1995). Insam & Domsch (1988) reported that as microbial biomass becomes more efficient, there is less loss of C as CO_2 through respiration, and a significant carbon fraction is incorporated into the microbial biomass. This effect occurs because the ecosystem tends to retain and conserve more nutrients as the succession progresses, decreasing the dependence on new entries; thus, increasing the efficiency of energy and nutrient use is a general succession strategy (Odum, 1983).

Metabolic quotient values of the forests evaluated in this study are within the range found for other forest ecosystems (Behera & Sahani, 2003) and can be classified as "low" compared to environments under stress (Gomide et al., 2011). These findings indicate that the forests in this study are at a more advanced succession stage, in which the energy accumulated in the system is greater (Odum, 1983; Insam & Haselwandter, 1989; Anderson & Domsch, 1990; Kieft et al., 1998). Therefore, it is worth emphasizing that, despite differences in the conservation stage of the forests evaluated, the microbial community of each forest has established a particular balance of function within its own limits.

The quantification of urease activity may indicate the soil potential to convert organic N into mineral N, thus initiating the N mineralization process. The urease activities for the forests evaluated did not differ significantly among the sampling periods, in agreement with a study of Gianfreda & Bollag (1996), who also reported a weak correlation between enzyme activity and seasonal variations.

In the PECJ forests, no significant differences in urease activity were stated. This finding can demonstrate that forests subjected to anthropogenic action (RF1 and RF2) recovered their nitrogen mineralization function.

The evaluated PETAR forests differed significantly in terms of urease activity, which were 86 and 79 % greater in NF those for RF and 62 and 60 % greater than those for ANF in the dry and rainy seasons, respectively. However, it is worth mentioning that chemical properties, such as vegetation cover, strongly influence enzyme activity. As described previously, only NF had nutrient-rich soil with a nearly neutral pH. Additionally, the soil had a more diverse vegetation cover, which made the litter of this forest more heterogeneous compared to the others. This characteristic provides a better energy source for microorganisms, thus favoring enzymatic activity.

In ecosystems where *Araucaria* was predominant (NF, RF1, RF2, RF, and ANF), the urease activity showed little variation, and PETAR NF was the most dissimilar to the other forests. The vegetation of this forest features is typical of a dense ombrophilous forest.

Fluorescein diacetate hydrolysis has been positively correlated with soil respiration (Schnürer & Rosswal, 1982); however, the heterotrophic activity was not high in terms of basal respiration in any of the forests evaluated. This suggests that these ecosystems are capable of conserving energy (lower losses of C-CO₂ and greater accumulation of C and N in microbial biomass), indicating their more advanced succession stage.

Enzymes are essential for element cycles in the soil. Because enzymes are synthesized mainly by soil-grown organisms, conditions that favor microbial activity, such as the presence of vegetation

(rhizosphere), also favor enzymatic activity. Eivazi & Tabatabai (1988) described the significant correlation between β -glucosidase activity and organic matter in the soil; the enzyme will hydrolyze both cellobiose and oligosaccharides with the release of sugar (glucose), an energy source for microorganisms. Generally, the results obtained in this study showed similar potentials for β -glucosidase activity in all forests, with no statistically significant differences among the sampling periods.

The results obtained in both parks evaluated in this study using principal component analysis (PCA) are in agreement with those reported by Salamanca et al. (2002) and Dinesh et al. (2003), who also found that the MB-N and MB-C contents were the properties most sensitive to changes in conservation stage and anthropogenic interference in the forests evaluated in those studies.

The scatter graph of the evaluated forests in this study, relative to the two ordination axes, shows dissimilarity between the seasons and between the conditions of conservation and anthropogenic interference in these areas. These effects were not reliably detected by univariate analysis.

The results by PCA demonstrate a seasonal influence on the soil chemical and biochemical properties, especially in the surface layers, where oscillations in moisture and temperature are greater (Campbell & Biederbeck, 1976; Martinez & Ramirez, 1978; Cattelan & Vidor, 1990).

CONCLUSIONS

1. The soil chemical and biochemical properties evaluated in this study were sensitive indicators (with the exception of the variable K) of differences in the conservation stage and anthropogenic interference in *Araucaria* forests.
2. Although some differences were found in the biological properties of the soil, the forests evaluated in this study were efficient in terms of use and conservation of energy. The qCO_2 was low in all forests, suggesting an advanced stage of succession.
3. The sampling season (dry/rainy) primarily influenced the properties MB-C, MB-N, BR, and qCO_2 .

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