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Soil Macrofauna as a Soil Quality Indicator in Native and replanted *Araucaria angustifolia* Forests

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ABSTRACT: Studies on soil quality in *Araucaria* forests contribute to understanding changes in this ecosystem and serve as a tool in conserving its biodiversity. The aim of this study was to evaluate the potential of soil macrofauna in discriminating native and replanted *Araucaria* forests for selection of soil quality indicators. Native (NF) and replanted (RF) *Araucaria angustifolia* forests were evaluated in three regions of the state of São Paulo, representing three true replications. Fifteen soil samples were collected in each area for evaluation of the physical, chemical, and microbiological properties and the macrofauna through use of monolith excavation and the manual screening method [Tropical Soil Biology and Fertility (TSBF) method]. In addition, we evaluated the weight of the surface litter dry matter and the C, N, and S contents. The abundance of macrofauna was subjected to Principal Components Analysis (PCA) and the physical, chemical, and microbiological properties were used as explanatory environmental variables for changes in the soil community. These variables and the macrofaunal properties were applied in analyses of variance and in canonical discriminant analysis (CDA) to indicate the best properties for discrimination of the forests studied. The abundance of macrofaunal groups was influenced by the state of conservation of the forest and by the sampling period; the native forest and the summer season provided greater diversity of taxonomic groups. The richness of taxonomic groups was the property that most contributes to discriminating reforested areas from native forests. The Oligochaeta group was a prominent indicator of soil quality and/or environmental disruption in *Araucaria* forests. Soil moisture, total porosity, and S content in the surface litter were likewise variables that contributed to distinguishing the areas. The ecological indexes (diversity, dominance, and equitability) were not sensitive to the changes in macrofaunal properties in the forests studied.

Keywords: multivariate analysis, biodiversity, bioindicators.

INTRODUCTION

The species *Araucaria angustifolia* (Bert.) O. Kuntze is a gymnosperm predominant in the Mixed Ombrophilous Forest in the Atlantic Forest domain (Souza et al., 2015). Intense use of *Araucaria* because of the quality of its wood, and increasing economic activity resulted in reduction of these forests (Ribeiro et al., 2009). In the state of São Paulo, *Araucaria* forests are quite fragmented and cover a small territorial area, less than 4 % of their original area (Mantovani et al., 2004; Kronka et al., 2005), leading to a critical situation for conserving the biodiversity of this ecosystem (Baretta et al., 2007a; Ribeiro et al., 2009; Souza et al., 2015) and placing *Araucaria angustifolia* on the list of "critically endangered" species worldwide (Thomas, 2013).

Conservation of remaining Mixed Ombrophilous Forests in fully protected conservation areas and restoration of this ecosystem in already degraded areas is a promising alternative in preserving this species and associated biodiversity, both above and below the soil level (Cardoso and Vasconcellos, 2015). The soil under native and reforested *Araucaria* forests in the state of São Paulo has high acidity and low exchangeable base and P contents, making the organic matter from decomposition of plant residues the main source of many nutrients for plants and the biological community (Carvalho et al., 2012; Pereira et al., 2013).

The soil biological community (mainly invertebrates), in the search for food and habitat, brings significant changes in the physical, chemical, and biological properties of the soil, although it too is regulated by climate, soil properties (soil structure and fertility), and the quality and amount of surface litter (Lavelle, 1996; Lavelle et al., 2006). The intensity of this regulation, together with the complex interaction between biotic and abiotic factors of the soil, defines the support capacity of this soil. In this respect, maintaining native *Araucaria* forests and improving areas in which it is replanted can ensure the sustainability and preservation of this ecosystem. Nevertheless, assessing soil quality is a complex task, which involves the use of indicators (Schloter et al., 2003; Cardoso et al., 2013).

In addition to the traditional physical and chemical properties (Schoenholtz et al., 2000; Baretta et al., 2008a; Pereira et al., 2013), biological (microbiological and macrofaunal) soil indicators should be included so that, together, they can better reflect the processes that act on soil quality (Bastida et al., 2008; Baretta et al., 2010; Cardoso et al., 2013; Vasconcellos et al., 2013a). Soil invertebrates perform different functions in land ecosystems and are represented by more than 20 taxonomic groups (Korasaki et al., 2013; Lavelle et al., 2014). In addition to their ecological function, soil invertebrates are also used as indicators of soil quality or of environmental disruptions because they are widely distributed and abundant, yet they are quite sensitive to the diverse human interventions made in the ecosystems (Baretta et al., 2010; Vasconcellos et al., 2013b; Segat et al., 2015).

Results from previous studies developed in native and replanted *Araucaria* forests show greater diversity and abundance of invertebrates associated with a more conserved forest, such as in the case of spider families (Baretta et al., 2007a), springtails (Baretta et al., 2008b), and earthworm species (Baretta et al., 2007b). A clear separation between the native forest, which was more conserved, and the replanted forests was observed in all these studies (Baretta et al., 2010). However, studies related only to soil quality in comparing preserved and degraded *Araucaria* forests could not be found.

Studies already published led to an understanding that soil macrofauna can also be used as a quality indicator in areas of native and replanted *Araucaria* forests. Thus, the aim of this study is to evaluate the potential of soil invertebrate macrofauna and of explanatory environmental variables in distinguishing native and replanted *Araucaria* forests and in selecting the best indicators of soil quality for this type of ecosystem, through multivariate analysis techniques.

MATERIALS AND METHODS

Description of the study areas

The study was conducted in Mixed Ombrophilous Forest areas of the Atlantic Forest biome in the state of São Paulo (SP). Plots of 0.5-1.0 ha were selected in areas of native forest (NF) and replanted forest (RF) with *Araucaria angustifolia* in three regions of the state. The areas of each region were considered true replicates. Detailed information on the physical-chemical properties of the soil and of the floristic composition of the areas studied can be obtained in Santos et al. (2009), and Pereira et al. (2013).

The first location for sampling NF and RF was subdivided into area 1 for native forest and area 2 for replanted forest (Pereira et al., 2013). The areas belong to the Bananal Ecological Station in the Serra da Bocaina. In the first sample collection period, in the winter (August), the mean temperature was 17 °C and rainfall was 40 mm. In the second period, in the summer (February), the mean temperature was 22 °C and the month of highest rainfall (January) registered more than 300 mm. In the NF area, there are 178 species of trees and bushes distributed in 46 families and 89 genera, according to Santos et al. (2009) and Pereira et al. (2013).

The second region studied (with area 3, NF) is in the Itaberá Ecological Station in the municipality of Itaberá, SP. Mean temperature in the winter was 17 °C and rainfall was 45 mm; in the summer, the mean temperature was 25 °C and the highest rainfall in January was 420 mm. The detailed floristic composition of this area can be found in Santos and Ivanauskas (2010) and the location and soil properties in Pereira et al. (2013). In this same region (with area 4, RF) is the Itapeva Experimental Station in the municipality of Itapeva, SP, that has been in replanted forest for 45 years, with predominance of *Araucaria* and understory species. More detailed descriptions regarding location (latitude, longitude, and altitude) and soil properties can be found in Pereira et al. (2013).

The third region is in the Alto Ribeira State Tourist Park (*Parque Estadual Turístico do Alto Ribeira*) (with area 5, NF) in the municipality of Barra do Chapéu and (with area 6, RF) in the municipality of Iporanga. In the winter sampling period, April was the driest month (<50 mm) and June was the coldest month (mean of 17 °C), whereas in the summer, the rainiest month was January (more than 400 mm) and mean temperature was 24 °C. Details regarding flora and soil can be found in Souza (2008) and Pereira et al. (2013). The climate in all these regions is humid mesothermal, according to the Köppen's climate classification system. Information on the physical and chemical properties can be found in Pereira et al. (2013). Each forest was represented by three true replications and, in each one, 15 soil samples were collected at random for evaluation of soil macrofauna and physical-chemical and microbiological properties.

Evaluation of soil macrofauna

The Tropical Soil Biology and Fertility (TSBF) method was used for evaluation of soil macrofauna (Anderson and Ingram, 1993). Samples were collected in the winter (August) and in the summer (February). A total of 15 soil monoliths were collected per area (0.25 × 0.25 m) at the depth of 0.00-0.20 m in each sample collection period (Baretta et al., 2007a). Before removing the soil monolith, the surface litter was sampled in the same area. The invertebrates were separated from the surface litter and from the soil and identified in taxonomic groups with the aid of a stereoscopic microscope. After this separation, the abundance, richness, Shannon's diversity index, Simpson's dominance index, and Pielou's evenness index were calculated (Odum, 1983).

For determination of physical, chemical, and microbiological properties, the soil was sampled with the aid of an auger at the depth of 0.00-0.20 m, and five simple samples were collected at each collection point (tree) to combine in a compound sample.

Dry matter, C, N, and S of the surface litter

The dry matter of the surface litter was determined (55 °C until constant weight). This dry matter was then ground, passed through a 100 mesh (1.7 mm) sieve, and the C, N, and S contents were determined by dry combustion in a simultaneous elemental analyzer (Elementar, Vario EL cube) for C, N, S, and H.

Soil chemical and physical properties

The soil samples were sieved through a 2 mm mesh and homogenized. The pH was determined by potentiometry in a 0.01 mol L^{-1} CaCl$_2$ solution. Phosphorus, Ca^{2+}, and Mg^{2+} were extracted with ion exchange resin. Potassium was extracted by Mehlich-1 and determined by flame emission spectrometry. Phosphorus was determined electrophotometrically by the molybdenum blue complex, and Ca^{2+} and Mg^{2+} by atomic absorption spectrometry. The H+Al were determined by a potentiometer in SMP solution at pH 7.0. We extracted Al from soil using a KCl solution (1 mol L^{-1}) and determined by titration with 0.025 mol L^{-1} NaOH. Organic carbon (OC) was determined by colorimetric procedures through oxidation of organic matter with Na$_2$Cr$_2$O$_7$.2H$_2$O and H$_2$SO$_4$, according to Raij et al. (2001).

Soil moisture content was determined after drying the soil in a laboratory oven at 105 °C for 48 h. For particle size analysis, the sand fraction was obtained through sieving (0.053 mm), clay fraction by the hydrometer method (Gee and Or, 2002), and silt by the difference between the quantity of soil and the quantity of sand plus clay in the sample. In the winter, at the same collection points of samples for chemical analyses, undisturbed soil samples were removed from the 0.00-0.05 m layer, which were saturated by capillarity, gradually raising the water layer up to two-thirds the height of the soil sample rings. They were then subjected to the matrix potentials of -1 and -6 kPa using a tension table. When water balance was reached, the samples were weighed and subsequently dried in a laboratory oven at 105 °C for 48 h to determine moisture content (Claessen, 1997) and soil density (Blake and Hartge, 1986). Macroporosity was obtained by the difference between the moisture content of the saturated soil and that corresponding to the application of 6 kPa (Claessen, 1997). Microporosity was estimated by the moisture content retained at the tension of 6 kPa. Total porosity was obtained by the sum of the macro and microporosity. Particle density was determined by the helium gas pycnometer method (Danielson and Sutherland, 1986).

Microbial biomass carbon, soil respiration, metabolic quotient, and dehydrogenase activity

Microbial biomass carbon (MBC) was determined by the fumigation-extraction method (Vance et al., 1987) using 100 g of soil. The samples were fumigated in ethanol-free chloroform for 24 h. The C of the fumigated and non-fumigated samples was extracted simultaneously with K$_2$SO$_4$ (0.5 mol L^{-1}) under shaking at 180 rpm for 30 min, oxidation with K$_2$Cr$_2$O$_7$ (66 mmol L^{-1}), and titration with ferrous ammonium sulfate (33 mmol L^{-1} [(NH$_4$)$_2$.Fe(SO$_4$)$_2$.6H$_2$O]) in the presence of barium diphenylamine sulfonate. The correction factor used in the calculations was 0.33, as proposed by Sparling and West (1988).

Microbial activity was evaluated by determination of basal respiration, the CO$_2$ released in incubated samples of 100 g of soil. The soil was incubated in glass containers, in the dark at 28 °C, containing a crucible with NaOH (0.5 mol L^{-1}), for 10 days. The C released as CO$_2$ was determined every 24 h through quantification of the remaining NaOH through titration with standard HCl solution (0.5 mmol L^{-1}), using phenolphthalein as an indicator and previous precipitation of the Na through addition of BaCl$_2$.2H$_2$O (4 mol L^{-1}), according to Alef and Nannipieri (1995). The results of CO$_2$-C and of microbial biomass carbon (MBC) were used to calculate the metabolic quotient (qCO$_2$), as described in Anderson and Domsch (1993).

Dehydrogenase enzyme activity (DehA) was determined in 5 g soil samples with natural moisture and the addition of 5 mL of a triphenyltetrazolium chloride (TTC) solution, 1.0 % (v/v), incubated at 37 °C for 24 h. Enzyme activity was measured in a spectrophotometer at 485 nm (Casida et al., 1964).

Statistical analysis

The results of density, richness of taxonomic groups, and the indexes of Shannon diversity (H), Simpson dominance (Is), and Pielou's evenness (*e*) were subjected to analysis of variance (two-way Anova), and the means were compared by the LSD test (p≤0.05) by the SAS 8.2 statistical program (Statistical Analysis Systems Institute Inc., 2002).

Principal Component Analysis (PCA) was conducted using the CANOCO 4.0 program (ter Braak and Smilauer, 1998). Chemical properties [$pH(H_2O)$, P, Ca^{2+}, Mg^{2+}, K^+, Al^{3+}, OC, and H+Al], physical properties [bulk density (BD), macroporosity (Ma), microporosity (Mi), total porosity (TP), and soil moisture (Moi)], properties of surface litter [carbon (C-lit), nitrogen (N-lit), and sulfur (S-lit)], surface litter dry matter (SLDM), soil microbiological properties (MBC, CO_2-C, DehA, and $q$$CO_2$), and the diversity indexes (H, Is, and *e*) were later used in the PCA as explanatory environmental variables for modifications in the macrofaunal community.

Only the physical properties (Moi, BD, Ma, and TP), soil chemical properties (P, OC, Ca, H+Al), surface litter chemical properties (C-lit, N-lit, S-lit), SLDM, microbiological properties (MBC, $q$$CO_2$, and DehA), and the diversity indexes (H, Is, and *e*) that were significant (p<0.05) were used as explanatory environmental variables, thus increasing the robustness of the analysis.

In addition, the environmental variables and the main properties of the macrofauna were subjected to canonical discriminant analysis (CDA) to identify which of them were most relevant in separation of the areas (Cruz-Castillo et al., 1994; Baretta et al., 2005). The standardized canonical coefficients (SCC), the correlation coefficient (*r*), and the parallel discrimination rate (PDR) (PDR = r × SCC) were calculated. The values of the coefficient of the parallel discrimination rate (PDR) indicate the force of the variable in distinguishing the areas studied in which the values are considered good (from 0.1 to 0.2), very good (from 0.2 to 0.4), optimal (from 0.4 to 0.8), and excellent (above 0.81) (Baretta et al., 2010).

RESULTS AND DISCUSSION

The density of soil invertebrates (ind m^{-2}) in the summer were higher in the NF (3,018 ind m^{-2}) than in the RF (2,115 ind m^{-2}) (p<0.05). However, in the winter, there were no significant differences (p>0.05) between NF (1,373 ind m^{-2}) and RF (1,132 ind m^{-2}). These values are higher than those found by Merlim (2005) and Baretta et al. (2010) in a native and reforested *Araucaria* area in the Campos de Jordão State Park and by Vasconcellos et al. (2013b) in the winter in forest restoration areas in the state of São Paulo. The increase that was observed may reflect the higher number of sampled areas of the same system (true replications), and the better state of conservation of the areas studied. This highlights the need to adopt true replications in the study of soil macrofaunal biodiversity and studies over several years.

Regardless of the sample collection period, the groups of social insects Isoptera and Hymenoptera were those with the greatest relative frequency in the soil community in all the areas. The relative frequency of ants increased in the summer compared to winter, regardless of the type of forest. These groups have been reported as most frequent in soils of different forest ecosystems (Aquino et al., 2008; Baretta et al., 2011; Nunes et al., 2012; Cardoso et al., 2013), including the soils of the *Araucaria angustifolia* forest (Baretta et al., 2010), mainly due to their competitiveness in the environment (Paula and Lopes, 2013).

The relative frequency of Oligochaeta was greater in RF areas, which shows the importance of this group as a good indicator of soil quality and/or environmental disruption (Baretta et al., 2007b). The other groups were Coleoptera, Chilopoda, and Diplopoda, which exhibit similar density and frequency among the areas and periods of the year evaluated. This is explained by the fact that such groups have similar life cycles and stable densities throughout the year (Tuf and Tufová, 2008; Brown et al., 2009).

The mean values of richness of taxonomic groups of invertebrate soil macrofauna in the two sample collection periods were significantly greater in the NF (10.04 in the winter and 11.82 in the summer) than in the RF (8.37 in the winter and 9.75 in the summer), and they were always greater in the summer. This indicates that the best sample collection period for studying the abundance and diversity of the soil macrofaunal community is the end of the rainy period. These results corroborate those of Merlim (2005) and Baretta et al. (2010), who obtained similar results, always greater in native and preserved forests. In general, the greatest richness of soil macrofaunal groups was associated with the native forest environment in the summer, where the greater diversity of plant cover may have resulted in greater heterogeneity and amount of surface litter and, consequently, greater food supply, as well as soil and climate conditions favorable to soil fauna.

Greater density and richness of soil fauna have been observed in soil under native forest cover in more advanced successional stages, compared to reforested and pasture areas (Moço et al., 2005; Menezes et al., 2009; Vasconcelos et al., 2013b), including native forest with *Araucaria angustifolia* (Baretta et al., 2010). In this respect, Gibson et al. (2011) alert that native and/or preserved secondary forests are irreplaceable in the task of preserving tropical biodiversity.

There were no significant differences in Shannon's diversity (H), Simpson's dominance (Is), and Pielou's evenness (*e*) ecological indices between NF and RF, although the NF exhibited greater richness of taxonomic groups in relation to RF. The predominance of some groups of soil invertebrates in NF, such as Isoptera, Hymenoptera, Coleoptera, and Chilopoda, with positive and higher scores in relation to the others, decreases equitability among the groups, and is directly reflected in diversity. As consequence of this effect, the ecological indices of diversity H, Is, and *e* showed weak association with the NF area (Figure 1a). Lima et al. (2010), studying the effect of soil use on macrofaunal diversity and its relation to chemical properties in different agrosystems, found that, regardless of the sample collection period, the soil under native forest cover had the lowest values of Shannon's and Pielou's indices, associated with the dominance of individuals of the Isoptera group. Thus, in the present study, considering not only the soil macrofaunal variables, but also the explanatory environmental variables, it was clear that the measure of diversity that best represented the areas of study was the richness of taxonomic groups, in comparison to the other ecological indices (H, Is, and *e*).

Principal Component Analysis (PCA)

Principal component analysis (PCA) revealed that the first two principal components (PC1 and PC2) explained 39.3 % of the total variability of the data in the winter, with 21.0 % explained by PC1 and only 18.3 % by PC2 (Figure 1a). In the summer, analysis of the variables by PCA in the first two axes explained 40.6 % of the variance (Figure 1b). The results of PCA show the separation of the forests studied, and that the groups of soil macrofauna and the explanatory environmental variables occurred in a differentiated manner between NF and RF in the two sample collection periods.

The RF area, in both periods, had greater earthworm density, especially *Ponthoscolex corethrurus* (a species native to Brazil, but a peregrine species in these areas) and the exotic genus *Amynthas* (data not shown) in relation to the other groups of macrofauna (Figures 1a and 1b). In contrast, the low abundance of these genera in the NF area is an indication of the good state of preservation of these areas, since these species are
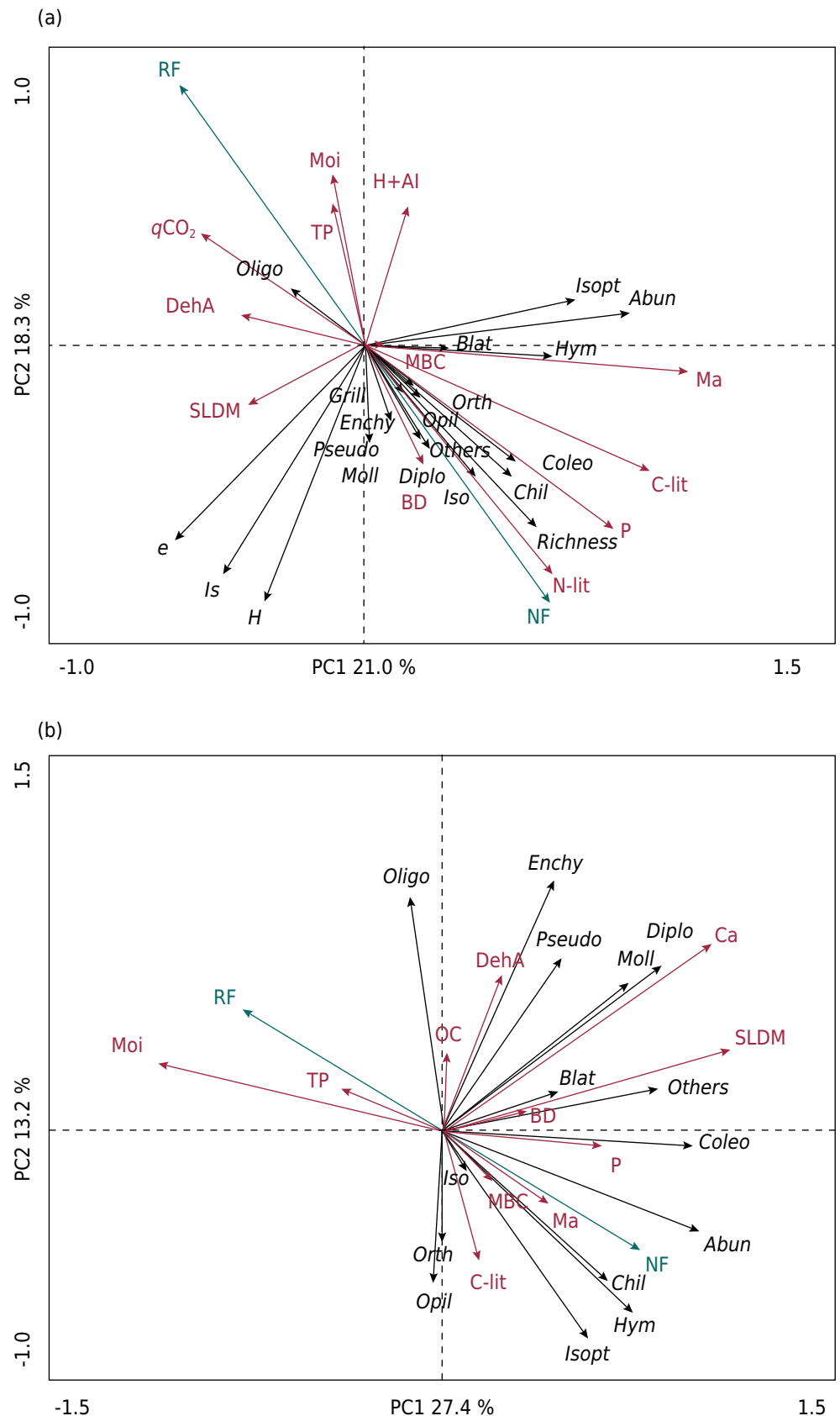
Figure 1. Relation between Principal Components 1 and 2, discriminating native Araucaria forests from reforested ones. Taxonomic groups: black arrows; explanatory environmental variables: red arrows, in winter (a) and summer (b). Diplo: Diplopoda; Iso: Isopoda; Coleo: Coleoptera; Hym: Hymenoptera; Isopt: Isoptera; Blat: Blattodea; Orth: Orthoptera; Pseudo: Pseudoscorpionidae; Chil: Chilopoda; Opil: Opilionidae; Moll: Mollusca; Enchy: Enchytraeidae; Grill: Grilloblatodea; Others: other less abundant groups. Dens: density; H: Shannon's diversity index; Is: Simpson's dominance index; e: Pielou's evenness index; OC: organic carbon; C-lit: litter carbon; N-lit: litter nitrogen; Moi: moisture; TP: total porosity; Ma: Macroporosity; BD: bulk density; SLDM: surface litter dry matter; MBC: microbial biomass carbon; DehA: dehydrogenase activity; $q\text{CO}_2$: metabolic coefficient.

considered to be invasive (Brown and James, 2007). Furthermore, the higher density of earthworms in RF is considered to be an indicator of improvement of soil quality in this location, especially through the presence of the genus *Amynthas*, which, though exotic, lives in soils with higher organic matter and surface litter content (Brown and James, 2006; Brown et al., 2006, 2009; Bartz et al., 2014). However, these two genera of earthworms (*Amynthas* and *Ponthoscolex*), found in the RF area, have already been observed in some ecosystems derived from the Atlantic Forest, especially areas with a certain human intervention, and have already been reported as indicators of disruptions in different ecosystems (Fragoso et al., 1997; Burtelow et al., 1998; Brown and James, 2006; Marichal et al., 2010), including native *Araucaria angustifolia* forests in the state of São Paulo (Baretta et al., 2007b).

The PCA also showed that the RF was more associated with soils with higher moisture content, total porosity (Figures 1a and 1b), exchangeable acidity, DehA and qCO$_2$ (Figure 1a). In this area, the higher soil moisture content, regardless of the sample collection period, brought about especially by the accumulation of a greater diversity of surface litter, provided a soil environment more favorable to colonization by Oligochaeta and less favorable to most of the other groups of macrofauna (Pimentel et al., 2006; Rosa et al., 2015), which helped to explain their greater density in RF. The greater density of Oligochaeta in the RF area may also indicate that they are pioneers in preparing this environment, favoring colonization by other groups of macrofauna in the long term, reflected in their being known as "ecosystem engineers" (Brown et al., 2000; Baretta et al., 2011).

In both sample collection periods, the groups Chilopoda, Coleoptera, Isopoda, and Diplopoda showed greater association with NF, resulting in greater richness of taxonomic groups in this forest (Figure 1a). These groups were also more abundant in preserved forests than in replanted forests of this species, whether subjected to human intervention or not (Baretta et al., 2010; Rosa et al., 2015). The presence of predator and saprophyte groups in the NF, even in the winter period, indicates greater availability of food related to the greater variety of plant cover, which favors the colonization and development of these groups (Rousseau et al., 2013). Besides these, other groups were associated with NF, such as Enchytraeidae, Mollusca, Pseudoscorpionida, and Hymenoptera (Figure 1a).

The forest environments with the more conserved native *Araucaria* forest provided the best soil conditions for colonization of macrofauna, with the exception of Oligochaeta, indicating that these groups were sensitive to environmental changes present in the RF area, probably related to changes in the soil/surface litter environment, the main habitat and source of resources necessary for invertebrates (Lavelle et al., 2006; Vasconcelos et al., 2013b). The NF was also associated with soils that had a higher content of MBC, C, and N in the surface litter, a high P content in the soil, and greater aeration ability and soil BD, which may explain the association of most of the macrofaunal groups with this area. The combined and more frequent occurrence of predators and saprophytes, associated with P content in the soil and C and N content in the surface litter, indicates improvement in the biological quality of the soil and suggests that the surface litter represents a nutrient and energy reserve that stimulates soil macrofauna (Haddad et al., 2012). In addition, the NF area is characterized by maintenance of more diversified and mature plant cover, which allows greater input of surface litter, root density, reduction in thermal oscillations in the soil, and nutrient cycling, essential for maintenance of a diversified soil community (Aquino et al, 2008; Baretta et al., 2011).

Canonical Discriminant Analysis (CDA)

The Wilks' Lambda multivariate statistical test for the invertebrate macrofaunal properties and environmental variables indicated significant differences between sampling periods and the areas studied ($p < 0.0001$) in regard to canonical discriminant function 1 (CDF1) and 2 (CDF2), and a CDA was performed for each sampling period (Figures 2a and 2b). The statistical model used in the CDA explained practically all the variability of the data.

High canonical correlation was also observed for the dry period (winter) (0.95) and rainy period (summer) (0.93) (p<0.0001). High canonical correlation values indicate high association among the properties analyzed and the areas where these properties dominated (Baretta et al., 2010).

Table 1 indicates the standardized canonical coefficients (SCC) of CDF1 and CDF2 for the two sampled areas with *Araucaria*, considering all the environmental properties and the soil macrofauna analyzed (Table 2). From the result of the CDA, we found that CDF1 clearly separated the NF area (with higher SCC values) from the RF area, in both sampling periods (Figures 2a and 2b).

The coefficient of the parallel discrimination rate (PDR) is considered the most adequate parameter for evaluating the effect of discrimination by the soil properties analyzed within the areas. Thus, the positive values of PDR indicate effects of separation between the areas in regard to the property analyzed, whereas the negative values indicate similarities (Baretta et al., 2008a). In general, the PDR value of each property was differentiated for each sampling period (Table 1).

In the winter sampling period, within CDF1, the properties of soil moisture (0.28), S-lit (0.18), TP (0.16), K (0.11), and CO_2-C (0.10) had the highest positive values of coefficients of PDR, better explaining the separation between the areas studied (Table 1). The environmental variables considered indicators of changes in the areas in CDF1 in the summer were TP (0.25), Moi (0.20), S-lit (0.17), and H+Al (0.10).

Total porosity (TP) of the soil is related to its capacity to provide conditions favorable for growth and development of the plant root system, and it is highly influenced by soil structure. Moisture content is related to the density of soil plant cover and the continuous presence of a thicker layer of surface litter, resulting in lower oscillation of temperature, besides the soil texture effect. The S content in the surface litter may vary according to the quality and diversity of plants in the areas of study; the same occurs with K, which is easily leached from the plant tissue to the soil (Torres and Pereira, 2008). Properties related to soil C and microbial activity (CO_2-C and MBC) also contributed to separation of the areas, as well as P and pH (H_2O), which modulate the action of CO_2-C and MBC, confirming their participation as soil quality indicators (Maluche-Baretta et al., 2006; Baretta et al., 2010).

In the winter sampling, within CDF1, the macrofaunal properties that had the highest positive values of the PDR coefficients were richness of taxonomic groups (0.13), Oligochaeta (0.03), and Chilopoda (0.03) (Table 2).
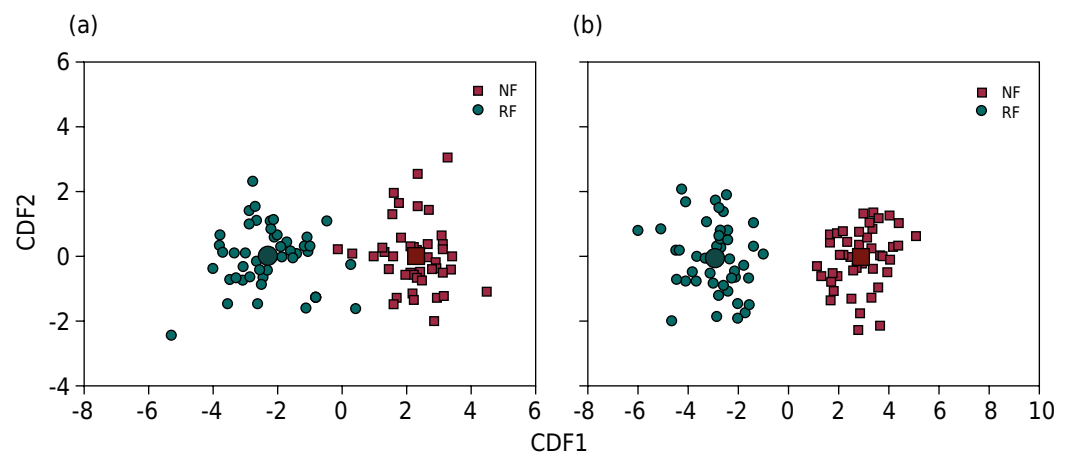

Figure 2. Standardized Canonical Coefficients (SCC) of the Canonical Discriminant functions 1 (CDF1) and 2 (CDF2), discriminating native Araucaria forests from reforested ones, when considering properties of environmental variables and of soil macrofauna sampled according to the TSBF methodology, in winter (a) and summer (b). The central black symbols represent the average of SCC for each área.

Table 1. Standardized canonical coefficients (SCC), canonical correlation (r), and Parallel Discrimination Rate (PDR: $r \times$ SCC) related to canonical discriminant analysis (CDA) containing environmental variables, in winter and summer, regardless of the site

Environmental variable	CDF1 (Winter)			CDF1 (Summer)		
	r	SCC	PDR	r	SCC	PDR
Microbial biomass carbon (MBC)	0.06	1.24	0.07	0.15	0.29	0.04
Basal soil respiration (CO_2-C)	-0.07	-1.46	0.10	-0.03	-0.64	0.02
Metabolic coefficient (qCO_2)	-0.16	0.57	-0.09	-0.21	-0.17	0.04
Dehydrogenase activity (DehA)	-0.13	0.32	-0.04	-0.09	0.15	-0.01
pH (H_2O)	-0.06	-1.16	0.07	-0.04	-1.19	0.05
Organic carbon (OC)	-0.04	-0.25	0.01	-0.02	0.30	-0.01
P	0.17	0.35	0.06	0.23	0.35	0.08
K^+	0.17	0.63	0.11	0.14	0.37	0.05
Mg^{2+}	0.01	0.44	0.01	0.05	-0.01	0.00
Al^{3+}	-0.02	0.41	-0.01	-0.11	0.97	-0.11
H+Al	-0.04	-0.46	0.02	-0.07	-1.35	0.10
Litter carbon (C-lit)	0.08	-0.93	-0.07	0.11	-0.41	-0.04
Litter nitrogen (N-lit)	0.26	-0.02	0.00	0.33	-0.18	-0.06
Litter sulfur (S-lit)	0.17	1.08	0.18	0.22	0.78	0.17
Soil moisture (Moi)	-0.35	-0.80	0.28	-0.31	-0.63	0.20
Soil bulk density (BD)	0.06	-0.50	-0.03	0.07	-0.86	-0.06
Macroporosity (Ma)	0.11	0.29	0.03	0.14	0.46	0.07
Total porosity (TP)	-0.18	-0.86	0.16	-0.23	-1.09	0.25
Surface litter dry matter (SLDM)	-0.01	0.47	0.00	0.11	0.14	0.02

Table 2. Standardized canonical coefficients (SCC), canonical correlation (r), and Parallel Discrimination Rate (PDR: $r \times$ SCC) related to canonical discriminant analysis (CDA) containing macrofaunal variables, in winter and summer, regardless of the site

Most frequent group	CDF1 (Winter)			CDF1 (Summer)		
	r	SCC	PDR	r	SCC	PDR
Coleoptera	0.04	0.50	0.02	0.14	0.05	0.01
Orthoptera	0.04	0.13	0.01	0.00	-0.19	0.00
Hymenoptera	0.05	0.08	0.00	0.11	0.02	0.00
Isoptera	0.02	-0.30	-0.01	0.05	-0.24	-0.01
Diplopoda	0.02	-0.16	0.00	0.00	-0.03	0.00
Chilopoda	0.15	0.18	0.03	0.16	0.00	0.00
Isopoda	0.10	0.00	0.00	0.01	0.13	0.00
Opilionidae	0.01	-0.13	0.00	-0.01	-0.50	0.00
Pseudoscorpionidae	0.03	-0.26	-0.01	-0.04	-0.13	0.00
Mollusca	0.10	0.09	0.01	0.14	0.41	0.06
Blattodea	0.06	-0.16	-0.01	0.07	-0.21	-0.01
Griloblattodea	0.06	0.14	0.01	0.00	0.00	0.00
Enchytraeidae	-0.06	-0.09	0.01	-0.12	-0.17	0.02
Oligochaeta	-0.15	-0.21	0.03	-0.19	-0.22	0.04
Others[1]	-0.01	-0.22	0.00	0.07	0.18	0.01
Other macrofaunal properties						
Abundance	0.03	0.00	0.00	0.08	0.00	0.00
Richness of taxonomical groups	0.11	1.18	0.13	0.17	0.91	0.15
Shannon's diversity index (H)	0.06	-1.28	-0.08	0.06	-1.24	-0.08
Simpson's dominance index (Is)	0.04	-1.07	-0.05	0.03	0.08	0.00
Pielou's evenness index (e)	0.03	2.22	0.06	0.01	1.26	0.01

[1] Others: Sum of less frequent groups

In the summer, within CDF1, the properties richness of taxonomic groups (0.15), Mollusca (0.06), and Oligochaeta (0.04) had the highest positive values of PDR coefficients (Table 2). Baretta et al. (2010) also found positive values of PDR coefficients for Mollusca, considered an indicator of changes in areas with *Araucaria*, regardless of the sample collection period. The other properties of the environmental variables and of macrofauna were less sensitive, with lower values of PDR, and they contributed less to the function recommended for separation between the areas (Table 2). This result confirms recommendation of the Oligochaeta group as important in discriminating between native and replanted *Araucaria* forests.

In the two sampling periods, we observed, through PDR values, that environmental and macrofaunal properties were effective in separating the areas studied, showing potential as indicators, because they provided separation between the areas, especially when a greater number of replications of the samples and of the ecosystems sampled (true replications) is observed, as in the present study (Rossi et al., 2006). Thus, for effective evaluation of soil quality in native and replanted *Araucaria* forests, there is the need for combined use of physical, chemical, and microbiological indicators, with the addition of the macrofauna, which play an important role in this ecosystem (Baretta et al., 2008a; Baretta et al., 2010).

CONCLUSIONS

The state of conservation of the *Araucaria* forest and the physical-chemical and microbiological properties of the soil were determining factors in the environmental conditions that influenced the distribution of the macrofaunal community in the areas with *Araucaria*. Such properties, together with the macrofauna, increase the robustness of evaluation of soil quality in areas with *Araucaria*.

The native forest with *Araucaria* that had greater floristic complexity provided greater diversity of soil macrofaunal groups, and this diversity is greatest in the summer.

Shannon's diversity, Simpson's dominance, and Pielou's evenness ecological indices were not sensitive to modifications in the soil macrofaunal properties in the forests studied.

Most of the soil macrofaunal groups were associated with the native *Araucaria* forest in the PCA and responded positively to improvement in environmental quality.

The most promising properties for separation between the areas with *Araucaria* were Oligochaeta, Chilopoda, Mollusca, group richness, soil moisture and total porosity, litter sulfur content, CO_2-C, and H+Al.

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