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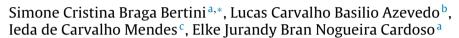
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Hierarchical partitioning for selection of microbial and chemical indicators of soil quality



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ABSTRACT

Statistical approaches, especially multivariate techniques such as hierarchical partitioning analysis (HP) and redundancy analysis (RDA), can be used to select appropriate variables for soil quality assessment. HP is usually applied to ecological data from plants and animals, but has not been applied to chemical and microbial properties such as those used as indicators of soil quality. Our aim was to show how these methods can be employed to find soil quality indicators, using soil microbiological, chemical and physical data to compare two forest types (native and reforested Brazilian Araucaria forests) in two locations in Southeast Brazil. We used RDA to investigate relationships among variables. Additionally, we quantified the independent effects of predictor variables: location, forest type, two specific seasons and some soil properties and used HP to examine how these environmental variables interact to influence soil microbial and chemical attributes. RDA showed that acid phosphatase and dehydrogenase activity, NO₂⁻ oxidizer numbers, basal respiration, metabolic quotient, pH, P and sand content were positive and significantly correlated with the native Araucaria forest, whereas arylsulphatase activity, denitrifier numbers, microbial biomass carbon, microbial quotient, TOC, S and clay levels were positively correlated with the reforested Araucaria. These preliminary results suggest that these variables are the best indicators of soil quality for Araucaria forests. However, HP, used as a complementary tool, showed that only dehydrogenase activity, pH and S variations were due more to forest type than to physical and chemical soil properties, and were resistant to the variation in the two seasons. Overall, our results indicated that dehydrogenase activity, pH and S are potential indicators that can be used to assess or monitor soil health in Araucaria forests. In conclusion, we demonstrated the usefulness of HP to find soil guality indicators. Similarly, other statistical methods, as RDA, could complement HP and increase the reliability of studies that consider simultaneous variables in soil science.

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Introduction

Since the advent of agriculture and the settlement of large populations, humans have interfered strongly in soil ecosystems (Goudie 2006). Technological development and population growth have caused greater impact on soil function from land use, agricultural practices, soil pollution, soil sealing and climate change (Goudie 2006; Holloway and Stork 1991). Soil quality monitoring is needed

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http://dx.doi.org/10.1016/j.pedobi.2014.06.001 0031-4056/© 2014 Elsevier GmbH. All rights reserved. to help recover soil resources and guide more sustainable soil use practices. According to Doran and Parkin (1994), soil quality is commonly defined by soil function, which considers the set of physical, chemical and biological properties that act on ecosystem function and contribute to plant growth, water and nutrient cycling, carbon storage, organic matter decomposition, and detoxification of hazardous compounds, among others. Thus, soil quality indicators are one way to evaluate and monitor soil function (Doran and Parkin 1994; Holloway and Stork 1991; Schoenholtz et al. 2000).

Questions have been raised about which soil properties could be reliable indicators of anthropogenic and natural impacts on soil function (Doran and Parkin 1994; Schoenholtz et al. 2000). Thus, soil parameters have been investigated to find suitable soil quality indicators for monitoring land use, soil degradation and the effects

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of different crop management systems (Doran and Parkin 1994; Holloway and Stork 1991; Schoenholtz et al. 2000; Ritz et al. 2009; Turbé et al. 2010).

Microorganisms and related processes are intimately linked to soil function and can consequently serve as sensitive indicators of recent modifications to the soil environment (Doran and Parkin 1994; Brejda et al. 2000). However, the sheer number of soil quality variables and relationships among soil microbial, chemical and physical properties make it difficult to interpret results using traditional statistical methods (Brejda et al. 2000; James and McCulloch 1990; Thioulouse et al. 2012). Consequently, the use of multivariate methods to obtain consistent soil quality assessments and a global view of soil function has increased in recent years (Brejda et al. 2000; Mele and Crowley 2008; Mendes et al. 2012).

Redundancy analysis (RDA), principal component analysis (PCA), non metric multidimensional scaling (NMS), discriminant analysis, Kohonen self-organizing maps (SOM) and sum of squares multivariate regression tree (SS-MRT) models are some of the multivariate methods that have been used to assess soil quality (Brejda et al. 2000; Floch et al. 2009; Mele and Crowley 2008; Mendes et al. 2012). These methods have also been used to determine soil quality indicators under different land use and management strategies in agricultural and agroforestry systems (Brejda et al. 2000; Floch et al. 2009; Mele and Crowley 2008; Mendes et al. 2009; Mele and Crowley 2008), differentiate between treatments or areas (Floch et al. 2009; Mendes et al. 2012) and determine the ecotoxicological effects of heavy metal pollution on soil microbial activity (Pereira et al. 2006).

Soil properties that contribute to soil quality assessment can be intercorrelated (Brejda et al. 2000). Thus, the challenge is to identify the environmental factors (predictor variables – e.g. management type, season, location, soil depth, and others soil properties) that most explain variation in soil microbial and chemical properties (response variables). In other words, it is important to identify the response variables that are most influenced by the factors that affect the soil. These response variables would be strong candidates for soil quality indicators. Hierarchical partitioning analysis (HP) is a multiple regression method that efficiently measures the independent effect of each environmental factor on response variables and consequently identifies which variables are most likely to influence variation in the response variables (Chevan and Sutherland 1991; Mac Nally 1996).

Unlike other types of multiple regression analysis (including multiple logistic and multiple Poisson regression models) HP is less susceptible to intercorrelation between predictor variables (Chevan and Sutherland 1991). HP multicollinearity decreases as model fit increases. This is achieved by averaging the influence of each predictor variable estimated for all possible combinations in which this variable appears (Chevan and Sutherland 1991; Mac Nally 2000). Therefore, HP includes predictor variables that contribute to best fit and that most likely influence variation in the response variable (Mac Nally 1996, 2000).

HP is commonly used in ecological studies such as investigations on the environmental factors that influence bird and butterfly abundance, fish and mammal species occurrence and amphibian species richness in different environments (Heikkinen et al. 2004, 2005; McAlpine et al. 2006; Pont et al. 2005; Werner et al. 2007). HP has also been used to determine the impact of exotic species on biodiversity (Walsh et al. 2012) and the effect of pollutants, anthropogenic activities (agricultural practices, human population distribution, point source pollution and atmospheric deposition), physiography and climate on water quality (Hatt et al. 2004; Lambert and Davy 2011; Morrice et al. 2008; Varanka and Luoto 2012). HP is a robust and powerful tool that can help identify and elucidate modifications to biological systems caused by natural and anthropogenic factors at several organizational levels (Mac Nally 2000; Pont et al. 2005; Arenas et al. 2006). Although HP has been applied in various ecological and conservation studies, this statistical method has not been used to identify microbial and chemical indicators of soil quality. Moreover, because it is difficult to define general principles and predict soil ecological responses due to the complexity and heterogeneity of soils (Fierer et al. 2009), HP could contribute to finding common mechanisms that drive ecological processes in soil.

Given the potential of HP and the difficulty of using traditional statistical methods to find effective soil guality indicators, our objective was to demonstrate the usefulness of RDA and HP for selecting an appropriate set of variables to assess soil quality in native and reforested Brazilian Araucaria forest areas. In order to exemplify the use of RDA and HP for soil quality investigations, we used a data set including physical, chemical and microbiological soil attributes, collected from native and reforested Brazilian Araucaria forests in two different regions in the southeast of São Paulo state, Brazil. Araucaria forests are part of the Atlantic Forest Biome (Shimizu and Oliveira 1981), composed mainly of the native pine Araucaria angustifolia (Bertoloni) Otto Kuntze, a plant species endangered by human exploitation (Thomas 2013). Although replanting has been used to restore Araucaria forests, the soil conditions of the planted forest have not been compared to those of the native Araucaria forest.

Thus, we used RDA to reduce data-set complexity and to determine relationships among variables (response and predictor) to identify the effect of forest type on soil attributes in two contrasting seasons. HP was used to determine the significant effects of each individual predictor variable on soil chemical and microbial properties and thus identify response variables that were mainly influenced by forest type and, to a lesser extent, by the two contrasting seasons.

Materials and methods

Study areas

The study was conducted at two state parks (separated by approximately 350 km) in Sao Paulo state: Alto Ribeira Tourist State Park (hereafter referred to as Alto Ribeira) located in the southeast of São Paulo state, Brazil (24°20' S, 48°36' W, 850 m above sea level, Cfb climate (Köppen)) and Campos do Jordão State Park (Campos do Jordão) in the northeast of the state (22°40' S, 45°27' W, 1630 m above sea level, Cwb climate (Köppen)). Two forest types were selected for each Park: Native Araucaria Forest (NAF) and Reforested Araucaria (RFA). The native Araucaria forests were dominated by A. angustifolia, Matayba elaeagnoides, Ocotea puberula and Ocotea elegans in the Alto Ribeira, and A. angustifolia, Podocarpus lambertii, Myrceugenia miersiana and Cinnamomum sellowianum in the Campos do Jordão. The reforested Araucaria areas were free from human intervention since planting (1984 at Alto Ribeira and 1959 at Campos do Jordão) and were mainly composed of A. angustifolia, P. lambertii, Tibouchina sp. and Paspalum grass. The Alto Ribeira soil was an Oxic Dystrudepts and the Campos do Jordão soil was a Humic Hapludox (Soil Survey Staff 2010).

Soil sampling and preparation

Soil samples were collected at a depth of 0-20 cm in September 2007 (end of the dry season) and March 2008 (middle of the rainy season) from areas of approximately 5000 m² in each forest type. Three transects ($40 \text{ m} \times 10 \text{ m}$) were selected at each representative area. Five *A. angustifolia* trees were randomly chosen along each transect and a soil probe was used to take three soil subsamples from under the canopy of each tree. The three subsamples were pooled to form a composite soil sample. Thus, five soil samples from

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