# Chapter 18 Diversity of the Shrub-tree Layer in a Brazilian Cerrado Under Nitrogen, Phosphorus and Nitrogen Plus Phosphorus Addition

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Abstract The aim of this study was to compare the diversity of the shrub-tree layer in fertilized and unfertilized plots in a cerrado stricto sensu area in Central Brazil. The experiment was conducted in 16 plots of 15 m×15 m arranged in a completely randomized design with tree fertilization treatments (+N, +P, +NP), and an unfertilized treatment (control) in the Ecological Reserve of the Instituto Brasileiro de Geografia e Estatística (RECOR-IBGE), Federal District, near Brasília. Treatments were applied from 1998 to 2006. Vegetation surveys were performed during January and July 2008 on all trees and shrubs with circumference > 5 cm at ground level. Indices of diversity of Shannon (H'), evenness of Pielou (J') and similarity of Sørensen were calculated. Control plots contained 479 individuals of 47 species, belonging to 29 families. In the nitrogen (N) plots, 461 individuals were registered, belonging to 53 species and 34 families while 448 individuals of 54 species from 31 families were registered in phosphorus (P) plots and 336 individuals of 40 species, belonging to 24 families in nitrogen + phosphorus (NP) plots. The N (H'=3.20; J'=0.80) and NP (H'=2.89; J'=0.78) plots showed lower Shannon (H') and Pielou (J') indices relative to the control plots (H'=3.40; J'=0.88). The Sørensen floristic similarity was high between fertilized and control plots but decreased in the following order: P plots (0.81), N (0.78) and NP (0.76) plots. Fertilization shifted species density and dominance patterns in comparison to unfertilized plots. Nitrogen and NP addition decreased the evenness, species diversity and provided the least floristic similarity relative to the control plots. Density and dominance changes resulted in differences in species importance values among treatments. Simultaneous addition of N and P affected density, dominance, richness and diversity patterns more significantly than addition of N or P separately.

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## 18.1 Introduction

Cerrado is the second largest Brazilian biome and it is characterized by a vegetation mosaic ranging from grassland to forest formations (Eiten 1972) where fire is a common occurrence (Kauffman et al. 1994). The attributes of Cerrado vegetation were considered to be driven by seasonality of water availability (Warming 1908). Later, Rawitscher (1948) observed that soil water is available to deep roots throughout the year. Alvim and Araújo (1952) linked the distribution of vegetation to low soil pH and calcium (Ca) concentration. Goodland (1971) proposed that aluminium (Al) would have a toxic effect on Cerrado plants, which, together with low soil nutrient content, determine the scleromorphic characteristics of Cerrado vegetation. Furthermore, floristic composition were associated with nutritional differences between dystrophic and mesotrophic soils (Lopes and Cox 1977). In addition to variation among vegetation types, phytosociological and floristic variations also occur due to fertility gradients and soil physical characteristics (Haridasan 1987). Among the Cerrado plant adaptive strategies, economy of nutrients is a crucial point in the establishment in highly weathered soils (Meinzer et al. 1999). Thus, phenological groups have different root system and resource exploration strategies (Scholz et al. 2008). Currently, it is known that soil fertility significantly influences species composition (Ratter and Dargie 1992). Changes in global biogeochemical cycles due to anthropogenic emissions and increasing human disturbance have affected processes, biotic interactions and resource availability patterns in different ecosystems, with changes in vegetation structure and composition (Vitousek et al. 1997; Bobbink et al. 2010). Particularly in tropical systems, land use changes due to agricultural intensification and urbanization have altered the nitrogen (N) cycle (Filoso et al. 2006). In Cerrado, one of world's biodiversity hotspots, land conversion has been substantial over the last 40 years. More than half of vegetation has been converted into pastures and croplands, with intensive use of chemical fertilizers (Klink and Machado 2005). Changes in soil chemical properties may possibly result in changes between soil-plant and plant-plant interactions patterns. In addition to individual nutritional adaptations, competition is an important factor in community establishment under altered nutrient availability conditions. The aim of this study was to compare shrub-tree layer diversity in fertilized (+N, +P, +NP)and unfertilized plots in a cerrado stricto sensu area in Central Brazil. The following hypotheses were tested: fertilizer addition will change soil nutrient availability (N and P) and species richness and diversity will be lower in fertilized plots. Changes in analyzed attributes will be greatest when N and phosphorus (P) are applied in combination.

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## 18.2 Material and Methods

#### 18.2.1 Study Area and Fertilization Treatments

The study was carried out in an area located in the Ecological Reserve of the Brazilian Institute of Geography and Statistics (RECOR/IBGE), near Brasília-Federal District, Brazil ( $15^{\circ}$  56' S,  $47^{\circ}$  53' N, average altitude = 1,100 m) in a cerrado sensu stricto area over dystrophic soil. RECOR-IBGE is part of environmental protection area Gama Cabeca de Viado, that has 10,000 ha of continuous protected native area. The climate is classified as Aw (Köppen's classification), with average annual rainfall varying between 1,100 and 1,700 mm. The average annual temperature is around 22 °C, daytime average relative humidity ranging from 80% in rainy season and 55% in dry season, with minimum values below 15%. Cerrado sensu stricto vegetation type is characterized by a continuous grass layer and a woody layer of trees and shrubs varying in cover from 10 to 60% and is the most common vegetation type, occupying approximately 43% of Cerrado region (Eiten 1972). The study area burned accidentally on two occasions, in 1994 and 2005. Soil is classified as Haplustox, which is deep, well drained, with 1:1 clay minerals and predominance of iron and aluminum oxides. This soil type is very acidic with low levels of base cations (Ca, Mg, K) and plant available P (Haridasan 1994). The fertilization experiment began in 1998 and the experimental design was completely randomized, with four nutrient addition treatments and four replicates randomly divided into 16 plots of 225 m<sup>2</sup>, separated by a 10 m buffer area. The treatments were: control (C; without fertilization), +N (single addition of ammonium sulfate ( $NH_{4}$ ),  $SO_{4}$ ), +1P (single addition of 20% superphosphate—Ca  $(H_2PO_4)_2 + CaSO_4$ .2H<sub>2</sub>O) and +NP (simultaneous addition of ammonium sulfate/20% superphosphate) applied in litter layer without incorporation. Each year, between 1998 and 2006, were annually added 100 kg ha<sup>-1</sup> of N, P and N+P, applied two times by year (beginning and end of rainy season). At the beginning of the experiment, soil nutrient concentrations did not differ significantly among plots (Kozovits et al. 2007).

### 18.2.2 Soil Sampling and Analysis and Vegetation Survey

In October 2007, a composite soil sample (two sub-samples) was collected, in each plot, at five depths (0–10, 10–20, 20–30, 30–40; 40–50 cm). Analysis were performed to determine pH in water and CaCl<sub>2</sub> (0.01 M), total N (micro Kjeldahl method), P (extraction with Mehlich 1 and colorimetric determination) and Al (extraction with 1 M KCl and titration with NaOH) (EMBRAPA 1999). In January and July 2008, we performed vegetation surveys in all plots including all trees and shrubs with circumference >5 cm at ground level. The phytosociological absolute and relative parameters of density, frequency and dominance were calculated for each species within each treatment. The phytosociological parameters were



Fig. 18.1 Soil chemical characteristics (mean and standard deviation) at five depths (0-10, 10-20, 20-30, 30-40, 40-50) of a cerrado sensu stricto under fertilization treatments (+N,+P,+NP) (n=4). \*Indicates significant differences in comparison to control treatment (unfertilized) (t-test, \*p <0.05). Reprinted from Jacobson et al. (2011) with permission from Elsevier

calculated according to Mueller-Dumbois and Ellenberg (1974). The index of importance value (IVI) was calculated for each species within each treatment according to Kent and Coker (1992).

#### 18.2.3 Statistical Analysis

Nutrient concentrations in each depth were tested for normality using Kolmogorov-Smirnov test (p < 0.05) and tested with F and student t test (p < 0.05). The data with no normal distribution even after transformation, were compared with Mann-Whitney nonparametric test (p < 0.05). Comparisons were made relative to control for each depth (n=4). Woody vegetation diversity was tested with Shannon-Wiever (H') diversity index and Pielou evenness index (J') for each treatment. Floristic similarity between fertilized and control plots, was tested with the Sørensen similarity index (Kent and Coker 1992). Diversity indices among fertilized plots were compared with control plots using Shannon diversity t test (p < 0.05). All tests (soil and woody vegetation) were performed using the statistical software PAST (Paleontological Statistics Software Package for Education and Data Analysis, UK).

#### 18.3 Results

Nitrogen, P and NP plots showed lower soil pH values (water and CaCl<sub>2</sub>) than those observed in control plots in the top 30 cm. Total soil N did not differ between fertilized and unfertilized plots, but available P concentrations were significantly higher in the P and NP plots at all depths. N and NP plots had higher exchangeable Al in comparison to control plots (all depths in NP plots and in the first 30 cm in N plots). Soil pH values (water and CaCl<sub>2</sub>) increased with depth in all treatments, while total N, P and exchangeable Al showed the inverse pattern (Fig. 18.1).

Relative to species diversity, 479 individuals of 47 species belonging to 29 families were sampled in control plots, 461 individuals belonging to 53 species and 34 families in N plots, 448 individuals from 54 species and 31 families in P plots and 336 individuals of 40 species belonging to 24 families in NP plots. Nitrogen and NP plots showed lower Shannon diversity (H') and evenness (J') indices compared to control plots (Table 18.1). Sørensen floristic similarity index was high among fertilized and unfertilized plots, decreasing in P plots (0.81) to N plots (0.78) and NP plots (0.76). *R. montana* (Proteaceae) had the highest absolute density in N, P and NP plots, and in control plots while *B. salicifolius* (Myrtaceae) (34.9) presented the highest importance value, with 26.2% of relative dominance. *B. salicifolius* (39.05) and *R. montana* (22.89) were the most important species in N plots. In P plots, the most important species were *R. montana* (27.35) and *C. brasiliense* (Caryocaraceae) (17.22), and the most dominant were *C. brasiliense* (12.02%) and *R. montana* (9.48%). *B. salicifolius* (56.73) and *R. montana* (30.0) were the most important species in NP plots, and *B. salicifolius*, the most dominant (44.59%).

**Table 18.1** Density, richness, number of genus and families, Shannon's diversity index (H'), Pielou's eveness index (J') of the shrub-tree flora in fertilized (+N,+P,+NP) and unfertilized (C) plots in a cerrado *sensu stricto* of Brasília—Brazil. \*Indicates significant differences with control plots (Diversity t-test, p < 0.05)

Treatment	Density	Richness	Genus	Families	Shannon (H')	Pielou (J')
C	479	47	38	29	3.40	0.88
+N	461	53	43	34	3.20*	0.80
+P	448	54	41	31	3.39	0.85
+NP	336	40	34	24	2.89*	0.78

# 18.4 Discussion

Species with greater ability to grow and establish under high nutrient availability conditions and lower susceptibility to disturbance (in this case, the occurrence of fire in 2005), had a greater competitive advantage. In fertilized plots, the dominant mature, established species responded to fertilization by increasing their dominance (*B. salicifolius*) and regenerating species with increased recruitment and abundance (*R. montana*), changing the community composition relative to control plots. These composition changes implied a decrease in evenness and diversity, especially in the N and NP plots, where P and Al availability and pH values were significantly different. Soil acidification and increased Al concentration is common under ammonium sulphate addition and is considered a key mechanism in plant diversity loss in several ecosystems (Bobbink et al. 2010). Comparing our results with earlier studies in the same area (Kozovits et al. 2007; Saraceno 2006), fertilization changed pH and P availability in deeper soil layers, on the other hand, total N decreased in the surface layer, but increased with depth.

Species density decreased, especially in NP plots, where we also observed lower richness. Changes in N and NP plot densities and dominance patterns led to a lower resemblance to the unfertilized control plots. Moreover, in these plots, R. montana increased its density ~2-fold and, despite the similar density, B. salicifolius increased in relative dominance with respect to control plots. The increase in herbaceous layer biomass and increase in exotic grass invasive potential is common in many ecosystems where N availability is increased (Bobbink et al. 2010). R. montana significantly increases its' height in competition with the exotic grass M. minutiflora (Poaceae) (Hoffmann and Haridasan 2008) and increases vegetative reproduction rates after fire (Hoffmann 1998). R. Montana's importance in the N and NP community can be explained by its high post-fire regenerative capacity by increasing height and vegetative reproduction rates (Hoffmann and Solbrig 2003). Moreover, species of Proteaceae family are associated with proteoid roots, which facilitate the uptake of P forms, not available forms to other species (Watt and Evans 1999; Turner 2008). Other studies showed that B. salicifolius is associated with plant communities in nutrients rich soils (Proença and Gibbs 1994) which might explain its increasing dominance under higher nutrient availability. However, observed changes in this study can be dynamic on a larger time scale as the relationship between adaptive strategies, competitive ability, composition and diversity have different rates of change over time. These results suggest that nutrient enrichment changes diversity and abundance patterns due to differences in individual species responses to fertilization. Simultaneous NP addition effects on density, dominance, richness and diversity patterns were more significant than those observed for N or P on their own. However, fertilization effects on diversity can be also influenced by other variables such as interaction with the herbaceous layer.

## 18.5 Conclusions

Our results indicated that nutrient addition decreased soil pH and increased exchangeable Al concentrations (N and NP plots) and increased available P concentrations (P and NP plots). Species richness was lower in NP plots, and N addition plots (single or combined) showed lower evenness and species diversity relative to control plots. Changes in species diversity were more intense in combined NP addition plots.

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