Herbaceous and shrubby species co-occurrences in Brazilian savannas: the roles of fire and chance

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Abstract: Competition and facilitation are expected to leave different signatures in the pattern of species co-occurrence. Competition may result in a given species pair occurring less often than expected by chance, whereas facilitation may result in a given species pair occurring more often than expected by chance. We assessed the co-occurrence of pairs of herbaceous and shrubby species in Brazilian savannas, asking (1) whether a given species pair occurs more often than expected by chance, (2) whether the number of species pairs in sites with frequent fires is higher than expected by chance, (3) whether the difference in the functional traits of heterospecific pairs is lower in sites with frequent fires, and (4) whether small environmental variations in each site – instead of species interactions – could explain the co-occurrence of species. We used null models to answer the first two questions, analyses of variance to answer the third question, and detrended correspondence analyses to answer the fourth question. In all studied sites, approximately half of the heterospecific pairs occurred more often than expected by chance. So, facilitation seems to be important in determining the co-occurrence of some species in Brazilian savannas. However, high fire frequencies changed the pattern of occurrence of the species pairs, resulting in a spatial signature indistinguishable from random. Frequent fires also promoted phenotypic clustering of species. Nevertheless, wherever fire frequency is reduced, competition may lead to phenotypic overdispersion of plant species. Thus, less harsh environmental conditions in savannas may increase the competition among plant species.

Introduction

Competition and facilitation are important ecological interactions determining species coexistence in plant communities (Callaway and Walker 1997, Grime 2001, Bruno et al. 2003, Butterfield 2009, Lamb et al. 2009). Competition occurs when neighbouring plants share the same light, nutrient, and space requirements (Grime 1973, 2001). Facilitation occurs when one plant species ameliorates the local environment for another, especially under harsh environmental conditions (Callaway 1995, Bruno et al. 2003). However, these ecological interactions are expected to be more evident within small neighbourhoods, because plants, as sessile organisms, only interact directly with nearest individuals (Stoll and Weiner 2000, Hubbell et al. 2001). Thus, competition and facilitation should leave different signatures in the pattern of co-occurrence of species at small spatial scales (Stoll and Weiner 2000, Perry et al. 2009). As competitively inferior species are unable to persist in the neighbourhood of competitively superior species, a lower number of co-occurring species than expected at random may arise from competition (Lieberman and Lieberman 2007, Perry et al. 2009). On the contrary, a higher number of co-occurring species may arise from facilitation (Cavieres et al. 2002, Arroyo et al. 2003, Cavieres and Badano 2009).

A straightforward way to evaluate whether competition or facilitation is predominant among species pairs in a community is considering the frequency of occurrence of neighbour pairs (Perry et al. 2009, Azaele et al. 2010). In other words, how often is the nearest neighbour of species *i* a member of species *j*? The frequency at which two species are neighbours is influenced by the kind of interaction between them (Grime 2001, Bruno et al. 2003) and their relative abundances (Perry et al. 2009). Thus, facilitation may result in a given pair of species occurring more often than expected by chance (Dullinger et al. 2007, Lieberman and Lieberman 2007, Maestre et al. 2008), whereas competition may result in a given pair of species occurring less often than expected by chance (Gotelli and Ellison 2002, Gotelli and Rohde 2002).

The outcome of competition and facilitation is frequently influenced by variation in the abiotic environment, and within a given habitat, the environmental conditions can vary through time, also influencing the outcome of biotic interactions and driving community dynamics (Butterfield 2009). Likewise, the intensity and importance of these biotic interactions vary spatially along gradients of environmental stress or resource availability (Greenlee and Callaway 1996, Goldberg and Novoplansky 1997), hereafter referred to as 'environmental harshness' (sensu Brooker et al. 2008). In plant communities, for instance, competition and facilitation are supposed to occur simultaneously among different species and to change as the age of the community advances. Seedlings may use larger individuals as 'nurses', but compete

with them when they become adults (Verdú and Valiente-Banuet 2008). Notwithstanding, empirical studies have demonstrated that facilitation increases with environmental harshness, whereas competition prevails under more benign conditions (that is, the stress-gradient hypothesis; Bertness and Callaway 1994, Greenlee and Callaway 1996, Brooker et al. 2008). As a consequence, the pattern of co-occurrence of plant species at small spatial scale is also expected to change when the environmental conditions change (Collins and Klahr 1991). The number of occurrences of pairs of neighbouring plant species should be, hence, greater than expected by chance in communities that experience an increasing environmental harshness (Lieberman and Lieberman 2007, Perry et al. 2009).

The assessment of the functional traits of species is also important to infer whether competition determines the co-occurrence of heterospecific pairs of plant species. At small spatial scales, plant species compete directly for limited soil nutrients (Hubbell et al. 2001, Grime 2001, Lamb et al. 2009). Such a competition is higher among species that have similar niches and consequently share more morphological and physiological characteristics, that is, share more functional traits (Weiher et al. 1998, Grime 2001). Thus, wherever competition is predominant, neighbouring heterospecific pairs of plants are expected to differ more than expected by chance in functional trait values (Weiher and Keddy 1995, Weiher et al. 1998).

In South America, the largest savanna region is the Brazilian cerrado (Gottsberger and Silberbauer-Gottsberger 2006). Like its counterparts elsewhere, the cerrado vegetation evolved with fire and, consequently, its plant species have well-developed capabilities of regenerating, either vegetatively or by seed (Coutinho 1990, Gottsberger and Silberbauer-Gottsberger 2006). Even though most fires in cerrado occur at the surface, by consuming mainly the herbaceous layer of the vegetation (Miranda et al. 1993), frequent burnings favour the herbaceous and shrubby species, due to their underground meristems, at the expense of trees (Moreira 2000, Gottsberger and Silberbauer-Gottsberger 2006). Many herbaceous species have the reproductive output increased by recurrent fires (Sarmiento 1992), and the persistence of some species in the community depends on high fire frequency (Canales et al. 1994).

Near neighbour interactions have been recently examined in forests (Lieberman and Lieberman 2007) and shrublands (Perry et al. 2009). These studies did not corroborate the stress-gradient hypothesis. Perry et al. (2009) argued that a great number of species pairs in rich plant communities are unlikely to occur, because the species are present at low densities in samples. In this case, most of near neighbour pairs are represented by a small fraction of species and, by chance alone, the number of observed possible pairs is lower than the theoretical maximum. Nevertheless, these previous studies did not evaluate explicitly the stress-gradient hypothesis, considering the species interactions along an environmental harshness gradient. Studies conducted in altitudinal (Choler et al. 2001) and moisture (Holzapfel et al. 2006) gradients have demonstrated that the frequency of facilitative interactions increases with the environmental harshness.

Here, we tested whether an annual fire frequency changes the pattern of co-occurrence of herbaceous and shrubby species in cerrado, taking into account the near neighbours. Based on the stress-gradient hypothesis (Brooker et al. 2008), we expected that annual fires would increase the co-occurrence of pairs of herbaceous and shrubby species and assemble neighbouring species with similar functional traits. In sum, we answered (1) whether a given species pair occurs more often than expected by chance, (2) whether the number of species pairs in sites with frequent fires is higher than expected by chance, (3) whether the difference in the functional traits of the heterospecific pairs is lower in sites with frequent fires, and (4) whether small environmental variations in each site – instead of species interactions – could explain the co-occurrence of species.

Methods

Study site

We surveyed three spatially proximate cerrado sites in Emas National Park (ENP), central Brazil ($17^{o}49$'-$18^{o}28$'S; $52^{o}39$'-$53^{o}10$'W). The ENP comprises 132,941 ha and is one of the largest and most important reserves of Cerrado (Unesco 2001). Regional climate is humid tropical, characterised by marked rainfall seasonality, with a pronounced dry season in the winter (Aw, Köppen 1931). Annual rainfall and mean temperature lie around 1,745 mm and 24.6°C, respectively. In general, the soils are poor in nutrients, well drained, and acid (Silva and Batalha 2008). Up to 1984, the ENP was annually burned in the dry season to promote forage regrowth to cattle (França et al. 2007). Afterwards, the ENP was fenced, and a fire exclusion policy was established (Ramos-Neto and Pivello 2000). As a consequence, catastrophic fires used to occur every 3-4 years, burning 80% of the park's area (Ramos-Neto and Pivello 2000). Since 1994, around 10 km^2 of preventive firebreaks have been burned annually at the end of the wet season, and a fire brigade works in the park during the dry season to prevent anthropogenic fires. Thus, there are currently few occurrences of anthropogenic fires inside the ENP (less than 2.2% of the burned area from 1994 to 2003), and the largest wildfires burn less than 30% of the total area (França et al. 2007).

Data collection

In the late rainy season of 2006, we sampled the plant species of the herbaceous layer in three nearby sites with different fire frequencies, but with similar physiognomies (grassland cerrado), on the same soils (Oxisols), in the southeastern portion of the ENP. The first and second sites were firebreaks, one burned annually since 1994 (18°18'50''S and 52°54'00''W) and other burned approximately biennially since 1994 (18°19'01''S and 52°54'10''W). The third site was a protected cerrado, without fires since 1994

(18°17'28''S and 52°53'41''W). The sites were distant less than 2 km apart.

In each site, we placed a 1,250 m line, with 125 points, 10 m apart. In each point, we sampled four individuals in the herbaceous layer with the point-quarter method (Mueller-Dombois and Ellenberg 1974), summing up 500 individuals in each site. We defined the herbaceous layer as all herbaceous individuals and all woody individuals with stem diameter at soil level lower than 3 cm. We did not sample seedlings, defined as those individuals with cotyledons. In cerrado, most plant species present clonal growth from subterranean organs (Gottsberger and Silberbauer-Gottsberger 2006), so that it is occasionally hard to distinguish ramets and genets in field. We tried to minimise this problem counting only those individuals that did not present clear connections to other sampled individuals at soil level. We identified the individuals sampled by comparing the collected material to reference collections from ENP (Batalha and Martins 2002) and vouchers lodged at the State University of Campinas herbarium. When identification at species level was not possible, we classified them as morphospecies.

Data analyses

First, we described the distribution of species abundances in the three sites to characterise the general structure of the plant community, considering all individuals sampled. For each site, we constructed a graph in which we plotted the relative abundance of species against species abundance rank order (Fig. 1). Then, in each sampling point, we considered that each individual could interact potentially with the other three individuals. Accordingly, we obtained six different pairs of co-occurring individuals per sampling point, summing up 750 pairs of plants in each site. First, we calculated the theoretical maximum number of species pairs for each site, computing the simple formula $S(S-1)/2$, where S was the total number of species. Then, we computed for each site the observed frequency of all possible ij species pairs.

To answer our first question, we generated a distribution of expected frequencies for each potential ij species pair in each site. We followed the null model proposed by Lieberman and Lieberman (2007), in which the combinations of species pairs in a site reflect a process of random sampling from the community: (1) we picked two plants at random, with replacement, from the complete list of individuals of the site; (2) we recorded the species identities of the pair and, if this randomly picked pair matched the ij species combination under consideration, we counted it; and (3) we repeated these steps until the number of pairs drawn was equal to the number of pairs in the site. We repeated this procedure 1,000 times, producing a null distribution of 1,000 expected counts for a given ij species combination.

To deal with a potential bias of the non-independence of the pairs at each sampling point, we also ran the previous analysis considering only two random pairs of individuals per point. So, we picked randomly two pairs of individuals without replacement at each point, summing up 250 pairs of plants in each site. In this second null model, we considered the highest number of species pairs picked by chance in each site for 1,000 randomisations. Then, we compared the frequency of these species pairs with a distribution of expected frequencies for each potential ij species pair in each site, as described above.

We used a Monte Carlo procedure to evaluate whether the observed number of species pairs in each site was different than expected by chance. We used the following procedure: (1) we computed the observed number of species pairs in each site; (2) we randomised the individuals in the sampling points and recorded the number of random pairs; (3) we repeated these steps 1,000 times and estimated a random mean and SD for the null communities; and (4) we calculated pseudo P-values for the observed number of species pairs to compare null and observed community characteristics. We also repeated this procedure considering only the conspecific pairs.

To answer our third question, we measured three functional traits: basal area, plant height, and specific leaf area. In each site, we sampled ten individuals of each species to estimate the mean of the functional traits. When there were not ten individuals of a given species, we used all individuals sampled. Basal area is a trait related to space occupation and resource acquisition (Navas and Moreau-Richard 2005). Plant height is associated to competitive ability, fecundity, and tolerance to stress (Cornelissen et al. 2003). Specific leaf area is positively related to maximum photosynthetic rates, since low values of it correspond to a high leaf structure investment (Cornelissen et al. 2003). We measured these traits according to Cornelissen et al. (2003). We calculated the leaf area with the ImageJ software (Rasband 2004). Then, we compared the absolute value of the differences in functional trait values of the heterospecific pairs of each site with an analysis of variance, followed by a Tukey test. The term 'phenotypic clustering' refers to high trait similarity among neighbouring species, whereas the term 'phenotypic overdispersion' refers to low trait similarity among neighbouring species (Weiher et al. 1998).

Finally, to answer our last question, we constructed a detrended correspondence analysis (Jongman et al. 1995), which allowed us to observe how spread species and sites scores were in the multivariate space. With this approach, we evaluated whether theoretical environmental variables could explain patterns of co-occurrence of species within sites. We ran all analyses in the R environment (R Development Core Team 2009). The R functions we wrote are available in http://www.cerradoecology.com.

Results

We found 97 species in the three sites; 58 species in the annually burned site, 50 species in the biennially burned site, and 62 species in the unburned site (Appendix 1). The distribution of species abundances in the three sites was similar, with the dominance of few species, such as *Axonopus suffultus* and *Tristachya leiostachya* (Fig. 1).

With the first null model, we found 182 species pairs in the annually burned site, 168 in the biennially burned site, and 221 in the unburned site. These values were proportionally much lower than expected by the theoretical maximum number of species pairs (1,711 for the annually burned site, 1,275 for the biennially burned site, and 1,953 for the unburned site). They represented 10.63, 13.17, and 11.31% of the theoretical maxima for the three sites respectively. Moreover, in the second null model, we found 101 pairs in the annually burned site, 97 pairs in the biennially burned site, and 127 pairs in the unburned site. These values accounted for 5.90, 7.60, and 6.50% of the theoretical maxima for the three sites respectively.

When we analysed each observed pair of species with the first null model, we found few pairs that occurred less often than expected by chance (two pairs in the annually burned site and three pairs in other sites (Appendix 2). However, approximately half of the observed heterospecific pairs in each site were very frequent. The observed frequencies of 50.9% of the heterospecific pairs in the annually burned site, 32.5% in the biennially burned site, and 45.5% in the unburned site were significantly higher than expected by chance ($P < 0.05$, Appendix 2). We found similar results with the second null model, given that one single pair occurred less often than expected by chance in the annually burned site, and two pairs in the biennially burned site (Appendix 3). Accordingly, 35.9, 42.8, and 41.2% of the observed heterospecific pairs in the annually burned, biennially burned, and unburned sites occurred at frequencies higher than those expected by chance ($P < 0.05$, Appendix 3).

The total number of observed pairs in the unburned site was significantly lower than expected by chance (mean of null pairs = 253.6 ± 9.7, $P < 0.001$). Nevertheless, the total numbers of observed pairs in the sites with high fire frequencies were not different from random (mean of null pairs = 195.7 ± 9.3, $P = 0.110$ for the annually burned site, mean of
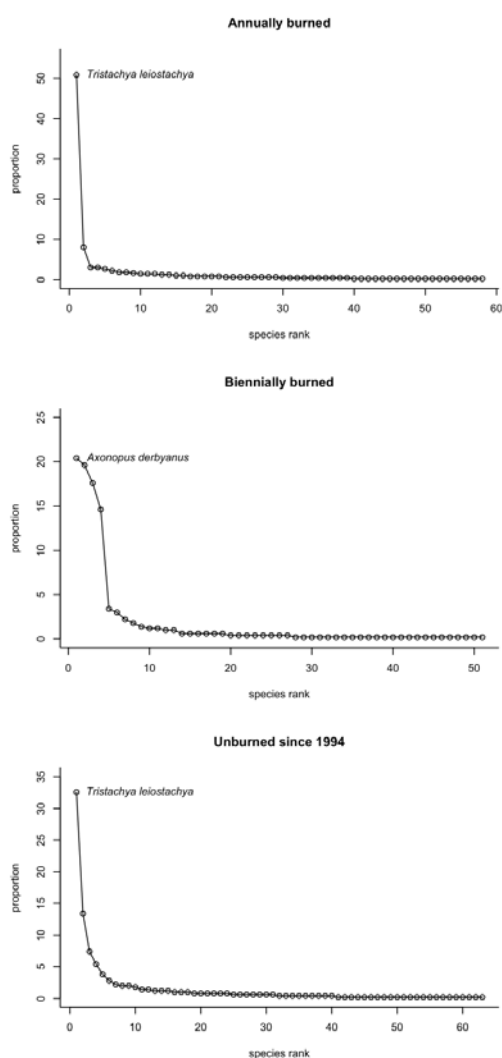

Figure 1. Rank-abundance curves of the herbaceous species found in three cerrado sites under different fire frequencies in Emas National Park, central Brazil.
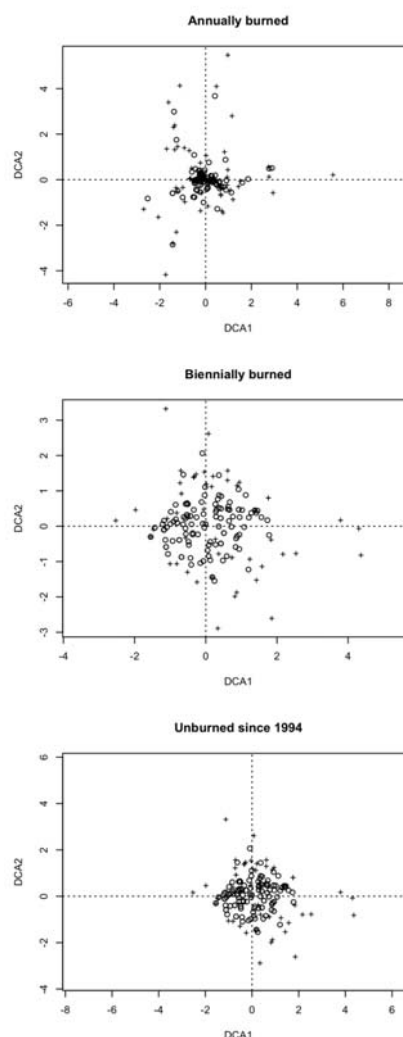

Figure 2. Detrended correspondence analysis of herbaceous species abundances in three cerrado sites under different fire frequencies at Emas National Park, central Brazil. Open black circles are species scores, and crosses are site scores.

Table 1. Mean differences ± standard deviations in basal area (cm^2), height (cm), and specific leaf area (m^2 kg^{-1}) between neighbouring species of herbaceous plants in cerrado, Emas National Park, central Brazil. Different letters indicate significant differences between means ($\alpha = 0.05$) in the annually burned site, the biennially burned site, and in the site unburned since 1994.

Functional trait	Annually	Biennially	Unburned	F	P
Basal area	3.562[a] ± 6.446	2.248[a] ± 6.120	3.22[a] ± 5.048	2.143	0.118
Height	19.782[a] ± 14.281	19.677[a] ± 12.831	22.042[a] ± 17.657	1.388	0.25
Specific leaf area	36.284[a] ± 32.909	70.781[b] ± 81.244	70.610[b] ± 71.063	15.083	0.001

null pairs = 177.0 ± 8.2, P = 0.240 for the biennially burned site). On the other hand, the total numbers of conspecific pairs in all sites were significantly higher than expected at random (P < 0.001). We observed 266 conspecific species pairs in the annually burned site (mean of null pairs = 202.1 ± 12.7), 194 in the biennially burned site (mean of null pairs = 101.3 ± 9.6), and 180 in the unburned site (mean of null pairs = 103.7 ± 9.6).

We also found a significant difference in the values of specific leaf area of neighbouring heterospecific pairs (F = 15.083, P < 0.001). The neighbouring plant species in the annually burned site presented a mean difference in the values of specific leaf area (36.284 m^2 kg^{-1}), lower than neighbouring species in the biennially burned (70.781 m^2 kg^{-1}) and unburned sites (70.610 m^2 kg^{-1}). The mean difference between biennially burned and unburned sites was not significant. When we analysed the other functional traits, we also did not find differences among the sites (Table 1, F = 2.143 for basal area and F = 1.388 for plant height, P > 0.100 for all comparisons).

The site scores in the detrended correspondence analyses were somewhat spread along the first two axes (Fig. 2), indicating that the species composition in the point-quarters along transects were different. This is an indication of environmental heterogeneity within sites, which may influence the species co-occurrences differently from expected at random. However, the detrended correspondence analyses did not show any particular clustering of species scores and the overall distribution of species scores in the multivariate space was quite overlapped. Thus, species interactions, rather than local environmental variations, are likely to explain the co-occurrence of species.

Discussion

Our results supported that more benign environmental conditions in cerrado may increase the competition among species plants. Although high fire frequencies did not assemble more pairs of species as predicted by stress-gradient hypothesis (Brooker et al. 2008), they may diminish the strength of competitive exclusion among plants, resulting in co-occurrences of species indistinguishable from random. Frequent fires also promoted phenotypic clustering of herbaceous species in cerrado. So, wherever fire frequency is reduced, competition may lead to phenotypic overdispersion of plant species.

The observed number of pairs of neighbouring species was lower than the expected maximum number for the cerrado sites. In other studies, this number was also very low (Lieberman and Lieberman 2007, Perry et al. 2009). This discrepancy is primarily due to differences in the relative abundance of species plants (Appendix 1, Perry et al. 2009). If there are many rare and few common species in the community, chance alone would make it unlikely that rare species would occur as neighbours (Lieberman and Lieberman 2007). As long as pairs of neighbouring species are determined to a certain extent by chance (see also Hubbell 2001), the reciprocal selective pressures acting on species are dependent on community context (Perry et al. 2009). Consequently, the interactions between species may be the product of a diffuse rather than a specific pairwise selection. Diffuse evolution among plants has been also evoked to explain the tolerance of saplings to a large number of neighbouring tree species in tropical forests (Uriarte et al. 2004). Further studies should consider competition and facilitation as a network of species in the community to place the ecological interactions in an appropriate evolutionary context (Johnson and Stinchcombe 2007, Verdú and Valiente-Banuet 2008)

If chance prevents most of plant species to interact with each other, ecological interactions may not be important in structuring the co-occurrence of plant species (Perry et al. 2009). However, a certain pattern emerges from that small proportion of heterospecific pairs that occurred as neighbours at frequencies different than expected at random. In the three cerrado sites, approximately half of the heterospecific pairs occurred more often than expected by chance and few species pair occurred less often than expected by chance. In forests and shrublands, most of the observed pairs also occurred at higher frequencies than expected by chance (Lieberman and Lieberman 2007, Perry et al. 2009). These findings indicate that facilitation is important in determining the co-occurrence of some species in plant communities. In cerrado, this positive interaction seems to be more important between rare species, of which the pairs were more frequent than rare-common species pairs (see Appendix 2).

The unburned cerrado showed a number of species pairs lower than expected by chance, what is expected to occur where competition is the predominant interaction among species (Lieberman and Lieberman 2007, Perry et al. 2009), since few species could survive in the neighborhood of competitively superior species (Grime 2001, Lamb et al. 2009). In addition, a decrease in the fire frequency promoted a phenotypic overdispersion of species relative to specific leaf area. Phenotypic overdispersion is also expected when competitive exclusion structures the community. Among neighbouring plants, however, phenotypic overdispersion is thought to be even higher (Weiher and Keddy 1995), because

species compete directly for limited soil nutrients (Grime 2001). Evidence of competition promoting phenotypic overdispersion among neighbours has been also found in other plant communities (prairies – Fargione et al. 2003; sand dunes – Stubbs and Wilson 2004; and tropical forests – Kraft et al. 2008). Thus, competition seems to be predominant when an environmental constraint is reduced, even though facilitation continues to occur among some species pairs.

However, for basal area and plant height, we did not find differences among the cerrado sites. Basal area and height are traits associated with the growth forms of plants (Cornelissen et al. 2003). Herbaceous and shrubby species present more restricted range in the values of basal area and height than trees, for instance. Thus, a clear pattern of phenotypic clustering or overdispersion relative to these traits may not emerge in the herbaceous layer of the savannas. On the other hand, annual fires promoted a phenotypic clustering of herbaceous plants, because frequent fires acting as an environmental filter select those species that can survive within a community on the basis of their tolerance to the abiotic conditions (Weiher and Keddy 1995). Consequently, they tend to assemble co-occurring species with similar niches, that is, species with similar functional traits (Fukami et al. 2005). Silva and Batalha (2010) have recently demonstrated that frequent fires may assemble functionally similar woody species in cerrado. In Mediterranean vegetation, high fire frequencies also promote phenotypic clustering of plants (Pausas and Verdú 2008). In other savannas (Williams et al. 1999, Silva et al. 2001), as well as in the California chaparral (see Syphard et al. 2006 for reference), frequent fires changed the plant functional types, favouring grass and shrub species. Although frequent fires favour herbaceous plants at the expense of woody ones (Moreira 2000), they reduce the range of functional traits and the number of functional types in fire-prone vegetation types, decreasing the overall functional diversity of the plant community.

Frequent fires, however, did not assemble species pairs at frequencies higher than expected by chance, favouring facilitation among plant species (Perry et al. 2009). Facilitation has been documented in alpine, arid, and Mediterranean-type plant communities (Brooker et al. 2008), that is, in environments that experience harsh abiotic conditions, as the savannas. In these vegetation types, facilitation takes the form of 'nurse plants', in which larger plants act as favoured recruitment sites for seedlings and for herbs by modifying microclimate, or reducing herbivory or both (Callaway 1995, Brooker et al. 2008). Here, we tested the stress gradient hypothesis with herbaceous and shrubby species, which are affected directly by fires in cerrado (Miranda et al. 1993). Consequently, possible nurse effects of large woody species were not captured by our analysis. Despite this bias, our results showed that frequent fires may decrease the strength of competitive exclusion among herbaceous plants, resulting in co-occurrences of species indistinguishable from random. Further studies in savannas should also assess the pairwise co-occurrence between herbs and trees when testing the stress gradient hypothesis.

The number of conspecific pairs of species was higher than expected at random in the three cerrado sites. In other vegetation types, the number of conspecific pairs was also higher than expected by chance (Perry et al. 2009, but see Lieberman and Lieberman 2007). This general pattern is a direct consequence of the spatial distribution of plant populations. In rich communities, most plant species present an aggregated spatial distribution (Condit et al. 2000, Perry et al. 2008). In cerrado, the spatial pattern of the populations is not different (San Jose et al. 1991, Silva et al. 2009). Aggregation of individuals of the same species is a response to patchiness of favourable soil conditions or to dispersal limitation (Condit et al. 2000). In cerrado, the conspecific aggregation is increased by the clonal growth of the large majority of their plant species (Gottsberger and Silberbauer-Gottsberger 2006). So, frequent fires do not seem to change the clumped pattern of spatial distribution of herbaceous species and, consequently, the co-occurrence of conspecific pairs in cerrado.

Moreover, it is worth noticing that the large dominance of some grasses in the studied sites, such as *Axonopus suffultus* and *Tristachya leiostachya* (Fig. 1), contributed largely to the significance of the results. Nevertheless, dominance of graminoid species are common features in other savannas (San Jose et al. 1991, Sarmiento 1992, Canales et al. 1994), so that the pattern in the co-occurrence of conspecific pairs we found was consistent in other savannas as well.

Our results also supported that the species interactions, rather than environmental heterogeneity, explain the co-occurrence of species pairs in cerrado. Other studies in temperate forests (Cavender-Bares et al. 2006) and sclerophyllous shrublands (Slingsby and Verboom 2006) also demonstrated similar trends at fine spatial scales. Thus, even though small variations in the environment might also influence the outcome of competition and facilitation (Butterfield 2009), species interactions seem to be the predominant ecological process in the assembly of neighbour species pairs.

It is also important to notice that we could not include genuine replications (sensu Hurlbert 1984) in our experimental design. In cerrado, where fire frequency is high, very few sites protected from fire during a decade can be found (França et al. 2007), so that replicated treatments may be hard to obtain. Hence, other non-controlled variables differing between the cerrado sites may also account for a fraction of the observed results.

In conclusion, the frequencies of neighbour species pairs in cerrado, as in other vegetation types, follow mostly a distribution expected by chance. However, competition and facilitation may also influence the co-occurrence of herbaceous and shrubby species. Among the heterospecific pairs that departed from the frequency expected at random, facilitation seems to be the predominant ecological interaction in cerrado. Competition, on the other hand, seems to take place in the absence of an environmental constraint. Frequent fires may hence decrease the strength of competitive exclusion

among plants, resulting in random co-occurrences of neighbouring species.

References

Arroyo, M.T.K., L.A. Cavieres, A. Peñaloza and M.A. Arroyo-Kalin. 2003. Positive associations between the cushion plant Azorella monantha (Apiaceae) and alpine plant species in the Chilean Patagonian Andes. *Plant Ecol.* 169: 121-129.

Azaele, S., R. Muneepeerakul, A. Rinaldo and I. Rodriguez-Iturbe. 2010. Inferring plant ecosystem organization from species occurrences. *J. Theor. Biol.* 262: 323-329.

Batalha, M.A. and F.R. Martins. 2002. The vascular flora of the cerrado in Emas National Park (Goiás, central Brazil). *Sida* 20: 295-312.

Bertness, M.D. and R.M. Callaway. 1994. Positive interactions in communities. *Trends Ecol. Evol.* 9: 191-193.

Brooker, R.W., F.T. Maestre, R.M. Callaway, C.L. Lortie, L. Cavieres, G. Kunstler, P. Liancourt, K. Tielbörger, J.M.J. Travis, F. Anthelme, C. Armas, L. Coll, E. Corcket, S. Delzon, E. Forey, Z. Kikvidze, J. Olofsson, F. Pugnaire, C.L. Quiroz, P. Saccone, K. Schiffers, M. Seifan, B. Touzard and R. Michalet. 2008. Facilitation in plant communities: the past, the present and the future. *J. Ecol.* 96: 18-34.

Bruno, J.F., J.J. Stachowicz and M.D. Bertness. 2003. Inclusion of facilitation into ecological theory. *Trends Ecol. Evol.* 18: 119-125.

Butterfield, B.J. 2009. Effects of facilitation on community stability and dynamics: synthesis and future directions. *J. Ecol.* 97: 1192-1201.

Callaway, R.M. 1995. Positive interactions among plants. *Bot. Rev.* 61: 306-349.

Callaway, R.M. and L.R. Walker. 1997. Competition and facilitation: a synthetic approach to interactions in plants communities. *Ecology* 78: 1958-1965.

Canales, J., M.C. Trevisan, J.F. Silva and H. Caswell. 1994. A demographic study of an annual grass (*Andropogon brevifolius* Schwartz) in burnt and unburnt savanna. *Acta Oecol.* 15: 261-274.

Cavender-Bares, J., A. Keen and B. Miles. 2006. Phylogenetic structure of Floridian plant communities depends on taxonomic and spatial scale. *Ecology* 87: S109-S122.

Cavieres L.A., M.T.K. Arroyo, A. Peñaloza, M. Molina-Montenegro and C. Torres. 2002. Nurse effect of *Bolax gummnifera* cushion plants in the alpine vegetation of the Chilean Patagonian Andes. *J. Veg. Sci.* 13: 547-554.

Cavieres, L.A. and E.I. Badano. 2009. Do facilitative interactions increase species richness at the entire community level? *J. Ecol.* 97: 1181-1191.

Choler, P., R. Michalet and R.M. Callaway. 2001. Facilitation and competition on gradients in alpine plant communities. *Ecology* 82: 3295-3308.

Collins, S.P. and S.C. Klahr 1991. Tree dispersion in oak dominated forests along an environmental gradient. *Oecologia* 86: 471-477.

Condit, R., P.S. Ashton, P. Baker, S. Bunyavejchewin, S. Gunatilleke, N. Gunatilleke, S.P. Hubbell, R.B. Foster, A. Itoh, J.V. LaFrankie, , H.S. Lee, E. Losos, N. Manokaran, R. Sukumar and T. Yamakura. 2000. Spatial patterns in the distribution of tropical tree species. *Science* 288: 1414-1418.

Cornelissen, J.H.C., S. Lavorel, E. Garnier, S. Díaz, N. Buchmann, D.E. Gurbich, P.B. Reich, H. Steege, H.D. Morgan, M.G.A. Heijden van der, J.G. Pausas and H. Pooter. 2003. A handbook of protocols for standardized and easy measurement of plant fuctional trait worldwide. *Aust. J. Bot.* 51: 335-380.

Coutinho, L.M. 1990. Fire in the ecology of the Brazilian cerrado. In: J.G. Goldammer (ed.), *Fire in the Tropical Biota.* Springer, Berlin, pp. 81-103.

Dullinger, S., I. Kleinbauer, H. Pauli, M. Gottfried, R. Brooker, L. Nagy, J.P. Theurillat, J.I. Holten, O. Abdaladze, J.L. Benito, J.L. Borel, G. Coldea, D. Ghosn, R. Kanka, A. Merzouki, C. Klettner, P. Moiseev, U. Molau, K. Reiter, G. Rossi, A. Stancisi, M. Tomaselli, P. Unterlugauer, P. Vittoz and G. Grabherr. 2007. Weak and variable relationships between environmental severity and small-scale co-occurrence in alpine plant communities. *J. Ecol.* 95: 1284-1295.

Fargione, J., C.S. Brown and D. Tilman. 2003. Community assembly and invasion: An experimental test of neutral versus niche processes. *Proc. Nat. Acad. Sci. USA* 100: 8916-8920.

França, H., M.B. Ramos-Neto, A. Setzer. 2007. *O fogo no Parque Nacional das Emas.* Instituto do Meio Ambiente e dos Recursos Naturais Renováveis (Ibama), Brasília.

Fukami, T., T.M. Bezemer, S.R. Mortimer and W.H. van der Putten. 2005. Species divergence and trait convergence in experimental plant community assembly. *Ecol. Lett.* 8: 1283-1290.

Goldberg, D. and A. Novoplansky. 1997. On the relative importance of competition in unproductive environments. *J Ecol.* 85: 409-418.

Gotelli, N.J. and A.M. Ellison. 2002. Assembly rules for New England ant assemblages. *Oikos* 99: 591-599.

Gotelli, N.J. and K. Rohde. 2002. Co-occurrence of ectoparasites of marine fishes: a null model analysis. *Ecol. Lett.* 5: 86-94.

Greenlee, J. and R.M. Callaway. 1996. Effect of abiotic stress on the relative importance of interference and facilitation. *Am. Nat.* 148: 386-396.

Grime, J.P. 1973. Competitive exclusion in herbaceous vegetation. *Nature* 242: 344-347.

Grime, J.P. 2001. *Plant Strategies, Vegetation Processes, and Ecosystem Properties.* John Wiley & Sons, Chichester.

Gottsberger, G. and I. Silberbauer-Gottsberger. 2006. *Life in the Cerrado: A South American Tropical Seasonal Vegetation. Vol. 1. Origin, Structure, Dynamics and Plant Use.* Reta, Ulm.

Holzapfel, C., K. Tielbörger, H.A. Parag, J. Kigel and M, Sternberg. 2006. Annual plant–shrub interactions along an aridity gradient. *Basic Appl. Ecol.* 7: 268-279.

Hubbell, S.P. 2001. *The Unified Neutral Theory of Biodiversity and Biogeography*. Princeton University, New Jersey.

Hubbell, S.P., J. A Ahumada, R. Condit and R.B. Foster. 2001. Local neighbourhood effects on long-term survival of individual trees in a neotropical forest. *Ecol. Res.* 16: 859-875.

Hurlbert, S.H. 1984. Pseudoreplication and the design of ecological field experiments. *Ecol. Monogr.* 54: 187-211.

Johnson, M.T.J. and J.R. Stinchcombe. 2007. An emerging synthesis between community ecology and evolutionary biology. *Trends Ecol. Evol.* 22: 250-257.

Jongman, R.H.G., C.F.J. ter Braak and O.F.R. Tongeren. 1995. *Data Analysis in Community and Landscape Ecology.* Cambridge University, Cambridge.

Köppen, W. 1931. *Grundriss der Klimakunde.* Gruyter, Berlin.

Kraft, N.J.B., R. Valencia and D.D. Ackerly. 2008. Functional traits and niche-based tree community assembly in an Amazonian forest. *Science* 332: 580-582.

Lamb, E.G., S.W. Kembel and J.F. Cahill Jr. 2009. Biodiversity and ecosystem stability in a decade-long grassland experiment. *J. Ecol.* 97: 155-163.

Lieberman, M. and D. Lieberman. 2007. Nearest-neighbor tree species combinations in tropical forest: the role of chance, and some consequences of high diversity. *Oikos* 116: 377-386.

Maestre, F.T., C. Escolar, I. Martínez and A. Escudero. 2008. Are soil lichen communities structured by biotic interactions? A null model analysis. *J. Veg. Sci.* 19: 261-266.

Miranda, A.C., H.S. Miranda, I.O. Dias and B.F. Dias. 1993. Soil and air temperatures during prescribed Cerrado fires in central Brazil. *J. Trop. Ecol.* 9: 313-320.

Moreira, A.G. 2000. Effects of fire protection on savanna structure in Central Brazil. *J. Biogeogr.* 27: 1021-1029.

Mueller-Dombois, D. and H. Ellenberg. 1974. *Aims and Methods of Vegetation Ecology.* Wiley, New York.

Navas, M.L. and J.M. Moreau-Richard. 2005. Can traits predict the competitive response of herbaceous Mediterranean species? *Acta Oecol.* 27: 107-114.

Pausas, J.G. and M. Verdú. 2008. Fire reduces morphospace occupation in plant communities. *Ecology* 89: 2181-2186.

Perry, G.L.W., N.J. Enright, B.P. Miller and B.B. Lamont. 2008. Spatial patterns in species-rich sclerophyll shrublands of southwestern Australia. *J. Veg. Sci.* 19: 705-716.

Perry, G.L.W., N.J. Enright, B.P. Miller and B.B. Lamont. 2009. Nearest-neighbour interactions in species-rich shrublands: the roles of abundance, spatial patterns and resources. *Oikos* 118: 161-174.

Ramos-Neto, M.B. and V.R. Pivello 2000. Lightning fires in a Brazilian savanna National Park: rethinking management strategies. *Environ. Manag.* 26: 675-684.

Rasband, W. 2004. *ImageJ: Image Process and Analysis in Java.* National Institute of Health, Bethesda

R Development Core Team 2009. *R: A Language and Environment for sTatistical Computing.* R Foundation for Statistical Computing, Vienna. URL http://www.R-project.org

San Jose, JJ., M.R. Farinas and J. Rosales. 1991. Spatial patterns of trees and structuring factors in a *Trachypogon* savanna of the Orinoco llanos. *Biotropica* 23: 114-123.

Sarmiento, G. 1992. Adaptative strategies of perennial grasses in South America savannas. *J. Veg. Sci.* 3: 325-336.

Silva, D.M. and M.A. Batalha. 2008. Soil–vegetation relationships in cerrados under different fire frequencies. *Plant Soil* 311: 87-96.

Silva, I.A. and M.A. Batalha. 2010. Woody plant species co-occurrence in Brazilian savannas under different fire frequencies. *Acta Oecol.* 36: 85-91.

Silva I.A, M.W. Valenti and D.M. Silva-Matos. 2009. Fire effects on the population structure of *Zanthoxylum rhoifolium* Lam (Rutaceae) in a Brazilian savanna. *Braz. J. Biol.*, 69: 631-637.

Silva, J.F., A. Zambrano and M.R Fariñas. 2001. Increase in the woody component of seasonal savannas under different fire regimes in Calabozo, Venezuela. *J. Biogeogr.* 28: 977-983.

Slingsby, J.A. and G.A.Verboom. 2006. Phylogenetic relatedness limits co-occurrence at fine spatial scales: evidence from the schoenoid sedges (Cyperaceae: Schoeneae) of the Cape Floristic Region, South Africa. *Am. Nat.* 168: 14-27.

Stoll, P. and J. Weiner. 2000. A neighborhood view of interactions among individual plants. In: U. Dieckmann, R. Law and J. A. J. Metz (eds.), *The Geometry of Ecological Interactions: Simplifying Spatial Complexity.* Cambridge University Press, Cambridge, pp. 11-27.

Stubbs, W.J. and B. Wilson. 2004. Evidence for limiting similarity in a sand dune community. *J. Ecol.* 92: 557-567.

Syphard, A.D., J. Franklin and J.E. Keeley. 2006. Simulating the effects of frequent fire on southern California coastal shrublands. *Ecol. Appl.* 16: 744-1756.

Unesco, United Nations Educational, Scientific, and Cultural Organization, 2001. *Cerrado protected areas: Chapada dos Veadeiros and Emas National Parks.* Unesco, Paris. URL http://www.unesco.org/whc/sites/1035.htm.

Uriarte, M., R. Condit, C.D. Canham and S.P. Hubbell. 2004. A spatially explicit model of sapling growth in a tropical forest: does the identity of neighbours matter? *J. Ecol.* 92: 348-360.

Verdú, M. and A. Valiente-Banuet. 2008. The nested assembly of plant facilitation networks prevents species extinctions. *Am. Nat.* 172: 751-760.

Weiher, E. and P.A. Keddy. 1995. Assembly rules, null models, and trait dispersion: new questions from old patterns. *Oikos* 74: 159-164.

Weiher, E., G.D.P. Clarke and P.A. Keddy. 1998. Community assembly rules, morphological dispersion, and the coexistence of plant species. *Oikos* 81: 309-322.

Williams, R.J., G.D. Cook, A.M. Gill and P.H.R. Moore. 1999. Fire regime, fire intensity and tree survival in a tropical savanna in northern Australia. *Aust. J. Ecol.* 24: 50-59.

Appendices

Appendix 1. Number of individuals of the species sampled in Emas National Park, central Brazil.

Appendix 2. Observed herbaceous species pairs for which the null hypothesis was rejected ($\alpha = 0.05$) with the first null model.

Appendix 3. Observed herbaceous species pairs for which the null hypothesis was rejected ($\alpha = 0.05$) with the second null model.

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