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Effect of livestock grazing in the partitions of a semiarid plant–plant spatial signed network

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A B S T R A C T

In recent times, network theory has become a useful tool to study the structure of the interactions in ecological communities. However, typically, these approaches focus on a particular kind of interaction while neglecting other possible interactions present in the ecosystem. Here, we present an ecological network for plant communities that consider simultaneously positive and negative interactions, which were derived from the spatial association and segregation between plant species. We employed this network to study the structure and the association strategies in a semiarid plant community of Cabo de Gata-Níjar Natural Park, SE Spain, and how they changed in 4 sites that differed in stocking rate. Association strategies were obtained from the partitions of the network, built based on a relaxed structural balance criterion. We found that grazing simplified the structure of the plant community. With increasing stocking rate species with no significant associations became dominant and the number of partitions decreased in the plant community. Independently of stocking rate, many species presented an associative strategy in the plant community because they benefit from the association to certain 'nurse' plants. These 'nurses' together with species that developed a segregating strategy, intervened in most of the interactions in the community. Ecological networks that combine links with different signs provide a new insight to analyze the structure of natural communities and identify the species which play a central role in them.

1. Introduction

Scientists have employed network theory to study the interactions within real systems for a long time (Boccaletti et al., 2006), including systems from a variety of fields as economics, sociology, physics, biochemistry, and ecology (Bascompte, 2007). In ecology, network analyses have become a powerful tool to study ecosystems as they address simultaneously the structural properties of a system (e.g. ecological community) and the role that its components (e.g. species) play in that structure (Newman, 2003). However, most research in ecological networks has focused in one particular type of interaction, such as trophic (feeding interactions between predator and prey, Dunne et al., 2002) and mutualistic relationships (mutually beneficial interaction between species, Bascompte and Jordano, 2007), while in nature various interactions happen at the same time. Only recently ecologists have started to analyze networks which include different types of interactions simultaneously, but mostly with theoretical perspective (Fontaine et al., 2011; Kefi et al., 2012; but see Melian et al., 2009). Here we propose an empirical ecological network, which combines positive and negative interactions (competition and facilitation among plants) for the first time.

One particular system of interactions present in nature is the competitive–facilitative interactions that plant species establish among them. Plant species coexist in a particular habitat, where they compete for space and resources with other plants (Fowler, 1986). In addition, some plant species benefit the establishment of other plants because they accumulate nutrients under their canopies or provide protection against environmental factors (Pugnaire et al., 1996). This beneficial effect is called facilitation and has been suggested as a major structural factor influencing the organization of stressed plant communities (Brooker et al., 2008). Interactions between plant species influence the spatial distribution of vegetation, as competitive interactions are related with spatial segregation between plants while positive interactions are related with spatial aggregation (Tirado and Pugnaire, 2005). Although spatial distribution of vegetation depends not only on biotic interactions but on seed dispersal patterns and soil

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heterogeneity (Escudero et al., 2005), it is possible to approximate positive and negative interactions between plant species studying their spatial association and it is a common practice to study plant–plant interactions at community level (Cavieres et al., 2006; Verdu and Valiente-Banuet, 2008; Wiegand and Moloney, 2004).

The study of plant–plant interactions is of particular interest in arid and semiarid ecosystems. In arid ecosystems vegetation is distributed in multi-specific patches within a matrix of bare soil. Typically, patches consist of assemblages of shrubs that act as shelters against harsh environmental conditions ('nurses'), and several other plants that can only survive within these shelters (Armas and Pugnaire, 2005; Sala and Aguiar, 1995). Thus, positive interactions between plant species result in a strong vegetation spatial distribution pattern (Sala and Aguiar, 1995). Spatial association between plant species has been already employed to study the interaction network between plant species in an arid ecosystem, but only considering positive interactions present between 'nurses' and facilitated species (Verdu and Valiente-Banuet, 2008, 2011). We propose to include the negative interactions within the plant community to the network by deriving them from the spatial segregation between species.

In arid and semiarid ecosystems, competition occurs primarily about water, but allelopathy and the exploitation of minerals can be operating as well (Fowler, 1986). These mechanisms mainly act on a local state among neighboring plants and become apparent in reduced size and higher individual plant mortality as compared to plants growing apart. Therefore, competitive interactions drive to spatial segregation of species. It is important to point out that spatial segregation does not only depend on competition but usually is the result of a complex interaction between competition and differences in microhabitat characteristics (Schenk et al., 2003; Schenk and Mahall, 2002). Nevertheless, it has been proposed that resource poor areas such as arid environments express the most intense positive and negative interactions, and consequently, they are the habitats where spatial pattern depends most on biotic interactions (Goldberg et al., 1999).

Overgrazing is one of the most important sources of stress in plant communities. Grazing reduces the abundance of plant species and modifies the composition and structure of plant communities (McNaughton, 1986; Milchunas and Lauenroth, 1993; Olff and Ritchie, 1998). For example, in dry grasslands grazing decreases plant species diversity while in wet environments grazing can promote it (Olff and Ritchie, 1998). In addition, grazing influences the spatial distribution of vegetation. On one hand, associative defense against grazers between grazing-sensitive species and grazing-resistance species may result in an increase of the clustering in the plant community (Baraza et al., 2006; Graff et al., 2007). On the other hand, intensive grazing may result in a random spatial distribution of the plant community with few grazing-resistance species that compete among them (Alados et al., 2004). This change of the spatial distribution may be modulated by grazing intensity, with low intensities related with positive associations and high intensities related with random patterns (Alados et al., 2004). To maintain sustainable grazing practices, it is essential to evaluate the effect of grazers on the entire plant community because this effect is mediated through the single species' tolerance to grazing in a non-additive and very dynamic way (Engel and Weltzin, 2008). The application of network theory aids to translate these species level interactions to the community level.

In this study, we used spatial association networks to investigate the effect of grazing in the structure of the interactions within a semiarid plant community. To our knowledge, this is the first empirical ecological network that employs positive and negative links simultaneously. The combination of different type of interactions allows a more reliable description of the structure of the system under study. Specifically, we divided the plant community in specific partitions, subsets of the species which share the same sign of interactions to other partitions. Within a partition all species associate in the same way to the rest of species in the network, so we consider that these partitions reflect species' association strategies in the plant community. We hypothesize that grazing modifies community association strategies; particularly a) an increase in grazing intensity results in a more randomized plant community and simplifies the association strategies in the community, and b) strategies that are best adapted to grazing are most important structuring the community. We predict that the number of partitions will decrease and the plant community will become more randomly associated as stocking rate increases. We also predict that spatial associations will concentrate in partitions that include the species that provide refuge from grazers to others ('nurse'), while partitions including other species (e.g. facilitated species) will present few spatial associations.

2. Methods

2.1. Study area

The study was conducted in Cabo de Gata-Níjar Natural Park (hereafter, Cabo de Gata NP), which is in the Mediterranean coast of southeastern Spain (36° 46′ N, 2° 09′ W). The park covers 37,570-ha, and the highest elevation is 493 m (El Fraile Peak). The climate is semiarid Mediterranean (drought in summer and most rainfall in spring and autumn. Average annual rainfall = 193.9 mm, Average annual temperature = 19.4 °C, Passera, 1999). Historically, the area has been used as an agro-pastoral system, with cereal cropping on floodplains and livestock (sheep and goats) grazing year-round on the slopes. The plant community is characterized by *Chamaerops humilis* L., *Rhamnus lycioides* L., *Pistacia lentiscus* L. and *Periploca laevigata* Aiton (Peinado et al., 1992). Vegetation data were collected from the southern section of the park, where highly stony soils predominate (Oyonarte et al., 1999). In that region, the vegetation is an open shrubland, with shrubs organized in patches embedded within a matrix of a large tussock grass, *Stipa tenacissima* L. *Stipa tenacissima* is a highly competitive species that colonizes the gaps created within patches by livestock and aridity, and can exclude other plant species from the area (Alados et al., 2006, 2003).

Within the study area, four sites with different stocking rate were selected near the El Romeral farm. One week per season in 2000, the movements of sheep and goats were monitored and the effective stocking rate at each site was calculated multiplying the average stocking rate of the farm (0.65 ind ha^{-1}) by a correction factor based on the proportion of time that livestock spent grazing on that site. Then, sites were ranked based on the stocking rate to which they were exposed ($G1 = 0$ ind/ha; $G2 = 0.27$ ind/ha; $G3 = 0.46$ ind/ha; and $G4 = 0.65$ ind/ha). Grazing carrying capacity of this plant community was 0.39–0.57 ind/ha (Robles and Passera, 1995); therefore, we assumed that G2, G3 and G4 sites had been exposed to low, medium and high grazing intensity, respectively. Furthermore, we confirmed that G1 site had not been grazed for decades.

2.2. Vegetation survey and network construction

In April, 2001, three 500-m-long linear transects were established randomly at each site, and the Point-Intercept Method was used to quantify the vegetation (Goodall, 1952). In each transect one survey point was established every 20 cm, and all species which intercepted a vertical line on each point were recorded (T = total surveyed points per transect = 2501). Only the presence of each species per point was recorded, independently of the number of individuals, and no distinction was made between the ontogenetic stages of individuals of the same species. We considered each point separated 20 cm as an independent unit which could lead to an overestimation of species co-occurrences for wider species. However, we confirmed that our main findings did not change when considering only points separated 1 m. Nevertheless, as several works in ecological networks have demonstrated that network indexes are very dependent on survey effort (Chacoff et al., 2012; Petanidou et al., 2008), we decided to present the results derived from the points separated 20 cm as a more realistic representation of the plant community ($T = 2501$ in 20 cm separated transects vs. $T = 501$ in 1 m separated transects). The abundance of each species i per transect was measured as the number of points where it occurred (n_i). Evenness ($E = H'/\ln(S)$, where H' is Shannon diversity index and S is the number of species present) was calculated for each transect. Linear transects ran parallel to the slope, separated by at least 50 m and at the same elevation, orientation and parent soil material.

For each site, three plant–plant spatial association networks were created using the data from each transect (one network per transect) and based on the plant–plant association matrix A_{SxS} (Saiz and Alados, 2012). In these networks nodes are plant species and links are spatial associations between pairs of species. In A_{SxS}, a_{ij} represents the spatial association between species i and j, with $a_{ij} = a_{ji}$ as A_{SxS} was symmetric. The spatial association between i and j was derived by comparing the number of times that species i and j co-occurred at the same sampling point in the transect (c_{ij}) and the expected number of co-occurrences based on species abundances ($e_{ij} = n_i/T \times n_j/T \times T$, probability of i appearing at a single point multiplied by the probability of j appearing at a single point multiplied by the total number of points). Each c_{ij} was compared to a Poisson distribution fitted with e_{ij} as parameter. When c_{ij} was significantly higher than e_{ij} ($c_{ij} > e_{ij}$, and fell outside the 95% confidence interval of e_{ij}), plant species were assumed to be positively associated and a_{ij} was set to $+1$. When c_{ij} was significantly lower than e_{ij} ($c_{ij} < e_{ij}$, and fell outside the 95% confidence interval of e_{ij}), plant species were assumed to be negatively associated and a_{ij} was set to -1. If c_{ij} fell within the confidence interval, the observed values did not differ significantly from those expected and the plant species were assumed to have a neutral association ($a_{ij} = 0$). It was not possible to estimate the number of co-occurrences of a species with itself from the point-intercept data; therefore, the diagonal terms of A_{SxS} were set to 0. The total number of links in the network was L and the association ratio was $Ratio = (L^{+}-L^{-})/(L^{+}+L^{-})$, where L^{+} was the total number of positive links and L^{-} was the total number of negative links in the network. Positive *Ratio* values indicated that positive associations were more frequent than negative ones, while negative *Ratio* values indicated the contrary (similar to relative interaction intensity index, Armas et al., 2004). We applied linear regression models with stocking rate as explanatory variable and S, E and *Ratio* as response variables.

Signed networks (*i.e.* networks with positive and negative links) can be partitioned attending to structural balance criterion (Doreian and Mrvar, 2009; Traag and Bruggeman, 2009). In a balanced signed network, all nodes within the same partition exhibit positive links among themselves and negative links with nodes in the other partitions. Under that criterion, however, not all signed networks can be balanced and a Frustration Index (e.g. $F =$ proportion of links that do not fulfill balance criterion) is used to measure the distance of a network from balance ($F = 0$ for a balanced network, Traag and Bruggeman, 2009). To deal with frustration, conditions for structural balance can be relaxed. In a signed network with relaxed balance, all links within a partition exhibit the same sign (positive or negative), which also applies to the links between pairs of partitions (*i.e.* all links between nodes in partition A and nodes in partition B have the same sign). Under this criterion, all signed networks can be balanced as long as they are divided in a sufficient number of partitions (Doreian and Mrvar, 2009). In our case each association network A_{SxS} was divided in partitions attending to relaxed structural balance criterion until balanced was achieved (all links between nodes within a partition and between nodes in pairs of partitions have the same sign). Moreover, three of the partitions were set specifically: P^{+}, which included all the species that only exhibited positive associations; P^{-}, which included all the species that only exhibited negative associations; and P^{0}, which included all the species with no significant associations. Other partitions were named as P^{x} (P^{I}, $P^{II}\ldots$).

Attending to relaxed structural balance criterion, all species within a partition associate in the same way to the rest of species in the network. Thus, we could argue that those species belonging to the same partition share the same association strategy in the plant community. This is quite straightforward for P^{+}, P^{-} and P^{0} partitions: P^{+} includes facilitated species (species that only present positive associations), P^{-} includes competitive species (species that only present negative associations) and P^{0} includes ruderal species (species that do not present significant associations). However, when species present positive and negative links (species in P^{x}) association strategy is not clear. The organization of these species in different partitions is related only with the associations that they establish among themselves, as they only establish positive associations with P^{+} and negative associations with P^{-}. In our case (semiarid environment where vegetation is organized in patches), one possible interpretation is that P^{x} includes, among others, ‘nurse’ species. Usually, ‘nurse’ species are abundant species adapted to the environmental conditions present in the area (Choler et al., 2001). These species form vegetation patches where other species establish, while segregate from their competitors. Nevertheless, our interpretation of P^{x} must be taken cautiously as P^{x} can also include species that only establish in patches formed by certain species and segregate from other patches. In our case, number of partitions (K) would reflect the diversity of association strategies in the community. Network analyses were performed using Pajek (http://pajek.imfm.si/doku.php).

2.3. *Data analysis*

In order to evaluate the effect of grazing in the organization of the networks, we applied linear regression models with stocking rate as explanatory variable and number of partitions, proportion of non-associative plants and links density as response variables. Number of partitions (K) represents the diversity of association strategies in the plant community, while proportion of non-associative plants ($NAs = S^{P0}/S$, where S^{P0} is the size of P^{0}) and number of links per species (linkage density, $D = L/S$) represents the spatial randomization of plant species. High NAs indicates that several species do not present any significant spatial association in the community, and low D indicates that most species spatially associate between them neutrally. As these variables could be related with the size of the network, we also included S as explanatory variable. Linear regression models were performed with R (http://www.Rproject.org).

To analyze the association strategies of the community we first compared the relative size of partitions (S^{Pi}/S, where S^{Pi} is the number of species in partition P^{i}) across all networks. We employed an ANCOVA with partition identity (P^{-}, P^{x}, P^{+} and P^{0}) and stocking rate as explanatory variables and relative size of partitions as response variable. Then, to confirm which strategy was more important, we compared the number of links per species that occurred in each partition ($D^{Pi} = L^{Pi}/S^{Pi}$, where L^{Pi} is the total number of positive and negative links for species within partition P^i) and the number of links per species with at least one link present in the network ($D' = L/(S-S^{P0})$, as only species that present at least one significant association were considered). In this way we can compare if there were differences between partitions and also if certain partitions gather more associations than expected in the network. If an association strategy plays a major role structuring the plant community, its corresponding partition would present more links per species than the expected number of links per species in the network without P^0 ($D^{Pi} > D'$). We applied an ANCOVA using partition identity (P^-, P^x, P^+ and *Network'* = all species that present at least one significant association) and stocking rate as explanatory variables and links per species (D^{Pi} and D') as response variable. The size and number of links for P^x were calculated for each transect after pooling all partitions that present both positive and negative associations. If significant differences were found, we applied a Tukey test on the residuals on the grazing effect to assess pair-wise comparisons between partitions. ANCOVAs and Tukey test were performed with R (http://www.Rproject.org).

3. Results

In Cabo de Gata NP, livestock grazing influenced plant community and its respective association networks (Table 1). Specifically, linear models showed significant negative effect of stocking rate on species richness and evenness of the plant community (S, Estimate = −54.627, adjusted R^2 = 0.554, p-value < 0.003; E, Estimate = −0.377, adjusted R^2 = 0.857, p-value < 0.001). Positive associations were more frequent than negatives in all sites (*Ratio* > 0 in all sites), and stocking rate did not present a significant effect on *Ratio* (*Ratio*, Estimate = −0.304, adjusted R^2 = 0.0758, p-value = 0.198).

Network parameters showed dependence on grazing intensity but not on species richness. Thus, we removed species richness as explanatory variable from the models. In simplified models, stocking rate presented a significant negative effect on the number of partitions and links per species and positive effect on the proportion of non-associative species (Table 2).

Plant species did not distribute equally among partitions. There was a significant effect of the identity of the partitions on partition's relative size, but stocking rate did not affect it (S^{Pi}/S, Table 3). Regardless of the grazing effect, Tukey test revealed that partitions P^+ and P^0 presented significantly more species than P^- and P^x (Fig. 1). There was also a significant effect of partition's identity and stocking rate on the number of links per species (D^{Pi}, Table 3). After removing the effect of grazing, Tukey test showed that D^{Px} and D^{P-} were significantly higher than D^{P+}, and D^{P-} was significantly higher than D' (Fig. 2). Thus, P^- and P^x included fewer species than P^+ and P^0 but concentrated more associations, specially P^- which presented more associations than expected in the network. In Fig. 3 we present an example of the signed networks along the stocking rate gradient to illustrate how associations distribute among partitions.

Table 1
Characteristics of the plant community in the study sites of Cabo de Gata NP.

Site	Transect	Stocking rate	S	E	L	D	L^+/L^-	Ratio	K
G1	1	0	96	0.694	174	1.813	142/32	0.632	5
	2	0	92	0.685	194	2.101	148/46	0.526	6
	3	0	81	0.642	162	2	116/46	0.432	7
G2	1	0.27	58	0.603	60	1.034	46/14	0.533	5
	2	0.27	53	0.607	82	1.547	54/28	0.317	6
	3	0.27	48	0.556	50	1.042	26/24	0.04	5
G3	1	0.46	46	0.58	76	1.652	58/18	0.526	6
	2	0.46	57	0.568	66	1.158	48/18	0.455	5
	3	0.46	49	0.528	56	1.143	34/22	0.214	5
G4	1	0.65	57	0.404	62	1.087	46/16	0.484	4
	2	0.65	45	0.399	34	0.756	18/16	0.059	4
	3	0.65	60	0.43	54	0.9	36/18	0.333	4

Stocking rate, livestock individuals/hectare; S, species richness; E, evenness; L, total number of links; D, links per species; L^+, number of positive links; L^-, number of negative links; *Ratio*, association ratio; K, number of partitions. Sites are ranked from 1 to 4 attending to increasing grazing intensity.

Table 2
Effect of stocking rate in plant–plant spatial association networks parameters.

Response variable	Explanatory variable	Estimate	Standard error	F-value	p-Value
K	Intercept	6.118	0.335	18.26	<0.001***
	Stocking rate	−2.757	0.797	−3.46	0.006**
NAs	Intercept	0.29	0.039	7.332	<0.001***
	Stocking rate	0.327	0.094	3.486	0.006**
D	Intercept	1.863	0.136	13.655	<0.001***
	Stocking rate	−1.477	0.325	−4.552	0.001**

K, number of partitions; *NAs*, proportion of non-associative species; D, number of links per species. Significant effects are highlighted by ** (p < 0.01) and *** (p < 0.001). K, adjusted R^2 = 0.499, p-value for the model = 0.006**; *NAs*, adjusted R^2 = 0.503, p-value for the model = 0.006**; D, adjusted R^2 = 0.642, p-value for the model = 0.001**.

4. Discussion

In the present work we studied the effect of grazing on a plant community employing network theory. The inclusion of positive and negative links in the same network supposes a step further in the development of ecological networks and improves our understanding about the structure of natural systems (Fontaine et al., 2011). Specifically, the analysis of the partitions in the semiarid plant–plant network in Cabo de Gata-Níjar NP reveals the effect that grazing had on the spatial structure of the community. Specifically, an increase in stocking rate randomized plant community spatial associations and reduced the number of association strategies present (Fig. 3). Furthermore, competitive and 'nurses' were the most important species structuring the plant community as they presented the highest linkage density.

Grazing influenced the semiarid shrubland community and its respective spatial association network in Cabo de Gata NP, Spain. The presence of livestock reduced species richness and as grazing became more intense, community presented lower evenness. This can be explained by the selective consumption by herbivores which was found to provide dominance of resistant plant species and reduce community biodiversity (Bisigato and Bertiller, 1997). The prevalence of positive associations in all sites (*Ratio* > 0, Table 1) coincide with the results suggesting that facilitation is a key factor structuring semiarid communities (Sala and Aguiar, 1995). As grazing-sensitive species can persist in grazed areas if they associate with grazing-resistant species that act as shelters against herbivores, we could expect an increase in the importance of

Table 3
Effect of partition's identity and stocking rate for the distribution of species and links among partitions in plant–plant spatial association networks.

Response variable	Explanatory variable	df	SS	MS	F-value	p-Value
S^{Pi}/S	Partition's identity	3	1.2848	0.4287	58.3975	<0.001***
	Stocking rate	1	0.0001	0.0001	0.0126	0.9113
	Residuals	42	0.308	0.0073		
D^{Pi}	Partition's identity	3	94.967	31.656	10.9452	<0.001***
	Stocking rate	1	22.613	22.613	7.81887	0.0078**
	Residuals	42	121.472	2.892		

S^{Pi}/S, relative size of partition P^i; D^{Pi}, number of links per species in partition P^i; df, degree of freedom; SS, sum square; MS, mean square. Significant effects are highlighted by ** (p < 0.01) and *** (p < 0.001).

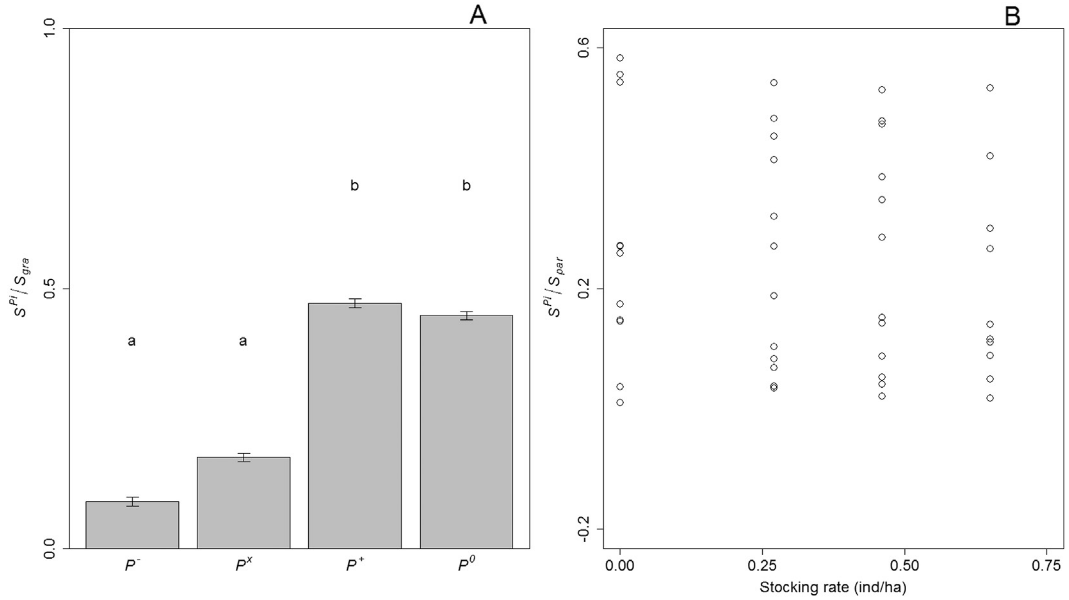

Fig. 1. Effect of grazing on the relative size of the partitions in plant–plant association networks of Cabo de Gata NP. A. S^{Mi}/S *gra*, residuals of the relative size of the partition P^i after removing the effect of stocking rate; P^-, partition with only negative links; P^x, partition with positive and negative links; P^+, partition with only positive links; P^0, partition with no links. S^{Pi}/S *gra* values are significantly different between partitions with different letters (differences were tested with Tukey test). B. S^{Mi}/S *par*, residuals of the relative size of the partition P^i after removing the effect of the identity of the partition. There was not a significant effect of stocking rate on S^{Mi}/S *par*.

positive association with grazing intensity (Baraza et al., 2006; Graff et al., 2007; Smit et al., 2007). However, we did not find a significant effect of stocking rate on the association ratio (*Ratio*). It has been proposed that the effect of grazing in vegetation spatial patterns and plant species interactions can interact with other source of stress (e.g. aridity, Milchunas and Lauenroth, 1993; Smit et al., 2009). Thus, it is possible that in arid areas predictions about a gain in positive associations with grazing intensity do not apply.

Attending to partitions in the networks, selective consumption of species and community randomization is visible. The number of partitions in the networks and the number of links per species decreased with stocking rate while the proportion of species with no significant associations increased. At the ungrazed site, the networks had the highest number of partitions which indicated diverse relationships among the species present in the plant community (*i.e.* the plant community presented many species that associated with some species but segregated from others). In

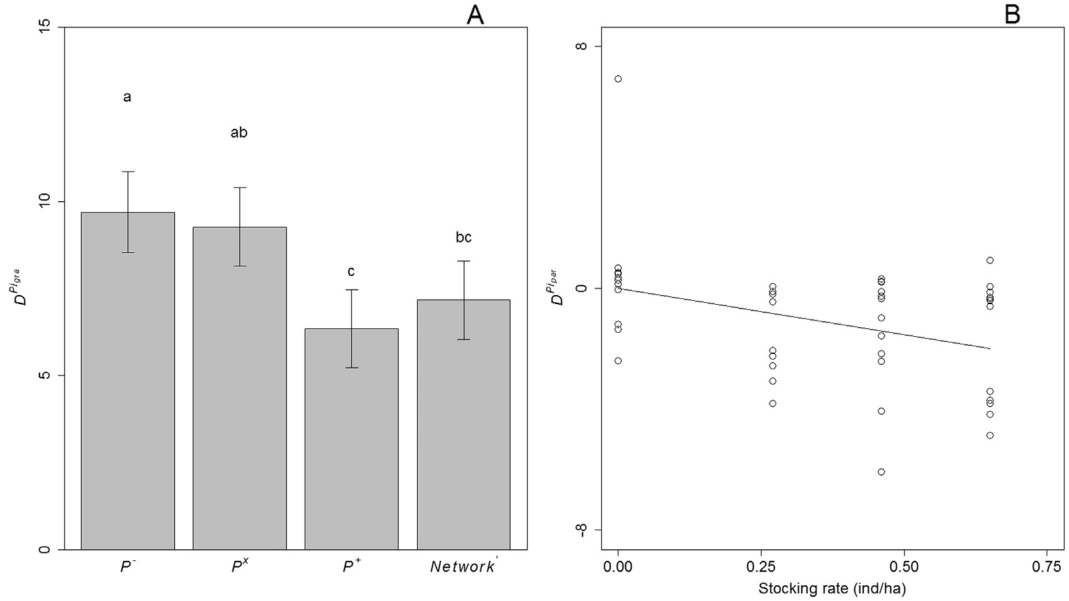

Fig. 2. Effect of grazing on the number of links per species for the partitions in plant–plant association networks of Cabo de Gata NP. A. D^{Pi} *gra*, residuals of the number of links per species in each partition P^i and in *Network'* after removing the effect of stocking rate; P^-, partition with only negative links; P^x, partition with positive and negative links; P^+, partition with only positive links; *Network'*, all the species with at least one significant association. D^{Pi} *gra* values are significantly different between partitions with different letters (differences were tested with Tukey test). B. D^{Pi} *par*, residuals of the number of links per species in the partition P^i after removing the effect of the identity of the partition. There was a significant effect of stocking rate on D^{Pi} *par* (stocking rate estimate = −3.065, adjusted R^2 = 0.104, *p*-value = 0.016*).

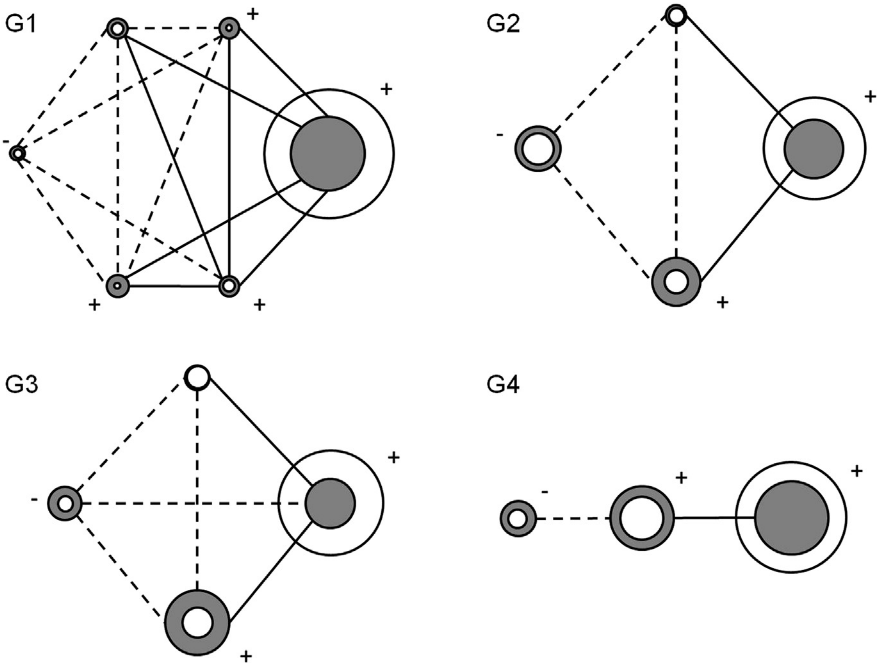

Fig. 3. Plant–plant spatial association networks along a grazing intensity gradient in Cabo de Gata NP. G1 to G4 are the vegetation sampling sites ranked attending to increased grazing intensity. Networks are built with the data of transect 3 of each site. Continuous lines are positive links and dashed lines are negative links. Circles are the association partitions. Symbols next to circles are the sign of the links within the partition. Size of white circles is proportional to the number of species in the partition, and the size of gray circle is proportional to the number of links within the partition. When gray circle is bigger than white, partition has more links than expected, while when gray circle is smaller than white, partition has fewer links. Partition P^0 is not represented.

semiarid plant communities where there is no grazing, functional traits of species become more diverse and the web of interactions within the community becomes more complex (Bisigato and Bertiller, 1997). We assume that with the appearing of livestock, some species disappeared from the community while several species that do not associate to others established, reducing the number of associations per species. The decrease in the number of partitions with stocking rate can be explained by the preferential feeding of grazers on palatable plants growing without protection through 'nurse' plants, which leads to a selection on association strategies (Smit et al., 2009, 2007). Both the establishment of non-associative species and simplification of association strategies were favored by an increase in stocking rate, particularly at the site with highest stocking rate (G4). In this site stocking rate is higher than the carrying capacity of the plant community, and we found that vegetation patches scarcely persist and community spatial structure differentiates from that of sites with stocking rates below carrying capacity (G2 and G3 in Fig. 3). These findings can be explained by a reduction of facilitative effects in plant community due to high herbivore pressures. The intense grazing caused severe damage to 'nurses', which could not act as proper shelters against grazing and/or aridity (Graff et al., 2007; Smit et al., 2007).

In our plant community, we found that independently of stocking rate competitive (species in P^{-}) and 'nurses' (species in P^{x}) exhibited more links than facilitated species (Fig. 2). This reflects the importance of these association strategies structuring the community. Each species in P^{x} exhibited negative associations with species in P^{-} and positive associations with many species in P^{+}. Namely, species in P^{x} included the species which occupied the gaps created by the competitive species of P^{-} and acted as refuges for species in P^{+}. 'Nurse' species, which concentrate most of the positive associations in semiarid plant communities are responsible for patch formation in these areas (Aguiar and Sala, 1999), and also can act as shelter against herbivores when grazing is present (Baraza et al., 2006). 'Nurse' species are important in maintaining ecosystems biodiversity and have been identified as key species for conservation purposes (Gomez-Aparicio et al., 2008; Maestre et al., 2001; Pueyo et al., 2009). Species in P^{+}, however, exhibited fewer associations than expected. Usually, in arid plant communities most species require facilitative interactions in order to survive (Verdu and Valiente-Banuet, 2008). The succession in these communities begins with the establishment of some 'nurse' species which attract several species under their canopies (Sala and Aguiar, 1995). Therefore, 'nurse' species gather many facilitated species under their canopy which results in high number of positive associations, while facilitated species associate to few 'nurse' and so, their number of associations is reduced.

In our study, spatial association between plants was identified based on the estimated abundances of the species. Many studies of community ecology do not include rare species in the analyses because low abundance precludes sufficient statistical power (Choler et al., 2001; Tirado and Pugnaire, 2005). For example, in our study when species are very infrequent, negative associations cannot be detected (the minimum observable number of co-occurrences, $c_{ij} = 0$, falls within the 95% confidence interval of e_{ij}). Thus, all the species within the partitions with negative associations (P^{-}, P^{x}) are abundant. However, we included all plant species found in the study area because positive associations could be detected for all pairs of species. Choler et al. (2001) indicated that the distribution of abundant species depends on abiotic conditions, while the distribution of rare species depends on biotic interactions with abundant species. In our case, our results agree with this hypothesis because more than half of the species that had a cover <1% exhibited at least one positive association with other species ($G1 = 77\%$, $G2 = 63\%$, $G3 = 58\%$, $G4 = 51\%$). As stocking rate increased, the proportion of species with positive associations decreased due to the appearance in the community of species that do not present any significant association.

It is important to consider that our approach presents some limitations. First, we considered plant association patterns between pairs of species in the community as an indicator of possible interactions between those plants, but spatial pattern is an indirect measure of ecological interactions. Spatial pattern is the net result of biotic interactions, seed dispersal, and environmental heterogeneity (Escudero et al., 2005). For example, at the ungrazed site in Cabo de Gata NP, plant species were positively associated within multi-species patches, where abiotic conditions were ameliorated, and not as result of defense against grazers (Pugnaire et al., 2011). Thus, we cannot grant that positive associations in grazed sites are only due to the effect of livestock. Second, plant species spatial strategy can vary over their life cycle, shifting from positive associations as seedlings to negative associations as adults (Miriti, 2006). As we did not distinguish among the life stages of the species, in our case the association strategy of each species was the net outcome of the different association strategies that species present over all its life stages. These limitations notwithstanding, our results are similar to those of other works on the organization of interactions and the effects of grazing in the structure of plant communities (Graff et al., 2007; Smit et al., 2007). Future analyses will attempt to validate the observed patterns in other plant communities and, specifically in our study area, to identify the functional traits related with each particular strategy and the role that each species plays in maintaining community structure.

5. Conclusions

Ecological networks that combine different types of links provide a new insight to analyze the structure of natural communities. In Cabo de Gata NP, plant–plant association networks showed that grazing influences the structure of the interactions within the plant community. At the study area, the complex association patterns that were apparent at the ungrazed site were diminished as the grazing pressure increased. Specifically, grazing randomized the spatial associations in the plant community and reduced the number of spatial association strategies in the study area. Independently of stocking rate, several rare species appeared associated with few abundant species. These abundant species played an important role in maintaining the structure of the plant community because they concentrated most of the positive and negative interactions. Species that only exerted positive associations exhibited fewer associations than expected. Ecological networks that combine links with different signs provide a new insight to analyze the structure of natural communities. Extending community studies to consider simultaneously different types of interactions represents a necessary step to identify the role that species fulfill in ecosystems.

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