

LETTER

Opposing effects of floral visitors and soil conditions on the determinants of competitive outcomes maintain species diversity in heterogeneous landscapes

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Abstract

Theory argues that both soil conditions and aboveground trophic interactions have equivalent potential to limit or promote plant diversity. However, it remains unexplored how they jointly modify the niche differences stabilising species coexistence and the average fitness differences driving competitive dominance. We conducted a field study in Mediterranean annual grasslands to parameterise population models of six competing plant species. Spatially explicit floral visitor assemblages and soil salinity variation were characterised for each species. Both floral visitors and soil salinity modified species population dynamics via direct changes in seed production and indirect changes in competitive responses. Although the magnitude and sign of these changes were species-specific, floral visitors promoted coexistence at neighbourhood scales, while soil salinity did so over larger scales by changing the superior competitors' identity. Our results show how below and aboveground interactions maintain diversity in heterogeneous landscapes through their opposing effects on the determinants of competitive outcomes.

Keywords

Coexistence, community assembly, demography, fitness, multitrophic interactions, mutualism, niche, pollinators, salinity, spatial structure.

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INTRODUCTION

One central aim in ecology is understanding how plant species diversity is maintained. Extensive empirical work has documented that variation in soil conditions and multitrophic interactions modulate key processes of plant population dynamics. For instance, plant offspring and the strength of competition depend on the combined species' ability to deplete shared limiting soil resources (Tilman 1982) and to cope with stressful soil conditions such as the amount of salt (Bertness & Shumway 1993; Crain *et al.* 2004). Likewise, mutualistic and antagonistic biotic interactions with herbivores (Hulme 1996; Olf & Ritchie 1998), soil biota (Bever 2003; Bennett *et al.* 2017; Teste *et al.* 2017), pathogens (Mordecai 2015; Parker *et al.* 2015) and floral visitors (Bastolla *et al.* 2009; Carvalheiro *et al.* 2014; Weber & Strauss 2016) can profoundly impact plant performance. It is obvious that outcrossing plants directly depend on their mutualistic floral visitors to maximise their reproductive success (Morris *et al.* 2010; Ollerton *et al.* 2011). Subtler is the fact that floral visitors indirectly mediate competition among plants through a wide variety of density-dependent processes including variation in the number and diversity of floral visitors as well as heterospecific pollen deposition (Moeller 2004; Arceo-Gómez & Ashman 2011; Runquist & Stanton 2013).

Although variation in soil conditions and multitrophic biotic interactions occurs simultaneously in nature, evaluations of their effects on modulating the strength and sign of plant competition have been explored individually. It remains unknown how these drivers can jointly maintain plant

diversity. Theoretical work has advocated that both types of interactions should be viewed symmetrically as they have equivalent potential to limit or promote diversity (Chesson & Kuang 2008). Although empirical tests of this prediction have remained so far elusive, we can progress by framing our research within recent advances of coexistence theory (Chesson 2000), and by learning from prior work applying these theoretical advances to multitrophic antagonistic interactions, mainly predators and pathogens (Chesson & Kuang 2008; Kuang & Chesson 2010; Stump & Chesson 2017).

According to Chesson's (2000) framework, both soil conditions and floral visitors can promote the stabilising niche differences that favour plant coexistence, which occur when intraspecific competitive interactions exceed interspecific competition, and the average fitness differences that favour competitive exclusion and determine the competitive winner in the absence of niche differences. Ecologists have paid much more attention to the relationship of these two factors with fitness differences (e.g. soil conditions (Tilman 1982; Casper & Jackson 1997; Rees 2013); floral visitors (Herrera 2000; Waites & Ågren 2004; Arceo-Gómez & Ashman 2011)) than with niche differences (Silvertown 2004; Levine & HilleRisLambers 2009) and this is particularly evident for floral visitors (Pauw 2013). Most likely both drivers modify niche and fitness differences simultaneously, yet the extent of such modifications is poorly understood. Therefore, a rigorous evaluation of the equivalent potential of soil conditions and floral visitors on maintaining plant diversity can only be done by a mechanistic understanding of how these two types of interactions relatively modify the determinants of competitive outcomes.

When relating theory to field experiments, it is important to consider two critical aspects. One is selecting ecological systems that are relatively easy to observe. For example, recent work in Mediterranean annual grassland (Godoy & Levine 2014) has described how niche and fitness differences influence species' population dynamics. Some of these grasslands are subjected to strong variation in soil salinity, which negatively correlates with soil fertility (Olf & Ritchie 1998; Hu & Schmidhalter 2005). Moreover, floral visitor assemblages in these Mediterranean environments are particularly interesting because they are composed of an array of insects including solitary bees, hover flies, beetles and butterflies. While some of these floral visitor types act as true mutualisms (Pauw 2013), others rob plant nectar or pollen or damage flowers (Morris *et al.* 2003). The second aspect is that these drivers of plant competition tend to show spatial structure (Tilman 1994; Weber & Strauss 2016). Coexistence theory predicts that plant diversity can be maintained at the neighbourhood scale when species niche differences overcome fitness differences. This can occur when either plant competitor equalises its fitness differences, increases its niche differences or a combination of both. If not, the superior competitor excludes the inferior species (Chesson 2000). However, competitive exclusion outcomes can also maintain plant diversity at larger scales if the spatial structure of variation in soil salinity and floral visitors change the identity of the superior competitor across locations. This latter process might be the cause of reported turnover patterns of species and functional attributes across soil salinity gradients (Bertness 1991; Pavoine *et al.* 2011).

Here, we considered three layers to test how the below-ground environmental conditions (i.e. soil conditions) and the aboveground trophic interactions (i.e. floral visitors) influence coexistence of the middle layers (i.e. plant species). We specifically focus on three questions: (1) How do soil salinity and floral visitors modify species' population dynamics via direct changes in per capita seed production and indirect changes in species' responses to competitive interactions? (2) Do these direct and indirect effects modify niche and fitness differences between plant species? and finally, (3) At which spatial scale are these modifications on the determinants of competitive outcomes limiting or promoting diversity?

We answered these three questions by first parameterising a general plant competition model from which the stabilising niche differences and average fitness differences were quantified. To parameterise these models of pairwise competition between six annual native grassland species, we quantified their vital rates and competition coefficients in field plots relating seed production of focal individuals to a density gradient of numerous different competitors. We then assessed how seasonal and spatial variation in the number of floral visits and in soil salinity changes species fecundity and their responses to competition (Question 1). Once, the model was parameterised, we estimated niche and fitness differences with and without considering the effect of soil salinity and floral visitors on species fecundity (Question 2), and compared how strong niche differences offset fitness differences between scenarios (Question 3). Our work is novel in quantifying the effects that distinct environmental conditions and trophic interactions have on modifying niche and fitness differences,

and showing under field conditions that they maintain diversity in heterogeneous landscapes through their opposing effects on the determinants of competitive outcomes.

METHODS

Study system

Our study was conducted in Caracoles Ranch (2680 ha), an annual grassland system located in Doñana NP, southwest Spain (37°04'01.5" N 6°19'16.2" W). The climate is Mediterranean with mild winters and average 50-year annual rainfall of 550–570 mm with high interannual oscillations (Muñoz-Reinoso & García Novo 2000). Soils are sodic saline (electric conductivity > 4 dS/m and pH < 8.5) and annual vegetation dominates the grassland with no perennial species present. The study site has a subtle micro topographic gradient (slope 0.16%) enough to create vernal pools at lower parts from winter (November–January) to spring (March–May) while upper parts do not get flooded except in exceptionally wet years. A strong salinity-humidity gradient is structured along this topographic gradient. Additionally, salt can reach upper parts of the soil by capillarity resulting overall in heterogeneous soil salinity patterns at the local and at the landscape scale (Appendix S1). This salinity gradient is strongly correlated with soil nutrient availability at our study location, and more saline conditions correlate with less fertile soils (Clemente *et al.* 2004).

We recorded 19 annual plants at the study site. Of this initial species set, three were not further considered due to their low abundance (only recorded in 5 out of the 324 subplots evaluated). The 16 species finally selected represent a broad range of taxonomic families, plant morphology and flowering phenology co-occurring at the scale of the entire study system. All species were considered for estimating competitive interactions, but we only observed enough visits of insects to the flowers of six species. Hence, we further focus on this particular set of species to compare the effect of soil salinity and floral visitors on niche and fitness differences (Table 1).

Modelling approach to quantify the niche and fitness differences between species pairs

Our observational study was designed to field-parameterise a mathematical model describing annual plant population dynamics (Levine & HilleRisLambers 2009). This model allows quantifying stabilising niche differences and average fitness differences between species within a trophic level (Godoy & Levine 2014). Importantly, there have not been previous attempts to quantify how soil condition or multitrophic interactions change the strength of niche and fitness differences between species within a single trophic level, and here we show how these effects can be incorporated into this model. The model is described as follows:

$$\frac{N_{i,t+1}}{N_{i,t}} = (1 - g_i)s_i + g_iF_i \quad (1)$$

where $\frac{N_{i,t+1}}{N_{i,t}}$ is the per capita population rate, and $N_{i,t}$ is the number of seeds of species i in the soil prior to germination in

Table 1 List of species observed in Caracoles Ranch. Code and taxonomic family of each species is provided. Sample size represents the total number of individuals sampled for each focal species, and it is correlated with their natural abundance at the study site

Species	Family	Code	Floral visitors	Sample size
<i>Beta macrocarpa</i>	Amaranthaceae	BEMA	Yes	289
<i>Chamaemelum fuscatum</i>	Asteraceae	CHFV	Yes	162
<i>Chamaemelum mixtum</i>	Asteraceae	CHMI	Yes	5
<i>Centaurium tenuiflorum</i>	Gentianaceae	CETE	No	23
<i>Frankenia pulverulenta</i>	Frankeniaceae	FRPU	No	5
<i>Hordeum marinum</i>	Poaceae	HOMA	No	289
<i>Leontodon maroccanus</i>	Asteraceae	LEMA	Yes	273
<i>Melilotus elengans</i>	Fabaceae	MEEL	Yes	77
<i>Melilotus sulcatus</i>	Fabaceae	MESU	Yes	229
<i>Plantago coronopus</i>	Plantaginaceae	PLCO	No	171
<i>Polypogon monspeliensis</i>	Poaceae	POMO	No	20
<i>Pulicaria paludosa</i>	Asteraceae	PUPA	Yes	124
<i>Scorzonera laciniata</i>	Asteraceae	SCLA	Yes	101
<i>Spergularia rubra</i>	Caryophyllaceae	SPRU	Yes	44
<i>Sonchus asper</i>	Asteraceae	SOAS	Yes	87
<i>Suaeda splendens</i>	Amaranthaceae	SUSP	No	29

winter of year t . The germination rate of species i , g_i , can be viewed as a weighting term for an average of two different growth rates: the annual survival of ungerminated seed in the soil (s_i), and the viable seeds produced per germinated individual (F_i). In past work, F_i was expanded into a function describing how the average fecundity of each germinated seed that becomes an adult (i.e. per germinant fecundity) declines with the density of competing number of individuals in the system (Godoy & Levine 2014). Now, we slightly modify this function to include the additional effect of soil conditions and floral visitors on the per germinant fecundity as follows:

$$F_i = \frac{\lambda_i(1 + \theta_{i,s}S_t + \gamma_{i,fv}A_t)}{1 + \sum(\alpha_{ij} + \psi_{ij,s}S_t + \omega_{ij,fv}A_t)g_jN_{j,t}} \quad (2)$$

where $\theta_{i,s}$ and $\gamma_{i,fv}$ control the effect of soil salinity (S_t) and floral visitors (A_t), respectively, on the per germinant fecundity of species i in the absence of competition (λ_i). In addition, λ_i is modified by the germinated densities of other species including its own ($g_jN_{j,t}$). To describe the per capita effect that species j is mediating on species i , we multiplied these germinated densities by a sum of three interaction coefficients ($\alpha_{ij} + \psi_{ij,s} + \omega_{ij,fv}$), which describes the additional direct effect of soil salinity (S_t) and the apparent effect of floral visitors (A_t) on the competitive interactions between species. Notice that we considered explicitly in our study the effect that soil salinity and floral visitors have on species' fecundity (F_i), but not on the other two vital rates, seed germination (g_i) and seed soil survival (s_i). This limitation is not due to the fact that the model cannot be extended to account for multitrophic effects on these two vital rates. Rather, it was because we did not follow seed performance over several years.

With the direct and apparent dynamics of competition described by this population model, we followed the approach of Chesson (2012) to determine fitness and niche differences between species pairs. Our procedure here parallels previous work described in Godoy & Levine (2014), and allows us to define stabilising niche differences and fitness differences

without and with considering the effect of floral visitors and soil salinity on plant coexistence. For the model described by eqns (1) and (2), we define niche overlap (ρ) as follows:

$$\rho = \sqrt{\frac{\alpha_{ij} + \psi_{ij,s}S_{i,t} + \omega_{ij,fv}A_{i,t}}{\alpha_{jj} + \psi_{jj,s}S_{j,t} + \omega_{jj,fv}A_{j,t}} \times \frac{\alpha_{ji} + \psi_{ji,s}S_{j,t} + \omega_{ji,fv}A_{j,t}}{\alpha_{ii} + \psi_{ii,s}S_{i,t} + \omega_{ii,fv}A_{i,t}}} \quad (3)$$

If multitrophic interactions are not considered by setting their values to zero (i.e. $S_{ij,t} = A_{ij,t} = 0$), then ρ collapses into an equation that reflects the average degree to which species limit individuals of their own species relative to heterospecific competitors based on their interaction coefficients (α 's) (Godoy & Levine 2014). Conversely, if multitrophic interactions are considered as present (i.e. $S_{ij,t} = A_{ij,t} = 1$) ψ and ω are the terms controlling changes in average niche differences between a pair of species. For example, two species with a similar set of floral visitors could increase niche overlap by having positive apparent competitive effects of each species on the other (i.e. $\omega_{ij,fv} > 0$). With (ρ) defining niche overlap between a pair of species, their stabilising niche difference is expressed as $1 - \rho$.

As an opposing force to stabilising niche differences, average fitness differences drive competitive dominance, and in the absence of niche differences, determine the competitive superiority between a pair of species. Addressing the modifications done in the annual population model described by eqns (1) and (2) to include the effect of floral visitors and soil conditions, we define average fitness differences between the competitors ($\frac{k_j}{k_i}$) as:

$$\frac{k_j}{k_i} = \frac{\eta_j - 1}{\eta_i - 1} \times \sqrt{\frac{\alpha_{ij} + \psi_{ij,s}S_{i,t} + \omega_{ij,fv}A_{i,t}}{\alpha_{jj} + \psi_{jj,s}S_{j,t} + \omega_{jj,fv}A_{j,t}} \times \frac{\alpha_{ii} + \psi_{ii,s}S_{i,t} + \omega_{ii,fv}A_{i,t}}{\alpha_{ji} + \psi_{ji,s}S_{j,t} + \omega_{ji,fv}A_{j,t}}} \quad (4)$$

and

$$\eta_i = \frac{g_i \lambda_i (1 + \theta_{i,s}S_t + \gamma_{i,fv}A_t)}{1 - (1 - g_i)s_i}$$

When the ratio $\frac{k_j}{k_i} > 1$ this condition indicates that species j has a fitness advantage over species i . Both soil salinity and floral visitors can be seen as equalising mechanisms promoting coexistence because they can reduce fitness differences between a species pair by two contrasted pathways. They can modify the 'demographic ratio' ($\frac{\eta_j - 1}{\eta_i - 1}$) which describes the degree to which species j produces more seeds ($g_j \lambda_j (1 + \theta_{j,s}S_t + \gamma_{j,fv}A_t)$) per seed loss due to death or germination ($1 - (1 - g_j)s_j$) than species i , and they can also modify the 'competitive response ratio' ($\sqrt{\frac{\alpha_{ij} + \psi_{ij,s} + \omega_{ij,fv}}{\alpha_{jj} + \psi_{jj,s} + \omega_{jj,fv}} \times \frac{\alpha_{ii} + \psi_{ii,s} + \omega_{ii,fv}}{\alpha_{ji} + \psi_{ji,s} + \omega_{ji,fv}}}$) which describes the degree to which species j is less sensitive to competition than species i (eqn (4)). Notice that these modifications can produce the opposing effect and promote species' competitive dominance by a combination of high demographic rates and low sensitivity to competition.

Competitors can coexist when niche differences overcome fitness differences, allowing both species to invade (i.e. increase its populations) when rare (Chesson 2012). This condition for mutual invasibility is satisfied when:

$$\rho < \frac{k_j}{k_i} < \frac{1}{\rho} \quad (5)$$

Therefore, coexistence at the neighbourhood scale occurs when niche differences between species pairs overcome their differences in fitness. If not the species with higher fitness exclude the inferior competitor from the neighbourhood. We used this condition to evaluate how strongly soil salinity and floral visitors increase or decrease the likelihood of coexistence between competitors across scales. Specifically, we computed for each species pairs the differences between observed and predicted niche differences needed to determine stable coexistence at the neighbourhood scale according to the observed asymmetry in fitness between species.

Field observations used to parameterise the model

In September 2015, we established nine plots of $8.5 \text{ m} \times 8.5 \text{ m}$ along a $1 \text{ km} \times 200 \text{ m}$ area. Three of these nine plots were located in the upper part of the topographic gradient, three at the middle and the last three at the lower part. Average distance between these three locations was 300 m and average distance between plots within each location was 15 m (minimum distance 10 m). In addition, each plot was divided into 36 subplots of $1 \text{ m} \times 1 \text{ m}$ with aisles of 0.5 m in between to allow access to subplots where measurements were taken (total of 324 subplots) (Appendix S2).

This spatial design was established to parameterise for each focal species the annual plant population model (eqns. (1) and (2)). Specifically, the core of the observations involved measuring per germinant viable seed production as a function of the number and identity of neighbours within a radius of 7.5 cm including individuals of the same species (see analyses below). This radius is a standard distance used in previous studies to measure competitive interactions among annual plant species (Levine & HilleRisLambers 2009; Mayfield & Stouffer 2017), and has been validated to capture the outcome of competition interactions at larger scales (1 m^2) under homogeneous environmental conditions (Godoy & Levine 2014). We measured one individual per subplot for widespread species and several individuals per subplot when species were rare (max. 324 individuals/species). To additionally incorporate the effect of soil salinity, from November 2015 to June 2016, we measured soil humidity (%) and soil salinity (dS/m) bimonthly at the subplot centre with a Time Domain Reflectometer (TDR) incorporating a 5-cm probe specially designed and calibrated for these sodic saline soils (EasyTest, Poland). We summarised the amount of soil salinity experienced by each germinant, which was highly correlated with soil moisture ($r = 0.77$), as the sum over their lifetime of the soil salinity measured at the subplot scale.

Moreover, floral visitors were measured during the complete phenological period of all species (from January to June 2016). We surveyed weekly the number of floral visitors for all species within each subplot. Visits were only considered when the floral visitor touched the reproductive organs of the plant, and insects were collected with a hand net. All subplots within a plot were simultaneously surveyed during 30 min each week. Plot survey was randomised between weeks to avoid sampling effects. Overall, this procedure rendered

approximately 90 h of overall floral visitors sampling. Floral visitors to each species and subplot were grouped in four main taxonomic groups (bees, beetles, butterflies and flies). We summarised the number of floral visits by insects to each germinant as the total sum of visits at the subplot scale.

Finally, we quantified the germination of viable seeds (g_i) by counting the number of germinants in 18 quadrats of $1 \text{ m} \times 1 \text{ m}$ placed close to the plots (2 quadrats per plot) from seeds collected the previous year and sown on the ground prior to the first major storm event after summer (September 2015). Similarly, we quantified seed bank survival (s_i) with the same seed material by burying seeds from September 2015 to September 2016 following the methods of (Godoy & Levine 2014).

ANALYSIS

To determine the role of soil salinity separated from the role of floral visitors in plant fecundity (eqn. 2), we used maximum likelihood methods to fit empirical observations in the following way. We fit changes in λ_i and α_{ij} (both bounded to be positive) as a function of the total number of floral visitors (A_i) and the accumulated soil salinity (S_i) experienced by each germinant at the subplot scale over their lifetime (optim method = 'L-BFGS-B'). The effect of floral visitors on fecundity was calculated by either considering all visits together or separating visits from each of the four taxonomic groups. Soil salinity ($\theta_{i,s}$, $\psi_{ij,s}$) and floral visitors ($\gamma_{i,fv}$, $\omega_{ij,fv}$) parameters were not bounded to any specific range as we hypothesised that they can have both positive and negative effects on per germinant fecundities. We further distinguished using AIC (Akaike Information Criterion) whether soil salinity and floral visitor effects on competition were specific to each pairwise interaction (model 1), common to all interactions (model 2), or had no effect (model 3) (Appendix S4 for details). For all three models, individuals of the other 10 species surveyed apart from our six focal species were separated into two groups depending on their degree of relatedness, either confamilial or heterofamilial, and their competitive effects on the focal species were summarised as independent parameters. Estimates of mean and standard error for each parameter of the best model selected by AIC across species are included in Appendix S3. Note that model estimations of λ_i values were not validated experimentally, so we do not know the species' selfing rates. All analyses were conducted in R (version 3.3.1) (R Core Team 2016).

RESULTS

The six focal species experienced a great variation in soil salinity and the type and number of floral visitors. Along the salinity gradient, *Beta macrocarpa* and *Pulicaria paludosa* grew mainly in high soil salinity concentrations, in contrast, *Melilotus elegans* and *Leontodon maroccanus* grew in relatively low saline soils, while *Chamaemelum fuscatum* and *Melilotus sulcatus* showed a more tolerant behaviour growing in a wider range of salt concentrations (Fig. 1). Number of floral visits by insects also varied greatly among plant species. Overall, the main groups of floral visitors in our system were flies (581 visits) and beetles (496 visits), followed by bees (161 visits)

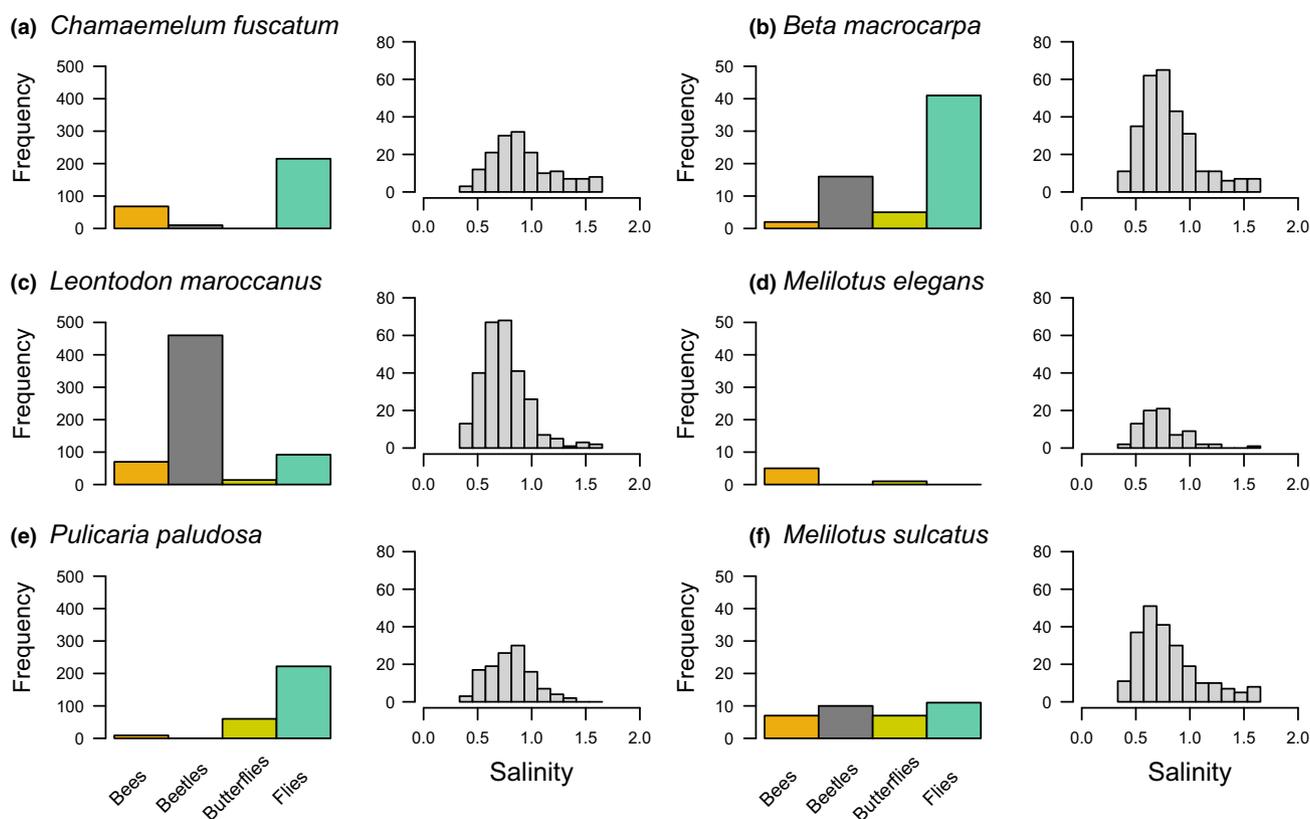


Figure 1 For the six focal species this figure shows: total number of visits of the four groups of floral visitors (bees, beetles, butterflies and flies) (left panel) and species abundance along the salinity gradient (right panel). The amount of salinity experience during the life span of each species was measured as the sum of the electric conductivity in Ds/m measured bimonthly. Note that the three Asteraceae species (a) *Chamaemelum fuscatum*, (c) *Leontodon maroccanus* and (e) *Pulicaria paludosa* had an order of magnitude more floral visits than the non-Asteraceae species (b) *Beta macrocarpa*, (d) *Melilotus elegans* and (f) *Melilotus sulcatus*.

and butterflies (87 visits). The three Asteraceae species were the most visited species. Among them, *L. maroccanus* received 636 visits followed by *C. fuscatum* 293 visits, and *P. paludosa* 291 visits. The rest of the species *B. macrocarpa* (64), and the Fabaceae congeners *M. sulcatus* (35) and *M. elegans* (6) had in comparison a much lower number of visits. Moreover, species also showed variation in the assemblage of floral visitors. Of the three plant species with higher number of visits, flies were the most abundant insects visiting *C. fuscatum* and *P. paludosa*, while beetles were the most abundant for *L. maroccanus* (Fig. 1).

The wide variation in soil salinity concentrations and the number of floral visits observed in our study modified the seed production in the absence of neighbours (λ_i) and the strength of the species' responses to competitive interactions (α_{ij}) of the three Asteraceae species plus *M. sulcatus* (model 2, lowest AIC values, Appendix S4), though AIC values did not help to distinguish between models 2 and 3 for *P. paludosa* (AIC < 10). Interestingly, the sign of the floral visitors' effects on λ_i and α_{ij} varied among these species. While higher number of visits to *C. fuscatum* increased its potential fecundity, and reduced the negative effect of both intra and interspecific competition on seed production, the opposite pattern was observed for *L. maroccanus* and *P. paludosa* (Fig. 2). The floral visitor groups that contribute greater to these positive and

negative effects were those that visited each focal plant species more frequently, bees and flies in the case of *C. fuscatum*, beetles in the case of *L. maroccanus* and flies in the case of *P. paludosa* (Appendix S5 and S6). Soil salinity, in contrast, had a similar effect across species increasing seed production in the absence of neighbours and promoting weaker competitive interactions. For the other two non-Asteraceae species, AIC values suggest that soil salinity and floral visitors did not have a strong effect on λ_i and α_{ij} . In neither case, did model selection support the view that floral visitors and soil salinity separately modified each pairwise competitive interaction (i.e. model 1 showed consistently higher AIC values) (Appendix S4).

Soil salinity and floral visitors exerted positive, negative or no effect on plant fecundity, yet they modified the determinants of competitive outcomes in opposite and specific directions (Fig. 3). While floral visitors tend to maintain stable coexistence at the neighbourhood scale (3 out of 15 species pairs), or to promote coexistence by equalising fitness differences (8 out of 15 pairs moved from the exclusion region into, or closer to, the coexistence region), soil salinity tended to promote competitive exclusion by increasing competitive asymmetries between species pairs (4 pairs moved out of the coexistence region) (Fig. 3). As a result, floral visitors significantly reduced the niche differences needed for coexistence at

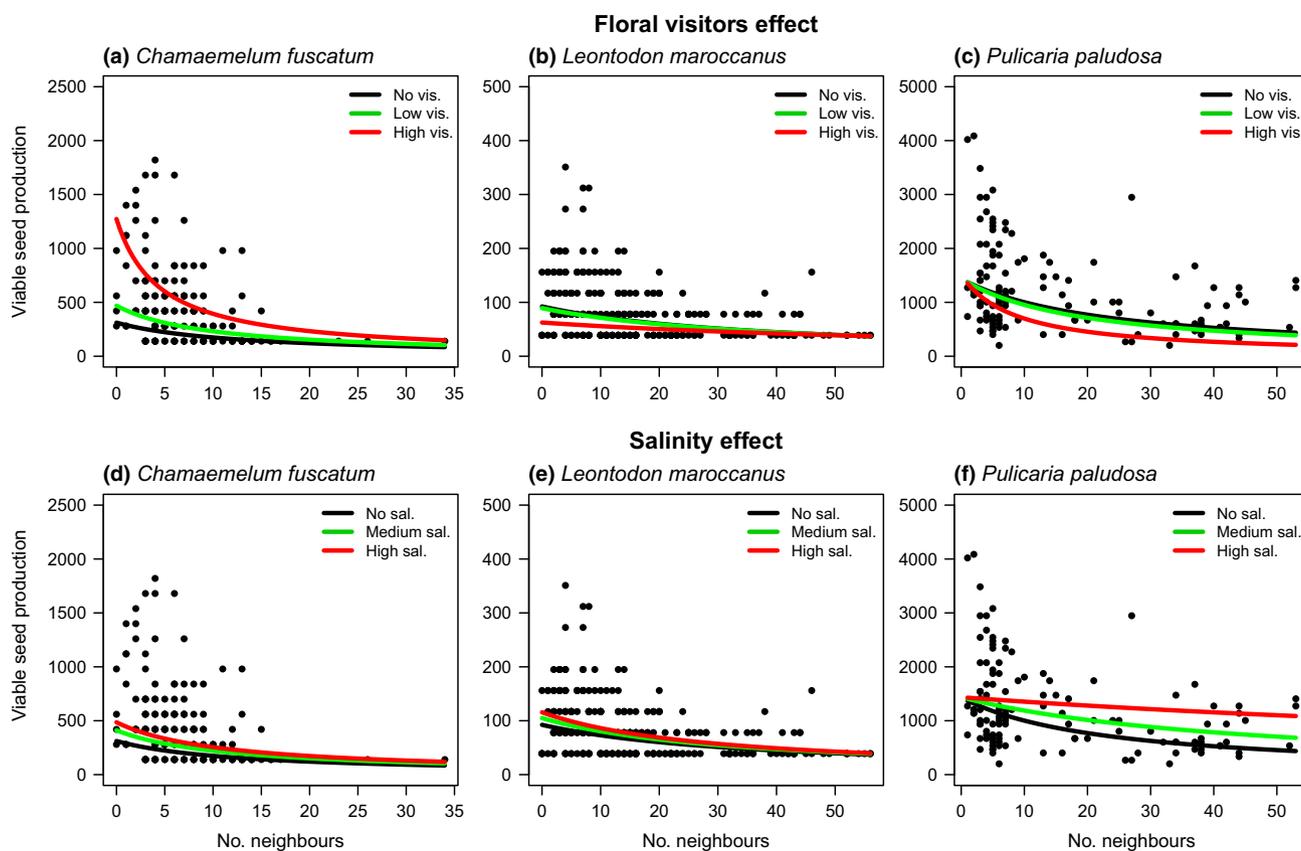


Figure 2 Relationship between per capita seed production as a function of the number of competitor within the neighbourhood according to three different conditions of floral visitors and soil salinity. Here, is represented the three focal Asteraceae species ((a and d) *Chamaemelum fuscatum*, (b and e) *Leontodon maroccanus* and (c and f) *Pulicaria paludosa*). Upper panels contains floral visitors' effects with black curves representing no floral visitation, green curves representing one or two visits, and red curves representing percentile 95 of floral visits (which ranges from six visits in *C. fuscatum* to nine visits in *L. maroccanus* and *P. paludosa*). Lower panels contain soil salinity effects with black curves considering no salt in the soil, and green and red curves representing percentiles 50 and 95, respectively, of the soil salinity sum over focal species life span. Here, we grouped both conspecific and heterospecific competitive interactions for visual purposes, but such interactions are represented separately in Appendix S6.

the neighbourhood scale (estimated from the mutual invasibility, eqn. 5) across species pairs (paired t -test, $t = 2.45$, $P = 0.046$) (for separated effects of each floral visitor guild, see Appendix S7), while soil salinity increased on average the niche differences needed for coexistence (paired t -test, $t = 5.72$, $P < 0.001$).

When comparing how strongly soil salinity and floral visitors modify the determinants of competitive outcomes, we observed that they do not have equivalent potential to limit or promote diversity at neighbourhood scales. The effect of soil salinity on increasing competitive asymmetries between species overwhelmed in most cases the equalising effect of floral visitors (Fig. 4). Nevertheless, the identity of competitive winners (estimated from eqn. 4) changed in one third of the species pairs (5 out of 15). Overall these results suggest that plant diversity in our system is primarily maintained by the effect of soil salinity on changing the dominant competitor across contrasting soil conditions.

DISCUSSION

Recent work has increased awareness among ecologists of the equivalent potential of soil conditions and aboveground

multitrophic interactions on promoting or impeding diversity maintenance (Chase *et al.* 2002; Chesson & Kuang 2008). However, empirical tests of this prediction have remained elusive due to the difficulties in connecting theory with detailed field observations. Our ability to combine coexistence theory advances with plant population models and spatially explicit observations provides direct evidence that variation in soil salinity content and floral visitor frequency modify the likelihood of plant coexistence, yet they do so in opposite directions. While floral visitors' effects on plant fecundity promoted plant diversity maintenance at the neighbourhood scale, soil salinity effects drove competitive exclusion. These modifications occurred via direct changes in per capita seed production and indirect changes in competitive responses. Nevertheless, the strength of these modifications differed between these two drivers. Variation in soil salinity overrode in many species pairs the effect of variation in floral visitors on promoting plant coexistence at the neighbourhood scale but it changed the identity of the superior competitor across contrasted saline conditions (Figs. 3 and 4). Overall, these results show that both soil conditions and aboveground multitrophic interactions are important for maintaining plant diversity, but suggest that, at least in our system, soil conditions

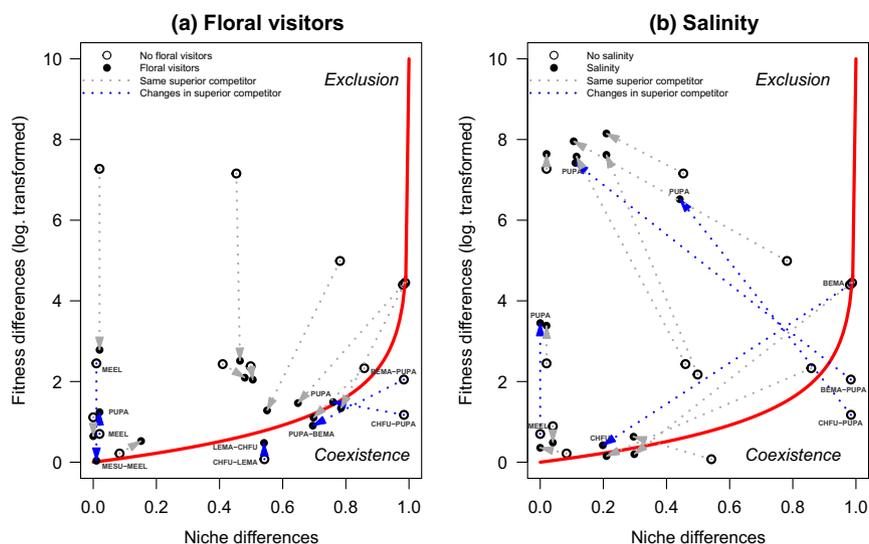


Figure 3 Average fitness and stabilising niche differences for each pair of species (denoted by a single point). Dashed arrows connect the scenario from not considering the effect of floral visitors (a) or soil salinity (b) on the determinants of competition outcomes (black open points) to the scenario in which each driver is considered separately (black solid points). Dashed lines are blue when the identity of the superior competitor changes across both scenarios. We used the condition for mutual invasibility (eqn. 5) to know the identity of the superior competitor. Such identity is written close to both ends of the lines (see Table 1 for species codes). For those cases where both species of the pair are predicted to coexist, the superior competitor is listed first. In case such identity does not change across scenarios then dashed lines are black. The red curve separates the exclusion region from the region where the condition for coexistence is met ($\rho < \frac{k_i}{k_j}$, where species j is the fitness superior)

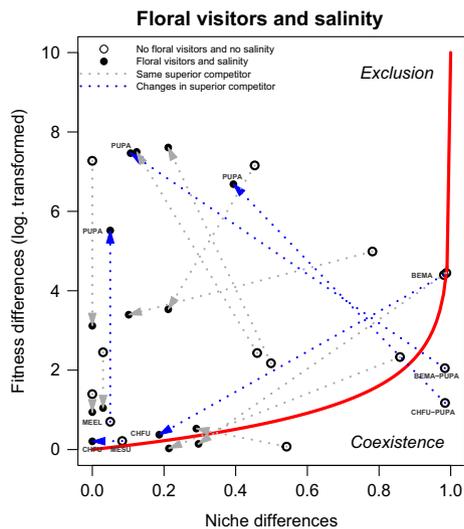


Figure 4 As in Fig. 3, average fitness and stabilising niche differences are represented for each pair of species (denoted by a single point). Dashed arrows connect the scenario from not considering the effects of floral visitors and soil salinity on the determinants of competition outcomes (black open points) to the scenario in which both drivers are jointly considered (black solid points). Dashed lines are blue when the identity of the superior competitor changes across both scenarios. We used the condition for mutual invasibility (eqn. 5) to know the identity of the superior competitor. Such identity is written close to both ends of the lines (see Table 1 for species codes). For those cases where both species of the pair are predicted to coexist, the superior competitor is listed first. In case such identity does not change across scenarios then dashed lines are black. The red curve separates the exclusion region from the region where the condition for coexistence is met ($\rho < \frac{k_i}{k_j}$, where species j is the fitness superior).

were more determinant at neighbourhood scales, and that regional diversity is primarily maintained by spatial changes in soil salinity conditions occurring over larger scales.

Floral visitors consistently promoted species coexistence at the neighbourhood scale by reducing the niche differences needed to overcome fitness differences between species pairs (Fig. 3a). The positive effect of floral visitors on diversity did not only occur due to a positive effect expected from mutualistic interactions. Rather, we observed both positive and negative effects on plant fecundity. For instance, floral visitors strongly increased the seed production in the absence of competition and reduced to a lesser extent the negative effect of competition on the seed production of *C. fuscatum* individuals (Fig. 2a). At the other extreme, floral visitors reduced the fecundity of species such as *L. marocanum* and *P. paludosa* by both reducing seed production in the absence of competition and increasing their sensitivity to competition (Fig. 2b,c). Detailed analyses of the effect of each particular group of floral visitors on plant fecundity showed that the strongest effects on each focal plant species were exerted by their main group of floral visitors (Fig. 1) (Appendix S5, S6 and S7). The most common groups visiting *C. fuscatum* individuals were bee and fly pollinators (mostly Syrphidae species), which are considered mutualistic species. In contrast, beetles visiting primarily *L. marocanum* flowers were pollen feeders belonging to the families Chrysomelidae and Melyridae (Wäckers *et al.* 2007) and the principal visitors of the late-flowering species *P. paludosa* were mainly Bombyliidae species, which are poor pollinators (Polidori *et al.* 2005).

Critically, the equalising effect of floral visitors on plant coexistence likely happened because positive and negative effects were influenced by the species' competitive ability. The

negative effect of floral visitors occurred for those species that were, on average, superior competitors, whereas positive effects occurred for the inferior competitors. This process arises from the fact that our system was dominated by non-specialist interactions and may be a common scenario in this type of system. For instance, beetles acted as herbivores that tend to focus on the most abundant resource, and therefore target the most abundant species (Table 1). Meanwhile, species with high pollinator dependence (i.e. self-incompatible mating system) tend to be subdominant and the ones that benefit substantially from pollinator visits (Tur *et al.* 2013). Although we did not observe that floral visitors increased niche differences between plant species in our system (Fig. 3a), this does not mean that this stabilising effect can occur in more specialised systems. It is reasonable to argue that equalising and stabilising effects occur in combination, as many plant species trade-off between being sufficiently specialised to differentiate in their pollination niche, while being able to attract a sufficient number of mutualistic partners (Vamosi *et al.* 2014; Coux *et al.* 2016).

Conversely to floral visitors, soil salinity promoted competitive exclusion at the neighbourhood scales of species interactions by reducing niche differences while increasing fitness differences among species pairs (Fig. 3b). Nevertheless, the identity of the competitive winner changed across contrasting soil salinity conditions. For instance, *B. macrocarpa* and *L. maroccanus* were competitive winners against *P. paludosa* under low soil salinity concentrations but losers under high soil salinity concentrations. For the particular case of *P. paludosa*, competitive superiority came mostly from the strong positive effect of salinity in reducing its sensitivity to competitive interactions rather than from an increase in the species' ability to produce seeds in the absence of neighbours (Fig. 2f). The consistent effect of soil salinity in determining competitive exclusion across species pairs predicts reduction in species diversity in homogeneous landscapes under constant soil salinity conditions, favouring species that either prefer or refuse salt. But in heterogeneous landscapes like our system, diversity is maintained because of the species' inability to be competitive superiors across all soil salinity conditions. Indeed, these results align with the well-known effect of environmental heterogeneity on promoting diversity (Chesson 2000), and agree also with spatial patterns of species turnover found for very similar salty grasslands in other Mediterranean areas (Pavoine *et al.* 2011). Yet, our results highlight that competitive interactions rather than niche partition (see Allouche *et al.* 2012) is likely the main mechanism driving documented patterns of species turnover.

Our methodological approach is novel in showing how to incorporate the effect of different abiotic and biotic variables into the estimation of niche and fitness differences between species pairs from models that describe species population dynamics via species' vital rates and interaction coefficients. This approach allows experimental testing of the prediction that soil conditions and aboveground multitrophic interactions have equivalent potential for promoting or limiting diversity maintenance. Our methodology is readily available to be extended to consider other soil conditions such as nutrient content or other kinds of interactions beyond the scope of

this study such as herbivores, leaf pathogens and root mutualisms. These soil conditions and interactions could potentially explain changes in fecundity of those species for which soil salinity and floral visitors did not have a significant effect (Landwehr *et al.* 2002; Pan *et al.* 2015), or could be indirectly influencing observed patterns.

Another important step when studying the effect of multitrophic interactions on plant coexistence is to move from direct pairwise effects to include 'higher order effects' among species (Mayfield & Stouffer 2017). Higher order effects occur when the presence of a third competitor changes per capita competitive interactions within a species pair. One main challenge to this is to achieve sufficient sampling size to capture the variability in species composition and multitrophic interactions (Levine *et al.* 2017). Yet, our study was not able to capture this complexity as model selection by AIC highlighted a common effect of floral visitors and soil salinity across species. However, this could be caused because we measured interactions in a relatively dry year and the abundance of some floral visitor groups such as bees and butterflies were relatively low. This last point also makes us aware that climatic variability across years is another layer of complexity that we do not include in our study. Variation between years in the amount of rainfall can change the spatial configuration of soil salinity conditions. Also it can change the abundance, the strength and the specificity of the effect of floral visitors on plant fecundity.

Together, our study shows that soil conditions and multitrophic interactions represented by floral visitors have contrasting outcomes in determining coexistence at the neighbourhood scale of plant species interactions. While variation in soil salinity promotes competitive exclusion, variation in floral visitors promotes coexistence. These differences were mostly explained by equalising processes rather than by stabilising processes. Nevertheless, soil salinity variation was the primary driver of plant diversity in our systems and promoted plant coexistence over larger scales by changing the identity of the competitive winner under contrasting soil salinity conditions. Our results highlight that the spatial structure of soil conditions and multitrophic interactions needs to be considered explicitly when evaluating their effects on maintaining species diversity.

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AUTHORSHIP

IB and OG designed the study. JBL and OG conducted field-work. All authors analysed the results, and JBL and OG wrote the manuscript with substantial contributions from IB

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