The outcome of shared pollination services is affected by the density and spatial pattern of an attractive neighbour

Merav Seifan[1]*, Eva-Maria Hoch[2], Sven Hanoteaux[2] and Katja Tielbörger[2]

[1]*Mitrani Department of Desert Ecology, Swiss Institute for Dryland Environmental and Energy Research, Jacob Blaustein Institutes for Desert Research, Ben-Gurion University of the Negev, 84900 Midreshet Ben-Gurion, Israel; and* [2]*Department of Ecology and Evolution, Plant Ecology Group, University of Tübingen, Auf der Morgenstelle 3, 72076 Tübingen, Germany*

Summary

1. Interactions among neighbouring plants are often mediated by foraging choices of pollinators. For example, the presence of a conspicuous species may increase the number of pollinators attracted to its vicinity, indirectly increasing visitation rates also to neighbouring plants. Because pollinator choices are frequently density-dependent, the presence of a conspicuous species at high densities may also increase competition for pollination services. Additionally, models predict that plant density will interact with spatial distribution in manipulating the pollinator behaviour, yet experimental evidence for this effect is missing.

2. We performed a field experiment in which we introduced a highly conspicuous species in different densities and spatial configurations in a full-factorial manner into a species-rich meadow and studied its effect on neighbouring plants.

3. We showed that the highly conspicuous species strongly contributed to the attractiveness of its local patch and thus benefited its neighbours. However, because of the strong density effect, the conspicuous species changed its role and became a competitor for pollinators when its density increased.

4. We supported our theoretical assumptions and showed that when the introduced conspicuous species was regularly distributed among other plants in the patch, it increased visitation rate, and in some cases also seed set, to conspicuous neighbours relative to when it was aggregated, at least at low densities.

5. *Synthesis.* We suggest that complex interactions between density and spatial distribution of plant species at the patch scale are highly relevant for the interpretation of pollinator behaviour and therefore should be treated as factors of floral attractiveness in future studies.

Key-words: competition, density dependence, facilitation, indirect interactions, magnet species, plant–plant interactions, plant–pollinator interactions, reproductive success, spatial distribution

Introduction

Plant–pollinator interactions are central to the survival of many plant species (Ollerton, Winfree & Tarrant 2011), but our understanding of the processes that govern pollinator activity at the community and patch scale is limited, particularly in relation to plant species distributions (Hegland & Boeke 2006). Nevertheless, it is accepted that pollinator choices at the patch scale are associated with two aspects of the plant community (Feinsinger 1987). The first is the flower density of the patch, which is often positively correlated with increase in pollinator visitation rate (e.g. Ghazoul 2005; Hegland & Boeke 2006; Dauber *et al.* 2010), probably due to an increase in patch attractiveness (e.g. Sih & Baltus 1987) and a minimization of foraging costs (Hegland & Boeke 2006). The second aspect is the patch species composition. The decision of a particular pollinator to visit a flower is a complex outcome of its innate preferences and its current learning experience (Lazaro, Lundgren & Totland 2009; Hegland & Totland 2012). Therefore, the available flower resources and the variation in the flowers' reward properties in a specific patch will play a major role in the choice process of the pollinators (Clegg & Durbin 2000; Klinkhamer, de Jong & Linnebank 2001).

*Correspondence author. E-mail: seifan@bgu.ac.il

The outcome of pollinator choices strongly affects the plant reproductive success in a particular patch. One of the well-known examples for a potential positive influence of a patch composition on the reproductive success of the species growing in it is the magnet species effect. Here, a rewarding, highly attractive species increases pollination success of less attractive species growing in its vicinity by locally increasing pollinator abundance. The extent of the positive effect depends on a close spatial association between the species (e.g. Laverty 1992; Johnson *et al.* 2003; Molina-Montenegro, Badano & Cavieres 2008). On the other hand, several studies have shown that the proximity of highly attractive species creates a competitive disadvantage for the less attractive species. Two mechanisms were shown to create this disadvantage. First, the attention of pollinators may be directed to the more attractive species, causing them to avoid other flowers in the patch (e.g. Yang, Ferrari & Shea 2011). Secondly, if the pollinators can chose between several flowers in a patch, this may increase heterospecific pollen transfer (e.g. Brown, Mitchell & Graham 2002; Cariveau & Norton 2009).

Recently, Hanoteaux, Tielbörger and Seifan (2013) suggested that the spatial arrangement of the species within a patch may be considered as a third relevant aspect for the pollinator's choice process. In their model, the spatial arrangement of two species, which varied in their attractiveness for pollinators, interacted with the species' relative density in governing the population persistence of the less attractive species. In particular, a less attractive species had higher pollination success at high densities when segregated from the attractive species, while at low densities, its pollination success was higher when the two species were uniformly distributed. Overall, the model suggests that changes in the spatial pattern of plant species may considerably affect the way pollinators perceive their pollination landscape (sensu Hanoteaux, Tielbörger & Seifan 2013). This may modify foraging behaviour and hence plant reproductive success, by both changing the probability of a certain plant patch to be approached by a potential pollinator from a distance and the way in which the pollinator then forages on individual flowers within the patch (Sieber *et al.* 2011; Yang, Ferrari & Shea 2011).

To date, only limited attempts have been made to experimentally test the effects of the three factors on pollinator choices (but see Caruso 2002; Feldman 2008; Yang, Ferrari & Shea 2011). To fill this knowledge gap, and to study the potential effects of the three factors together, we experimentally manipulated the presence of a highly conspicuous plant species within a species-rich meadow, and tested the response of pollinators to the patch's flower density, flower composition and spatial arrangement. By working within a natural meadow, we were able not only to test the combined effect of the three factors, but also to maintain a more realistic experiment, in which more than two plant species interact with each other and affect the pollinator's foraging decisions.

Pollinator response was considered as a two-step process, in which the pollinators first selected a particular patch in the meadow and then choose among flower species within the patch. Namely, we first studied whether the flower density and the composition of the patch (i.e. the addition of a highly conspicuous species) affected pollinator patch choices. Secondly, we studied whether the visit choices within a patch were affected by the density and spatial arrangement of a highly conspicuous species. We expected (i) the addition of a conspicuous species to increase the number of visits to a plant patch. Within a patch, we predicted that (ii) neighbouring species will be differently affected by the experimental manipulations. Specifically, the presence of a conspicuous species will have a stronger effect on neighbouring plants that are considered attractive by the pollinators (see also Gibson, Richardson & Pauw 2012). Furthermore, we predicted that (iii) an aggregated distribution of the conspicuous species will increase the number of visits to the neighbouring plants relative to a uniform distribution. Moreover, this pollinator response will generate a positive effect on the reproductive success of neighbours (in terms of visits and seed production). (iv) Finally we assumed that different pollinator groups will respond differently to the experimental manipulations.

Material and methods

STUDY SITE

The experiment was conducted in a semi-natural meadow in southwestern Germany (48°26′26.9″N, 8°55′20.3″E). The meadow, lying at an altitude of 450 m above sea level, is part of the lowlands of the Swabian Alb mountain range. The region is interspersed by a mixture of small settlements and towns, forests, agriculture fields and nature reserves characterized by juniper heathlands and semi-dry pastures. The meadow is considered productive relative to the region (Seifan *et al.* 2010) with plant height reaching approximately 140 cm at the peak of the vegetation period in mid-June.

STUDY SPECIES

Experimentally manipulated species

As a potentially conspicuous species for our manipulations we chose *Centaurea cyanus* L. (Asteraceae). This species is a widely spread annual in the study region, although absent from the particular meadow in which the experiment was conducted. The species, which is self-incompatible, has inflorescences appearing blue to human eyes. Seeds of *C. cyanus* were sown in a glasshouse in March 2011 and transferred as young seedlings into 15 × 15 cm pots. The pots were placed in a common garden and grown until the development of flowers in May 2011, when they were transferred to the field.

Neighbouring species

We selected neighbouring species among the common species growing naturally in the study site (see Tables S1 and S2 in Supporting Information). The criteria for the selected species were an overlapping flowering time with *C. cyanus* and a relatively even distribution among the experimental plots. The chosen species were divided into three groups. a. A congeneric species – *Centaurea jacea* L. (Asteraceae). b. Unrelated species with a relatively large floral display – *Knautia arvensis* (L) Coult. (Dipsacaceae), *Salvia pratensis* L. (Lamiaceae) and *Ranunculus acris* L. (Ranunculaceae). These species have

similar inflorescence height (ca. 60–100 cm) and display size as *C. cyanus* and *C. jacea*. Therefore, these species were regarded as conspicuous species whose attractiveness may be comparable to that of the experimentally manipulated species *C. cyanus*. c. other common species in the plots – *Trifolium pratense* L. (Fabaceae), *Lotus corniculatus* L. (Fabaceae) and *Achillea millefolium* L. (Asteraceae). These species have a relatively large inflorescence, but their flowering heights lay below the above-mentioned species. Therefore, they are regarded in the following analysis as 'less conspicuous species'.

EXPERIMENTAL DESIGN

At the beginning of May 2011, 15 plots of 2 × 2 m were defined within a designated area of approximately 25 × 25 m at the study site. The plots were located so that their vegetation was as homogeneous as possible but the distance between them was larger than one metre. This distance was sufficient to exclude the possibility that flower density of the plot was biased by spatial autocorrelation (see Fig. S1). Each of the plots was assigned randomly to one of five treatments created by the arrangement of the *C. cyanus* pots: (i) regularly spaced, low-density treatment, created by arranging 10 pots in three rows separated by approximately 50 cm from each other; (ii) regularly spaced, high-density treatment, created by arranging 28 pots in five rows separated by approximately 30 cm from each other; (iii) aggregated, low-density treatment, created by arranging 10 pots in three groups, at least 50 cm apart from each other; (iv) aggregated high-density treatment, created by arranging 28 pots in three groups at least 40 cm apart from each other; (v) control, where no *C. cyanus* pots were added. Each treatment had three replicates. The number of pots was chosen to accommodate relatively high and low natural densities of flowering plants after estimating the average plant and inflorescence density in the meadow. All flower heads between the plots were removed twice a week during the observation period to ensure that foraging was not affected by natural variation in flower density around the experimental plots (see below).

FIELD OBSERVATIONS

Pollinator activity in each of the experimental plots was observed for several 5 min rounds in a random order. The observation rounds were repeated three times per observation day between 09:00 and 17:00. The three sessions of 5 min per plot were summed up to create a 15-min observation session that represented the pollinator activity per plot and day. Observations were conducted only on days with air temperatures above 15°C, with no precipitation and a minimal wind speed, limiting the valid observations to 9 days during the season.

For each plot, we recorded the identity of each insect visitor and of each flower visited (the above-mentioned plant species and 'others' representing landing on other species in a plot). A visit was defined valid when an insect contacted the reproductive organs of a flower and stayed in the position for a short while (i.e. events in which an insect only inspected a flower but did not come in clear physical contact with it were ignored; see also Lazaro & Totland 2010). Because of the nature of the observations and the conservation status of most pollinator groups in the region, a complete identification of the visitors was not possible. Instead, we assigned them to the following pollinator groups (see also Hegland & Boeke 2006; Hegland & Totland 2012): the honeybee, *Apis mellifera* (hereafter called *Apis*), bumblebees, *Bombus* species (here after *Bombus*), hoverfly species (Syrphidae); fly species which were not hoverflies (flies); and butterflies (Lepidoptera such as Nymphalidae and Lycaenidae). In our original data set, we also

collected information about other hymenopteran species (mainly solitary bees, e.g. Andrenidae, Halictidae, Colletidae) and Coleopteran species (e.g. Scarabeidae, Miridae), but the low number of visits recorded did not allow for separate analyses of these groups.

Because we assumed that plant density played a significant role in determining plant–pollinator interactions, all inflorescences in all plots were counted before each observation day. Because different inflorescences have different morphology and size, we weighted the number of inflorescence by an estimation of their relative cover area in the plot. To do so, we recorded the size of 20 inflorescences per plant species and calculated the area of its two-dimensional shape (Tables S1 and S2; see also Hegland & Boeke 2006). Because visitation rate is assumed to affect plant reproductive success, but the connection between the two components is not necessarily linear (e.g. Wirth *et al.* 2011), we also assessed the studied species' seed set by randomly tagging three individuals of each species in each plot and collecting all the ripe inflorescences of these individuals. The number of fertilized seeds in each of the flower heads was counted (i.e. seeds which did not contain a living embryo were disregarded; see also Yang, Ferrari & Shea 2011). For each studied species in a plot, the average number of viable seeds per inflorescence was calculated.

STATISTICAL ANALYSIS

The aim of our study was to test for the contribution of plant density and species presence to the various levels of pollinator foraging activities when approaching a plant patch. Therefore, we first checked whether our results could be biased by a spatial autocorrelation. To test whether the overall plant species distribution in the field may have affected pollinator choices, we calculated correlograms based on Moran's I for the total number of visits to each plot in each of the observation days (Dormann *et al.* 2007). After testing for overall plant distribution effects, we used a generalized linear mixed model (GLMM) to test whether total plant density and the density of *C. cyanus* affected the overall number of pollinators attracted to a plot (represented by the 15-min observations per day). Due to the discrete nature of the observation data, we used a model with a Poisson distribution and a log link. Because each manipulation of *C. cyanus* was replicated in three plots in the field, we used plots as a random factor.

To study the potential effect of the density of *C. cyanus* and its spatial arrangement on the particular visitation choices within a plot, we used a set of GLMMs with a Poisson distribution and a log link. Our main goal was to analyse how the number of pollinator visits to each of the studied species was affected by the density of *C. cyanus*, its spatial pattern and the interaction between them. Because we assumed that flower density has a strong effect on pollinator foraging behaviour, we followed the suggestion of Hegland & Boeke (2006) and added as covariates the total density of the flowers and the focal species density per plot. The plot was used as a random factor. For seed set analysis of the same species, we used a generalized linear model (GLM, with a normal distribution and a log link) in which the density and spatial pattern of *C. cyanus* and their interaction were the main factors and the specific plant species density and the plot total density were used as covariates.

For our third goal, that is studying whether different pollinator groups showed different plant choices within a plot, we used a set of GLMMs in which the number of visits of a specific pollinator group to each of the studied species was analysed using the same explanatory factors as described above. In most cases, a Poisson distribution with a log link was used. In few cases, we used a negative binomial

distribution with a log link to prevent overdispersion (see results). The spatial autocorrelation analysis was conducted with R (R Core Development Team 2012) with the package ncf (Bjornstad 2013). The GLMM and GLM analyses were conducted with SPSS (Version 21.0, IBM, Armonk, NY, USA).

Results

Overall, 272027 visits from potential pollinators were observed on nine plant species (the above-mentioned focal species plus all 'others') in 27 observation rounds.

CHOICES AMONG PLOTS

Overall, the number of pollinator visits in each of the observation dates was not significantly affected by the location of the plot within the field (Fig. S2). However, the number of pollinators visiting a plot was positively affected by the total flower density ($F_{1,132} = 277.72$; $P < 0.001$; Fig. 1a) and the density of *C. cyanus* ($F_{1,132} = 42.23$; $P < 0.001$; Fig. 1b) in the plot.

CHOICES WITHIN PLOTS

The number of pollinator visits to the manipulated species *C. cyanus* was positively affected by its density (Table 1; Fig. 2a) regardless of its spatial distribution. In addition, the interaction between density and spatial pattern was significant, showing a faster positive response to density when *C. cyanus* was aggregated. Although not significant, there was a tendency for more visits and higher seed set in plots in which *C. cyanus* was aggregated (Table S4 and Fig. S3).

Effect on neighbouring species

While the number of visits to each of the neighbouring species had a different response to the total plot density, their response to their intraspecific density and to the density of the manipulated species *C. cyanus* was similar (Table 1). In all observed neighbouring species, except *R. acris*, the number of pollinator visits was positively associated with the species' own density (Fig. 3), and in all cases except for *T. pratense* and *L. corniculatus*, the number of visits was negatively associated with *C. cyanus* density.

The interaction between *C. cyanus* density and its spatial pattern showed a different effect on the visits to the conspicuous and less conspicuous neighbours (Table 1). The effect on the conspicuous species group (including the congeneric species) was twofold. First, although generally *C. cyanus* density decreased pollination visits to the neighbouring conspicuous species, the number of pollinators visiting these neighbours was higher at low *C. cyanus* density than in the control plots, where no *C. cyanus* was present (Fig. 2b,c; S4). Secondly, the number of pollinator visits to conspicuous neighbours, at least at low densities of *C. cyanus*, was higher when the manipulated species was regularly distributed relative to when it was aggregated. The response of the less conspicuous neighbouring species was less unified, but in all three cases the response to aggregated *C. cyanus* was more positive than when it was regularly distributed, at least at low densities (Fig. 2d; S4).

The effects of *C. cyanus* density and its spatial pattern on the seed production of the neighbouring species were relatively weak and showed only significant associations with intraspecific densities (see Table S4). However, although not significant, the response of each of the neighbour groups to *C. cyanus* spatial distribution was detected also in the neighbours' seed production (Fig. S3).

POLLINATOR GROUP RESPONSES

All pollinator groups responded positively (i.e. increased visitation rate) to increased density of all focal plant species (Table 2). An increased density of *C. cyanus* generally had a negative effect on the number *Apis* visits to neighbouring species (Table 2; Fig. 4). However, in the cases of *C. jacea* and *K. arvensis,* the number of visits in the presence of low densities of *C. cyanus* was significantly higher than in the control plots (Fig. 4b,d). These two neighbouring species also responded significantly to the spatial arrangement of *C. cyanus* spatial pattern, though the effect was not consistent.

Unlike the *Apis* individuals, the number of visits to neighbouring plants made by *Bombus* individuals was generally positively associated with an increase in the density of *C. cyanus* (Table 2). In the case of *C. cyanus* and *K. arvensis, Bombus* individuals responded to *C. cyanus* spatial distribution, visiting
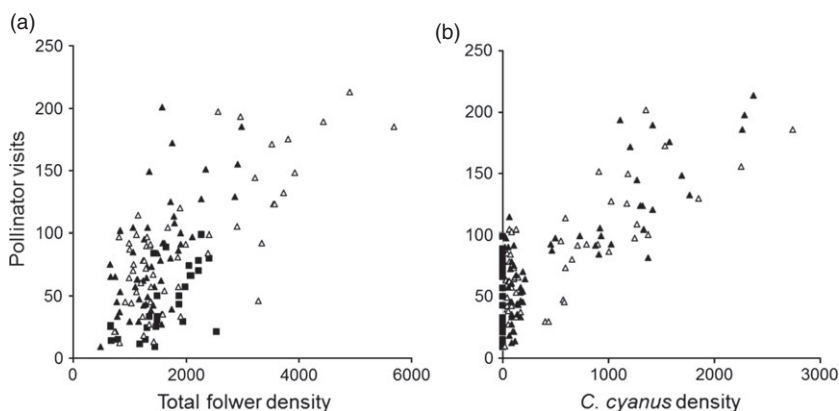
Fig. 1. Total pollinator visits (per 15 min) to plot as a function of the total plant inflorescence density (a) and the density of the manipulated species *Centaurea cyanus* (b). □/■ – plot with regularly spaced low/ high density of *C. cyanus* pots; ○/● – plots with aggregated low/high density of *C. cyanus* pots; ▲ – control plots with no *C. cyanus* plants.

Table 1. Effect of density and spatial pattern on the number of pollinator visits to the studied plant species (per 15 min)

Species	Total	Target	cyanus	Spatial	cyanus × spat
Manipulated					
C. cyanus	53.03↓***		835.80↑***	0.05	61.73***
Congeneric					
C. jacea	16.13↑***	297.36↑***	24.59↓***	4.40*	5.85*
Conspicuous					
K. arvensis	0.01↓	205.71↑***	97.01↓***	0.89	5.45*
S. pratensis	14.22↑***	54.87↑***	0.58↓	0.03	0.07
R. acris	7.72↑**	0.01↓	20.54↓***	3.41*	4.48*
Less conspicuous					
L. corniculatus	1.01↓	7.92↑**	0.88↑	0.09	4.55*
T. pratense	4.47↓*	44.18↑***	7.05↑**	0.26	0.08
A. millefolium	0.32↓	54.02↑***	3.80↓*	1.65	8.38**

The following effects were tested in the models: total – total inflorescence density per plot; target – intraspecific density of the studied species per plot; cyanus – the manipulated species *Centaurea cyanus* density per plot; spatial – the spatial arrangement of *C. cyanus* per plot; cyanus × spat – the interactions between spatial arrangement and *C. cyanus* density. Numbers represent *F* values (with 1, 103 d.f. for *C. cyanus* and 1, 123 d.f. for the other studied species, except spatial pattern, where 2, 103 and 2, 123 d.f. were used, respectively). ↑/↓ – indicate a positive/negative coefficient values (values can be found in Table S3).

*0.01 < *P*-value < 0.05; **0.001 < *P*-value < 0.01; ***P*-value < 0.001.
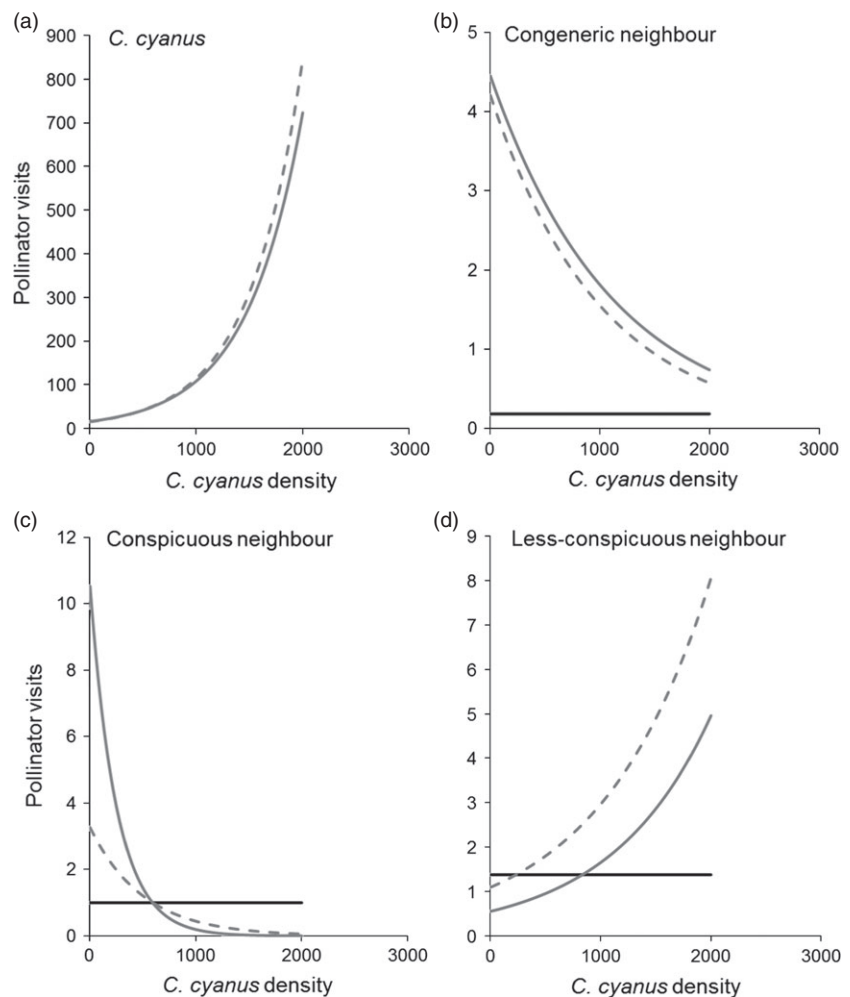
Fig. 2. Examples of the estimated number of pollinator visits (per 15 min) to different plant species as a function of *Centaurea cyanus* density and spatial arrangement in a plot. (a) *C. cyanus*; (b) the congeneric species *C. jacea*; (c) a conspicuous neighbouring species *R. acris*; (d) a less conspicuous neighbouring species *T. pratense*. Grey lines represent the estimated visits when *C. cyanus* was regularly distributed in a plot; dashed lines represent the estimated visits when *C. cyanus* was aggregated in a plot; black lines represent the estimated visits in control plots, with no *C. cyanus* plants. For details on all studied species, see Fig. S4.

the focal species more in plots in which *C. cyanus* was regularly distributed relative to those in which it was aggregated. The response of flies was similar to those of *Bombus*. Generally, there was an increase in the number of visits to neighbouring species with increased density of *C. cyanus*. In addition, flies visiting *K. arvensis* and *A. millefolium* showed a response
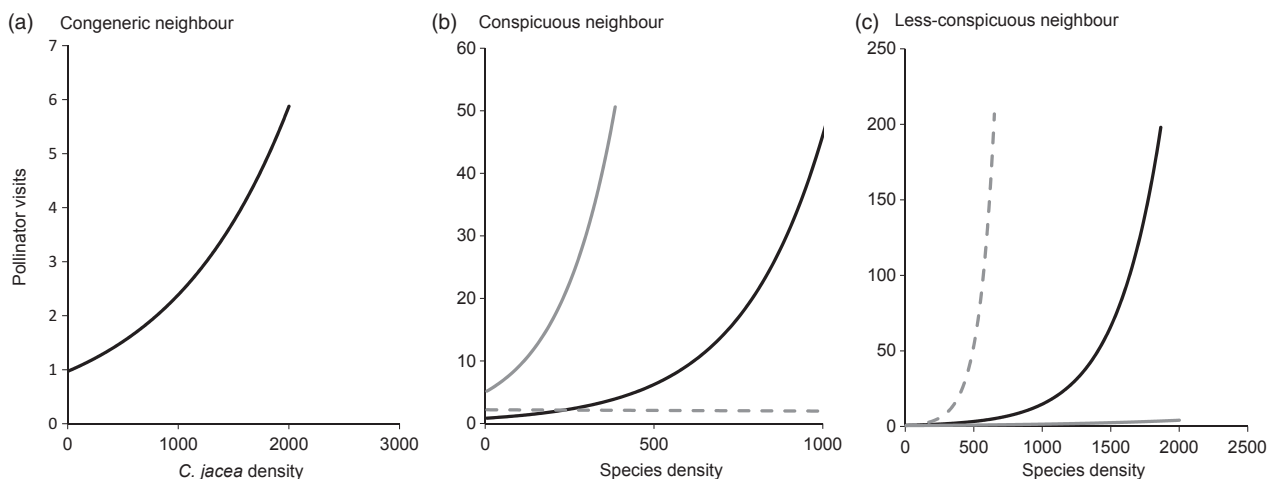
Fig. 3. Estimated number of pollinator visits (per 15 min) to each of the studied species as a function of their intraspecific density in a plot. (a) *Centaurea jacea*; (b) the conspicuous neighbour group: *Salvia pratensis* (black line), *K. arvensis* (grey line), *R. acris* (dashed grey line); (c) the less conspicuous neighbour group: *Trifolium pratense* (black line), *Achillea millefolium* (grey line), *Lotus corniculatus* (dashed grey line).

to *C. cyanus* spatial arrangement, in which at least at low *C. cyanus* densities, aggregated distribution increased the number of fly visits to the two neighbouring species. The hoverfly and butterfly groups did not show any significant trend in their response to the presence of *C. cyanus* or its spatial arrangement (Table 2).

Discussion

Overall, our study supports the assumption that flower density is a crucial factor in the determination of pollinator choices at the patch level (Goulson 1999; Hegland & Boeke 2006). In addition, we showed that the presence of a highly conspicuous species strongly contributed to the attractiveness of the patch and that this increase in attractiveness had an effect also on within patch choices of pollinators. In particular, we demonstrated that the conspicuous species may function as a magnet species to its neighbours, but that this role may change into strong competition for pollinators when density of the conspicuous species is increasing (see also Munoz & Cavieres 2008). Furthermore, the contribution of the conspicuous species to its neighbours shifted predictably not only with density but also with its spatial arrangement.

The results support our assumption that spatial arrangement of flowers within a patch will influence pollinator choices and that this influence is different according to the conspicuousness of the neighbouring plant. This is apparent when looking at the pollinator choices of neighbouring flowers at low densities of the manipulated species: while the more conspicuous neighbours (i.e. the congeneric and conspicuous neighbour groups) received more visits when the manipulated, highly attractive species was regularly distributed, the less conspicuous neighbours received more visits when the highly attractive species was aggregated within the patch. These results may indicate that the two groups of neighbouring flowers benefited from different aspects of the pollinator foraging behaviour. On the one hand, assuming that the pollinators

considered the conspicuous neighbours as relatively attractive (even if less than *C. cyanus*), when the conspicuous species were interspersed with *C. cyanus,* they might have offered a sufficiently attractive alternative to persuade some of the pollinators to switch between flower resource. In accordance with this interpretation, only when *C. cyanus* was aggregated within the plot, did it provide enough closely located flower resources to cause the pollinators to avoid other neighbours. On the other hand, the less conspicuous neighbours probably 'trapped' pollinators in the local plant choices when separated from the aggregated *C. cyanus*, even when more attractive alternatives were available in the patch, as predicted in our recent theoretical study (Hanoteaux, Tielbörger & Seifan 2013). A similar observation was reported in a laboratory experiment by Keasar (2000). She found that less attractive flowers (non-rewarding in her case) were visited more often when separated from the more attractive (i.e. rewarding) ones. This may be because the aggregation of the attractive species reduced its encounter rate in other parts of the plot, which led to a reduction in the pollinators' ability to compare between the attractive and less attractive species, thus increasing visitation rate to other plant species (see also Internicola *et al.* 2007).

The effect of the composition and spatial pattern of plant species on pollinator choices may also shed light on the interactions between native and invasive plants. Invasive species are often regarded as highly attractive because of their relatively large flowers and reward (e.g. Bjerknes *et al.* 2007). Nevertheless, recent experimental studies showed that although visitation rates to plots to which invasive species were introduced usually increased, seed set of native species within those plots was reduced (Totland *et al.* 2006; Bjerknes *et al.* 2007). Such outcomes may be interpreted in the light of our experiment, because highly attractive invasive species probably show opposing effects at different scales of the pollinator foraging process: at a larger scale, the invasive species attract more pollinators to their vicinity, thus increase

Table 2. Effect of density and spatial pattern on the number of visits (per 15 min) to the studied plant species of specific pollinator groups

Effect	Manipulated	Congeneric	Conspicuous		Less conspicuous		
	C. cyanus	C. jacea	K. arvensis	S. pratensis	L. corniculatus	T. pratense	A. millefolium
Apis							
Total	63.5 (1,103)↓***	25.1 (1,83)↑***	4.6 (1,123)↑*	15.2 (1,83)↓*	6.2 (1,123)↑*		
Target	867.3 (1,103)↑***	226.4 (1,83)↑***	195.1 (1,123)↑***	54.6 (1,83)↑***	0.1 (1,123)↑		
cyanus		47.0 (1,83)↓***	163.2 (1,123)↓***	2.3 (1,83)↓	24.5 (1,123)↓***		
Spatial	0.2 (2,103)	5.5 (1,83)**	3.6 (1,123)*	0.1 (1,83)	1.5 (1,123)		
cyanus × spat	77.9 (1,103)***	10.6 (1,83)**	0.1 (1,123)	0.4 (1,83)	0.1 (1,123)		
Bombus							
Total	50.9 (1,103)↓***	7.2 (1,83)↓**	0.3 (1,123)↓		12.0 (1,123)↓**	1.7 (1,123)↑	
Target	81.3 (1,103)↑***	24.3 (1,83)↑***	15.2 (1,123)↑**		1.2 (1,123)↑	1.4 (1,123)↑	
cyanus		14.4 (1,83)↑***	0.8 (1,123)↓		9.0 (1,123)↑**	0.1 (1,123)↑	
Spatial	5.4 (2,103)*	1.6 (1,83)	4.1 (1,123)*		0.3 (1,123)	0.1 (1,123)	
cyanus × spat	6.1 (1,103)*	11.3 (1,83)**	0.4 (1,123)		11.2 (1,123)**	3.2 (1,123)	
Flies							
Total	0.8 (1,103)↓	2.6 (1,83)↓	39.8 (1,123)↓***				0.9 (1,117)↑
Target	7.2 (1,103)↑**	4.3 (1,83)↑*	5.5 (1,123)↑*				33.4 (1,117)↑***
cyanus		6.9 (1,83)↑*	62.4 (1,123)↑***				11.7 (1,117)↓***
Spatial	1.8 (1,103)	1.9 (1,83)	3.8 (1,123)*				0.8 (1,117)*
cyanus × spat	0.5 (1,103)	0.3 (1,83)	5.2 (1,123)*				10.3 (1,117)**
Hoverflies							
Total	0.6 (1,39)↑†	0.5 (1,30)↑	0.5 (1,46)↓				2.8 (1,50)↑†
Target	1.1 (1,39)↑	3.4 (1,30)↑	0.4 (1,46)↑				0.4 (1,50)↑
cyanus		0.2 (1,30)↑	0.1 (1,46)↑				2.6 (1,50)↓
Spatial	0.1 (1,39)	1.4 (1,30)	0.2 (1,46)				0.5 (2,50)
cyanus × spat	1.5 (1,39)	0.1 (1,30)	0.1 (1,46)				0.4 (1,50)
Butterflies							
Total	3.7 (1,43)↓†	2.0 (1,27)↑	0.7 (1,27)↓				
Target	14.7 (1,43)↑***	9.1 (1,27)↑**	9.9 (1,27)↑**				
cyanus		0.2 (1,27)↓	0.4 (1,27)↓				
Spatial	0.1 (2,43)	0.4 (2,27)	1.3 (2,27)				
cyanus × spat	3.7 (1,43)	0.4 (1,27)	0.1 (1,27)				

The following effects were tested in the models: total – total inflorescence density per plot; target – intraspecific density of the studied species per plot; cyanus – the manipulated species *C. cyanus* density per plot; spatial – the spatial arrangement of *C. cyanus* per plot; cyanus × spat – the interactions between spatial arrangement and *C. cyanus* density. Numbers represent *F* values (with the relevant d.f.). ↑/↓ – indicate a positive/negative coefficient values (values can be found in Table S5).
*0.01 < *P*-value < 0.05; **0.001 < *P*-value < 0.01; ***P*-value < 0.001.
†Negative binomial distribution.
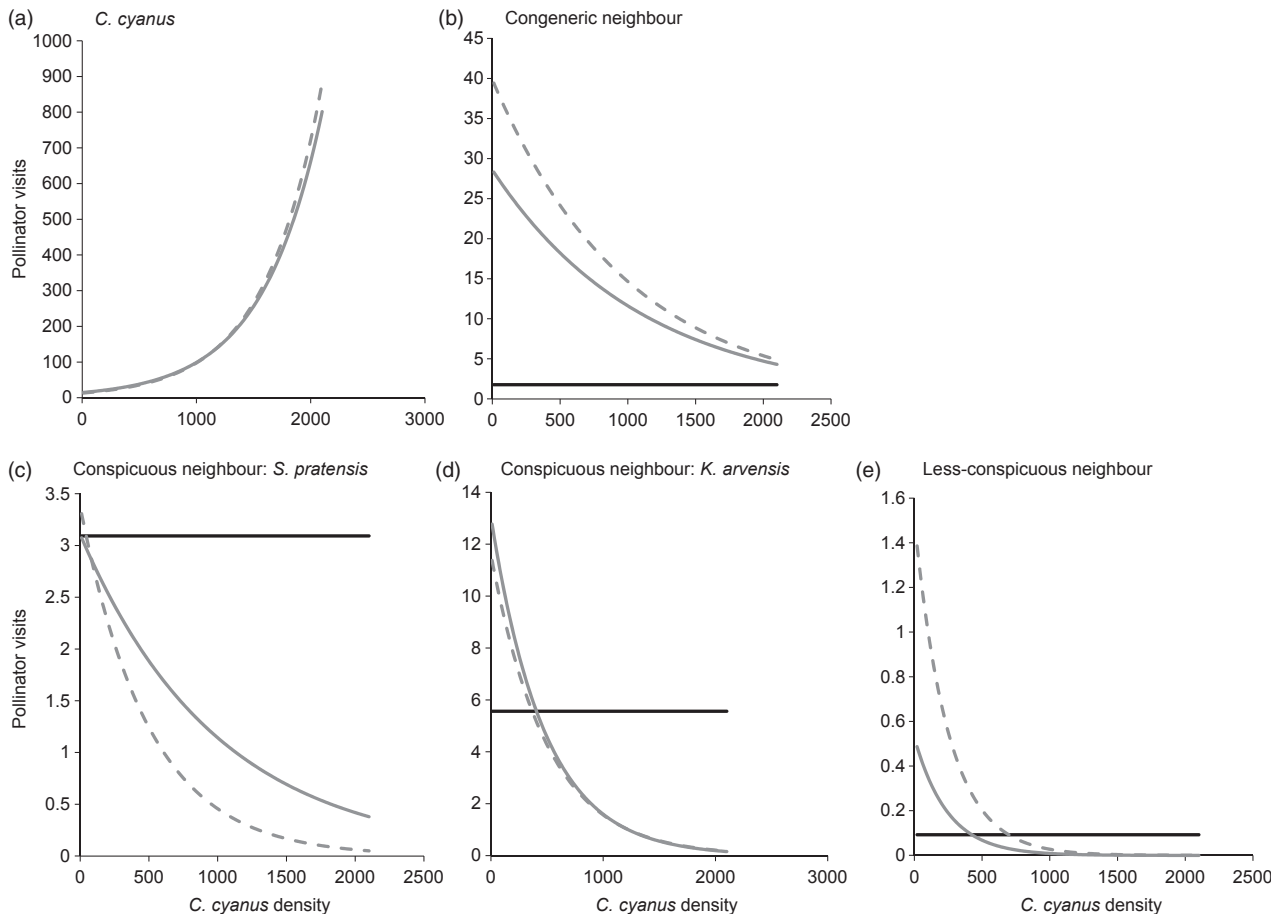
Fig. 4. Estimated number of *Apis mellifera* visits to the studied species as a function of *Centaurea cyanus* density and spatial arrangement in a plot. (a) *C. cyanus*; (b) the congeneric species *C. jacea*; (c and d) the conspicuous neighbouring species *Salvia pratensis* and *K. arvensis*, respectively. (e) the less conspicuous neighbouring species *Lotus corniculatus*. Grey lines represent the estimated visits when *C. cyanus* was regularly distributed in a plot; dashed lines represent the estimated visits when *C. cyanus* was aggregated in a plot; black lines represent the estimated visits in control plots, with no *C. cyanus* plants.

visitation rate to patches in which they are present (e.g. Bjerknes *et al.* 2007). However, at the patch scale, native neighbours may benefit from the additional pollinators only if the manipulated density of the invasive species is low (Munoz & Cavieres 2008; Morales & Traveset 2009) and when there is a spatial segregation between the plant species (Cariveau & Norton 2009).

Interestingly, the effect of the attractive species on shared pollination services in our study cannot be simply categorized according to the floral traits of the species (e.g. morphology, colour). Numerous classical works connected floral trait syndromes with particular pollinator groups (e.g. Goulson 1999; Johnson & Steiner 2000), and especially predicted that more simple floral structures, in which plant rewards are easily collected, will attract more pollinator types (e.g. Johnson & Steiner 2000; Pellissier *et al.* 2010). However, only few studies focused on the potential effect of floral traits on indirect plant–plant interactions (Gibson, Richardson & Pauw 2012). These studies generally assumed that plant species with similar floral traits will share similar pollinator groups (Danieli-Silva *et al.* 2012) and thus will have a higher potential to affect each other's pollination success (Hegland &

Totland 2005). Nevertheless, our study indicated that within patches (unlike at larger scales, e.g. Pellissier *et al.* 2010), most of the neighbouring plants were affected (positively or negatively) by the presence of *C. cyanus*, regardless of their floral traits. This general result suggests that indirect plant–plant interactions were strongly affected by shifts in pollinator choices even if the plant species have different floral traits (e.g. they belong to different 'pollination guilds'). A possible explanation may lie in the major role of floral display size and nectar level in determining flower attractiveness in temperate regions (Hegland & Totland 2005). Unlike our prediction, because the inflorescences of all our studied species had similar sizes and all of the species are known to produce relatively high amount of nectar (BiolFlor data bank, Klotz, Kühn & Durka 2002), their attractiveness level to the pollinators may have been more similar than we assumed.

Conclusions about group-specific pollinator behaviour should be made with caution, because observations on the pollinator group level included many confounded effects. The coarse classification we made may have grouped together species whose foraging activities and energetic needs are different. Nevertheless, the analyses revealed some common

patterns. The most general is that all pollinator groups responded positively to the visited plant intraspecific density, supporting the general notion that plant density is the central factor affecting pollinator foraging behaviour (e.g. Hegland & Boeke 2006).

Among the analyses of the various pollinator groups, the most striking result was the highly negative effect of increased *C. cyanus* density on the number of visits of *Apis* individuals to neighbours. This observation may be explained by the species' fast learning ability, which is combined with highly constant foraging behaviour (see review in Goulson 1999). Because of this combination, the presence of a highly attractive resource (such as the *C. cyanus*) might have caused *Apis* individuals to rapidly switch their preference towards the new resource and avoid other plant species. Nevertheless, the presence of *C. cyanus* at low densities increased *Apis* individual visits also to many neighbouring plants, and the behaviour was often sensitive to changes in the spatial pattern of the plants. Interestingly, *S. pratensis* (Lamiaceae) was the only conspicuous neighbour with enough observations that always responded negatively to the presence of *C. cyanus*. The strong negative response of this species to the presence of another highly attractive species may be a consequence of the clear morphological differences in the shape of the two species and an indication to the lower tendency of honeybees to switch their constancy during a bout (Chittka, Thomson & Waser 1999). *Bombus* and fly groups also showed a general sensitivity to the presence of *C. cyanus* and its spatial pattern, although the direction of their response was sometimes contradictory. This may be explained by the lower fidelity of flies and bumblebees to specific flower species within about (Goulson 1999). Unlike the above-mentioned groups, the hoverfly and butterfly groups did not respond to our manipulations. Although a potential explanation may be that the groups were too heterogeneous for a clear trend, our results fit previous conclusions that these groups are highly generalist and are therefore not expected to show clear flower preferences or to act as effective pollinators (e.g. Branquart & Hemptinne 2000; Danieli-Silva *et al.* 2012). Hoverflies in particular were suggested to be less mobile and to require less energy than bee species, suggesting that they discriminate less among food resources within a patch while foraging (Hegland & Boeke 2006).

Conclusions

Our study created a unique bridge between theoretical explanations concerning pollinators and their effect on the plant community and shared pollination services. We demonstrated that relative density and spatial distribution of highly conspicuous species interact in dictating pollinator choices not only between plant patches but also in their preferences of visited plants within a patch. Namely, as long as the density of the conspicuous species was relatively low and its distribution regular, it induced visits to other relatively conspicuous species in the patch. However, with increasing density, and when its distribution was more aggregated, the effect shifted towards a strong competition for pollinator visits. Less conspicuous neighbours showed a less uniformed response, but often benefitted more from aggregated distribution of the highly conspicuous species. These results may shed light on the process of pollinator foraging and its potential outcome for the visited plants. In particular, our study supports our theoretical prediction that to predict the outcome of plant–pollinator interactions, the specific composition and community structure, including spatial distribution and species identity, should be taken into considerations in addition to the more traditional focus on species density.

Data accessibility

Data available from the Dryad Digital Repository (Seifan *et al.* 2014).

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Figure S1. Spatial autocorrelation of the plots' flower density per date.

Figure S2. Spatial autocorrelation of the number of visits per date.

Figure S3. Seed production of the studied species as a function of *C. cyanus* spatial arrangement.

Figure S4. Pollinator visits as a function of *C. cyanus* density and spatial arrangement.

Table S1. Inflorescence traits of focal species.

Table S2. Average inflorescence size of the remaining plant species in the grassland.

Table S3. Coefficients for the GLMM model in Table 1.

Table S4. GLM analysis of fertilized seed production.

Table S5. Coefficients for the GLMM model in Table 2.