ARTICLE

Indirect interactions between pollinators drive interaction rewiring through space

Ainhoa Magrach[1,2] | Maddi Artamendi[1,3] | Paula Dominguez Lapido[1,4] | Clara Parejo[1] | Encarnacion Rubio[1,5]

[1]Basque Centre for Climate Change-BC3, Leioa, Spain

[2]IKERBASQUE, Basque Foundation for Science, Bilbao, Spain

[3]Universidad del Pais Vasco, Euskal Herriko Unibertsitatea (UPV-EHU), Leioa, Spain

[4]Enebada Entorno, S.L., Santiago de Compostela, Galicia, Spain

[5]Departamento de Biología Vegetal y Ecología, Universidad de Sevilla, Seville, Spain

Correspondence
Ainhoa Magrach
Email: ainhoa.magrach@bc3research.org

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Abstract

In recent years, an extended body of literature has focused on the importance of either temporal or spatial dynamics in shaping the structure of interacting plant and pollinator communities. This improvement from a previously static and aggregated perspective has allowed us to understand many of the ecological processes that shape community assembly. However, fewer are the studies that have simultaneously focused on spatial and temporal dynamics, and even fewer are those that collect data across different habitat types to assess the generality of their findings. Here, we used a dataset collected weekly throughout the full flowering season for two consecutive years and within two contrasting habitat types in N and SW Spain: a mountain grassland area and the understory of sparse pine forests. We evaluated species and interaction persistence through space and time, pollinator fidelity, and turnover patterns in interaction composition while providing a potential mechanistic explanation for the patterns observed. Our results show that although species generalization does not explain species or interaction persistence, moderately generalist species are those showing the greatest fidelity to the subset of plant species they visit through space and time. Further, we find that interaction turnover through time is mostly driven by changes in species composition, while through space it is mostly driven by interaction rewiring resulting from indirect competitive interactions between pollinator species. Our results help to shed light on the potential mechanisms driving community assembly patterns beyond niche or neutral processes by adding within-trophic-level interactions that can modify pollinator preferences.

KEYWORDS
competition, indirect interactions, interaction networks, interaction persistence, interaction rewiring, interaction turnover, plant–pollinator interactions

INTRODUCTION

In recent years, community-level analyses of the interactions between plants and their pollinators have allowed us to gain a significant understanding of how these communities are structured (Bascompte & Jordano, 2013), as well as initial insights into how this structure affects some ecosystem functions (e.g., plant reproductive success, Magrach et al., 2021). However, a major challenge continues to be accounting for and understanding the mechanisms behind the temporal and spatial dynamics that affect interaction composition, community assembly, and overall community structure. Specifically, a major challenge resides in understanding why a large proportion of the interactions between plants and their pollinators are only observed during a given year but not in others. Is it due to the nonpresence of one or both species, or are species present in both years but their interactions not taking place?

In the past decades, research focusing on temporal and spatial dynamics in plant and pollinator communities (Alarcón et al., 2008; Burkle et al., 2013; Burkle & Alarcón, 2011; CaraDonna et al., 2017, 2021; CaraDonna & Waser, 2020; Chacoff et al., 2017; MacLeod et al., 2016; Resasco et al., 2021; Schwarz et al., 2020, 2021; Trøjelsgaard & Olesen, 2016; Vázquez et al., 2009; Vizentin-Bugoni et al., 2014; Weinstein & Graham, 2017) has tried to shed some light on patterns of interaction persistence and turnover. Although research has advanced greatly in the description of patterns, far less efforts have been set on understanding the mechanisms driving these patterns. For instance, we know of the importance of phenology, which leads to temporal variations in species composition and consequently to changes in community structure (CaraDonna et al., 2021). We also know that plant and pollinator species vary in their distribution in space, which leads to spatial changes in the composition of the community and thus to differences in the composition of interactions (Olesen & Jordano, 2002; Vázquez et al., 2009). But changes in species composition are not the only reason why interaction composition can shift. Plant and pollinator species do not necessarily interact in the same way across space or time (Poisot et al., 2015). In some cases, interaction rewiring, or switching of partners, can also occur. Traditionally, these changing interaction patterns have been explained by two processes: (1) a stochastic (neutral) process by which changes in species abundances and hence encounter probabilities influence interaction frequencies and (2) a niche-driven deterministic process by which trait-matching between species (e.g., that related to phenological overlap or morphology) is the main driver explaining differences in interaction frequencies. However, this dichotomy largely ignores the

effect of the presence and abundance of other species in the community, the potential indirect interactions between these co-occurring species, and their effect on interaction frequencies (e.g., through competition for floral resources).

Indeed, competition between pollinators for floral resources has been shown to affect pollinator behavior both under experimental (Brosi & Briggs, 2013) and field conditions (Magrach et al., 2017). This research has shown that the absence (Brosi & Briggs, 2013) or presence (Magrach et al., 2017) of a highly competitive pollinator species modifies the behavior and visitation patterns of other pollinator species in the community (Bain et al., 2022). However, a larger community-level analysis of pollinator competitive interactions and how they influence pollinator preference patterns is currently lacking (Bartomeus et al., 2021).

Importantly, many of the patterns observed in plant–pollinator interaction networks depend highly on the aggregation level used to conduct analyses. For example, CaraDonna and Waser (2020) found that pollinator species considered to be generalist across the cumulative or aggregated network were actually much more specialized when week-to-week dynamics were considered. Although other research has suggested that the macroscopic patterns of these networks are robust to the aggregation level used (Olesen et al., 2011, yet CaraDonna & Waser, 2020 for contrasting results), microscopic patterns, such as those at the species level, have been shown to be hugely scale-dependent (CaraDonna & Waser, 2020), with implications for their use as explanatory variables for species and interaction persistence patterns.

Here, we use data collected within two different habitat types (mountain grasslands and the understory of Mediterranean woodlands) across two consecutive years. Our aim is first to understand whether microscale features of species (e.g., their degree of generalization) can explain species and interaction persistence patterns. Second, we ask whether these microscale features are sensitive to the aggregation scale at which they are calculated (e.g., obtained from seasonally aggregated networks vs. weekly networks). Third, we explore whether these same microscale features can explain pollinator fidelity to plant species. Fourth, we focus on examining the drivers of interaction turnover through space and time, with a particular focus on understanding the contribution of species turnover and interaction rewiring. Finally, we explore whether interaction rewiring is driven mostly by neutral (e.g., changing species abundances) or niche (e.g., species generalization degree) processes, or rather by the composition of the larger community and the potential indirect interactions between co-occurring species (e.g., competitive interactions).

Specifically, our research aims to answer the following questions: (1) are species and interaction persistence

through space and time determined by a species' degree of generalization, (2) does the degree of pollinator generalization explain its fidelity to the plant species it visits through space and time, and (3) do changes in species composition and interaction rewiring contribute similarly to interaction turnover through space and time? We expect that species generalization will explain a large portion of the patterns in species and interaction persistence, as shown in previous research (Resasco et al., 2021), given the greater tolerance that generalist species have to environmental variation. However, we also expect that the level of generalization for a given species will be different if the aggregated network of interactions is used as compared to the weekly version. Nevertheless, we expect the same patterns regardless of the method used to calculate generalization. Further, we expect that pollinator species fidelity will be greater for generalist species, as their visitations will include many generalist plant species, who will also be more persistent through time and space. We further expect that, given the changes observed in both pollinator and plant communities through time because of their different phenologies, interaction turnover will be mainly driven by species turnover. Conversely, given the smaller turnover in species composition expected through space, we expect that interaction turnover will be much lower. In addition, we expect that together with species attributes (such as their degree of generalization), or their abundances, other factors related to indirect interactions with the larger community will also impact interaction composition. Specifically, we expect that competitive interactions with other species will also be important in driving the patterns observed in interaction turnover. By collecting data across two different habitat types, we can further assess the generality of the patterns observed and the potential mechanisms identified.

METHODS

Study sites

We collected data within two different study areas and habitat types in Spain: within the vicinity of Doñana National Park in SW Spain and at Gorbea Natural Park, located in N Spain (Appendix S1: Figure S1). Doñana features a Mediterranean climate, with warm, dry summers and cool, humid winters. Annual precipitation is 500 mm, and mean temperatures during the study period range from 12.5 to 22.5°C (Pizarro et al., 2021). Surveys were conducted within fragmented woodlands of stone pines (*Pinus pinea*) that host a rich understory of flowering shrubs and annual plants (Aparicio, 2007).

In turn, surveys in Gorbea were conducted within an area characterized by the influence of the Atlantic Ocean, with mild winters and summers and high annual precipitation values, ~1300 mm. Here, mean temperatures during the study period range from 10 to 17.5°C (Pizarro et al., 2021). Vegetation in the area is composed of beech forests in conjunction with abundant coniferous plantations. There is also a substantial presence of shrubland and meadows, dispersed across a rocky environment, present mainly in the steepest areas of the mountain (Albizu et al., 2002).

Surveys were conducted in two consecutive years, 2020 and 2021. Within each area, we selected five plots located at similar elevations (ranging from 50 to 150 m above sea level [asl] in the case of Doñana and between 800 and 900 m asl in the case of Gorbea), as well as within similar habitats and soil types, reducing potential confounding factors. Similarity in plant composition between plots was 0.42 ± 0.05 and 0.39 ± 0.01, respectively, in the case of Doñana and Gorbea (plant mean Sørensen beta-diversity). The average distance between plots ranged from 3.2 to 24.2 km in the case of Doñana and from 1.3 to 4.6 km in Gorbea. In Doñana, one of the plots had to be substituted in 2021 due to the impossibility of access in the second year of sampling, but it was changed to a very similar area nearby.

Within each plot, we established one 250×2 m transect. Transects were visited every week (from now on referred to as period) when weather conditions allowed. To record visits by pollinator species foraging at different times during the day, each transect was surveyed at three different times during the day it was visited: one in the early morning (9:30–10:30), one in the middle of the day (13:30–14:30), and one at the end of the day (16:30–17:30), to obtain a complete picture of the community of plants and pollinators present throughout the day. Each census lasted for 1 h. During the whole flowering season (from February to May in the case of Doñana and from March to July in the case of Gorbea), we were able to sample each of the plots between six and nine times, depending on the year. Sampling was interrupted for two weeks in 2020 due to the lockdowns derived from the COVID-19 pandemic. The objective was to sample all five plots per study site for one full day each during the week and then repeat the sampling again the following week in a randomized order, such that each week represented a period with a difference of 4 days in the sampling of each site. This was however not always possible due to lockdowns, restrictions, and bad weather, and therefore, on a small number of occasions, a period covers a maximum of a 10-day difference between the sampling of the first plot and that of the last. At each census, along each transect, we identified all plant species and recorded all the floral visitors that landed on their flowers and touched the plant's

reproductive parts. Only floral visitors (from now on referred to as pollinators) that could not be identified in the field were captured, stored, and identified in the laboratory by an expert entomologist (see *Acknowledgments*). In addition, for each period, we recorded the number of open flowers or flowering heads (for species with small clusters of flowers) produced by each plant species present in those same transects. All surveys were done under similar weather conditions, avoiding windy or rainy days.

Sampling completeness

We assessed sampling completeness for plant and pollinator species as well as for plant–pollinator links using the Chao1 estimator of asymptotic species richness for abundance data (Chao, 1984). To this end, we estimated the richness of plant, pollinator species, and plant–pollinator links accumulated as sampling effort increased up to 100% sampling coverage using package iNEXT (Hsieh et al., 2016). We estimated completeness for the fully aggregated network for each year and study area, as well as for each individual site across the different time periods and for each time period across all sites, which are the different levels of network aggregation used throughout the analyses.

Interaction and species persistence through time and space

First, we focused on interaction persistence through space and time, and on the potential drivers of this persistence. Following previous research (Chacoff et al., 2017; Resasco et al., 2021), we evaluated whether the degree of generalization of the species involved in an interaction determined interaction persistence. To this end, we used the subset of data where pollinator specimens were fully identified to the species level, which represents 86% of the total sampled specimens. We then created a plant–pollinator interaction matrix where plant and pollinator species were sorted to maximize nestedness, that is, plants were sorted in rows and pollinators into columns based on their decreasing degree (number of interacting partners), such that the more generalist species (those that interact with a greater number of partners) were located close to the top left corner of the matrix. For each focal interaction, generalization was approximated as one minus the combined Euclidean distance of the plant and pollinator species in the nested matrix to the upper left cell (see Chacoff et al., 2017 and Resasco et al., 2021 for further details on calculations). Relationships between generalization and spatial and temporal persistence were then tested by means of Spearman rank correlation tests

between this generalization degree (distance to the upper left cell) and four measures of spatial and temporal persistence: the number of years in which an interaction was observed, the number of plots it was observed in, the number of surveys per day in which it was observed, and its phenophase. Phenophase was calculated as the difference between the last and the first day of the year in which the interaction was observed, therefore providing a measure of the number of days in which an interaction could be potentially observed.

Similarly, at the species level, we tested for relationships between the pollinator and plant species' distance to the upper left cell of the nested matrix and their spatial and temporal persistence (number of years in which a species was present, number of plots it was observed in, and phenophase) by means of linear regressions with independent variables fitted to second-order polynomials. In the case of pollinators, we also analyzed the relationship between generalization level of the species and the number of surveys per day in which it was recorded. All analyses were conducted separately for the two study areas, Doñana and Gorbea.

This approach uses the aggregated network of interactions recorded across all plots and periods to calculate interaction and species generalization levels, therefore assuming that a pollinator or plant species is equally generalized across the whole flowering period and study area, which is not necessarily the case as species can change their foraging patterns (Magrach et al., 2020). To account for these changing generalization levels, we redid this same analysis, but we calculated both interaction and species generalization values for each census (i.e., for each combination of period and plot). We then obtained the median value for each interaction, plant, and pollinator species, which we related to the previous four measures of persistence using Spearman rank correlations in the case of interactions and linear regressions for species.

Interaction fidelity

Following the analysis of interaction and species persistence, we focused on understanding pollinator species fidelity to plant species across space and time. In this case, we use the definition of interaction fidelity used by McLeod et al. (2016), which defines it as a change in a pollinator species' preferences for plant species that is smaller than that expected by chance. We calculated this metric as the change in pollinator preferences between years, between plots, and between periods within the same year. In the case of between-year fidelity, for each pollinator species, we created a year-by-plant matrix

where cell values represented the number of pollinator individuals observed at each plant species each year. Then, based on this observed matrix, we generated 1000 null matrices that maintained the annual abundances of each pollinator species (as in MacLeod et al., 2016). For each pollinator species, we determined the between-year dissimilarity in the composition of plants visited using the Morisita–Horn dissimilarity index (Horn, 1966), as well as the mean value of dissimilarity across all null matrices, and obtained the 95% CIs for them. We then compared the observed dissimilarity values to the null CIs. A value of observed dissimilarity below that expected by chance indicated significant fidelity in plant species used by a particular pollinator species, while an observed value above that expected by chance indicated interaction turnover. Then, we evaluated whether these values of fidelity or turnover were related to the degree of species generalization, using both the value calculated using the aggregated network of interactions (as per Resasco et al., 2021) as well as the version we calculated earlier that takes into account potential differences in generalization level through time and space and is calculated as the median across all censuses. We repeated the same process in the case of between-plot and between-period dissimilarity. By using this null model approach, we eliminate confounding effects of species abundances and partner choices (MacLeod et al., 2016).

Interaction turnover

We then focused on understanding the patterns and potential drivers of spatial and temporal interaction turnover. To this end, we calculated the turnover in interaction composition across space and time (within-year). To assess interaction turnover through space, we used data for each site aggregated for the different time periods per year, while in the case of turnover through time, we used data aggregated across all sites for each of the different time periods. Interaction turnover was calculated using the function *betalinkr* (Fründ, 2021; Poisot et al., 2012) in package bipartite (Dormann et al., 2009). This function calculates dissimilarity in interaction composition between pairs of communities and further partitions this dissimilarity into dissimilarity due to changes in species composition (ST) and that due to interaction rewiring (OS). In this case, interaction rewiring is considered as dissimilarity in species interactions between species that are shared by both communities (as opposed to other definitions used in the literature), which consider rewiring as a change in pollinator partner choice not restricted to co-occurring species (MacLeod et al., 2016). We used the Ruzicka dissimilarity index and the

quantitative version of our plant–pollinator networks. We further decomposed interaction turnover due to changes in species composition into changes in pollinator species composition (ST.h), plant species composition (ST.l), or both (ST.hl).

Potential drivers of interaction rewiring

Next, we focused on understanding the potential mechanisms driving interaction rewiring in space (given the prevalence of this process driving spatial interaction turnover, see *Results*). First, we assessed whether there were differences in species abundance between species involved in interaction rewiring compared to those that did not rewire. For plants, we used the number of floral resources as a measure of abundance and the observed occurrence in the case of pollinator species. In both cases, we fit linear mixed models with floral abundance or pollinator occurrence as dependent variables and plot, period, year, and their interaction with species type (rewiring vs. not rewiring) as independent variables. Plant or pollinator species and study area were included as random factors. Then, we evaluated whether interaction rewiring involved species whose abundance was more variable in space than those that did not rewire. Variability was calculated as the CV of plant or pollinator abundance. As above, we fit linear mixed models, this time including only year and its interaction with species type as explanatory variables and plant and study area as random effects.

We then also assessed whether interaction rewiring could be explained by changing preference patterns of pollinator species. Pollinator preference by a specific plant species was defined as the nonrandom association of a pollinator species with a plant species; that is, if a pollinator species was observed feeding on a plant species more often than expected by chance, then this pollinator species would be assumed to show a preference for that particular plant species (similar to habitat preference analysis used in Collado et al., 2019). To this end, we developed 1000 null matrices of plant and pollinator interaction frequencies using Patefield's algorithm (Patefield, 1981) in package bipartite (Dormann et al., 2008), which maintains the row and column sums, such that the proportional abundance of species is maintained but interactions are reshuffled. A pollinator species was considered to prefer a plant species if the interaction frequency observed between both was greater than the 0.95 quantile of the expected frequency extracted from the 1000 null models.

We fit a logistic regression with the preference (binary variable describing preference or neutrality) of each pollinator species for each plant species within a plot and

period as a response variable. We included year, flower abundance, plant and pollinator species richness, the degree of generalization of the pollinator species (measured as the median distance to the top left corner of the nested network across each census), and the average potential competition between a focal pollinator species and other pollinators in the community. In addition, we included the two-way interaction between these variables and the binary variable reflecting whether pollinator species were involved in interaction rewiring. Plant species and study area were included as random factors. We ran all combinations of models with these variables using function "dredge" in package MuMIn (Bartoń, 2020) and selected the best one based on the lowest AIC_c value.

Potential competition reflects a measure of possible influence of each pollinator species on the rest of pollinator species in the community via shared plant resources. We calculated potential competition using an index originally proposed by Müller and collaborators to quantify apparent competition between resource species through shared consumers (Müller et al., 1999), but that has been used in other pollination studies to quantify indirect interactions between plants (Carvalheiro et al., 2014) and pollinators (Magrach et al., 2017). The index quantifies, for each pair of pollinator species, the potential for showing competition with another species, mediated through the plant species level, using function PAC in package bipartite (Dormann et al., 2008). We computed this value for each co-occurring pair of pollinator species and then obtained a single measure for each species as the average value across all pairwise calculations the species is involved in.

RESULTS

Across both our study areas and years, our sampling spanned 435 h of observation. During this time, we recorded a total of 61 and 82 plant species in Doñana and Gorbea, respectively. Out of these, between 68.57% and 100% of the plant species found in the different sites in Doñana, and between 61.54% and 82.35% of the plant species present in Gorbea received at least one visit from floral visitors. Floral visitors included a total of 145 and 125 species or morphospecies of flower visitors in Doñana and Gorbea, respectively, across both study years (see Appendix S1: Table S1 for distribution in different families and Appendix S1: Figure S2 for their different phenologies), mainly belonging to families Apidae and Syrphidae. Collectively, these species performed 2287 and 2641 flower visits in Doñana and Gorbea, respectively. We recorded 405 and 435 unique

combinations of plants and flower visitors in each of the study areas.

Sampling completeness

Our analysis of sampling completeness across our two areas sampled shows that with our sampling efforts we were able to obtain high sampling coverage for both pollinator species (81% and 67% of pollinator species detected in 2020 and 2021 in the case of Doñana and 63% and 54% in the case of Gorbea) and plant communities in both study years (95% and 87% of plant species, respectively, for 2020 and 2021 in the case of Doñana and 92% and 94% in the case of Gorbea). For plant–pollinator links, sampling completeness was lower, as expected, in both study areas (60% and 49%, respectively, for 2020 and 2021 in the case of Doñana and 42% and 51% in the case of Gorbea). Aggregating data across all sites for each of the different periods showed values ranging between 49% and 64% for pollinator species, 70% and 84% for plant species, and 35% and 51% for plant–pollinator links across both study areas and years (Appendix S1: Table S2 and Figure S3), while aggregations through time for each site showed values ranging between 57% and 61% for pollinator, 75% and 90% for plant species, and 32% and 41% for plant–pollinator links, respectively (Appendix S1: Table S2 and Figure S3).

Interaction persistence

In response to our first question related to individual species and interaction persistence through space and time and their relationship with a species' degree of generalization, our results show different outcomes depending on the aggregation level used to calculate the degree of generalization. Our results using generalization values calculated using the aggregated network of interactions show that interactions involving more generalist species were those that persisted more through time (across a greater number of years: Spearman's rank correlation coefficient = 0.39, $p < 0.001$ and 0.26, $p < 0.001$, respectively, for Doñana and Gorbea, days: Spearman's rank correlation coefficient = 0.51, $p < 0.001$ and 0.48, $p < 0.001$, and present more frequently throughout different censuses per day: Spearman's rank correlation coefficient = 0.53, $p < 0.001$ and 0.43, $p < 0.001$). Interactions involving generalist species could also be found more widespread in space (across a greater number of plots: Spearman's rank correlation coefficient = 0.53, $p < 0.001$ and 0.49, $p < 0.001$). The same holds true at the species level for both pollinator and plant species, with more generalist

species (i.e., those closer to the upper left cell in the nested matrix) being present across both years, having larger phenophases and spatial representation across more sites (Appendix S1: Table S3 and Figure S4).

A reanalysis of the relationship between interaction persistence and the degree of generalization of the species involved using the median value of generalization calculated for each species at each census, however, shows a different pattern. In this case, we find that although the correlations between the Euclidean distance to the core of the nested network and interaction persistence through space (plots: Spearman's rank correlation = 0.29 and 0.21, $p < 0.005$ in Gorbea and Doñana, respectively), and time (number of years: Spearman's rank correlation = 0.12 and 0.18, $p < 0.05$, and days: 0.24 and 0.23, $p < 0.005$ in Gorbea and Doñana, respectively) are maintained, their strength decreases. Further, when analyzing species-level relationships between generalization and species persistence, we found that the previously significant relationships now disappear, such that generalist species are not

more prone to persist through space and time (Figure 1; Appendix S1: Table S4).

Interaction fidelity

In response to our second question, related to whether a species' generalization degree can also explain its fidelity to the group of plant species it visits through time and space, our results show that fidelity is a rare phenomenon in our studied communities. Pollinator species that visit the same species of plants across years more often than expected by chance represent between 4% and 9.5% of the species in both study areas. In the case of plots, fidelity is present in 16%–17.6% of the species, while this value represents 10%–17% of the pollinator species that visit the same species of plants across periods. Conversely, pollinator species that change the plant species they visit more often than expected by chance (i.e., unfaithful pollinator species) across years represent 24%–28.6% of the
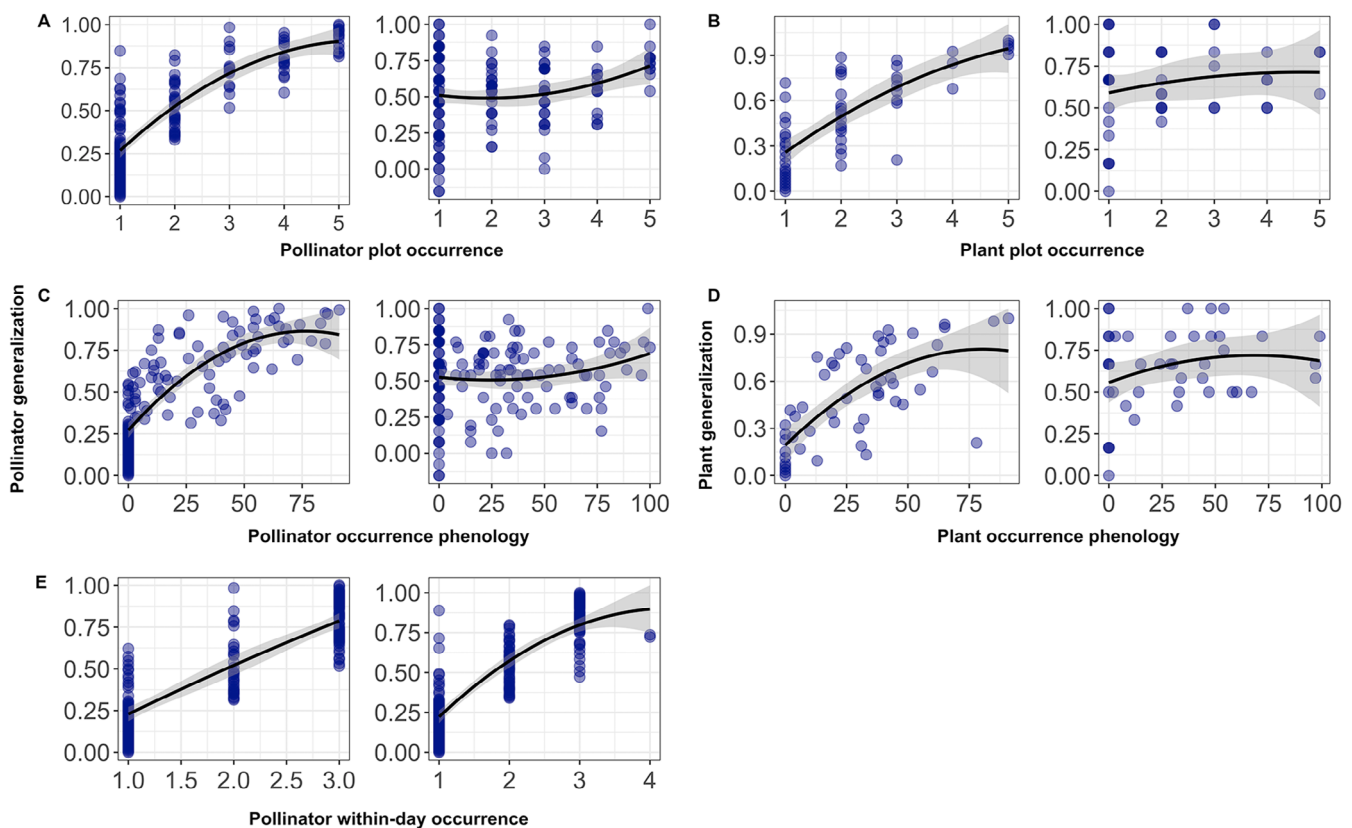

FIGURE 1 Species-level relationships between generalization, measured as the median of the proximity to the core of the nested network across each of the censuses in which that species is recorded, and number of plots in which the (A) pollinator or (B) plant species is recorded, phenophase lengths for (C) pollinators and (D) plants, and (E) number of censuses per day in which a pollinator species is recorded. Plant phenophases are defined as the last (maximum) minus the first (minimum) day of the year in which each species was recorded interacting with pollinators, while pollinator phenophases are defined as the last minus the first day of the year in which each species was recorded interacting with flowers. Gray shaded areas represent 95% CIs around means of model.

species, 21.6%–23.2% across plots, and across periods of time 24.5%–28.3%, with the remainder of the species showing no deviations from a normal expectation (Figure 2). A visualization of the extent to which fidelity or turnover was related to species generalization levels (calculated using weekly data) shows that more generalist species tend to show larger values of interaction turnover than expected by chance, although pollinator species that change the identity of their interactions are found across the whole spectrum of generalization levels (Figure 2). The few species that show significant fidelity to plant species through space and time show medium-high values of generalization (Figure 2). This result is essentially the same when calculating the degree
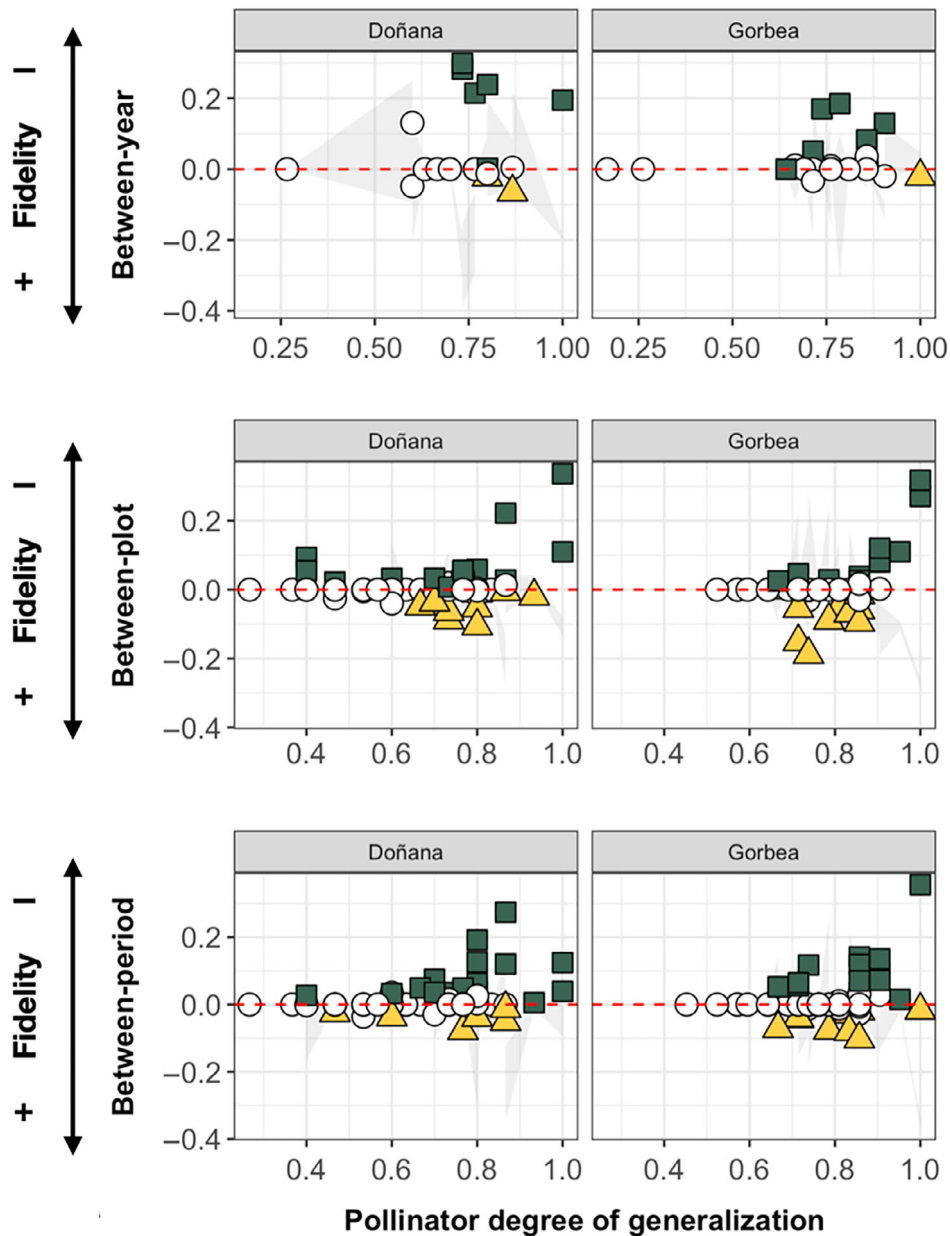

FIGURE 2   Results of fidelity analysis using null models of field data, ranked in increasing order of pollinator species generalization (calculated as the median distance to the core across nested networks obtained from each census) on the *x*-axis. The shaded area is bounded by each pollinator species' empirical change in preference, subtracted from the upper or lower bound of the null model 95% CIs for changes in preference. Changes in preference were measured as mean Morisita–Horn dissimilarity across all pairwise comparisons of years. The points represent species divergence from the mean Morisita–Horn dissimilarity obtained from the null model (observed − expected). Open circles in the shaded area indicate no difference from random expectations, solid yellow triangles below the shaded area indicate significant fidelity, and solid green squares above the shaded area indicate interaction turnover. CIs and shaded areas are uneven because each null model is based on the data for a particular pollinator species.

of generalization of a species using the aggregated network (Appendix S1: Figure S5).

Interaction turnover

In response to our third question related to the drivers of interaction turnover through space and time, we found that 68%–83% of the interactions observed in one particular year are unique to that year, not being observed the other year in both study areas (Appendix S1: Table S5). Although these unique interactions are less frequent than common ones, interactions that are uniquely observed one year can represent 15.5%–95% of the interactions observed at a given plot and period of time in the case of Doñana and 12%–80% of the interactions observed in Gorbea.

Our decomposition of total interaction turnover into its interaction rewiring and changes in species composition components shows that the former seem to be more prevalent across space, while the latter determine the main differences in interaction composition observed through time (Table 1, Figure 2). This means that despite there being the same composition of plant and pollinator species across plots, interaction frequencies between them are different (Figure 3, Table 1). A further decomposition of species composition differences shows that it is changes in the composition of pollinator species that mainly drive interaction turnover through time (Figure 3).

Specifically, our results show that 53%–62% of the interactions between plants and their pollinators are only observed within one of the surveyed plots at each study area, and 59%–76% of the interactions are only recorded in one period and not observed in the one immediately following. A decomposition of this turnover in interactions shows that both inter- and intra-year changes in interaction composition are mostly due to changes in species composition, while interaction rewiring represents most of the variability in interaction composition between plots (Appendix S1: Table S5).

Potential drivers of interaction rewiring

Finally, our analysis of the potential drivers of interaction rewiring through space shows that although flower abundance varies across plots and through time, plants that were involved in interaction rewiring were not more variable in the number of floral resources they produced than those plants that are consistently visited by the same pollinator species (Appendix S1: Table S6). In the case of pollinators, we found that the frequency at which pollinator species were observed changed through time while remaining constant through space. Although, in general, pollinator species that rewired tended to be more frequently observed than those that did not (Appendix S1: Table S6), both types of species (those that rewire and those that do not) were equally variable (no differences in their CV through space; Appendix S1: Table S6).

Our analysis of pollinator preference patterns shows that interaction frequencies between plants and pollinators that are more frequent than expected by chance can be explained by two main factors: plant species richness and the average potential competition between a focal pollinator species and all of the co-occurring pollinator species (Table 2). In this case, we find that species of pollinators that do not rewire their interactions in space suffer much less potential competition than species that do rewire (Appendix S1: Figure S6), and they respond to these lower levels of competition by focusing on a specific plant resource more often than expected by chance (Figure 4). Our best model did not include either the number of floral resources produced by a plant or the level of generalization of a pollinator species, suggesting these two variables do not affect preference patterns in both of our studied systems.

DISCUSSION

Our results show that although species generalization does not seem to affect their persistence through space and time, generalist species show both the greatest levels of interaction turnover through space and time as well as the greatest levels of fidelity to plant species, suggesting they are a rather heterogenous group of species. We further find that, contrary to our expectations, interaction turnover is large both through space and through time. Through time, turnover is mostly driven by changes in species composition, particularly by changes in pollinator
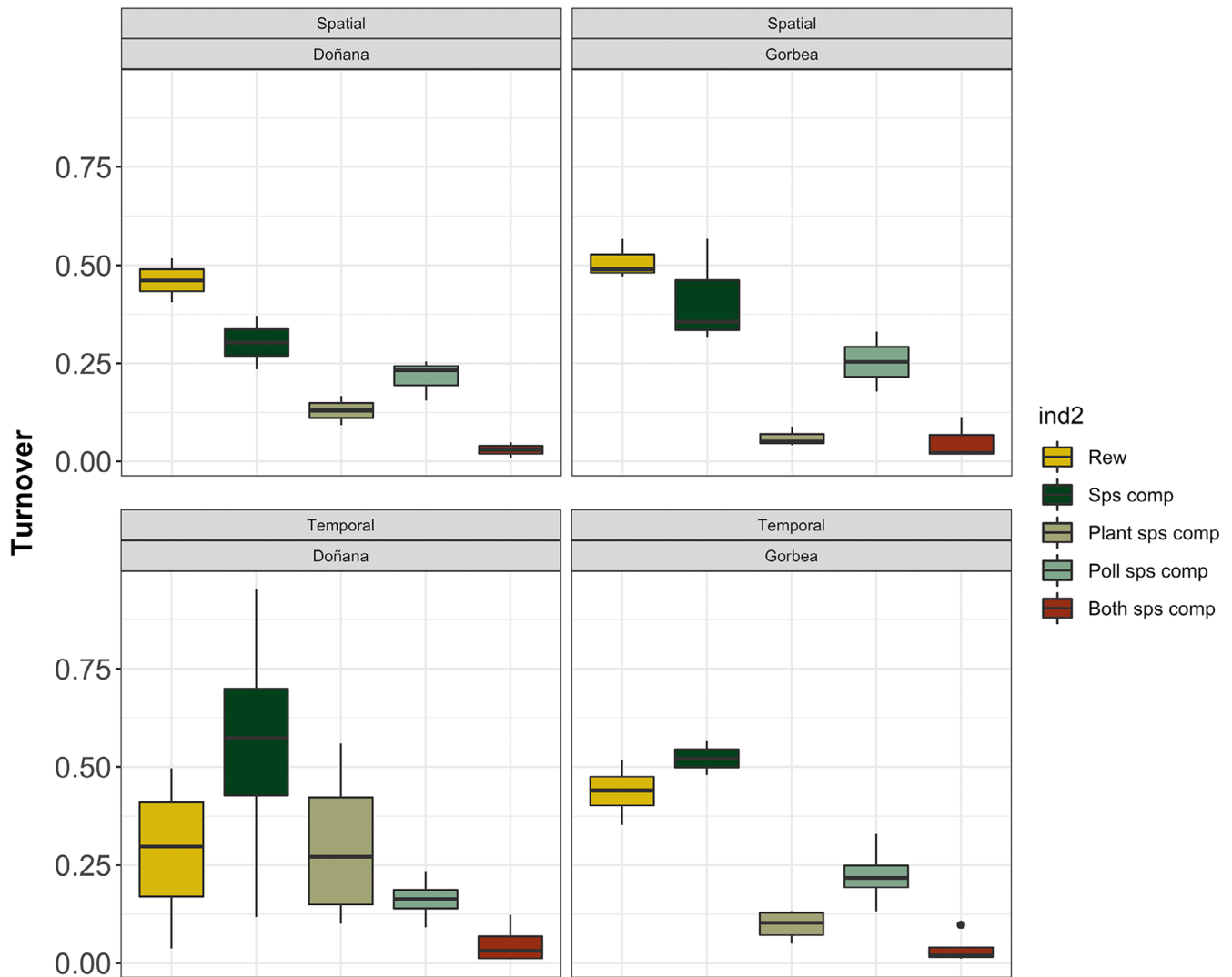
TABLE 1 Results of linear mixed model evaluating differences in the magnitude of turnover due to different components (rewiring and changes in species composition) across space and time.

Variable	2.5%	Estimate	97.5%	t value
(Intercept)	**0.34**	**0.49**	**0.64**	**5.99**
Turnover decomposition	−0.32	−0.12	0.08	−1.12
Type (spatial vs. temporal)	−0.31	−0.13	0.05	−1.34
Year	−0.12	0.01	0.13	0.08
Turnover decomposition × type (spatial vs. temporal)	**0.05**	**0.30**	**0.55**	**2.17**

Note: Values represent variable estimates and 95% CIs. Variables and values in boldface indicate significant variables whose CI does not overlap 0.

FIGURE 3 Boxplots showing the decomposition of interaction turnover into interaction rewiring and changes in species composition components across space and time for both study areas. Turnover due to changes in species composition is further decomposed into changes in species composition of plants, pollinators, or both. Box represents interquartile range, midline indicates median value, and whiskers represent minimum and maximum data values.

species composition, while interaction turnover through space is mostly a consequence of interaction rewiring. Rewiring in this case is not driven by spatial changes in plant or pollinator species abundances but rather by indirect competitive interactions between pollinators through their shared plant resources. Here, we show that pollinator species that do not rewire are involved in fewer competitive interactions, and their immediate response to a slight increase in competition is to focus on a preferred plant resource. In turn, pollinator species that do rewire are able to sustain greater levels of potential competition before focusing on specific resources.

Following recent research presented by Resasco et al. (2021), we focused on the relationship between species and interaction generalization levels and their persistence through space and time. When using the measure of

generalization based on the aggregated version of the plant–pollinator interaction network, we find results that are similar to those presented by Resasco et al. (2021). However, a focus on a value of generalization that considers the dynamic nature of species preference patterns (Magrach et al., 2020) shows different results. In this case, correlations between interaction generalization and persistence decrease, and relationships between species generalization level and persistence through space and time become nonsignificant. Here, we focus on a description of species generalization from a functional perspective as a measure that gives an idea of how efficient a pollinator will be from the plant's perspective, assuming that a more generalist species visits a greater variety of plants and brings heterospecific pollen to plant individuals (Brosi & Briggs, 2013). By using the aggregated

TABLE 2   Results of linear mixed model analyzing potential drivers of pollinator preferences.

Variable	2.5%	Estimate	97.5%	z
(Intercept)	−2.04	−0.81	0.55	−1.11
Pollinator species richness	−0.05	0.01	0.05	0.20
Plant species richness	**−0.45**	**−0.23**	**−0.06**	**−2.03**
Potential competition	**0.21**	**1.08**	**1.96**	**2.52**
Rewiring	−2.03	−0.60	0.53	−0.80
Year	−0.53	0.07	0.58	0.25
Plant species richness × rewiring	−0.05	0.11	0.36	1.00
Potential competition × rewiring	**−1.74**	**−0.82**	**0.00**	**−1.86**

Note: Values represent variable estimates and 95% CIs. Variables and values in boldface indicate significant variables whose CI does not overlap 0.

network to calculate generalization, Resasco et al. (2021) are assuming that a species is equally generalist across the whole flowering period and across the whole study area, something that has proven to not be the case for many species that shift from more generalist to more specialist roles (Magrach et al., 2020).

When focusing on interaction fidelity, we find that generalist species are the ones that shift more of their interactions through time and space (greater turnover than expected by chance), but medium and greatly generalist species are also the few that are prone to visit similar groups of plants through space and time. Therefore, generalist species represent an idiosyncratic group of species that includes highly faithful species as well as species that shift interactions frequently. Generalist species are also the most abundant ones, whose degree of generalization is driven by greater encounter probabilities with potential partners (Fort et al., 2015). However, our results show that despite having large abundances as a common feature, the way in which they interact with their partners is different.

A subsequent analysis of interaction preferences shows that the level of generalization of a pollinator species does not explain differences in their preferences for different plant species. Rather, here we find that it is the diversity of plant species present at a given plot and time, and even more so, the indirect interactions between pollinator species through shared plant resources, that drive these differences. Our results are compatible with previous research showing that the composition of the wider pollinator community and their use of available floral resources are major factors shaping interaction composition (Brosi & Briggs, 2013; Magrach et al., 2017). For example, Brosi and Briggs (2013) found that the

removal of a highly generalist species from a grassland community affected the preferences of other pollinators for plant resources, as they expanded their foraging niches in the absence of the strong competitor.

Within our dataset, we find that while most plant species were consistently recorded across both study years, a significant proportion of the interactions and pollinator species were uniquely recorded during one of the study years. In the case of pollinator species, we find values of pollinator persistence across years ranging from 54% to 73%. These values are intermediate compared to those found in other studies. For example, Olesen et al. (2008) found that 80% of the pollinator species present in an arctic heathland were recorded during two consecutive study years, while other studies have found this value decreased to 20.5% (Petanidou et al., 2008) or <25% or of the total pollinator fauna (Dupont et al., 2009). We find these values despite our large sampling efforts, spanning multiple locations and times, and including several surveys throughout the day to account for daily dynamics. This could mean that our sampling was still too low to uncover all existing species and interactions, or it could mean that some of these interactions follow neutral processes through time and are a consequence of random encounters between plants and pollinators based on species abundances (Vázquez et al., 2009), which do not necessarily replicate every year as species abundances shift through time (e.g., bet hedging; Danforth, 1999).

Interestingly, we find that the patterns and the processes that explain interaction persistence and turnover are very similar for both of our studied areas, despite the different habitat types they represent. The main difference between both habitat types seems to be the level of competition between pollinators, which is larger in Gorbea than in Doñana. This might reflect a greater scarcity of resources. Indeed, in Gorbea, located in a mountain area that features much larger environmental differences throughout the day as well as between weeks, pollinators might have less feeding opportunities and might be more adaptive to changing conditions (Ploquin et al., 2013), showing a more plastic response in their use of resources. This might reflect the greater ability of this community to adapt better to future perturbations, such as climate change. Or maybe our findings show that they are already adapting to it, as mountain areas are one of the most impacted by changing climates (Inouye, 2019; Schmeller et al., 2022). Data should be collected across more habitat types to be able to confidently assess whether this is the case.

While our dataset represents a significant sampling effort, encompassing two different habitat types and five sites per habitat sampled multiple times during the flowering season and multiple times during the day for
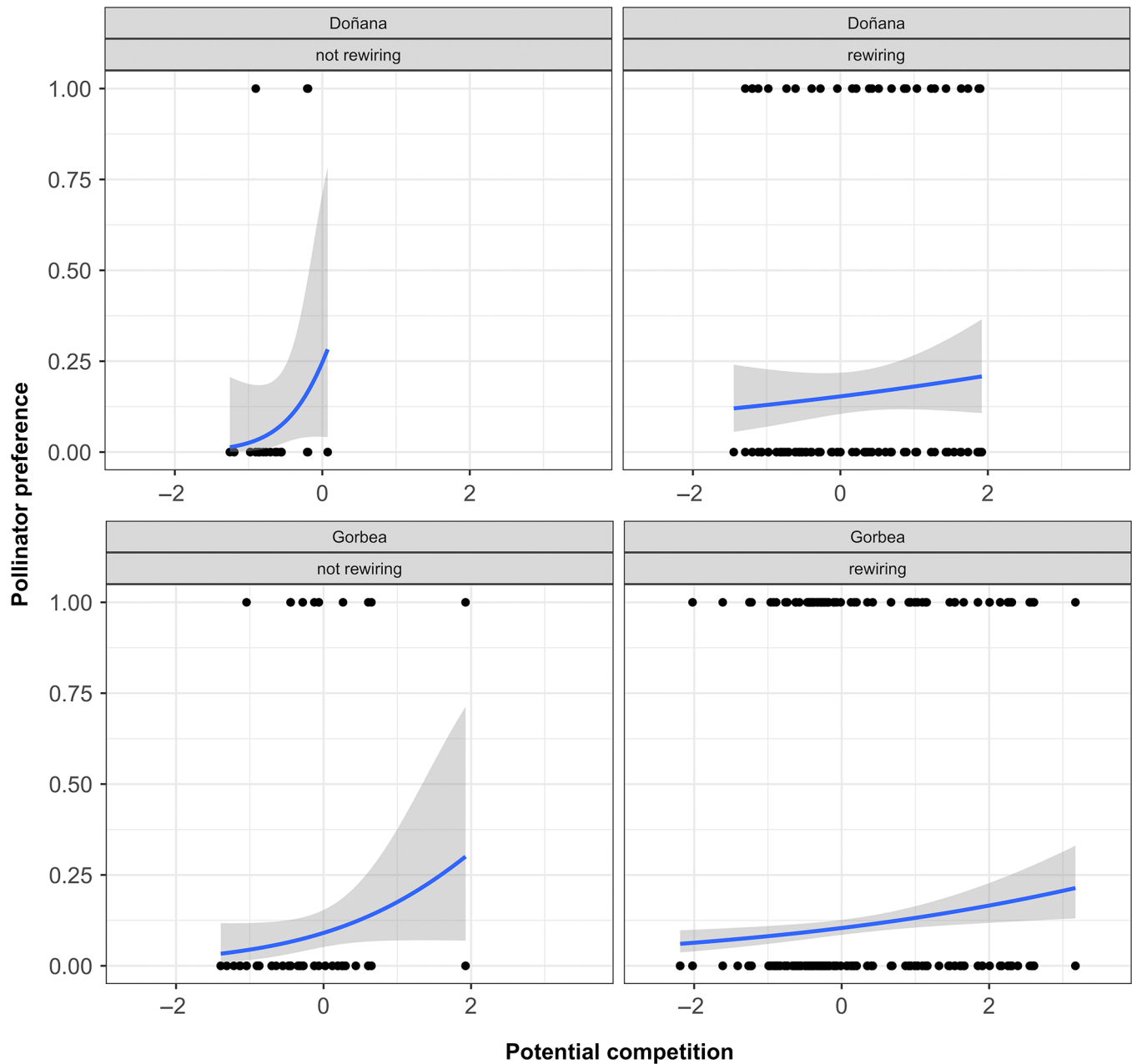
FIGURE 4 Scatterplots showing the effect of average potential competition between each focal pollinator species and all pollinator species it co-occurs with and their preference for a particular plant species. Preference is defined as an observed interaction frequency between a plant and a pollinator species that is greater than that expected by chance. Results are shown for the two study areas separately and for pollinator species that show interaction rewiring through space and those that do not.

two consecutive years, our weekly sampling completeness is still relatively low, particularly for plant–pollinator interactions. And this could be impacting some of the results we find. Specifically, the pattern we observe where temporal variation in interactions is driven by species turnover while spatial variation is a consequence of rewiring might occur because spatial networks aggregated across the entire season have much more room for rewiring compared to weekly networks aggregated across plots. In addition, in our analyses, we aggregate networks at the scale of

the entire season to look at variation across space. By contrast, temporal networks are examined across weeks for intra-annual variation in interaction composition, and we therefore have spatial variation in full-season networks and temporal variation in full-site networks. Another approach could involve examining spatial variation at the scale of the week, but given the relatively low sampling completeness we obtain, these analyses could be challenging. In this case, we would not expect significant changes in our results since our analyses of rewiring are done for

species that are common across the different sites, species that would only be present simultaneously for a shorter period of time than the full flowering season, in many cases for a period of time similar to our sampling frequency.

The research we present here answers, at least partially, the question posed by Trøjelsgaard and Olesen (2016): "why do some species interact one year but not the subsequent although they co-occur in both years?" Our results show that together with trait-matching (measured here through phenological overlap) and neutral processes driven by differences in species abundances (Vázquez et al., 2009), the composition of both interacting communities, and the indirect interactions between species within a trophic level, play an important role in determining species interactions. This probably explains why predictive exercises have been somewhat successful in predicting macroscopic features of plant and pollinator communities but not so much in predicting pairwise interactions (Olito & Fox, 2014; Vázquez et al., 2009), which vary across space and time (Trøjelsgaard & Olesen, 2016) as a consequence of all these different processes.

Furthermore, our research results demonstrate that the way we choose to build ecological networks can have dramatic effects on the results we find. In this case, our main differences with previous research lie in the definition of what a generalist species is. Here, rather than defining a generalist pollinator species as one that interacts with multiple plant species throughout its full active period across the whole flowering season and that includes plant species that do and do not temporally co-exist, we define its degree of generalization by the number of plant species it interacts with during a given time period (a week in this case) in which all the different plant species co-exist in time. This measure of generalization focuses more on a functional perspective, such that the more generalist a species is, the higher the probability that it will deposit heterospecific pollen (although this species-level measure of generalization also ignores individual foraging behaviors, which can be different from the species-level ones) (Tur et al., 2013). The temporal scale-dependence of different network metrics has previously been shown by Schwarz et al. (2020) in their reanalysis of 30 individual datasets of plant–pollinator interaction networks aggregated at five different temporal scales, which demonstrated that the aggregation level had a significant impact on five of the six metrics they calculated. Therefore, our results also support the idea that network aggregation should be based on the questions to be explored, with macroscopic features being more robust to the aggregation scale, while meso (e.g., motifs) and microscale features show a greater sensitivity to the aggregation used.

In summary, our results show the importance of simultaneously considering spatial and temporal dynamics within natural communities to understand how they assemble and point towards other determinants of interaction composition beyond niche or neutral processes, such as indirect interactions between species.

AUTHOR CONTRIBUTIONS

Ainhoa Magrach originally formulated the idea. Ainhoa Magrach, Maddi Artamendi, Paula Dominguez Lapido, Clara Parejo, and Encarnacion Rubio conducted fieldwork. Ainhoa Magrach performed statistical analyses and wrote the first version of the manuscript. All authors contributed to revisions and improvements to the manuscript.

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CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

DATA AVAILABILITY STATEMENT

Data (Magrach, 2023) are available from Figshare: https://doi.org/10.6084/m9.figshare.22492432.

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