

# Increasing dominance by a native shrub effects plant-pollinator interactions but not pollen transfer

Tomas Mitchell-Storey<sup>1\*</sup>

\*tomasmitchellstorey@gmail.com

<sup>1</sup>Research School of Biology, ANU, Canberra, ACT, Australia

## Abstract

Climate change is causing plant species to shift distributions, especially in alpine environments. These changes could potentially impact plant-pollinator interactions and pollen transfer. This study assessed how *Nematolepis ovatifolia* (F. Muell.) Paul G. Wilson, a mass flowering shrub in Kosciuszko National Park, New South Wales, Australia, effected the pollination ecology of co-flowering. Visitation surveys in small plots with differing abundances of *N. ovatifolia* were conducted to construct plant-pollinator networks. A compound light microscope was used to count conspecific (CP) and *N. ovatifolia* pollen grains on plant stigmas. Bipartite networks and generalized linear mixed models were used to find patterns in the data. There was evidence for *N. ovatifolia* effecting visitation patterns at high densities. However, *N. ovatifolia* abundance was found not to significantly effect CP and *N. ovatifolia* pollen deposition onto the stigmas of other plants. CP deposition was significantly lower in *Pimelea alpina* F. Muell. ex Meisn. and *N. ovatifolia* pollen deposition was very low across all species. Visitation was not a good predictor for pollen transfer. Further studies should include more plant species over longer time periods and analyze seed set. These findings demonstrate that the effect of range shifting native plants on plant-pollinator networks may not be analogous to that of invasive species.

## Introduction

Anthropogenic-driven climate change is resulting in a shifting distribution of plant species. Shifts in plants are likely to be caused by changes in air temperature and precipitation, rather than fire regime or air pollution (Kelly and Goulden 2008). As the climate continues to warm, it is likely that distribution shifts will become more evident over time (Mansfield et al. 2020). These range shifts are pronounced in alpine environments, where temperature gradients closely follow elevational gradients. While shifts up elevational gradients are common, plants are variously sensitive to local climate, sometimes resulting in downwards shifts (Koorem et al. 2018; Lenoir and Svenning 2015). In Australian alpine ecosystems, alpine plants have been recorded as shifting altitude by as much as 10 meters per year (Auld et al. 2022). This presents a problem to alpine and cold adapted plant species, which have an already limited range. Furthermore, the rates of expansion are not equal amongst growth forms. Under experimental warming, forbs were shown to increase in cover by 9%, however shrubs were shown to increase in cover by twice that amount (Wahren et al. 2013). The composition of vegetative

assemblages prior to anthropogenic disturbances was likely to have been strongly mediated by competition, and sometimes facilitation, for resources, such as nutrients, space, and pollinating insects (Callaway and Walker 1997). Pollination of plants by insects is a mutualistic interaction which is predicted to be locally stable, so that competition for pollinators between plants is minimal (Ugo et al. 2009). It is therefore important to describe how anthropogenic-driven changes to plant communities might affect historically stable plant-pollinator interactions.

The response of plant-pollinator networks to the effects of climate change is an area which has not been fully explored. Kosciuszko National Park in New South Wales, Australia, is a useful study system to test such theories because of the high spatial turnover of flowering plant communities. If certain plants are expanding across relatively small spatial scales into adjacent areas, it may result in novel competitive interactions with other plants. These interactions may manifest in pollen transfer, by increasing heterospecific pollen (HP) transfer and decreasing conspecific pollen (CP) transfer (Parra-Tabla & Arceo-Gómez 2021). Plant-pollinator structure would also change due to the addition of a new plant species. These effects may be analogous in nature to those of exotic invasive plants which are more thoroughly documented (McDougall et al. 2018; Muñoz & Cavieres 2008; Pickering et al. 2011). Parra-Tabla and Arceo-Gómez provide an excellent outline of the integration of invasive plants into a native plant-pollinator community (Parra-Tabla & Arceo-Gómez 2021). Whether the climate-change driven range expansion of native plants have analogous impacts on plant-pollinator interactions remains to be seen (Valdovinos 2019).

*Nematolepis ovatifolia* (F.Muell.) Paul G.Wilson is a mass flowering supergeneralist shrub which grows in the alpine regions of KNP (Encinas-Viso et al. 2022). For this reason, it is a good candidate to predict how range shifting native plants will impact nearby plant-pollinator communities. Insect visitation surveys are often used demonstrate patterns in pollinator distribution (Solís-Montero et al. 2022); while pollen transfer (CP and HP) can be used to estimate reproductive success (Lopes et al. 2022). Research that incorporates both effects onto a plant-pollinator community can provide far more predictive power (Albrecht et al. 2016).

Visitation networks are often used as a simple measurement for the success of pollinator recruitment (Lundemo 2007). While visitation may not necessarily imply pollination, combining many observations can help estimate whether animals are preferentially visiting certain plants. This is usually visualized with a bipartite network formed from interactions between plants and pollinators, from which calculated metrics can be used to derive other useful information about the system (Young et al. 2021). Additionally, visual analyses allow for easy interpretations of network structure, such as modularity, which in this case is a measure of how much pollinators tend to share interactions with a flowering plant (Carstensen et al. 2016).

Various observational studies show that the Australian alpine is a highly nested and generalist system (Inouye & Pyke 1988; Encinas-Viso et al. 2022). A generalist plant in a highly nested network would strongly compete with native generalist plants but could also reduce the pollination services received by specialist plants if it attracts a significant amount of generalist pollinators. This seems a likely scenario considering the abundance and diversity of generalist flies in the community. On the other hand, invading species may not always negatively affect the reproductive success of nearby plants. Models by Rathcke (1983) demonstrate that co-flowering plants can engage in facilitatory interactions, although at high densities this can result in competition, when interactions benefit one species more than the other. There has been existing and an increasing number of studies in KNP related to pollination. Inouye and Pyke (1988) completed a historic overview of the plant-pollinator community, and recent studies have begun to further unravel plant-pollinator dynamics in the area (Encinas-Viso et al. 2022).

These studies were completed sampled at a similar place and time of year, and so will provide key points of comparison to the findings in this study.

While visitation data is useful for describing plant-pollinator interactions, a mounting collection of evidence shows that there is no reliable correlation between visitation and reproductive success (Thompson and Knight 2018). Measurements such as pollen deposition or seed set are more direct indicators of reproductive success, primarily because pollinator species differ in their efficiency to pollinate. Therefore, a plant may only require one or few pollinators to avoid pollen limitation even if it interacts with many (Yu et al. 2012). Counting conspecific pollen (CP) and *N. ovatifolia* on the stigmas of native flowers along an abundance gradient of *N. ovatifolia* could highlight how broad functional traits, such as height, of native plants impact their response to invasive plants (Daniels & Arceo-Gómez 2020). Linking visitation and pollen deposition data could bridge the gap between observational analyses of network structure and real ecosystem function (Parra-Tabla & Arceo Gómez 2021). Specifically, comparing pollen deposition on flora in the vicinity of differing abundances of *N. ovatifolia* may reveal what influence *N. ovatifolia* has on those species. While it may be anticipated that there will be high HP deposition between nearby flowering plants, several studies have found that deposition of HP occurs only at very low frequencies (Gilpin et al. 2019; Taveira et al. 2023). These studies mentioned that low HP deposition could be explained by high rates of specialization, whereas KNP has low rates of specialization. A meta-analysis by Arceo-Gómez et al agrees with these conclusions and predicts that the elevation and latitude of KNP could result in higher rates of HP transfer (Arceo-Gómez et al. 2019).

Floral traits affect pollinator recruitment, for example having large or colorful, generalized flowers with high nectar rewards can increase visitation (Daniels and G. Arceo-Gómez 2020). *N. ovatifolia* flowers are small but sprout in great abundance with flowers that overlap in colour with nearby flowering plants. This may result in increased competition with those flowers. To receive pollen, invading plants must induce visitation from native pollinators. Because it is native to the area, *N. ovatifolia* is likely to be already adapted to the native pollinator community at a broad scale. However, its presence in new areas may change local pollinator behavior. A network construction by Encinas-Viso et al in 2022 found *N. ovatifolia* to be the most generalized plant in the community of which this study has also sampled from. Therefore, invasion by *N. ovatifolia* into new areas may result in the attraction of existing insects, and the introduction of new ones, thus altering the insect community structure. This assumption arises from the correlation between plant and insect communities evident in other surveys (Losapio et al. 2016; Zhu et al. 2015). The issue of physical structure in relation to pollinator interactions should also be considered. Ground level forb species are a key component of the KNP flora, such as *Pimelea alpina* F.Muell. ex Meisn and *Ranunculus grainiticola* Melville. These species may experience reduced pollination services if they are physically covered or obscured by *N. ovatifolia*, especially if pollinators forage opportunistically (Essenberg 2013). One study by Klecka et al shows how temperate Syrphid flies forage opportunistically and are more likely to visit taller inflorescences (Klecka et al. 2018A). Shrub species such as *Grevillea australis* R. Br. may be facilitated by *N. ovatifolia*, due to pollinator spillover, a well-studied mechanism. However, at high densities, similarities in growth form or flower type could result in competition.

This study addressed the following questions: (1) Does *N. ovatifolia* density impact insect visitation to flowers of other native plant species? (2) Does *N. ovatifolia* flower density impact conspecific and heterospecific pollen receipt on other native flowering plants? (3) Are there plant-pollinator network structural changes along a gradient of *N. ovatifolia* flower density? Whether patterns of pollinator visitation and network structure impact pollen transfer is

another important question, but could not be directly and reliably determined due to low sample size. However, connections between visitation, network structure, and pollen transfer may be inferred through answering the previous questions.

## Methods

### *Spatial and temporal information*

Twelve 3x3 m plots were established in Charlottes Pass (36.255S, 148.19E) in the Australian alpine at Kosciuszko National Park, NSW, at an elevation of approximately 1830m. The plots were dispersed amongst alpine herb field, alpine heath, and subalpine woodland plant communities. *N. ovatifolia* was the focal species of this study. Five locally abundant flowering plants were assessed; *Ranunculus graniticola* and *Pimelea alpina*, both small forbs, *Olearia phlogopappa* (Labill.) DC., a relatively taller herb, and *G. australis*, a mass flowering shrub, similar in form to *N. ovatifolia*. Sampling took place on 4 intervals during early summer, from the 21<sup>st</sup>-24<sup>th</sup> November 2023 from 09:00-15:00PM. Temperature during observations varied between 13-19°C. Flower counts were conducted between the 21<sup>st</sup> and 23<sup>rd</sup>. In plots with a large abundance of flowers, a proportion of flowers were counted and extrapolated. *P. alpina* inflorescences were counted and multiplied by an average number of flowers per inflorescence taken in the field. Density within plots was initially judged qualitatively, selected to maximize variability of different flower abundances. 3 plots with an absence of *N. ovatifolia* were selected.

For the purposes of visualizing network structure at low and high densities of *N. ovatifolia*, plots with less than 500 *N. ovatifolia* were grouped into 'low-density' plots, and above 500 into 'high-density' plots. Low-density plots contained far more *R. graniticola* and far less *P. alpina* flowers compared to high-density plots, in most cases *R. graniticola* grew in meadow-like communities and was therefore not established in the shrub-dominated areas. *G. australis* abundance was more or less uniform (table 1). The density of 500 flowers was chosen so that the quantity of visits was similar in both networks (101 and 119 respectively).

### *Visitation*

Four total observation rounds were completed per plot on separate days. Each round was 5-7 minutes long, the total time spent observing was equal between plots. Time spent observing flowers was equal regardless of the abundance of each plant, so that observed visitation was independent of flowering density.

The timer was paused during handling time. If insects could not be confidently ascertained to at least family level in the field, they were euthanized and identified using a dissecting microscope later with the assistance of a reference collection. An observation was only counted when contact was made with the reproductive parts of the flower.

### *Pollen deposition*

Stigmas or whole flowers were collected in the field and placed in 1.5ml Eppendorf tubes. Between 2 and 5 stigmas were collected for each plant and species in each plot, each plant was assigned a separate Eppendorf. Stigmas were prepared on microscope slides by melting glycerine gel on a slide using a lighter and were then squashed and covered with a coverslip. Stained pollen grains on or nearby stigmas were counted using a compound light microscope at 100-400x magnification.

### Statistical analysis

To assess the impact of *N. ovatifolia* density on insect visitation, plant-pollinator networks are presented for the the whole community (figure 1) and for high and low-density plots respectively (figure 2). Additionally, the proportion of observed visits (normalized degree) was calculated for each plant species in each plot and plotted against *N. ovatifolia* density.

To determine the effect of *N. ovatifolia* density on conspecific and *N. ovatifolia* pollen deposition on plant stigmas, separate GLMMs for both types of pollen deposition were constructed for each plant species, *R. granitica* was excluded due to low sample size. *N. ovatifolia* density and focal plant density were treated as fixed interactive effects, and plot as a random effect. Furthermore, conspecific and *N. ovatifolia* pollen counts were plotted against *N. ovatifolia* density to visualize pollen deposition.

To understand whether *N. ovatifolia* flower density affected plant-pollinator network structure, connectance was calculated for each plot and regressed against *N. ovatifolia* density. However, this approach was limited by sample size, which also prevented calculating other important network metrics such as nestedness. To better visualize changes in network structure due to *N. ovatifolia* density, plots more than and less than 500 *N. ovatifolia* flowers were grouped and the resulting networks displayed.

**Table 1** Flower abundance in different plots, where n is the number of plots sampled. High-density is more than 500 *Nematolepis ovatifolia* flowers, and low-density is less than 500

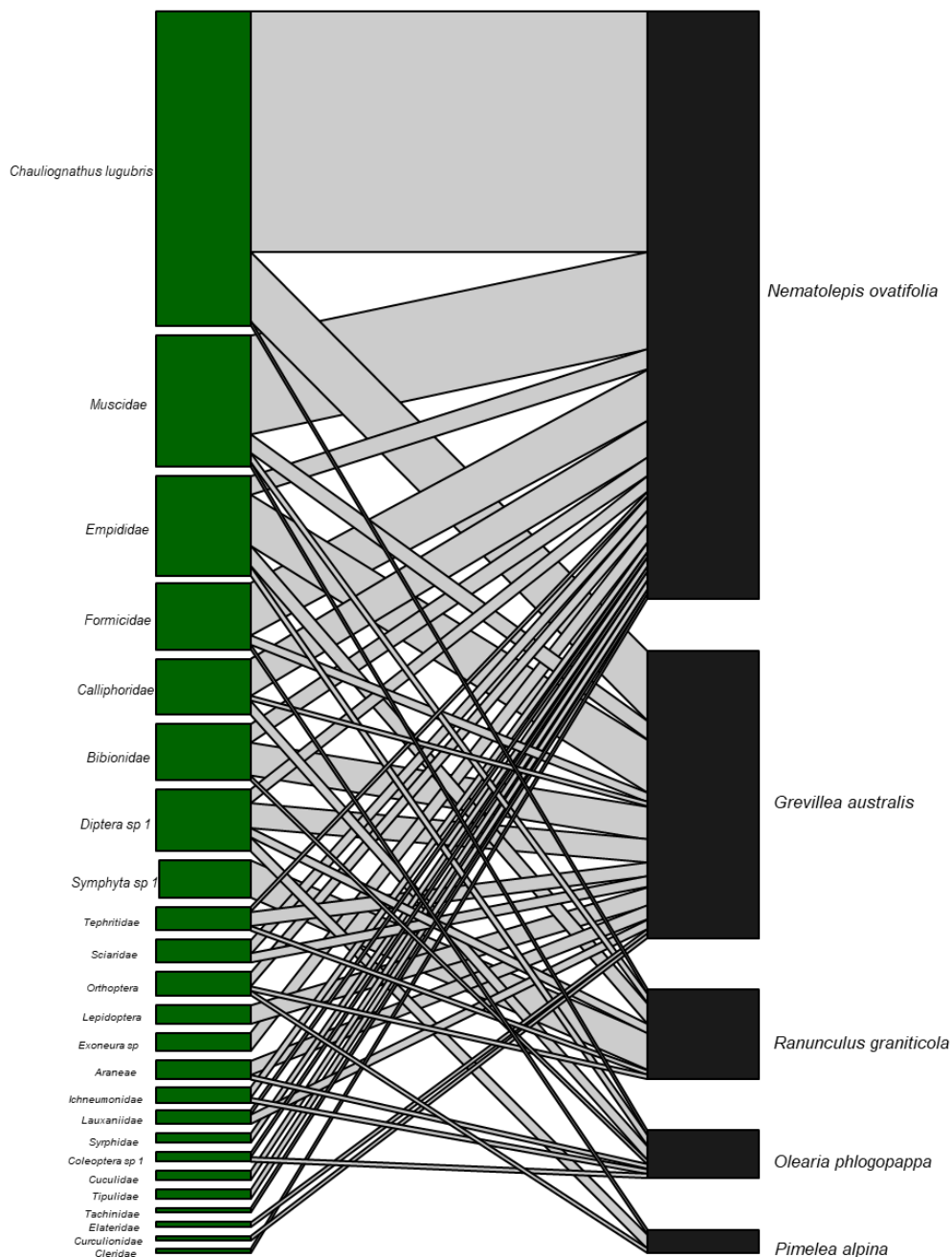
	<i>Nematolepis ovatifolia</i>	<i>Grevillea australis</i>	<i>Ranunculus granitica</i>	<i>Olearia phlogopappa</i>	<i>Pimelea alpina</i>	Observations
Total abundance in high-density plots (n=4)	5384	2307	5	20	1639	101
Total abundance in low-density plots (n=8)	1530	4085	117	133	1571	119

## Results

### *Insect visitation*

There were 220 visits observed in total. Most observations were of the orders Diptera and Coleoptera, the former spanning 11 families, mostly consisting of Muscidae, Empididae, Calliphoridae, and Bibionidae. However, Lepidoptera, Orthoptera, Araneae, and Hymenoptera were also observed contacting flower stigmas. Almost all Coleoptera visits were by *Chauliognathus lugubris*, often found mating during visitation and observed traversing between *G. australis* and *N. ovatifolia*. Notably, Lepidoptera were rarely observed visiting flowers, and bees were largely absent, apart from in plots located in subalpine woodland, where they nest in the deadwood. Most Formicidae observations took place in a single plot which contained a nest.

The community-level network was both highly generalized and nested (figure 1). *Chauliognathus lugubris* was the most abundant visitor, mainly observed visiting *N. ovatifolia* and *G. australis*. On the other hand, only visitors from the order Diptera visited more than 3 plant species.

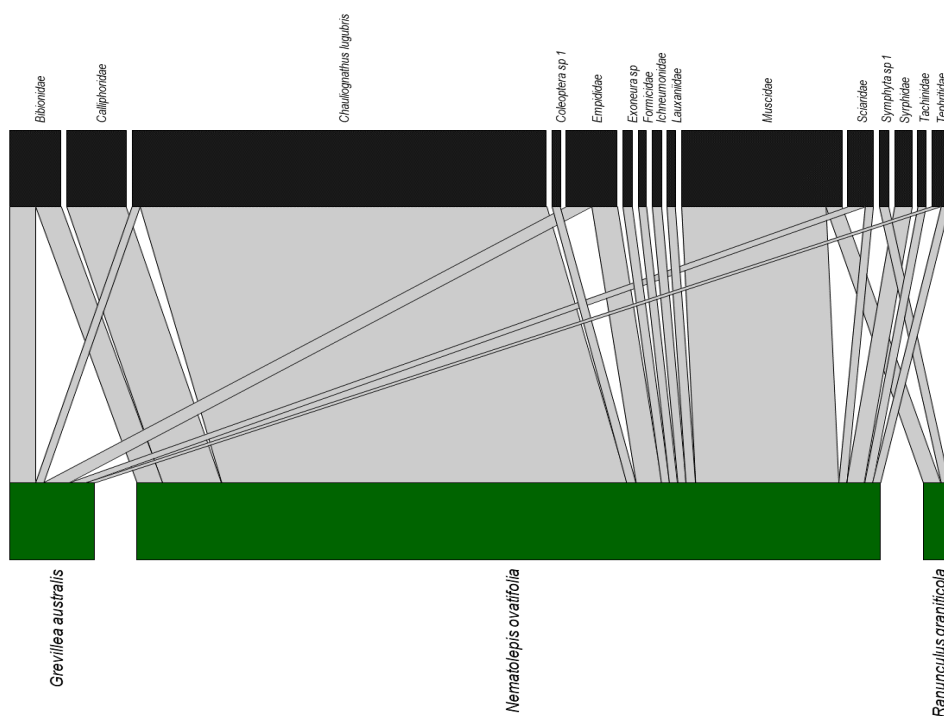


**Figure 1** Community plant-pollinator network for all observations. Thickness of lines indicates number of observed visits, green boxes on the left are plant species, black boxes on the right are insects grouped into taxonomic or functional groups

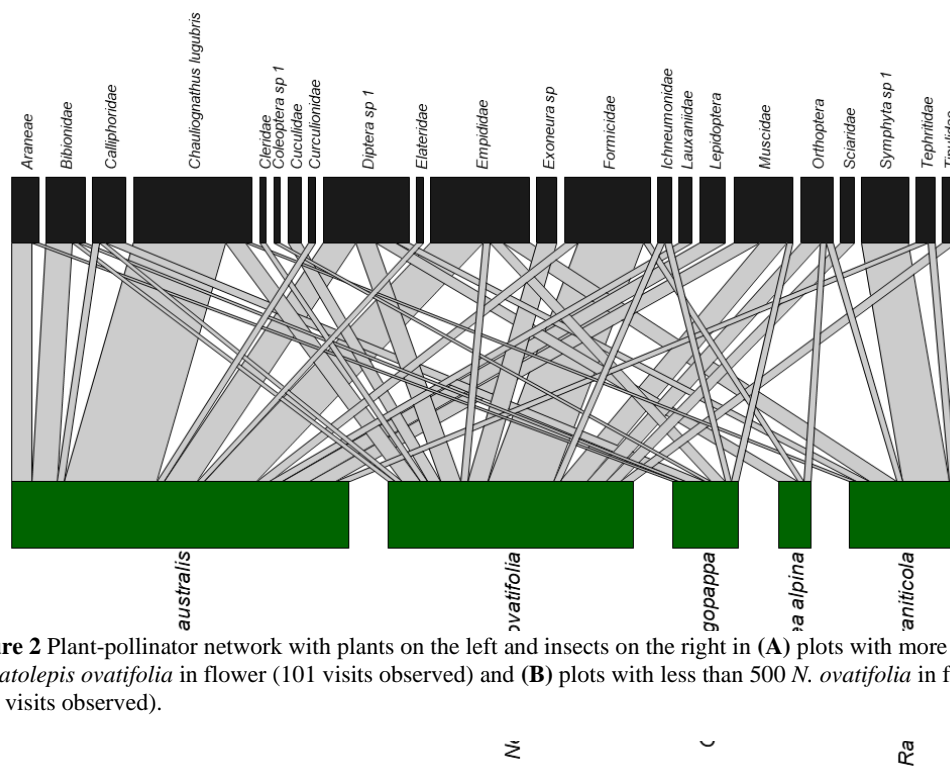
Notably, *Symphyta sp.* was the only species displaying strong specialization, as it was only observed visiting *R. graniticola* (figure 1). Other species with only one link were observed very few times. The two shrub species, *N. ovatifolia* and *G. australis*, were highly generalized and received the most visits (figure 1). Despite counting for 20% of the flowers counted in all plots,

*P. alpina* made up only 2.27% of observations. *O. phlogopappa* was only ever present in relatively small populations and received few visits (table 1; figure 1).

A



B



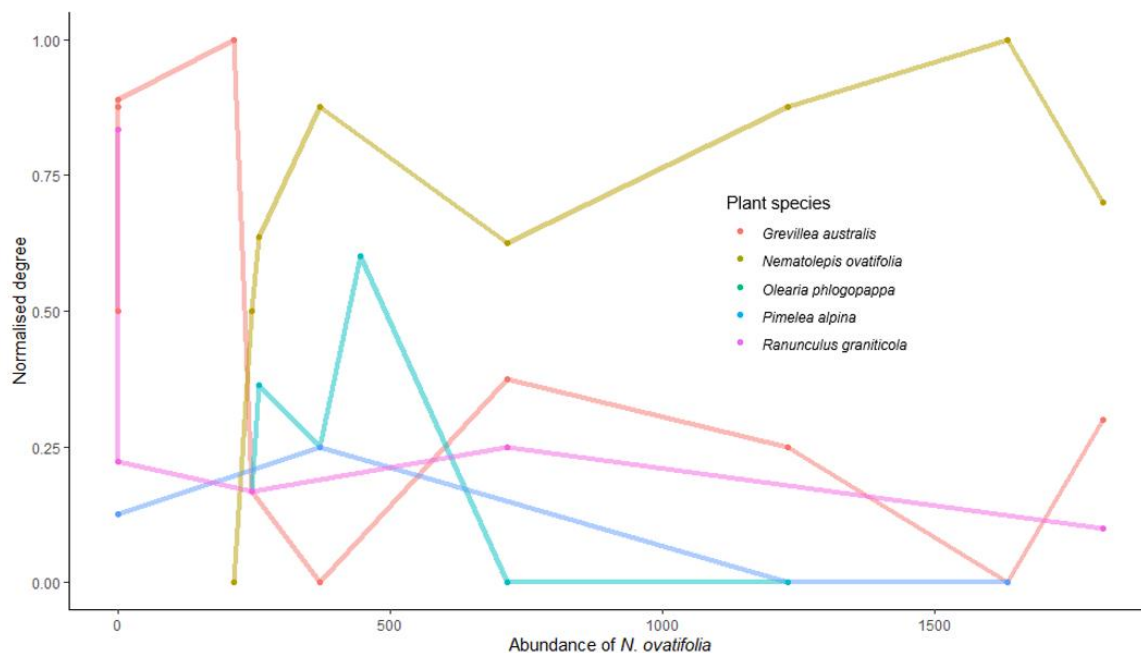
**Figure 2** Plant-pollinator network with plants on the left and insects on the right in (A) plots with more than 500 *Nematolepis ovatifolia* in flower (101 visits observed) and (B) plots with less than 500 *N. ovatifolia* in flower (119 visits observed).



**Table 2** Network and species level metrics calculated for high- and low-density networks, H2 is a network level measure of specialization and normalized degree is the proportion of all animals present in the network found contacting the flower stigma, expressed as a percentage

Metric	High-density network	Low-density network
Number of observations	101	119
Number of plants	3	5
Number of animals	15	24
Normalized degree of <i>N. ovatifolia</i>	0.93	0.79

Visitation in high density plots showed far less overlap compared to low density plots (figure 2). The diversity of visiting species was also lower. This may have been due to there being twice as many low-density plots, however, the cumulative visitation was similar (101 in high density, 119 in low density plots). Additionally, the generalized structure seen in low-density plots was not seen in high density plots, where almost all visitors were observed at *N. ovatifolia* (figure 2). Despite being present at high densities (table 1), *P. alpina* received no visits in high density plots. *O. phlogopappa* was only present in small populations in high density plots and received no visits.



**Figure 3** Number of visits to each plant species as a proportion of total visits (normalised degree) calculated from plots over a range of *Nematolepis ovatifolia* abundances.

The normalized degree of *N. ovatifolia* was far greater in plots with a high density of co-flowering (figure 3). As a result, the normalized degree of co-flowering species was low in plots with high densities of *N. ovatifolia*. This pattern is despite all co-flowering species receiving equal observation periods. At intermediate densities of *N. ovatifolia*, all co-flowering species were still able to maintain normalized degrees exceeding 20%, which would be the expected value if insects visited plants evenly.

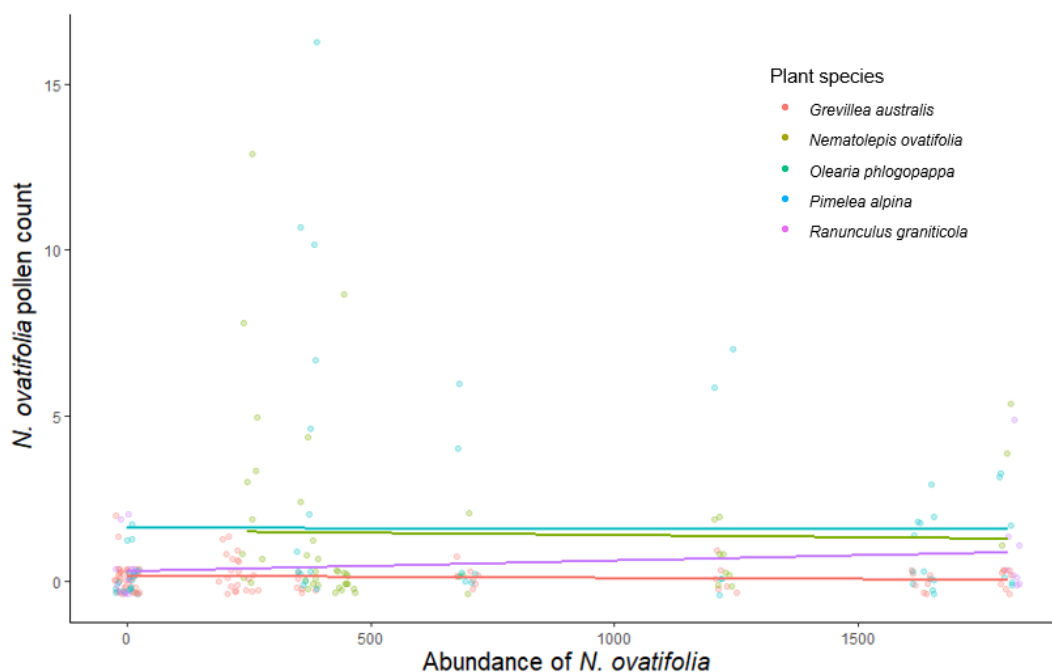


## Pollen deposition

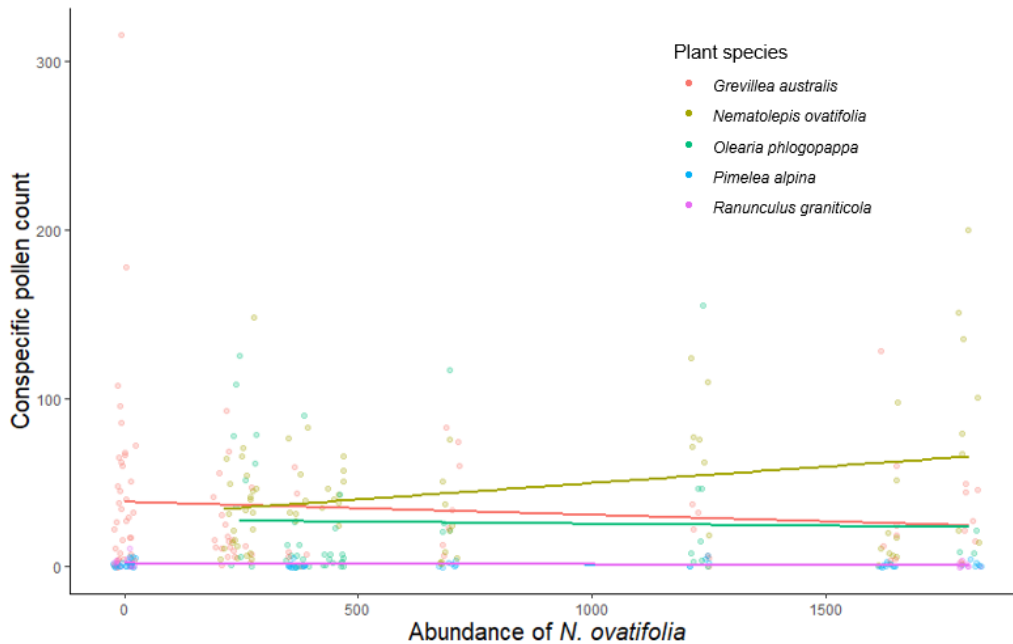
**Table 3** Results of generalized linear mixed effects models where the response variables are conspecific and *Nematolepis ovatifolia* pollen deposition respectively. The interactive fixed effect variables are log-transformed plot-level flower abundances of *N. ovatifolia* and conspecific plant species, and the random effect variable is plot. The final column includes number of stigmas analyzed (N).

Plant species	Conspecific pollen count			<i>N. ovatifolia</i> pollen count			
	Conspecific density	<i>N. ovatifolia</i> density	Interactive effect	Conspecific density	<i>N. ovatifolia</i> density	Interactive effect	N
<i>G. australis</i>	P = 0.26	P = 0.77	P = 0.67	P = 0.44	P = 0.53	P = 0.55	91
<i>P. alpina</i>	P = 0.126	P = 0.21	P = 0.21	P = 0.16	P = 0.19	P = 0.24	61
<i>O. phlogopappa</i>	<b>P = 0.0038 (+)</b>	<b>P = 0.0028 (+)</b>	<b>P = 0.0055 (-)</b>	P = 0.30	P = 0.13	P = 0.35	47
<i>N. ovatifolia</i>	P = 0.21	-	-	-			74

CP deposition varied significantly only for *O. phlogopappa* in response to conspecific and *N. ovatifolia* density, as well as their interactive effects (table 3). No other significant responses were detected due to the fixed variables. Intercept values from models of CP were significantly higher than zero for *N. ovatifolia* ( $P < 0.001$ ) and *O. phlogopappa* ( $P = 0.0058$ ), indicating that, theoretically, CP transfer would occur even with no conspecific flowers in the plot. No significant intercepts were found for the *N. ovatifolia* pollen count models.



**Figure 4** Scatterplot showing how *N. ovatifolia* pollen counts on heterospecific stigmas changed with plot-level abundance of *N. ovatifolia*



**Figure 5** Scatterplot showing how conspecific pollen counts changed with plot-level abundance of *N. ovatifolia*

A GLMM with species and *N. ovatifolia* density as interacting fixed effects and plot as a random effect revealed that *P. alpina* and *R. graniticola* received significantly lower amounts of CP compared to other species ( $P < 0.001$  for both, figure 5). A GLMM with the same structure revealed that *O. phlogopappa* and *P. alpina* received significantly more *N. ovatifolia* pollen than did other species, although no species received more than 3 on average (figure 4).

## Discussion

This analysis of visitation and pollen deposition demonstrates *Nematolepis ovatifolia* density impacted insect visitation but not pollen transfer. Conspecific and *N. ovatifolia* deposition remained similar between plots despite differences in local flowering assemblage. These results demonstrate that co-flowering plants maintain conspecific pollen transfer and avoid heterospecific pollen transfer despite differences in insect visitation.

### Visitation

The community level plant-pollinator network structure observed in this study is reasonably similar to that of others in the area, especially in the large relative abundance of Diptera flower visitors (Inouye and Pyke 1988). Due to the small number of plants surveyed, it is difficult to draw similarities with specific network level metrics found by other studies. That being said, low rates of specialization and a fairly asymmetric network mirrored the findings of similar studies (Encinas-Viso et al. 2022). Blüthgen et al (2008) notably discussed how this type of nested network structure may be an artifact of sample size, as rare-rare interactions are numerically less likely to occur than rare-abundant or abundant-abundant interactions. While it is true that increased sampling would detect more rare interactions, the actual network structure is unlikely to be affected.

The reduced overlap of visitation in the high-density network could be a cause for concern if *N. ovatifolia* or similar plant species are expanding in range, as network complexity is found to relate directly to long term ecological stability (Huang et al. 2021). Because of the spatial and temporal overlap with research conducted recently by Encinas-Viso et al, it is difficult to understand why significantly fewer Lepidoptera visits were recorded in this study. The Lepidoptera were mostly seen perching on leaves rather than visiting flowers. One explanation could be annual variation in climate and small-scale variation in community assemblage in the Australian alpine.

Despite representing twice as many plots, visitation to plots with >500 *N. ovatifolia* in flower had almost the same number of total visits (table 2). One explanation for this pattern is that the diversity and abundance of insect visitors is maximized by the presence of co-flowering plants. This effect is well understood in the context of agricultural systems and would also explain the loss of visitor diversity observed in high-density plots (figure 2; Gilpin et al. 2022). In another study floral density was also found to correlate positively with visitation abundance and diversity (Lázaro et al. 2009). However, the authors also found that the floral density of a focal species generated competitive or facultative interactions between plants resulting in varying effects on the composition of visitors. A clear pattern of increased normalized degree of *N. ovatifolia* was found (figure 3), despite equal observation time being given to co-flowering species. This is unsurprising, given that Diptera, the most diverse and abundant insect group, is generally considered to be an opportunistic flower visitor. For example, it was found that for temperate Syrphid flies, vertical stratification of flowers resulted in foraging further from the ground where there were inflorescences present both closer and further from the ground (Klecka et al. 2018B). Because *P. alpina*, *R. graniticola*, and *O. phlogopappa* tend to flower nearer to the ground than shrubs, it could be expected that *N. ovatifolia* would directly reduce visitation to these plants.

Although improved in more recent research, Coleoptera is an insect order typically neglected in relation to being an efficient pollinator. For example, the study by Inouye and Pyke in 1988 did not consider Coleoptera to be effective pollinators. This is contrary to the network constructed by Encinas-Viso and this study, where *C. ligubris* was found to visit a diversity of flora and in high abundances. During visitation surveys, *C. ligubris* was found to be carrying visible quantities of pollen on their body and an isolated study shows that the species is an effective pollen vector (Gaffney et al. 2018). In general, plant-pollinator research should consider all species as potential pollen vectors, rather than relying on traditionally accepted species such as bees and Lepidoptera. Visible pollen loads were even observed on species of Orthoptera perched in the flowers of *R. graniticola*. *C. ligubris* was often observed mating while collecting pollen and nectar from flowers. Another species observed mating in the presence of flowers was *Symphyta sp. 1*, a sawfly only ever observed on the flowers of *R. graniticola*. Apart from typical floral rewards, heliotropic flowers with ‘satellite’ shaped inflorescences may provide thermal rewards for visitors by redirecting UV light into the center of the inflorescence, especially onto the stigmatic surface (Lamprecht et al. 2007; Orueta 2002). This may explain the preference for *Symphyta sp. 1* to bask in *R. graniticola*. More studies should investigate complex plant-pollinator interactions such as insect mating and thermal reward, and how that may link to the maintenance of specific plant-pollinator mutualisms (Pellmyr and Thien 1986).

On the community level, very few visits to *P. alpina* were observed. Because of its long and tubular flower shape, it could be possible that *P. alpina* is mostly nocturnally pollinated. However, there is quite limited research on the pollination of *P. alpina*, and studies on other

species of *Pimelea* reveal differing results (Blackall et al. 2023; Buxton et al. 2022). Studies of nocturnal pollination are more rare than diurnal studies. However, nocturnal pollination can help fill gaps in diurnal studies, and are necessary to capture the full network of plant-pollinator interactions (Souza et al. 2022). In high density plots, there appeared to be a large increase in some of the more generalist pollinators such as *C. ligubris*, Calliphoridae, and Muscidae, at the expense of other insect groups (figure 2). This raises concern over how a more homogenized plant community structure consisting of an overstory of shrubs may affect insect communities and their ecosystem services. Outcomes of research on the correlation between plant and pollinator communities show evidence both for and against this hypothesis, so further research on general plant-insect community dynamics in KNP would be required (Losapio et al. 2016; Szigeti et al. 2020).

### *Pollen deposition*

*O. phlogopappa* was the only plant species found to have significantly variable CP deposition, where increased CP was significantly increased by conspecific and *N. ovatifolia* floral density. Additionally, CP counts for *O. phlogopappa* decreased as a result of the interactive effects of the two floral abundance variables. Why this pattern occurred is unclear and does not correlate with the findings for pollen transfer in the other plant species. One possible reason is the presence of pollen grains generated by the plant itself, rather than deposited by insects, occurring to the closeness of the stamen and stigma in Asteraceae. A greater number of plots containing both *O. phlogopappa* and *N. ovatifolia* would be needed to attribute these differences as being the result of biologically significant factor, such as increased asexual reproduction in areas of low-conspecific floral density or facilitation due to *N. ovatifolia* density.

The finding that CP counts were largely unaffected by *N. ovatifolia* density is not an uncommon finding. One study assessing the impact of a native dominant flowering plant on other native plants found the same pattern (Hernández-Castellano et al. 2020). The authors found only small differences in the rate of HP deposition explained by dominant plant presence. Interestingly, there was a decline in seed set among one of the native plants. Therefore, any long-term studies on the impact of dominant plants in KNP should measure seed set, which in many cases is not well predicted by pollen deposition (Waites and Ågren 2004; Wang et al. 2017).

Very few CP grains were found on *R. graniticola* and *P. alpina*. The latter species also received very few visits, and its stigma is often secluded within its tubular corolla. Furthermore, its main observed visitor was very small species of Diptera (<3mm), which would be unlikely to carry large pollen loads. In this context, the visitation data was in support of the pollen counts. *R. graniticola* was more successful in terms of attracting pollinators, so the low pollen deposition could be partially explained by the fact that its primary visitor, *Symphyta sp. 1* is a relatively small and hairless insect. However, the species was only witnessed on *R. graniticola* and in fairly abundant numbers, so it is difficult to explain the lack of pollen deposition without measuring single visit deposition data. Additionally, *R. graniticola* stigmas are thick and difficult to manipulate using the stained glycerine slide method, which could have led to miscounting. Acetolysis is a commonly used method for pollen counting which circumvents this issue (Smith et al. 2021). HP deposition was also found to be nonsignificant in other studies and is overall an enigmatic process (Peuker et al. 2020). CP deposition on *N. ovatifolia*, *G. australis*, and *O. phlogopappa* was much higher than *N. ovatifolia* pollen deposition. Because of the low rates of specialization at the community level, it is possible that flowers are significantly more receptive to CP grains and are effectively 'choosing' which pollen they receive from insects. Further

studies are needed into the dynamics of pollen deposition, a complex situation which is not well understood.

## Conclusions

These findings show that plant species pollination services are not sensitive to increased dominance by a native plant. It is possible that this is because the plants evolved in the same community. Furthermore, visitation results did not correlate with pollen deposition, as plants were resilient to *N. ovatifolia* pollen deposition, again possibly due to their shared evolutionary history. As climate change continues to effect plant distributions in alpine environments, longer-term studies on plants responses to these changes, at the functional level if not resolved to species. Further studies on the flow-on effects on pollination services and insect community responses to these changes are also needed, perhaps using seed set as a more accurate assessment of reproductive success. Furthermore, repeated, or improved versions of the methods used here should be applied over longer sampling periods and inclusive of a larger diversity of plant species.

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## References

- Albrecht, M., Ramis, M. R. & Traveset, A. 2016. Pollinator-mediated impacts of alien invasive plants on the pollination of native plants: the role of spatial scale and distinct behaviour among pollinator guilds. *Biological Invasions*, 18, 1801-1812.
- Arceo-Gómez, G., Schroeder, A., Albor, C., Ashman, T.-L., Knight, T. M., Bennett, J. M., Suarez, B. & Parra-Tabla, V. 2019. Global geographic patterns of heterospecific pollen receipt help uncover potential ecological and evolutionary impacts across plant communities worldwide. *Scientific Reports*, 9, 8086.
- Auld, J., Everingham, S. E., Hemmings, F. A. & Moles, A. T. 2022. Alpine plants are on the move: Quantifying distribution shifts of Australian alpine plants through time. *Diversity and Distributions*, 28, 943-955.
- Blackall, A. G., Mackay, D. A. & Whalen, M. A. 2023. Impact of reserve area on reproduction of a moth-pollinated *Stackhousia* Sm. (Celastraceae) species in a fragmented landscape. *Austral Ecology*, 48, 2053-2075.
- Buxton, M. N., Anderson, B. J. & Lord, J. M. 2022. Moths can transfer pollen between flowers under experimental conditions. *New Zealand Journal of Ecology*, 46, 1-5.
- Callaway, R. M. & Walker, L. R. 1997. COMPETITION AND FACILITATION: A SYNTHETIC APPROACH TO INTERACTIONS IN PLANT COMMUNITIES. *Ecology*, 78, 1958-1965.
- Carstensen, D. W., Sabatino, M. & Morellato, L. P. 2016. Modularity, pollination systems, and interaction turnover in plant-pollinator networks across space. *Ecology*, 97, 1298-306.

- Daniels, J. D. & Arceo-Gómez, G. 2020. Effects of invasive *Cirsium arvense* on pollination in a southern Appalachian floral community vary with spatial scale and floral symmetry. *Biological Invasions*, 22, 783-797.
- Encinas-Viso, F., Bovill, J., Albrecht, D. E., Florez-Fernandez, J., Lessard, B., Lumbers, J., Rodriguez, J., Schmidt-Lebuhn, A., Zwick, A. & Milla, L. 2022. Pollen DNA metabarcoding reveals cryptic diversity and high spatial turnover in alpine plant-pollinator networks. *Molecular Ecology*, n/a.
- Essenberg, C. J. 2013. Explaining the effects of floral density on flower visitor species composition. *Am Nat*, 181, 344-56.
- Gaffney, A., Bohman, B., Quarrell, S. R., Brown, P. H. & Allen, G. R. 2018. Frequent Insect Visitors Are Not Always Pollen Carriers in Hybrid Carrot Pollination. *Insects*, 9.
- Gilpin, A.-M., Denham, A. & Ayre, D. 2019. Do mass flowering agricultural species affect the pollination of Australian native plants through localised depletion of pollinators or pollinator spillover effects? *Agriculture, Ecosystems and Environment*, 277, 83-94.
- Gilpin, A.-M., O'brien, C., Kobel, C., Brettell, L. E., Cook, J. M. & Power, S. A. 2022. Co-flowering plants support diverse pollinator populations and facilitate pollinator visitation to sweet cherry crops. *Basic and Applied Ecology*, 63, 36-48.
- Hernández-Castellano, C., Rodrigo, A., Gómez, J. M., Stefanescu, C., Calleja, J. A., Reverté, S. & Bosch, J. 2020. A new native plant in the neighborhood: effects on plant-pollinator networks, pollination, and plant reproductive success. *Ecology*, 101.
- Huang, H., Tu, C. & D'odorico, P. 2021. Ecosystem complexity enhances the resilience of plant-pollinator systems. *One Earth*, 4, 1286-1296.
- Inouye, D. W. & Pyke, G. H. 1988. Pollination biology in the Snowy Mountains of Australia: Comparisons with montane Colorado, USA. *Australian Journal of Ecology*, 13, 191-205.
- Janovský, Z., Mikát, M., Hadrava, J., Horčíčková, E., Kmecová, K., Požárová, D., Smyčka, J. & Herben, T. 2013. Conspecific and Heterospecific Plant Densities at Small-Scale Can Drive Plant-Pollinator Interactions. *PLoS ONE*, 8.
- Kelly, A. E. & Goulden, M. L. 2008. Rapid shifts in plant distribution with recent climate change. *Proceedings of the National Academy of Sciences of the United States of America*, 105, 11823-11826.
- Klecka, J., Hadrava, J., Biella, P. & Akter, A. 2018A. Flower visitation by hoverflies (Diptera: Syrphidae) in a temperate plant-pollinator network. *PeerJ*, 2018.
- Klecka, J., Hadrava, J. & Koloušková, P. 2018B. Vertical stratification of plant-pollinator interactions in a temperate grassland. *PeerJ*, 6, e4998.
- Koorem, K., Kostenko, O., Snoek, L. B., Weser, C., Ramirez, K. S., Wilschut, R. A. & Van Der Putten, W. H. 2018. Relatedness with plant species in native community influences ecological consequences of range expansions. *Oikos*, 127, 981-990.
- Lamprecht, I., Maierhofer, C. & Röllig, M. Infrared thermography and thermometry of phototropic plants. *Journal of Thermal Analysis and Calorimetry*, 2007. 49-54.
- Lázaro, A., Lundgren, R. & Totland, O. 2009. Co-flowering neighbors influence the diversity and identity of pollinator groups visiting plant species. *Oikos*, 118, 691-702.

- Lenoir, J. & Svenning, J. C. 2015. Climate-related range shifts - a global multidimensional synthesis and new research directions. *Ecography*, 38, 15-28.
- Lopes, S. A., Bergamo, P. J., Najara Pinho Queiroz, S., Ollerton, J., Santos, T. & Rech, A. R. 2022. Heterospecific pollen deposition is positively associated with reproductive success in a diverse hummingbird-pollinated plant community. *Oikos*, 2022.
- Losapio, G., Gobbi, M., Marano, G., Avesani, D., Boracchi, P., Compostella, C., Pavesi, M., Schöb, C., Seppi, R., Sommaggio, D., Zanetti, A. & Caccianiga, M. 2016. Feedback effects between plant and flower-visiting insect communities along a primary succession gradient. *Arthropod-Plant Interactions*, 10, 485-495.
- Lundemo, S. 2007. Within-population spatial variation in pollinator visitation rates, pollen limitation on seed set, and flower longevity in an alpine species. *Acta Oecologica*, 32, 262-268.
- Mansfield, L. A., Nowack, P. J., Kasoar, M., Everitt, R. G., Collins, W. J. & Voulgarakis, A. 2020. Predicting global patterns of long-term climate change from short-term simulations using machine learning. *npj Climate and Atmospheric Science*, 3, 44.
- Mcdougall, K., Wright, G. & Peach, E. 2018. Coming to terms with Ox-eye Daisy (*Leucanthemum vulgare*) in Kosciuszko National Park, New South Wales. *Ecological Management and Restoration*, 19, 4-13.
- Muñoz, A. A. & Cavieres, L. A. 2008. The presence of a showy invasive plant disrupts pollinator service and reproductive output in native alpine species only at high densities. *Journal of Ecology*, 96, 459-467.
- Norberg, R. A. 1977. An Ecological Theory on Foraging Time and Energetics and Choice of Optimal Food-Searching Method. *Journal of Animal Ecology*, 46, 511-529.
- Orueta, D. 2002. Thermal relationships between *Calendula arvensis* inflorescences and *Usia aurata* bombyliid flies. *Ecology*, 83, 3073-3085.
- Parra-Tabla, V. & Arceo-Gómez, G. 2021. Impacts of plant invasions in native plant-pollinator networks. *New Phytologist*, 230.
- Pellmyr, O. & Thien, L. B. 1986. Insect Reproduction and Floral Fragrances: Keys to the Evolution of the Angiosperms? *Taxon*, 35, 76-85.
- Penberthy, T. M., Dillon, M. K., Chen, X. & Donaldson-Matasci, M. C. 2023. Honey bee foraging density depends on plant size. *Animal Behaviour*, 206, 39-51.
- Peuker, M. A., Burger, H., Krausch, S., Neumüller, U., Ayasse, M. & Kuppler, J. 2020. Floral traits are associated with the quality but not quantity of heterospecific stigmatic pollen loads. *BMC Ecology*, 20, 54.
- Pickering, C. M., Mount, A., Wichmann, M. C. & Bullock, J. M. 2011. Estimating human-mediated dispersal of seeds within an Australian protected area. *Biological Invasions*, 13, 1869-1880.
- Rathcke, B. 1983. CHAPTER 12 - Competition and Facilitation among Plants for Pollination. In: Real, L. (ed.) *Pollination Biology*. Academic Press.
- Smith, G. X., Swartz, M. T. & Spigler, R. B. 2021. Causes and consequences of variation in heterospecific pollen receipt in *Oenothera fruticosa*. *American Journal of Botany*, 108, 1612-1624.



- Solís-Montero, L., Vega-Polanco, M., Vázquez-Sánchez, M. & Suárez-Mota, M. E. 2022. Ecological niche modeling of interactions in a buzz-pollinated invasive weed. *Global Ecology and Conservation*, 39, e02279.
- Souza, C. S., Oliveira, P. E., Rosa, B. B. & Maruyama, P. K. 2022. Integrating nocturnal and diurnal interactions in a Neotropical pollination network. *Journal of Ecology*, 110, 2145-2155.
- Szigeti, V., Fenesi, A., Soltész, Z., Berki, B. & Kovács-Hostyánszki, A. 2020. Neutral effect of an invasive plant species with specialized flower structure on native pollinator communities. *Biological Invasions*, 22, 3017-3030.
- Taveira, R., Magnusson, W. E. & Hipólito, J. 2023. Heterospecific pollen deposition in understory plants of a forest in central Amazon. *Flora*, 305, 152336.
- Thompson, A. H. & Knight, T. M. 2018. Exotic plant species receive adequate pollinator service despite variable integration into plant–pollinator networks. *Oecologia*, 187, 135-142.
- Valdovinos, F. 2019. Mutualistic networks: moving closer to a predictive theory. *Ecology Letters*, 22.
- Wahren, C. H., Camac, J. S., Jarrad, F. C., Williams, R. J., Papst, W. A. & Hoffmann, A. A. 2013. Experimental warming and long-term vegetation dynamics in an alpine heathland. *Australian Journal of Botany*, 61, 36-51.
- Waites, A. R. & Ågren, J. 2004. Pollinator visitation, stigmatic pollen loads and among-population variation in seed set in *Lythrum salicaria*. *Journal of Ecology*, 92, 512-526.
- Wang, H., Cao, G. X., Wang, L. L., Yang, Y. P., Zhang, Z. Q. & Duan, Y. W. 2017. Evaluation of pollinator effectiveness based on pollen deposition and seed production in a gynodioecious alpine plant, *Cyananthus delavayi*. *Ecology and Evolution*, 7, 8156-8160.
- Young, J.-G., Valdovinos, F. S. & Newman, M. E. J. 2021. Reconstruction of plant–pollinator networks from observational data. *Nature Communications*, 12, 3911.
- Yu, W. B., Li, D. Z. & Wang, H. 2012. Highly efficient pollination by bumblebees ensures seed production in *Pedicularis lachnoglossa* (Orobanchaceae), an early-flowering Himalayan plant. *Journal of Systematics and Evolution*, 50, 218-226.
- Zhu, H., Zou, X., Wang, D., Wan, S., Wang, L. & Guo, J. 2015. Responses of community-level plant-insect interactions to climate warming in a meadow steppe. *Scientific Reports*, 5.