Defensive mutualisms affect plant and ant island biogeography at a global scale

Drin Brown[1,2] and Megan E. Frederickson[1,3]

[1]Department of Ecology and Evolutionary Biology, University of Toronto, 25 Willcocks Street, Toronto, Ontario M5S 3B2, Canada

[2]Author for correspondence: Drin Brown, email: drin.brown@gmail.com

[3]Email: m.frederickson@utoronto.ca

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Novelty statement: Delavaux et al. (2024) claimed that mutualism filters plant diversity on islands, thereby weakening the latitudinal diversity gradient on islands. We re-analyzed their data, adding a new mutualism type: the defensive mutualism between plants with extrafloral nectaries (EFNs) and the ants that protect them from herbivores. By analyzing the geographic distributions of mutualistic plants and their ant partners on islands and mainlands, we found that ant and plant species that interact mutualistically via EFNs are over-represented on oceanic islands compared to other taxa. Thus, counter to Delavaux et al. (2024), this defensive ant-plant mutualism enhances rather than filters species richness on islands. Furthermore, this mutualism does not weaken the LDG on islands.

Abstract

Mutualism can shape biogeographic patterns at macroecological scales. Recently, Delavaux et al. (2024) found that plant mutualisms with mycorrhizae, nitrogen-fixing bacteria, and animal pollinators limit island colonization, weakening the latitudinal diversity gradient (LDG) on oceanic islands. Here, we examine whether ants and plants engaged in a common defensive mutualism mediated by extrafloral nectaries (EFN) are similarly under-represented on oceanic islands and whether this mutualism also weakens the LDG on islands. We used global trait and occurrence databases to compare ant and plant species richness between oceanic islands and their likely source mainlands for taxa that do and do not interact mutualistically via EFNs. The species richness of EFN-visiting ants and EFN-bearing plants positively covary across islands and plants with EFNs and EFN-visiting ants are significantly over-represented on islands, equally so across latitudes. Thus, EFNs facilitate rather than limit colonization on islands, and not all mutualisms dampen the LDG on islands.

Introduction

Reciprocally beneficial species interactions, or mutualisms, can shape biodiversity at macroecological scales. Mutualism can enlarge a species' realized niche (Fowler et al., 2023) or geographic range (Afkhami et al., 2014, Nathan et al., 2023) and may be especially likely to facilitate range expansion if the interaction is generalized (Harrison et al., 2018), because local species may cooperate with the immigrating species and encourage its establishment (Chomicki et al., 2019). However, in specialized, obligate mutualisms, both partners may need to be present to establish in a new location, potentially limiting colonization opportunities for mutualist species (Simonsen et al., 2017, Delavaux et al., 2024). Here, we examined how defensive

ant-plant mutualisms affect species richness on oceanic islands, where mutualism has been proposed to act as a strong filter on biodiversity (Delavaux et al., 2021, 2022, 2024).

Species richness on islands depends on the balance among colonization, speciation, and extinction or emigration, with island size and isolation from mainland sources of colonists being the primary determinants of species richness on islands (MacArthur and Wilson, 2001, Delavaux et al., 2021). However, biotic interactions may also limit colonization on islands, particularly on oceanic islands that were never part of a continent (Delavaux et al., 2024). Recently, Delavaux et al. (2021, 2022, 2024) found that mutualists have a reduced likelihood of colonizing oceanic islands, potentially because of a scarcity of compatible partners.

Specifically, Delavaux et al. (2021, 2022, 2024) found that plants that associate with mycorrhizal fungi, nitrogen-fixing bacteria (i.e., rhizobia), and animal pollinators are more under-represented on islands than plants that lack these mutualistic associations (but see Pichler and Hartig, 2024), with important consequences for island ecosystems and biodiversity. For example, because biological nitrogen fixation is the primary means by which nitrogen enters the biosphere from the atmosphere, if the symbiosis between plants and N-fixing bacteria prevents these species from colonizing islands, island soils may not receive the same inputs of fixed N as mainland soils (Delavaux et al., 2022). Furthermore, although species richness generally increases from the poles to the equator, a pattern known as the latitudinal diversity gradient (LDG, Hillebrand, 2004; Kinlock et al., 2018), Delavaux et al. (2024) found that the mutualism filter on island species richness was strong enough to flatten the LDG on oceanic islands. In other words, because of mutualism, islands showed a different global pattern of biodiversity than mainlands (but see Pichler and Hartig, 2024). However, previous studies have focused only on nutritional and pollination mutualisms.

Here, we study defensive ant-plant mutualisms mediated by extrafloral nectaries (EFNs). Well over 4000 plant species produce EFNs, which are glands on plant parts other than flowers (Weber and Keeler, 2013). EFNs secrete sugar-rich nectar that attracts mainly ants and sometimes other arthropods that defend the plant from arthropod herbivores (Marazzi et al., 2013). Multiple meta-analyses across plant species, families, and biogeographic regions have shown that EFNs generally reduce herbivory and increase plant survival, growth, or reproductive success (Chamberlain and Holland 2009, Rosumek et al. 2009, Trager et al. 2010, Leal et al. 2023). Ant-plant interactions mediated by EFNs are thus a type of facultative and generalized mutualism—one in which the presence of the partner is usually not required for plant recruitment and partners are highly interchangeable (Marazzi et al., 2013). Globally, EFN-bearing plants follow the LDG (Luo et al., 2023). Phylogenetic studies have also shown that in general, the origination of EFNs in a lineage promotes diversification (Weber and Agrawal, 2014). This increase in diversity is expected because the presence of EFNs promotes population growth by providing defense, which can result in greater ecological opportunity leading to adaptive radiation. By the same token, EFNs may also accelerate plant range expansion (Nathan et al., 2023). In particular, more EFN-bearing plants are legumes than any other plant family, and EFN-bearing legumes have spread to more new regions of the globe than legumes that do not have EFNs (Nathan et al. 2023). Thus, unlike the results of Delavaux et al. (2024) for plants that associate with mycorrhizae, rhizobia, and animal pollinators, we predicted that EFN-bearing plants may experience enhanced colonization on islands (Chomicki et al., 2019). If so, then counter to Delavaux et al. (2024), mutualism does not always filter species richness on islands in ways that weaken the LDG.

Materials and methods

We used five datasets to determine whether EFN-bearing plants and their EFN-visiting ant partners are over- or under-represented on oceanic islands compared to mainlands. All analyses were performed in R Studio using R version 4.4.2 (R Studio R Core Team, 2024). We used the following datasets: the Global Inventory of Floras and Traits (GIFT) database (Weigelt et al., 2020) for plant occurrence checklists, plant trait data (e.g., plant habit and life history), and geographic information, the World List of Plants with Extrafloral Nectaries (Weber et al., 2024) for plant EFN trait status, the Global Ant Biodiversity Index (GABI) database (Guenard et al., 2017) and the Global Ant Biodiversity Index-Islands (GABI-I) database (Liu et al., 2023) for ant occurrence and geographic information, and data from Kaur et al. (2019) on which ant species visit EFNs. These data sources are described in more detail in the supplemental appendix.

We fit two types of models to the data: 1) species-level models in which each data point is a species, and the (binary) response variable is whether it lives on islands, and 2) island-level models in which each data point is an island, and the (continuous) response variable is the log response ratio of island species richness to mainland species richness. We modelled the data two ways because the species-level model allowed us to account for phylogeny and other traits like plant habit and life history, but not island characteristics, while the island-level model allowed us to account for the classical predictors of species richness on islands like island size and isolation, but not phylogeny and species-level traits.

For the species-level plant model, we used data from Delavaux et al. (2024), which included a list of oceanic islands, vascular plant checklists, and trait data on associations with animal pollinators, nitrogen-fixing symbionts, and mycorrhizae. We added EFN trait status from Weber et al. (2024). We treated island presence as a binary trait, such that each species either

occurs on any oceanic island or exclusively on mainlands. To account for the phylogenetic non-independence of species, we built a phylogeny of all vascular plants in the GIFT checklists using the R package V.Phylomaker2 (Jin & Qian, 2022) and then fit a phylogenetically adjusted GLM with island presence as the response variable and EFN status, mycorrhizal association, nitrogen fixation, biotic pollination, plant habit, and plant life history as predictors. We also fit a species-level model for ants examining island presence as a function of EFN association, using the ant phylogeny from Nelsen et al. (2018). For both species-level models, we used the phylogenetically adjusted logistic regression method of Ives and Garland (2010) as implemented in the phyloglm package in R.

Next, for the island-level models, we needed to compare species richness between oceanic islands and mainland sources of colonizing species. Delavaux et al. (2024) interpolated mainland species richness using only absolute latitude as a predictor and then compared island and estimated mainland species richness. Pichler and Hartig (2024) re-analysed the same data using random forest models parameterized with either latitude alone or latitude and longitude to predict mainland species richness; however, they found no support for mutualism dampening the LDG on oceanic islands with these models (but see Delavaux et al., 2025). Thus, instead of using a model to predict mainland species richness, we compared species richness on oceanic islands with the observed, pooled species richness of their five most likely source mainlands, following König et al. (2021) to identify likely source mainlands. We then measured the difference in richness of EFN-bearing plants and EFN-visiting ants on oceanic islands compared to their likely source mainlands.

To obtain a set of oceanic islands, we used data in Delavaux et al. (2024), which used the GIFT R package (Denelle et al., 2023), to extract geographic data from the GIFT database,

including information on island geological origins, which distinguishes oceanic from non-oceanic islands. Following Delavaux et al. (2024), we analyzed only oceanic islands because they were not originally part of a mainland, so they represent unique archives of evolution and colonization. To infer source mainlands for each island, we followed König et al. (2021), who modelled species turnover as a function of environmental and geographic distance between islands and mainlands. For plants, we constrained the possible source mainlands to those with vascular plant checklists available in GIFT, and for ants, we use GABI polygons as possible source mainlands. We pooled the species in the five most likely source mainlands for each oceanic island. To determine which plant species have EFNs, we joined GIFT plant checklists with data on EFN presence from Weber et al. (2024) by species name, after first resolving to best taxonomic match following Nathan et al. (2023). We assumed that plants not in the Weber et al. (2024) list did not have EFNs, as this list is the most complete in the literature to date; for comparison, 1.5% of the plant species in GIFT have EFNs (Figure 1), while Weber and Keeler (2013) estimate that EFNs occur in 2-3.6% of all flowering plant species. The result was a list of geographic areas, either island or mainland, with counts of plant species present, either with or without EFNs.

For ants, we joined ant occurrence data from GABI and GABI-I to the geographic data in GIFT. Ant occurrence data in GABI-I had latitudes and longitudes that sometimes differed from the coordinates supplied for islands in GIFT. We used the sf package in R to generate a distance matrix between each island as represented in GIFT and in GABI-I, using the minimum distance to pair GABI-I and GIFT islands, and then used a 1km distance cutoff for a pair to be treated as the same island. In four cases, there were islands appearing twice in GABI-I with the same coordinates but different names. In these cases, the islands with fewer occurrence records were

dropped. Finally, we joined the GABI and GABI-I data with data from Kaur et al. (2019) to obtain trait data on whether each ant species did or did not visit EFNs, and we grouped each oceanic island with a pool of its five most likely source mainlands, again following the modelling approach of König et al. (2021).

The final dataset was a list of island-mainland groups, with plant and ant species lists, and whether the plants possessed EFNs, or the ants visited EFNs. For both plants and ants, we used species occurrence records from the Global Biodiversity Information Facility (GBIF) to filter out species that are very rare (< 100 GBIF occurrences), in order to exclude understudied species in which the presence or especially the absence of a mutualistic trait might be unreliable (Chamberlain et al., 2022). When testing for a mutualism filter on island colonization (as in Delavaux et al., 2024), the most relevant species on islands are those present in their likely source mainlands. As such, we required that for an island species to be counted, it had to be present in at least one of that island's five most likely source mainlands. This excluded island endemics, which likely originated via speciation on islands and not as colonists from mainlands, as well as island species that arrived from mainlands not captured in our source pool for each island; for comparison, we also include models with all island species in the supplemental material (Table S1 and Figures S1 and S2). For each island, we calculated the log response ratio (LRR) of island to source mainland species richness for plants with and without EFNs and ants that do or not visit EFNs.

We fit generalized linear models (GLMs) separately for the ant and plant trait data. The response variable was the LRR between mainland and island species richness. Because log response ratios can be calculated only from positive, non-zero values, we added a constant of 1 to all count data. The main predictor of interest was whether plants have EFNs, or ants visit

EFNs. Additional biogeographical and climatic variables in the models included absolute latitude of the island in each island-mainland group, average distance from island to source mainlands, island area and elevational range, average mainland area and elevational range, and mean annual precipitation on the island. We log-transformed all geographic variables to improve normality of the residuals.

Finally, to examine the relationship between plant and ant species richness, we also fit separate GLMs with the island species richness of either EFN-visiting or non-EFN-visiting ants as response variables and the species richness of EFN-bearing and non-EFN bearing plants, as well as biogeographical variables, as predictors.

Results

To describe the final plant dataset, we visualized how often the different mutualistic traits (i.e., biotic pollination, mycorrhizae, nitrogen-fixing symbionts, and extrafloral nectaries that attract ants) occur among island and mainland plants, dividing plant species into three groups: plants that are endemic to one or more oceanic islands, plants that occur on both one or more oceanic islands and at least one mainland, or plants found only on mainlands ("endemics", "colonists", and "mainland" species in Figure 1, respectively). Among the 77,874 plant species in GIFT with appropriate trait data, island colonists (species that occur on both islands and mainlands) are more likely to be pollinated by abiotic agents (especially wind), to be non-mycorrhizal, and to lack nitrogen-fixing symbionts than mainland-only plant species, recapitulating the general finding of Delavaux et al. (2024) that these plant mutualisms are less well represented on oceanic islands than on mainlands (Figure 1). However, island colonists are more likely, not less likely, to

have EFNs (Figure 1). Interestingly, island endemics are more likely to be biotically pollinated, mycorrhizal plants that lack EFNs and nitrogen-fixing symbionts than island colonists (Figure 1).

Figure 1 also reveals that island colonists are more often herbaceous annuals than either island endemics or mainland-only plant species, meaning it is important to control for plant habit and life history when modelling the influence of plant mutualisms on island colonization. We fit a species-level model to the plant data that included these traits and the four mutualism-related plant traits, and confirmed that, indeed, plants on oceanic islands are significantly more likely to be herbaceous annuals, compared to woody or perennial plants (Table 1). In this model, EFN-bearing plants were also significantly more likely to occur on oceanic islands than plants without EFNs (Table 1), as were plants with arbuscular mycorrhizal fungi; for AMF, this is the opposite of what Delavaux et al. (2024) found, but they did not account for plant habit and life history. Nonetheless, consistent with Delavaux et al. (2024), biotically pollinated plants were significantly under-represented on islands, compared to abiotically pollinated plants (Table 1, Figure 2). There was no significant effect of nitrogen-fixing symbionts on island presence (Table 1, Figure 2). In the ant species-level analysis, EFN-visiting ants were also significantly more likely to occur on oceanic islands than ant species that do not visit EFNs (Table 1). There was also phylogenetic signal for whether ant and plant species occur on islands, with ants having stronger phylogenetic signal for insularity than plants (see Table 1 for alphas).

The mainland-island grouping procedure yielded 169 groups (of one oceanic island and five likely source mainlands) for plants (Figure 3A) and 178 for ants (Figure 4A). Log response ratios of island to mainland species richness were always negative, reflecting the well-known deficit of species on islands relative to mainlands. In the island-level plant model, EFN-bearing plants were again over-represented on oceanic islands (Table 2A, Figure 3B); the LRR of island

to mainland species richness was less negative for plants with than without EFNs. For plants, islands that were farther from their source mainlands, smaller islands, islands with smaller source mainlands, and drier islands all had larger species deficits compared to mainlands (i.e., more negative LRRs of island to mainland species richness) (Table 2A). There was also a significantly positive main effect of latitude, meaning that tropical islands are more plant species-poor relative to tropical mainlands than are temperate islands relative to temperate mainlands (Table 2A, Figure 3B). However, there was no significant interaction between latitude and EFN status, indicating that the effect of EFNs on island plant species richness does not vary with latitude (Table 2A, Figure 3B).

In the island-level ant model, EFN-visiting ants were also over-represented on islands (Table 2B, Figure 4B); the LRR of island to mainland species richness was less negative for ants that visit EFNs than for ants that do not visit EFNs. Island area was the only other significant predictor in the island-level ant model, again with smaller islands having a larger species deficit, as evidenced by a more negative LRR of island to mainland species richness (Table 2B). Just like for plants, there was no significant interaction between island latitude and EFN status for ants, meaning that the effect of visiting EFNs on ant species richness does not vary latitudinally (Table 2B, Figure 4B).

Because island endemics differ from island colonists in mutualistic traits (Figure 1), in Table 2, we modelled the log response ratio of island to mainland species richness including only island species that also occur on a corresponding mainland and likely arrived from there as colonists. When we included all island species, adding island endemics and island species originating from beyond the most likely source mainlands, a significantly positive effect of EFN trait status remained for ants, but not plants (Table S1, Figures S1 and S2). Nonetheless, EFN

trait status never interacted significantly with latitude. In other words, regardless of modelling approach, ant-plant mutualisms mediated by EFNs do not weaken the latitudinal diversity gradient on islands for either ants or plants.

Across oceanic islands, the species richness of island ants that visit EFNs was correlated with the species richness of plants with EFNs, but not with the species richness of plants that do not have EFNs (Table S2A). In contrast, the species richness of ants that do not visit EFNs was significantly correlated with the species richness of non-EFN-bearing plants, but not with the richness of plants with EFNs (Table S2B). For both EFN-visiting and non-EFN visiting ants, species richness followed the typical latitudinal diversity gradient, and there was higher ant species richness on larger islands. For EFN-visiting ants only, more distant islands also had fewer ant species.

Discussion

Ant-plant mutualisms mediated by EFNs are generalized and facultative (Marazzi et al., 2013), and previous work has shown that this mutualism facilitates plant and ant introductions to new ranges (Nathan et al., 2023) and spurs plant diversification (Weber and Agrawal, 2014). As such, we expected that, contrary to Delavaux et al. (2024), this mutualism type would not act as a filter on the diversity of oceanic islands, and might instead enhance island colonization from mainlands. The results of both the species- and island-level models support our prediction; plants that have EFNs and ants that visit EFNs are over-represented on oceanic islands, compared to taxa lacking these traits. Furthermore, the positive effect of this ant-plant mutualism on island colonization does not vary latitudinally, meaning that it neither intensifies nor weakens the LDG on islands. Finally, the richness of EFN-visiting ants and EFN-bearing plants positively covary

across oceanic islands, suggesting that the mutualistic ant and plant taxa that interact via EFNs either respond in similar ways to climatic or other environmental variation among islands, or are subject to positive feedback fueled by the mutualism itself.

The raw proportions of mutualistic versus non-mutualistic taxa vary among island endemics, colonists, and mainland plant species in ways that are consistent with Delavaux et. al (2021, 2022, 2024); colonists are less likely than mainland plant species to have animal pollinators, mycorrhizae, and rhizobia (Figure 1). However, colonists are more likely than mainland species to have EFNs (Figure 1), suggesting that not all mutualism types affect island colonization equally. Of course, mutualism does not evolve independently of other functional traits, making it challenging to disentangle the effects of mutualism from the effects of correlated traits on island biogeography. For example, mycorrhizal associations are not equally common across plant growth forms; non-mycorrhizal plants are more often herbaceous, compared to ectomycorrhizal and arbuscular mycorrhizal plants, which are more often woody trees or shrubs (Soudzilovskaia et al., 2020). In contrast to the study by Delavaux et al. (2024), which fit only island-level models that did not account for other plant traits beyond mutualism, we fit a species-level model that included not only mutualism-related traits, but also plant habit and life history as covariates. Unsurprisingly, these covariates significantly predicted insularity; herbaceous, annual plants are over-represented on islands, compared to woody, perennial plants (Figure 1, Table 2). After controlling for plant habit and life history, biotic pollination still had a negative effect on whether plant species live on islands, as in Delavaux et al. (2024), but the effect of nitrogen-fixing symbionts was non-significant, and the effects of arbuscular mycorrhizal fungi and EFNs were significantly positive (Table 2, Figure 2); plants with EFNs and arbuscular mycorrhizal fungi are significantly more likely, not less likely to occur on on

oceanic islands than non-EFN and non-mycorrhizal plant species. These findings cast doubt on the generality of the mutualism filter on island plant diversity reported by Delavaux et al. (2024).

In the species-level model, biotic pollination still significantly reduced the likelihood that a plant species lives on islands, compared to abiotic pollination, and indeed Delavaux et al. (2024) found that biotic pollination results in the largest island species deficit among the mutualism types they examined. Delavaux et al. (2024) ascribe this mutualism filter on island colonization to "the reduced likelihood of simultaneous co-colonization of partners" on islands. However, this mechanism should mainly affect plants that are highly specialized on a specific taxon of animal pollinator, whereas most plant-pollinator interactions are highly generalized and dynamic (Waser et al., 1996). Mutualisms can also re-assemble even if colonization by the partners is not simultaneous (Hembry and Tocora, 2025). Nonetheless, the idea that animal pollination is less reliable on islands than on mainlands is old and has solid empirical support; both selfing and wind pollination can be favored on islands (Barrett 1996). Wind pollination may be favored not only because of a dearth of animal pollinators, but also because open, wind-swept coastal habitats on islands favor plant groups, like grasses, in which wind pollination is widespread (Barrett et al. 1996, Friedman and Barrett, 2009).

Delavaux et al. (2024) did not distinguish between taxa that arrived on islands from elsewhere and taxa that have radiated on islands–i.e., whether a plant species has allochthonous or autochthonous origins. For the island-level models, we excluded potentially autochthonous island endemics because they do not give information about the limits to colonization; instead they may reflect the evolutionary drivers on the island itself, although endemics still evolve from successful colonists, and so may retain their traits. Comparing the traits of island endemics and colonists, as in Figure 1, reveals differences in their mutualism-related traits; mutualism may

affect the process of dispersing to and establishing on an island differently from how mutualistic traits and taxa subsequently evolve on islands. For example, while reduced dependence on animal pollinators might favor island colonization, adaptive radiation and diversification on islands might depend on 'escaping homozygosity' by outcrossing (Barrett 1996); this is consistent with the greater proportion of biotically pollinated plants among island endemics than island colonists in Figure 1. When we observe a species living on islands as having particular traits, it reflects the end result of multiple processes, including dispersal, successful establishment in the face of abiotic factors and biotic interactions, and subsequent adaptation and speciation on islands (Losos and Ricklefs, 2009), that are difficult to capture in coarse comparisons of the traits of island and mainland taxa. Mutualism is likely to impact many of these processes, shaping island faunas and floras.

We also added a new mutualism type to the Delavaux et al. (2024) analysis: the mutualism between plants with EFNs and their ant defenders. Although this mutualism type is much rarer among plants than biotic pollination (Figure 1), these two mutualisms have similar effect sizes on insularity (Table 1), meaning that they have almost equivalent effects on shaping island floras, albeit in opposite directions. In other words, in the species-level model, a biotically pollinated plant with EFNs is about equally likely to live on islands as an abiotically pollinated plant without EFNs. Similarly, in the island-level models, mutualism via EFNs positively predicts both ant and plant species richness on islands. Afkhami et al. (2014) proposed that defensive mutualisms, by protecting organisms from biotic interactions with negative fitness consequences, might facilitate range expansion, and Nathan et al. (2023) found evidence of this pattern in legumes that have EFNs and ant defenders. Establishment on an island involves both colonization and niche expansion, and generalist mutualists, such as in defensive ant-plant

mutualisms mediated by EFNs, may be more likely to undergo both the expansion and ecological release phases of species establishment (Chomicki et al., 2019). Alternatively, rather than helping taxa colonize islands, species engaged in ant-plant mutualisms might be overrepresented on islands because they have larger population sizes and therefore reduced extinction rates (Weber and Agrawal, 2014).

Because there are few plant taxa with some combinations of mutualistic traits (e.g., very few plants have no mutualisms at all), we did not explore the interactive effects of multiple mutualisms on insularity or island species richness. However, plants very commonly participate simultaneously or sequentially in multiple mutualisms, which can have non-additive ecological or evolutionary effects (Afkhami et al., 2014, Laurich et al., 2023). For example, legumes with extrafloral nectaries that attract ants may also have some combination of rhizobia, mycorrhizae, and animal pollinators (Nathan et al., 2023). Even if biotic pollination constrains island colonization, the net result of multiple mutualisms on insularity might be positive, given that the positive effects of EFNs and mycorrhizae on insularity swamp the negative effect of biotic pollination in Table 1. However, multiple mutualisms could also result in non-additive effects on island species richness that are difficult to predict.

Mutualism can beget or hinder species diversity in ecological communities, with effects often depending on whether the benefits of mutualism are private to only one or a few community members, or widely available to many species (Bshary 2003, Frederickson et al., 2005, Rudgers and Clay, 2008). We examined the correlation between ant and plant species richness across oceanic islands, and found that the species richness of EFN-visiting ants was significantly predicted by the species richness of EFN-bearing plants, but not by plant diversity more generally. In contrast, the species richness of ants that do not visit EFNs was significantly

predicted by non-EFN plant richness, but not the richness of EFN-bearing plants. These correlations suggest that ant and plant species that engage in mutualism via EFNs either respond more similarly to environmental variation across islands than, for example, EFN-visiting ants and non-EFN plant species, or that the establishment of one mutualist on an island accelerates the establishment of other mutualists in a process akin to invasional meltdown (Simberloff and Von Holle, 1999, Prior et al., 2025). However, the benefits of this kind of positive feedback appear restricted to plant and ant species that directly participate in the mutualism. In a global biogeographic analysis of ant-plant mutualisms, Luo et al. (2023) also found that the presence of potential ant partners was a driver of EFN-bearing plant species richness, suggesting that the pattern we observed might hold on both islands and mainlands.

Understanding what factors shape biodiversity on islands is critical because islands have unique biotas and island species are more vulnerable to extinction than mainland species (Losos and Ricklefs, 2009). We wholeheartedly agree with Delavaux et al. (2024) that mutualism plays an underappreciated role in island biogeography, but our findings suggest that mutualism does not universally filter island species richness, weakening the LDG. Instead, the facultative, generalized mutualism between ants and EFN-bearing plants is associated with increased insularity and island species richness. Furthermore, after accounting for phylogeny and other plant functional traits, only biotic pollination, and not mutualism with mycorrhizae or nitrogen-fixing symbionts, appears to reduce plant colonization of oceanic islands. Because mutualism links the fates of interacting species, mutualism makes key contributions to global biodiversity patterns, but most plant mutualisms do not prevent species from living on tropical islands as Delavaux et al. (2024) claimed.

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References

Afkhami, M. E., McIntyre, P. J., & Strauss, S. Y. (2014). Mutualist‐mediated effects on species' range limits across large geographic scales. *Ecol. Lett.*, 17(10), 1265-1273.

Afkhami, M. E., Rudgers, J. A., & Stachowicz, J. J. (2014). Multiple mutualist effects: conflict and synergy in multispecies mutualisms. *Ecology*, 95(4), 833-844.

Barrett, S. C. H. (1996). The reproductive biology and genetics of island plants. *Philos. Trans. R. Soc. B.*, 351(1341), 725-733.

Bshary, R. (2003). The cleaner wrasse, Labroides dimidiatus, is a key organism for reef fish diversity at Ras Mohammed National Park, Egypt. *J. Anim. Ecol.*, 72(1), 169-176.

Chamberlain, S. A., & Holland, J. N. (2009). Quantitative synthesis of context dependency in ant–plant protection mutualisms. Ecology, 90(9), 2384-2392.

Chamberlain, S., Oldoni, D., & Waller, J. (2022). rgbif: interface to the global biodiversity information facility API.

Chomicki, G., Weber, M., Antonelli, A., Bascompte, J., & Kiers, E. T. (2019). The Impact of Mutualisms on Species Richness. *TREE*, 34(8), 698-711.

Delavaux, C. S., Crowther, T. W., Bever, J. D. & Gora, E. M. (2025). Response to comment on "Mutualisms weaken the latitudinal diversity gradient among oceanic islands. *arXiv preprint arXiv:2411.15105*

Delavaux, C. S., Crowther, T. W., Bever, J. D., Weigelt, P., & Gora, E. M. (2024). Mutualisms weaken the latitudinal diversity gradient among oceanic islands. *Nat.*, 627(8003), 335-339.

Delavaux, C. S., Crowther, T. W., Bever, J. D., Weigelt, P., & Gora, E. M. (2024). Data from: *Mutualisms weaken the latitudinal diversity gradient among oceanic islands*. GitHub Digital Repository. Available at: https://github.com/c383d893/IslandLDG.

Delavaux, C. S., Weigelt, P., Dawson, W., Essl, F., van Kleunen, M., König, C. *et al*. (2021). Mycorrhizal types influence island biogeography of plants. *Commun. Biol.*, 4(1), 1128-1128.

Delavaux, C. S., Weigelt, P., Magnoli, S. M., Kreft, H., Crowther, T. W., & Bever, J. D. (2022). Nitrogen-fixing symbiotic bacteria act as a global filter for plant establishment on islands. *Commun. Biol.*, 5(1), 1209-1209.

Denelle, P., Weigelt, P., & Kreft, H. (2023). GIFT—An R package to access the Global Inventory of Floras and Traits. *Methods Ecol Evol*, 14(11), 2738-2748.

Etienne, R. S., Cabral, J. S., Hagen, O., Hartig, F., Hurlbert, A. H., Pellissier, L. *et al*. (2019). A minimal model for the latitudinal diversity gradient suggests a dominant role for ecological limits. *Am. Nat.*, 194(5), E122-E133.

Fowler, J. C., Donald, M. L., Bronstein, J. L., & Miller, T. E. (2023). The geographic footprint of mutualism: How mutualists influence species' range limits. *Ecol. Monogr.*, 93(1), e1558.

Frederickson, M. E., Greene, M. J., & Gordon, D. M. (2005). 'Devil's gardens' bedevilled by ants. *Nature*, 437(7058), 495-496.

Friedman, J., & Barrett, S. C. (2009). Wind of change: new insights on the ecology and evolution of pollination and mating in wind-pollinated plants. *Ann. Bot.*, 103(9), 1515-1527.

Guenard, B., Weiser, M., Gomez, K., Narula, N., & Economo, E. P. (2017). The Global Ant Biodiversity Informatics (GABI) database: synthesizing data on the geographic distribution of ant species (Hymenoptera: Formicidae). *Myrmecol. News*, 24, 83-89.

Hembry, D. H., & Tocora, M. C. (2025). Biogeography of Interactions. In Reference Module in Life Sciences. Elsevier.

Hillebrand, H. (2004). On the Generality of the Latitudinal Diversity Gradient. *Am. Nat.*, 163(2), 192-211.

Ho LST, Ane C (2014). "A linear-time algorithm for Gaussian and non-Gaussian trait evolution models." *Syst. Biol.*, 63, 397-408.

Ives, A. R., & Garland Jr, T. (2010). Phylogenetic logistic regression for binary dependent variables. *Syst. Biol.*, 59(1), 9-26.

Kaiser-Bunbury, C. N., Traveset, A., & Hansen, D. M. (2010). Conservation and restoration of plant–animal mutualisms on oceanic islands. *PPEES*, 12(2), 131-143.

Kaur, K. M., Malé, P.-J. G., Spence, E., Gomez, C., & Frederickson, M. E. (2019). Using text-mined trait data to test for cooperate-and-radiate co-evolution between ants and plants. *PLoS Comput. Biol.*, 15(10), e1007323.

Kaur, K. M., Malé, P.-J. G., Spence, E., Gomez, C., & Frederickson, M. E. (2019). Data from *Using text-mined trait data to test for cooperate-and-radiate co-evolution between ants and plants*. Dryad Digital Repository. Available at: https://doi.org/10.5061/dryad.3630t8j.

Kinlock, N. L., Prowant, L., Herstoff, E. M., Foley, C. M., Akin-Fajiye, M., Bender, N. *et al*. (2018). Explaining global variation in the latitudinal diversity gradient: Meta-analysis confirms known patterns and uncovers new ones. *Glob. Ecol. Biogeogr.*, 27(1/2), 125-141.

König, C., Weigelt, P., Taylor, A., Stein, A., Dawson, W., Essl, F. *et al*. (2021). Source pools and disharmony of the world's island floras. *Ecography*, 44(1), 44-55.

König, C., Weigelt, P., Taylor, A., Stein, A., Dawson, W., Essl, F. *et al*. (2021). Data from: *Source pools and disharmony of the world's island floras*. Dryad Digital Repository. Available at: https://doi.org/10.5061/dryad.6wwpzgmwm.

Laurich, J. R., Reid, C. G., Biel, C., Wu, T., Knox, C., & Frederickson, M. E. (2023). Genetic architecture of multiple mutualisms and mating system in Turnera ulmifolia. *J. Evol. Biol.*, 36(1), 280-295.

Leal, L. C., Nogueira, A., & Peixoto, P. E. (2023). Which traits optimize plant benefits? Meta‐analysis on the effect of partner traits on the outcome of an ant–plant protective mutualism. *J. Ecol.*, 111(1), 263-275.

Liu, C., Economo, E. P., & Guénard, B. (2023). GABI‐I: The global ant biodiversity informatics‐island database. In: Wiley Online Library.

Liu, C., Economo, E. P., & Guénard, B. (2023). Data from: GABI‐I: *The global ant biodiversity informatics‐island database*. Figshare Digital Repository. Available at: https://doi.org/10.6084/m9.figshare.21701285.

Losos, J. B., & Ricklefs, R. E. (2009). Adaptation and diversification on islands. *Nature*, 457(7231), 830-836.

Luo, Y., Taylor, A., Weigelt, P., Guénard, B., Economo, E. P., Nowak, A., Inderjit *et al*. (2023). Climate and ant diversity explain the global distribution of ant‐plant mutualisms. *Ecography*, e06841.

MacArthur, R. H., & Wilson, E. O. (2001). *The theory of island biogeography* (Vol. 1). Princeton university press.

Marazzi, B., Bronstein, J. L., & Koptur, S. (2013). Diversity, ecology and evolution of extrafloral nectaries: current perspectives and future challenges. *Ann. Bot.*, 111(6), 1243-1250.

Mittelbach, G. G., Schemske, D. W., Cornell, H. V., Allen, A. P., Brown, J. M., Bush, M. B. *et al*. (2007). Evolution and the latitudinal diversity gradient: speciation, extinction and biogeography. *Ecol. Lett.*, 10(4), 315-331.

Nathan, P., Economo, E. P., Guénard, B., Simonsen, A. K., & Frederickson, M. E. (2023). Generalized mutualisms promote range expansion in both plant and ant partners. *Proc. R. Soc. B.*, 290(2006), 20231083.

Nathan, P., Economo, E. P., Guénard, B., Simonsen, A. K., & Frederickson, M. E. (2023). Data from *Generalized mutualisms promote range expansion in both plant and ant partners*. Dryad Digital Repository. Available at: https://doi.org/10.5061/dryad.jm63xsjh6.

Nelsen, M. P., Ree, R. H., & Moreau, C. S. (2018). Ant–plant interactions evolved through increasing interdependence. *PNAS*, 115(48), 12253-12258.

Nelsen, M. P., Ree, R. H., & Moreau, C. S. (2018). Data from *Ant–plant interactions evolved through increasing interdependence*. Dryad Digital Repository. Available at: https://datadryad.org/dataset/doi:10.5061/dryad.ft4sn88.

Pebesma, E., & Bivand, R. (2023). *Spatial data science : with applications in R* (First edition ed.). CRC Press.

Pichler, M., & Hartig, F. (2024). Is there a robust effect of mainland mutualism rates on species richness of oceanic islands? *arXiv preprint arXiv:2411.15105*.

Pichler, M., & Hartig, F. (2024). Data from: *Is there a robust effect of mainland mutualism rates on species richness of oceanic islands?*. GitHub Digital Repository. Available at: https://github.com/MaximilianPi/IslandLDG.

Prior, K. M., Robinson, J. M., Meadley Dunphy, S. A., & Frederickson, M. E. (2015). Mutualism between co-introduced species facilitates invasion and alters plant community structure. *Proc. R. Soc. B*, 282(1800), 20142846.

Rosumek, F. B., Silveira, F. A., de S. Neves, F., de U. Barbosa, N. P., Diniz, L., Oki, Y., Pezzini, F. *et al*. (2009). Ants on plants: a meta-analysis of the role of ants as plant biotic defenses. *Oecologia*, 160, 537-549.

Rudgers, J. A., & Clay, K. (2008). An invasive plant–fungal mutualism reduces arthropod diversity. *Ecol. Lett.*, 11(8), 831-840.

Schemske, D. W., Mittelbach, G. G., Cornell, H. V., Sobel, J. M., & Roy, K. (2009). Is There a Latitudinal Gradient in the Importance of Biotic Interactions? *Annu. Rev. Ecol. Evol. Syst.*, 40(1), 245-269.

Simberloff, D., & Von Holle, B. (1999). Positive interactions of nonindigenous species: invasional meltdown?. *Biol. Invasions*, 1(1), 21-32.

Soudzilovskaia, N. A. et al. FungalRoot: global online database of plant mycorrhizal associations. *New Phytol.* 227, 955–966 (2020).

Tedersoo, L., Bahram, M., & Zobel, M. (2020). How mycorrhizal associations drive plant population and community biology. *Sci*, 367(6480).

Trager, M. D., Bhotika, S., Hostetler, J. A., Andrade, G. V., Rodriguez-Cabal, M. A., McKeon, C. S. *et al*. (2010). Benefits for plants in ant-plant protective mutualisms: a meta-analysis. *PLoS One*, 5(12), e14308.

Waser, N. M., Chittka, L., Price, M. V., Williams, N. M., & Ollerton, J. (1996). Generalization in pollination systems, and why it matters. *Ecology*, 77(4), 1043-1060.

Weber, M., Porturas, L., & Keeler, K. (2024). *World list of plants with extrafloral nectaries*. Available at: http://www.extrafloralnectaries.org/. Last accessed 14 08 2025.

Weber, M. G., & Agrawal, A. A. (2014). Defense mutualisms enhance plant diversification. *PNAS*, 111(46), 16442-16447.

Weber, M. G., & Keeler, K. H. (2013). The phylogenetic distribution of extrafloral nectaries in plants. *Ann. Bot.*, 111(6), 1251-1261.

Weigelt, P., König, C., & Kreft, H. (2020). GIFT–A global inventory of floras and traits for macroecology and biogeography. *J. Biogeogr.*, 47(1), 16-43.

Werner, G. D., Cornwell, W. K., Sprent, J. I., Kattge, J. & Kiers, E. T. A single evolutionary innovation drives the deep evolution of symbiotic N_2-fixation in angiosperms. *Nat. Commun.* 5, 4087 (2014).

Zeng, Y., & Wiens, J. J. (2021). Species interactions have predictable impacts on diversification. *Ecol. Lett.*, 24(2), 239-248.

Figures

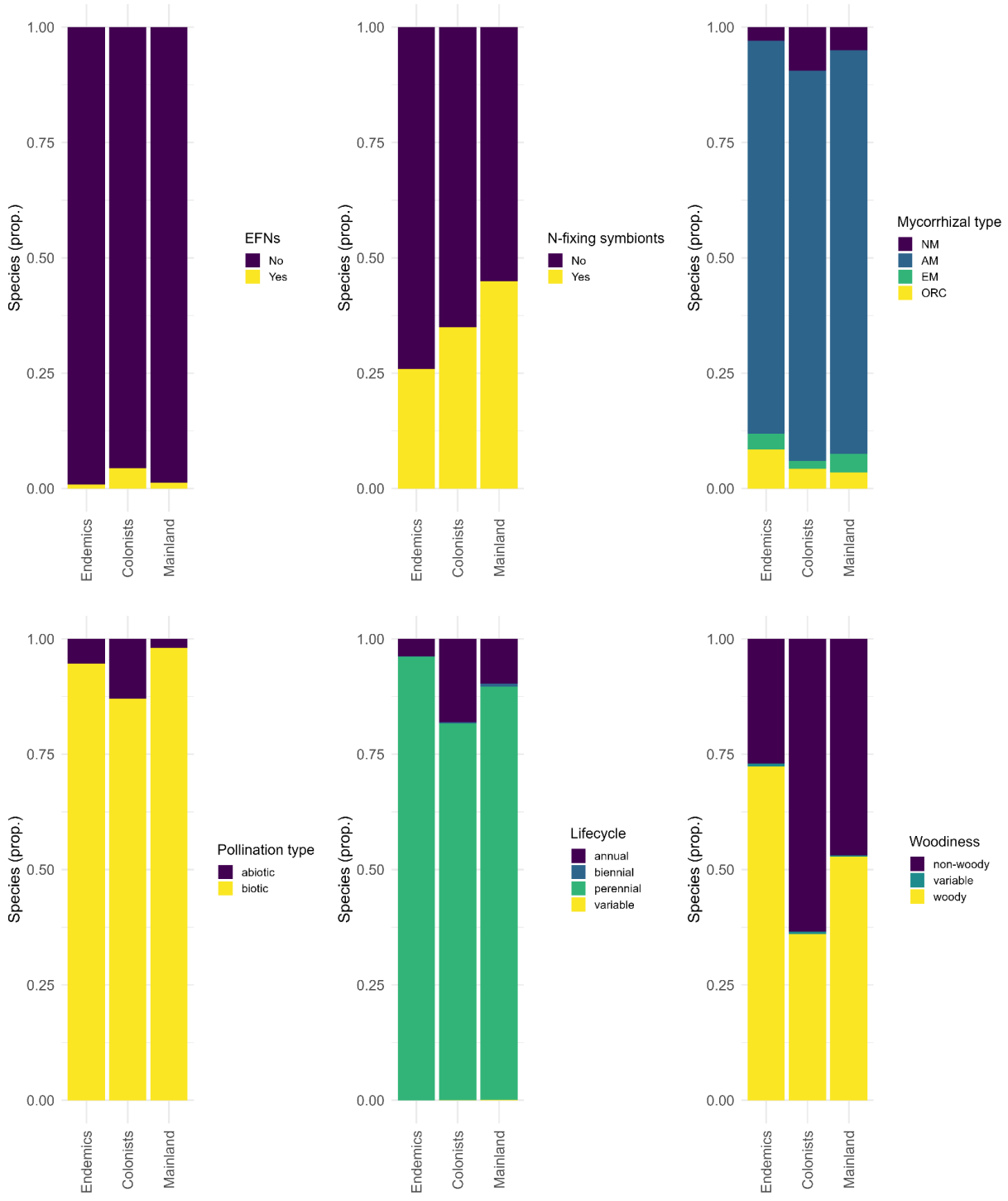


Figure 1. Plant traits among island endemics, island colonists, and mainland species after combining data from GIFT and Weber et al. (2024).

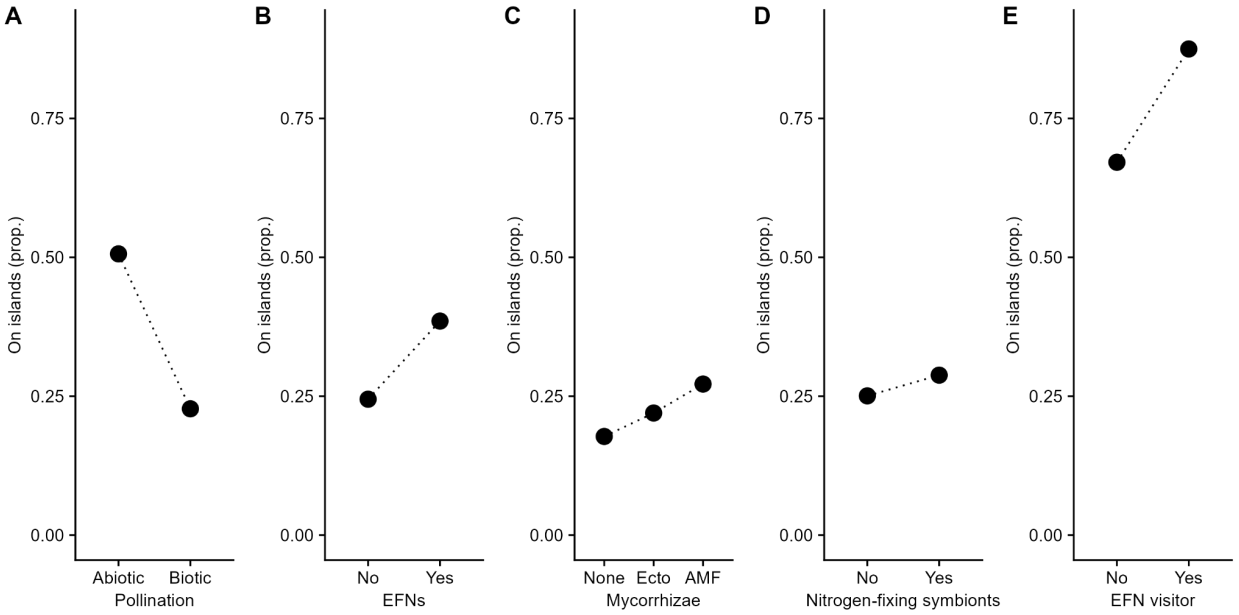
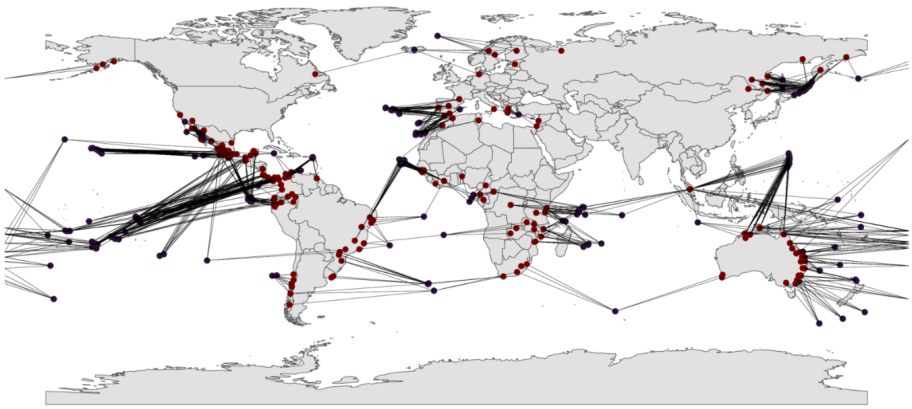


Figure 2. Proportions of plant (A-D) or ant (E) species that occur on islands, for plant species that A) are biotically versus abiotically pollinated, B) do or do not have EFNs, C) associate with arbuscular mycorrhizal fungi (AMF), ectomycorrhizal fungi (ecto), or are non-mycorrhizal (none), and D) do or do not associate with nitrogen-fixing symbionts, and for ant species that E) do or not visit EFNs.

A



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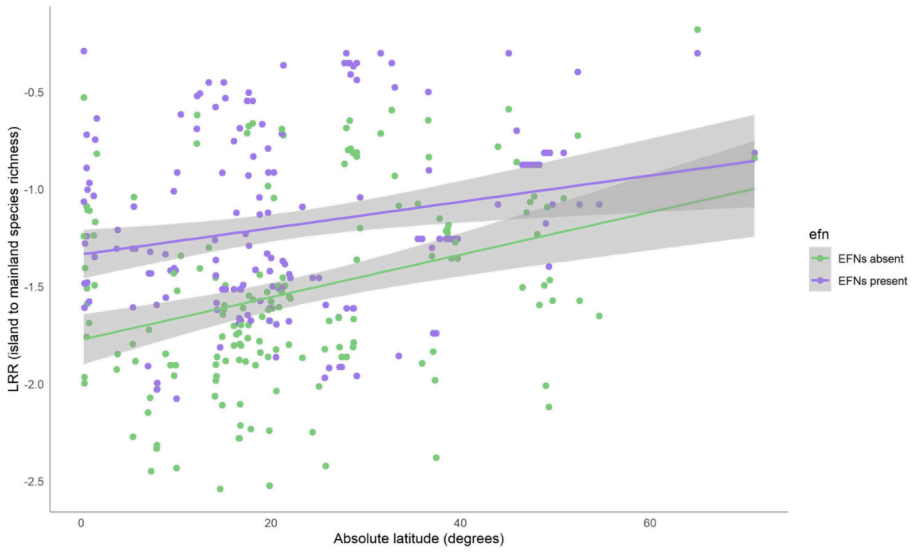
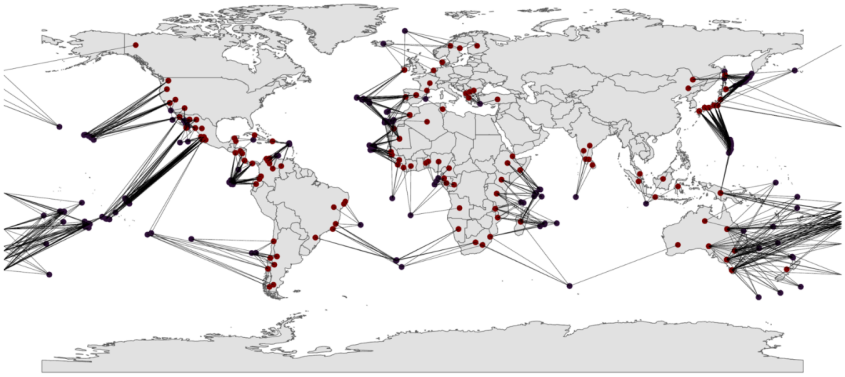


Figure 3. A) Map of five likely source mainlands (red) for each island (blue) for plants. B) Plot of log response ratio of island to mainland species richness for EFN- (purple) and non-EFN-bearing (green) plants against absolute latitude of the island.

A



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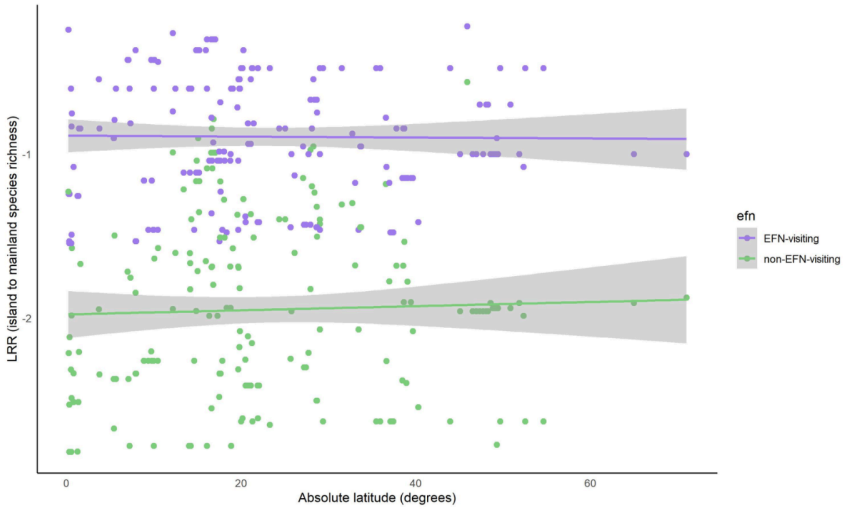

Figure 4. A) Map of five likely source mainlands (red) for each island (blue) for ants. B) Plot of log response ratio of island to mainland species richness for EFN-visiting (purple) and non-EFN-visiting (green) ants against absolute latitude of the island.

Tables

Table 1. Estimates and standard errors (s.e.) from phylogenetic logistic regression models for A) plants (N = 1517 species with complete trait data) and B) ants (N=1660 species with trait data). Island species presence was modelled as a function of EFN status only for ants, or EFN status, mycorrhizal association, nitrogen-fixing symbionts, biotic pollination, woodiness, and lifecycle for plants. Alpha is a measure of phylogenetic signal. In A), the intercept represents abiotically pollinated, annual, herbaceous plants with no EFNs, no mycorrhizae, and no nitrogen-fixing symbionts. In B) the intercept represents ants that do not visit EFNs.

	Estimate	s.e.
A - Plants		
Intercept	0.79	0.42
EFN	*0.91****	*0.20*
Arbuscular mycorrhizal	*0.68**	*0.29*
Ectomycorrhizal	0.32	0.38
Orchid mycorrhizal	1.18	1.45
Nitrogen fixer	0.19	0.23
Biotic pollination	*-1.19****	*0.24*
Variable woodiness	-13.07	840.39
Woody	*-0.96****	*0.19*
Biennial	-1.06	1.06
Perennial	*-0.69**	*0.30*
Variable lifecycle	-13.64	793.66
Mean tip height	137.69	

Alpha	0.046	
B - Ants		
Intercept	*0.760****	*0.067*
EFN status	*1.510****	*0.427*
Mean tip height	136.99	
Alpha	0.085	

Significant effects are *italicized* and indicated by *** for $p \leq 0.001$, ** for $p \leq 0.01$ and * for $p \leq 0.05$.

Table 2. Estimates and standard errors (s.e.) from Gaussian generalized linear models of the log response ratio of island to mainland species richness for A) plants (N = 169 groups) and B) ants (N = 178 groups) as a function of EFN mutualism status and biogeographical variables. Only island species that occur in at least one of the five likely source mainlands per island were included. Parameters ending in .i are island, .ml are mainland.

	Estimate	s.e.
A - Plants		
Intercept	*-1.64****	*0.05*
Absolute latitude.i	*0.005**	*0.002*
EFN?	*0.44****	*0.07*
Distance.ml	*-0.23****	*0.03*
Area.i	*0.22****	*0.03*
Area.ml	*0.17****	*0.05*
Elevational range.i	0.004	0.03
Elevational range.ml	0.05	0.03
Annual precip.i	*0.19****	*0.03*
Latitude * EFN	-0.004	0.003
B - Ants		
Intercept	*-1.952****	*0.081*
Absolute latitude.i	0.0001	0.003
EFN?	*1.089****	*0.087*
Distance.ml	-0.052	0.033

Area.i	*0.071***	*0.025*
Area.ml	-0.013	0.048
Annual precip.i	-0.039	0.032
Latitude * EFN	-0.002	0.003

Significant effects are *italicized* and indicated by *** for $p \leq 0.001$, ** for $p \leq 0.01$ and * for $p \leq 0.05$.