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Research

Small traits with big consequences: how seed traits of nitrogen-fixing plants might influence ecosystem nutrient cycling

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Symbiotic nitrogen (N)-fixing plants have important effects on the biogeochemical processes of the sites they inhabit, but their ability to reach these sites is determined by the dispersal of their seeds. Differences in seed size and dispersal vectors of N-fixing and non-fixing plants could influence the spatial and temporal distributions of N fixers, and thus could have important impacts on biogeochemical cycling. Using seed mass, dispersal vector, and biome data retrieved from online public databases, we ask if there are systematic differences in seed mass and dispersal vectors between N-fixing and non-fixing plants. We demonstrate that rhizobial N fixers tend to have larger seeds that are more likely to be biotically dispersed than seeds of non-fixers, whereas actinorhizal N-fixing trees tend to have small, abiotically dispersed seeds. We then synthesize existing evidence from the literature to draw links between these dispersal traits and the spatio–temporal patterns of N fixers, as well as their biogeochemical effects on terrestrial ecosystems. Using this literature, we argue that the spatio–temporal distributions of N fixers are influenced by their seed dispersal characteristics, and that these distribution patterns have important effects on the total amount of N fixed at a site and the timing of N inputs during processes such as succession.

Keywords: dispersal vector, nitrogen fixation, seed dispersal

Introduction

Nitrogen (N) is a commonly limiting nutrient to primary production in terrestrial ecosystems worldwide (LeBauer and Treseder 2008, Thomas et al. 2013). Symbioses between N-fixing bacteria and certain higher plants (hereafter we refer to the plants as 'N fixers') represent the largest potential natural input of N into ecosystems through their conversion of atmospheric di-nitrogen gas (N_2) into bio-available forms of N (Sprent 2009). These symbioses have the potential to bring well over 100 $\mathrm{kg\,N\,ha^{-1}\,year^{-1}}$ into terrestrial ecosystems (Binkley et al. 1994, Binkley and Giardina 1998, Carlsson and Huss-Danell 2003, Ruess et al. 2009), a potential that exceeds that of asymbiotic N fixation (Reed et al. 2011), rock weathering (Houlton and Dahlgren 2018), and atmospheric N deposition at all but the most polluted locations (Galloway et al. 2008). Where symbiotic N fixers are sufficiently abundant and active, the N they fix can have

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dramatic effects on both local (Vitousek et al. 1987, Binkley and Giardina 1998) and ecosystem-level (Binkley et al. 1992, Compton et al. 2003, Perakis et al. 2011) nutrient cycling and primary production (Vitousek et al. 1987, Binkley and Giardina 1998, Batterman et al. 2013a).

The potential for symbiotic N fixation is ultimately controlled by the distribution and relative abundance of N fixers in an ecosystem. Although N fixers are widely distributed (Benson and Dawson 2007, Sprent 2009), they are conspicuously absent from some ecosystems. The abundance and distribution of N fixers, like all plants, depend in part on their dispersal in addition to niche-based and demographic processes. Seed characteristics such as seed size, shape, color, appendages, sugar and nutrient content, and the presence of secondary compounds determine the primary dispersal vector(s) of a seed and how effectively seeds disperse to environments that are conducive to germination (Howe and Smallwood 1982, Cipollini and Levey 1997, Schupp and Jordano 2010, Howe 2016). For example, species with heavy seeds that are primarily dispersed by terrestrial animals can have different distribution patterns than species with light seeds that are primarily wind dispersed (Moles et al. 2005, Eriksson 2008, Thomson et al. 2011, Galetti et al. 2013). Thus, seed traits that are common for N fixers may influence the dispersal vectors, distribution patterns in time and space, and ultimately the biogeochemical roles of these plants in an ecosystem.

Seed size and dispersal vectors help determine the spatio–temporal patterns of seed deposition (Nathan and Muller-Landau 2000, Thomson et al. 2011). Biotic dispersal (animal or self) is often associated with seeds deposited in clumped distributions (Howe and Smallwood 1982, Wenny 2001, Russo and Augspurger 2004, Karubian et al. 2010), whereas abiotic dispersal (especially via wind) is associated with more evenly distributed seeds (Nathan et al. 2002, McEuen and Curran 2004, Vittoz and Engler 2007). Furthermore, within animal-dispersed plant species, those with larger seeds are more likely to be dispersed in clumped distributions than those with smaller seeds (Howe 1989). Seed size and dispersal vectors can also affect the temporal patterns of seed dispersal. Seed dispersal is one of the most important processes driving the succession of forests (Chazdon 2003), and whether a species is dispersed by biotic or abiotic vectors can have important impacts on when its seeds arrive to a site following disturbance (Wunderle 1997, Howe and Miriti 2004).

Given the substantial effects that seed dispersal can have on species distributions in space and time (Wunderle 1997, Wang and Smith 2002, Chazdon 2003, Seidler and Plotkin 2006, Wiegand et al. 2009), any systematic differences between N fixer and non-fixer seed characteristics, their dispersal vectors, or both, could drive important patterns of N fixer abundance distributions. If N fixers tend to have larger, more biotically dispersed seeds than non-fixers, then we would expect N fixer seed dispersal within the ecosystem to be spatially clumped and occur later in successional time. If, however, N fixers have smaller, more abiotically dispersed seeds than non-fixers, then we would expect N fixers to be distributed more evenly across space and to arrive earlier in successional time. Although distributions of adult plants in any ecosystem are the result of complex processes of dispersal, recruitment, growth, and survival (Wang and Smith 2002), the dispersal of N fixers to a given location at a given time is the first step toward their potential ecological and biogeochemical impacts.

One of the challenging issues in any N-fixer versus non-fixer comparison is the strong phylogenetic signal in the trait of symbiotic N fixation (Felsenstein 1985). The vast majority of N-fixing plant taxa are found in a single family (legumes), and all nodulating (including rhizobial and actinorhizal) N fixers are confined to the Rosid I clade (Soltis et al. 1995, Werner et al. 2014), so an association of seed traits with N-fixing plants might reflect the trait of N fixation or it might reflect a property of legumes or the Rosid I clade more broadly. One pioneering study examined how seed size, N content, and a variety of other characteristics varied between N-fixing and non-fixing legumes (Corby et al. 2011). They found that N-fixing legumes had smaller seeds with higher N concentrations than non-fixing legumes (Corby et al. 2011). Because their analysis targets traits within the phylogenetic clade where N fixation occurs, it speaks to how N fixation and seed traits co-evolve. Their study, however, did not give a full picture of how the seeds of N fixers compared to those of all relevant non-fixers (including non-legumes as well as legumes). If there are fundamental differences in seed characteristics and dispersal vectors between N fixers and non-fixers, then the biogeochemical consequences of these differences matter for ecosystem-level studies, regardless of how those differences arose evolutionarily. Some studies have examined N fixer versus non-fixer seed differences within specific ecosystems. For example, Vargas et al. (2015) examined germination rates of legumes compared to non-legumes in a tropical dry forest, and argued that legumes' higher germination rates help drive their success in dry forests. At present, however, there has been no systematic investigation of N fixer versus non-fixer seed traits across ecosystems.

In this study, we assessed the potential for seed dispersal characteristics of N-fixing plants to drive the ecosystem-level biogeochemical impacts of N fixers in terrestrial ecosystems. We use a combination of global seed trait and dispersal vector data to address two questions: 1) how do seed masses of N fixers differ from those of non-fixers? 2) How do dispersal vectors differ between N fixers and non-fixers? We then draw on evidence from the seed dispersal literature to evaluate the argument that: 3) the seed mass and dispersal vector differences we document influence patterns of N-fixer abundances in space and/or time. Finally, we draw on the biogeochemistry literature to evaluate the argument that: 4) the spatio–temporal patterns of N fixers have direct impacts on their biogeochemical effects in an ecosystem, and therefore that the seed traits of N-fixing plants influence ecosystem-level biogeochemistry in terrestrial ecosystems.

Material and methods

Data collection and preparation

To assess the first two questions – differences in seed mass and dispersal vectors between N-fixing and non-fixing plants – we used data from the TRY Plant Trait Database (Kattge et al. 2011) and the Diaspore Dispersal Database (D3; Hintze et al. 2013). We requested data from TRY on seed mass, dispersal vector, plant growth form, and spatial coordinates of data collection for all publicly available angiosperm species. Given that the D3 database does not provide spatial coordinates, we requested dispersal vector data for all angiosperm species available from D3, and corroborated those results with the growth form and spatial data requested from TRY. For consistency across taxa and dispersal syndromes, we focused exclusively on dry seed mass rather than the mass of the entire diaspore. We used spatial data for global biome distributions from the World Wildlife Fund to assign spatially referenced seed trait data to a given terrestrial biome (Olson et al. 2001).

We classified species in our dataset as putative N fixers based on a series of criteria. For actinorhizal N fixers, we used the list of actinorhizal N-fixing species presented in Werner et al. (2014), which is largely based on the list composed by Huss-Danell (1997). Rhizobial N fixers were first classified as such based on the list of confirmed rhizobial N-fixing species in Sprent (2009). Because the trait of N fixation is thought to be largely conserved at the genus level, we also assigned species as putative N fixers if they were congeners of the rhizobial N fixers in Sprent (2009) and actinorhizal N fixers in Werner (2014).

To assess differences in seed characteristics, we compared seed mass data between N fixers and non-fixers in our combined dataset. Seed mass data were natural-log transformed to meet assumptions of normality. In addition to analyzing global differences in seed size between N fixers and non-fixers, we also assessed these differences within growth-form categories and biomes. We removed any growth-form classifications that did not have at least one N fixer and one non-fixer species represented, resulting in four growth form categories: herb, shrub, tree and vine. We also removed any biomes that we did not have at least one N fixer and one non-fixer species for, resulting in six terrestrial biomes: desert, Mediterranean forest, montane grassland, temperate broadleaf forest, temperate grassland and tropical moist broadleaf forest.

To assess our second assumption, we compared primary dispersal vectors between N fixers and non-fixers. Given that these data were collected in data repositories (TRY and D3), verification of dispersal vector for each entry was done by the researcher(s) contributing to each database. Dispersal vector data were first placed into eight vector categories to group redundant vector names, which resulted in the following dispersal vector classifications: anemochory (wind dispersal), anthropochory (human dispersal), autochory (self dispersal), ballochory (ballistic dispersal), barochory (gravity dispersal), hydrochory (water dispersal), zoochory (animal dispersal, including dispersal by ants), multiple (vector information was listed as 'multiple' in databases), and other (dispersed by some other vector not listed here). To investigate differences in abiotic versus biotic dispersal between N fixers and non-fixers, we grouped dispersal vectors into groups of abiotic (wind and water dispersed) and biotic (human, self, ballistic, gravity and animal dispersed). Our dataset included 6700 unique species with dispersal vector data, but because many species had multiple unique vectors listed, our dispersal vector analyses were conducted on 13 902 unique species–vector combinations (Supplementary material Appendix 1 Table A1).

Statistical analyses

To test for differences in seed mass between N fixers and non-fixers, we ran bootstrapped analysis of variance (ANOVA) models testing for differences in ln(seed mass) by fixer type (N fixer or non-fixer) using the mean of each species' seed mass as an individual observation. Seed mass data were natural-log transformed and bootstrapped to meet assumptions of normality and equal sample sizes for ANOVA models. Seed mass results were back-transformed for figure presentation. We also tested for the effects of biome and plant growth form on the relationship between fixer type and seed mass using bootstrapped ANOVAs testing ln(seed mass) by fixer type, biome (or growth form), and their interaction. We ran all bootstrapped ANOVAs for 1000 iterations, and averaged the statistical output across these iterations. To test for differences in dispersal vectors between N fixers and non-fixers, we used two-sample χ^2- tests with Yates's continuity correction testing the proportion of N-fixing and non-fixing species dispersed by each of our dispersal vector categories. χ^2 comparisons for N fixers and non-fixers were performed independently for each dispersal vector category. We then ran all statistical models (ANOVA and χ^2) as above but testing for differences in fixation symbiosis (Rhizobial fixers versus Actinorhizal fixers versus non-fixers). All analyses were run in the base package of R statistical software (<www.r-project.org>).

Data deposition

Data available from the TRY digital repository: <https://www.try-db.org/TryWeb/Data.php doi: 10.17871/TRY.3> (Wilcots et al. 2018).

Results

We obtained data from 52 unique datasets from the TRY and D3 databases that were widely distributed globally (Fig. 1) (Kattge et al. 2011, Hintze et al. 2013). These data included 7890 individual species for which some combination of seed mass, growth form, and dispersal vector data were available – 730 N-fixing species (655 Rhizobial, 75 Actinorhizal) and 7160 non-fixing species (Supplementary material Appendix

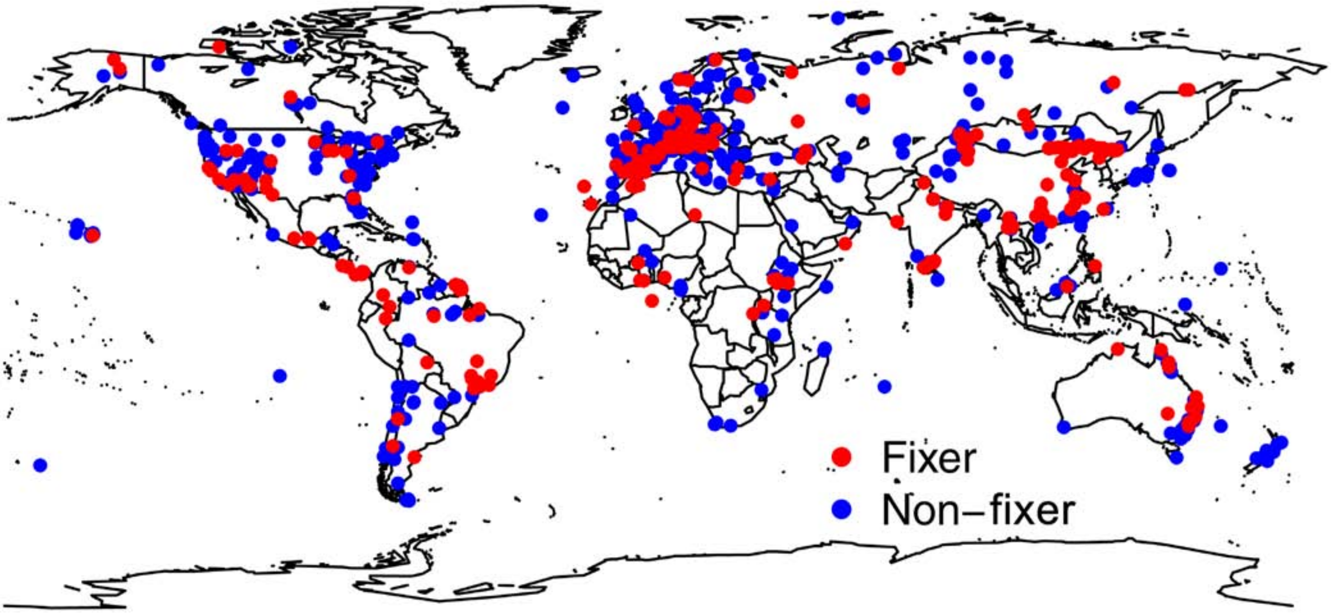

Figure 1. Geographic distribution of geo-referenced data in this study. Red dots indicate plants capable of symbiotic N fixation ('N fixers'), and blue dots indicate non-fixers. Points represent the 2830 unique species entries that contained geographic reference data. Although this represents only 36% of the total 7890 data points in our study, even this subset of our data demonstrates the broad geographic extent of our analyses. All species data were taken from the TRY Plant Trait Database (Kattge et al. 2011) and Diaspore Dispersal Database (D3, Hintze et al. 2013).

1 Table A1, A2). A total of six terrestrial biomes held both N-fixing and non-fixing species in our dataset – deserts, Mediterranean forests, montane grasslands, temperate broadleaf forests, temperate grasslands, and tropical moist broadleaf forests.

Differences in seed traits between N fixers and non-fixers

When species in all growth forms, dispersal vectors, and biomes were pooled, N-fixing species had seeds that were over four times larger, on average, than non-fixing species ($p < 0.001$; Fig. 2a). Geometric mean seed mass also varied by a factor of 50 between plant growth forms ($p < 0.001$), with herbs having the smallest seeds and trees having the largest (Supplementary material Appendix 1 Fig. A1). Within each of the four growth forms we assessed, N fixers had larger seeds than non-fixers in all categories except for trees: approximately five times larger in herbs, three times larger in shrubs, and four times larger in vines (although the difference was not statistically significant for vines, which were variable and had low sample sizes; Supplementary material Appendix 1 Fig. A1a). The discrepancy in the pattern of seed size in trees was driven by differences between the two functional groups of N-fixing symbioses – rhizobial and actinorhizal N fixers (Supplementary material Appendix 1 Fig. A1b). Rhizobial tree species, which in our dataset are all in the plant family Fabaceae and are capable of housing symbiotic bacteria from the family Rhizobia (Sprent 2009), had similarly-sized seeds as non-fixing trees (Supplementary material Appendix 1 Fig. A1b). However, actinorhizal N-fixing trees, which occur in a variety of plant families and house bacteria from the genus *Frankia* (Huss-Danell 1997, Werner et al. 2014), had significantly smaller seeds than non-fixing and rhizobial N-fixing trees ($p < 0.001$ for both comparisons; Supplementary material Appendix 1 Fig. A1b).

Because the effect of a species' dispersal ecology relative to its neighboring species is most important for potentially sympatric species, we also analyzed differences in seed mass by biome. Of the six terrestrial biomes that included both N fixers and non-fixers in our dataset, N fixers had significantly heavier seeds in four of them (Supplementary material Appendix 1 Fig. A2, A3) – tropical moist broadleaf forests ($p = 0.006$), deserts ($p < 0.001$), temperate broadleaf forests ($p < 0.001$), and temperate grasslands ($p = 0.034$). N fixers also tended to have higher average seed mass in the remaining two terrestrial biomes, but these differences were not statistically significant (montane grasslands: $p = 0.126$, and Mediterranean forests: $p = 0.086$).

Differences between N-fixer functional groups were also present in several terrestrial biomes (Supplementary material Appendix 1 Fig. A2b). In desert systems, rhizobial N fixers had much larger seeds than actinorhizal N fixers ($p = 0.040$) and non-fixers ($p < 0.001$), but actinorhizal N fixers were not different than non-fixers ($p = 0.235$). Actinorhizal N-fixing species in Mediterranean forests had larger seeds than rhizobial N fixers ($p = 0.001$) and non-fixers ($p = 0.032$), which were not significantly different from each other ($p = 0.211$). In temperate broadleaf forests, rhizobial N fixers had larger seeds than non-fixers ($p < 0.001$), with actinorhizal N-fixing

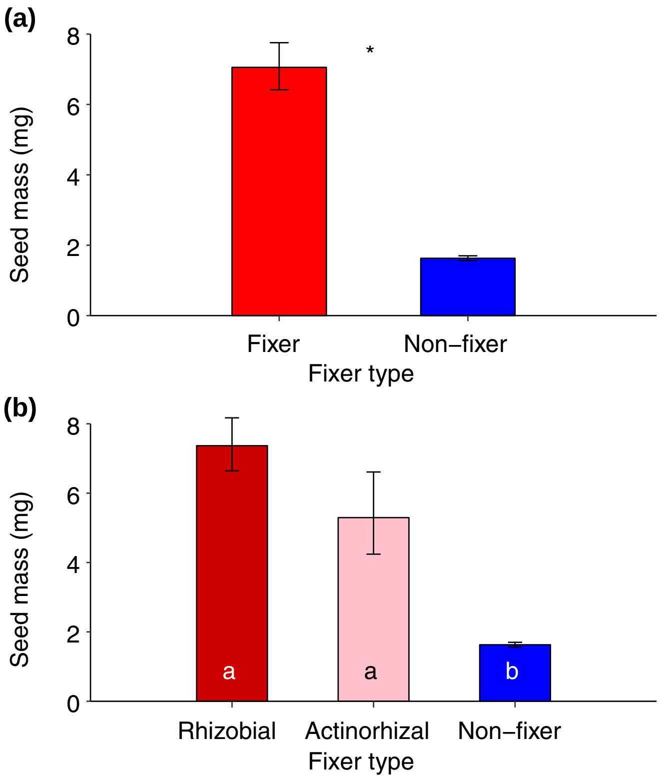

Figure 2. (a) Geometric mean seed mass (mg) of N fixers and non-fixers. (b) Geometric mean seed mass for rhizobial N fixers (dark red), actinorhizal N fixers (pink), and non-fixers (blue). Error bars represent ±1 SE. For (a) N fixers had significantly higher mean seed mass than non-fixers ($p < 0.0001$). For (b) seed mass did not differ between rhizobial and actinorhizal N fixers ($p = 0.686$), but both rhizobial and actinorhizal N fixers had significantly higher seed mass than non-fixers ($p = 0.005$ and $p < 0.0001$, respectively).

species having intermediate-sized seeds that were not statistically different from either rhizobial N fixers ($p = 0.603$) or non-fixers ($p = 0.101$).

Differences in dispersal vectors between N fixers and non-fixers

In addition to differences in seed size, we found significant differences in the proportions of N-fixing and non-fixing species dispersed by most of the dispersal vectors that we assessed. N-fixing species were more likely than non-fixers to be dispersed via ballistic propulsion ($p < 0.001$), autochory (self; $p < 0.001$), and barochory (gravity; $p = 0.006$) (Fig. 3a). Conversely, non-fixers were more likely than N fixers to be dispersed by hydrochory (water; $p < 0.001$) and anemochory (wind; $p < 0.001$) (Fig. 3a). When we grouped our dispersal vectors into coarser categories, we found that N fixers were more likely than non-fixers to be biotically dispersed (animal, self, gravity, and ballistic dispersal grouped; $p < 0.001$), while non-fixers were more likely to be abiotically dispersed (water and wind dispersal grouped; $p < 0.001$; Fig. 3c, d).

Although we did not find differences in the proportion of N-fixing versus non-fixing species dispersed by animals ($p = 0.096$), this was due to different dispersal mechanisms of the two N-fixer functional groups. Differences between N-fixer functional groups were important for many of our dispersal vectors (Fig. 3b, d). For example, the lack of a significant difference in animal dispersal between N fixers and non-fixers was due to the drastically different propensity of rhizobial and actinorhizal N fixers to be animal dispersed. Rhizobial N fixers were significantly more likely to be animal dispersed than non-fixers ($p = 0.002$), and both groups were more likely to be animal dispersed than actinorhizal N fixers ($p < 0.001$ for both comparisons). Important differences also existed between N-fixer functional groups for wind dispersal. Actinorhizal N fixers were more likely to be wind-dispersed than non-fixers ($p = 0.026$), and both groups were more likely to be wind-dispersed than rhizobial N fixers ($p < 0.001$ for both comparisons). For gravity dispersal, significant differences between N fixers and non-fixers were primarily driven by actinorhizal N fixers, which had significantly more gravity-dispersed seeds than both rhizobial N fixers and non-fixers ($p < 0.001$ for both comparisons), but rhizobial N fixers and non-fixers were similarly likely to be gravity dispersed ($p = 0.595$). Despite non-fixers being 1.5 times as likely to be water-dispersed as rhizobial N fixers ($p < 0.001$) and twice as likely as actinorhizal N fixers ($p = 0.063$), the difference between non-fixers and actinorhizal N fixers was non-significant due to the low sample size of actinorhizal N fixers.

Discussion

Our dataset comprises nearly 8000 species from six continents and four plant growth forms. As such, our analyses present a broad view of the differences in seed size and dispersal vectors between N-fixing and non-fixing plants. Results from these analyses reveal clear answers to our two questions. 1) On average, rhizobial N fixers had substantially larger seeds than non-fixers (Fig. 2), although actinorhizal N-fixing trees had smaller seeds than non-fixing trees. 2) In general, rhizobial N fixers are dispersed more by animals, ballistic propulsion, autochory, and barochory than non-fixers, which tend to be dispersed more by water and wind. Actinorhizal N fixers are even more likely to be dispersed by wind and gravity (Fig. 3). These patterns were generally consistent across plant growth forms (Supplementary material Appendix 1 Fig. A1) and biomes (Supplementary material Appendix 1 Fig. A2, A3). Although both the TRY and D3 plant trait databases are somewhat biased toward European sampling (Fig. 1), the large number of species in our dataset and our ability to separate our data geographically by biome make our analyses more robust to this bias. Below, we discuss how the evidence from our analyses relates to the relevant literature, then we draw on additional literature to connect the dispersal characteristics of N fixers to their abundance patterns and ecosystem consequences.

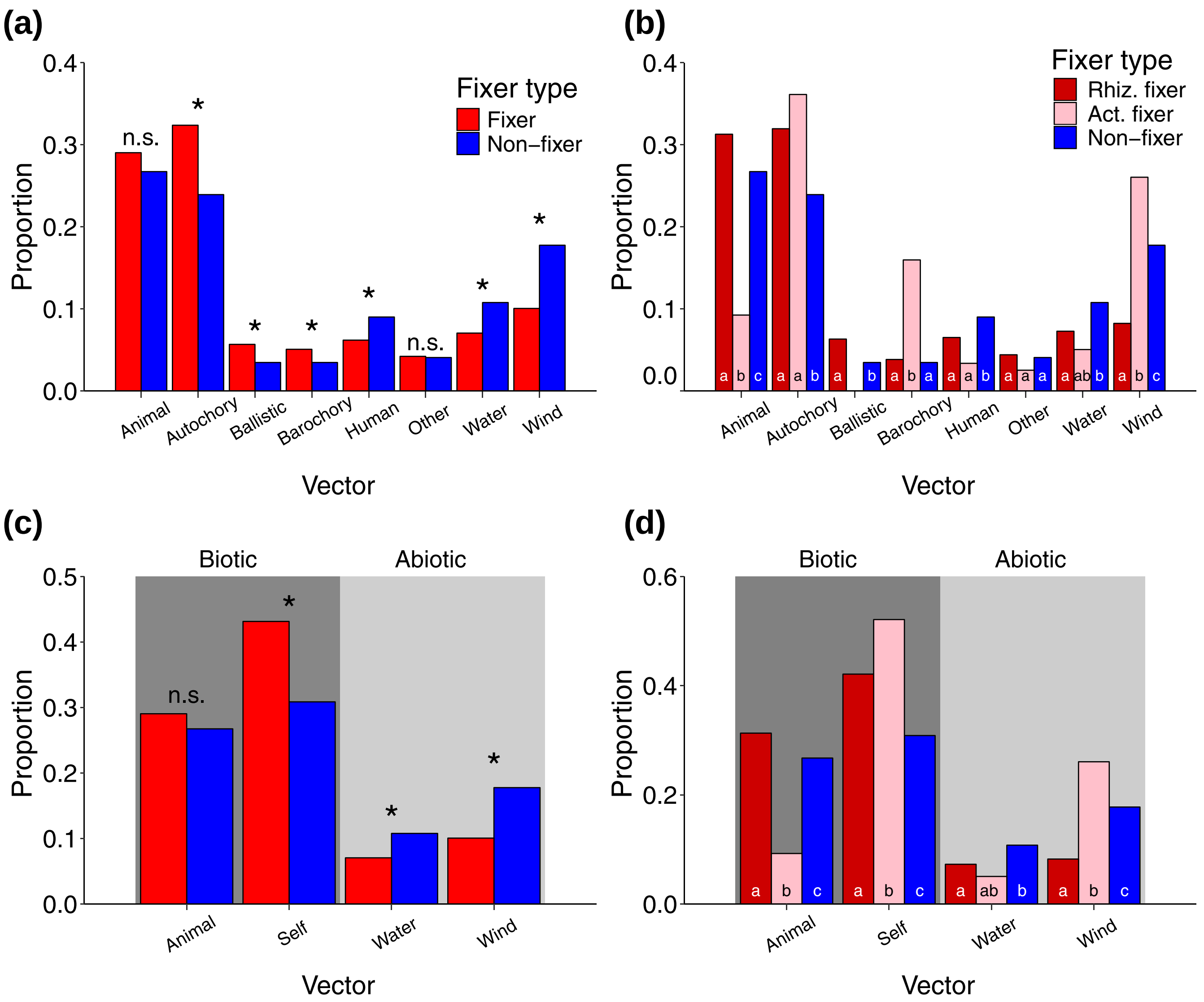

Figure 3. (a) Proportion of N-fixing and non-fixing species dispersed by individual dispersal vectors, and (b) these same dispersal vectors shown with N fixers separated by fixer functional types. (c) Proportion of N fixers and non-fixers dispersed by broader dispersal vector categories. (d) The same broad dispersal categories are shown with N fixers separated by fixer functional type. * and letters indicate significant differences for two- and three-way comparisons, respectively, as in Fig. 2.

Question 1. How does seed mass differ between N fixers and non-fixers?

Evidence from the almost 8000 species in our dataset suggests that rhizobial N fixers have larger seeds than non-fixers, which in turn have larger seeds than actinorhizal N-fixing trees. Our finding about rhizobial N fixers seems diametrically opposed to the finding of a previous study (Corby et al. 2011), which found that rhizobial N fixers had smaller seeds than non-fixers. However, the seeming conflict is easily explained by the different phylogenetic contexts of the two studies. Corby et al. (2011) compared N-fixing legumes to non-fixing legumes, whereas we compared N-fixing legumes to all non-fixing plants. Together, these two studies provide a comprehensive picture of the relationship between seed size and N fixation. Corby et al. (2011)'s study, which had a more targeted phylogenetic context, suggests that N fixation has been more successful when it appears with smaller seeds. But our study shows that despite this potential co-evolutionary trend, N fixers in general have larger seeds than non-fixers because legumes have large seeds compared with other plant families. Because larger seeds represent a large resource expenditure for the parent plant but also provide an important competitive advantage for germinating offspring, the relative costs and benefits of producing large seeds are likely context specific and contribute to the relative success of N fixers in various global biomes.

Although seed trait information was limited to seed mass in our dataset, other seed traits such as nutrient content, color, fruit type, and secondary compounds may also differ systematically between N fixers and non-fixers and may also have important implications for the dispersal and distribution of this plant group. For example, Corby et al. (2011) found that N-fixing legumes have higher seed N concentrations than non-fixing legume species, and evidence shows that even non-fixing legumes have higher seed N concentrations than non-legumes (McKey 1994). It seems likely, therefore, that N fixers also have higher seed N content than a broader class of non-fixers. We are aware of no systematic comparisons of traits such as fruit type, color, or seed defense between N fixers and non-fixers; however, related evidence from the literature suggests that N fixers may allocate more to seed defense than non-fixers. N-based defensive compounds are diverse within the legume family (Janzen 1969, McKey 1994), and among non-legumes, seed size and seed N content have both been shown to increase plant allocation to seed defense (Grubb et al. 1998). These two lines of evidence suggest that the relatively large, N-rich seeds of rhizobial N fixers may promote high allocation to seed defense, and that many of these N fixers have evolved the chemical capacity for seed defense, but systematic studies are needed to directly assess this possibility.

Question 2. How do dispersal vectors differ between N fixers and non-fixers?

We also found N fixers and non-fixers to have different dispersal vectors (Fig. 3). At the broadest scale, we found that N fixers were more likely to be dispersed by biotic vectors than were non-fixers. Notably, biotic vectors dominated the dispersal of both N fixers and non-fixers in our data (as has been noted previously for angiosperms, Howe and Smallwood 1982, Tiffney 2018), but N-fixer dispersal was more strongly dominated by biotic vectors than non-fixers. As with our analysis of seed size, these patterns were generally consistent across plant growth forms (Supplementary material Appendix 1 Fig. A1), biomes (Supplementary material Appendix 1 Fig. A2, A3), and symbiotic N-fixing functional groups with the exception of actinorhizal N fixing trees, which were more likely to be wind-dispersed than both rhizobial N fixers and non-fixers (Fig. 3, Supplementary material Appendix 1 Fig. A1b).

Of the two response variables in our dataset (seed mass and dispersal vector), dispersal vector is inherently less precise due to the possibility of many dispersal events going unobserved and the common difficulty of separating dispersal from predation for animal-dispersed seeds (Schupp 1993). However, our analysis of seed size lends additional support to our results for dispersal vectors. Seed size tends to be positively related to, and often the evolutionary result of, a propensity for biotic dispersal vectors (Moles et al. 2005, Tiffney 2018). Thus, the strong evidence for rhizobial N fixers to have larger seeds than non-fixers, but actinorhizal N-fixing trees to have smaller seeds than non-fixers (Fig. 2, Supplementary material Appendix 1 Fig. A1), corroborates our results that rhizobial N fixers are more likely to be biotically dispersed, but actinorhizal N-fixing trees are more likely to be abiotically dispersed, than non-fixers (Fig. 3).

Argument 3. Differences in dispersal ecology between N fixers and non-fixers likely influence N-fixer abundance patterns in space and time

We are aware of no studies that have directly assessed the link between the dispersal ecology of N fixers and their resulting spatio–temporal distributions. However, evidence from the literature suggests that large, biotically-dispersed seeds tend to be distributed spatially in clumps and temporally in mid- to late-successional landscapes relative to smaller, abiotically dispersed seeds (Fig. 4; Fragoso et al. 2003, Russo and Augspurger 2004, Clark et al. 2005). There is wide variation in seed masses and dispersal vectors for all of our plant groups, but the fact that rhizobial N fixers have large, biotically dispersed seeds on average (Fig. 2, 3a) should predispose them to be spatially clumped across a landscape and temporally distributed late in successional time.

A substantial body of literature has documented the clumped species distributions resulting from non-random animal dispersal (Russo and Augspurger 2004, Clark et al. 2005, Fedriani et al. 2010, Razafindratsima and Dunham 2016, Trolliet et al. 2017). Much of this literature focuses on the latrines and roosts of bird and monkey dispersers, where studies have demonstrated that roost sites result in high densities and high genetic structure of the preferred food species of the animal disperser (Russo and Augspurger 2004, Clark et al. 2005, Trolliet et al. 2017). This effect has also been demonstrated for a variety of terrestrial mammal dispersers (Fedriani et al. 2010). Thus, with the notable exception of seeds that pass through the digestive tracts of large vertebrates, seeds dispersed either by the parent plant or by animal dispersers (e.g. invertebrates, terrestrial mammals, reptiles, bats, birds) are likely to be dispersed either in clumps around the parent plant (within a few meters) (Vittoz and Engler 2007, Kuprewicz 2013) or in clumped distributions near feeding roosts (Janzen et al. 1976, Russo and Augspurger 2004). Given the large seed size and propensity for biotic dispersal vectors that we demonstrate for rhizobial N fixers (Fig. 2, 3) and the abundance of evidence that animal dispersal often leads to spatially clumped species distributions, we deem it likely that these dispersal characteristics predispose rhizobial N fixers to be more spatially clumped, but actinorhizal species less spatially clumped, than non-fixers.

A species' seed traits and dispersal vectors also control the temporal distributions of its seeds, largely determining when a species is present in dynamic successional landscapes. The temporal effects of a species' seed dispersal are especially important during the process of forest succession (Chazdon 2003). Abiotically dispersed seeds are often the first to reach a site following disturbance (Howe and Miriti 2004), with large, animal-dispersed seeds only reaching a site once sufficient perches (Zanini and Ganade 2005, Graham and Page 2012, McClanahan and Wolfe 2012), vegetation structure (McDonnell and Stiles 1983, Debussche and Isenmann

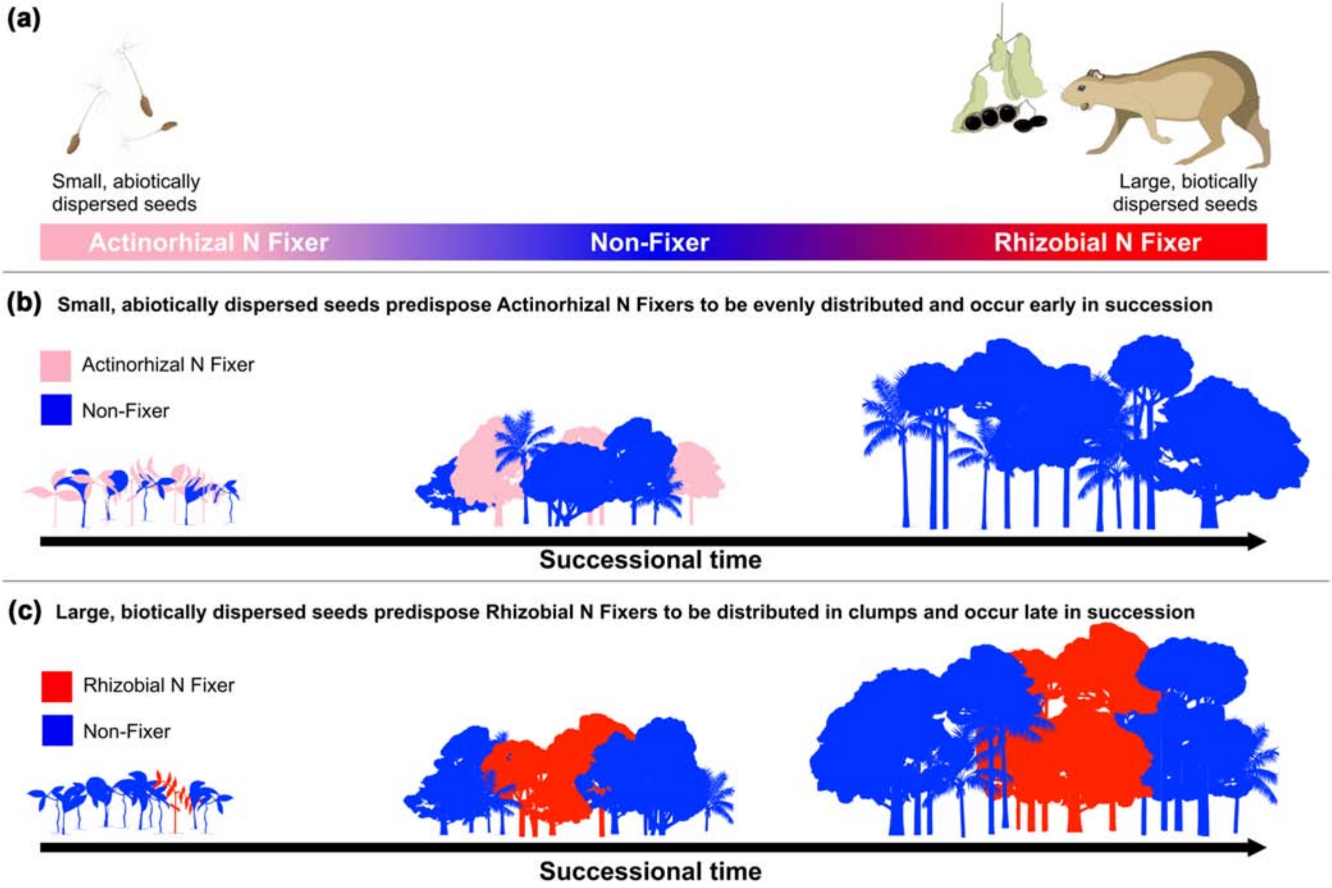

Figure 4. Conceptual diagram showing potential links between seed traits of N fixers and their spatio–temporal patterns. (a) Our results (Fig. 2, 3) indicate that rhizobial N fixers tend to have larger, more often biotically dispersed seeds than non-fixers, and that actinorhizal N-fixing trees tend to have small, abiotically dispersed seeds. (b) The small, wind-dispersed seeds of actinorhizal N-fixing trees predispose them to be evenly spaced and relatively common in early successional stages. (c) The large, biotically dispersed seeds of rhizobial N fixers predispose them to being spatially clumped and relatively common in later successional stages.

1994, Ferguson and Drake 2010), and fruit (Guevara et al. 1992, Vieira et al. 1994, Silva and Murray 1996) exist to attract and protect animal dispersal vectors (Finegan 1984, Wunderle 1997, Chazdon 2003). In general, the seed dispersal literature points to a temporal shift from abiotic dispersal in early succession to biotic dispersal in later-successional sites (Janzen 1969, Wunderle 1997).

Together, this evidence from the literature suggests that the tendency for rhizobial N fixers to have large, biotically dispersed seeds may predispose them to be spatially distributed in clumps and temporally distributed later in successional time than non-fixing species. However, the small, wind-dispersed seeds of actinorhizal N-fixing trees may lead to evenly spaced and early successional distributions of N fixers (Fig. 4b) in systems like temperate and boreal forests where actinorhizal is the dominant N-fixer tree type (Menge et al. 2014, 2017). In the next section we argue that these spatio–temporal patterns of N-fixer distributions can have important consequences for the amount, location, and timing of N brought into an ecosystem by N fixers.

Argument 4. Spatio–temporal abundance patterns influence the biogeochemical effects of N fixers

Spatial distributions

The spatial pattern of N fixers likely has important effects not only on where N inputs occur, but also on how much total N is brought into an ecosystem. N fixers provide N to the surrounding ecosystem via litterfall of their N-rich leaf tissue, potentially creating soil N ‘hotspots’ throughout a landscape where N fixers are abundant. A large body of evidence demonstrates that tree species influence local soil chemistry in a range of ecosystems (Zinke 1962, Binkley and Giardina 1998, Finzi et al. 1998, Waring et al. 2015, Russell et al. 2017, Soper et al. 2018) and the high leaf N concentrations of N fixers (McKey 1994, Fyllas et al. 2009, Nasto et al. 2014, Adams et al. 2016) make their potential to affect local soil N availability particularly strong. It should be noted that identifying the effects of individual trees on soil chemistry has been less successful in some highly diverse tropical forests (Powers et al. 2004, Gei and Powers 2013, but see Osborne et al. 2017, Soper et al. 2018), but this may be a homogenization effect of highly diverse litter inputs rather than canopy trees simply not affecting local soil chemistry.

In addition to the location of N inputs into an ecosystem, the aggregation of N fixers may influence the magnitude of N inputs by driving survival and relative abundance patterns of N fixers as well as their individual N fixation rates. Recent work has shown that particularly strong competitive effects between neighboring N fixers mean that N fixers within clumps of other N fixers have low growth and survival rates (Taylor et al. 2017). This suggests that clumped distributions of N fixers may lead to lower relative abundances than if N fixers are evenly spaced throughout the landscape. Additionally, clumping might reduce total N inputs because of how the soil N hotspots created by clumping affect the symbioses. Many N fixers decrease N fixation rates when soil N availability is high (Barron et al. 2011, Batterman et al. 2013a,b, Menge et al. 2015), so when N-fixing individuals are within clumps they might down-regulate N fixation.

In a recent study, Menge and Levin (2017) used spatially explicit models to support the idea that spatial clumping of N fixers decreases ecosystem-level rates of N fixation. Based on a realistic assumption of higher foliar N content in N fixers (McKey 1994, Fyllas et al. 2009, Nasto et al. 2014, Adams et al. 2016), and assuming that some fraction of an individual's litter moves beyond its rooting zone, their models demonstrate that total ecosystem N inputs from N fixation are substantially lower when N fixers are arranged in clumped distributions than when they are distributed evenly throughout a landscape (Fig. 5). The mechanism, as explained above, is that N fixers at the center of N-fixer clumps down-regulate fixation when receiving N-rich litter transfers from neighboring N fixers rather than N-poor litter from non-fixers. Their results suggest that in forests where N-fixers comprise 50% of the trees (higher than the Neotropical average of ~10–20%) (ter Steege et al. 2006, Menge et al. 2014, Menge and Levin 2017, Gei et al. 2018), but not uncommon for individual stands (Sullivan et al. 2014, Gei et al. 2018) the difference in the spatial arrangement of N fixers alone can drive changes in ecosystem N inputs that are equivalent or greater than N deposition for many of these sites (Galloway et al. 2008). This suggests that N fixers with large, biotically-dispersed seeds such as rhizobial N-fixing trees, may not bring as much N into ecosystems as they would if they had seed characteristics that distributed these plants more evenly across landscapes.

Temporal distributions

One of the hallmark roles of N fixers in ecosystem processes is their ability, via the N they fix, to facilitate ecosystem development during the early stages of succession (Chapin et al. 1994, Walker and Del Moral 2003, Batterman et al. 2013a). N fixers are often associated with early succession on highly degraded soils due to their ability to supply their own biologically available N (Walker and Del Moral 2003). However, our findings suggest that the reliance on biotic dispersal for many rhizobial N fixers means their seeds may have difficulty reaching early successional sites due to limited animal dispersal during the earliest successional stages. This evidence indicates that rhizobial N fixers could potentially have an even larger role in the development of nutrient cycling in early succession if their dispersal characteristics were more suited to rapid colonization of recently disturbed sites.

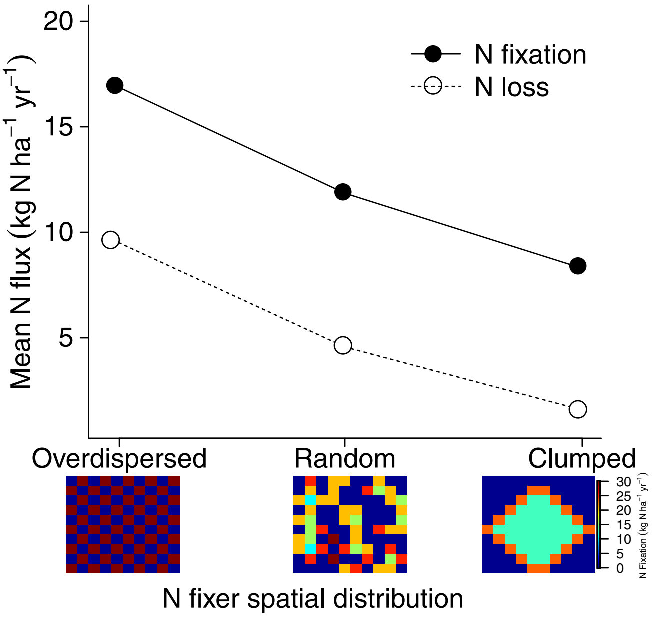

Figure 5. Increasingly clumped N-fixer distributions reduce N inputs and losses. Modeled mean N-fixation (closed dots) and available N loss (open circles) values for overdispersed, random, and clumped spatial distributions of N fixers are shown, adapted with permission from Menge and Levin (2017). X-axis labels show a visual representation of the spatial distributions of N fixer abundance (blue is non-fixer, other colors are fixers) and N fixation inputs. In their model, Menge and Levin show that total ecosystem-level N fixation decreases as N-fixer clumping increases because N fixers fertilize their neighbors, resulting in greater down-regulation of N fixation than when their neighbors are non-fixers.

One notable exception to this successional pattern can be seen in temperate N-fixing trees. Ample evidence shows that N-fixing trees above 35°N latitude in North America are predominantly actinorhizal (Menge et al. 2014, 2017), which have substantially smaller and more abiotically dispersed seeds than their average non-fixing neighbors (Fig. 3b, d, Supplementary material Appendix 1 Fig. A1). These dispersal characteristics may make temperate actinorhizal N fixers better suited to early successional colonization than sympatric non-fixing trees. Demographic analyses of temperate N fixers could help corroborate this pattern. In the temperate United States, N-fixing trees are largely confined to the earliest stages of succession (Menge et al. 2010), partially due to their inability to recruit into later-successional forests (Liao and Menge 2016). This recruitment limitation during later stages of succession may be partially due to the shade intolerance of the small seeds of actinorhizal north American N fixers. Alternatively, the notable abundance of N fixers in late-successional Neotropical forests (Gei et al. 2018) may result, in part, from their large, biotically-dispersed seeds, which are dispersed more frequently to these habitats and have the seed resources to tolerate the deep shade of mature tropical forest understories (Baraloto and Forget 2007).

Overall, we find reasonable evidence in the literature to suggest that the dispersal characteristics of N fixers could have important effects on the timing of their N inputs during succession. However, this evidence indicates that the effects of dispersal on the timing of N fixer abundances may be very different between large, biotically dispersed rhizobial N fixers and small, abiotically dispersed actinorhizal N-fixing trees. Similar to the spatial patterns of N fixers discussed above, future empirical studies are needed to directly assess these potential mechanisms in nature.

Conclusion

Although the success and ecological influence of N fixers are determined by a combination of factors, including demographic traits (Liao and Menge 2016, Menge and Chazdon 2016) and soil nutrient availability (Batterman et al. 2013a,b), here we demonstrate that the seed traits and dispersal vectors common to N fixers may be important for the timing, amount, and distribution of N inputs from symbiotic fixation in terrestrial ecosystems. Our analyses show N fixers have, on average, larger and more biotically-dispersed seeds than non-fixers, with the exception of actinorhizal N-fixing trees which tend to have small, abiotically-dispersed seeds. These significant differences in the dispersal mechanisms and seed sizes of N fixers and non-fixers could impact terrestrial biogeochemical cycling, demonstrating how seemingly small traits of individual organisms can have important effects at the ecosystem scale.

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References

Adams, M. A. et al. 2016. Legumes are different: leaf nitrogen, photosynthesis and water use efficiency. – Proc. Natl Acad. Sci. USA 113: 4098–4113.

Baraloto, C. and Forget, P.-M. 2007. Seed size, seedling morphology, and response to deep shade and damage in Neotropical rain forest trees. – Am. J. Bot. 94: 901–911.

Barron, A. R. et al. 2011. Facultative nitrogen fixation by canopy legumes in a lowland tropical forest. – Oecologia 165: 511–520.

Batterman, S. A. et al. 2013a. Key role of symbiotic dinitrogen fixation in tropical forest secondary succession. – Nature 502: 224–227.

Batterman, S. A. et al. 2013b. Nitrogen and phosphorus interact to control tropical symbiotic N_2 fixation: a test in *Inga punctata*. – J. Ecol. 101: 1400–1408.

Benson, D. R. and Dawson, J. O. 2007. Recent advances in the biogeography and genecology of symbiotic *Frankia* and its host plants. – Physiol. Plant. 130: 318–330.

Binkley, D. and Giardina, C. 1998. Why do tree species affect soils? The warp and woof of tree–soil interactions. – Biogeochemistry 42: 89–106.

Binkley, D. et al. 1992. Biogeochemistry of adjacent conifer and alder–conifer stands. – Ecology 73: 2022–2033.

Binkley, D. et al. 1994. Soil nitrogen availability in some arctic ecosystems in northwest Alaska: responses to temperature and moisture. – Ecoscience 1: 64–70.

Carlsson, G. and Huss-Danell, K. 2003. Nitrogen fixation in perennial forage legumes in the field. – Plant Soil 253: 353–372.

Chapin, F. et al. 1994. Mechanisms of primary succession following deglaciation at Glacier Bay, Alaska. – Ecol. Monogr. 64: 149–175.

Chazdon, R. L. 2003. Tropical forest recovery: legacies of human impact and natural disturbances. – Perspect. Plant Ecol. Evol. Syst. 6: 51–71.

Cipollini, M. L. and Levey, D. J. 1997. Secondary metabolites of fleshy vertebrate – dispersed fruits: adaptive hypotheses and implications for seed dispersal. – Am. Nat. 150: 346–372.

Clark, C. et al. 2005. Comparative seed shadows of bird-, monkey- and wind-dispersed trees. – Ecology 86: 2684–2694.

Compton, J. E. et al. 2003. Nitrogen export from forested watersheds in the Oregon Coast Range: the role of N_2-fixing red alder. – Ecosystems 6: 773–785.

Corby, H. D. L. et al. 2011. Size, structure and nitrogen content of seeds of *Fabaceae* in relation to nodulation. – Bot. J. Linn. Soc. 167: 251–280.

Debussche, M. and Isenmann, P. 1994. Bird-dispersed seed rain and seedling establishment in patchy Mediterranean vegetation. – Oikos 69: 414–426.

Eriksson, O. 2008. Evolution of seed size and biotic seed dispersal in angiosperms. – Paleoecol. Neoecol. Evid. 169: 863–870.

Fedriani, J. M. et al. 2010. Spatial pattern of adult trees and the mammal-generated seed rain in the Iberian pear. – Ecography 33: 545–555.

Felsenstein, J. 1985. Phylogenies and the comparative method. – Am. Nat. 125: 1–15.

Ferguson, R. N. and Drake, D. R. 2010. Influence of vegetation structure on spatial patterns of seed deposition by birds. – New Zeal. J. Bot. 37: 671–677.

Finegan, B. 1984. Forest succession. – Nature 312: 109–114.

Finzi, A. C. et al. 1998. Canopy tree–soil interactions within temperate forests: species effects on soil carbon and nitrogen. – Ecol. Appl. 8: 440–446.

Fragoso, J. M. V. et al. 2003. Long-distance seed dispersal by tapirs increases seed survival and aggregates tropical trees. – Ecology 84: 1998–2006.

Fyllas, N. M. et al. 2009. Basin-wide variations in foliar properties of Amazonian forest: phylogeny, soils and climate. – Biogeosciences 6: 2677–2708.

Galetti, M. et al. 2013. Functional extinction of birds drives rapid evolutionary changes in seed size. – Science 340: 1086–1091.

Galloway, J. N. et al. 2008. Transformation of the nitrogen cycle: recent trends, questions and potential solutions. – Science 320: 889–893.

Gei, M. G. and Powers, J. S. 2013. Do legumes and non-legumes tree species affect soil properties in unmanaged forests and plantations in Costa Rican dry forests? – Soil Biol. Biochem. 57: 264–272.

Gei, M. et al. 2018. Legume abundance along successional and rainfall gradients in Neotropical forests. – Nat. Ecol. Evol. 2: 1104–1111.

Graham, L. L. B. and Page, S. E. 2012. Artificial bird perches for the regeneration of degraded tropical peat swamp forest: a restoration tool with limited potential. – Restor. Ecol. 20: 631–637.

Grubb, P. J. et al. 1998. Nitrogen-richness and protection of seeds in Australian tropical rainforest: a test of plant defence theory. – Oikos 82: 467–482.

Guevara, S. et al. 1992. Floristic composition and structure of vegetation under isolated trees in Neotropical pastures. – J. Veg. Sci. 3: 655–664.

Hintze, C. et al. 2013. D3: the dispersal and diaspore database – baseline data and statistics on seed dispersal. – Perspect. Plant Ecol. Evol. Syst. 15: 180–192.

Houlton, B. Z. and Dahlgren, R. A. 2018. Convergent evidence for widespread rock nitrogen sources in Earth's surface environment. – Science 360: 58–62.

Howe, H. F. 1989. Scatter- and clump-dispersal and seedling demography: hypothesis and implications. – Oecologia 79: 417–426.

Howe, H. F. 2016. Making dispersal syndromes and networks useful in tropical conservation and restoration. – Global Ecol. Conserv. 6: 152–178.

Howe, H. F. and Smallwood, J. 1982. Ecology of seed dispersal. – Annu. Rev. Ecol. Syst. 13: 201–228.

Howe, H. F. and Miriti, M. N. 2004. When seed dispersal matters. – Bioscience 54: 651–660.

Huss-Danell, K. 1997. Actinorhizal symbioses and their N_2 fixation. – New Phytol. 136: 375–405.

Janzen, D. 1969. Seed-eaters versus seed size, number, toxicity, and dispersal. – Evolution 23: 1–27.

Janzen, D. H. et al. 1976. Two Costa Rican bat-generated seed shadows of *Andira inermis* (Leguminosae). – Ecology 57: 1068–1075.

Karubian, J. et al. 2010. Destination-based seed dispersal homogenizes genetic structure of a tropical palm. – Mol. Ecol. 19: 1745–1753.

Kattge, J. et al. 2011. TRY – a global database of plant traits. – Global Change Biol. 17: 2905–2935.

Kuprewicz, E. K. 2013. Mammal abundances and seed traits control the seed dispersal and predation roles of terrestrial mammals in a Costa Rican forest. – Biotropica 45: 333–342.

LeBauer, D. and Treseder, K. 2008. Nitrogen limitation of net primary productivity in terrestrial ecosystems is globally distributed. – Ecology 89: 371–379.

Liao, W. and Menge, D. N. L. 2016. Demography of symbiotic nitrogen-fixing trees explains their rarity and successional decline in temperate forests in the United States. – PLoS One 11: 1–13.

McClanahan, T. and Wolfe, R. 2012. Accelerating forest succession in a fragmented landscape: the role of birds and perches. – Conserv. Biol. 11: 279–288.

McDonnell, M. J. and Stiles, E. W. 1983. The structural complexity of old field vegetation and the recruitment of bird-dispersed plant species. – Oecologia 56: 109–116.

McEuen, A. B. and Curran, L. M. 2004. Seed dispersal and recruitment limitation across spatial scales in temperate forest fragments. – Ecology 85: 507–518.

McKey, D. 1994. Legumes and nitrogen: the evolutionary ecology of a nitrogen-demanding lifestyle. – In: Sprent, J. and McKey, D. (eds), Advances in legume systematics 5: the nitrogen factor. – Royal Botanic Gardens, pp. 211–228.

Menge, D. N. L. and Chazdon, R. L. 2016. Higher survival drives the success of nitrogen-fixing trees through succession in Costa Rican rainforests. – New Phytol. 209: 965–977.

Menge, D. N. L. and Levin, S. A. 2017. Spatial heterogeneity can resolve the nitrogen paradox of tropical forests. – Ecology 98: 1049–1061.

Menge, D. N. L. et al. 2010. Phylogenetic constraints do not explain the rarity of nitrogen-fixing trees in late-successional temperate forests. – PLoS One 5(2): e12056.

Menge, D. N. L. et al. 2014. Nitrogen fixation strategies can explain the latitudinal shift in nitrogen-fixing tree abundance. – Ecology 95: 2236–2245.

Menge, D. N. L. et al. 2015. Diversity of nitrogen fixation strategies in Mediterranean legumes. – Nat. Plants 1: 15064.

Menge, D. N. L. et al. 2017. Nitrogen-fixing tree abundance in higher-latitude North America is not constrained by diversity. – Ecol. Lett. 20: 842–851.

Moles, A. T. et al. 2005. A brief history of seed size. – Science 307: 576–580.

Nasto, M. K. et al. 2014. Interactions among nitrogen fixation and soil phosphorus acquisition strategies in lowland tropical rain forests. – Ecol. Lett. 17: 1282–1289.

Nathan, R. and Muller-Landau, H. 2000. Spatial patterns of seed dispersal, their determinants and consequences for recruitment. – Trends Ecol. Evol. 15: 278–285.

Nathan, R. et al. 2002. Mechanisms of long-distance dispersal of seeds by wind. – Nature 418: 409–413.

Olson, D. M. et al. 2001. Terrestrial ecoregions of the world: a new map of life on Earth. – Bioscience 51: 933–938.

Osborne, B. B. et al. 2017. Climate, topography, and canopy chemistry exert hierarchical control over soil N cycling in a Neotropical lowland forest. – Ecosystems 20: 1089–1103.

Perakis, S. S. et al. 2011. $\delta^{15}N$ constraints on long-term nitrogen balances in temperate forests. – Oecologia 167: 793–807.

Powers, J. S. et al. 2004. Tree species do not influence local soil chemistry in a species-rich Costa Rica rain forest. – J. Trop. Ecol. 20: 587–590.

Razafindratsima, O. H. and Dunham, A. E. 2016. Frugivores bias seed–adult tree associations through nonrandom see dispersal: a phylogenetic approach. – Ecology 97: 2094–2102.

Reed, S. C. et al. 2011. Functional ecology of free-living nitrogen fixation: a contemporary perspective. – Annu. Rev. Ecol. Evol. Syst. 42: 489–512.

Ruess, R. W. et al. 2009. Disease-mediated declines in N-fixation inputs by *Alnus tenuif* in interior and south-central Alaska to early-successional floodplain. – Ecosystems 12: 489–502.

Russell, A. E. et al. 2017. Tropical tree species traits drive soil cation dynamics via effects on pH: a proposed conceptual framework. – Ecol. Monogr. 87: 685–701.

Russo, S. E. and Augspurger, C. K. 2004. Aggregated seed dispersal by spider monkeys limits recruitment to clumped patterns in *Virola calophyllai*. – Ecol. Lett. 7: 1058–1067.

Schupp, E. W. 1993. Quantity, quality and the effectiveness of seed dispersal by animals. – Vegetatio 107: 15–29.

Schupp, E. W. and Jordano, P. 2010. Seed dispersal effectiveness revisited: a conceptual review. – New Phytol. 188: 333–353.

Seidler, T. G. and Plotkin, J. B. 2006. Seed dispersal and spatial pattern in tropical trees. – PLoS Biol. 4: 344.

Silva, J. and Murray, G. 1996. Plant succession, landscape management, and the ecology of frugivorous birds in abandoned Amazonian pastures. – Conserv. Biol. 10: 491–503.

Soltis, D. E. et al. 1995. Chloroplast gene sequence data suggest a single origin of the predisposition for symbiotic nitrogen fixation in angiosperms. – Proc. Natl Acad. Sci. USA 92: 2647–2651.

Soper, F. M. et al. 2018. Remotely sensed canopy nitrogen correlates with nitrous oxide emissions in a lowland tropical rainforest. – Ecology 99: 2080–2089.

Sprent, J. I. 2009. Legume nodulation: a global perspective. – Wiley.

Sullivan, B. et al. 2014. Spatially robust estimates of biological nitrogen (N) fixation imply substantial human alteration of the tropical N cycle. – Proc. Natl Acad. Sci. USA 111: 8101-8106.

Taylor, B. et al. 2017. Nitrogen-fixing trees inhibit growth of regenerating Costa Rican rainforests. – Proc. Natl Acad. Sci. USA 114: 8817-8822.

ter Steege, H. et al. 2006. Continental-scale patterns of canopy tree composition and function across Amazonia. – Nature 443: 444–447.

Thomas, R. Q. et al. 2013. Global patterns of nitrogen limitation: confronting two global biogeochemical models with observations. – Global Change Biol. 19: 2986–2998.

Thomson, F. J. et al. 2011. Seed dispersal distance is more strongly correlated with plant height than with seed mass. – J. Ecol. 99: 1299–1307.

Tiffney, B. H. 2018. Seed size, dispersal syndromes, and the rise of the angiosperms: evidence and hypothesis. – Ann. Miss. Bot. Gard. 71: 551–576.

Trolliet, F. et al. 2017. Frugivorous birds influence the spatial organization of tropical forests through the generation of seedling recruitment foci under zoochoric trees. – Acta Oecol. 85: 69–76.

Vargas, G. G. et al. 2015. Explaining legume success in tropical dry forests based on seed germination niches: a new hypothesis. – Biotropica 47: 277–280.

Vieira, I. C. G. et al. 1994. The role of the shrub *Cordia multispicata* Cham as a 'succession facilitator' in an abandoned pasture, Paragominas, Amazônia. – Vegetatio 115: 91–99.

Vitousek, P. M. et al. 1987. Biological invasion by *Myrica faya* alters ecosystem development in Hawaii. – Science 238: 802–804.

Vittoz, P. and Engler, R. 2007. Seed dispersal distances: a typology based on dispersal modes and plant traits. – Bot. Helv. 117: 109–124.

Walker, L. R. and Del Moral, R. 2003. Primary succession and ecosystem rehabilitation. – Cambridge Univ. Press.

Wang, B. C. and Smith, T. B. 2002. Closing the seed dispersal loop. – Trends Ecol. Evol. 17: 8–10.

Waring, B. G. et al. 2015. Pervasive and strong effects of plants on soil chemistry: a meta-analysis of individual plant 'Zinke' effects. – Proc. R. Soc. B 282: 20151001.

Wenny, D. G. 2001. Advantages of seed dispersal: a re-evalution of directed dispersal. – Evol. Ecol. Res. 3: 51–74.

Werner, G. D. A. et al. 2014. A single evolutionary innovation drives the deep evolution of symbiotic N_2 fixation in angiosperms. – Nat. Commun. 5: 1–9.

Wiegand, T. et al. 2009. Recruitment in tropical tree species: revealing complex spatial patterns. – Am. Nat. 174: 106-140.

Wilcots, M. E. et al. 2018. Data from: small traits with big consequences: how seed traits of nitrogen-fixing plants might influence ecosystem nutrient cycling. – TRY digital repository, <https://www.try-db.org/TryWeb/Data.php doi: 10.17871/TRY.3>.

Wunderle, J. M. 1997. The role of animal seed dispersal in accelerating native forest regeneration on degraded tropical lands. – For. Ecol. Manage. 99: 223–235.

Zanini, L. and Ganade, G. 2005. Restoration of *Araucaria* forest: the role of perches, pioneer vegetation, and soil fertility. – Restor. Ecol. 13: 507–514.

Zinke, P. J. 1962. The pattern of influence of individual forest trees on soil properties. – Ecology 43: 130–133.

Supplementary material (available online as Appendix oik-05798 at <www.oikosjournal.org/appendix/oik-05798>). Appendix 1.