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Quantifying the effects of seed arrival and environmental conditions on tropical seedling community structure

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Abstract Though it is recognized that both stochastic and deterministic processes structure all communities, empirical assessments of their relative importance are rare, particularly within any single community. In this paper, we quantify the dynamic effects of dispersal assembly and niche assembly on the seedling layer in a diverse neotropical rain forest. The two theories make divergent predictions regarding the roles of seed arrival and environmental heterogeneity in generating community structure. Put simply, dispersal assembly posits that the stochasticity inherent to seed arrival structures communities, whereas niche assembly suggests that heterogeneity in post-dispersal environmental conditions is more important. We experimentally sowed 15,132 seeds of eight tree species at varying levels of density and diversity. Every six months we censused the seedlings that germinated and assessed the abiotic and biotic conditions of each plot. We assessed the density, diversity, and species composition of three nested subsets of the seedling layer: seedlings germinated from sown seeds, all seedlings germinated between July 2003 and 2004, and all woody seedlings. We partitioned the variance in density and diversity of each subset of the seedling layer into components representing seed-addition treatments and environmental conditions at 6- to 12-month intervals. Seed additions initially explained more variance in the density and diversity than did environmental heterogeneity for seven of eight sown species, but explained little variance in the density or diversity of the entire seedling layer. Species composition was better explained by seed-addition treatments than by environmental heterogeneity for all three subsets and in all time periods. Nevertheless, the variance in community structure explained by seed-addition treatments declined over the two years following germination, presaging shifts in the relative importance of dispersal assembly and niche assembly. Our study quantifies how dispersal assembly and niche assembly may vary among the components of an ecological community and shift dynamically through time.

Keywords Community assembly · Dispersal · Niche differentiation · Seed addition · Variance partitioning

Introduction

Niche assembly and dispersal assembly, two bodies of theory that seek to explain the structure of ecological communities, have frequently been interpreted as adversarial. Niche assembly suggests that community structure arises through the interplay of relatively deterministic interspecific interactions and tradeoffs across a spatially heterogeneous landscape (Chase and Leibold 2003). Dispersal assembly, by contrast, takes the view that community

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structure arises from stochasticity in dispersal and demography (Bell 2001; Hubbell 2001). Many aspects of niche assembly have found empirical support: species distributions often track environmental conditions (Harms et al. 2001; Arii and Lechowicz 2002; John et al. 2007), competition can structure community composition (Fargione et al. 2003; Harpole and Tilman 2006), and species differ in trait values, which can affect demographic performance (Bloor and Grubb 2003; Shipley et al. 2006). While challenged by abundant evidence of non-neutral patterns, dispersal assembly theories predict some aspects of community structure, such as relative species abundance, accurately and with extreme parsimony (Hubbell 2001; Volkov et al. 2005). Hybrid models that combine stochastic dispersal with deterministic niche assembly have been developed recently (Tilman 2004; Gravel et al. 2006), but empirical evaluation of these assembly theories has been scant.

Previous examinations of the roles of dispersal assembly and niche assembly in shaping community structure have generally yielded a single estimate per community. As data sources, these studies have used snapshots of relative species abundance distributions (Volkov et al. 2003, Etienne and Olff 2004), spatial distributions of species at local and regional scales (Condit et al. 2002; Karst et al. 2005), and community dynamics data at local and continental scales (Clark and McLachlan 2003; McGill et al. 2005; Harpole and Tilman 2006). Even so, it is increasingly evident that dispersal assembly and niche assembly simultaneously affect all ecological communities (Chase 2003), and that their roles may shift both among and within communities (Chase 2007). Previous studies have often averaged over within-community variation in dispersal assembly and niche assembly to generate a single estimate of their relative importance (but see Gilbert and Lechowicz 2004 for a refreshing counterexample). This study takes an experimental approach to quantify within-community variation in the roles of dispersal assembly and niche assembly.

We base our study on the divergent predictions of dispersal assembly and niche assembly regarding the relationship between community structure and the two stages of seedling recruitment: seed arrival and seedling establishment. Under strict dispersal assembly, the arrival of seeds would generate community structure, whereas under strict niche assembly, heterogeneity in the environmental conditions encountered by juveniles would structure communities. We define “environmental conditions” broadly, including all factors, biotic and abiotic, that may affect seedling growth and survival. Much effort has been dedicated to elucidating the myriad environmental conditions in tropical forests that affect the structure of the seedling layer. They include, among others, soil moisture (Engelbrecht and Kursar 2003), light availability (Nicotra et al. 1999, Montgomery and Chazdon 2002), competition (Lewis and Tanner 2000) and seed predation (Paine and Beck 2007). By contrast, the role of seed arrival in generating seedling community structure has received less attention, directed primarily towards its effects on seedling density and diversity (Harms et al. 2000; Norden et al. 2007).

We performed two very simple experiments to quantify the effects of seed arrival and environmental heterogeneity on the structure of the seedling layer. In the density experiment, we added seeds of a single species at varying levels of density to plots. In the richness experiment, we added exactly 24 seeds at varying levels of richness to plots. In both experiments, we then monitored the germination and survival of the resultant seedlings. By simultaneously monitoring the environmental conditions of each plot, we were able to partition the variance in seedling community structure (density, richness and species composition) into components representing seed arrival and environmental heterogeneity. These partitions represent the degree to which dispersal assembly and niche assembly structure the seedling community. We performed these analyses on three nested subsets of the seedling layer, and repeated them every 6 (or 12) months to better understand the within-community dynamics of these processes.

Materials and methods

Study site and species

This study was conducted at Estación Biológica Cocha Cashu (EBCC) in the moist floodplain forest of Peru’s Manu National Park (~12°S, 71°W, ~350 m elevation, see site descriptions in Terborgh 1990; Paine 2007). The site is characterized as tropical moist forest (Holdridge 1947). The forested floodplain of the Manu River is extremely diverse, with an average of 174 species that attain a diameter of 10 cm at breast height per hectare, and at least 825 in the floodplain as a whole (Pitman et al. 2002). Average annual precipitation is 2,200 mm, falling mainly between October and April (Paine 2007).

Tree species were included in the seed-addition experiments of this study based upon three criteria. First, their seeds had to be sufficiently large to be easily cleaned and sown. Second, fruit needed to be available between April–May 2004, the period in which seeds were added. Third, fruiting adults had to be sufficiently common and fecund to provide 1,600–3,100 seeds per species for sowing. These criteria yielded eight species, all of which reach the canopy as adults, and all of which are primarily mammal-dispersed (Foster and Janson 1985). The eight species are members of widely separated phylogenetic lineages, including one basal eudicot, two palms, and dicots from four other orders (Davies et al. 2004). Adult stem density of the eight species encompassed the entire range of abundances observed among canopy trees in this forest, ranging from 0.08 to 32 ha^{-1} (median = 1.3 ha^{-1}; see Paine and Beck (2007) for methods). Seeds for sowing were cleaned as in Paine and Beck (2007); briefly, fruit pulp and dispersal structures were removed manually, using water only when necessary. Seeds that bore any evidence of insect or fungal damage were discarded. Average seed mass ranged from 0.4 to 3.9 g (median = 0.8 g). Seed viability was assessed by observing germination in a shadehouse at EBCC. Ten seeds of each species were sown into ten pots (six for *T. pleeana*) filled 1:1 with forest soil and sand and monitored for one year. Species names and distinguishing characteristics are presented in Table 1. Further natural history information about the species employed in this experiment is available in Paine (2007).

Experimental design

We conducted two seed-addition experiments to quantify the roles of seed arrival and environmental heterogeneity in shaping seedling community structure. In July 2003, 230 plots were laid out in 23 blocks of ten plots each. Plots were circular, with an area of 1 m^2, and permanently marked with a central iron rebar. The following April, each block was expanded to 24 plots, and a 24th block was added for a total of 576 study plots. Blocks were randomly located throughout a 4-km^2 area of mature floodplain forest at EBCC. Blocks were located to avoid recent tree-fall gaps or frequently inundated swales. Within a block, plots were arranged in two (occasionally three) parallel rows, separated by 10 m. Spacing between neighboring plots was varied between 5 and 10 m to minimize autocorrelation of environmental variables. The geographic distance among plots therefore ranged from 5 m to 2 km. Over the three-year study, four of the 576 plots were destroyed by fallen trees and excluded from analysis.

The density and richness experiments were conducted in separate plots. Sixteen plots in each block were used by the density experiment, four for the richness experiment, and the remaining four served as unsown controls for both experiments. The natural recruitment rates of the eight sown species were estimated by their emergence in control plots, where no seeds were sown.

The 16 density experiment plots in each block were, in turn, divided evenly among the eight sown species. Due to an insufficiency of seeds, not every sowing density was represented in every block, and "block" terms were not included in the statistical models. The density experiment was repeated with each of the sown species. In it, seeds of a single species were sown at doubling classes of density up to a species-specific maximum, as suggested by Clark et al. (2007). The species-specific maximum sowing density was the maximum density of the natural seed fall observed after exhaustively searching for seeds in ten randomly located 1-m^2 quadrats beneath the crowns of at least five fruiting adult trees of each sown species (50 m^2 per species, in total). The adult trees under which we searched were separated by at least 100 m. The maximum observed density varied among species from 48 seeds/m^2 for *Duguetia quitarensis* and *Diospyros subrotata* to 120 seeds/m^2 for *Iriartea deltoidea*.

The four plots per block randomly assigned to the richness experiment received exactly 24 seeds, representing two, four, six or eight species, with species drawn randomly and evenly from the pool of eight sown species. In this way, evenness was maximized, and the species richness of the added seeds was confounded neither with total sown density nor species composition. Annually, a median of six vertebrate-dispersed species (quartiles: 4–8) arrive in

Table 1 Natural history of species added as seeds in this experiment, sorted by family

Species (family)	Seed rain (seeds m^{-2} $year^{-1}$)	Seed mass (g)	Adult density (ha^{-1})	Seed viability (%)
Duguetia quitarensis Benth. (Annonaceae)	0.36	0.41	3.65	55
Iriartea deltoidea Ruiz and Pav. (Arecaceae)	0.24	3.86	32.3	61
Socratea exorrhiza (Mart.) H. Wendl. (Arecaceae)	<0.01	3.44	0.08	88
Buchenavia grandis Ducke (Combretaceae)	<0.01	1.93	0.17	52
Diospyros pavonii (A. DC.) J.F. Macbr. (Ebenaceae)	0.02	0.61	0.17	42
Diospyros subrotata Hiern. (Ebenaceae)	<0.01	0.79	2.33	56
Trichilia pleeana (A. Juss.) C. DC. (Meliaceae)	0.13	0.86	3.35	25
Heisteria nitida Engl. (Olacaceae)	0.06	0.22	0.25	55

All species are canopy trees, except *I. deltoidea* and *S. exorrhiza*, which are canopy palms. Annual seed rain was estimated from fortnightly collections of 289 0.5-m^2 seed traps in the floodplain forest at Estación Biológica Cocha Cashu (EBCC) between January 2003 and 2006. Adult density was determined in three permanent plots totaling 10.25 ha in the EBCC trail system. Data on seed rain and adult density was kindly provided by Dr. John Terborgh. Seed mass is the air-dry weight of fresh seeds with all dispersal structures removed (see Paine and Beck 2007 for details). Seed viability is reported as the mean percentage of seeds germinating within 12 months of sowing in a shadehouse (see text for details) a square meter of forest floor, according to an ongoing seed-trapping project at EBCC (J. Terborgh, unpublished data). Seed additions in the richness experiment thus spanned the range of species richness that would annually arrive in the seed rain.

For both experiments, seeds were manually sprinkled over plots without disturbing the soil or leaf litter in any manner (as in Tilman 1997). The vegetation in each plot was then lightly shaken to ensure that no seeds remained perched atop their leaves. We began the density experiment in July 2003 with one species, *Diospyros pavonii,* as a pilot study immediately following the first census. The richness experiment began in May 2004. The density experiments for the remaining seven species also began at that time.

All plots were observed every 6 months between July 2003 and 2006, at the height of the dry season (July) and the wet season (January). At each census, we tagged all new individuals of the eight sown species that had germinated since the previous census. In addition, we monitored three aspects of the environment of each plot: the biotic neighborhood, soil moisture, and global light index. We evaluated two components of each plot's biotic neighborhood. At every July census, we tagged and measured seedlings and small saplings of free-standing nonsown woody species between 10 and 50 cm tall, excluding any that bore only cotyledons. For brevity, we refer to both seedlings and small saplings as "seedlings," regardless of whether they bore cotyledons. At all censuses except those of January 2004 and 2006, we estimated the percent cover of understory palms, ferns, herbaceous vegetation (mostly Marantaceae, Heliconiaceae, and Araceae), and coarse woody debris (limbs, trunks and roots $\geq$5 cm in diameter). We used the four estimates of cover, along with the density and species richness of nonsown seedlings, as descriptors of the biotic neighborhood of each plot. Soil moisture was evaluated gravimetrically to a depth of 5 cm in July 2004, and with time-domain reflectometry in July 2005, January 2005 and 2006 (depth: 15 cm; Field Scout TDR 200, Spectrum Technologies). Light availability was quantified as the global light index in Gap Light Analyzer 2.0 (Frazer et al. 2000) from canopy photos taken 67 cm above each plot in July 2004.

Identifying seedlings in such a diverse forest is a challenging task. To facilitate identification, we developed living and digital reference collections. Seeds of all available and identifiable species were sown into a greenhouse at EBCC between 2003 and 2005. Greenhouse-grown seedlings were studied and photographed repeatedly throughout their germination and establishment (all images are publicly available at http://manuplants.org). All tagged seedlings were identified in the field based on field characteristics and comparisons with greenhouse-grown individuals. Voucher photographs of unidentified seedlings were taken in the field. Post hoc comparisons of voucher photographs with the reference collection allowed further identifications to be made. Nevertheless, not all seedlings could be confidently identified to species. Recognizable seedlings that could not be assigned to a species were assigned to higher taxa, and/or assigned to morphotypes. We assume that morphotypes represent distinct species; every morphotype that we identified in the course of the project was indeed a distinct species. The identification of each seedling was re-evaluated and refined (if necessary) at each census. By the final census, in July 2006, 75, 18 and 4% of seedlings were identified to species, genus or family level, respectively; 3% remained unidentified.

Data analyses

Prior to assessing the effects of our seed-addition treatments, we checked for correlations between establishment conditions and geographic distance. Such correlations could confound their relationships with seedling community structure. We assessed the correlation between environmental conditions and geographic distance with Mantel tests (Mantel 1967; Gilbert and Lechowicz 2004). Mantel tests test the significance of the correlation between two matrices through permutation (McCune and Grace 2002). We calculated the Euclidian distances among plots and the scaled Euclidian dissimilarity of environmental conditions of each plot and calculated a Pearson correlation coefficient. Then we permuted the elements of one matrix, recalculated the correlation, and determined how often a stronger correlation would be expected by chance. The significances of observed correlations were assessed with 1,000 permutations.

For both experiments, we assessed the variance explained by seed-addition treatments, environmental conditions, and their one-way interactions. Data from the density experiment and the richness experiment were analyzed in an identical fashion, with only a change in response variables: stem density in the density experiment, and species richness, Shannon index of diversity, and species composition in the richness experiment. We assessed these relationships using three nested sets of seedlings: seedlings germinated from experimentally added seeds (henceforth "sown species"), all seedlings (both sown and nonsown) that recruited between July 2003 and 2004 (henceforth "2004 cohort"), and the entire seedling layer together (regardless of individual ages).

To quantify the effects of seed arrival and environmental conditions on seedling stem density and species diversity, we used stepwise model selection based on Akaike's information criterion (AIC, Burnham and Anderson 2002). We began with a multiple regression that included log-transformed sown density (or log-transformed sown species richness) and all measured environmental variables as predictor variables. Stepwise selection was used to select the best combination of variables (including one-way interactions) for predicting each response variable. Models were compared on the basis of AIC, and both forward and backward steps were possible (Venables and Ripley 1999). Multicollinearity was not an issue, as the variables representing environmental conditions were essentially uncorrelated (mean $|r| = 0.03$). Repeated measures analyses were not conducted, as not all environmental conditions were measured at all census periods. Instead, separate models were created for each census period. We extracted the variance explained by the seed-addition treatment, the sum of the variance explained by environmental conditions, and the variance explained by seed-addition by environment interactions using Type I sums of squares. Because seed arrival is the first stage of recruitment, the seed-addition variable always entered the model first. Thus, the percent variance explained by environmental conditions represented that which remained after accounting for seed arrival; i.e., the degree to which environmental conditions change the community structure generated by seed additions.

Finally, we quantified the effects of seed arrival and establishment conditions on seedling species composition. These analyses were performed for the richness experiment only. Mantel tests assessed the strength of (partial) correlations between sown species composition, environmental conditions, and seedling species composition (McCune and Grace 2002). The correlation between sown and seedling species composition was calculated with a Mantel test as described above. The relationship between environmental conditions and seedling species composition was calculated with a partial Mantel test, allowing an estimation of the partial correlation of environmental conditions and seedling species composition, after accounting for the correlation of sown and seedling species composition. Two sets of distance matrices for species composition were calculated, based on Chao-Jaccard and Sorenson (Bray–Curtis) dissimilarity indices (Chao et al. 2005). The use of different dissimilarity indices did not change the resulting correlations in any meaningful way, and for brevity, only results based on the Chao-Jaccard index are presented. We tested the significance of the (partial) correlations by permuting the seedling species composition matrix 1,000 times (holding the sown species composition and environmental similarity matrices constant), and recalculating the correlation coefficient. Mantel tests were repeated at each census period to assess changes over time in the relative importance of seed arrival and environmental conditions.

All analyses were performed in the R statistical package (R Development Core Team 2007). The MASS and vegan packages were used to implement the stepwise selection and Mantel tests, respectively (Venables and Ripley 1999; Oksanen et al. 2007).

Results

Over three years, 1,360 seedlings germinated from the 15,132 seeds that we added, constituting a mean germination rate of 9.0%. Germination varied by an order of magnitude among sown species, from 2.1% in *D. quitarensis* to 21.3% in *D. pavonii*. One hundred six seedlings of sown species recruited in control plots to which no seeds were experimentally added; a mean natural recruitment rate for each species of 0.008 seedlings m^{-2} $year^{-1}$. Three relatively common and fecund species recruited more than ten seedlings each into control plots, whereas no seedlings of *Socratea exorrhiza*, which is rare in the immediate study area, recruited naturally.

Correlations between environmental variables and geographic distances were very weak at all census periods (Mantel's $r \leq 0.09$), but were significant in July 2004, January 2005 and July 2005 ($P \leq 0.03$, Table 2). Seed-addition treatments were randomly assigned to plots, and should, therefore, have been uncorrelated with geographic distance. We tested this assumption with another set of Mantel tests, and found it to be valid (Mantel's $|r| \leq 0.01$, $P \geq 0.55$, Table 2).

Table 2 Mantel tests compared the correlation of geographic distance and environmental dissimilarity, and geographic distance and seed-addition treatments

	Environmental dissimilarity		Seed-addition treatments	
	Mantel's *r*	*P*	Mantel's *r*	*P*
July 2004	0.05	0.030	0.00	0.553
January 2005	0.09	0.001	0.00	0.686
July 2005	0.08	<0.001	−0.01	0.743
January 2006	0.03	0.087	−0.01	0.839
July 2006	0.03	0.166	−0.01	0.920

Significance was tested with 1,000 randomizations of the geographic distance matrix. Environmental dissimilarity was twice weakly and positively, but significantly, correlated with geographic distance. Seed-addition treatments were uncorrelated with geographic distance

We tagged 4,792 non-sown seedlings over the three-year study, representing 320 species, 185 genera and 66 families (Paine 2007). Each 1-m^2 study plot contained, on average, 6.4 ± 4.8 individual stems, and 5.0 ± 3.0 species (mean ± SD; Fig. 1). Nonsown seedling density peaked in July 2004 and declined thereafter. We did not monitor natural seed rain in the context of this study, but this degree of temporal variation in seedling stem density has been associated with fluctuations in seed rain density elsewhere in tropical South America (Norden et al. 2007).

Density experiment

Seed arrival was a better predictor of seedling stem density than were environmental conditions (Figs. 2, 3). During the early stages of the density experiment, sown density explained more of the variance in seedling density than did all environmental variables combined, indicating that seed arrival strongly limited seedling density [21 ± 2% vs. 11 ± 2% (mean ± SEM), respectively; $F_{(1,1)} = 15.63$, $P = 0.0002$]. For one species, *D. quitarensis*, seedling stem density was low and unrelated to sown density, suggesting that environmental conditions strongly affected the performance of this species. Models of seedling stem density explained an average of 39% of total variance, leaving 61% of the variance unexplained. The variance explained by sown density was initially substantial (mean over eight sown species ± SEM, January 2005: 27 ± 5%), and declined over time. Nevertheless, seed arrival still significantly limited stem density at the final census, explaining 16 ± 4% of the variance. At the final census, environmental conditions explained more variance in stem density than did sown density for three species. Statistical interactions among seed-addition treatments and environmental variables were frequently significant, but rarely substantial (6 ± 1%). In the 2004 cohort, seed-addition treatments explained only a small fraction of the variance in stem density (Fig. 3). Instead, the stem density of the 2004 cohort was best explained by environmental variables ($R^2 > 0.66$). Neither seed arrival nor environmental conditions explained the stem density of the entire seedling layer ($R^2 < 0.09$).

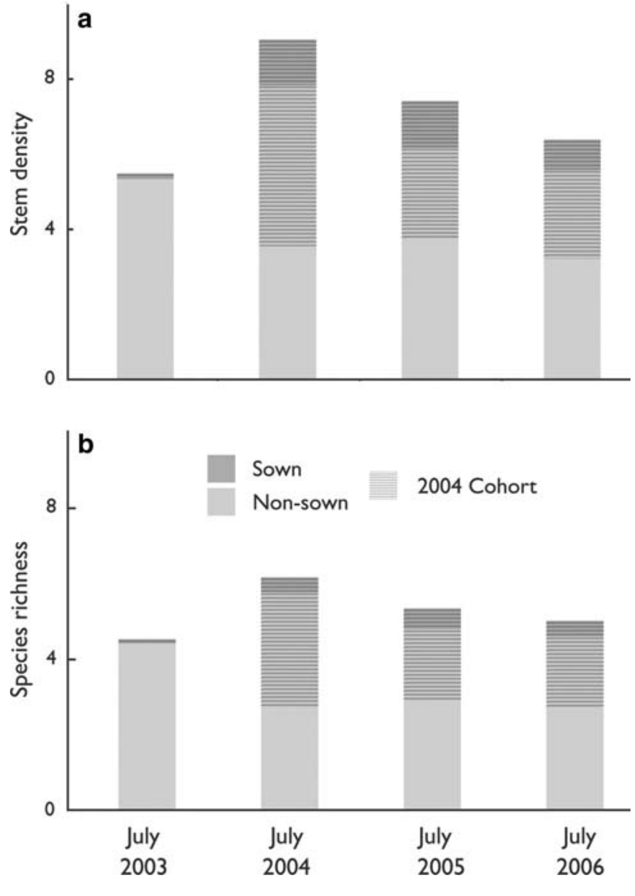


Fig. 1 **a** Stem density and **b** species richness of sown and nonsown seedlings over three years. Individuals and species that constitute the 2004 cohort are *stippled*. The 2004 cohort comprised both sown and nonsown species

Richness experiment

The species richness of seedlings germinated from sown seeds was strongly affected by seed arrival, but was also influenced by environmental heterogeneity (Figs. 4, 5). In January 2005, sown species richness explained 27% of the variance in seedling species richness; environmental variables explained 2%. Eighteen months later, at the final census, the variances explained by seed arrival and environmental conditions had roughly equalized, at 8 and 11%, respectively. The analyses of the Shannon diversity index closely paralleled those of species richness, and are not shown. The species richness of the 2004 cohort was well explained by environmental variables ($R^2 > 0.42$), and not by sown species richness ($R^2 < 0.03$). Neither seed-addition treatments nor environmental variables explained the species richness of the entire seedling layer ($R^2 < 0.06$).

Seedling species composition was also assessed in the richness experiment. Seedling species composition was more tightly correlated with the species composition of added seeds than with environmental conditions (Table 3). The correlation between sown species composition and seedling species composition was strongest in July 2005 (Mantel's $r = 0.41$), and weakened over the following two years ($r = 0.29$). Partial correlations between environmental conditions and the species composition of sown seedlings, after accounting for seed arrival, were significant at no time ($|r| \leq 0.02$, $P > 0.094$). The species composition of the 2004 cohort was significantly, though weakly, correlated with seed-addition treatments. The strength of this correlation also declined between July 2004 and 2006, from

0.14 to 0.03. Even so, the correlation of the 2004 cohort species composition and environmental conditions was weaker, and only significant in July 2005 ($r = 0.05$, $P = 0.007$). Looking at the entire seedling layer, seedling species composition was more tightly correlated with sown species composition than with environmental variables (Mantel's r_{arrival}: 0.23–0.28, Mantel's $r_{\text{environment}}$: 0.04–0.08).

Discussion

Seed arrival limits density, diversity, and shapes species composition

The density experiment showed the arrival of seeds to affect strongly the density of tree seedlings in a highly diverse neotropical forest. This outcome was expected, and is consistent with the results of previous seed-addition studies (Makana and Thomas 2004; Svenning and Wright 2005), and observational studies of adult and seedling community structure (Webb and Peart 2001; Terborgh et al. 2008). The richness experiment further showed seed arrival to limit seedling species richness, a novel result in tropical forests. This finding parallels results from temperate grasslands (Tilman 1997; Foster and Tilman 2003; Foster et al. 2004), and is consistent with tropical forest studies that have inferred dispersal limitation to be strong, through seed trapping (Clark et al. 1998, Clark et al. 1999, Hubbell et al. 1999; J. Terborgh, unpublished data), or observations of adult distributions at local (Condit et al. 2000; Seidler and Plotkin 2006) and regional scales (Condit et al. 2002; Tuomisto et al. 2003).

A novel result of the richness experiment is that even a single seed arrival event can shift the species composition of the seedling layer. This finding is robust, in that the species composition of experimentally added seeds was significantly and positively correlated with not only the composition of the seedlings that germinated from those seeds, but also with the seedlings comprising the 2004 cohort, and with the entire seedling layer (Table 3). In this diverse forest, the arrival of a seed of a rare species to a small plot can make its composition dissimilar from that of neighboring plots, moving it to a remote part of multidimensional "species-composition space." Though common species were included in our pool of sown species, others, such as *Buchenavia grandis* and *S. exorrhiza*, are locally rare as adults and germinated many seedlings. Thus, the one-time seed additions we performed were sufficient to shift the species composition of the entire seedling layer, and this compositional shift persisted for two years following germination.

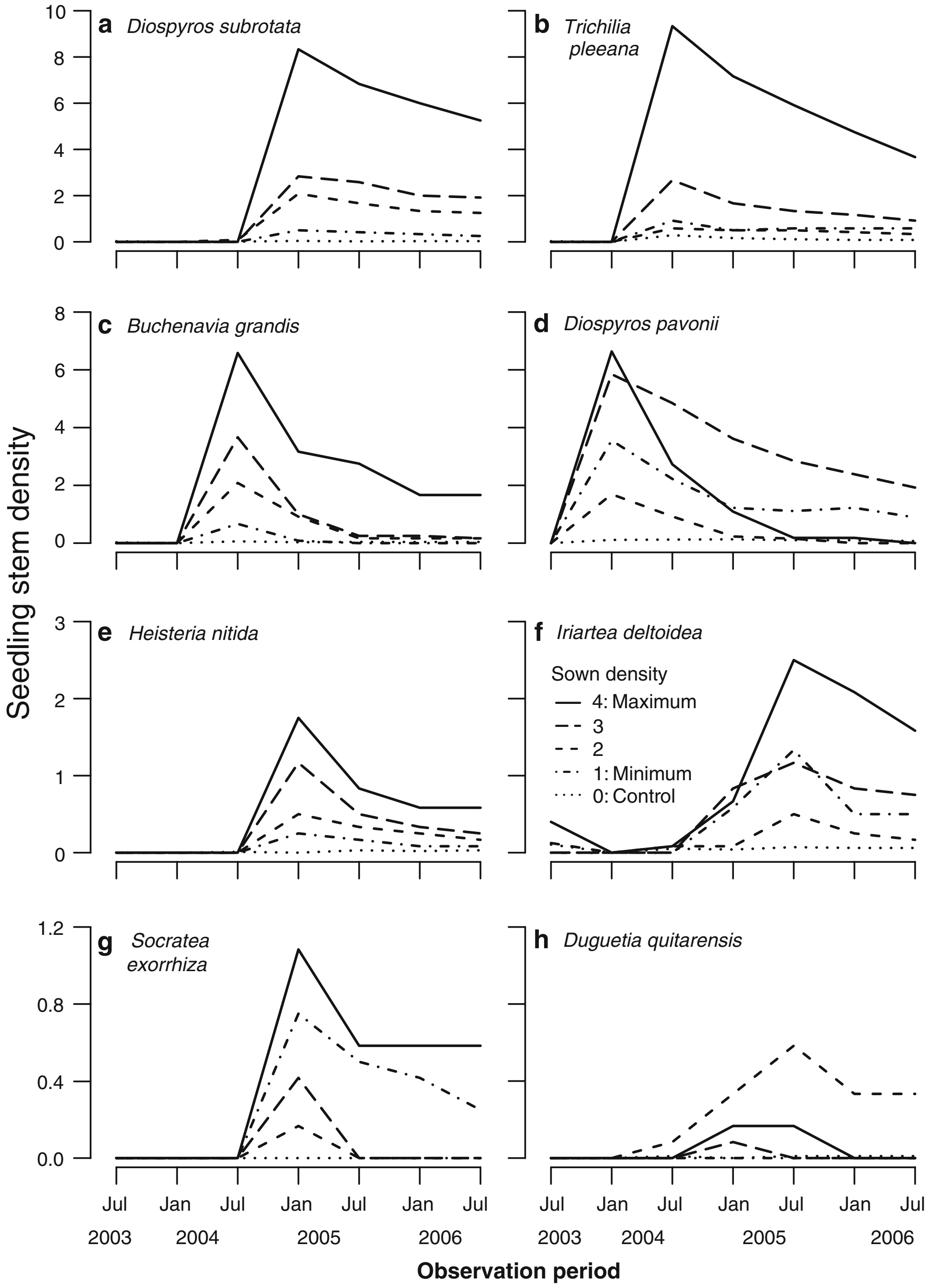
At face value, the finding that seed arrival influences seedling community composition is unsurprising. On the other hand, decades of research in tropical forests have supported the view that recruitment patterns are primarily generated by environmental conditions, including—among other factors—the availability of light, water, and nutrients, competition and seed predation (Nicotra et al. 1999; Lewis and Tanner 2000; Montgomery and Chazdon 2002; Engelbrecht and Kursar 2003; Paine and Beck 2007). With effects on density, diversity and species composition, seed arrival also affects the community structure of the seedling layer.

Within-site variation in the roles of dispersal assembly and niche assembly

After showing seed limitation to be strong, we partitioned the variance in seedling community structure to infer the roles of dispersal assembly and niche assembly on each subset of the community over time. Put simply, seed arrival and environmental conditions imposed complex and dynamic forces on the seedling community. Combining the results of the density and richness experiments, seed-addition treatments initially explained more variance in stem density, diversity and species composition than did environmental heterogeneity (Fig. 3, 5, and Table 3), suggesting the dynamics of the youngest seedlings to be consistent with dispersal assembly. Nevertheless, the variance explained by the seed-addition treatments decreased through time, indicating an increase in the importance of niche assembly through time. Broad conclusions are complicated by results from the 2004 cohort and the entire seedling layer, in which heterogeneity in environmental conditions explained far more variance in stem density and species richness than did seed arrival, even though their species compositions were significantly correlated with those of the seed-addition treatments. In the following paragraphs, we highlight some salient themes in these results.

One would expect the inferred role of niche assembly to increase through ontogeny. Dispersal is a singular event for sessile organisms; it establishes the initial distribution of individuals and species. Thereafter, bouts of ecological filtering, such as those imposed by drought, occur at irregular intervals (e.g., Delissio and Primack 2003). In longitudinal studies like ours which track a cohort through time (e.g., Makana and Thomas 2004; Svenning and Wright 2005), the inferred role of niche assembly will increase over time as incidents of ecological filtering accumulate. Simultaneously, longitudinal studies of experimentally sown cohorts may underestimate the long-term effects of seed arrival. As in this study, a one-time addition of seeds may not affect the density or diversity of the entire seedling layer. Even so, if seed arrival frequently affects the structure of cohorts, its influence will accrue in community-wide stem density, species richness, and species composition. In

Fig. 2 Density of recruited seedlings of eight sown species over time, by sown density, sorted by the abundance of recruited seedlings. Each *line* represents the mean stem density in plots where there were a variable number of seeds in July 2004 (July 2003 for *Diospyros pavonii*). Seed-addition treatments increased seedling density for seven of eight species. Note that the scales of the *y*-axes differ



this context, it is important to note that our one-time seed additions slightly increased the density of the 2004 cohort and affected that cohort's species composition. This suggests that the effects of multiple seed-arrival events may accumulate in the structure of the seedling layer as a whole. "Press" experiments, in which seeds are repeatedly added to plots over time, would be essential to assess this hypothesis.

The roles of dispersal and niche differentiation may also vary among subsets of a community. Among seven of our eight sown species, stem density was strongly limited by seed arrival, indicating the early dynamics of these species to be consistent with theories of dispersal assembly. The eighth, *D. quitarensis*, was affected primarily by heterogeneity in environmental conditions. The low recruitment rate of *D. quitarensis* is not simply a result of low viability, as 55% of its seeds germinated in the shadehouse (Table 1). It is important to note that all eight of the species sown in this study are vertebrate dispersed and relatively large-seeded. A recent meta-analysis suggests establishment limitation may be more intense on species with smaller seeds and abiotic dispersal (Clark et al. 2007). Although the generality of

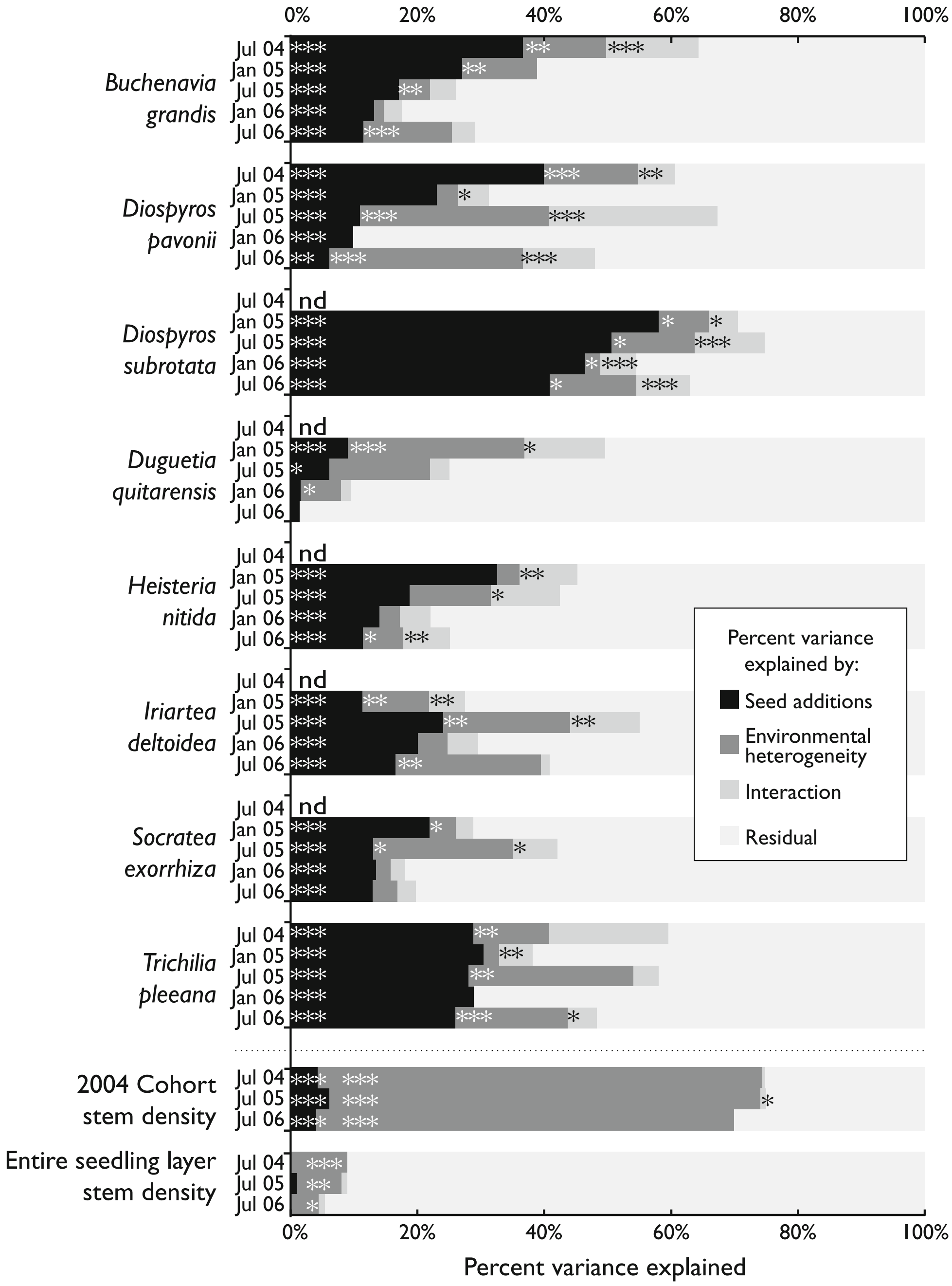


Fig. 3 Percent variance in the stem density of seedlings germinated from sown seeds was better explained by seed-addition treatments than by environmental heterogeneity. Each *bar* represents the percent variance explained by seed-addition treatment, environmental heterogeneity, their interactions, or unexplained (residual). *Bars* are grouped by species and, within species, sorted by observation period. The lowermost two sets of *bars* represent the effects of sown density on the density of the 2004 cohort and the entire seedling layer density, respectively. The environmental heterogeneity variance represents the variance explained by all environmental variables. Only species that germinated rapidly upon sowing were analyzed in July 2004; *nd* indicates that no data were available. * $P < 0.01$; ** $P < 0.001$; *** $P < 0.0001$

our results for smaller-seeded taxa is presently unknown, our results are consistent with the findings of the meta-analysis: *D. quitarensis* had the second-smallest seeds among the sown species.

Finally, the breadth of environmental conditions investigated, together with the number of variables assessed to quantify them, are likely to limit the imputed role of niche assembly. In this study, the range of environmental conditions to which seedlings were exposed was limited by experimental design to the habitat to which the great majority of seeds arrive: relatively undisturbed closed-canopy forest. Even so, the range of environmental conditions experienced by seedlings in the understory of closed-canopy forest is not trivial (Montgomery and Chazdon 2002). All studies that seek to quantify the degree of niche differentiation are susceptible to a fundamental criticism: the variables utilized by the investigators to summarize the environment may fail to adequately represent the conditions experienced by the studied organisms. This charge may be levied against the present study: many environmental variables remained unquantified, including (among others) soil nutrients. We doubtlessly would have found a more prominent role of environmental variation in predicting community structure had we measured a wider variety

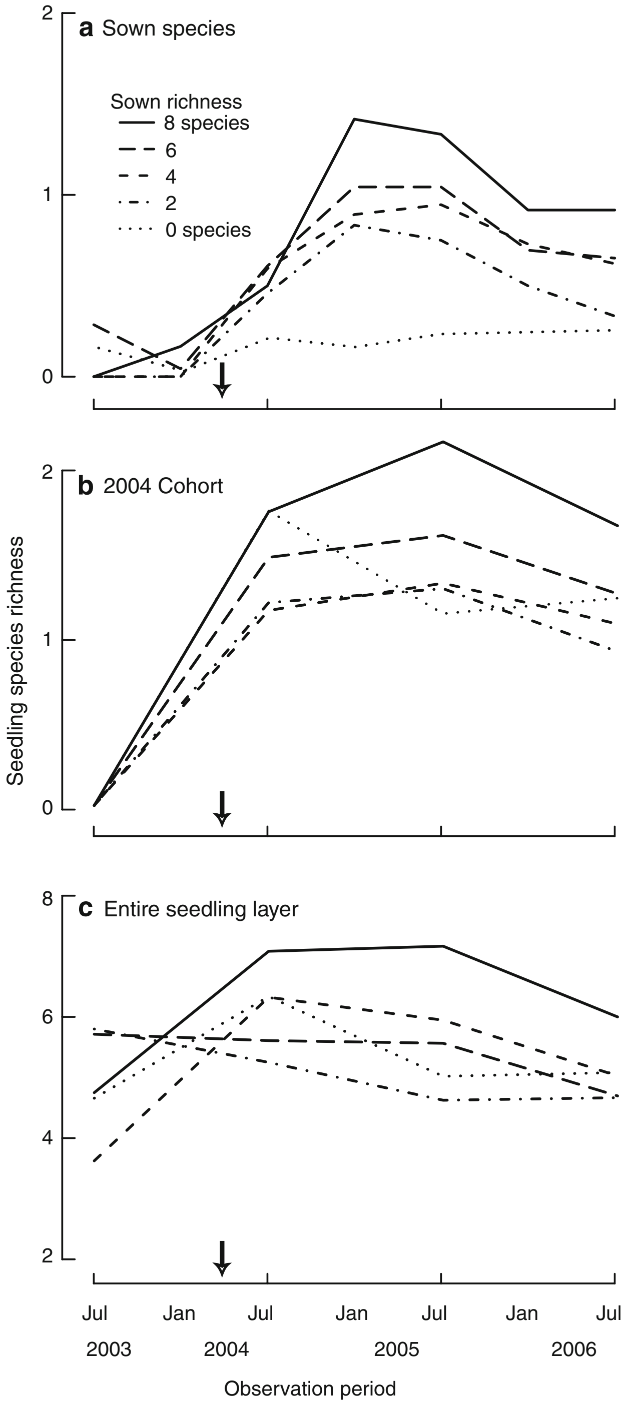


Fig. 4 Species richness of sown species (**a**), the 2004 cohort (**b**), and entire seedling layer (**c**). Each *line* represents the species richness in plots where zero, two, four, six or eight species were added as seeds in July 2004. Adding a diverse set of seeds significantly increased the species richness of the subsequent recruits, but had limited effects on cohort richness and the richness of the entire seedling layer, since their inter-plot variance was great. *Arrows* indicate the timing of seed additions. Note that the scales of the *y*-axes vary

of environmental variables or extended the experiment across broader environmental gradients to include, for example, flooded swales or tree-fall gaps.

Among-site variation in the roles of dispersal assembly and niche assembly

Among-site differences in the intensity of ecological filtration will affect the relative importance of dispersal assembly and niche assembly. Where ecological filtration is relatively weak, dispersal assembly may dominate community dynamics. Just as neutral drift is expected for genomes in the absence of selection (Kimura 1983), dispersal assembly may be expected for communities in the absence of ecological filtering. Similarly, just as selection nonrandomly alters the relative abundance of alleles, ecological filtering may fall more strongly upon individuals of some species than upon others, thus driving a community toward niche assembly. Accordingly, communities subject to intense predation or disturbance regimes appear resistant to stochasticity in community structure, and are well fitted by niche-assembly models (McGill 2003; McGill et al. 2006; Chase 2007). On the other hand, communities subject to relatively benign environments and moderate rates of predation and disturbance appear more sensitive to stochasticity, and are well fitted by models of dispersal assembly (Fukami 2004).

Despite the simplicity of our manipulations, they afforded a degree of resolution into dispersal assembly and niche assembly unavailable to observational studies. The paired seed trap–seedling plot design employed in many observational studies (e.g., Harms et al. 2000; Norden et al. 2007) has a key weakness: the seed rain captured by the seed traps is not identical to that which arrives in the neighboring seedling plots. In contrast, by experimentally adding seeds, we knew with greater (though still not complete) certainty the composition of the seed rain arriving in our seedling plots. Furthermore, by conducting independent manipulations of the density and the species richness of the seed rain, we were able to disentangle their influences, which are frequently confounded in natural seed rain. The combination of seed additions and variance partitioning permitted a detailed dissection of the interacting roles played by dispersal assembly and niche assembly within a single diverse community.

Conclusion

This study showed that seed arrival not only limits the density and diversity but that it also affects the species composition of the seedling layer of a diverse tropical forest. The complex roles played by the main forces structuring the seedling layer, dispersal assembly and niche assembly, were illustrated by a variance-partitioning analysis. The relationship between seed-addition treatments and community structure varied among subsets of the seedling layer, and became weaker over the course of the experiment. We

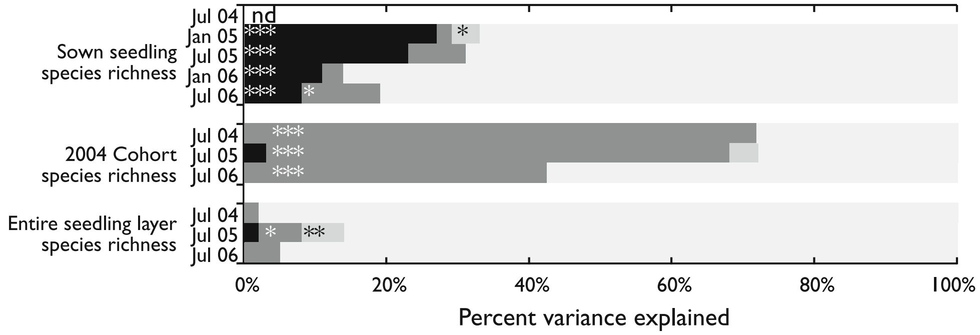


Fig. 5 Variance in species richness of sown seedlings, the 2004 cohort, and the entire seedling layer was explained by seed additions, environmental conditions, and their interactions. Only three sown species had germinated by July 2004, so the species richness of sown seedlings was analyzed only after January 2005. Abbreviations are as in Fig. 3

Table 3 Seed-addition treatments better predicted seedling species composition than does environmental heterogeneity, even two years after germination

	Seed arrival		Environmental heterogeneity after arrival	
	Mantel's *r*	*P*	Mantel's *r*	*P*
Sown species				
July 2004	nd	nd	nd	nd
January 2005	0.41	<0.001	−0.02	0.878
July 2005	0.30	<0.001	0.02	0.094
January 2006	0.32	<0.001	0.01	0.255
July 2006	0.29	<0.001	−0.02	0.902
2004 cohort				
July 2004	0.14	<0.001	0.03	0.109
July 2005	0.05	<0.001	0.05	0.007
July 2006	0.03	0.005	0.01	0.336
Entire seedling layer				
July 2004	0.28	<0.001	0.05	0.014
July 2005	0.24	<0.001	0.08	0.000
July 2006	0.23	<0.001	0.04	0.021

The correlation of the species composition of sown seeds with the species composition of three subsets of the seedling layer was assessed with Mantel tests. The correlation of post-dispersal environmental conditions with the species composition of the three subsets of the seedling layer was assessed with partial Mantel tests that controlled for the species composition of the sown seeds

thus infer dispersal assembly to play a relatively strong role in the initial stages of seedling recruitment, but that role is likely to weaken over time for most species. When (or if) niche assembly assumes a dominant role, its timing remains unclear. We note that our scope for inference is limited by several factors; prominent among them is the fact that we studied only the earliest stages of recruitment in long-lived trees. If the effects of seed arrival persist through time, despite ongoing ecological filtering, stochastic processes may play an important role in generating the structure and dynamics of diverse tropical forests.

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References

Arii K, Lechowicz MJ (2002) The influence of overstory trees and abiotic factors on the sapling community in an old-growth Fagus–Acer forest. Ecoscience 9:386–396

Bell G (2001) Neutral macroecology. Science 293:2413–2418

Bloor JMG, Grubb PJ (2003) Growth and mortality in high and low light: trends among 15 shade-tolerant tropical rain forest tree species. J Ecol 91:77–85

Burnham KP, Anderson DR (2002) Model selection and inference: a practical information-theoretic approach, 2nd edn. Springer, New York

Chao A, Chazdon RL, Colwell RK, Shen T-J (2005) A new statistical approach for assessing similarity of species composition with incidence and abundance data. Ecol Lett 8:148–159

Chase JM (2003) Community assembly: when should history matter? Oecologia 136:489–498

Chase JM (2007) Drought mediates the importance of stochastic community assembly. Proc Natl Acad Sci USA 104:17430–17434

Chase JM, Leibold MA (2003) Ecological niches: linking classical and contemporary approaches. University of Chicago Press, Chicago

Clark JS, McLachlan JS (2003) Stability of forest biodiversity. Nature 423:635–638

Clark JS, Macklin E, Wood L (1998) Stages and spatial scales of recruitment limitation in southern Appalachian forests. Ecol Monogr 68:213–235

Clark CJ, Poulsen JR, Levey DJ, Osenberg CW (2007) Are plant populations seed limited? A critique and meta-analysis of seed addition experiments. Am Nat 170:128–142

Condit R et al (2000) Spatial patterns in the distribution of tropical tree species. Science 288:1414–1418

Davies TJ, Barraclough TG, Chase MW, Soltis PS, Soltis DE, Savolainen V (2004) Darwin's abominable mystery: insights from a supertree of the angiosperms. Proc Natl Acad Sci USA 101:1904–1909

Delissio LJ, Primack RB (2003) The impact of drought on the population dynamics of canopy-tree seedlings in an aseasonal Malaysian rain forest. J Trop Ecol 19:489–500

Engelbrecht BMJ, Kursar TA (2003) Comparative drought-resistance of seedlings of 28 species of co-occurring tropical woody plants. Oecologia 136:383–393

Etienne RS, Olff H (2004) A novel genealogical approach to neutral biodiversity theory. Ecol Lett 7:170–175

Fargione J, Brown CS, Tilman D (2003) Community assembly and invasion: an experimental test of neutral versus niche processes. Proc Natl Acad Sci USA 100:8916–8920

Foster SA, Janson CH (1985) The relationship between seed size and establishment conditions in tropical woody plants. Ecology 66:773–780

Foster BL, Tilman D (2003) Seed limitation and the regulation of community structure in oak savanna grassland. J Ecol 91:999–1007

Frazer GW, Canham CD, Lertzman KP (2000) Gap light analyzer (GLA), version 2.0: image-processing software to analyze true-color, hemispherical canopy photographs. Bull Ecol Soc Am 81:191–197

Fukami T (2004) Assembly history interacts with ecosystem size to influence species diversity. Ecology 85:3234–3242

Gilbert B, Lechowicz MJ (2004) Neutrality, niches, and dispersal in a temperate forest understory. Proc Natl Acad Sci USA 101:7651–7656

Gravel D, Canham CD, Beaudet M, Messier C (2006) Reconciling niche and neutrality: the continuum hypothesis. Ecol Lett 9:399–409

Harms KE, Wright SJ, Calderon O, Hernandez A, Herre EA (2000) Pervasive density-dependent recruitment enhances seedling diversity in a tropical forest. Nature 404:493–495

Harms KE, Condit R, Hubbell SP, Foster RB (2001) Habitat associations of trees and shrubs in a 50-ha Neotropical forest plot. J Ecol 89:947–959

Harpole WS, Tilman D (2006) Non-neutral patterns of species abundance in grassland communities. Ecol Lett 9:15–23

Holdridge LR (1947) Determination of world plant formations from simple climatic data. Science 105:367–368

Hubbell SP (2001) The unified theory of biodiversity and biogeography. Princeton University Press, Princeton

Hubbell SP et al (1999) Light gap disturbances, recruitment limitation, and tree diversity in a neotropical forest. Science 283:554–557

John R et al (2007) Soil nutrients influence spatial distributions of tropical tree species. Proc Natl Acad Sci USA 104:864–869

Karst J, Gilbert B, Lechowicz MJ (2005) Fern community assembly: the roles of chance and the environment at local and intermediate scales. Ecology 86:2473–2486

Kimura M (1983) The neutral theory of molecular evolution. Cambridge University Press, London

Lewis SL, Tanner EVJ (2000) Effects of above- and belowground competition on growth and survival of rain forest tree seedlings. Ecology 81:2525–2538

Makana J-R, Thomas SC (2004) Dispersal limits natural recruitment of African mahoganies. Oikos 106:67–72

Mantel N (1967) The detection of disease clustering and a generalized regression approach. Cancer Res 27:209–220

McCune B, Grace JB (2002) Analysis of ecological communities. MjM Software Design, Gleneden Beach

McGill BJ (2003) A test of the unified neutral theory of biodiversity. Nature 422:881–885

McGill BJ, Hadly EA, Maurer BA (2005) Community inertia of Quaternary small mammal assemblages in North America. Proc Natl Acad Sci USA 102:16701–16706

McGill BJ, Maurer BA, Weiser MD (2006) Empirical evaluation of neutral theory. Ecology 87:1411–1423

Montgomery RA, Chazdon RL (2002) Light gradient partitioning by tropical tree seedlings in the absence of canopy gaps. Oecologia 131:165–174

Nicotra AB, Chazdon RL, Iriarte SVB (1999) Spatial heterogeneity of light and woody seedling regeneration in tropical wet forests. Ecology 80:1908–1926

Norden N et al (2007) Is seedling dynamics determined by environment or by seed arrival? A test in a neotropical forest. J Ecol 95:507–516

Oksanen J, Legendre P, O'Hara B, Stevens MHH (2007) vegan: Community Ecology Package. R package 1.8–8. http://cran.r-project.org/; http://r-forge.r-project.org/projects/vegan/

Paine CET (2007) Ecological factors affecting the diversity of tropical tree seedlings. Louisiana State University, Baton Rouge

Paine CET, Beck H (2007) Seed predation by neotropical rainforest mammals increases diversity in seedling recruitment. Ecology 88:3076–3087

Pitman NCA et al (2002) A comparison of tree species diversity in two upper Amazonian forests. Ecology 83:3210–3224

Rosenzweig ML (1995) Species diversity in space and time. Cambridge University Press, London

Seidler TG, Plotkin JB (2006) Seed dispersal and spatial pattern in tropical trees. PLoS Biol 4:2132–2137

Shipley B, Vile D, Garnier E (2006) From plant traits to plant communities: a statistical mechanistic approach to biodiversity. Science 314:812–814

Svenning JC, Wright SJ (2005) Seed limitation in a Panamanian forest. J Ecol 93:853

R Development Core Team (2007) R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna

Terborgh J (1990) An overview of research at Cocha Cashu Biological Station. In: Gentry AH (ed) Four neotropical forests. Yale University Press, New Haven, pp 48–59

Terborgh J et al (2008) Tree recruitment in an "empty" forest. Ecology 89:1757–1768

Tilman D (1997) Community invasibility, recruitment limitation, and grassland biodiversity. Ecology 78:81–92

Tilman D (2004) Niche tradeoffs, neutrality, and community structure: a stochastic theory of resource competition, invasion, and community assembly. Proc Natl Acad Sci USA 101:10854–10861

Tuomisto H, Ruokolainen K, Yli-Halla M (2003) Dispersal, environment, and floristic variation of western Amazonian forests. Science 299:241–244

Turnbull LA, Crawley MJ, Rees M (2000) Are plant populations seed-limited? A review of seed sowing experiments. Oikos 88:225–238

Venables WN, Ripley BD (1999) Modern applied statistics with S-Plus. Springer, New York

Volkov I, Banavar JR, Hubbell SP, Maritan A (2003) Neutral theory and relative species abundance in ecology. Nature 424:1035–1037

Volkov I, Banavar JR, He FL, Hubbell SP, Maritan A (2005) Density dependence explains tree species abundance and diversity in tropical forests. Nature 438:658–661

Webb CO, Peart DR (2001) High seed dispersal rates in faunally intact tropical rain forest: theoretical and conservation implications. Ecol Lett 4:491–499