Fungi, insects, and mammals differentially impact the diversity of tropical tree seedling communities

Running Title: Natural enemies contribute to tree diversity

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Statement of Authorship

All authors formulated and designed the study, KH and HB established the experiment, all authors collected data, KH and CETP performed the data analysis, KH wrote the first draft and all authors contributed to the revisions.

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Should this manuscript be accepted, all data supporting these results will be archived in a public repository.

Abstract

Natural enemies have been implicated as agents of negative density dependence (NDD) in tropical forests, but their relative contributions to NDD and thus the maintenance of diversity are largely unknown. We assessed the influence of natural enemies on density-dependent interactions among tropical seedlings, monitoring survival and relative growth rates in plots that excluded fungal pathogens, insects, small mammals, or large mammals. Only fungal pathogens caused density dependent mortality, and their exclusion reduced species diversity. Insects reduced relative growth rates when stem density was high, but because this was not driven by conspecific density, insects had little effect on species diversity. Mammals did not cause NDD interactions. We conclude that both fungal pathogens and insects drive NDD interactions among seedlings. Even so, only fungal pathogens increased species diversity, and consequently contribute critically to the structure of tropical tree communities.

Introduction

The maintenance of tropical tree diversity has piqued the interest of community ecologists for many years (Janzen 1970, Connell 1971, Wright 2002, Terborgh 2012). Avoiding competitive exclusion is central to the persistence of rare species (Chesson 2000). Although mechanisms of species coexistence are extensively debated in the literature (reviewed by Wright 2002), negative density dependence (NDD) is one that has received extensive empirical support (Harms et al. 2000, Liu et al. 2012, Zhu et al. 2015). NDD promotes coexistence by increasing individual performance at low conspecific density, thereby favouring rare species (Chesson 2000). Much of tree community structure is determined during the seedling stage, when mortality rates are high and non-random (Green et al. 2014).

Mechanisms that drive NDD are widely debated (Wright 2002, Terborgh 2012). NDD was once thought to be driven by competition among neighbouring plants for shared resources (Yoda et al. 1963), but little evidence for competition among tropical rainforest seedlings has emerged (Paine et

al. 2008, Svenning et al. 2008). Multiple studies, on the other hand, have shown natural enemies to cause NDD by disproportionately allowing rare species to succeed (Packer and Clay 2000, Bell et al. 2006, Mangan et al. 2010, Terborgh 2012, Bagchi et al. 2014, Paine et al. 2016).

Few studies to date have elucidated the mechanisms by which NDD occurs (Packer and Clay 2000, Bell et al. 2006, Liu et al. 2012), and even fewer have assessed the relative importance of the taxa of natural enemies that cause it (Bagchi et al. 2014, Gripenberg et al. 2014, Paine et al. 2016). Bagchi et al. (2014) found both fungal pathogens and insect herbivores to cause NDD among seedlings in Belize, although only fungi affected species diversity. Paine et al. (2016) evaluated the contributions of mammals to NDD during seedling recruitment in Peru, finding that small and medium mammals, but not large mammals, affected mortality and thus diversity. We build upon these studies by evaluating the relative contributions of fungi, insects, small mammals, and large mammals to NDD in seedling performance.

The most commonly implicated natural enemies are pathogenic fungi or oomycota (referred to as fungal pathogens hereafter; Bell et al. 2006, Mangan et al. 2010), since they are commonly highly host specific, or have a limited host range (Gilbert et al. 2012). Herbivorous insects can also cause NDD (Bagchi et al. 2014, Fricke et al. 2014), though their contribution is debated (Bagchi et al. 2010, Gripenberg et al. 2014). Furthermore, granivorous mammals can cause NDD (Theimer et al. 2011, Beck et al. 2013), but studies have shown limited consequences for diversity (except see Paine et al. 2016). Considering the defaunation occurring in tropical forests worldwide, there are serious implications regarding the contribution of mammals to the generation of species diversity (Dirzo et al. 2014). The loss of large vertebrate seed or seedling predators may alter plant community composition (Theimer et al. 2011) and reduce species diversity (Paine et al. 2016). We expect each of the four taxa to contribute to NDD, as all consume seeds and seedlings (Packer and Clay 2000, Paine et al. 2016). We further expect that smaller natural enemies will make larger contributions (Bagchi et al. 2014), because these are exponentially more abundant, and tend to be host specific (Gilbert et al. 2012). The effects of natural enemies on plant communities may interact. For example, García-Guzman and Dirzo (2001) found that insect herbivores accelerate rates of pathogen infection by

creating wounds through which pathogens can attack. To test this we experimentally combined treatment groups, expecting to find stronger effects on NDD in combined herbivore (insects or mammals) and fungal pathogen exclusion plots.

We assess two aspects of seedling performance; mortality and relative growth rate (RGR). Because generalist natural enemies impact the heterospecific community, we expect seedling RGR to be more strongly affected by generalist natural enemies such as insects (Novotny et al. 2002) and mammals (Beck et al. 2013). We expect mortality, in contrast, to be more strongly driven by conspecific crowding (Comita et al. 2010, Hazelwood et al. in prep), and we therefore expect fungi to more strongly contribute to NDD in mortality (Bagchi et al. 2014, Gilbert et al. 2012). This study aims to determine the degree to which different taxa (fungi, insects and mammals) contribute to NDD and consequently maintain diversity.

Methods

Study Site

This study was carried out at the Cocha Cashu Biological Station (CCBS). CCBS is located in Amazonian South-East Peru in lowland tropical rain forest, at 11°51'S, 71°19'W, 350 m elevation, and 2200 mm average annual precipitation. The site is in a highly diverse and remote area of the Manu National Park, with over 350 tree species with a diameter >10 cm DBH (Foster 1990), and has experienced minimal hunting, and no logging or mining during the last century (Terborgh, 1990).

Experimental Design

Circular 1 m^2 experimental plots were located in a random blocked design throughout a 4 km^2 trail system in mature floodplain rain forest. 24 plots were spaced between 5 and 10 metres apart in each of 24 blocks, avoiding trails and new treefalls. Within each experimental plot, all woody seedlings >10 cm high and <1 cm dbh were identified, tagged, and had their height and number of leaves recorded over four censuses from 2014 to 2017. Censuses were carried out 294 to 330 days apart. Within each

104 block, 8 plots were randomly selected for the application of 8 taxon-excluding treatments: none (a
105 control), fungi, insects, large mammals, and all mammals, and combined fungi and insects, all
106 mammals and fungi, and all mammals and insects.

107 We applied fungicide and insecticide to assess the effects of fungi and insects on seedlings,
108 respectively. The fungicide Amistar (Syngenta Ltd, active ingredient: azoxystrobin) provides a broad
109 spectrum of protection against fungal attack, has low toxicity in non-target organisms, and was found
110 to be effective by Bagchi et al. (2014). The insecticide Karate (Syngenta Ltd. active
111 ingredient: lambda cyhalothrin), provides protection against a broad spectrum of insect herbivores,
112 leaving low rates of residue and has low impact on non-target organisms. Pesticides were applied
113 according to manufacturer's instructions, mixing 1.25 ml of pesticide with 1 litre of water, and
114 applying 50 ml of the mixture to 1 m^2 plot with spray bottles. Pesticides were applied to treatment
115 plots every 10 to 14 days, in equal amounts over 31 months, with some treatment breaks when it was
116 logistically impossible to apply treatments (max 1 month). Control plots were misted with an amount
117 of water equivalent to that applied to pesticide plots.

118 Mammals were excluded from the study plots using 2x2 m wire mesh exclosures, which were 150 cm
119 high and included a 50 cm buffer around the perimeter of each plot to reduce potential germination
120 bias from perching birds. Half of the mammal exclosures allowed entry of small mammals. They had
121 15 by 15 cm holes cut into the base of the mesh large enough to allow agoutis (*Dasyprocta sp.*) or
122 smaller rodents to enter, but too small for peccaries (*Tayassu sp.*), deer (*Mazama americana*), or tapir
123 (*Tapirus terrestris*). Exclosures that excluded all mammals were constructed flush to the ground.

124 Non-experimental seedling plots where no treatment was applied (16 additional plots in each block)
125 were also monitored. Mortality and RGR were modelled against conspecific and heterospecific
126 neighbourhood density in control plots (where water was applied) and non-treatment plots (where no
127 water was applied). Significant differences were found between control and non-treatment plots
128 neither in mortality models (conspecific density: $P = 0.68$, heterospecific density: $P = 0.61$) nor in

RGR models (conspecific density: $P = 0.16$, heterospecific density: $P = 0.79$). Therefore, we combined control and non-treatment plots for all analyses.

Data Analysis

In 4 censuses conducted over 31 months, 5164 seedlings were monitored from 237 unique species or morphospecies, plus 787 individuals that were unidentified or identifiable only to family; the latter were excluded from the analysis. Rarer species that do not occur frequently enough to have many conspecific neighbours were removed from the dataset by including only species that had a mean conspecific neighbourhood density of >1 individual/m^2. The resulting dataset consisted of 174 unique species and morphospecies, with 71% identified to species level and the remaining 29% identified to genus level. Seedlings excluded from the dataset were counted among heterospecific neighbours.

Conspecific or heterospecific neighbourhood density, and neighbouring seedling size may all impact mortality or RGR. We therefore calculated a neighbourhood crowding index for each seedling (Canham et al. 2004). Neighbourhood crowding indices (NCI) were calculated per plot and census as:

$$NCI_k = \Sigma(height_n / height_k)$$

Where *k* is the focal individual, and *n* are neighbouring seedlings. Indices were calculated separately for conspecific neighbours and heterospecific neighbours.

To assess the effects of fungi, insects, and mammals as sources of mortality, we used generalised linear mixed effect models with binomial errors and a complementary log-log link. Mortality was predicted by interacting conspecific crowding and treatment, and heterospecific crowding and treatment. Larger seedlings experience lower mortality risk (Green et al. 2014, Paine et al. 2012), therefore log-transformed seedling height entered these models as a fixed effect. An offset of the log-transformed time between censuses was included to account for differing census intervals and to yield parameter estimates in units of years. RGR was assessed using linear mixed effect models, in which log-transformed seedling height was predicted by seedling age, and model coefficients represented relative growth rates (Paine et al. 2012). Seedling RGR was assumed to be exponential since growth

154 rates do not decrease until trees reach a height far exceeding 1 m (Fig. S1). Model coefficients from
155 interactions between age, crowding indices (conspecific and heterospecific) and treatment indicated
156 RGR for each treatment and their association with crowding indices. Species was included as a
157 random intercept in all models to account for different mortality or RGR among species. Crowding
158 indices were included as a random slope to assess whether the strength of NDD varied among species.
159 The inclusion of random slopes was assessed using model ΔAIC comparisons (Burnham and
160 Anderson 2004). Model fits were better with the inclusion of random slopes for conspecific crowding
161 for all mortality models. The inclusion of random slopes improved the fits of the growth models for
162 both conspecific and heterospecific crowding.

163 To compare responses between treatment types the eight treatments were split into four groups: 1)
164 non-combination plots; exclusion of fungi, insects, large mammals and all mammals, 2) exclusion of
165 fungi, insects and both combined, 3) exclusion of all mammals, fungi, and both combined, and 4)
166 exclusion of all mammals, insects, and both combined. Fungicide, insecticide, all mammal and large
167 mammal treatments were each compared to control plots. To assess the impact of small mammals,
168 large mammal and all mammal treatments were included in one model as separate treatment levels,
169 and in a second model combined large mammals and all mammals into one treatment level. The
170 significance of these separate treatments was compared using analysis of variance, comparing ΔAIC
171 for the two models (Burnham and Anderson 2004). Combination treatments (e.g. fungicide and
172 insecticide treatments in one plot) were compared to control and to each constituent non-combination
173 treatment (e.g. fungicide and insecticide treatments compared to 1) control, 2) fungicide and 3)
174 insecticide treatments).

175 Diversity was measured using exponent of the Shannon-Weiner diversity index for each plot at each
176 census. Change in diversity was calculated as the within-plot difference in Shannon-Weiner diversity
177 index from one census to the next, measuring from the second census, 9 months after the initiation of
178 treatments. Change in diversity was modelled against the number of days since the initiation of the
179 experiment, interacting with treatment. Shannon-Weiner diversity was included as a random intercept
180 to allow for differences in diversity among plots at the outset of the study.

Analyses were performed in R 3.5.0 (R Core Team 2017), using packages lme4 (Bates et al. 2015), lmerTest (Kuznetsova et al. 2016), effects (Fox 2003), vegan (Oksanen et al. 2017), and ggplot2 (Wickham 2009).

Results

Over 31 months and four censuses, we monitored 237 species. For seedlings in all treatments, the estimated annual probability of mortality was around 22% per year in the absence of neighbours. Mortality increased with conspecific crowding in control plots ($P = 0.008$, Fig. 1), with a doubling of conspecific crowding resulting in a 2% increase in the annual probability of mortality. Heterospecific crowding weakly and non-significantly increased mortality risk ($P = 0.36$, Fig. 1). RGR declined significantly with both conspecific and heterospecific crowding ($P \leq 0.001$), though the decline was stronger with heterospecific crowding (Fig. 2).

Mortality

The impact of neighbourhood density on mortality was assessed for each taxon of natural enemies. The effect of conspecific crowding on mortality was significantly reduced by fungicide application ($P = 0.01$, Fig. 3A), with a doubling of conspecific crowding resulting in a 2% decrease in mortality. There was no significant difference between large mammal and all mammal models, meaning that excluding small mammals tended to reduce mortality rates, but not to a significant extent ($P = 0.37$, Fig. 3C). Excluding insects and all mammals had no impact on the relationship between conspecific crowding and mortality ($P > 0.1$, Fig. 3B&D). No change in mortality was seen in relation to heterospecific crowding in any treatments ($P > 0.1$ for all treatments, Fig. S2), nor was heterospecific crowding a driver of mortality in control plots ($P = 0.36$, Fig. 1).

The effects of insects and mammals on seedling mortality were interactive, as mortality risks in plots from which they were simultaneously excluded did not significantly differ from those in plots from which they were separately excluded ($P > 0.05$ for all combination treatments, Fig. 4). Mortality rates in plots from which fungi were excluded in combination with other natural enemies, on the other

206 hand, tended to be elevated when mortality interacted with conspecific crowding (Fungicide and
207 Insecticide: $P = 0.01$, Fungicide and Mammals: $P = 0.06$) and heterospecific crowding (Fungicide and
208 Mammals: $P = 0.01$). No combination treatment showed any evidence of conspecific crowding
209 driving mortality when compared to control plots, with exceptionally high variance in mortality rates
210 (Fig. 4).

211 **Relative Growth Rates**

212 Relative growth rates were negatively impacted by conspecific crowding (Fig. S3A), a pattern that did
213 not vary in any exclusion treatments (Fig. S3B-E). High heterospecific crowding negatively impacted
214 RGR ($P < 0.001$), and this effect was significantly weakened by the application of insecticide ($P =$
215 0.003, Fig. 5C. In control plots, a doubling in heterospecific crowding reduced RGR by 5%, whereas
216 RGR was reduced by only 1% in plots from which insects were excluded. Under the influence of
217 heterospecific crowding there was no effect of excluding fungal pathogens, large mammals or all
218 mammals on RGR ($P = 0.11$, $P = 0.89$, $P = 0.18$ respectively, Fig. 5), and no effect of small mammals
219 ($P = 0.37$). Plots from which insects were excluded in combination with other treatments showed a
220 reduced effect of heterospecific crowding on RGR compared to control plots (insects and fungi: $P =$
221 0.005, insects and mammals: $P < 0.001$, Fig. S4). The effect of heterospecific crowding on RGR in
222 combined fungi and mammal exclusion treatments was not significantly different to control plots ($P =$
223 0.69).

224 **Diversity**

225 The effect of exclusion treatments on diversity was assessed by monitoring the change in Shannon-
226 Weiner diversity in each plot over the duration of the experiment. Control plots showed no change in
227 diversity throughout the experiment ($P = 0.32$). The application of fungicide lead to a significant
228 decrease in diversity, which intensified over the experimental period ($P = 0.006$, Fig. 6A). The
229 exclusion of large mammals significantly increased diversity at the first census ($P = 0.006$), though
230 this effect later dissipated (Fig. 6A). The model with all mammals and large mammals separate, and
231 the model with the treatments combined were not significantly different ($P = 0.96$), indicating that

small mammals were not a significant driver of declining diversity. The application of insecticide lead to a similar pattern in diversity as large mammals, with a small initial increase in diversity followed by a decline, but this change was not significant ($P = 0.16$). When treatments were combined, diversity decreased significantly in plots where mammals were excluded in combination with fungi ($P = 0.005$) or insects ($P = 0.05$, Fig. 6B). The combined exclusion of fungi and insects had no significant effect on diversity in comparison to control plots ($P = 0.22$).

Discussion

Natural enemies have been widely implicated as causes of NDD in seedling recruitment (Bell et al. 2006, Mangan et al. 2010). While their effects have been tested individually (Packer and Clay 2000), our study is among very few that have compared the interacting effects of four key taxa of natural enemies simultaneously (Paine et al. 2016, Bagchi et al. 2014, Gripenberg et al. 2014). We find strong evidence that fungal pathogens generate more NDD mortality than do insects, consistent with Bagchi et al. (2014). Our study expands upon theirs, being the first to simultaneously assess NDD among seedlings driven by fungi, insects, small mammals, and large mammals. The comparison of four groups of natural enemies allows us to assess their relative contributions to NDD. Moreover, we examined both mortality and growth. Whereas density dependent mortality was driven exclusively by fungal pathogens, density dependent RGR was driven by insects. Furthermore, fungal pathogens drove conspecific NDD, while insects drove heterospecific NDD. Only fungal pathogens, however, generated changes in diversity through NDD. This indicates that while NDD among plants may have multiple drivers, not all are important in maintaining diversity as only conspecific NDD leads to increased diversity.

It has been widely demonstrated that NDD structures tree communities at early life stages (Harms et al. 2000, Green et al. 2014). Here, we demonstrated that conspecific interactions are the primary contributor to NDD mortality, consistent with previous research (Comita et al. 2010). NDD interactions were detected in control plots. We tested the mechanism driving NDD using treatment plots, where natural enemy groups were excluded. Our results indicate that fungal pathogens alone are

the key drivers of conspecific NDD mortality. These results are consistent with studies that found strong associations between NDD and fungal pathogens (Packer and Clay 2000, Bell et al. 2006, Bagchi et al. 2014), and validate a mechanism originally proposed by Janzen (1970). Fungal pathogens are often highly host specific (Gilbert and Webb 2007); it is this host specificity that allows pathogens to pass between hosts effectively under high conspecific crowding, rapidly causing mortality to conspecifics and allowing heterospecifics to persist. Where fungal pathogens were excluded in this study, mortality at high conspecific crowding not only decreased but the trend was reversed, with mortality decreasing with conspecific density after the application of fungicide. This indicates that, once released from the negative effects of fungal pathogens, the environmental conditions were favourable enough for high densities of conspecifics to thrive. This further supports the argument that intraspecific competition among seedlings is unlikely to drive NDD interactions (Paine et al. 2008), because no NDD mortality was recorded in this study among high numbers of conspecifics with natural enemies supressed, even though conspecifics are likely to be competing for identical resources.

A comparable study by Bagchi et al. (2014) found NDD mortality driven by both fungal pathogens and insects, whereas we found no evidence for insect driven NDD mortality. Both studies used robust and similar methods. Differing biotic and abiotic conditions among experimental sites may account for this difference, however it would be beneficial to extend studies excluding insects to monitor long-term effects. NDD interactions driven by fungal pathogens in this study are driving changes in diversity through the suppression of competitive exclusion in seedling communities (Harms et al. 2000). This process is key to the maintenance of diversity in seedling communities (Wright 2002, Comita et al. 2010), and our study presents strong evidence that fungal pathogens are by far the strongest driver of this process.

Relative growth rates were negatively impacted by overall crowding effects, with RGR diminished under both high conspecific and heterospecific crowding. With fungal pathogens driving mortality but not RGR under high conspecific crowding, it appears that seedling mortality driven by fungal pathogens is rapid, with little time for a detectable reduction in growth rate before mortality occurs (

285 Bell et al. 2006). The exclusion of insects increased RGR significantly under high heterospecific
286 crowding, an effect that did not consequently reduce mortality during this study. It is, however,
287 possible that negative effects on RGR lead to mortality in the long term, reducing plant health until
288 they are unable to compete successfully. Detecting such long-term effects was beyond the scope of
289 this study, however. Although many insect herbivores are host-specific (Forister et al. 2015), insects
290 more commonly consume a host clade, which can be variable in species richness (Novotny et al.
291 2002). With this semi-generalist nature, insects generate hetero- and conspecific NDD and RGR, thus
292 not generating a significant change in diversity, since diversity is elevated only when intraspecific
293 effects are stronger than interspecific effects (Chesson 2000).

294 If insect herbivores accelerated rates of pathogen infection (García-Guzman and Dirzo 2001) NDD
295 processes would be stronger in combined herbivore (insects or mammals) and fungal pathogen
296 exclusion plots. Surprisingly, not only did we detect no change in rates of NDD mortality, the effect
297 of excluding fungal pathogens became undetectable. We found, however, that in all cases the
298 simultaneous exclusion of multiple natural enemy groups lead to an overall loss of conspecific NDD
299 mortality, with highly variable mortality rates under any conspecific crowding. This may be because
300 removing multiple deterministic mechanisms simultaneously (e.g. fungi impacting mortality and
301 insects impacting RGR) makes the occurrence of mortality highly stochastic, or at least unconnected
302 to the biotic neighbourhood. Combined treatments impacted RGR similarly to plots with individual
303 exclusions; in plots excluding insects in combination with other treatments, the effect of
304 heterospecific crowding on RGR was diminished as with plots excluding insects alone. We conclude
305 that insects alone are driving the change detected in combination plots. Our results show that natural
306 enemies are not interacting to influence NDD, and are therefore impacting different sections of the
307 seedling community. This provides evidence for variation among species in vulnerability to natural
308 enemy attack, and thereby vulnerability to NDD. This could be further examined by investigating
309 changes in species composition in treatment plots.

310 There was no change in mortality or RGR under the influence of crowding after the exclusion of large
311 or small mammals, in contrast with Paine et al. (2016), who found that small and medium sized

312 mammals contribute to NDD in a seed removal experiment. However Paine et al. (2016) did not
313 observe fungal pathogens or insects, and our study indicates that the effects of mammals are minimal
314 compared to the prevailing effects of natural enemies in the smaller size classes. Studies have
315 implicated small mammals as seed predators in structuring tree communities (Dematttia et al. 2006,
316 Paine et al. 2016). It is possible this study overlooks a component of NDD interactions at the seed
317 stage, however we do not detect any changes in diversity driven by small mammals.

318 A caveat in this study is the unusually low densities of *T. pecari* (white-lipped peccary), which can
319 influence seed mortality (Beck 2005), at the site during this experiment. *T. pecari* populations
320 undergo large fluctuations, becoming locally scarce for years at a time (Reyna-Hurtado et al. 2009).
321 This could account for the contrasting results found by Beck et al. (2013) and Theimer et al. (2011),
322 who found that large mammals contributed to NDD. Nevertheless, we detect a change in diversity
323 when large mammals were excluded (Fig 6A). Diversity initially increased after the first year,
324 implying that large mammals suppress diversity when present, though this effect diminishes during
325 the following 2 years. We did not detect NDD in large mammal plots, and it is unlikely that NDD
326 would suppress diversity, so we must assume that large mammals are suppressing plant diversity via
327 alternative mechanisms. A secondary mechanism appears to be stabilizing diversity after the initial
328 release from herbivory. Where mammals and fungal pathogens were simultaneously excluded,
329 diversity increased initially as with large mammal exclusion, showing no signs of the significant
330 decline seen in fungicide treated plots (Fig. 6B), indicating the diversity effects of large mammals are
331 more powerful than the effects of fungal pathogens. Moreover, there is a subsequent decline in
332 diversity in large mammal plots in the presence and absence of fungal pathogens or insects, it is
333 therefore unlikely that fungal pathogens or insects are causing the re-stabilizing effect on diversity
334 through NDD interactions.

335 Large mammals initially had a stronger impact on diversity than small mammals, contrasting with
336 studies that indicate small mammals are more likely to drive NDD interactions (Dematttia et al. 2006,
337 Paine et al. 2016), or show little evidence for altered diversity in relation to large mammals (Theimer
338 et al. 2011). Large vertebrates suffer population declines in hunted forests (Endo et al. 2010) and

339 defaunated forests have seen substantial changes in tree community assemblage (Terborgh et al. 2008,
340 Peres et al. 2015). Our results indicate that the extirpation of large terrestrial mammals would have
341 little effect on tree diversity, with the initial boost in diversity after the loss of large mammals
342 levelling out before trees reach reproductive age. This does not negate impacts of hunting however,
343 since hunting impacts both terrestrial and arboreal mammals (Endo et al. 2010), and many trees are
344 heavily reliant on arboreal frugivores for seed dispersal (Peres and van Roosmalen 2002). While the
345 loss of large terrestrial mammals has minimal consequences for tree communities, hunting
346 conceivably alters community assembly in hunted forests due to a loss of large frugivores (Terborgh
347 et al. 2008).

348 # Conclusions

349 Our study demonstrates that while fungal pathogens and insects drive NDD interactions, diversity is
350 promoted by the actions of fungal pathogens. Diversity is also driven by large mammals; but while
351 fungal pathogens maintain diversity through NDD, large mammals impact diversity through non-
352 NDD processes. Furthermore, after a loss of large mammals destabilizes diversity, fungal pathogens
353 are not associated with the following restabilization. In response to this, we emphasize that in the
354 absence of large mammals, disruption to tree diversity is not compensated for by natural enemies,
355 implying non-biotic mechanisms play an essential role in maintaining diversity. While our results
356 suggest that diversity is maintained by multiple drivers and should not be assigned a single
357 mechanism, we present strong evidence that fungal pathogens are uniquely important in shaping
358 tropical tree communities above other groups of natural enemies, and are disproportionately important
359 drivers of diversity in tropical forests.

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Figures

Figure 1: Predicted neighbourhood crowding effects on probability of mortality (y^{-1}) in control plots. Conspecific but not heterospecific crowding lead to higher mortality rates.

Figure 2: Model predictions showing height as predicted by age under the influence of A) conspecific and B) heterospecific crowding. Colours represent minimum, median and maximum crowding indices. Difference in slope indicates a difference in relative growth rates (RGR) among crowding indices, with steep lines represent high RGR. RGR decreases with increased crowding index for heterospecific crowding only.

Figure 3: Probability of mortality (y^{-1}) predicted by conspecific crowding indices under the exclusion of A) fungal pathogens, B) insects, C) small mammals and D) large mammals. Coloured lines represent the excluded group of natural enemies. Only plots excluding fungal pathogens significantly reduced mortality at high conspecific crowding. Control treatment is repeated in each panel to facilitate comparisons.

Figure 4: Mortality driven by conspecific crowding indices for plots excluding A) fungal pathogens and insects, B) mammals and fungal pathogens, and C) mammals and insects. Coloured lines represent the excluded group of natural enemies. Combination treatments are indicated in the plot heading. Conspecific crowding influences mortality under control treatments, but does not significantly impact mortality under any combination treatments.

Figure 5: Model predictions showing relative growth rates (RGR) as height predicted by age under the influence of heterospecific crowding in A) control plots (as seen in 2B) and under the exclusion of B) fungal pathogens, C) insects, D) small mammals and E) large mammals. Colours represent minimum, median and maximum heterospecific crowding indices. Difference in slope indicates a difference in RGR among crowding indices, with steep lines represent high RGR. RGR decreases with increased heterospecific crowding in all plots except those in which insects were excluded. Figure 2A is duplicated in Figure 5A in order to facilitate comparison.

513 **Figure 6:** Effective change in Shannon-Weiner Diversity index under the exclusion of A) fungal
514 pathogens, insects, small mammals and large mammals, and B) combination treatments.

515 Diversity change is within plot over time from 10 months after the initial application (the second
516 census). Treatment effects are represented as the ratio of Shannon diversity in treatment vs. control
517 plots. Small mammal effects are represented as the ratio of Shannon diversity in all mammal vs. large
518 mammal plots.
519

Figure 1

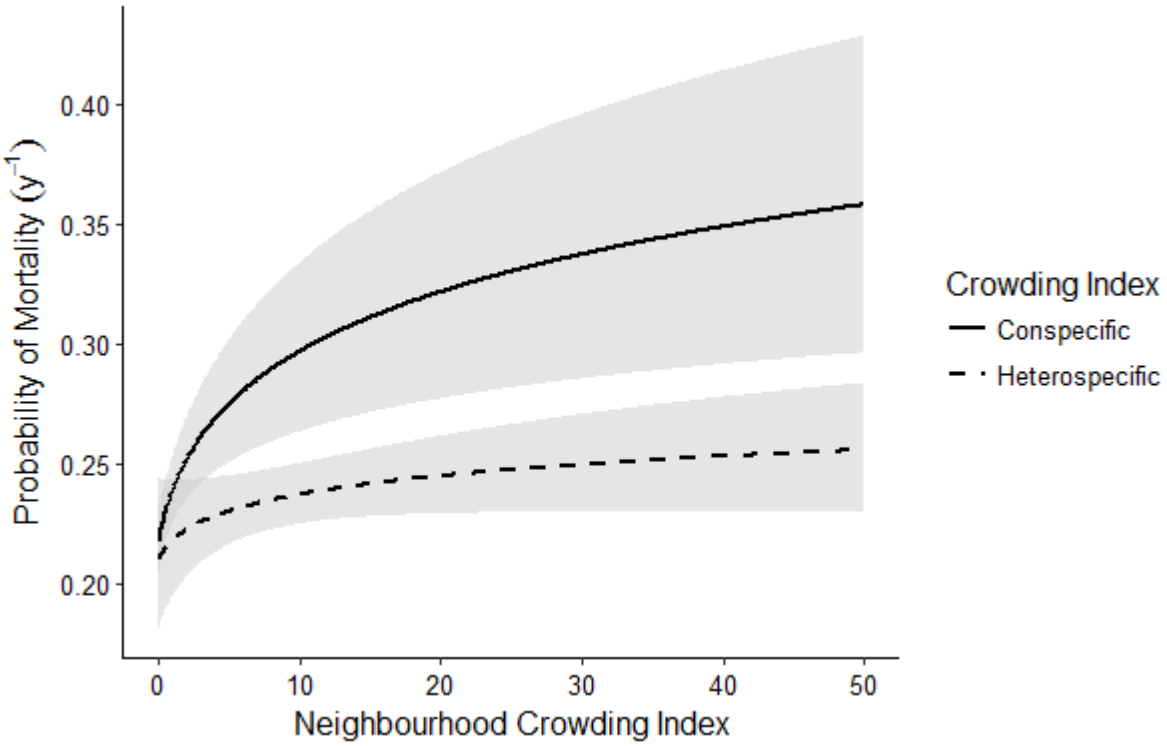

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Figure 2

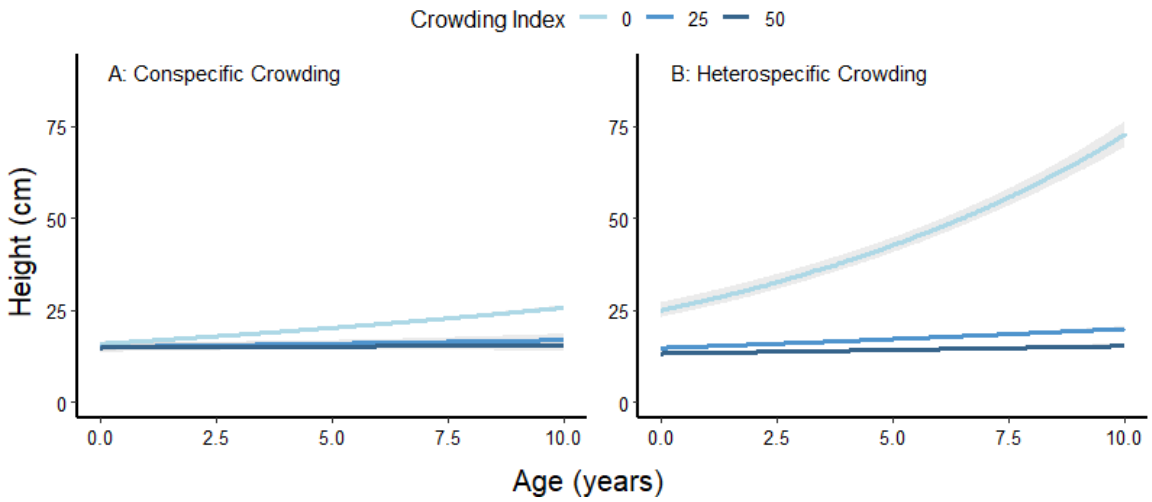

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Figure 3

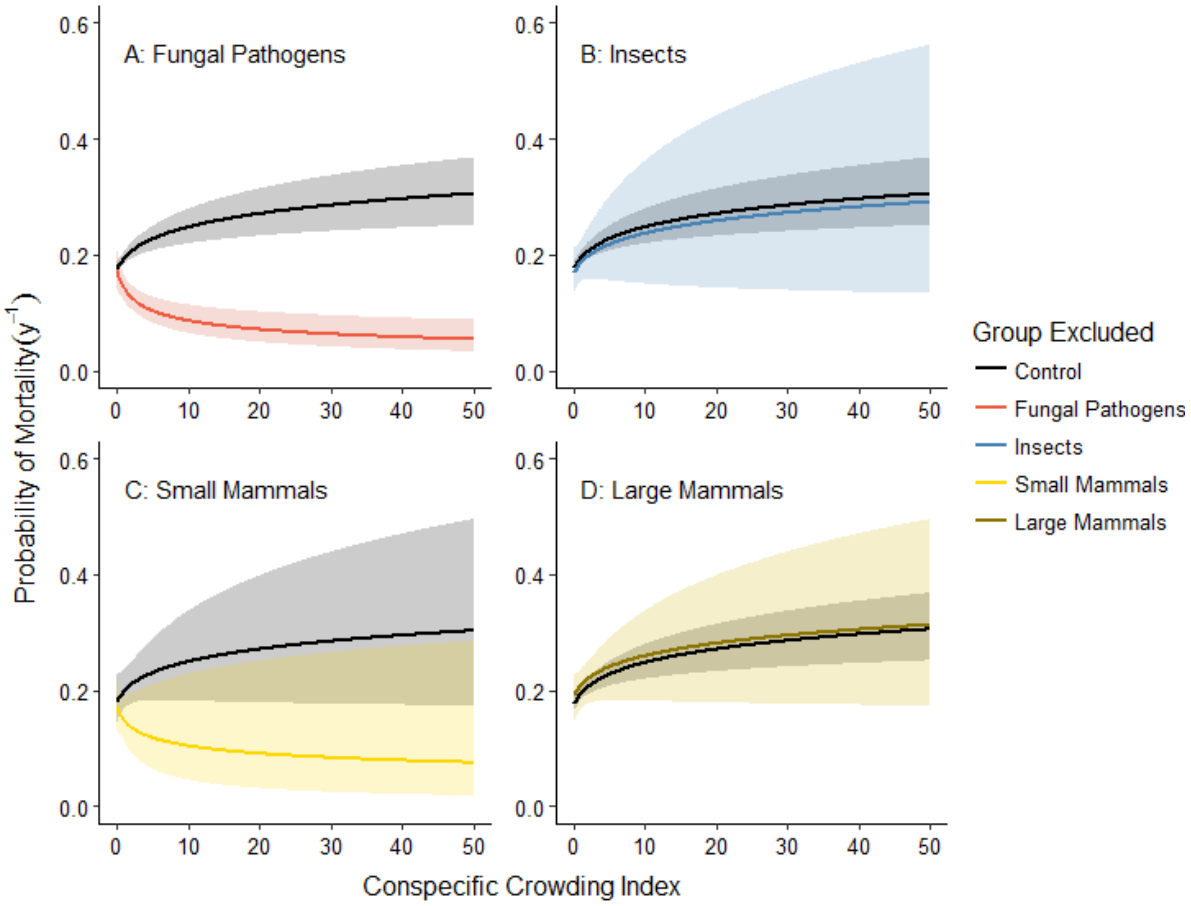

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Figure 4

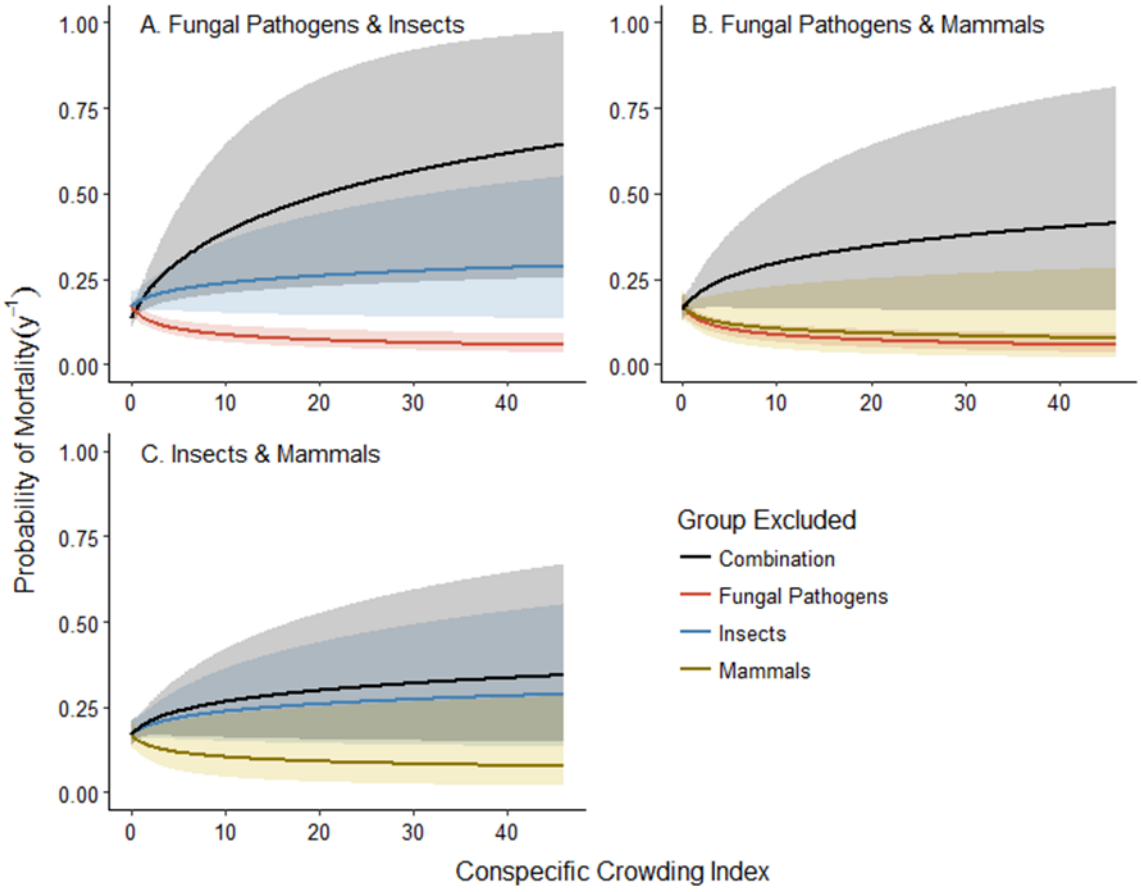

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Figure 5

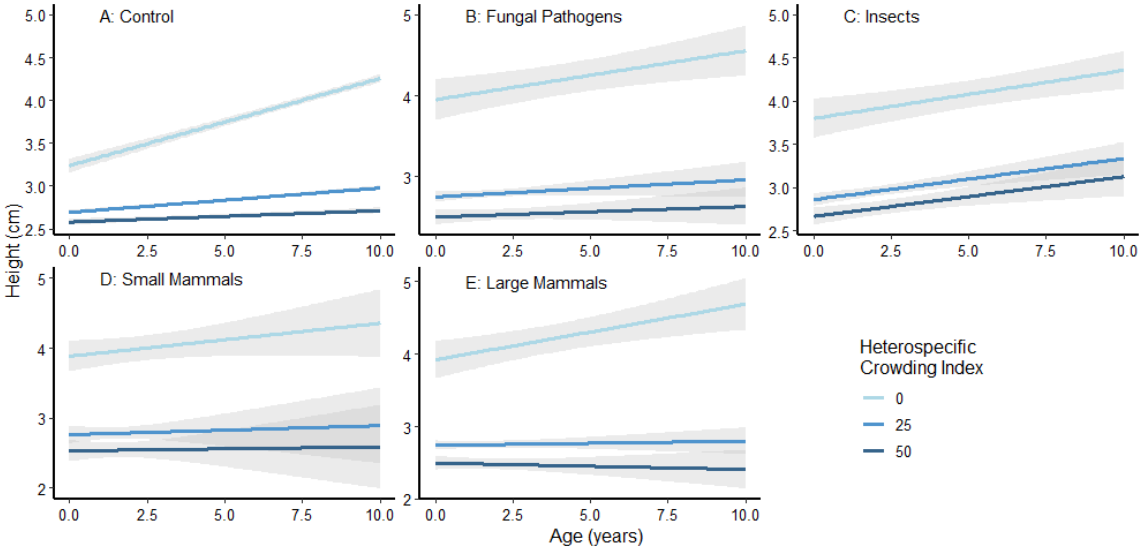

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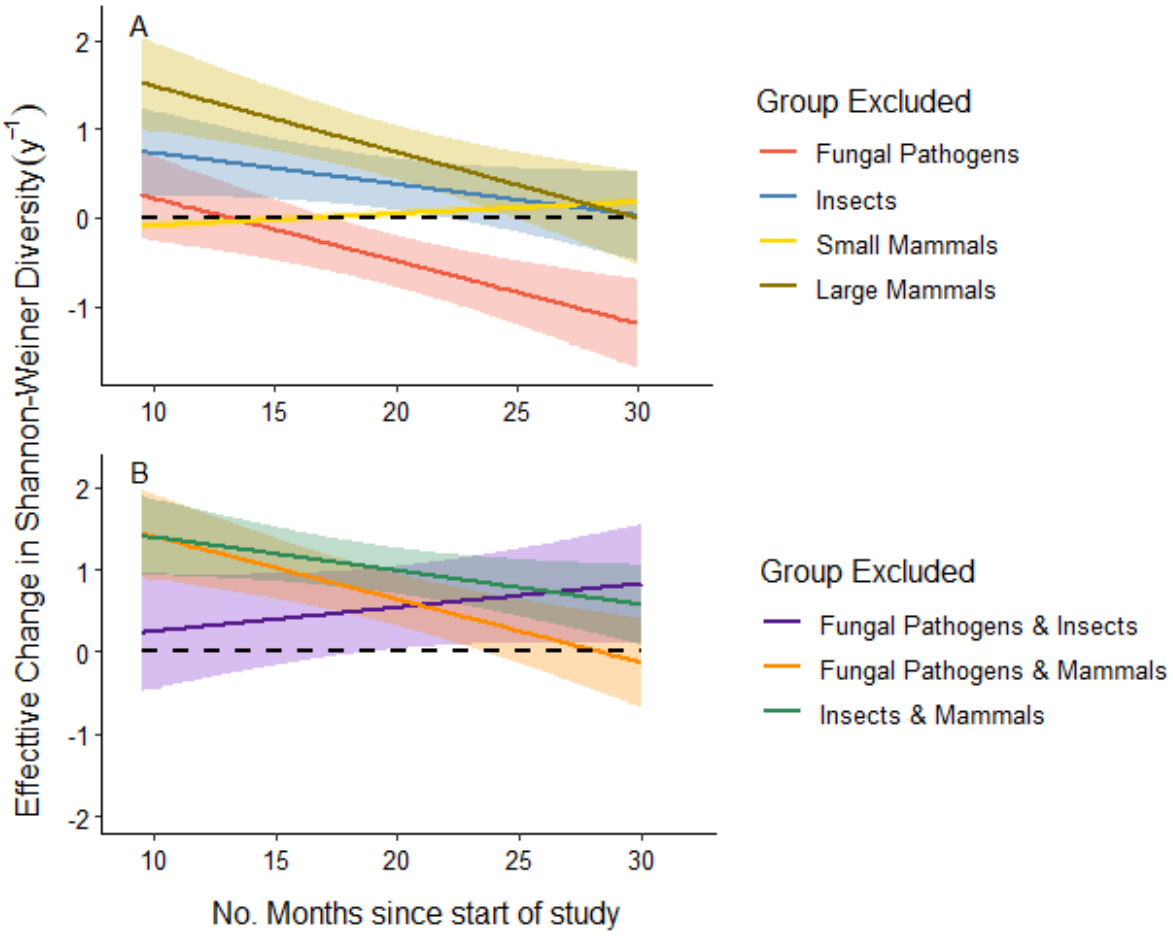
Figure 6


A
B
2
1
0
-1
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Effective Change in Shannon-Weiner Diversity (y^{-1})
No. Months since start of study
Group Excluded
Fungal Pathogens
Insects
Small Mammals
Large Mammals
Group Excluded
Fungal Pathogens & Insects
Fungal Pathogens & Mammals
Insects & Mammals

530