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THE MUTUALISM–PARASITISM CONTINUUM IN ECTOMYCORRHIZAS: A QUANTITATIVE ASSESSMENT USING META-ANALYSIS

Justine Karst,[1,5] Laurie Marczak,[2] Melanie D. Jones,[3] and Roy Turkington[1,4]

[1]*Department of Botany, University of British Columbia, 3529-6270 University Boulevard, Vancouver, British Columbia V6T 1Z4 Canada*
[2]*Centre for Applied Conservation Research, Department of Forest Sciences, University of British Columbia, 2424 Main Mall, Vancouver, British Columbia V6T 1Z4 Canada*
[3]*Biology and Physical Geography Unit, University of British Columbia Okanagan, 3333 University Way, Kelowna, British Columbia V1V 1V7 Canada*
[4]*Biodiversity Research Center, University of British Columbia, Vancouver, British Columbia V6T 1Z4 Canada*

Abstract. Context dependency is deemed to position the outcomes of species interactions along a continuum of mutualism to parasitism. Thus, it is imperative to understand which factors determine where a particular interspecific interaction falls along the continuum. Over the past 20 years research on the ectomycorrhizal symbiosis has resulted in sufficient independent studies to now generalize about the factors and mechanisms that affect host response to ectomycorrhizas. Using meta-analysis we quantitatively evaluated the role of biotic (partner identity and colonization levels of ectomycorrhizal fungi) and abiotic (phosphorus levels) factors in determining host biomass, height, and shoot : root responses to ectomycorrhizal associations. On average, seedlings across multiple host genera increased in total biomass when inoculated with ectomycorrhizal fungi regardless of the identity of the fungal associate; host genera differed in the magnitude of response for both total biomass and shoot : root ratio. Association with different fungal genera modified only host allocation of biomass to shoots and roots. Neither level of colonization on inoculated seedlings nor the level of contamination on control seedlings relative to colonization levels by target fungi on inoculated seedlings was important in explaining variation in effect sizes for any growth response. None of our proposed factors (identity of partners, colonization level, magnitude of contamination, or duration of association) explained variation in effect sizes for shoot height, although in general seedlings were taller when inoculated with ectomycorrhizal fungi. Phosphorus additions did not influence effect sizes. Although the general trend across studies was for a positive response of hosts to ectomycorrhizal inoculation, publication bias and methodological issues effectively reduce and distort the spectrum on which we evaluate host responses to ectomycorrhizal inoculation. Our results indicate that the variation in ectomycorrhizal fungi perceived by the host may be of a discrete (presence/absence of ectomycorrhizal fungi) rather than continuous nature (variation in identity or abundance of ectomycorrhizal fungi).

Key words: coevolution; context dependency; ectomycorrhizal fungi; host responses; inoculation; meta-analysis; pairwise interactions; publication bias.

Introduction

The degree to which and the mechanisms by which the outcome of species interactions is dependent on endogenous and exogenous factors are questions central to ecology (Thompson et al. 2001). Within putative mutualisms in particular, context dependency has been recognized to generate conditional responses (Thompson 1988, Bronstein 1994, Herre et al. 1999, Egger and Hibbett 2004). As a consequence, the outcome of such interactions between partner species often falls on a continuum of mutualism to parasitism. To determine the factors predicting the outcome of interspecific interactions along this continuum requires an understanding of the spatial, temporal, and taxonomic context for a given system (Herre et al. 1999).

Variation in the functioning of mutualisms caused by context dependency has important ecological consequences. For example, invasions by exotic plant species may be facilitated or hindered, depending on their response to the abiotic and biotic milieu of a new habitat. In particular, plants gain and lose interactions with species, and this dynamic is often situated within new abiotic conditions (Mitchell et al. 2006). Mycorrhizal fungi are important components of the biotic milieu in most soils and research on mycorrhizas has been "paradigmatic" in developing our understanding of the existence and functioning of the mutualism–parasitism continuum (Sapp 2004). Because host plants do not always respond positively to mycorrhizas, the continu-

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[5] E-mail: justine@karst.ca

um concept has been put forward as an appropriate framework to describe the range of outcomes between mycorrhizal partners (Francis and Read 1995, Johnson et al. 1997, Brundrett 2004, Jones and Smith 2004). In addition, variation in mycorrhizal functioning is believed to be important in maintaining species coexistence (Bever 2003, Umbanhowar and McCann 2005) and to impact ecosystem-level processes (e.g., van der Heijden et al. 1998).

Given that the concept of the mutualism–parasitism continuum is well established, we now require a better understanding of the factors and mechanisms that determine where a particular interspecific interaction falls along the continuum. Ectomycorrhizal symbioses are particularly well-studied examples; there is a full literature examining the role of biotic and abiotic factors in affecting host response to ectomycorrhizas (e.g., Gehring and Whitham 1994, Johnson et al. 1997, Setala et al. 1997, van der Heijden and Kuyper 2001*a*, Kennedy and Peay 2007). Of the biotic factors deemed important to the mutualism–parasitism continuum, fungal identity has been most thoroughly investigated. Functional variation among taxa of ectomycorrhizal fungi is well documented for characteristics including nutrient uptake (e.g., Abuzinadah and Read 1989, Dighton et al. 1990, Jongbloed et al. 1991, Lilleskov et al. 2002), and drought (Parke et al. 1983, Boyle and Hellenbrand 1991, Dixon and Hiol-Hiol 1992) and pH tolerance (Wallander 2002, Yamanaka 2003, Dunabeitia et al. 2004). In nature, host plants will encounter variation in both the presence and abundance of fungal species with which they form associations and abundance is often quantified by colonization levels. The relationship, however, between levels of colonization and host growth is inconsistent (e.g., Jones et al. 1990, Thompson et al. 1994), and root tip ectomycorrhizal biomass does not necessarily correspond to the biomass of fungi occurring as extramatrical hyphae (Genney et al. 2006) or in fruiting structures (Gardes and Bruns 1996). Abiotic factors, such as the nutrient status of soils, are hypothesized to also be key in determining host position on the mutualism–parasitism continuum (Johnson et al. 1997) and are spatially and temporally heterogeneous at scales relevant to ectomycorrhizas (Nantel and Neumann 1992, Toljander et al. 2006). In soils of high fertility, the net benefit a fungus confers to a plant is expected to decrease because the nutrient acquisition abilities a plant gains through mycorrhizal associations become superfluous (Smith and Read 1997).

The past few decades have generated sufficient individual studies on plant host responses to ectomycorrhizal associations that some generalizations can now be made about the nature of the association (mutualistic to parasitic) across different host–fungus pairings and nutrient regimes; however, there has been no quantitative synthesis that allows us to determine the variation in these responses. Meta-analysis is an increasingly common analytical tool used by ecologists to quantitatively summarize and explain the results of multiple independent studies (e.g., Gurevitch et al. 2000, Treseder 2004, Cardinale et al. 2006, Lortie and Callaway 2006) and is particularly useful when published studies have conflicting results. Meta-analyses have also been used to highlight gaps in the data and to identify common methodological problems or constraints. More importantly, by treating separate empirical studies as independent data points weighted by their replication and precision, meta-analysis allows us to discern general patterns already existing in the data that might not be otherwise evident. We used meta-analysis to determine: (1) whether host response to ectomycorrhizal fungi is host or fungal specific; (2) whether levels of colonization modify the response; (3) whether soil nutrient conditions modify host growth responses. We addressed three additional questions about the role of experimental conditions in modifying host response: (4) Has the perception of mycorrhizas as mutualisms (i.e., defining interactions between hosts and fungi as strictly beneficial) biased publication of results? (5) Does contamination of controls modify detectable host response to ectomycorrhizas? (6) Does host response change with the length of association between host and fungus (i.e., experiment length)?

Methods

Data collection

We searched the Institute of Scientific Information (ISI) Web of Science (1965 to the present) using the key word "ectomycorrhiza." Of the 3591 hits, we selected papers written in English reporting either total biomass (in grams), shoot height (in centimeters), or shoot : root ratio of tree seedlings inoculated with ectomycorrhizal fungi paired with non-inoculated control seedlings. We also checked the literature-cited sections of these papers for additional references. Total biomass is a measure of productivity. Shoot height may be indicative of competitive ability in the seedling establishment phase and changes in shoot : root ratio may identify factors that increase seedling survival in nutrient-limited environments or that control the potential carbon supply to ectomycorrhizal fungi, the currency mediating the association. For each study, we recorded the mean, standard deviation, and sample size for inoculated and control seedlings, but eliminated those studies that did not report both mean and sample size. When necessary, we digitized graphs to obtain this information. We also did not include studies in which inoculation resulted in no colonization or in which there were no control data (non-inoculated treatments).

We recorded the species of host and, when given, ectomycorrhizal fungus (in some cases the fungus was an unknown isolate or the species epithet was not provided). In cases in which experimental treatments involved several combinations of host species with ectomycorrhizal fungal species or fungal isolates, we treated each combination as a separate study, although

not all studies were completely independent. Inclusion of several studies from one paper tends to reduce the overall heterogeneity in effect sizes, but excluding multiple results from a paper could underestimate effect sizes (Gurevitch and Hedges 1999).

Duration of association was quantified by the number of weeks each experiment ran. This measure was the only consistent proxy to evaluate the influence of experimental duration on host outcome to ectomycorrhizal associations; however, we recognize that extreme differences in growth rates among host species would render absolute length of time irrelevant. When repeated measures were taken in a study, we used data from the last sampling period to capture the maximal length of association between host and fungus.

Colonization level (percentage of root tips colonized or percentage of root length colonized) of inoculated seedlings and control seedlings was also recorded; when it was given as a range, we used the median value. Contamination of non-inoculated seedlings reduces differences in colonization levels between control seedlings and inoculated seedlings. Consequently, the perceived response of hosts to ectomycorrhizal inoculation may be reduced as a result of contamination. We determined the magnitude of contamination by calculating the level of colonization on control seedlings relative to that measured on inoculated seedlings according to the proportion

$$C_C/(C_C + C_{TR})$$

where C_{TR} is the percentage of colonization of target fungi on inoculated seedlings and C_C is the percentage of colonization of contaminant fungi on control seedlings. Because levels of contamination are expected to be lower in situations in which sterility can be strictly maintained, we also noted whether experiments were conducted in growth chambers, greenhouses, nurseries, or in the field.

In cases in which results from papers involved inoculation trials in combination with explicit manipulations of the environment, other than nutrient levels (e.g., pH, pathogen abundance, nematode density, salinity, soil moisture, CO_2), we used data from "ambient treatments." For example, we recorded data for inoculated and control seedlings from ambient CO_2 levels while excluding data from treatments featuring elevated CO_2 levels. Among those papers that manipulated fertilizer types and amounts, only the manipulation of inorganic phosphorus levels was reported in a sufficient number of studies to merit an additional, separate meta-analysis. We converted phosphorus additions to a common unit, milligrams of P per kilogram substrate, with values ranging from 0 to 136 mg/kg substrate.

Data analysis

The effect size of ectomycorrhizal inoculation for total biomass, shoot height, and shoot : root ratio was calculated as the natural log of the response ratio of inoculated to control seedlings. The response ratio (R) is the ratio of the mean outcome in the experimental (inoculated) group to that of the control (non-inoculated) group (Rosenberg et al. 2000). Only 12% of the studies in our analysis reported measures of variation around means. Consequently, the variance for each effect size was calculated by sample sizes and means alone (Shurin et al. 2002, Lajeunesse and Forbes 2003, Marczak et al. 2007), and while this increases the probability of Type II errors, it avoids underestimating effect sizes (Gurevitch and Hedges 1999).

We tested the null hypothesis that all effect sizes were equal, based on the statistic Q_T, with larger values indicating greater heterogeneity in effect sizes among comparisons (Rosenberg et al. 2000). If rejected, we examined the continuous (colonization levels, magnitude of contamination, and duration of association) and categorical (fungal and host genus identity) explanatory variables using fixed models. We regressed effect size against all continuous predictor variables. For the categorical variables, we grouped species into genera for both host and fungus, and when testing for differences among genera, we included only those that were represented by at least 10 studies. Effect sizes for all analyses were not normally distributed, so we relied on randomization tests (4999 iterations) to assess significance levels. Explanatory variables were considered significant at $\alpha = 0.05$. When categorical predictors were significant, we assessed differences among groups based on 95% bootstrapped confidence intervals. For any significant explanatory variable, we only report those explaining >5% of the variation in effect sizes as estimated by Q_M/Q_T, where Q_M is the variation in effect sizes that is explained by a particular model (Rosenberg et al. 2000).

We assessed the importance of publication bias for those analyses including at least 25 papers (Begg and Mazumdar 1994) using a nonparametric rank correlation test (Spearman's rho). A significant correlation between standardized effect size and sample size across studies would indicate bias in the publication of extreme effect sizes. All data analyses were performed in MetaWin software version 2.1.4 (Rosenberg et al. 2000).

Results

Seedling response to ectomycorrhizal inoculation

Overall we extracted 459 studies of inoculation response of total biomass from 36 papers, 329 studies of shoot height from 24 papers, and 235 studies of shoot : root ratio from 20 papers (Appendix A). Across all growth traits, we assessed the outcome of 21 host genera inoculated with 31 fungal genera; however, these inoculations were not represented in all possible combinations.

On average, seedlings increased in total biomass and shoot height, but did not change in shoot : root biomass allocation when inoculated with ectomycorrhizal fungi (mean cumulative effect sizes = 0.208, 0.113, −0.0174,

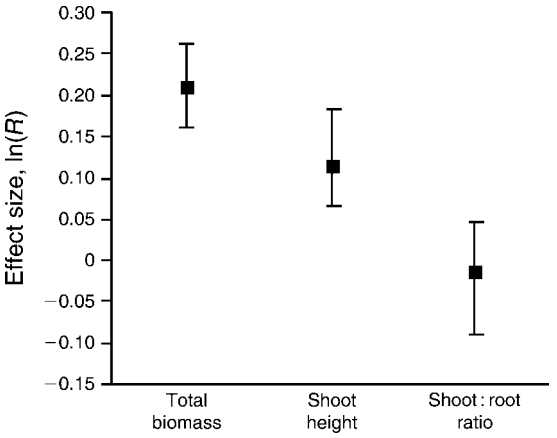

FIG. 1. Cumulative mean effect sizes for total biomass, shoot height, and shoot : root ratio. Error bars are 95% bootstrapped confidence intervals.

respectively; Fig. 1). However, there was significant heterogeneity in the data ($Q_T = 10152$, df = 458; $Q_T = 95389$, df = 328; $Q_T = 705$, df = 234, respectively; all $P < 0.001$), indicating that there was substantial variation around the means.

The identity of the host genus was significant in explaining variation in effect sizes for both total biomass ($P = 0.028$, df = 4, 409, $Q_M/Q_T = 0.18$) and shoot : root ratio ($P < 0.001$, df = 3, 191, $Q_M/Q_T = 0.22$). In particular, inoculated seedlings of the genera *Quercus*, *Pseudotsuga*, and *Eucalyptus* increased in total biomass more than those of *Pinus* and *Picea* (Fig. 2A), while *Picea* seedlings allocated more biomass to shoots than seedlings of *Quercus*, *Pseudotsuga*, and *Pinus* when inoculated (Fig. 2B). Although there was a positive relationship between total biomass and shoot height ($P < 0.001$, df = 1, 567, $r^2 = 0.37$), neither categorical nor continuous predictors explained variation in effect sizes of shoot height. Fungal genus influenced allocation of biomass to shoots vs. roots ($P < 0.001$, df = 5, 199, $Q_M/Q_T = 0.26$), but did not explain variation in effect sizes for total biomass or shoot height. Seedlings inoculated with fungi from the genus *Scleroderma* allocated more biomass to roots than that observed for other genera (Fig. 3).

Level of colonization of inoculated seedlings, ranging from 0.5% to 98%, was not important in explaining variation in effect sizes for total biomass ($P = 0.043$, df = 1, 349, $Q_M/Q_T = 0.03$), shoot height ($P = 0.30$, df = 1, 220), or shoot : root ratio ($P = 0.03$, df = 1, 211, $Q_M/Q_T = 0.03$; Fig. 4). (Note that although level of colonization was significant, $Q_M/Q_T < 0.05$ for both total biomass and shoot : root ratio [see *Methods*].) Heterogeneity in effect sizes was unrelated to the magnitude of contamination for total biomass ($P = 0.20$, df = 1, 324), shoot height ($P = 0.48$, df = 1, 211), and shoot : root ratio ($P = 0.063$, df = 1, 197; Fig. 5). Contamination levels were highest in those experiments performed in nurseries and

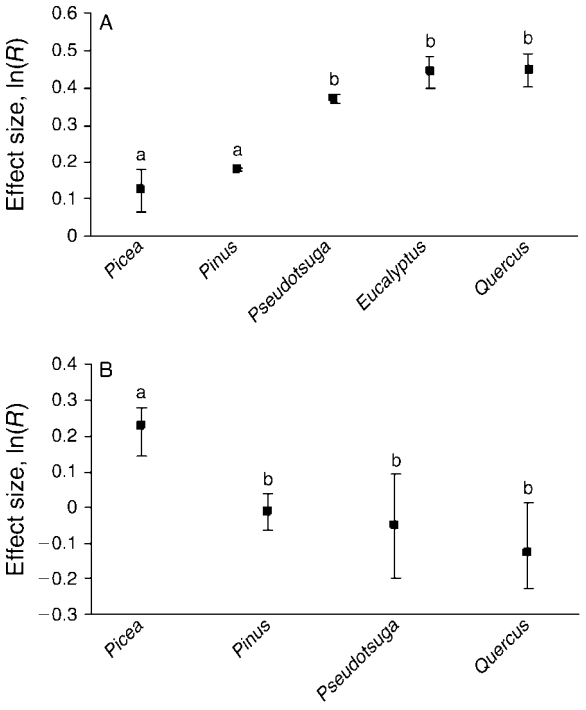

FIG. 2. Effect size for (A) total biomass and (B) shoot : root ratio by host genus. Means with 95% bootstrapped confidence intervals are shown. Means designated by the same letter are not statistically different (95% bootstrapped confidence intervals overlap). For (B), positive values indicate that allocation of biomass to shoots was higher than allocation to roots.

in the field and lowest in those in growth chambers (one-way ANOVA, $P < 0.001$, $F_{3,416} = 76.9$; Table 1). Effect sizes for total biomass and seedling height were unrelated to location of experiment ($P = 0.73$, df = 3, 454; $P = 0.92$, df = 3, 324) but shoot : root ratios were

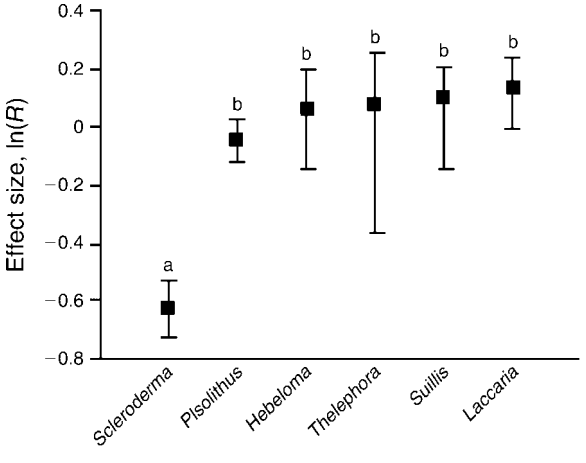

FIG. 3. Mean effect size for shoot : root ratio by fungal genus. Means with 95% bootstrapped confidence intervals are shown. Means designated by the same letter are not statistically different (95% bootstrapped confidence intervals overlap). Positive values indicate that allocation of biomass to shoots was higher than allocation to roots.

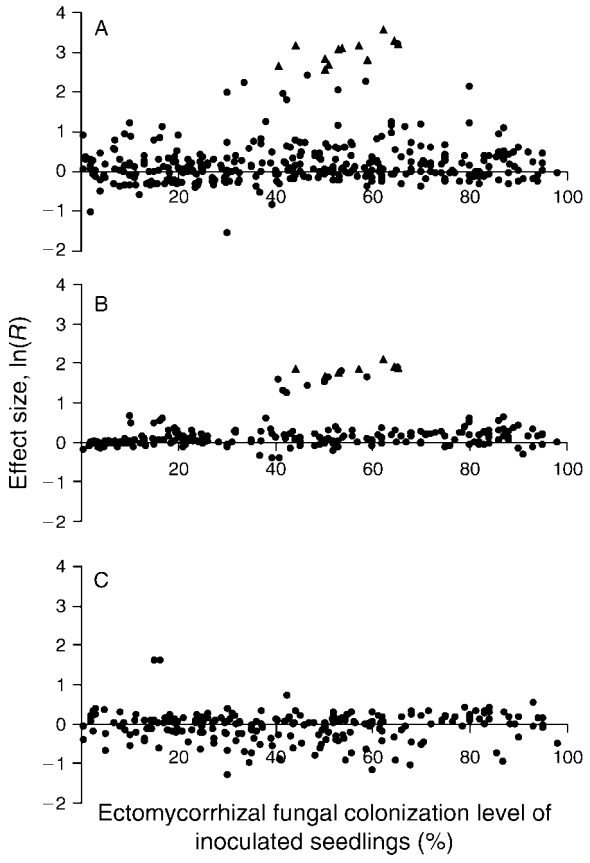

FIG. 4. Relationship between mean effect sizes and level of ectomycorrhizal fungal colonization of inoculated seedlings for (A) total biomass, (B) shoot height, and (C) shoot : root ratio. Outliers in (A) and (B) (those data points above the 97th percentile of the distribution) are indicated as triangles; these were retained in the analysis.

higher for those seedlings grown in greenhouses vs. growth chambers ($P = 0.001$; df = 3, 119, $Q_M/Q_T = 0.24$).

The mean length of experiments was 21 weeks (range = 8–104 weeks), slightly less than the mean age of seedlings used in experiments (23 weeks, range = 10–104 weeks). Duration of association between host plant and fungus did not explain variation in effect sizes for total biomass ($P = 0.86$, df = 1, 457) or shoot height ($P = 0.97$, df = 1, 327; Fig. 6A, B). On average, seedlings allocated more biomass to roots than shoots, with increasing duration of association ($P < 0.001$, df = 1, 233, $Q_M/Q_T = 0.06$; Fig. 6C). Experiments performed in growth chambers tended to be shorter in duration than those in greenhouses, nurseries, or in the field (one-way ANOVA, $P < 0.001$, $F_{3,578} = 335.8$; Table 1). The magnitude of contamination was positively related to the duration of experiments ($P < 0.001$, df = 1, 418, $r^2 = 0.14$; Fig. 7).

There was evidence for significant publication bias in data for total biomass, the only measure to be comprised of data from >25 papers. Spearman's rho for the correlation between effect size and sample size was −0.28 ($P < 0.001$), indicating that there was an overrepresentation of studies with positive effect sizes at low replication.

Seedling response to ectomycorrhizal inoculation and phosphorus addition

We analyzed 234 studies (six host and 15 fungal genera) from 10 papers for changes in total biomass of seedlings inoculated with ectomycorrhizal fungi under phosphorus (P) additions ranging from 0 to 136 mg P/kg (Appendix B). The cumulative effect size was positive (0.0769), but the 95% bootstrapped confidence intervals overlapped zero, indicating there was no average change in total biomass of seedlings inoculated with ectomycorrhizal fungi subjected to manipulated phosphorus levels when all levels of substrate P, including no additions, were included. There was underlying structure in the data ($P < 0.001$, df = 232, $Q_T = 1236$); however, of the explanatory variables tested (host genus, fungal genus, and amount of P added), only host genus explained a

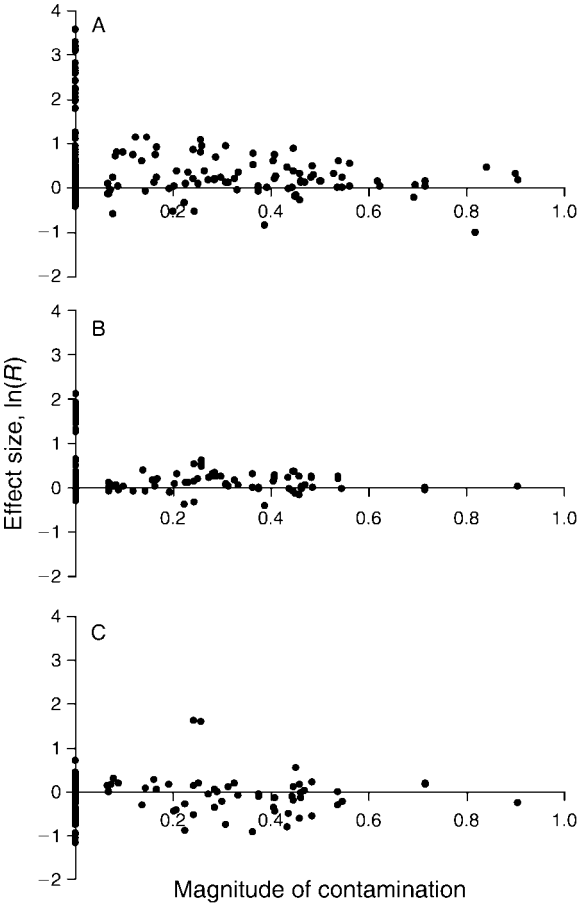

FIG. 5. Relationship between mean effect sizes and magnitude of contamination for (A) total biomass, (B) shoot height, and (C) shoot : root ratio.

TABLE 1. The relationship between the locations of the experiments and the magnitude of contamination and the duration of association.

Location	*n*	Mean	SE
Magnitude of contamination			
Growth chamber	106	0.0026^{A}	0.0160
Greenhouse	243	0.10^{B}	0.0105
Nursery	28	0.43^{C}	0.0311
Field	43	0.33^{C}	0.0251
Duration of association (weeks)			
Growth chamber	122	13^{A}	0.9
Greenhouse	374	18^{B}	0.5
Nursery	40	26^{C}	1.6
Field	43	67^{D}	1.6

Note: Means followed by the same letter are not statistically different (Tukey-Kramer honestly significant difference test, $\alpha = 0.05$).

significant amount of variation in effect size ($P < 0.001$, df = 3, 24, $Q_M/Q_T = 0.31$). Specifically, seedlings of the genera *Eucalyptus*, *Pinus*, and *Larix* responded less positively than those of *Picea* when inoculated, regardless of phosphorus level.

DISCUSSION

Context dependency of the mutualism–parasitism continuum: biotic and abiotic factors

Our meta-analysis shows that the position along the mutualism–parasitism continuum on which hosts fall is contingent on biotic factors, namely the identity of plant partners involved in ectomycorrhizal associations. The abiotic context, i.e., phosphorus levels, did not modify host response to ectomycorrhizal inoculation. We also determined that on average, seedlings across multiple host species had more biomass when inoculated with ectomycorrhizal fungi, regardless of the identity of the fungal associate. This finding supports those from research on non-symbiotic interspecific interactions; for example, host plants are often generalists with response to different pollinators (Zamora 2000). This result conforms to theory predicting the outcome of multispecific mutualistic systems, i.e., interactions involving many species tend to result in the evolution of generalists because reciprocal specialization is unlikely (Howe 1984). In forest stands, the number of species of ectomycorrhizal hosts is typically an order of magnitude less than that of its fungal symbionts (Bruns 1995). Reciprocal specialization is unlikely in this system due to the changing composition of ectomycorrhizal fungi both spatially (Izzo et al. 2005, Genney et al. 2006, Toljander et al. 2006) and temporally (Izzo et al. 2005, Koide et al. 2007). Thus, hosts may adapt to "landscapes" (sensu Howe 1984) of ectomycorrhizal fungi in which fungal species diversity diffuses selection from one source.

Although the mutualism–parasitism continuum has become well accepted in the mycorrhizal literature, prior to our analysis the empirical data supporting the concept came primarily from studies on arbuscular-mycorrhizal systems. Our study provides the most thorough quantitative characterization of the range of host outcomes to ectomycorrhizas. The continuum of responses (i.e., confidence intervals) we observed for measures of host biomass was generated by the specific response a particular genus of host had to ectomycorrhizal inoculation. In particular, *Quercus* seedling biomass and biomass allocation to roots ranked highest, and *Picea* lowest, with ectomycorrhizal inoculation. Mycorrhizal dependency has been hypothesized to relate to various root morphological traits such as root thickness, surface area, and incidence of root hairs (Brundrett 2002). In addition, dependency on arbuscular mycorrhizas seems to be higher for hosts that have small seeds or have had seed reserves experimentally reduced (Janos 1980, Allsop and Stock 1995, Siqueira et al. 1998, Zangaro et al. 2003). In our data set, we cannot say whether host rankings are taxon- or trait-specific, due to

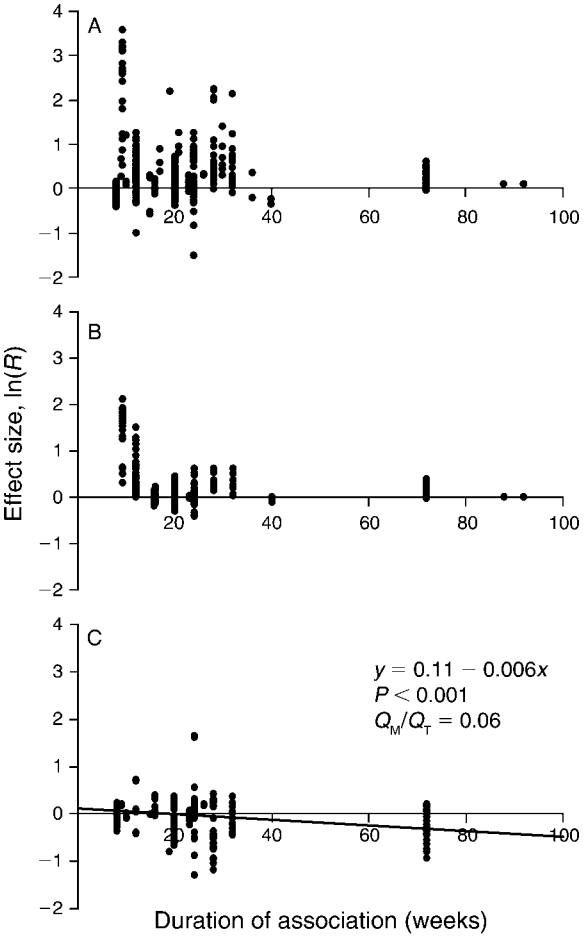

FIG. 6. Relationship between effect sizes and duration of association of ectomycorrhizal fungus and host for (A) total biomass, (B) shoot height, and (C) shoot : root ratio. Q_M/Q_T is the amount of total heterogeneity in the data due to variation in effect sizes explained by the model. Statistics and regression line are shown for significant models only.

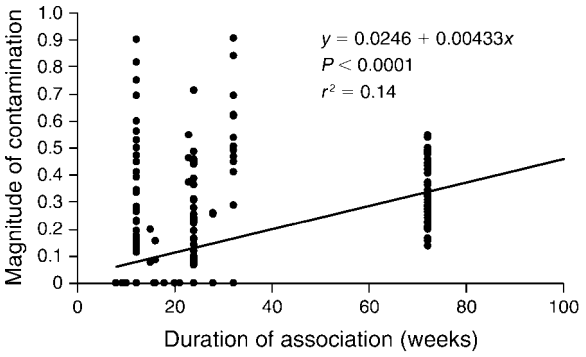

FIG. 7. Relationship between the magnitude of contamination and the duration of association of ectomycorrhizal fungus and host.

the relatively few genera included in the analysis. Multiple fungal taxa at various levels of these traits would be required to effectively test the impact of traits on mycorrhizal dependency. Going beyond taxonomic correlations with inoculation responses and identifying those host traits that correlate to specific outcomes would enrich our understanding of ectomycorrhizal interactions. In particular, further research within a broad framework, such as that developed for leaf traits (Wright et al. 2004), would be especially fruitful to understand trade-offs among plant traits and mycorrhizal responsiveness.

Levels of colonization were not important in explaining host responses to ectomycorrhizal inoculation. Our results indicate that the positive responses measured by increases in seedling biomass and height may be equally expressed through colonization levels ranging from 1% to 98%. Given that positive effects of inoculation were observed at such low levels of colonization for both seedling biomass and shoot height, we suggest that chemical mechanisms may underlie host responses to ectomycorrhizal inoculation. It is well established that plant-associated microorganisms are capable of synthesizing phytohormones that are used for communication between a host and its microflora (Tsavkelova et al. 2006). For example, small amounts of auxins increase shoot elongation and dry mass of wheat inoculated with rhizobacteria (Khalid et al. 2004). Auxins, which are involved in a wide variety of physiological responses that influence growth of woody plants (Kozlowski and Pallardy 1997), are also produced by ectomycorrhizal fungi (Barker and Tagu 2000). Though some research has been conducted on the effects of auxins on ectomycorrhizal development (e.g., Niemi et al. 2002, Rincon et al. 2003), their role at the level of the host has been neglected.

That hosts are unresponsive to the identity of fungal partner and abundance of fungi occurring on root tips yet are sensitive to the presence or absence of ectomycorrhizas indicates that the variation in ectomycorrhizal fungi perceived and selected for by the host plant may be of a discrete rather than continuous nature. But the manner in which hosts perceive ectomycorrhizas is not concordant with the distribution of ectomycorrhizal fungi at the scale of an individual host. While particular species of ectomycorrhizal fungi may occur in discrete patches, seldom do we find abrupt boundaries representing "mycorrhizal" and "non-mycorrhizal" patches (Lilleskov et al. 2004), nor do we often find ectomycorrhizal hosts that are free of mycorrhizas. That is, the strongest host response is conditional on a situation that infrequently occurs in nature.

Admittedly, our results are not definitive on the role of fungal identity with regard to host response to ectomycorrhizas for two reasons. First, although seedling biomass and height were not modified by the particular genus of fungi used in inoculation trials, this factor appears to have a role in altering host allocation to shoot : root ratios. Seedling allocation to roots increased by almost three times when inoculated by fungi from the genus *Scleroderma*. Diédhiou et al. (2005) concluded that *Scleroderma dictyosporum* has a higher requirement for glucose relative to thelephoroid species, perhaps related to construction costs of its network-like mycelium (Newton 1991). Plants growing in nutrient-depleted soils allocate more biomass to roots than shoots (Gedroc et al. 1996). If association with fungi from this taxon is perceived by the host as equivalent to growing in nutrient-depleted soils, this would explain allocation patterns. Second, some distinction among fungal genera by hosts must be present since there appeared to be a difference between those fungi that contaminate seedlings and those used to inoculate seedlings. Because there was no effect of magnitude of contamination on all three growth measures despite a cumulative positive effect, contaminant fungi were likely neutral in their effects. Species of contaminant fungi were for the most part unidentified but included those from the genera *Thelephora* and *Cenococcum*. These fungi are common, widespread, and widely dispersed via airborne spores; whether such characteristics of fungi and magnitude of host response covary should be further studied.

In addition, the role of fungal identity in our analysis could be masked if there were an interaction between fungal taxon and nutrient levels. For example, Bougher et al. (1990) have indicated there is an interaction between the effects of fungal taxa and P additions. Specifically, at low P additions (2–12 mg P/kg soil) differences among *Desoclea maculate*, *Laccaria laccata*, and *Pisolithus tinctorius* in host dry mass production are apparent, but these differences are not apparent at >16 mg P/kg soil. A similar interaction was reported for seedlings colonized by *Laccaria bicolor* or *Thelephora terrestris* along a P gradient (Jones et al. 1990). Our meta-analysis could not detect such an interaction because not all host–fungi combinations were present across the range of P additions.

Context dependency of the mutualism–parasitism continuum: experimental conditions

Factors unrelated to inoculation per se have potentially influenced interpretation of host responses to ectomycorrhizal inoculation, namely publication bias towards large positive effects, the pairing of host and fungal symbionts not known to occur together, and the duration of experiments. The presence of these factors effectively reduces and distorts the spectrum on which host responses to ectomycorrhizal inoculation are evaluated.

1. *The spectrum is reduced: publication bias inflates measures of effect sizes.*—Under a model of no publication bias, estimated effects should be distributed around the unknown true effect, with the spread of the effects representing their variances. As sample sizes increase, the spread of the distribution should decrease, resulting in a funnel-shaped distribution of effect sizes. Publication bias against studies with negative results will produce a negative correlation between sample size and the magnitude of effect (Begg and Mazumdar 1994), and this inflates the magnitude of overall effect sizes calculated in a meta-analysis. Because the lower limit of the cumulative effect on total biomass is well above zero, there may indeed be a change in seedling biomass upon inoculation. However, if publication bias were absent, the spread of effect sizes may also include negative to neutral values. Thus, the presence of publication bias reduces the spectrum of positive to negative outcomes within mycorrhizal associations.

Among the papers used in this meta-analysis, Dixon et al. (1984) and Hung and Molina (1986) explicitly reported that data had been omitted due to nonsignificant differences between control and inoculated seedlings. It is unlikely that these particular omissions alone caused publication bias in our data set, but they may be symptomatic of bias in the selection of data reported in published papers. At the other extreme, although they did not affect the results of the meta-analysis, host–fungus pairings extracted from Burgess et al. (1994; identified as outliers in Fig. 4A, B) were irregularities in our data set, reporting highly positive responses to ectomycorrhizal inoculation by various strains of *Pisolithus*. Due to the tradition of categorizing mycorrhizal fungi as mutualists, such extreme positive results are unlikely to go unpublished. Negative results in mycorrhizal research may be more likely to go unpublished compared to other fields in which no a priori expectation exists of the magnitude or direction of the outcome of species interactions.

2. *The spectrum is distorted: effects of crossing hosts and ectomycorrhizal fungi not known to co-occur remain poorly understood.*—Inoculation trials are often performed using artificial pairings of host and fungus (e.g., Chen et al. 2006). The geographic origin of fungi and hosts used in trials may affect inoculation responses in unpredictable ways. Similar to plants, some but not all species of ectomycorrhizal fungi are cosmopolitan in their distribution. One corollary to this pattern is that not all host and ectomycorrhizal fungal species will interact and at any given location a host species will encounter a subset of the global pool of ectomycorrhizal fungi. This geographic variation in plant–mycorrhizal community structure has likely resulted in a mosaic of coevolution between plants and mycorrhizal fungi (Thompson 2005), but we still have very few data on the consequences of this mosaic on mycorrhizal inoculation responses (but see Monzon and Azcon 1996, Klironomos 2003, Sylvia et al. 2003, Hoeksema and Thompson 2007). This lack of knowledge of the range of host responses to exotic symbionts also carries over to conservation research; the ecological consequences of mycorrhizal fungal species' introductions are unpredictable (Schwartz et al. 2006).

In our meta-analysis, we could not categorize each host–fungus pairing as "local" or "foreign," as such information was either unavailable or it was not clear at what scale we should consider a host and fungal species to co-occur (e.g., within a forest stand, region, or country). Studies on arbuscular mycorrhizas have shown that crossing local plants and fungi produces a greater range in responses measured by plant biomass than for crosses involving foreign symbionts (Klironomos 2003). Conversely, variation in plant growth was independent of fungal isolates when different geographic populations of three host plant species were crossed with four populations of the ectomycorrhizal fungus *Rhizopogon occidentalis* (Hoeksema and Thompson 2007). Origin of fungal isolate also was not found to be important in modifying growth of *Eucalyptus globulus* (Thompson et al. 1994) and of *Salix repens* (van der Heijden and Kuyper 2001*b*). These findings are consistent with our results that variation in fungal identity bears little consequence to variation in shoot height or seedling biomass.

3. *The spectrum is distorted: factors that covary with time may cause spurious effects.*—Over time, inoculated seedlings allocated more biomass to roots vs. shoots compared to control seedlings. If inoculated seedlings were to become root-bound in less time than control seedlings (because of increased total biomass), allocation patterns in seedlings would be time-dependent and not necessarily determined by ectomycorrhizas. We also noted that experiments performed in growth chambers were of shorter duration than experiments performed in greenhouses, nurseries, or in the field. Thus, allocation patterns appear to be confounded with the duration of experiments and ultimately the location of experiment. The duration of experiments was also positively correlated to the magnitude of contamination; however, contamination levels had no impact on host response to ectomycorrhizal inoculation.

Conclusions

The reliance on comparisons between mycorrhizal and non-mycorrhizal individuals, beyond its heuristic

purpose, is somewhat artificial because these conditions are seldom found in nature. As such, we add realism to our knowledge on the ecological and evolutionary consequences of ectomycorrhizal associations when we consider host responses to variation in the identity and abundance rather than just the presence or absence of fungi. Our meta-analysis, however, showed that for the most part, hosts do not respond to variation in the identity or abundance of ectomycorrhizal fungi. Differences among host genera appear to govern the magnitude of host response to ectomycorrhizal inoculation. And although we found that the role of fungal genus modified shoot : root ratios, the relevance of this measure of host response is uncertain because it was also related to the duration and the setting in which experiments were performed. Thus, to conclude upon results from >20 years of inoculation trials: the effect of ectomycorrhizal partners on host position along the mutualism–parasitism continuum appears unilateral.

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APPENDIX A

Identity of host plant and fungal species pairings and effect sizes [ln(R)] for seedling biomass, shoot height, and shoot : root ratio for each study used in the meta-analysis and full literature citations (*Ecological Archives* E089-062-A1).

APPENDIX B

Identity of host plant and fungal species pairings with associated effect sizes [ln(R)] for seedling biomass and full literature citations (*Ecological Archives* E089-062-A2).