Mycorrhiza (2012) 22:227–235
DOI 10.1007/s00572-011-0398-7

ORIGINAL PAPER

Do arbuscular mycorrhizal fungi affect the allometric partition of host plant biomass to shoots and roots? A meta-analysis of studies from 1990 to 2010

Stavros D. Veresoglou · George Menexes · Matthias C. Rillig

Received: 11 January 2011 / Accepted: 10 June 2011 / Published online: 28 June 2011

Abstract Arbuscular mycorrhizas (AM) are ubiquitous root symbioses with often pervasive effects on the plant host, one of which may be above- and belowground biomass allocation. A meta-analysis was conducted on 516 trials that were described in 90 available articles to examine whether AM colonization could result in a modification of partitioning of plant biomass in shoots and roots. It was hypothesized that alleviating plant nutrient limitations could result in a decrease of root to shoot (*R*/*S*) ratio in AM plants or, alternatively, the direction of shifts in the *R*/*S* ratio would be determined by the changes in total dry biomass. In our analysis, we considered four types of stresses: drought stress, single heavy metal stress, multiple heavy metal stress, and other potential abiotic plant stress factors. When disregarding any factors that could regulate effects, including stress status and mode of propagation, the overall AM effect was a significant modification of biomass towards shoot growth. However, the responses of stressed and clonally propagated plants differed from those of seed-grown unstressed plants. Our meta-analysis detected a considerable decline in the *R*/*S* ratio when plants were grown from seeds in the absence of abiotic stresses. Moreover, we demonstrate that additional regulators of the AM-mediated impact on *R*/*S* ratio were presence of competition from other plants, plant growth outcome of the symbiosis, growth substrate volume, experimental duration, and the identities of both plant and AM fungus. Our results indicate that a prediction of AM effects on *R*/*S* allocation becomes more accurate when considering regulators, most notably propagation mode and stress. We discuss possible mechanisms through which stress and other regulators may operate.

Keywords Arbuscular mycorrhizas · Root-to-shoot ratio · Physiological plant responses · Plant growth stress factors · Allometric partitioning of biomass

Introduction

Arbuscular mycorrhizas (AM) represent a ubiquitous symbiosis between terrestrial plant roots and fungi of the phylum *Glomeromycota* (Schüßler et al. 2001). The symbiosis entails a carbon cost for the plant host, which receives in return improved access to nutrients, protection from pathogens, and a range of other benefits (Smith and Read 2008). The physiological responses of plants to AM inoculation are numerous and include an increase in size (Hoeksema et al. 2010), higher plant tissue phosphorus concentrations, and higher photosynthetic rates (Smith and Read 2008).

An allometric analysis of plant responses to environmental factors represents a widely investigated topic in plant physiology (Farrar and Gunn 1998). The vast majority of studies have confined allometric analysis to “shoot” and “root” partitions, although, ideally, further compartmentalization of plant biomass should be conducted (Poorter and Nagel 2000). In brief, root to shoot (*R*/*S*) ratio appears to be

---

Electronic supplementary material The online version of this article (doi:10.1007/s00572-011-0398-7) contains supplementary material, which is available to authorized users.

S. D. Veresoglou (✉) · M. C. Rillig
Institut für Biologie, Freie Universität Berlin,
Altensteinstr. 6,
14195 Berlin, Germany
e-mail: seby31@zeroone.net

G. Menexes
Laboratory of Agronomy, School of Agriculture,
Aristotle University of Thessaloniki,
Thessaloniki, Greece

dependent on the identity of the plant species and plant growth stage (Ledig and Perry 1966). Additional variation results from seasonality, as higher *R*/*S* values are recorded in the beginning and the end of a growth season (Haolin et al. 2008). Increased nutrient availability causes decreased belowground plant biomass allocation (e.g., Reynolds and D'Antonio 1996; Hermans et al. 2006). By contrast, higher irradiance can favor biomass allocation to the shoots (e.g., Poorter and Nagel 2000). Allocation towards nutrient acquisition or light capture reflects a dynamic equilibrium between root and shoot allocation.

The attention to *R*/*S* allometric analysis of plants originated from the fact that for individual plant species this parameter represents a good index of soil fertility and consequently plant health (e.g., Harris 1992). Moreover, challenges in determining root biomass and, thus, accurately assessing crop biomass mean that an estimate of *R*/*S* ratio could represent an alternative way of conducting biomass assessment with data on the aboveground part of the plant only (e.g., Monk 1966); similarly, plant competitive dynamics in the absence of root data could be better described (Robinson et al. 2010) or the contribution of root biomass to soil carbon (Rasse et al. 2005). Clearly, *R*/*S* allometric allocation could have important implications on decomposition rates and terrestrial carbon storage. *R*/*S* ratios, however, in several cases may be misleading. Primarily, the combination of stems and leaves in a single compartment, despite their functional differences, renders interspecies comparisons of *R*/*S* ratios precarious (e.g., Poorter and Nagel 2000). Moreover, the *R*/*S* ratio lacks a qualitative assessment of the extent of the root system.

With regards to AM colonization, evidence suggests that upon colonization many physiological changes occur, including increases in lateral root length, branching, and fine root length (Hooker et al. 1992; Yao et al. 2009). However, we are not aware of a systematic study of how overall plant allometric partitioning to shoot and root may be affected by AM colonization. Alleviation of host nutrient limitation as a result of AM fungal establishment (e.g., Smith and Read 2008) could be expected to result in decreases of the *R*/*S* ratio (e.g., Mamolos et al. 1995; Wilson and Tilman 1995). Isolated studies, however, have not been informative because both increases (e.g., Nuortila et al. 2004; Davies et al. 2005; Rabie 2005) and decreases (e.g., Pérez and Urcelay 2009; Zandavalli et al. 2004) in the *R*/*S* ratio have been reported. Reviews on this topic are not available, and thus standard textbooks also do not deal with such effects in detail (e.g., Smith and Read 2008). Therefore, we have conducted a meta-analysis on existing literature of a period spanning the last 20 years (1990–2010) in an attempt to quantitatively address the effect of AM inoculation on *R*/*S* ratio of plants. Analysis was confined to data on shoot and root partitions so that a maximum number of studies could be integrated. We hypothesized that AM inoculation would result in decreases in the *R*/*S* ratio through ameliorating plant species nutrient status and that this would occur irrespective of the fitness outcome of the symbiosis for the plant. We further hypothesized that the identity of both the plant host and the AM fungus would be key regulators of such a decrease in the *R*/*S* ratio.

Materials and methods

Sources of data

On the 3rd of November 2010, we conducted a search in the ISI Web of Science ® database using the keyword combination "arbuscular and shoot and root." The 814 articles retrieved were screened so that they met the following criteria: (1) Plants had been grown on a soil or soil–sand mix substrate; (2) Full control of AM propagules was possible through prior sterilization of the growth substrate; and (3) Comparisons were possible in the absence of interactions with organisms other than rhizobia and microbes present in the soil/growth substrate. A random subset of 50% of the screened articles, totaling 90, was used to construct the database (others have also used randomly sampled subsets of studies, e.g., Hoeksema et al. 2010). In agreement with other studies, several trials may be extracted from each article (e.g., Curtis and Wang 1998; Lekberg and Koide 2005). In some cases, data were extracted from graphs, provided that they were legible.

Data on *R*/*S* ratio in the presence and absence of AM fungi were collected along with the following nine characteristics to be used as categorical explanatory variables in the meta-analysis:

Stress type: The variable had five levels: non-stressed plants, water-stressed plants, heavy metal stress, other stresses (comprised elevated CO_2 effect and acid precipitation stress), and multiple stress (which included the cases where more than one heavy metal stress concurred—no cases of combined stress factors other than heavy metal pollution were retrieved).

Seed: The variable had two levels (plants grown from seed and plants propagated clonally).

AM impact: The variable had two levels (positive/neutral host plant response to AM inoculation and negative response of host plant to AM inoculation).

Plant competition: The variable had two levels (plants grown alone and plants grown in intra-specific or inter-specific competition).

Space available for plant growth: The variable was calculated as the quotient of soil weight (or, where absent, pot size in liters) to the number of seedlings grown. To allow a classification of studies based on the available space for plant growth (ASPG), we faced the following two problems: (1) this had to be done at a crude scale so that the sample sizes may yield meaningful results; and (2) it is difficult to find relevant references on what may be large or small and these had to be somewhat arbitrarily selected. We decided to group ASPG based on our experience on plant responses to pot sizes. The first category included microcosms of such size that were unable to support the growth of the majority of plants for a substantial period of time (up to 200 g), and the second category represented microcosm sizes where growth and reproduction of most plants is possible but this results in a high root/soil volume. This roughly described microcosms with a size up to 1–1.5 kg. The final category described conditions that resembled field growth. A common source of misunderstandings with classifications occurs when volumes that match the limits of categories are encountered—we dealt with this issue through modifying the classes so that a limited number of trials were of ASPG close to the limits of the classes. Therefore, the independent categorical variable had three levels (low, less than 300 g of growth substrate or 300 ml of pot space; intermediate, 300–1,500 g of soil or 300 ml–1.5 l of pot space; high, higher than 1,500 g of soil or 1.5 l of pot space).

Duration of the experiment: The logic behind classifying studies according to duration was similar to ASPG. Two months represent a minimum time span to allow for AM colonization, whereas studies longer than 2 years (or 25 months so that classification will be easier) represent long-term controlled studies. Thus, the variable had three levels (short, studies with duration of 2 months or less; intermediate, studies with duration up to 25 weeks; long, studies with duration longer than 25 weeks).

Plant functional group: The variable had five levels (non-woody grasses, non-woody legumes, non-woody non-legume dicotyledons, woody legumes, woody non-legume plants).

Woody: The variable had two levels (non-woody, woody).

AM group: The variable had five levels (mixture of more than one AM isolate, *Glomus mosseae* isolate, *Glomus intraradices* isolate, other *Glomeraceae* AM isolate, *Gigasporaceae* isolate).

Candidate categorical dependent variables were selected so that they represented key factors that potentially affect the *R*/*S* ratio of plants. Plant growth under stress conditions often results in responses to the stress factors that include modification of *R*/*S* ratio. For example, heavy metal-stressed plants tend to have a lower *R*/*S* ratio (Roosens et al. 2003). Plant competition status is a factor that has recently been detected to initiate increased plant biomass allocation belowground (Murphy and Dudley 2007). Pot size may, too, have strong implications for the resulting *R*/*S* ratio as small growth substrate volumes may limit the allocation of additional plant growth to root biomass (e.g., Schroeder and Janos 2005). For other plant symbionts, e.g., *Azospirillum* spp., the interactive effects of pot size and inoculation on host *R*/*S* ratio have also been found (Veresoglou and Menexes 2010). With respect to the duration of an experiment, herbaceous plants are believed to decrease their *R*/*S* ratio with time, whereas woody plants increase it (Poorter and Nagel 2000). Therefore, an additional analysis of experiment duration was conducted separately for woody and herbaceous plants. We separately examined instances when AM colonization had a positive effect on plant biomass with those when it had a negative effect so that we could address whether the AM-mediated effect on *R*/*S* ratio is actually a result of improved overall plant growth. Finally, clonal plants face distinct ecological challenges and this could potentially result in distinct *R*/*S* ratio responses (e.g., Bernier et al. 1995).

Transformations and effect sizes

The *R*/*S* ratio index suffers from the following two problems as described by Körner (1994): (1) deviation from normality and (2) variability in shoot and root biomass does not have the same absolute impact on the *R*/*S* ratio. For the above reasons, a logarithmic transformation is usually proposed (e.g., Poorter and Nagel 2000). Therefore, the chosen effect size was the natural log of the ratio of *R*/*S* responses, $\mathrm{ES}_{R/S} = \ln\left(\frac{R/S_{\mathrm{AM}}}{R/S_{\mathrm{control}}}\right)$, that equals the difference of the log-transformed *R*/*S* responses.

To account for the lack of variance information in many studies and in order to make the analysis as inclusive as possible, the non-parametric weight statistic, $w_{ij} = \frac{N_{ij}{}^{\mathrm{E}} N_{ij}{}^{\mathrm{C}}}{N_{ij}{}^{\mathrm{E}} + N_{ij}{}^{\mathrm{C}}}$, was adopted, where $N_{ij}{}^{\mathrm{E}}$ and $N_{ij}{}^{\mathrm{C}}$ are the experimental and control sample sizes, respectively (Adams et al. 1997; Gurevitch and Hedges 1999). This method has been widely used in ecological studies in the past (e.g., Hoeksema and Forde 2008; Lekberg and Koide 2005). To exclude the possibility that the $\mathrm{ES}_{R/S}$ reflects the *R*/*S* ratio of the control plants, a weighted Spearman's rho (ρ) statistic ($\mathrm{ES}_{R/S}$ vs. $\ln_{R/S\mathrm{control}}$) was calculated, resulting in $\rho = -0.396$, $P < 0.001$ (see Fig. A1 in "Appendix"). However, further analysis with Loess regression (a smoothing technique that uses locally weighted polynomials; Jacoby 2000) revealed that the relationship of the two variables explained a negligible part of the variation in the effect sizes (we retrieved an overall slope close to zero) and was not linear (see Fig. A1 in “Appendix”). In the past, the authors have applied correction procedures (e.g., linear interpolation) in studies where a systematic error factor, with a continuous distribution, was present (e.g., Veresoglou and Menexes 2010). Despite the significance of the ρ correlation coefficient, because of our inability to accurately specify a systematic source of error, we were reluctant to adopt a similar correction procedure.

Meta-analysis

Meta-analysis was conducted in MetaWin v2.0 software (Rosenberg et al. 2000) based on a random effects model (Borenstein et al. 2009; Gurevitch and Hedges 1999). We estimated 95% confidence intervals (CIs) of the means following a bootstrapping procedure of the dataset with 3,999 iterations. Meta-analytical 95% CIs based on bootstrapping are known to be wider than standard asymptotic confidence intervals (Adams et al. 1997); however, they still suffer from high likelihood of type I errors when used for multiple comparisons.

Hence, for treatment comparisons, the probability of inter-treatment differences (Q statistic) was examined through a randomization procedure with 3,999 iterations. Treatment groups were considered statistically different if the P value resulting from the randomization test was significant and the corresponding 95% CIs for means did not overlap (Adams et al. 1997). The resampling approach adopted, when combined with bootstrapped CIs, does not assume normality of the dataset (Gurevitch and Hedges 1999; Rosenberg et al. 2000). Therefore, we did not test the dataset for normality. Medians and their 95% CIs were calculated according to a non-parametric method reported in Conover (1980): the width of the CIs (in the form of a percent value) is determined by the number of records analyzed (narrower CIs when many records are available). The next step is sorting of the studies according to their reported effect sizes and the addition of a column where the cumulative weight of studies is calculated. Medians and 95% CIs are then calculated following a transformation of the percent values to the corresponding cumulative weight equivalents and are projected to the initial ESs. This procedure does not always lead to symmetric CIs. The medians and their 95% CIs are not discussed in the manuscript but are provided as a means to assess the robustness of the estimates on bootstrapped mean CIs. Significance was assumed for $P<0.05$.

Results

An overall negative effect of AM colonization on *R*/*S* ratios was detected when the full dataset was used (Fig. 1a). The first step of the analysis aimed at increasing the homogeneity of the dataset. A preliminary inspection of data revealed that clonally propagated plants reacted differently to AM colonization. Analysis confirmed that, whereas the *R*/*S* ratios of plants that were raised from seedlings declined with AM addition, *R*/*S* ratios of clonally propagated plants increased (Fig. 1b, $P=0.0003$). We thus excluded clonally propagated plants from further analysis. We also wanted to test whether the stress to which plants had been exposed could have an effect on the AM-mediated impact on *R*/*S* ratios. Given that the vast majority of trials had been grown in the absence of stresses, this could have resulted in unwanted heterogeneity. The effect of stress was significant (Fig. 1c, $P=0.0018$) and the “reduced dataset” comprised all trials but those on clonally propagated material and on stressed plants. It is interesting to note that non-stressed plants were the only “stress type” level with a mean ES$_{R/S}$ significantly different from zero (Fig. 1c). The “reduced dataset” included 342 trials out of a total of 516 trials and had a mean ES$_{R/S}$ of −0.2377 (Fig. 2a).

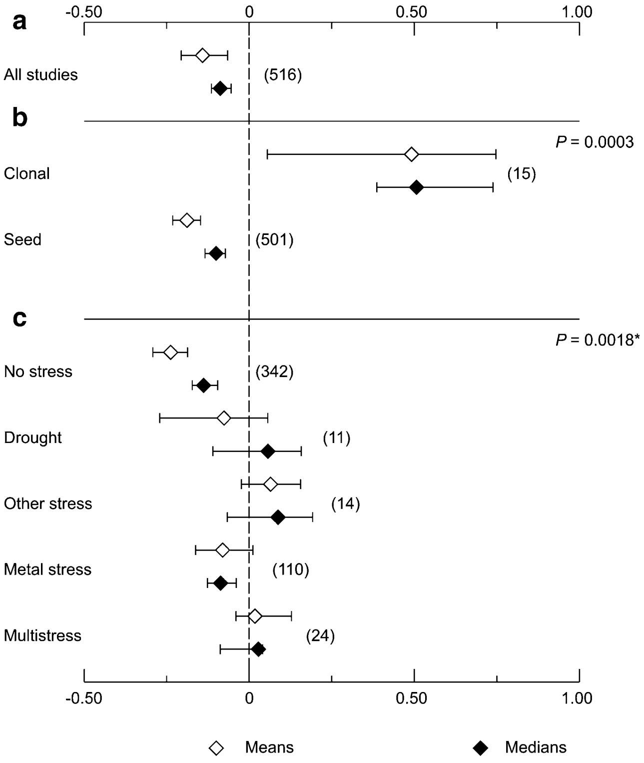

Fig. 1 Effect sizes (ES$_{R/S}$)—means and 95% CIs (*white rhombs*) and medians and 95% CIs (*black rhombs*)—for **a** all studies included in the database, **b** studies grouped according to their mode of propagation, and **c** studies grouped according to the stress factors experienced by the plants. *Numbers in parentheses* refer to the number of trials that were present in the specific group. P values reported are obtained from the permutation test. *Asterisk* is used to denote that the specific analysis was carried out exclusively to plants grown from seeds

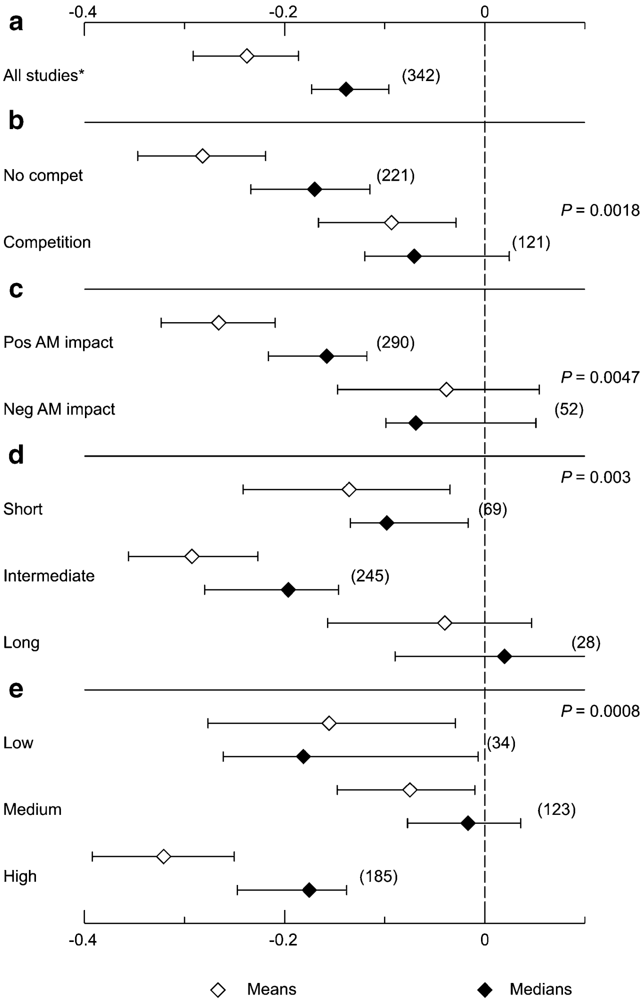

Fig. 2 Effect sizes ($\mathrm{ES}_{R/S}$)—means and 95% CIs (*white rhombs*) and medians and 95% CIs (*black rhombs*)—for trials of the "reduced dataset" grouped as following: **a** mean effect of all studies, **b** plants grown in the absence vs. presence of competition, **c** trials where a positive and negative AM plant growth effect was reported, **d** trials grouped according to the duration of the study, and **e** trials grouped according to plant growth substrate available volume per seedling. *Numbers in parentheses* refer to the number of trials that were present in the specific group. *P* values reported are obtained from the permutation test. *Asterisk* is used to denote that analysis was carried out exclusively to unstressed plants grown from seeds

An analysis of the "reduced dataset" (see above) revealed that competition was a significant regulator of the AM impact on *R*/*S* ratios as the AM-mediated decline in *R*/*S* ratio was stronger in plants grown in isolation (Fig. 2b, P=0.0018). Moreover, the plants that had responded to AM colonization with reduced plant growth exhibited less negative $\mathrm{ES}_{R/S}$ than when colonized hosts reacted with increased growth (P=0.0182) (Fig. 2c). The effect of the duration of the experiment on the $\mathrm{ES}_{R/S}$ was significant, but we failed to retrieve specific patterns of AM-mediated plant responses to experimental length (Fig. 2d). However, when herbaceous and woody plants were separately analyzed, there was a trend for the $\mathrm{ES}_{R/S}$ to decrease with age in herbaceous plants (Fig. 3b), whereas $\mathrm{ES}_{R/S}$ increased with age in woody plants (P=0.0076) (Fig. 3c). It should be noted that, amongst the nine studies with herbaceous plants that lasted more than 25 weeks, eight reported $\mathrm{ES}_{R/S}$ below −0.34. However, a single, heavily weighted study reported a positive $\mathrm{ES}_{R/S}$ and could have accounted for the non-significant result of the comparison in Fig. 3b as well as the poor assessment of the median. A key determinant of the allometric response of plants to AM colonization appeared to be the space available for plant growth (P=0.0008). Plants with more available space tended to further promote aboveground growth (more negative $\mathrm{ES}_{R/S}$ values were recorded for the "high" group when compared to the

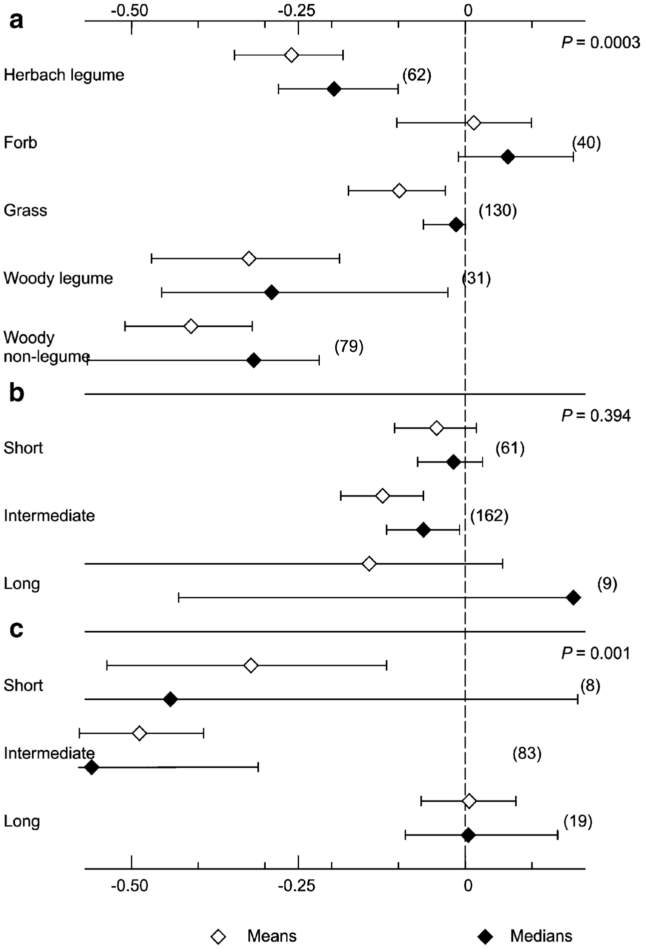

Fig. 3 Effect sizes ($\mathrm{ES}_{R/S}$)—means and 95% CIs (*white rhombs*) and medians and 95% CIs (*black rhombs*)—for **a** trials grouped according to the functional group of the host plant, **b** studies with herbaceous plants grouped according to their duration, and **c** studies with woody plants grouped according to their duration. *Numbers in parentheses* refer to the number of trials that were present in the specific group. *P* values reported are obtained from the permutation test. All analyses were conducted on the "reduced dataset" that did not include clonally propagated or stressed plants

"intermediate" group, which represented the two groups with the most trials) in the presence of AM fungi (Fig. 2e).

Both identities of plant functional group (Fig. 3a) and AM inocula (Fig. 4a) appeared to have significant effects on $ES_{R/S}$. For the effect of plant functional group, differences could be mainly observed amongst woody (that tended to have more negative $ES_{R/S}$) and herbaceous plants, although a significant difference was also present between legumes and other herbaceous plants (Fig. 3a). Mixed AM inocula appeared to have a stronger effect ($P=0.0043$) on mediating R/S allometric responses when compared with single isolates (Fig. 4b). No differences could be detected amongst *Glomeraceae* isolates (Fig. 4c), but isolates from the *Glomeraceae* family appeared to be less efficient in promoting shoot growth than *Gigasporaceae* isolates (Fig. 4d). For this last comparison, caution is needed due to the low level of replication of the *Gigasporaceae* trials.

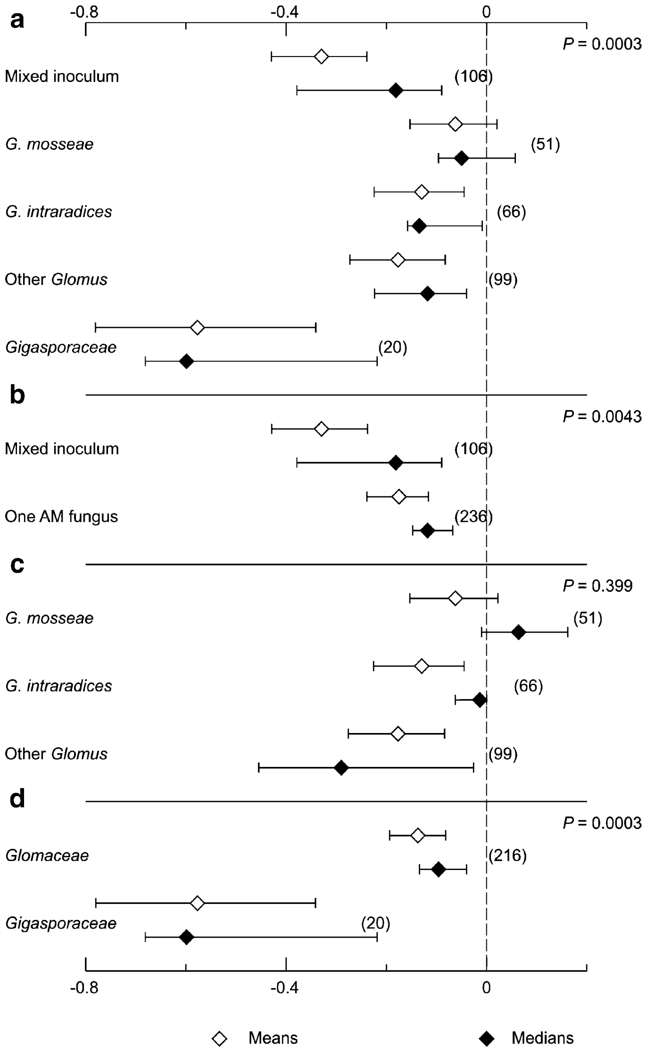

Fig. 4 Effect sizes ($ES_{R/S}$)—means and 95% CIs (*white rhombs*) and medians and 95% CIs (*black rhombs*)—for **a** studies grouped according to the identity of the AM symbiont, **b** studies with one or more AM isolates, **c** studies that have used *Glomeraceae* isolates, and **d** *Glomeraceae* vs *Gigasporaceae* comparison. *Numbers in parentheses* refer to the number of trials that were present in the specific group. *P* values reported are obtained from the permutation test. All analyses were conducted on the "reduced dataset" that did not include clonally propagated or stressed plants

Discussion

Methodology

A compilation of the available literature of AM manipulation studies on R/S ratios resulted in a heterogeneous dataset. To account for the within-group variance heterogeneity problem, often encountered in ecological meta-analyses (Gurevitch and Hedges 1999), a bootstrapping approach was utilized. The bootstrapping methodology applied is becoming a default methodology for inter-treatment comparisons in meta-analyses as it results in higher accuracy than respective maximum likelihood estimation methods (Adams et al. 1997; van den Noortgate and Onghena 2005). Information on CIs, when used alone, may be prone to type I errors but is occasionally adopted in meta-analytical studies as the sole statistic for inter-treatment comparisons (e.g., Lekberg and Koide 2005; Wang et al. 2008). This is why a permutation test was conducted here as a means of establishing significance of inter-treatment differences. A drawback of the present study is that it addresses available categorical dependent variables separately instead of incorporating them in more advanced multifactor models. As highlighted by Chaudhary et al. (2010), methodologies for conducting multi-factor mixed-effects meta-analyses remain largely underdeveloped and our study could not overcome that problem.

Ecology of AM effect on *R*/*S* ratio

The present study detected an overall negative effect of AM on allometric partitioning of plants towards root growth when there had been no consideration of any regulators. Following consideration of propagation mode and stress type, non-stressed plants grown from seeds when inoculated with AM fungi demonstrated a more pronounced ability to promote plant shoot growth (Fig. 1b). AM colonization of clonally propagated material appeared to have an apparent stimulating effect on root growth. We remain skeptical of this result due to the low number of available trials. However, we attribute this effect to the growth stimulation resulting from AM inoculation. It is well known that the way clonally propagated plant cuttings are buried in soil results in a high initial R/S ratio. Following propagation, bigger plants may be more flexible in recovering their optimal R/S ratio (Farrar and Gunn 1998). In this respect, the short duration of most controlled experiments could render the plant growth difference resulting from AM inoculation an important factor in shaping the plant's *R*/*S* ratio and reflect the ability of AM-inoculated plants to produce more biomass. Moreover, we were able to demonstrate the absence of a significant effect of AM status on allometric partitioning of biomass in stressed plants. Reduced plant fitness of host plants exposed to stresses may limit their ability to modify their allometry and, consequently, they are less responsive to factors that may mediate allometric modifications such as AM colonization.

An important point in our meta-analysis was the test for significance of positive vs negative plant growth outcome of AM inoculation. As stated earlier, this analysis could be informative on whether AM mediation of *R*/*S* ratio could actually reflect the modified plant size. Significance of the variable "AM impact" was detected (Fig. 2c). However, even in the absence of positive AM size outcome, there was a trend for plant species to increase biomass allocation to their shoots (mean $ES_{R/S}$ remained negative). This result favors the view that modified biomass allocation to aboveground growth, following AM colonization, is a result of improved plant nutrition. However, targeted experimentation is needed to verify the presence of the specific mechanism. The significance of the effect of competition could be attributed to the expected increased plant biomass allocation belowground in the presence of competitors (Murphy and Dudley 2007). Alternatively, it could be an indirect result of the decline in available plant growth space that plants in competition face in controlled experiments. Experimentation procedures do not always correct for the plant growth space per plant and competition experiments are commonly carried out in microcosms of standard sizes that do not differ from those for plants grown in isolation.

Both the identity of plant and AM symbionts appeared to have a significant effect on the $ES_{R/S}$ with plant-related differences mainly observed between herbaceous and woody plants. A possible explanation is that woody plants tend to allocate aboveground and belowground biomass more asymmetrically, in favor of aboveground growth, and could potentially reflect the ability of AM fungi to further promote plant growth. However, when analyzed separately for the effect of duration of the experiments, woody and herbaceous plants appeared to respond to AM inoculation differently. "Young" herbaceous plants and "old" woody plants were least responsive to AM inoculation (less negative $ES_{R/S}$ values were reported). Interestingly, these two categories of plants (i.e., "young" herbaceous and "old" woody) are those that are expected to have the highest *R*/*S* ratio amongst herbaceous and woody plants, respectively (Poorter and Nagel 2000). The relationship appears to support the idea that is presented in Fig. A1 in "Appendix" that the *R*/*S* ratio of the plants in the absence of AM colonization exerts a negative influence on $ES_{R/S}$. Another striking result in Fig. 4a is the different responses of herbaceous legumes and forbs to AM inoculation. Both functional groups of plants, though ubiquitous in nature, are considered characteristic of low fertility biomes (e.g., Stevens et al. 2009). However, their adaptation strategy is different. Many forbs develop a deep rooting system that allows access to a more extensive soil volume. By contrast, most legumes take advantage of a symbiotic pathway to meet their nitrogen demands. The differences in their allometric responses to AM fungi could be attributed to (a) a possible higher dependency of legumes on symbiotically obtained phosphorus (P) due to high P requirements of nitrogen fixation (Puppi et al. 1994) or (b) a tendency of forbs to further invest in belowground growth which represents their competitive advantage.

Single AM isolates appeared to be less effective in promoting aboveground biomass allocation than AM mixtures as well as *Glomeraceae* isolates when compared with *Gigasporaceae*. Both results could be adequately justified on the basis of physiological differences amongst AM isolates. With regards to multifunctionality of AM symbionts, Fitter (2005) suggested a model according to which the AM community possessed the potential to perform multiple functions, but these functions are mutually incompatible for single AM isolates. AM ability to acquire P is known to differ amongst isolates (e.g., Jakobsen et al. 1992) and a similar result is expected for other functions such as protection from pathogens. Evidence in favor of such a divergence of AM fungal functional characters accumulates (e.g., Powell et al. 2009; Sikes et al. 2010). Possibly, the coexistence of more than one AM fungal isolate in the roots increased the chances of incorporating an AM fungus capable of alleviating the plant growth nutrient limitation which could be the mechanism through which AM fungi impact the *R*/*S* ratio of plants. Alternatively, the co-occurrence of more than one AM isolate could result in a complementary effect in plant nutrition (van der Heijden et al. 1998). The lower $ES_{R/S}$ recorded for *Gigasporaceae* isolates could be explained on the basis of physiological differences (Hart and Reader 2002; Klironomos and Hart 2002). *Gigasporaceae* are known to invest more extensively to extraradical mycelia growth (Hart and Reader 2002), and this could have resulted in improved nutrient acquisition and lower *R*/*S* ratios.

Applications

The ubiquity of AM fungi in nature has often raised skepticism on the necessity of experimentation using non-inoculated control plants (e.g., Augé 2001). However, terrestrial plant communities, especially when managed intensively, are AM propagule-limited (Lekberg and Koide 2005). Moreover, the commercialization of AM fungi in agriculture and plant nurseries and their incorporation in phytostabilization projects necessitate our ability to accurately forecast the outcome, which can only be achieved following a detailed understanding of the physiology of both symbionts alone and in association. The positive impact of AM fungi on *R*/*S* ratios is not always desirable. In nurseries, it is commonly perceived that low *R*/*S* ratio is one of the primary reasons associated with transplanting shock risk and low ability to withstand water stress following transplanting (Bernier et al. 1995). Increased belowground allocation following AM inoculation might thus represent an additional argument for their commercial use. Similarly, in phytoremediation, a higher *R*/*S* ratio could enhance phytostabilization but concurrently, unless accompanied with more intense heavy metal translocation to shoots, complicate phytoextraction (e.g., Göhre and Paszkowski 2006). In the present study, the effect of AM fungi on *R*/*S* ratio of metal-stressed plants was not significant (Fig. 1c) and further research may be needed to address this specific point. By contrast, in agriculture, for most crops, an increased yield may necessitate increased biomass allocation aboveground and the mediation of AM fungi could thus be desirable. Crops where an increased biomass allocation aboveground may be desirable include not only those where the whole aboveground biomass is harvested such as bamboos, some vegetables (e.g., lettuce, spinach), tobacco, and biofuel crops but also crops where the standing biomass could have alternative application such as fodder.

Conclusions

In summary, AM colonization of host plants led to increased biomass allocation to the shoots and this occurred irrespective of the dry weight biomass outcome of the AM symbiosis. Upon further examination, however, we found evidence that responses had not been uniform throughout the dataset since clonally propagated plants reacted to AM colonization with increased root growth and stressed plants demonstrated a neutral allometric reaction. AM colonization in non-stressed plants grown from seeds mediated an extensive promotion of aboveground growth that was significant in all cases except when (1) AM colonization had been detrimental for the growth of plant host and when (2) experimentation involved either a forb host or a *G. mosseae* inoculant. In the specific instances, a neutral effect was detected. Moreover, analysis provided evidence in favor of the additional hypothesis since both host plant identity (mainly differences existing between woody and herbaceous plants) and AM inoculant identity (differences arising from the number) were significant factors that regulated AM impact on non-colonized plant species *R*/*S* ratio. More work should target a mechanistic understanding of the AM impact on plant *R*/*S* biomass allocation.

Acknowledgements The project was funded through an award to Stavros Veresoglou from Freie Universität Berlin, Center for International Cooperation. We are grateful to two anonymous referees for their constructive comments.

References

Adams DC, Gurevitch J, Rosenberg MS (1997) Resampling tests for meta-analysis of ecological data. Ecology 78:1277–1283

Augé RM (2001) Water relations, drought and vesicular–arbuscular mycorrhizal symbiosis. Mycorrhiza 11:3–42

Bernier PY, Lamhanedi MS, Simpson DG (1995) Shoot:root ratio is of limited use in evaluating the quality of container conifer stock. Tree Planters' Notes 46:102–106

Borenstein M, Hedges LV, Higgins JPT, Rothstein HR (2009) Introduction to meta-analysis. Wiley, New York

Chaudhary VB, Walters LL, Bever JD, Hoeksema JD, Wilson GWT (2010) Advancing synthetic ecology: a database system to facilitate complex ecological meta-analyses. B Ecol Soc Am 91:235–243

Conover WJ (1980) Practical nonparametric statistics. Wiley, New York

Curtis PS, Wang X (1998) A meta-analysis of elevated CO_2 effects on woody plant mass, form and physiology. Oecologia 113:299–313

Davies FT Jr, Calderon CM, Hueman Z (2005) Influence of arbuscular mycorrhizae indigenous to Peru and a flavonoid on growth, yield, and leaf elemental concentration of "Yangay" potatoes. Hortscience 40:381–385

Farrar JF, Gunn S (1998) Allocation: allometry, acclimation—and alchemy? In: Lambers H, Poorter H, van Vuuren MMI (eds) Inherent variation in plant growth. Physiological mechanisms and ecological consequences. Backhuys, Leiden, pp 183–198

Fitter AH (2005) Darkness visible: reflections on underground ecology. J Ecol 93:231–243

Göhre V, Paszkowski U (2006) Contribution of the arbuscular mycorrhizal symbiosis to heavy metal phytoremediation. Planta 223:1115–1122

Gurevitch J, Hedges LV (1999) Statistical issues in ecological meta-analysis. Ecology 80:1142–1149

Haolin G, Yuhui W, Fengyu W, Bingrui J (2008) Dynamics of root–shoot ratio and environmental effective factors of recovering *Leymus chinensis* steppe vegetation in Inner Mongolia, China. Acta Ecol Sinica 28:4629–4634

Harris RW (1992) Root–shoot ratios. J Agr 18:39–42

Hart MM, Reader RJ (2002) Taxonomic basis for variation in the colonization strategy of arbuscular mycorrhizal fungi. New Phytol 153:335–344

Hermans C, Hammond JP, White PJ, Verbruggen N (2006) How do plants respond to nutrient shortage by biomass allocation? Trends Plant Sc 11:610–617

Hoeksema JD, Forde SE (2008) A meta-analysis of factors affecting local adaptation between interacting species. Am Nat 171:275–290

Hoeksema JD, Chaudhary VB, Gehring CA, Johnson NC, Karst J, Koide RD, Pringle A, Zabinski C, Bever JD, Moore JC, Wilson GWT, Klironomos JN, Umbanhowar J (2010) A meta-analysis of context dependency in plant response to inoculations with mycorrhizal fungi. Ecol Let 13:394–407

Hooker JE, Munro M, Atkinson D (1992) Vesicular–arbuscular mycorrhizal fungi induced alteration in poplar root system morphology. Plant Soil 145:207–214

Jacoby W (2000) Loess: a nonparametric, graphical tool for depicting relationships between variables. Elect Stud 19:577–613

Jakobsen I, Abbot LK, Robson AD (1992) External hyphae of vesicular–arbuscular mycorrhizal fungi associated with *Trifolium subterraneum* L. 1. Spread of hyphae and phosphorus inflow into roots. New Phytol 120:371–380

Klironomos JN, Hart MM (2002) Colonization of roots by arbuscular mycorrhizal fungi using different sources of inoculum. Mycorrhiza 12:181–184

Körner C (1994) Biomass fractionation in plants: a reconsideration of definitions based on plant functions. In: Roy J and Garnier E (eds) A whole plant perspective on carbon–nitrogen interactions. SPB Academic Publishing, The Hague, pp. 173–185

Ledig FT, Perry TO (1966) Physiological genetics of the shoot–root ratio. In Proceedings of the Society of American Foresters, Society of American Foresters, Washington DC, pp. 39–43

Lekberg Y, Koide RT (2005) Is plant performance limited by abundance of arbuscular mycorrhizal fungi? A meta-analysis of studies published between 1988 and 2003. New Phytol 168:189–204

Mamolos AP, Elisseou GK, Veresoglou DS (1995) Depth of root activity of coexisting grassland species in relation to N and P additions, measured using nonradioactive tracers. J Ecol 83:643–652

Monk C (1966) Ecological importance of root/shoot ratios. Bull Torrey Bot Club 93:402–406

Murphy GP, Dudley SA (2007) Above- and below-ground competition cues elicit independent responses. J Ecol 95:261–272

Nuortila C, Kytöviita M-M, Tuomi J (2004) Mycorrhizal symbiosis has contrasting effects on fitness components in *Campanula rotundifolia*. New Phytol 164:543–553

Pérez M, Urcelay C (2009) Differential growth response to arbuscular mycorrhizal fungi and plant density in two wild plants belonging to contrasting functional types. Mycorrhiza 19:517–523

Poorter H, Nagel O (2000) The role of biomass allocation in the growth response of plants to different levels of light, CO_2, nutrients and water: a quantitative view. Aust J Plant Physiol 27:595–607

Powell JR, Parrent JL, Hart MM, Klironomos JN, Rillig MC, Maherali H (2009) Phylogenetic trait conservatism and the evolution of functional trade-offs in arbuscular mycorrhizal fungi. P Roy Soc B Biol Sci 276:4237–4245

Puppi G, Azcón R, Höflich G (1994) Management of positive interactions of arbuscular mycorrhizal fungi with essential groups of soil microorganisms. In: Gianinazzi S, Schüepp H (eds) Impact of arbuscular mycorrhizas on sustainable agriculture and natural ecosystems. Birkhäuser, Basel, pp 201–215

Rabie GH (2005) Influence of arbuscular mycorrhizal fungi and kinetin on the response of mungbean plants to irrigation with seawater. Mycorrhiza 15:225–230

Rasse DP, Rumpel C, Dignac M-F (2005) Is soil carbon mostly root carbon? Mechanisms for a specific stabilization. Plant Soil 269:341–356

Reynolds HL, D'Antonio C (1996) The ecological significance of plasticity in root weight ratio in response to nitrogen: opinion. Plant Soil 185:75–97

Robinson D, Davinson H, Trinder C, Brooker R (2010) Root–shoot growth responses during interspecific competition quantified using allometric modelling. Ann Bot (London) 106:921–926

Roosens N, Verbruggen N, Meerts P, Ximenez-Embun P, Smith JAC (2003) Natural variation in cadmium tolerance and its relationship to metal hyperaccumulation for seven populations of *Thlaspi caerulescens* from western Europe. Plant Cell Environ 26:1657–1672

Rosenberg NJ, Adams DC, Gurevitch J (2000) MetaWin: statistical software for meta-analysis version 2.0. Sinauer, Sunderland

Schroeder MS, Janos DP (2005) Plant growth, phosphorus nutrition, and root morphological responses to arbuscular mycorrhizas, phosphorus fertilization, and intraspecific density. Mycorrhiza 15:203–216

Schüßler A, Schwarzott D, Walker C (2001) A new fungal phylum, the *Glomeromycota*: phylogeny and evolution. Mycol Res 105:1413–1421

Sikes BA, Powell JR, Rillig MC (2010) Deciphering the relative contributions of multiple functions within plant–microbe symbiosis. Ecology 91:1591–1597

Smith SE, Read DJ (2008) Mycorrhizal symbiosis. Elsevier Science, New York, p 605

Stevens CJ, Maskell LC, Smart SM, Caporn SJM, Dise NB, Gowing DJG (2009) Identifying indicators of atmospheric nitrogen deposition impacts in acid grasslands. Biol Conserv 142:2069–2075

Van Den Noortgate W, Onghena P (2005) Parametric and nonparametric bootstrap methods for meta-analysis. Behav Res Method 37:11–22

van der Heijden MGA, Klironomos JN, Ursic M, Moutoglis P, Streitwolf-Engel R, Bolle T, Wiemken A, Sanders IR (1998) Mycorrhizal fungal diversity determines plant biodiversity, ecosystem variability and productivity. Nature 396:69–72

Veresoglou SD, Menexes G (2010) Impact of inoculation with *Azospirillum* spp. on growth properties and seed yield of wheat: a meta-analysis of studies in the ISI Web of Science from 1981 to 2008. Plant Soil 337:469–480

Wang X, Ngigi AR, Smith DL, McPeek TM (2008) Biomass responses to interspecific competition differ between wild species and CO_2. J Plant Ecol 1:25–32

Wilson SD, Tilman D (1995) Competitive responses of 8 old-field plant-species in 4 environments. Ecology 76:1169–1180

Yao Q, Wang LR, Zhu HH, Chen JZ (2009) Effects of arbuscular mycorrhizal fungal inoculation on root system architecture of trifoliate orange (*Poncirus trifoliata* L. Raf.) seedlings. Sci Hortic Amst 121:458–461

Zandavalli RB, Dillenburg LR, de Souza PVD (2004) Growth responses of *Araucaria angustifolia* (*Araucariaceae*) to inoculation with the mycorrhizal fungus *Glomus clarum*. Appl Soil Ecol 25:245–255