FULL PAPER

Plant cheaters preferentially target arbuscular mycorrhizal fungi that are highly connected to mutualistic plants

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Abstract

- To address how arbuscular mycorrhizal networks sustain cheaters – mycoheterotrophic plants that obtain both carbon and soil nutrients from fungi – here we investigate how mutualistic and antagonistic mycorrhizal networks are interlinked.
- We sampled root tips of mutualistic and cheater plants in two tropical forest plots and assembled the combined network between fungi linked to mutualistic and cheater plants (i.e., tripartite network) using DNA sequencing. We compared the interactions of the fungi in the mutualistic and antagonistic networks and searched for motifs (simple subgraphs that are overrepresented in complex networks) representing simultaneous links of particular mutualist and cheater plants to pairs of fungi to investigate fungal association preferences.
- Within the tripartite network, cheater plants are highly connected and interact with the majority of available fungal partners (74%), and we find that fungi that share interactions with the same mutualistic plants also share interactions with sets of cheaters. The motif analysis demonstrates that cheaters preferentially interact with sets of fungi that are linked to particular plant species.
- Our findings indicate that cheaters preferentially interact with fungi that are well-connected to particular mutualistic plants. We hypothesize that these non-random interactions may result from trait-based selection and that this strategy maximizes carbon availability for cheaters.

Introduction

Since the early history of life, interspecific mutualisms have been paramount in the functioning of ecosystems (Thompson, 2005; Bascompte & Jordano, 2013). Mutualisms can form complex networks of interdependence between dozens or even hundreds of species. A prime example of this 'web of life' is the 450-million-year-old mutualism between the majority of land plants and arbuscular mycorrhizal (AM) fungi (Smith & Read, 2008). In this interaction plants supply root-associated Glomeromycotina fungi with carbohydrates, essential for fungal survival and growth. In return, the fungi provide their host plants with mineral nutrients and water from the soil (Smith & Read, 2008). One of the key characteristics of the arbuscular mycorrhizal interaction is its low specificity. A mycorrhizal plant typically associates simultaneously with multiple fungi and a mycorrhizal fungus often associates simultaneously with multiple plants. This creates complex underground networks in which plants of different species are linked by shared arbuscular mycorrhizal fungi (Toju *et al.*, 2015; Chen *et al.*, 2017).

Experiments demonstrate that the arbuscular mycorrhizal mutualism is evolutionarily stable because control is bidirectional, and partners offering the best rate of exchange are rewarded (Bever *et al.*, 2009; Kiers *et al.*, 2011). These results suggest that arbuscular mycorrhizal networks consist of 'fair trade' interactions, which are akin to trade in a market economy (Kiers *et al.*, 2011; Wyatt *et al.*, 2014). However, the universal importance of these market dynamics in arbuscular mycorrhizal interactions is challenged by the occurrence of cheaters (Selosse & Rousset, 2011). Mycoheterotrophic plants receive not only soil nutrients but also carbon from arbuscular mycorrhizal fungi (Leake, 1994; Merckx, 2013). Hence, mycoheterotrophic plants exploit the arbuscular mycorrhizal symbiosis for nutrients and carbon without being sanctioned by the fungal partner (Walder & Van Der Heijden, 2015). Therefore, it has been suggested that mycoheterotrophic plants may display a truly biotrophic parasitic mode, digesting the fungus colonizing their roots (Imhof *et al.*, 2013). The mechanism underpinning carbon transfer from arbuscular mycorrhizal fungi to mycoheterotrophic plants remains unclear, but it is unlikely that resource exchange in these mycoheterotrophic associations relies on reciprocal dynamics.

68 Phylogenetic studies demonstrate that these cheaters evolved from mutualistic ancestors
69 (Merckx *et al.*, 2013). Within obligate mutualisms, the critical barrier to mutualism breakdown
70 and to the evolutionary stability of the resulting cheater species is thought to be a
71 requirement for three-species coexistence: a cheater species relies on a mutualistic partner
72 which simultaneously interacts with a mutualistic species (Pellmyr & Leebens-Mack, 1999). In
73 species-rich mutualisms such as the arbuscular mycorrhizal symbiosis, where multi-species
74 coexistence is the rule, a high potential for the occurrence of these tripartite linkages
75 (cheater–fungus–mutualistic plant) is expected (Merckx & Bidartondo, 2008). Indeed, while
76 evolution of cheating in specialized obligate mutualisms is relatively rare (Sachs & Simms,
77 2006), cheating in the arbuscular mycorrhizal symbiosis evolved in more than a dozen of plant
78 clades, including over 250 species which together cover nearly all tropical and subtropical
79 forests (Gomes *et al.*, 2019a).
80
81 Previous work shows that cheaters target a subset of the mycorrhizal fungi available in the
82 local community (e.g Bidartondo *et al.* (2002); Gomes *et al.* (2017a); Sheldrake *et al.* (2017)).
83 Their associated fungal communities can vary in specificity (Merckx *et al.*, 2012), and in
84 communities of co-occurring cheaters their fungal diversity appears to increase
85 proportionally to their fungal overlap (Gomes *et al.*, 2017b). However, which arbuscular
86 mycorrhizal fungi cheaters select from the available community is not well understood. From
87 an evolutionary perspective there are two mechanisms that may be involved: (1) cheaters,
88 because of their increasingly parasitic interaction with fungi, have been 'denied' access to
89 some members of the fungal community except for fungal lineages that fail to detect or
90 exclude the cheaters, and (2) cheaters have selected from the potential fungal community
91 the best target to meet their nutrient demands (Bruns *et al.*, 2002; Egger & Hibbett, 2004;
92 Bidartondo, 2005). We address the potential importance of the latter mechanism and
93 hypothesise that mycoheterotrophic plants preferentially associate with 'keystone' (Mills &
94 Doak, 1993) fungi that are connected to many different mutualistic plants, since these fungi
95 are potentially more resilient to perturbance (Bascompte & Jordano, 2007) and may be the
96 most reliable source of carbon (Waterman *et al.*, 2013). In addition, since fungal traits play an
97 important role in the arbuscular mycorrhizal symbiosis – phylogenetically related AM fungi
98 (assumed to have similar functional traits), preferentially interact with similar plant species
99 (Chagnon *et al.*, 2015) – we hypothesize that if partner selection in tripartite networks is trait-

100 driven we will be able to detect an influence of the phylogenetic relationships of the fungi.
101 Here we test these hypotheses on a combined tripartite mycorrhizal network of co-occurring
102 mycoheterotrophic and surrounding mutualistic plants linked by shared arbuscular
103 mycorrhizal fungi compiled by high-throughput DNA sequencing.
104
105 **Materials and methods**
106
107 *Sampling*
108
109 Since mycoheterotrophic plants are relatively rare and often have patchy distributions
110 (Gomes *et al.*, 2019b), we sampled two proximate 4 x 4 m plots (plot 1 and plot 2) in a coastal
111 rainforest in French Guiana (5°28'25"N 53°34'51"W) on 28 July 2014, with overlapping
112 mycoheterotrophs as replicates. In both plots, root tips of mycoheterotrophic plants
113 (cheaters) and surrounding autotrophic plants (mutualists) were sampled, cleaned with
114 water, and stored on CTAB buffer at -20°C until further processing. Plot 1 contained the
115 cheater species *Dictyostega orobanchoides* (3 specimens), *Gymnosiphon breviflorus* (2
116 specimens) *Voyria aphylla* (3 specimens), *Voyriella parviflora* (7 specimens), and *Soridium*
117 *spruceanum* (6 specimens). Plot 2 contained *D. orobanchoides* (2 specimens), *G. breviflorus*
118 (5 specimen), *V. parviflora* (15 specimens), and *S. spruceanum* (2 specimens). Around each
119 cheater, 10 root tips of mutualistic plants where collected. In addition, we sampled 5
120 additional mutualistic plant root tips randomly from each quadrant of each plot aiming to
121 represent the belowground mutualistic community. Root tips of mutualistic plant species
122 were identified by sequencing the genetic markers *matK or trnL* as described in (Gomes *et al.*,
123 2017a). Fungi associated with the roots of mutualist and cheater plants were sequenced as
124 described in Gomes *et al.* (2017b) in two separate runs, following the same bioinformatics
125 methods to process the raw reads from the two runs combined until clustering into 97%
126 Operational Taxonomic Units (OTUs). The OTUs that were represented by less than five reads
127 in each sample were excluded to avoid spurious OTUs (Lindahl *et al.*, 2013). The taxonomical
128 assignment of OTUs was done by querying against the UNITE database (Kõljalg *et al.*, 2013).
129 Because the mycoheterotrophic plants of this study are associated with fungi that belong to
130 the sub-phylum Glomeromycotina (Merckx *et al.*, 2012), we only retained fungal OTUs from
131 this sub-phylum in the subsequent analysis. In this study, we sequenced the fungal

132 communities of mutualistic and cheater plants, which are partially and totally representative
133 of the entire root systems, respectively, and may vary in the degree of specificity towards
134 their fungal interactions (Yamato *et al.*, 2014; Mennes *et al.*, 2015; Gomes *et al.*, 2017a,b).
135 Thus, because rarifying the OTU matrix has been shown to greatly increase the false positive
136 rate of OTUs per sample (McMurdie & Holmes, 2014), we decided not to rarify the data, and
137 only used incidence data, since our main interest was to determine which interactions can be
138 established and not how abundant they are. Hereafter we refer to OTUs as 'fungi'. The
139 simultaneous presence of fungi in the roots of both cheater and mutualistic plants does not
140 necessarily reflect that these fungi are functionally active but is indicative of a potential link
141 between plants.
142
143 *Tripartite network compilation*
144
145 We built three tripartite networks: (1) the tripartite network of plot 1 with 21 mutualists, 5
146 cheaters, and 108 fungi; (2) the tripartite network of plot 2 with 16 mutualists, 4 cheaters,
147 and 99 fungi; and (3) the combined tripartite network of plot 1 and plot 2 with 33 plants (28
148 mutualists and 5 cheaters) and 121 fungi, of which 89 (74%) are shared between mutualists
149 and cheaters. The tripartite network and the corresponding plant-plant interaction strength
150 network including mutualists and cheater plants were visualized with the *igraph* R package
151 (Csardi & Nepusz, 2006). Plot 1 and plot 2 overlapped in 9 mutualists (32%), 4 cheaters (80%),
152 and 86 fungi (71%). In each network, a plant was linked to a fungus if that fungus was found
153 in the roots of that plant. To examine the congruence between the mycorrhizal interactions
154 of plot 1 and plot 2, we calculated the normalized interaction degree of plants and fungi of
155 both tripartite networks using the *bipartite* R package (Dormann *et al.*, 2018). We measured
156 the correlation between the normalized interaction degree of plants and fungi which occur in
157 both plots using the Pearson correlation coefficient.
158
159 *Phylogenetic signal*
160
161 For the combined tripartite network, we infer the phylogenetic relationships between the
162 fungi following the strategy of Gomes *et al.* (2017b). Phylogenetic relationships of the plants
163 were derived from the Angiosperm Phylogeny Website (Stevens, 2012). To test the influence

of the evolutionary relationships of the interacting partners in both mutualists and cheater mycorrhizal networks, the phylogenetic signal on each bipartite network was evaluated using an estimated generalized least squares (EGLS) analysis that fits the phylogenetic variance–covariance matrix to the plant-fungi association matrix (Ives & Godfray, 2006). Using this EGLS method, we calculated the independent phylogenetic signals of the plant (d_{plant}) and fungi (d_{fungi}) phylogenies on the binary association matrix and assessed the overall strength of the signal of both phylogenies combined using mean-squared error (MSE) values.

Comparing mutualistic and antagonistic networks

To understand how fungi are preferentially linked to the cheater and mutualistic plants, first we compared the fungal strength association of the individual mutualistic and antagonistic bipartite interactions of the combined tripartite network. The fungal strength association of a pair of fungi represents similarity in their interactions through shared plants. This measure is a proxy for the ecological similarity between fungi. We selected the fungal OTUs shared between both interaction types in the subsequent analysis, and calculated dissimilarity matrices between the fungi linked to mutualistic and cheater plants separately, using two different measures. The first measure was the Jaccard index, and the second was the Overlap measure (Saavedra *et al.*, 2013), which corresponds to the number of shared plants relative to the maximum number of plants that can be shared. We computed the Mantel test correlation between the distance matrices for the mutualistic and antagonistic interactions using the two different measures. Because a single fungal species is often not represented exclusively by an individual OTU, and multiple OTUs can in fact represent a single species (Flynn *et al.*, 2015), we accounted for the phylogenetic relatedness among the fungi: we computed the partial Mantel test correlation between the distance matrices calculated using the Jaccard and Overlap measures, controlling for the phylogenetic relatedness of the shared fungi. The phylogenetic distances among the shared fungi were calculated by pruning the fungal phylogenetic tree to the fungi shared by the mutualistic and antagonistic interaction networks and summing up the total branch lengths using the *ape* R package (Paradis & Schliep, 2019).

195 Then, we searched for simple motifs in the combined tripartite network (reduced to only
196 include the fungal OTUs shared between both interaction types). Motifs are subgraphs of
197 interconnections that appear significantly more often than expected by chance in complex
198 networks (Milo *et al.*, 2002; Bascompte & Melián, 2005; Stouffer *et al.*, 2007). We aimed to
199 test whether the plants in the mutualistic and antagonistic networks had simultaneous
200 preferential links to specific fungi, or each interaction type was disconnected in terms of their
201 fungal association preferences. Therefore, we selected motifs that translate the simultaneous
202 link of a mutualist and antagonist plants to pairs of fungi (Fig. 1a). We generated null models
203 to evaluate the fidelity of plants in associating with specific fungi (consistency in partner
204 choice). Both datasets have imbalanced number of plant species (5 cheaters and 28
205 mutualistic plants), and because the nature of the plant-fungus interactions is unknown for
206 each plant type, null models were generated separately for cheater and mutualistic plants.
207 We generated randomly resampled null matrices using the *shuffle.web* method in *bipartite*
208 (Dormann *et al.*, 2009), where each fungal OTU and plant species keeps exactly the same
209 number of interactions as in the observed network, resampling 1000 times. Mutualistic and
210 antagonistic random matrices were combined each time before counting the number of
211 motifs. We compared the empirical motif result to the null model's 95% confidence interval.
212 An empiric result above the null confidence interval indicates that pairs of fungi are shared
213 between particular cheater and mutualistic plants more often than expected by chance,
214 reflecting a simultaneous preferential association to particular fungi between species in the
215 antagonist and mutualist network. An empirical result below the null confidence interval
216 indicates that pairs of fungi are shared between particular cheater and mutualistic plants less
217 often than expected by chance, reflecting separate dynamics and disconnection between
218 antagonist and mutualist networks.
219
220 **Results**
221
222 *Plant – fungus interactions*
223
224 In total, we successfully sequenced 145 root tips samples of 28 mutualistic and 5 antagonistic
225 plant species (see Table S1 for detailed species list). From all the root tips analyzed, the Ion
226 Torrent sequencing runs produced 10,453,711 quality filtered reads, of which 368,005 were

227 classified as Glomeromycotina and represented by 121 OTUs (Fig. 1b). Mutualist plants were
228 associated with 110 fungi (51% of the reads) and cheaters were associated with 101 fungi
229 (49% of reads). The plant-plant interaction strength network is shown in Fig. 1c. Plants belong
230 to 21 different families (20 angiosperms, 1 fern), arbuscular mycorrhizal fungi are part of
231 Gigasporaceae (4 fungi), Acaulosporaceae (14 fungi), and Glomeraceae (103 fungi). We find a
232 strong significant linear correlation between the interaction degree of overlapping fungi of
233 plots 1 and 2 (R^2 = 0.81, p < 0.001; Fig 2), but not for the overlapping plants (R^2 = 0.01, p =
234 0.318; Fig 2). Due to the similarities in fungal interaction strength, the subsequent analyses
235 were performed on the combined tripartite network.
236
237 *Phylogenetic signal*
238
239 We measured a low significant phylogenetic signal of the mutualistic plant phylogeny (d_{plant} =
240 0.14; 95% CI: 0.11 – 0.19) and a strong significant signal of the fungal phylogeny (d_{fungi} = 0.34;
241 95% CI: 0.29 – 0.40; Fig 3) on the mutualistic interactions of the combined tripartite network.
242 The overall strength of the phylogenetic signal for the linear model fitted to the actual data
243 (MSE_d = 0.18) was closer to that found under the assumption of no phylogenetic covariances
244 (MSE_{star} = 0.18) than for the assumption of maximum phylogenetic signal (MSE_b = 0.28). We
245 measured a low but significant phylogenetic signal of the cheater phylogeny on the
246 antagonistic network (d_{plant} = 1.05 x 10^{-9}; 95% CI: 3.85 x 10^{-14} – 5.96 x 10^{-4}) and a strong
247 significant signal of the fungal phylogeny on the interactions (d_{fungi} = 0.59; 95% CI: 0.32– 0.85).
248 The overall strength of the phylogenetic signal for the linear model fitted to the actual data
249 (MSE_d = 0.25) was closer to that found under the assumption of no phylogenetic covariances
250 (MSE_{star} = 0.25) than for the assumption of maximum phylogenetic signal (MSE_b = 0.67).
251
252 *Comparison between mutualistic and antagonistic plant networks*
253
254 The mutualists and cheaters shared 89 fungal OTUs. We found a similar fungal association
255 strength between both interaction types (Mantel tests: Jaccard r = 0.31, p = 0.001; Overlap r
256 = 0.28, p = 0.001), and also when accounting for the phylogenetic relatedness among the
257 shared fungi (partial Mantel tests: Jaccard r = 0.29, p = 0.001; Overlap r = 0.24, p = 0.001). The
258 tripartite network had 23,261 motifs (*z-score* = 2.675, p = 0.005), suggesting that particular

259 pairs of fungi are being preferentially targeted within the antagonist and mutualistic networks
260 simultaneously.
261
262 **Discussion**
263
264 The tripartite networks from the two sampled plots share the majority of their fungi and
265 interestingly, these overlapping fungi have a similar interaction degree in both plots (Fig. 2a).
266 The ability of fungi to interact with many plant species simultaneously therefore seems a
267 taxon-specific characteristic. In contrast, the shared mutualistic plant species differ in fungal
268 richness in each plot (Fig. 2b), indicating that for mutualistic plants the level of specificity may
269 be context dependent, and preferential association with particular arbuscular mycorrhizal
270 fungi may be independent from their identity or position in the interaction network.
271 Alternatively, our sampling method may have failed to identify all fungi linked to each plant.
272
273 In our study, the cheater plant community is highly connected to arbuscular mycorrhizal fungi,
274 interacting with 74% of the available fungi. This is congruent with previous studies which
275 found communities of co-occurring cheaters of the same families from tropical America to
276 associate with a diverse array of fungi (Gomes *et al.*, 2017b; Sheldrake *et al.*, 2017). Within
277 the combined network, the four most generalist fungi of the mutualistic interaction network,
278 representing 13% of all mutualistic interactions, were also among the highest-ranking
279 generalists in the antagonistic network (Fig. 2). In fact, these Glomeraceae fungi were found
280 to associate with all cheater species (Fig. 3). Glomeraceae are usually not only the most
281 dominant clade in natural arbuscular mycorrhizal communities, often accounting for c. 70%
282 of all species (Montesinos-Navarro *et al.*, 2012), but they also have consistently been found
283 to include the most generalist arbuscular mycorrhizal fungi in other network studies (e.g.
284 Montesinos-Navarro *et al.,* 2012; Chagnon *et al.*, 2015; Chen *et al.*, 2017). The ability to
285 interact with many mutualistic plant species can be a potential reason for why cheaters
286 generally target Glomeraceae fungi (Merckx *et al.*, 2012; Renny *et al.*, 2017). Ecological theory
287 predicts that generalist species tend to have large distribution ranges (Brown, 1984) and,
288 consequently, are less vulnerable to (local) extinction than specialized species (Schleuning *et*
289 *al.*, 2016). Therefore, associations to generalist fungi may be advantageous for the
290 evolutionary persistence of cheaters. In addition, in the context of mycorrhizal fungi, which

291 can be linked to different plant species simultaneously, generalist fungi are likely to be more
292 reliable carbon sources for cheaters. Associations with multiple plant partners may increase
293 fungal resilience to disturbance, and mediate temporal fluctuations in carbon flow and
294 interaction dynamics (Bennett *et al.*, 2013) and thus guarantee continuous carbon supply to
295 the cheaters, without pronounced negative effects on particular fungal hosts. An alternative
296 and perhaps not mutually exclusive explanation for why cheaters preferentially target well-
297 connected fungi may be that these fungi are less effective in detecting and excluding non-
298 mutualistic plant partners (van der Heijden & Walder, 2016).
299
300 Within Glomeraceae, the most generalist fungi are part of multiple distantly related clades,
301 belonging to different species in at least two distinct genera (*Rhizophagus* and *Glomus*; Fig.
302 3), potentially representing distinctive functional traits (Chagnon *et al.*, 2015). The tendency
303 of plants to simultaneously associate with distantly-related fungi may reflect functional trait
304 complementarity among co-occurring species (Maherali & Klironomos, 2007). Overall, we
305 measured a significant influence of the fungal phylogenetic relationships on both the
306 mutualistic and antagonistic interactions, showing that closely related fungi interact with
307 similar mutualistic and cheater plants respectively. We detected a lower but significant
308 phylogenetic signal of the mutualistic plants on the mutualistic network, while the
309 phylogenetic signal of the cheater plants of the antagonistic interactions was near zero. The
310 latter likely results from the fact that all cheater plants mainly target similar Glomeraceae
311 fungi. These results reinforce the hypothesis that partner selection in the arbuscular
312 mycorrhizal symbiosis is driven by fungal traits (Chagnon *et al.*, 2015), also for antagonistic
313 interactions.
314
315 The positive and statistically significant correlation between the ecological distances of the
316 fungi in the antagonistic and mutualistic networks strongly suggests that fungi that are
317 targeted by the same cheaters tend to interact with a similar set of mutualistic plants species.
318 This result is not the consequence of the phylogenetic signal of the fungi on the interactions
319 reported above as shown by the partial mantel test. Similarly, the motif analysis demonstrates
320 that pairs of fungi that share an interaction with a cheater species have a high chance to also
321 share a link with a particular mutualistic plant. These results indicate that antagonistic and
322 mutualistic mycorrhizal networks are not linked randomly but are symmetrically influenced

323 by mutualistic and cheater plant identity. Whether this is the consequence of spatial patterns
324 in the distribution of mutualists, cheaters, and fungi remains to be determined. Alternatively,
325 the detected pattern may reflect species-specific association preferences. The plant
326 interaction strength network indicates that each cheater species is linked to several
327 mutualistic plants through shared fungi, and each cheater species tends to be closely
328 connected to a different mutualistic plant (Fig. 1c). Earlier work suggested that antagonistic
329 mycorrhizal interactions can be better explained by understanding cheater–cheater
330 interactions generated by sharing fungal associations (Gomes *et al.*, 2017b), and indicate that
331 antagonistic interactions may respond to an ecological mechanism driven by maximizing co-
332 occurrence and avoiding competitive exclusion among cheaters. The results obtained here
333 expand this view and suggest that not only cheater-cheater interactions drive patterns of co-
334 occurrence of cheater plants, but also the indirect interactions between mutualistic and
335 cheater plants.
336
337 **Conclusions**
338 This is the first study assessing how plant cheaters are embedded in mutualistic mycorrhizal
339 networks. We find that arbuscular mycorrhizal cheater plants simultaneously interact with a
340 wide range of fungal partners, but generally prefer those that are well-connected to
341 mutualistic plants. The phylogenetic relationships between the fungi, likely a proxy for fungal
342 traits, have a significant influence on these non-random tripartite interactions. We also detect
343 that the antagonistic and mutualistic and mutualistic networks are not linked randomly.
344 Instead, sets of cheaters plants target fungi that are simultaneously linked to similar
345 mutualistic plants. Therefore, we conclude that the persistence of cheaters in arbuscular
346 mycorrhizal networks is dependent on well-connected 'keystone' mycorrhizal fungi. The
347 finding that well-connect nodes in arbuscular mycorrhizal networks are more prone to
348 cheating remains to be tested in other diffuse mutualisms.
349
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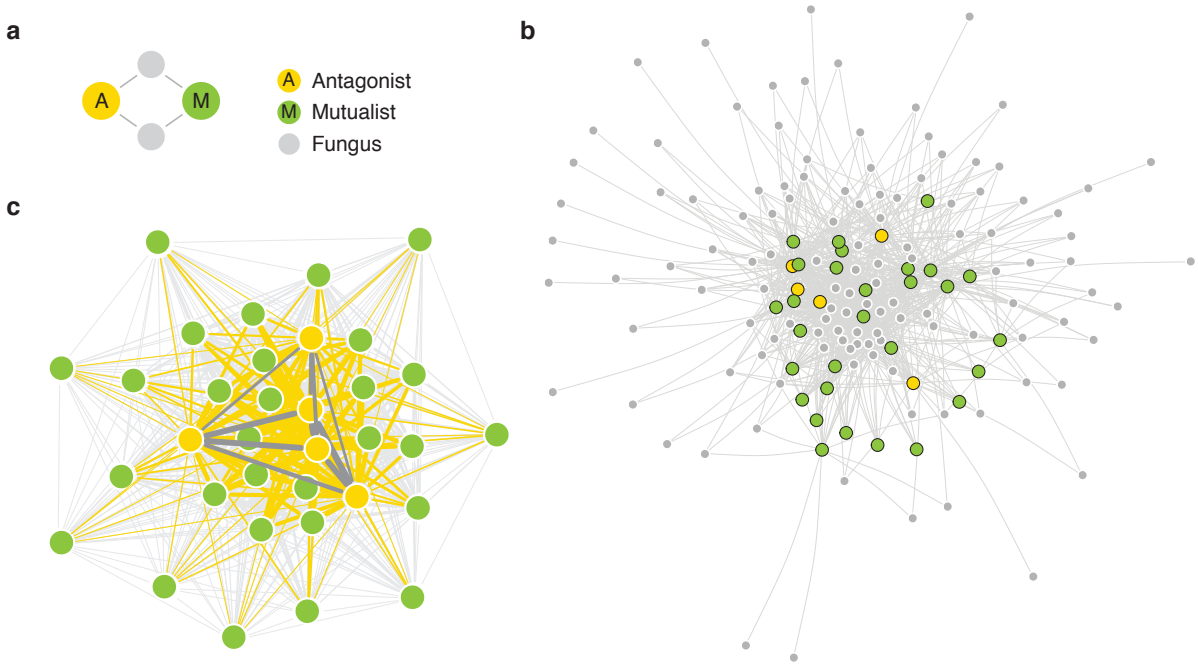
487 **Figures**



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490 **Figure 1**. Tripartite arbuscular mycorrhizal interactions. (a) Motif depicts pairs of fungi
491 simultaneously linked to a cheater and a mutualistic plant. This motif appears significantly
492 more often than expected by chance in the empirical network; (b) Visualization of the
493 combined tripartite network between cheaters (yellow), arbuscular mycorrhizal fungi (grey),
494 and mutualistic plants (green), where lines represent a connection between a plant and a
495 fungus; (c) The plant-plant interaction strength network, in which lines represent a link
496 through a shared arbuscular mycorrhizal fungus. The thickness of the lines represents the
497 interaction strength between the plants (the thicker, the more fungi are shared). Yellow lines
498 link cheater to mutualistic plants; light grey lines link mutualistic to mutualistic plants; and
499 dark grey lines link cheater to cheater plants. The closer plants are together in the graph, the
500 higher their interactions strength.

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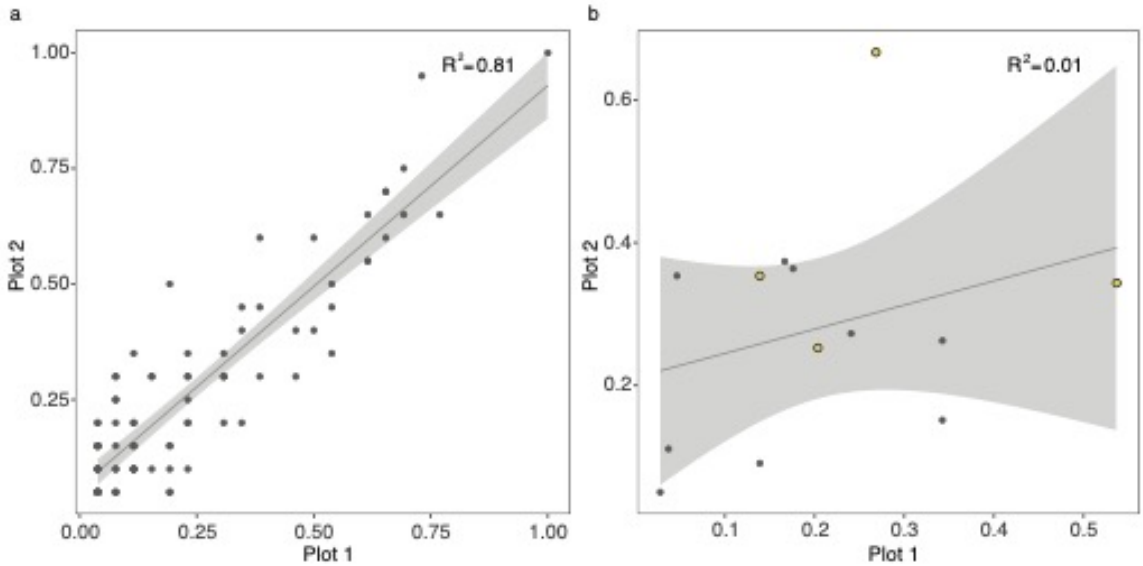

Figure 2. Comparison of interaction degree of taxa shared between plot 1 and plot 2. (a) Shared arbuscular mycorrhizal fungi (n = 86) show a strong significant linear correlation in their interaction degree of plot 1 and plot 2; (b) Shared plants do not have similar interaction degrees in the plots. Grey dots represent the 9 shared mutualistic plants and yellow dots represent the 4 shared cheater plants.

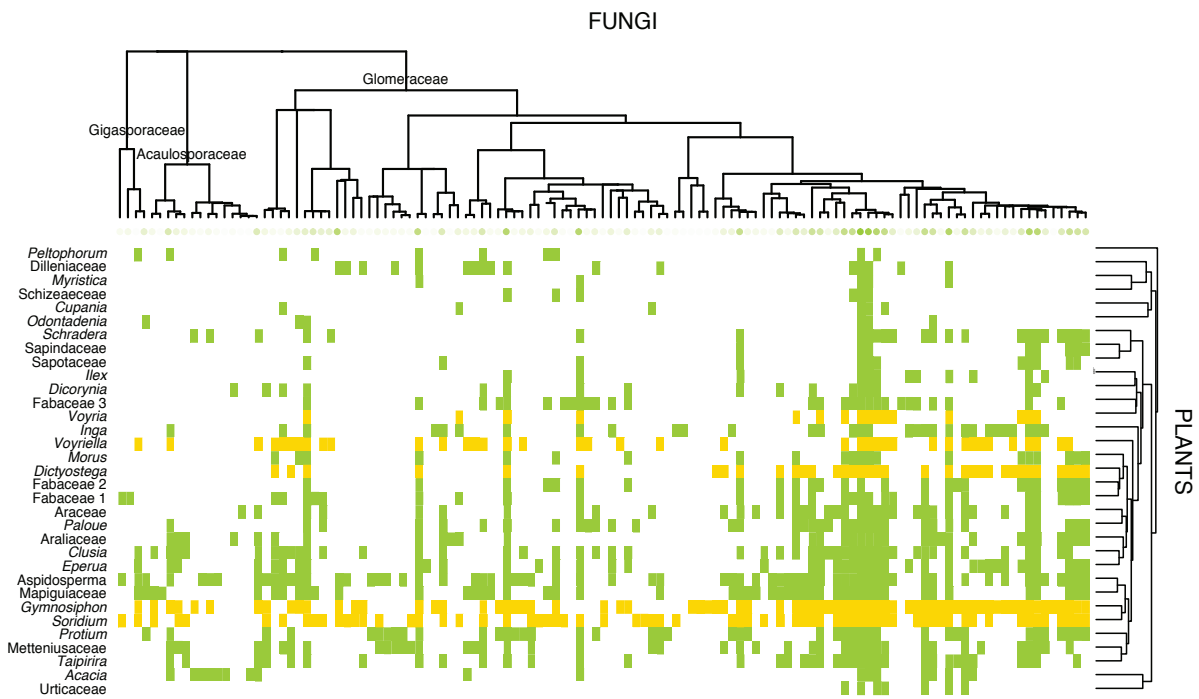

Figure 3: Plant-fungus interactions of mutualistic (green rectangles) and cheater (yellow rectangles) plants. Phylogenetic relationships between the fungi are shown on top. The intensity of the green dots on the tips of the fungal phylogeny represent the interaction degree of each fungus in the total network. Plant species are listed on the left, hierarchy clustered dendrogram based on the Bray-Curtis distance of their fungal communities is shown on the right.