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Host Specificity in Ectomycorrhizal Communities: What Do the Exceptions Tell Us?[1]

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SYNOPSIS. Classic ectomycorrhizal symbioses are mutualisms that involve the exchange of fixed carbon for mineral nutrients between plant roots and fungi. They are unique in the way they contain features of both intimate and diffuse symbioses. The degree of host specificity varies, particularly among the fungi. Here we examine two exceptional cases of specificity to see what they tell us about the advantages of specificity, how it is initiated, and the potential role that it plays in complex ecosystems. The first case involves non-photosynthetic epiparasitic plants, which contrary to virtually all other plants, exhibit high levels of specificity toward their fungal hosts. The second case involves suilloid fungi; this is the largest monophyletic group of ectomycorrhizal fungi that is essentially restricted to associations with a single plant family. In both cases, new symbioses are initiated by dormant propagules that are stimulated to germinate by chemical cues from the host. This reduces the cost of wasting propagules on non-hosts. The advantages of specificity remain unclear in both cases, but we argue that increased benefit to the specialist may result from specialized physiological adaptations. We reexamine the idea that specialist fungi may help their hosts compete in complex ecosystems by reducing facultative epiparasitism by other plants, and suggest an alternative hypothesis for the observed pattern.

INTRODUCTION

The purpose of this paper is to examine the patterns of host specialization within several ectomycorrhizal symbioses that we have studied, and to ask what these patterns tell us about the forces that select for specialization and what effect specialized associations may have on complex ecosystems. For the purposes of this paper we define a specialist as a species that associates with a phylogenetically narrow range of hosts, and we use the word ''host'' to include plants in the case of specialized fungi, and fungi in the case of specialized plants.

Ectomycorrhizal (**EM**) symbioses involve a mutualism between a plant root and a fungus; the plant provides fixed carbon to the fungus and in return, the fungus provides mineral nutrients, water and protection from pathogens to the plant. For most plants and fungi the interaction appears to be essentially obligate. Pines, for example, can not grow in exotic settings unless EM fungi are introduced (Brisco, 1959), and there is no evidence that the fungi can grow in nature without connection to a plant host. Thus, EM systems appear to be classic mutualisms, and in their simplest form might be conceptualized as shown in Figure 1a. Such a model might be fitting for a potted plant inoculated with a single fungus, but natural ectomycorrhizal systems are much more complex.

EM symbioses are unique in a number of ways that are likely to affect host specificity. Both the fungi and the plants that participate in EM symbioses are polyphyletic with multiple origins of the symbiosis accounting for at least a large part of the pattern (Bruns *et al.*, 1998; Hibbett *et al.*, 1997, 2000). The plants are typically large woody species such as pines, oaks, birches, eucalyptus, and many other temperate, and, to a lesser extent tropical, tree species. Hoeksema (1999) points out that the size and longevity of the plants may make them more predictable targets for specialist EM fungi. The fungi include species from multiple families in the Basidiomycota and Ascomycota and some species of *Endogone* from the Zygomycota. Ectomycorrhizae are an ''intimate symbiosis'' in the sense that the fungus penetrates the root, establishes contact at the cellular level with the plant, and sets up a biotrophic association that lasts weeks or months. Intimate parasitic systems often exhibit high specificity, perhaps because tight physiological coordination is required between two partners (Borowicz and Juliano, 1991), while mutualisms such as the *Rhizobium*/*Legume* system exhibit a range of specificity (Perret *et al.*, 2000) rather similar to what we discuss below for EM fungal specialization. EM symbioses are also ''diffuse symbioses'' in the sense that both plants and fungi have large parts of their thalli that are not associated directly with their partner and are free to simultaneously associate with other unrelated hosts. This arrangement means that individual fungi can be simultaneously associated with several plants and that individual plants can be simultaneously associated with multiple fungi. A more complex model of the interactions between plants and fungi is the result (Fig. 1b). In addition, both plants and fungi disperse independently (*i.e.*, horizontally), and with a few exceptions, do this by meiotically-produced propagules. These latter features mean that the fates of the mutualistic partners are not tightly linked; each is free to try to optimize the interaction in a selfish way. This is predicted to lead to low specificity, and possibly cheating (Borowicz and Juliano, 1991; Law, 1988).

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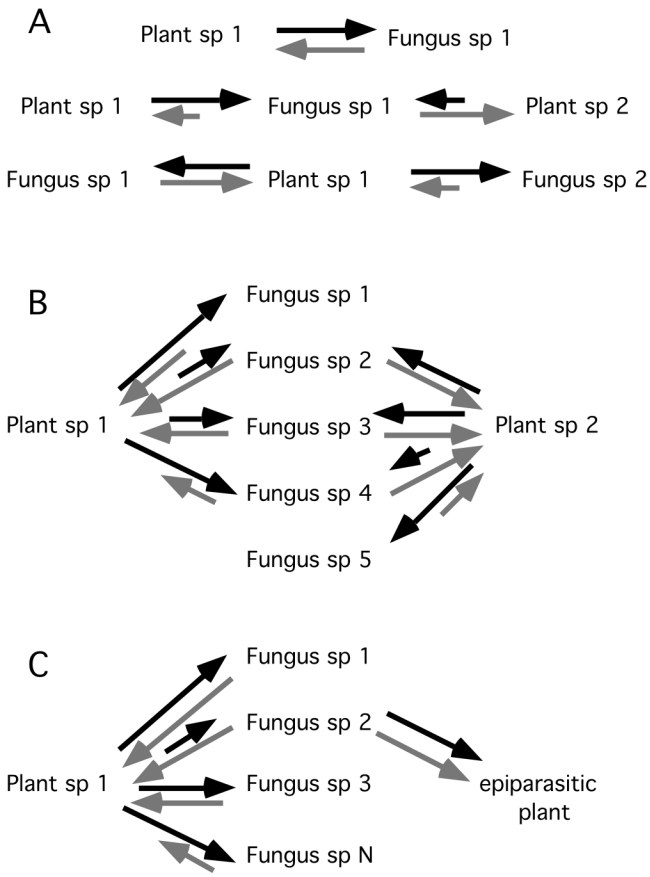

FIG. 1. Conceptual models of plant-fungal interactions in ectomycorrhizal systems. **A.** A ''textbook'' model involves a single plant and a single fungus that exchange fixed carbon (black arrows) for mineral nutrients (gray arrows). This can become more complex as two different fungi are connected to the same plant, or as two different plants are connected to the same fungus. In the latter cases, the cost/benefit relationship, as indicated by the length of the arrows may differ between plant/fungal combinations. In some cases one plant may benefit indirectly from another plant's investment (*i.e.*, facultative epiparasitism). **B.** A more realistic model shows two plants connected by multiple fungi with differences in cost/benefit relationship between each plant/fungal combination. Some specialist fungi (sp. 1 and sp. 5) have restricted host association patterns. **C.** An epiparasitic model shows a non-photosynthetic plant that specializes on one fungal species and received both carbon and nutrients from it.

SPECIFICITY OF PLANTS FOR FUNGI—EPIPARASITIC PLANTS ARE THE EXCEPTION

The vast majority of ectomycorrhizal plants associate with large numbers of distantly related fungi (Borowicz and Juliano, 1991; Molina *et al.*, 1992). Over the entire geographic range of a plant species, the number of fungal associates can be in the thousands (Trappe, 1977). At a local scale, tens of ectomycorrhizal fungi are often found on single trees or in single small patches of monoculture forests (Bruns, 1995). This generalized association pattern seems to make sense for plants for at least two reasons: 1) it increases the chances that seedlings will not be limited in new settings to which they are dispersed, and thereby increases the habitats in which the plants can live; and 2) it may increase plant access to mineral nutrients both within and between sites if individual fungal species are habitat-adapted or if the EM fungal species vary with respect to their access to different nutrient pools.

Exceptions to the broad host association pattern are rare within photosynthetic plants. *Alnus* species are often mentioned in this regard (Molina *et al.*, 1992) because their fungal associates are few and highly specialized to *Alnus* (Miller and Koo, 1991; Miller *et al.*, 1992; Molina, 1979), but *Alnus*-associated fungi are drawn from several distantly related lineages. Thus, *Alnus* does not show phylogenetic specificity; it only associates with a set of fungi that do. The tropical tree *Pisonia grandis* is perhaps the only example of a photosynthetic plant that exhibits high levels of specificity; it appears to associate only with species in the Thelephoraceae (Chambers *et al.*, 1998), but details of this interaction are not well documented. Some photosynthetic orchids provide additional examples, but they differ in that they receive fixed carbon from their fungal associates, rather than providing carbon to them (Taylor *et al.*, 2001). In this respect they are more similar to the non-photosynthetic epiparasitic plants discussed below.

In contrast to the above examples, host specificity appears to be the rule for non-photosynthetic plants that obtain all of their fixed carbon through EM fungi. These have been called ''**epiparasites**'' or ''mycorrhizal cheaters'' because they indirectly obtain fixed carbon from surrounding photosynthetic plants through the EM fungi that are associated with both plants (Björkman, 1960). Cullings *et al.* (1996) used several molecular identification methods, based primarily on the fungal mitochondrial large subunit rRNA gene, to show that *Pterospora andromedea* (Monotropoideae) associated only within a single species group of *Rhizopogon*. Based on smaller sample sizes, they also suggested that *Monotropa uniflora* was restricted to members of the Russulaceae. *Sarcodes sanguinea* appeared to associate with at least three distantly related groups of fungi, but it was the only apparent generalist among the members of the Monotropoideae sampled (Cullings *et al.*, 1996).

We have since followed up on this work with larger sample sizes and with identifications that were primarily based on nucleotide sequences of the ribosomal DNA internal transcribed spacer region (ITS). The increased sample gave us better confidence that the patterns were general and the ITS sequences gave us better phylogenetic resolution for the fungal identifications. One major difference we found was that *Sarcodes sanguinea* is also a specialist; it is consistently associated with the *Rhizopogon ellenae* species group over most of its geographic range (Bidartondo and Bruns, 2001; Kretzer *et al.*, 2000). At the very northern part of its range it is associated with *R. subpurpurascens*, which is the closest relative of the *R. ellenae* complex (Bidartondo and Bruns, 2002). We were also able to show that all other members of the Mon-

otropoideae appear to have specific associations, although different monotrope species target different fungal lineages (Bidartondo and Bruns, 2001). Thus, no exceptions exist within this plant subfamily to the rule that epiparasitic plants are specialists.

This pattern extends beyond the Monotropoideae, as non-photosynthetic orchids that associate with ectomycorrhizal fungi also exhibit specific fungal associations. Taylor and Bruns (1997) showed that *Cephalanthera austinae* and *Corallorhiza maculata* are associated exclusively with members of the Thelephoraceae and the Russulaceae, respectively. *Corallorhiza maculata* and *Corallorhiza mertensiana* were later investigated in more detail and found to associate with mutually exclusive subsets of species from the Russulaceae (Taylor and Bruns, 1999). Association patterns varied among populations; in *C. maculata* this variation correlated with habitat differences such as forest type and altitude, while in *C. mertensiana*, host associations patterns varied with geography. It was suspected that the underlying cause of the host differences was at least partially genetic, because sympatric populations of the two orchids shared no hosts in common, and sympatric color variants of *C. maculata* also shared no associates. Recent work has now confirmed that genetic differences within *C. maculata* correlate with host association patterns (unpublished results, D.LT.). A similar fine-scale host-race pattern also occurs in the Monotropoideae, where sympatric plant genotypes of *Pterospora andromedea* discriminate between the *Rhizopogon arctostaphyli* or *R. salebrosus* species groups (Bidartondo and Bruns, 2002). These are the first examples of host-race formation in mycorrhizal plants.

The high levels of specificity, the geographic mosaic pattern associated with it, and the evidence for host-races are all reminiscent of more traditional parasites (Price, 1980; Thompson, 1994; Thompson and Burdon, 1992). But are these plants truly parasitic, or, as suggested by Miller and Allen (1992), is there some mutualistic aspect to this interaction that is not obvious? Early work by Björkman (1960) showed that extracts from *Monotropa hypopithys* stimulated its fungal associate. Similarly, Bidartondo *et al.* (2000) found that the fungal associate of *S. sanguinea*, *Rhizopogon ellenae*, increased its abundance on *Abies* roots by three orders of magnitude near *S. sanguinea* plants. Furthermore, the abundance of *Abies* roots also increased significantly around *S. sanguinea*. Of course, many plant parasites stimulate their hosts causing hypertrophy and gigantism; broom formation in mistletoes, and gall formation by insects and rust fungi are examples. These types of growth stimulations, however, are at least mildly detrimental to the host. Thus, the question remains, is fungal stimulation by the monotropes simply another example of parasite manipulation of the host? The fact that *Abies* roots have no direct connection to *S. sanguinea* makes it seem unlikely that this pattern is simply due to hypertrophic stimulation by a parasitic interaction. However, we do not know whether the stimulation of *Rhizopogon* has a cost to the fungus (*e.g.*, reduced sporulation or longevity). In any case, the interactions between these non-photosynthetic plants and the rest of the mycorrhizal community should be relatively simple compared to the complexity of green plants, and this simplicity is the result of the specificity (Fig. 1c).

We argued above that being a generalist with respect to fungal associations provided some obvious advantages to photosynthetic plants. Why then is specialization so common in non-photosynthetic plants, and how can these plants overcome the problem of establishment that is presented by specialization? If non-photosynthetic plants are simply parasites, then the ''why'' part of this question has two potential answers: 1) specialization may allow the plant to better adapt to the physiology of a particular host, thereby maximizing its ability to obtain resources from it; 2) parasites could be driven toward specialization by development of resistance among potential hosts in a gene-for-gene arms race. Unfortunately, there are no data to support or reject either of these alternatives in the case of these plants. In contrast, the answer to how these plants can specialize without incurring tremendous cost at the establishment phase does have a fairly clear answer with some experimental support: seed dormancy, coupled with host-specific stimulation of germination, may allow these plants to disperse and wait for the host to come to them. Bruns and Read (2000) have recently shown that two monotropes, *Sarcodes sanguinea* and *Pterospora andromedea* behave in this way *in vitro*, and they showed that the fungal chemical signal that the seeds react to is either diffusible or volatile. Similarly, McKendrick *et al.* (2000) have shown that seeds of the orchid *Corallorhiza trifida* lie dormant and germinate in response to the ''correct'' fungus in the field. In addition, seeds of some photosynthetic orchids are stimulated to germinate by specific fungi (Taylor *et al.*, 2001).

In vitro stimulation of seed germination is not perfectly correlated with observed natural host pattern; typically it is broader. Fungi that are not observed to associate with mature plants can stimulate seeds to germinate. This pattern has been reported in photosynthetic orchids and has led to an underestimation of the level of specificity in mature plants (Taylor *et al.*, 2001). In the case of two monotropes, *Sarcodes sanguinea* and *Pterospora andromedea*, only *Rhizopogon* species have been found to stimulate germination *in vitro*. However, some closely related *Rhizopogon* species, that are not observed to associate with adult plants, also stimulate seed germination *in vitro*. This difference may be due to some artifact of the *in vitro* conditions, or it may indicate that closely related fungi produce similar chemical signals that these plants can not distinguish. Recently, we conducted field experiments, in which we planted seeds in retrievable packets, and we found that *Sarcodes sanguinea* makes the same ''mistakes'' in the field; its seeds germinate in response to *Rhizopogon salebrosus* and *R. arctosta-*

phyli, two species that are closely related to its ''correct'' host, *R. ellenae* (M.I.B. and T.D.B., unpublished results). These mistakes are likely to occur frequently, as all three *Rhizopogon* species and *S. sanguinea* often occur at the same sites. However, the fate of these mistakes is unclear (*i.e.*, death or replacement?). What is clear is that mature *Sarcodes sanguinea* are never found to be associated with the ''wrong'' species (Kretzer *et al.*, 2000; Bidartondo and Bruns, 2001).

SPECIFICITY OF FUNGI FOR PLANTS

Who gains from fungal specialization?

On the fungal side of the ectomycorrhizal symbiosis it is well known that various levels of specificity exist (Borowicz and Juliano, 1991; Molina *et al.*, 1992); it is less clear what individual species of fungi gain from host specialization, or what plants gain from associating with specialist fungi. One would assume that there is a cost to specialization for the fungus, that being the inability to colonize other hosts. Advantages are less obvious, but if specialization provides greater physiological compatibility with the targeted plant host, it could result in greater competitive ability on that host or greater access to the host's resources.

The disadvantages for a plant that associates only with specialist fungi would be similar to those discussed above for plants that are themselves specialists; the chance that the necessary specialist fungi will be somewhere that the plant does not already exist should be less than for fungi that are generalists. This problem could be reduced if the spores or other propagules can remain viable for long periods of time, as in the case of *Alnus* and *Alpova diplophloeus* (see Miller *et al.*, 1992, 1994).

Molina *et al.* (1992) hypothesized one advantage for plants that associate with specialist fungi: specialized associations would reduce the chances of indirectly helping competing plant species. We will refer to this process as ''**facultative epiparasitism**.'' It occurs when one plant indirectly parasitizes a second plant through an EM fungus connected to both. This would happen when one plant reaps the benefits of another plant's investment in an EM fungus. A simple case is diagramed in Figure 1a, line 2, where plant species 2 is acting as a facultative epiparasite. It gains more nutrients for its carbon investment than does plant species 1. Thus, plant species 1 is indirectly subsidizing species 2. This subsidy could either involve direct transfer or simply differential return on investment, as diagramed. Rewards or transferred commodities could be fixed carbon, mineral nutrients, or water (Newman, 1988). Direct carbon transfer has received most of the attention (Newman, 1988; Simard *et al.*, 1997), but its significance remains controversial (Robinson and Fitter, 1999). For the purposes of this paper, let us assume that facultative epiparasitism could be important in some settings; it then follows that plants associated with specialized fungi are more protected from interspecific epiparasitism than plants associated with generalist fungi.

The case of Suillus

It was with these ideas in mind that Gardes and Bruns (1996) studied *Suillus pungens*. This species exhibits a very narrow host range. In nature it fruits almost exclusively with *Pinus muricata* and *Pinus radiata*, two West Coast endemic species with small and highly scattered natural ranges. Furthermore, it is a member of the **suilloid group**, which is composed of *Suillus*, *Rhizopogon*, *Truncocolumella*, *Gomphidius*, and *Chroogomphus*. This is a monophyletic lineage (Bruns *et al.*, 1998; Kretzer and Bruns, 1999; Kretzer *et al.*, 1996) that is almost entirely restricted to hosts in the Pinaceae, and it is the largest group of ectomycorrhizal fungi that exhibits this degree of host specificity. In addition, most suilloid species are restricted to single plant genera, subgenera, or species groups within the Pinaceae, and individual clades within both *Suillus* and *Rhizopogon* frequently exhibit these narrow host association patterns (Grubisha, 1998; Kretzer *et al.*, 1996). Because of this evolutionary history, one might expect suilloid species to have capitalized on whatever advantages specialization might offer.

Suillus pungens is one of the most abundant fruiters in the coastal pine forests that we have studied. Based on this observation, Gardes and Bruns (1996) hypothesized that *S. pungens* was likely to be a dominant species on roots as well, but this was not the case. Instead, it was found to be a minor component of a complex community that was dominated by *Russula* species, and *Tomentella sublilacina* (Gardes and Bruns, 1996). Several hypotheses were put forth to explain this discrepancy between heavy fruiting and limited below-ground abundance:

1. *S. pungens* invests more in fruiting than in vegetative growth and competition. Essentially, it is ''r-selected.''
2. *Suillus* mycorrhizae may have been missed somehow; either because of seasonal shifts or clumped spatial distribution.
3. *Suillus* may obtain significant amounts of carbon saprobically.
4. *Suillus* may be more efficient at obtaining carbon from its host.

The first hypothesis predicts that *S. pungens* is a poor vegetative competitor and that it invests less in mycelial growth than dominant species such as *Russula amoenolens* and *Tomentella sublilacina*. Several lines of evidence contradict these predictions. First, *S. pungens* can expand vegetatively over areas of at least 360 m^2 and persist for decades (Bonello *et al.*, 1998). Second, we have observed large mats of mycelium associated with mushrooms in the field and with seedlings in laboratory microcosm settings. Both observations are in contrast with *Russula* and *Tomentella* species, which have relatively small genets (Redecker

et al., 2001, and unpublished results, T.D.B.), and produce little visible mycelium.

The second hypothesis is harder to eliminate because the spatial sizes of below-ground samples are always small; therefore the chance for missing a species is high. However, in four other nearby sites where we sampled the below-ground community of the undisturbed forest along transects, we never saw *S. pungens* mycorrhizae again (Gardes and Bruns, 1996; Horton and Bruns, 1998; Taylor and Bruns, 1999; and unpublished data, T.D.B.). Only when samples were taken below or adjacent to mushrooms of *Suillus pungens*, did we find mycorrhizae of this species, and even then, they were a minor component of a complex community.

The third hypothesis, that *S. pungens* obtains additional carbon saprobically, is rooted in the knowledge that *Suillus* has some ability to degrade cellulose (Colpaert and Van Laere, 1996), and the suilloid group is closely related to the Coniophoraceae, a family of wood decay fungi (Bruns *et al.*, 1998). However, *S. bovinus* seems to have a fairly limited ability to decompose leaf litter (Colpaert and Van Laere, 1996; Colpaert and Van Tichelen, 1996), and fruitbodies of *Suillus* have a carbon isotope signature that is typical of other pine-associated mycorrhizal fungi and distinct from that of saprobic fungi (Högberg *et al.*, 1999). These results make the saprobic carbon hypothesis unlikely.

The fourth hypothesis, that *Suillus* gets more pine carbon per mycorrhiza than other species, is interesting, because it could relate to the question of ''what does it gain from specialization?''—it gained the ability to adapt better to its host and extract more carbon. Note, however, that if this hypothesis is correct, it would suggest a larger cost to the tree in associating with *Suillus*. This could explain why *S. pungens* colonizes a minority of the root tips; perhaps the tree invests more growth in parts of its root system that provide a more economic return on its investment. As long as *Suillus* colonizes a minority of root tips, the tree may not be able to discriminate against it, but if *Suillus* comes to dominate a part of the root, then that part of the root system may become a bad investment relative to other parts that cost less and return the same level of nutrients. These arguments all assume that the ability of *Suillus* to deliver water and nutrients to the roots are not proportionality greater, if it were, it could make the higher carbon investment by the plant a bargain.

One last point is worth making about how *Suillus* specificity is initiated. Spores of at least some species are stimulated to germinate by abietic acid, a diffusible secondary chemical released from pine roots (Fries *et al.*, 1987). Spores of the closely related genus, *Rhizopogon*, are also stimulated to germinate by pine roots, although the chemical signal has not been identified (Miller *et al.*, 1993). So just like epiparasitic plants, the first active step in establishing specificity is taken by the specialist; it chooses the host based on specific chemical signals. Another similarity to epiparasitic plant seeds is that there is some evidence of dormancy in *Suillus* and *Rhizopogon* spores (Miller *et al.*, 1993 and see below). Growing mycelium shows a different pattern with respect to specificity. Mycelium of *Suillus grevillei*, a *Larix* specialist, will initiate mycorrhizae with the wrong host (*e.g.*, *Pinus* or *Pseudotsuga*), but when they do, phenolics accumulate in the mycorrhizal root (Duddridge, 1986). This reaction appears similar to the way a host reacts to a pathogen; thus at the mycelial stage, the specificity of *Suillus* may be partially enforced through rejection by non-hosts. This suggests that there may be advantages for non-hosts in not associating with these fungi. However, mycelium from species of *Rhizopogon* in section Amylopogon can colonize *Arbutus* and *Arctostaphylos* species (Molina *et al.*, 1997), demonstrating that not all suilloids are so restricted. Interestingly, these are the same *Rhizopogon* species that are the hosts for the epiparasites *Sarcodes sanguinea* and *Pterospora andromedea*.

Generalist fungi dominate natural systems and connect dissimilar hosts

Fungal taxa that are numerically dominant below ground in the California systems we have studied, are members of the Thelephoraceae and the Russulaceae (Bidartondo *et al.*, 2000; Gardes and Bruns, 1996; Horton and Bruns, 1998; Horton *et al.*, 1999; Stendell *et al.*, 1999; Taylor and Bruns, 1999). Many of the species such as *Tomentella sublilacina*, *Russula amoenolens*, *Russula xerampelina*, *Russula brevipes*, *Lactarius rufus*, and *Lactarius xanthogalactus* are reported to associate with a wide range of hosts. From such reports it is temping to call these species generalists. Caution is necessary in interpreting such records, because host trees often co-occur in the field and so it is difficult to say which trees are the actual hosts. In addition, the widespread occurrence of cryptic species within morphologically defined fungal species always has the potential to make fungal species appear less host-specific than they really are.

Two recent molecular studies removed these doubts for several species and show that most of the dominant species were acting as generalists under field conditions. The first study examined soil cores in which pine and Douglas-fir roots overlapped in space, and found that the top five most abundant species, which accounted for more than 80% of the biomass, were associated with both hosts. In addition many of the rare fungal species were also found to be associated with both tree species (Horton and Bruns, 1998). Most fungi were found to be associated with both tree species within single, 10×40 cm, soil cores. This is a spatial scale that makes it likely that dissimilar tree species were associated not only with the same fungal species, but also the same fungal genotypes. In the second study, Douglas-fir seedlings were found to associate with many of the same fungi found on the EM roots of a hardwood shrub, *Arctostaphylos glandulosa*.

Again, these associations occurred within single soil cores, and thus, it was likely that the same fungal individuals were associated with both hosts (Horton *et al.*, 1999). What makes this result particularly interesting is that the survivorship of *Pseudotsuga* seedlings increased under *Arctostaphylos* chaparral relative to adjacent *Adenostoma fasciculatum* chaparral, and historically *Pseudotsuga* primarily invaded the *Arctostaphylos* sites (Horton *et al.*, 1999). The main difference between these chaparral types is that *Adenostoma* is not generally associated with EM fungi and therefore, provides no resident fungal mycelium for *Pseudotsuga* seedlings. Moisture, soil depth, light, and nutrients were similar between the two chaparral types, and if anything, were more favorable under *Adenostoma*. (Horton *et al.*, 1999). These results provide suggestive evidence for ectomycorrhizal-facilitated plant succession, a process that requires fungal generalists.

Disturbance—a setting where Rhizopogon*, and therefore fungal specialization, thrives.*

In mature California pine forests that we have examined, *Rhizopogon salebrosus*[3] is often frequent but in low abundance, usually accounting for less than 6% of the ectomycorrhizal biomass (Gardes and Bruns, 1996; Horton and Bruns, 1998; Stendell *et al.*, 1999; Taylor and Bruns, 1999). Several other species such as *R. occidentalis* and *R. vulgaris* are common fruiters in mature coastal pine forests, but we have not encountered roots colonized by them in any mature forest settings. Thus, the pattern is similar to that discussed above for *Suillus pungens*—high levels of fruiting associated with low observable root colonization.

In these same forests, however, the soils harbor a rich spore bank of *Rhizopogon* species. This was shown by bioassays—planting pine seedlings into forest soil under laboratory conditions. Over half the assays in which soil was diluted 1:100 with sterile soil produce *Rhizopogon* (Taylor and Bruns, 1999). This means that the density of *Rhizopogon* propagules was very high. The fact that the main propagules were spores was inferred from the facts that 1) *Rhizopogon* was not found on the roots that were present in the soil—thus mycelium was unlikely to be present; 2) the soil was dried prior to the bioassays—a treatment likely to kill active mycelium; and 3) adjacent non-forested areas, where mycelium could not have been present, also yield *Rhizopogon* (Horton *et al.*, 1998). We refer to this abundant stockpile of spore inoculum as a spore bank, because of its obvious similarity to seed banks in plants. The similarity to the behavior of *Pterospora* and *Sarcodes* seeds should also be obvious, and as in that case, dormancy coupled with response to specific host signals could be one way that *Rhizopogon* reduces the cost of specialization.

When and how does this spore bank function? A partial answer came as the result of studies that followed the 1995 Mt. Vision wild fire. This was a severe stand-replacing fire that killed the overstory and incinerated the organic layer in four of our study sites at Pt. Reyes National Seashore. It left us in the unique position of having detailed knowledge of the below-ground species composition prior to the fire. One of the first ways we sampled the post-fire environment was to re-run the soil bioassays from the same site from which they had been conducted prior to the fire. The pre- and post-fire results were almost identical (Baar *et al.*, 1999; Taylor and Bruns, 1999); thus, the spore bank had largely survived the fire. However, the more interesting result came from the seedlings that established naturally at that site; they too were largely colonized by the species present in the spore bank (Baar *et al.*, 1999). This showed that the spore bank played a crucial role in recolonization, and this shift toward spore bank species increased the presence of *Rhizopogon* dramatically.

Judging from species composition in the prefire forest, we can infer that the abundance of *Rhizopogon* species will drop as the forest ages and other competing taxa reestablish their dominance. In fact, even within the post-fire community there are indications that these *Rhizopogon* species are poor competitors. They are most abundant on seedlings that established in the formerly non-forested areas adjacent to burned forests; these are settings where inoculum for other taxa appears to be low (Horton *et al.*, 1998). While in the formerly forested areas, *Rhizopogon* species are less abundant on the naturally established seedlings; this is in spite of the fact that bioassays show that their spores are perhaps the most abundant inoculum in these soils (Baar *et al.*, 1999; Taylor and Bruns, 1999). What did these *Rhizopogon* species gain from specializing?—apparently not a competitive advantage, but just like *Suillus*, their abundant fruiting suggests that they obtain a large amount of fixed carbon.

What effect did the increase in specialized fungi have on the plant community? Kropp and Trappe (1982) and Molina *et al.* (1992) noted that pioneer tree species tend to associate with specialist fungi and later successional species tend to associate with less specific fungi. Molina *et al.* (1992) went on to hypothesize that the specific associations of pioneer trees are of value because they protect them from facultative epiparasitism by competing tree species. However, in our system, such an effect was unlikely in the critical early stages of recolonization, because the fire resulted in a pine monoculture by eliminating the only competing ectomycorrhizal tree species, *Pseudotsuga menziesii*. We also know that by the time these forests are about 35 years old, and co-dominated by both tree species in some areas, the below-ground community is no longer dominated by fungi that would discriminate between these hosts (Gardes and Bruns, 1996; Horton

[3] *R. salebrosus* and *R. occidentalis* were referred to respectively as *R. subcaerulescens* and *R. ochraceorubens* in our previous publications, but ITS sequence analysis of holotype collections revealed that the former names are correct. (Bidartondo and Bruns, 2001; Grubisha, 1998).

and Bruns, 1998; Taylor and Bruns, 1999). However, the bishop pine system that we studied is a much younger system with a more frequent and severe disturbance regime than old growth Douglas-fir/Western Hemlock forests that were discussed by Molina *et al.* (1992). In addition, the host specific *Rhizopogon* species associated with Douglas-fir in the latter forest achieve a more prominent position in the community were they account for 12% of the mycorrhizal roots (Goodman and Trofymow, 1998). Thus, in this and other settings where fire is less severe, or where more than one tree species is involved in the early recolonization, a predominance of specialist fungi could indeed have the effect of reducing interspecfic facultative epiparasitism. How common such settings are, and whether specialist fungi are abundant enough to have such an effect is unknown, but worthy of further research. One additional caveat is that the effect of host specific fungi would be limited to interspecific interactions (*i.e.,* those between different tree species). Intraspecific epiparasitism would not be differentially affected by the specificity of EM fungi, and in many post-disturbance settings the most intense competition is likely to be among individuals of the same species.

Interestingly, Molina *et al.*'s (1992) theory is based primarily on the association of *Rhizopogon* and other Suilloid species with *Pseudotsuga menziesii* and *Pinus*, and the specialist fungi associated with alder. This being the case, an alternative explanation for the correlation of these pioneer tree species and specialist fungi exists. Early successional settings are probably most conducive to those species that colonize primarily via a spore bank, and as argued above, a spore- or seed-bank strategy is one way to increase the chances of finding the correct host by the specialist. Under this scenario, the correlation between fungal specialization and pioneer tree species comes from selective pressure on the specialist rather than the host.

CONCLUSIONS

The patterns of specialization in ectomycorrhizal communities are becoming clearer due to quantitative descriptions of below-ground communities and molecular analyses of epiparasitic plants. Nevertheless, the reasons for these patterns are still unknown. In particular it is unclear what advantage specialization provides to either the plants or the fungi that offsets the lost opportunity to associate with additional hosts. Results from orchids, monotropes, and Suilloid fungi suggest that the cost of specialization is reduced by producing propagules that lie dormant until they encounter the correct host. However, dormant propagules are also employed by generalists, so although this strategy may lessen the cost of specialization, it is unlikely to be the advantage that selects for it.

We hypothesize that the main advantage in EM specialization is that it allows the specialist to derive more resources from its host than are available to generalists that associate with these same hosts. This hypothesis predicts that the main advantage in specialization is for the specialist, not its host. This does not predict that mycorrhizal specialists are devoid of benefits to their hosts (*i.e.,* that they are parasites), it only predicts that if benefits are similar to those of generalists, costs will be greater. This makes testing this hypothesis challenging, because costs and benefits in mycorrhizal systems are likely to be conditioned by the environment.

A second unanswered question is whether specialist fungi affect plant competition by reducing facultative epiparasitism (*i.e.,* Molina *et al.,* 1992). Again the answer to this question is likely to be condition-dependent. A good first step toward structuring a test would be to assemble additional quantitative data on the abundance of specialists in a wider set of communities.

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