

THREE-WAY INTERACTIONS AMONG ECTOMYCORRHIZAL  
MUTUALISTS, SCALE INSECTS, AND RESISTANT  
AND SUSCEPTIBLE PINYON PINES

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*Abstract.*—Herbivores and mycorrhizal fungi are important associates of most plants, but little is known about how these organisms interact. In a 9-yr experiment, we examined how the pinyon needle scale (*Matsucoccus acalyptus*) affects and is affected by the ectomycorrhizal mutualists found on the roots of scale-resistant and -susceptible pinyon pines (*Pinus edulis*). Three major results emerged. First, removal experiments demonstrated that scales negatively affected ectomycorrhiza. Second, although ectomycorrhiza could either positively or negatively influence scale performance by improving plant vigor or increasing plant investment in antiherbivore defenses, we found no ectomycorrhizal effect on scale mortality when we experimentally enhanced levels of ectomycorrhiza. This represented the first test of whether ectomycorrhiza promote plant resistance and contrasted with studies showing that arbuscular mycorrhiza negatively affected herbivores. Third, pinyon resistance to scales mediated the asymmetrical interaction between fungal mutualists and scale herbivores. High scale densities suppressed ectomycorrhizal colonization, but only on trees susceptible to scales. Similarities between mycorrhiza-herbivore interactions and competitive interactions among herbivores suggest broader generalities in the way aboveground herbivores interact with belowground plant associates. However, because mycorrhiza are mutualists, mycorrhiza-herbivore interactions do not fit within traditional competition paradigms. The widespread occurrence and importance of both herbivores and mycorrhiza argue for incorporating their interactions into ecological theory.

Herbivores and mycorrhizal fungi can have significant effects on the population and community dynamics of their host plants. By consuming plant tissues, herbivores influence plant growth, recruitment, mortality rates, community composition, and diversity (Huntly 1991). The extent of herbivory may become so great that the plant becomes a limiting resource, resulting in competition among insect herbivores (Denno et al. 1995). Competitive interactions among herbivores may in turn be influenced by plant resistance traits, but only a few studies have explored the potential for such plant-mediated interactions among herbivores. For example, balsam aphids (*Mindarus abietinus*) reduced the survival of spruce budworms (*Choristoneura fumiferana*) but only on trees that were susceptible to aphids (Mattson et al. 1989).

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The effects of mycorrhizal fungi on plant growth, survival, and reproduction (Harley and Smith 1983; Stanley et al. 1993) can also influence plant communities (Allen et al. 1992; Molina et al. 1992). Mycorrhiza are associations between many land plants (Newman and Reddell 1987) and an estimated 5,000+ species of fungi (Molina et al. 1992). Mycorrhizal fungi are generally considered to be mutualists that provide plants with increased nutrient uptake and protection from pathogens in exchange for photosynthate (Harley and Smith 1983). However, although mycorrhizal fungi may be present on roots continuously, they may benefit plants only during some portions of the plant's life cycle (Fitter 1991) or only under certain environmental conditions (e.g., West et al. 1993*b*). Mycorrhizal fungi can influence plant community composition by mediating competition among plant species that vary in their dependence on the mycorrhizal association and/or by facilitating the transfer of materials among plants via mycorrhizal linkages (Miller and Allen 1992).

Because herbivores consume/damage photosynthetic tissue and mycorrhizal fungi depend on the products of photosynthesis for most or all of their energy needs (Allen 1991), these two groups of organisms are likely to interact with one another. Mycorrhizal fungi both consume photosynthate and assist in its production by enhancing nutrient uptake by plants (Allen 1991). Therefore, the relationship between herbivores and mycorrhizal fungi is likely to be complex and may lie outside the normal definitions of the types of interactions that occur among species. For example, by consuming photosynthetic tissue, herbivores are likely to reduce the amount of photosynthate available to mycorrhizal fungi. Reductions in mycorrhizal colonization following herbivory have been observed in plants colonized by both arbuscular mycorrhizal (e.g., Daft and El-Giahmi 1978; Same et al. 1983; Bethlenfalvay et al. 1988; Trent et al. 1988) and ectomycorrhizal fungi (e.g., Gehring and Whitham 1991; reviewed in Gehring and Whitham 1994*b*). Mycorrhizal fungi, however, are unlikely to have an equivalent reverse effect on herbivores through their consumption of photosynthate unless they are acting as parasites rather than mutualists (e.g., Koide 1985; Johnson et al. 1992).

However, mycorrhizal fungi still have the potential to significantly affect herbivore performance. First, by increasing the vigor of their host plants, mycorrhiza could improve the performance of herbivores that selectively attack and perform better on more vigorous hosts (Price 1991). Alternatively, herbivore performance is negatively associated with plant vigor in many plants (White 1984; Waring and Cobb 1992), so that mycorrhiza could improve plant resistance by improving plant performance. Finally, mycorrhiza could alter plant carbon-to-nutrient ratios, allowing plants to increase their investment in antiherbivore defenses, which thereby decreases herbivore performance (Jones and Last 1991). Regardless of the mechanisms involved, in the few studies that have examined the effects of mycorrhiza on herbivores, both above- and belowground insect herbivore performances were lower on plants colonized by arbuscular mycorrhiza than on control plants (Rabin and Pacovsky 1985; Gange and West 1993, 1994; Gange et al. 1994).

Our long-term studies of pinyon pine offered the opportunity to examine the

relationships among a herbivorous insect, the pinyon needle scale (*Matsucoccus acalyptus*), ectomycorrhizal fungal mutualists, and their common host, pinyon pine (*Pinus edulis*). Although we have examined some of these interactions in a previous study (Del Vecchio et al. 1993), the present article extends that work in three important ways. First, we integrate plant resistance, herbivory, and ectomycorrhizal mutualism by examining this three-way interaction from the perspective of each organism. Second, we examine the role of ectomycorrhiza in affecting herbivore performance; previous studies have examined only arbuscular mycorrhiza. Ectomycorrhiza are formed by an estimated 2,000 species of woody perennials (Kendrick 1992) and differ from arbuscular mycorrhiza both morphologically and in the major divisions of fungi involved (Reid 1990). Ectomycorrhiza also have been hypothesized to differ from arbuscular mycorrhiza in a variety of functional characteristics, including the degree of protection they provide plants against both natural enemies and stressful physical conditions (Connell and Lowman 1989). Finally, we discuss the prevalence and importance of these interactions in nature and their similarities to interactions among competing herbivores.

#### METHODS

##### *Description of the Study Site and Background Biology of Study Organisms*

We examined the relationships among pinyon pines, ectomycorrhiza, and pinyon needle scale near Sunset Crater National Monument, northeast of Flagstaff, Arizona (2,000-m elevation). Soils in the area consisted of lava, ash, and cinders low in nutrients and moisture (Mopper et al. 1991*b*; Gehring and Whitham 1994*a*). Pinyon root systems were shallow; most of the fine roots occurred in the upper 20–25 cm of cinders (Gehring and Whitham 1991; Del Vecchio et al. 1993). When grown in cinders, pinyon seedlings had a size positively linearly correlated with the number of ectomycorrhiza, indicating that ectomycorrhiza were mutualists in this environment (Gehring and Whitham 1994*a*).

The pinyon pine needle scale, *Matsucoccus acalyptus* (Homoptera: Margarodidae), feeds intracellularly on the mesophyll tissue of *Pinus edulis* needles by inserting its stylet through stomata. Feeding begins in May and lasts 6 mo for males and 10 mo for females (Cobb and Whitham 1993). At high scale densities, scale feeding resulted in extensive chlorosis and premature needle abscission (Cobb and Whitham 1997), leading to an altered tree architecture resembling a poodle's tail with only the most recent 1–2 yr of needles remaining (Mopper et al. 1991*a*). Pinyons varied markedly in susceptibility to scales; scale mortality was two to five times higher on resistant trees than that on susceptible trees (Cobb and Whitham 1993; Del Vecchio et al. 1993).

##### *Experimental Tests of the Interaction between Ectomycorrhiza and Scales*

To examine the interactions between ectomycorrhiza and scales, we performed two experiments on trees that varied in resistance to scale herbivory. In the first experiment, we removed scales from susceptible trees and compared

their ectomycorrhizal levels with those of naturally resistant and susceptible trees. If scales negatively affected ectomycorrhiza, we predicted that ectomycorrhizal densities would be greater on scale-resistant trees than on scale-susceptible trees and that ectomycorrhiza would rebound following scale removal from susceptible trees. In the second experiment, we added equal quantities of scales to resistant trees and to susceptible trees that varied in the amount of ectomycorrhiza present on their roots and monitored scale performance. If ectomycorrhiza positively affected scales by improving host plant vigor, we predicted that scale performance would be poorer on less vigorous susceptible trees with lower levels of ectomycorrhiza than on more vigorous susceptible trees with higher levels of ectomycorrhiza. If ectomycorrhiza negatively affected scales, we predicted that scales would have lower performance and/or cause less damage on trees with higher levels of ectomycorrhiza (i.e., scale performance on susceptible trees with high ectomycorrhiza would be more similar to scale performance on resistant trees than on susceptible trees with low ectomycorrhiza).

#### *Experiment 1: Scale Removal*

We removed scales from 13 susceptible trees in 1986 (hereafter referred to as susceptible scale removal trees). An additional 13 susceptible trees and 13 naturally resistant trees were not manipulated and served as controls. Trees in the three groups grew intermixed within the study site. In April 1994, after 8 yr of scale removal, we collected roots from treatment and control trees and measured ectomycorrhizal colonization as an estimate of the density of ectomycorrhiza.

In 1986, we eliminated scales by removing egg masses from the base of the trees by hand and applying a sticky barrier to the tree trunk to prevent colonization of the tree by larvae from eggs that we missed. We removed the sticky barrier the following spring. We examined the trees annually for the presence of scales and removed any scales we observed. The pinyon needle scale is an extremely poor disperser (Cobb and Whitham 1993) and did not reach damaging levels on any of the trees from 1986 to 1994. We measured ectomycorrhizal colonization by collecting 40–60 cm of long root from each tree and calculating the percentage of short roots that were actively ectomycorrhizal as described previously (Gehring and Whitham 1991, 1994a, 1995; Del Vecchio et al. 1993).

Del Vecchio et al. (1993) examined the ectomycorrhiza of susceptible, resistant, and susceptible scale removal trees in 1990 and 1991 and found that ectomycorrhizal colonization was approximately 30% greater in the latter two groups than in susceptible trees. Because our present study was designed to integrate both the effects of scales on ectomycorrhiza and the effects of ectomycorrhiza on scales, we continued the scale removal experiments of Del Vecchio et al. (1993) for an additional 3 yr (total length of scale removal = 8 yr) to allow the trees to recover fully from scale attack. In addition to measuring ectomycorrhiza, we also measured foliage retention as an indicator of recovery following scale removal. We quantified the amount of foliage retained in the three groups of trees over the last 5 yr (i.e., percentage foliage retention = the

length of 5 yr of shoot growth with a full complement of needles divided by the total length of 5 yr of shoot growth). We measured five branches per tree and averaged them. We then added scales to these same trees in a second experiment to examine whether enhanced ectomycorrhizal colonization affected scale performance.

#### *Experiment 2: Scale Addition*

In 1994, we added an equal quantity of scale eggs (approximately 10,000 eggs) to the base of each resistant (high ectomycorrhiza), susceptible (low ectomycorrhiza), and susceptible scale removal tree (high ectomycorrhiza). To ensure that scale densities were nearly equal in all groups, we removed scale eggs from the susceptible trees before adding eggs to them and measured scale densities on all trees. Before scale removal and readdition, scale densities were high on susceptible trees (approximately 10 scales/centimeter needle length), whereas the resistant and susceptible removal trees lacked scales. Following the experimental scale addition, scale densities were comparable among groups ( $\bar{x} \pm 1$  SE scales/centimeter needle length for resistant trees,  $0.30 \pm 0.028$ ; for susceptible trees,  $0.35 \pm 0.024$ ; and for susceptible scale removal trees,  $0.30 \pm 0.024$ ;  $F = 1.691$ ,  $P = .199$ ). Scales were added in April 1994, and scale performance and damage due to scale feeding were measured in April 1995.

We estimated scale mortality by collecting 60 needles (30 fascicles) per tree from the inner and outer portions of the canopy at all compass points. We collected only second-year needles for two reasons. First, scales settle on needles before the current-year needles have emerged. Second, susceptible trees only had 2 yr of needles, the current-year needles and the second-year needles. Scales were examined under a dissecting microscope and classified into three groups: individuals that died as first instars, individuals that died as second instars, and individuals that emerged as adults. We used the characteristics described elsewhere (Cobb 1990; Cobb and Whitham 1993) to distinguish between instars and to recognize emerged adults. We measured cumulative scale mortality as the percentage of scales that died before emergence (i.e., the number of scales that died as first or second instars divided by the total number of scales). As a second test of the hypothesis that ectomycorrhiza affected scale performance, we regressed ectomycorrhizal colonization against cumulative scale mortality in resistant trees.

Ectomycorrhiza may influence herbivore damage to plants and herbivore performance directly. Therefore, we assessed the damage that resulted from scale feeding by visually estimating the percentage chlorosis of each of 30–40 needles per tree. Because scales that died as first or second instars caused less damage than did scales that reached maturity, we restricted our estimates to chlorosis caused by mature scales that fed alone on a needle. We also measured the sizes of two male and two female scales for each of 33 trees (11 trees per group) to establish whether differences in scale size contributed to the variation among groups in chlorosis. Scale size was determined by measuring the length, width, and depth of individual scale exoskeletons under a dissecting microscope and calculating a volume in cubic millimeters. Again, we only measured scales that

fed alone on a needle. Values of the four scales per tree were averaged to obtain one value per tree.

Finally, to test predictions of the carbon-nutrient exchange ratio concept of Jones and Last (1991), we determined the percentage of carbon (C) and nitrogen (N) in needles collected from resistant, susceptible, and susceptible scale removal trees. Jones and Last (1991) predicted that trees growing in low-nutrient, high-light environments would have higher foliage C:N ratios if they were ectomycorrhizal than if they were not. These higher relative levels of C were hypothesized to allow greater investment in carbon-based defenses, enhancing resistance to herbivores (Jones and Last 1991). We expected that trees with more ectomycorrhiza (resistant and susceptible scale removal trees) would have higher C:N ratios than would those with fewer ectomycorrhiza (susceptible trees). We analyzed needles that lacked scales but that were part of a fascicle on which scales had fed, limiting our sample size to seven to eight per group. Needles were analyzed by IAS Laboratories in Phoenix, Arizona.

### *Statistical Analyses*

Ectomycorrhizal colonization, foliage retention, scale size, scale density, and percentage chlorosis data were analyzed using one-way ANOVAs followed by Tukey's tests to locate treatment differences. The foliage retention data were log-transformed before analysis to correct variance inequalities. Cumulative scale mortality and scale mortality at the first and second instars were compared among groups using a multivariate ANOVA. Univariate  $F$  ratios were also calculated followed by Tukey's tests to locate treatment differences. Needle carbon and nitrogen data were also analyzed using a multivariate ANOVA followed by univariate  $F$ -tests and Tukey's tests. All tests were performed on SYSTAT for Windows, version 5.0 (SYSTAT 1992).

## RESULTS

### *Effects of Scale Herbivory on Ectomycorrhizal Colonization*

Two lines of evidence demonstrated that scales negatively affected ectomycorrhiza. First, young pinyon pines susceptible to scale herbivory had 37% lower levels of ectomycorrhizal colonization than did resistant trees. Second, susceptible trees had 44% less ectomycorrhiza than did susceptible trees from which scales had been removed for 8 yr ( $F = 34.921$ ,  $df = 2, 36$ ,  $P < .0001$ ) (fig. 1A). These findings were consistent with those of Del Vecchio et al. (1993), who observed similar patterns in a previous year.

### *Effect of Ectomycorrhizal Colonization on Scale Mortality and Scale-Induced Damage*

Our measurements of foliage retention demonstrated that 8 yr of scale removal resulted in susceptible trees with foliage resources comparable with those of resistant trees. Foliage retention exceeded 85% for scale-resistant and suscep-

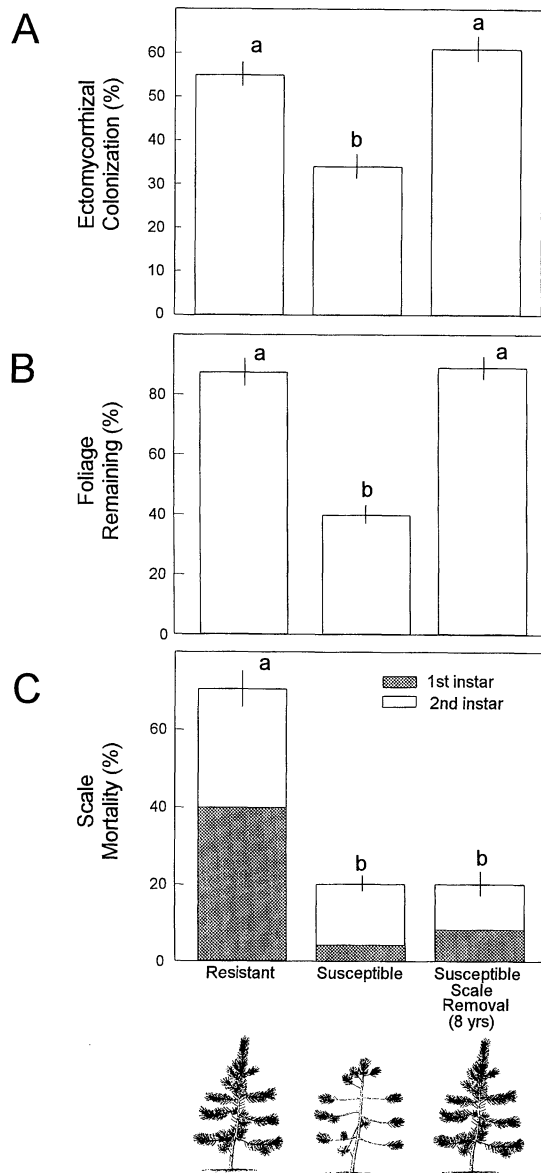


FIG. 1.—Levels of ectomycorrhizal colonization (A), the percentage of foliage retained (B), and the percentage mortality of scale insects (C) on scale-resistant, scale-susceptible, and susceptible trees from which scales were removed for 8 yr. Data are presented as means  $\pm 1$  SE. Different letters above the bars denote significant differences at  $P < .05$ . In graph C, cumulative scale mortality is represented by bar height, first instar mortality is represented by the portion of the bar shaded gray, and second instar mortality is represented by the open portion of the bar. The responses were the same for first instar, second instar, and cumulative mortality, so the letters above the bars in C represent differences among groups in all of these variables.

tible scale removal trees but was less than 40% for scale-susceptible trees ( $F = 176.74$ ,  $df = 2, 36$ ,  $P < .0001$ ) (fig. 1B).

Comparisons of scale mortality on resistant, susceptible, and susceptible scale removal trees demonstrated that variation in levels of ectomycorrhiza did not affect scale performance. Although scale-resistant and susceptible scale removal trees had similar levels of ectomycorrhizal colonization (fig. 1A), cumulative scale mortality was 3.4 times higher on the resistant trees than on the susceptible scale removal trees (fig. 1C). These differences in scale mortality among groups were statistically significant when first, second, and cumulative scale mortality were analyzed together using a MANOVA (Wilks's  $\lambda = 0.166$ ,  $F = 16.466$ ,  $df = 6, 68$ ,  $P < .0001$ ) and when using univariate  $F$ -tests ( $F = 38.487$ ,  $df = 2, 36$ ,  $P < .0001$  for first instar mortality;  $F = 7.353$ ,  $df = 2, 36$ ,  $P = .002$  for second instar mortality; and  $F = 82.232$ ,  $df = 2, 36$ ,  $P < .0001$  for cumulative scale mortality). Tukey's tests indicated that scale mortality on resistant trees differed significantly ( $P < .05$ ) from scale mortality on both scale-susceptible and susceptible scale removal trees in all univariate comparisons (fig. 1C). In contrast, scale mortality did not differ significantly between the latter two groups. Despite substantial differences in level of ectomycorrhizal colonization, scale mortality was nearly identical on scale-susceptible and susceptible scale removal trees. These findings indicate that ectomycorrhiza did not promote resistance to scales in susceptible pinyon pines.

Ectomycorrhiza also were not involved in the resistance mechanism of scale-resistant trees. The variability in scale mortality among resistant trees was not correlated with the level of ectomycorrhizal colonization ( $r^2 = 0.1495$ ,  $F = 1.934$ ,  $P = .192$ ). For example, the resistant tree with the lowest level of ectomycorrhizal colonization (35.4%) had a scale mortality of 84.7%, while the resistant tree with the highest level of ectomycorrhizal colonization (68.6%) had a scale mortality of 58.8%.

Finally, ectomycorrhiza did not reduce the damage resulting from scale feeding. Levels of leaf chlorosis due to mature scales were similar in susceptible trees with lower and higher levels of ectomycorrhiza (fig. 2A). Resistant trees experienced less chlorosis than did the other groups; this difference was significant for susceptible versus resistant trees ( $F = 11.651$ ,  $df = 2, 36$ ,  $P < .0001$ ) (fig. 2A). This finding suggests not only that resistant trees were more resistant to scales but that they were also better able to withstand scale herbivory. The lower chlorosis in resistant trees was not due to smaller scales; scale size did not differ significantly among groups ( $F = 0.2579$ ,  $df = 2, 27$ ,  $P = .6171$ ) (fig. 2B).

#### *Needle C:N Ratios and Scale Resistance*

Differences among scale-resistant, scale-susceptible, and susceptible scale removal trees in needle nitrogen or carbon contents did not help explain the variation among groups in scale mortality. Needle carbon contents were approximately equal in all three groups (univariate  $F = 1.349$ ,  $df = 2, 20$ ,  $P = .282$ ) (fig. 3A). In contrast, needle nitrogen content was significantly greater in the susceptible trees than in the resistant trees or the susceptible scale removal trees



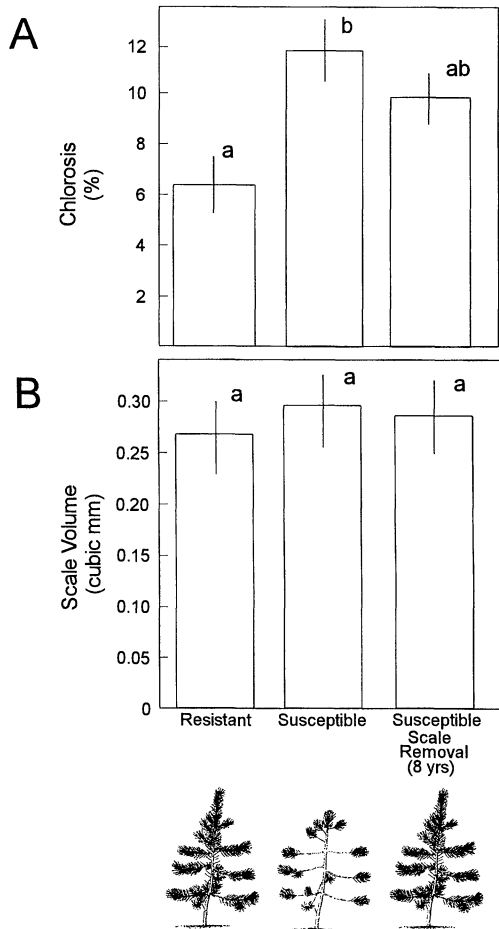


FIG. 2.—The levels of leaf chlorosis caused by scale feeding (A) and scale size (B) on scale-resistant, scale-susceptible, and susceptible trees from which scales were removed for 8 yr. Data are presented as means  $\pm$  1 SE. Different letters above the bars denote significant differences at  $P < .05$ .

(univariate  $F = 14.688$ ,  $df = 2, 20$ ,  $P < .0001$ ) (fig. 3B). Similar differences were observed among groups in carbon-to-nitrogen ratios (univariate  $F = 8.102$ ,  $df = 2, 20$ ,  $P = .003$ ) (fig. 3C). The multivariate ANOVA was significant (Wilks's  $\lambda = 0.265$ ,  $F = 5.665$ ,  $df = 6, 36$ ,  $P < .0001$ ). Thus, although the foliage nitrogen content decreased and the C:N ratio of susceptible trees increased to levels similar to those of resistant trees following scale removal, transfer experiments demonstrated that scale mortality was nearly equal on scale-susceptible and susceptible scale removal trees.

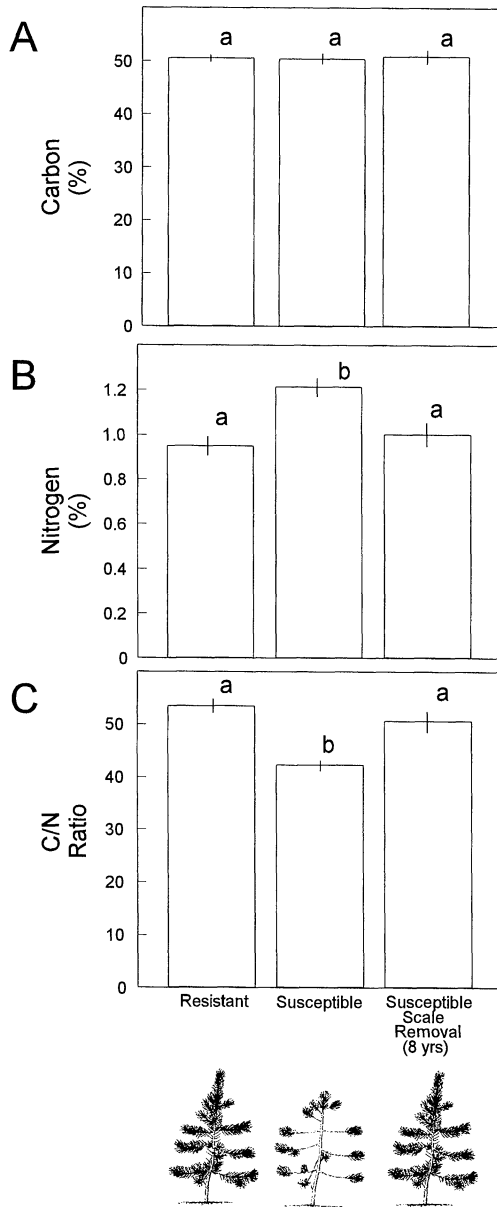


FIG. 3.—The percentage of pinyon needle tissue composed of carbon (A) and nitrogen (B) and the C:N ratios (C) of resistant, susceptible, and susceptible trees from which scales were removed for 8 yr. Data are presented as means  $\pm$  1 SE. Different letters above the bars denote significant differences at  $P < .05$ .

## DISCUSSION

*Importance of the Long-term Experiment*

Because many evergreens retain foliage for several years, recovery from defoliation is a long-term process. For example, young pinyons typically had 7–8 yr of foliage on their branches, but this was reduced to 1–2 yr of foliage on trees that suffered chronic herbivory by scales (Cobb 1990). In the present study, it took several years of scale removal for susceptible pinyons to have levels of ectomycorrhizal colonization similar to those of resistant trees and a full 8 yr of scale removal for susceptible pinyons to accumulate a full complement of needles. Furthermore, foliage carbon and nitrogen levels were also similar in resistant and susceptible scale removal trees. These data demonstrated that long-term removal of scales was critical to examine this plant-herbivore-ectomycorrhiza interaction. This example is not unusual; it took 2–7 yr for the ectomycorrhiza of pinyons to recover from reductions caused by another insect, a stem-boring moth (Gehring and Whitham 1995).

*Ectomycorrhiza and Resistance to Scale Herbivory*

Three lines of evidence demonstrated that ectomycorrhizal colonization did not affect the ability of pinyon pines to either resist or withstand herbivory by the pinyon needle scale. First, although levels of ectomycorrhizal colonization were 44% higher in susceptible scale removal trees than those in susceptible trees, levels of scale mortality were virtually equivalent. Second, ectomycorrhizal colonization and scale mortality were not correlated with one another in scale-resistant trees. Third, the increase in ectomycorrhiza that resulted from the removal of scales from susceptible trees did not improve the ability of pinyons to withstand scale herbivory at the needle level.

By using indirect means (herbivore removal) to increase ectomycorrhizal colonization on susceptible trees, our experiment had additional treatment effects. The only way to remove ectomycorrhizal fungi directly from plants in nature is through the use of fungicides. Although this can be a powerful technique, it also has problems. Fungicides can negatively affect fungi not involved in the mycorrhizal symbiosis (West et al. 1993a), and different ectomycorrhizal fungal species can vary in their responses to the same fungicide (Trappe et al. 1984). Thus, with several hundred species of microbes living within 1 cm of the fine roots of pinyons at our study site (C. Kuske, unpublished data), we judged that the experimental removal of a single herbivore to achieve an increase in ectomycorrhiza was more realistic than the use of a nonspecific fungicide that could affect the entire microbial community. However, as a result of using herbivore removal as a means of increasing ectomycorrhizal colonization, susceptible trees also differed from resistant and susceptible scale removal trees in foliage retention and foliage nitrogen content. Although, these two parameters and unmeasured variables could have affected scale mortality, we detected no effect. Resistant and susceptible scale removal trees differed threefold in scale mortality even though their levels of ectomycorrhiza, foliage retention, and foliage nitrogen were virtually equivalent. We conclude that neither ectomycorrhiza, quantity

of foliage resources, nor foliage nitrogen content significantly affected scale mortality.

This lack of effect of ectomycorrhiza in promoting herbivore resistance contrasts with the results of researchers examining aboveground insect herbivore performance on plants colonized by arbuscular mycorrhizal fungi. Rabin and Pacovsky (1985) observed that corn earworm (*Heliothis zea*) and fall army worm (*Spodoptera frugiperda*) larvae performed better when fed leaves from nonmycorrhizal soybeans than when fed leaves from mycorrhizal soybeans. These differences were not associated with variations in plant nutrients or defensive chemicals but were observed in both insect-susceptible and insect-resistant soybean cultivars. Because the magnitude of arbuscular mycorrhiza-induced resistance was not related to cultivar susceptibility, Rabin and Pacovsky (1985) concluded that mycorrhiza-mediated resistance and cultivar resistance operated by different mechanisms. In our study, initial resistance and susceptibility of pinyons predicted scale mortality regardless of ectomycorrhizal levels, indicating that ectomycorrhiza played no role in resistance and suggesting that other factors such as plant genetics were better predictors of scale mortality.

Our findings regarding nitrogen concentrations and C:N ratios were similar to those observed in a study on a *Plantago lanceolata*-arbuscular mycorrhiza-insect herbivore interaction (Gange and West 1994). Gange and West (1994) found that *Plantago* plants with high arbuscular mycorrhizal colonization had more leaves, lower levels of nitrogen, higher C:N ratios, increased levels of carbon-based compounds that were suspected to be defenses, and reduced herbivore performance than did plants with low arbuscular mycorrhizal colonization (fungicide-treated plants). We also found that pinyons with higher ectomycorrhizal colonization (resistant trees and susceptible scale removal trees) had more foliage, lower nitrogen concentrations, and higher C:N ratios than did pinyons with lower ectomycorrhizal colonization. These data were also consistent with the carbon-nutrient exchange hypothesis (Jones and Last 1991). However, these chemical differences did not correlate with scale mortality or with one class of the presumed carbon-based defenses produced by conifers, the terpenes (Howe and Westley 1988). Scale-resistant and scale-susceptible pinyons did not differ in the concentration of 13 terpenes (Cobb 1990). Pinyons also produce carbon-based phenolics that may defend them against scales. However, if phenolics are associated with scale mortality, our results indicate that their concentrations will not be predicted by levels of ectomycorrhizal colonization or needle C:N ratios.

Thus far, studies of arbuscular mycorrhizal and ectomycorrhizal fungal effects on herbivores have yielded different patterns: arbuscular mycorrhizal fungi had negative effects on insect herbivores, while ectomycorrhizal fungi had no effect (Rabin and Pacovsky 1985; Gange and West 1993, 1994; Gange et al. 1994; this study). However, only four studies have been conducted, with only the present study focusing on ectomycorrhizal fungi. Although most plants form only one type of mycorrhizal association (Molina et al. 1992), the study of the few groups of plants that can form both ectomycorrhizal and arbuscular mycorrhizal associations (e.g., members of the genera *Populus* and *Eucalyptus*) may be particularly informative.

*Comparisons between Mycorrhiza-Herbivore Interactions and  
Herbivore-Herbivore Competition*

Comparing and contrasting the few studies of mycorrhiza-herbivore interactions with the numerous studies of herbivore-herbivore competition suggest important similarities in these interactions. First, with both mycorrhiza-herbivore and herbivore-herbivore interactions, the plant is a common resource that may become limiting and result in competition. The increases in ectomycorrhiza that we observed following scale removal were consistent with release from interspecific competition. Rather than producing chemicals that interfere with ectomycorrhiza, scales more likely affected ectomycorrhiza by removing resources, and the interaction could therefore be described as exploitative competition. In contrast, ectomycorrhiza had no effect on scales, demonstrating that the interaction was strongly asymmetrical. Similarly, most of the competitive interactions documented between insect herbivores were also asymmetrical (Denno et al. 1995).

Second, the outcome of competition between above- and belowground herbivores is similar to the outcome of the interaction between scales and ectomycorrhiza. All documented cases of competition between root-feeding and foliage-feeding insects were asymmetrical, in which the foliage feeders negatively affected the root feeders but not vice versa (Denno et al. 1995). In two studies, foliage feeders even benefited from the presence of root feeders (Gange and Brown 1989; Masters and Brown 1992). Furthermore, in the two studies that contrasted the impacts of root and foliage feeders on plants, foliage feeders had greater negative effects on plants than did root feeders (Moran and Whitham 1990; Masters and Brown 1992). Similar patterns occur with other aboveground herbivore-mycorrhiza interactions. Mycorrhizal reductions were observed following aboveground herbivory in the majority of plants examined (62%) (Gehring and Whitham 1994b), and the occurrence and extent of herbivore-induced mycorrhizal reductions depended on the degree of damage to the host plant (Daft and El-Giahmi 1978; Bethlenfalvay et al. 1985, 1988). Although the mechanism that results in this asymmetry is unclear, the negative effect of aboveground herbivores on both belowground herbivores and mycorrhiza is similar.

Third, plant resistance traits have been shown to influence herbivore-herbivore interactions just as they influenced the interaction between ectomycorrhiza and scales. In our study, the indirect interactions between ectomycorrhiza and scales were mediated by the resistance traits of pinyon pine (fig. 4). Scales and ectomycorrhiza only interacted on susceptible trees, while the ectomycorrhiza of resistant trees were unaffected by scales and remained at high levels. Similarly, an earlier study (Moran and Whitham 1990) found that competition between a leaf-galling and a root-feeding aphid occurred only on plants susceptible to the leaf galler. The role of plant resistance traits in mediating interactions has been little studied, but it is likely to gain in importance as genetic factors are more broadly incorporated into ecological studies.

Although there are many parallels between ectomycorrhiza-scale interactions and herbivore-herbivore competition, the former interaction cannot be consid-

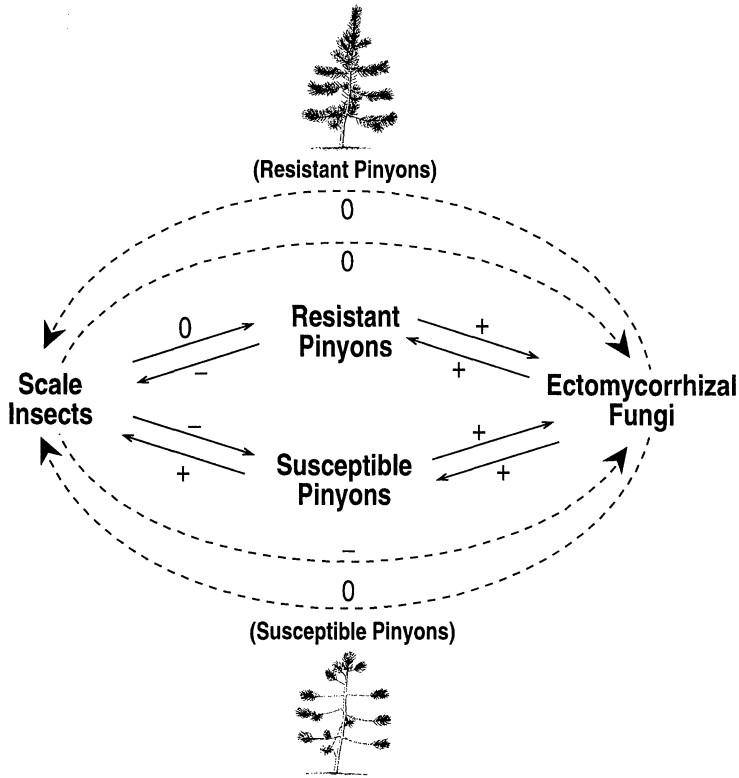


FIG. 4.—A summary of the direct and indirect interactions among scale insects, scale-resistant, and scale-susceptible pinyon pines and ectomycorrhizal fungi. The solid lines represent direct interactions, and the dashed lines represent indirect interactions. The indirect interactions are mediated through the host plant.

ered competition. Although both ectomycorrhiza and scales use plant resources, only scales are capable of negatively affecting resource acquisition by their potential competitor, a necessary condition of exploitation competition (Petren and Case 1996). Ectomycorrhiza consume photosynthate, but because of their role in plant nutrient uptake, they also assist in photosynthate production and thus do not deplete photosynthetic resources. Instead, pinyon ectomycorrhiza, or any mycorrhiza, can only interact with scales or other herbivores by changing plant vigor, by stimulating the production of antiherbivore compounds by plants, or by producing antiherbivore compounds themselves.

Instead of competition, the interaction between ectomycorrhiza and scales may be better classified as an indirect parasitism or a trophic cascade. Scales negatively affected susceptible pinyons and, as an apparent consequence, indirectly and negatively affected the ectomycorrhizal fungal mutualists associated with susceptible pinyon roots (fig. 4). In contrast, resistant pinyons negatively affected scales (fig. 4). Ectomycorrhizal fungi had no reciprocal effect on scales

in either resistant or susceptible trees so that the interaction between scales and ectomycorrhiza could be described as  $(-, 0)$ , consistent with indirect parasitism (fig. 4). Alternatively, the negative effects of scales on ectomycorrhiza were mediated through a consumer-resource interaction (i.e., scale-pinyon pine), and therefore could be classified as a trophic cascade (Wootton 1994).

Regardless of how they are classified, the potential interactions among mycorrhiza, their herbivores, and host plants have not been incorporated into discussions of indirect interactions. We propose that these interactions may be particularly widespread for three reasons. First, both herbivores and mycorrhiza are ubiquitous, and both are dependent on host plants for most or all of their energy needs. Nearly 90% of plants belong to families that are commonly mycorrhizal (Newman and Reddell 1987), and herbivores are a common component of ecosystems. Second, herbivorous insects commonly compete with one another (Lawton and Hassell 1981; Denno et al. 1995), which suggests that plant resources are often limited. For example, interspecific competition was observed in 76% of 193 pairwise interactions between phytophagous insects (Denno et al. 1995). Third, both arbuscular mycorrhiza and ectomycorrhiza are known to interact with plant pathogens (e.g., Perrin 1990) and root-feeding nematodes (e.g., Ingham 1988); they have been shown to protect their hosts from a variety of root-borne diseases and to increase their susceptibility to some viral diseases (Dehne 1982; Perrin 1990). The mechanisms for these effects range from improving plant nutrition to stimulating defensive chemical production by plants (Perrin 1990). A similar range of mechanisms may also occur between mycorrhiza and aboveground herbivores. We suggest that integrated examination of the above- and belowground communities of plants is essential if we are to understand a wide range of important indirect interactions that should be incorporated into the body of theory dealing with ecological interactions.

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