

Mycorrhizal networks counteract competitive effects of canopy trees on seedling survival

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Abstract. The dynamics of forest ecosystems depend largely on the survival of seedlings in their understories, but seedling survival is known to be limited by preemption of light and soil resources by overstory trees. It has been hypothesized that “common mycorrhizal networks,” wherein roots of seedlings are linked to overstory trees by symbiotic mycorrhizal fungi, offset some or all of the negative effects of trees on seedlings. Here we report the results of an unambiguous experimental test of this hypothesis in a monodominant *Pinus radiata* forest. We also tested the hypothesis that adaptive differentiation among plant populations causes local plant genotypes to respond more positively to mycorrhizal networks than nonlocal plant genotypes. Our results demonstrate large positive effects of overstory mycorrhizal networks on seedling survival, along with simultaneous negative effects of tree roots, regardless of whether plant genotypes were locally derived. Physiological and leaf-chemistry measurements suggest that seedlings connected to common mycorrhizal networks benefited from increased access to soil water. The similar magnitude of the positive and negative overstory effects on seedlings and the ubiquity of mycorrhizal networks in forests suggest that mycorrhizal networks fundamentally influence the demographic and community dynamics of forest trees.

Key words: common mycorrhizal network; ectomycorrhizal fungi; local adaptation; Monterey pine forest (California, USA); *Pinus radiata*; seedling growth cf. seedling survival.

INTRODUCTION

In many forests, seedling survival is the chief demographic bottleneck influencing stand structure, species diversity, and ecosystem properties such as carbon storage and nutrient cycling (Shugart 1984, Oliver and Larson 1996). Survival of seedlings is conventionally thought to be determined by light availability and species’ “shade tolerances” or light-response curves (Shugart 1984, Chapin et al. 1987, Canham 1989, Oliver and Larson 1996, Walters and Reich 1997). Recent trenching studies have also shown that root competition for soil resources between seedlings and overstory trees can influence survival, even in deep shade (Coomes and Grubb 2000). Yet, observations that mycelia of mycorrhizal fungi will colonize roots of neighboring trees and seedlings (Newman 1988, Simard and Durall 2004) and that such common mycorrhizal networks (CMNs) can transfer resources between plants or influence their resource budgets (reviewed by Simard and Durall [2004] and Selosse et al. [2006]) have led to the hypothesis that CMNs offset the negative effects of trees on seedling

survival (Newman 1988, Read 1997, Perry 1998, Wilkinson 1998).

Reports of directional carbon, nitrogen, and water movement among plants through CMNs (e.g., Simard et al. 1997, He et al. 2004, 2005, Egerton-Warburton et al. 2007) demonstrate the potential for CMNs to mitigate the effects of overstory competition on understory regeneration. Increased growth and survival of compatible seedlings near network-hosting adults suggest that, in some cases, CMNs might even result in net facilitation of seedlings by older plants (Horton et al. 1999, Newbery et al. 2000, Dickie et al. 2002, Onguene and Kuyper 2002, Nara 2006). A handful of studies have attempted to manipulate networks experimentally to estimate the effects of established CMNs on tree seedling performance (Booth 2004, McGuire 2007, Teste and Simard 2008, Teste et al. 2009). Results of these experiments generally support the notion that CMNs exert a positive influence on networked seedlings, improving growth and survivorship of CMN-compatible seedlings and limiting the survival of CMN-incompatible species. Authors of two studies also attempted to identify mechanisms by which CMNs facilitate tree seedlings in the field, including water redistribution from overstory trees to seedlings (Schoonmaker et al. 2007) and carbon and nitrogen transfer from saplings to seedlings in a clearcut (Teste et al. 2009). However, these studies did not isolate CMN pathways for resource movements, and their results cannot yet be generalized.

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We conducted an experiment in a seasonally dry Monterey pine (*Pinus radiata* D. Don) forest on the Pacific coast of central California, USA, to test the hypotheses that (1) CMNs offset some of the negative effects of trees on growth and survival of seedlings and (2) CMNs reduce seedling water stress in the forest understory. In addition to examining the basic effects and functions of CMNs in the Monterey pine forest, we also explored the possibility that seedling responses to CMNs might vary among different plant genotypes. It has been shown that plant populations often are locally adapted to their biotic and abiotic environments (Linhart and Grant 1996). The role of important soil biota, including mycorrhizal fungi, in such adaptations is not well understood (Schultz et al. 2001). If forest trees are locally adapted to the mycorrhizal fungi prevalent in mature forests, then local plant genotypes are predicted to benefit more from access to CMNs than nonlocal genotypes. Monterey pine is substantially differentiated genetically among populations (Wu et al. 1998, Rogers 2004), so we tested this prediction by comparing the responses to CMNs of Monterey pine seedlings from local and nonlocal populations.

METHODS

Study site

Two simultaneous field experiments were established in a 125 × 200 m plot in a native stand of Monterey pine (*Pinus radiata*) located in coastal Santa Cruz County, California, USA, at Swanton Pacific Ranch, a facility administered by California State Polytechnic University, San Luis Obispo (37°03'47" N, 122°14'17" W). The habitat was dominated by Monterey pines of mixed age, with shallow loamy sand to sandy loam soils classified as Haploixerolls in the Santa Lucia series (Santa Cruz County Soil Survey; USDA 1976). Additional characteristics of the soil were determined from samples collected in August 2006 and January 2008 (Appendix A). Most of the annual average rainfall of 530 mm occurs during the winter months (NOAA National Climatic Data Center, Central Coast Drainage Climatic Division, 1895–2007), during which the minimum monthly average temperature is 4.0°C. The prolonged dry summer season, typical of mediterranean climates, has a maximum monthly average temperature of 22.9°C (WorldClim global climate database, 1950–2000; *available online*).⁴ The sandy soils and limited, seasonal rainfall at the study site make it likely that soil resources limit plant growth and/or survival. Although cones of Monterey pine are serotinous, they often open during warm, dry seasons in the absence of fires. Thus, seedling establishment is common in the understory of this forest and in the other native Monterey pine stands in California (Vogl et al. 1977, McDonald and Laacke 1990).

Experimental setup

Experiment 1 compared performance of *Pinus radiata* seedlings in two treatments, CMNs (common mycorrhizal networks) only ($n = 100$ plots) and no CMNs ($n = 100$ plots) to estimate the effects of CMNs alone on seedlings, without overstory roots. The CMNs only treatment used heavy-duty PVC cylinders (15 cm internal diameter, 20 cm deep, sealed on the bottom with a PVC disk) bored to 50% openness and fitted with 44-μm stainless-steel mesh screens to exclude overstory tree roots while allowing colonization by mycorrhizal fungal networks associated with overstory trees (see Plate 1). The no CMNs treatment utilized a 0.5 m diameter, conical, slit trench around identical cylinders, excluding both mycorrhizal networks and roots. Experiment 2, conducted simultaneously and randomly interspersed with Experiment 1, compared seedling performance in two treatments to estimate the net effect of overstory tree roots and CMNs on seedlings: no roots and no CMNs ($n = 50$ plots), which utilized a 0.5 m diameter, conical, slit trench cut around seedlings planted directly in the forest floor, and roots and CMNs ($n = 100$ plots), which included non-trenched seedlings. Whereas in the earlier study these four treatments were compared altogether, here we divide them into two separate experiments to better delineate the effects of CMNs alone (Experiment 1) from the net effects of CMNs and overstory roots together (Experiment 2). Details about implementation of these four treatments are as described by Booth (2004), except that in Experiment 1, we allowed mycorrhizal networks to colonize experimental cylinders for one full year before transplanting seedlings into them. At that point, ectomycorrhizal sporocarps were observed fruiting in the interior of multiple cylinders, and a destructively harvested subset of cylinders revealed substantial ingrowth of fungal mycelium from the surrounding soil.

Important advantages of this experimental design include (1) the planting of seedlings into intact forest soil cores to minimize colonization of networked seedlings by ruderal fungi, (2) thorough recolonization of soil cores by overstory CMNs before planting and trenching, and (3) a constant mesh size across networking and non-networking treatments. This last feature allowed the mycorrhizal fungi of nonnetworked seedlings to explore relatively unrestricted volumes of soil.

Conical slit trenches, which were re-cut every six weeks during the course of the experiment, were used instead of an alternative “rotated-core” method that has been used recently to sever mycorrhizal networks in physiology studies (Johnson et al. 2002), in order to allow nonnetworked seedlings to maintain intact mycorrhizal mycelia (Booth 2004). Although trenching has been shown in some previous studies to increase mineral and water resources available to trenched seedlings beyond what would be demanded by excluded competitors, in this experiment any mobilization of resources due to trenching would lead to a more conservative

⁴ <http://www.worldclim.org/>

estimate of the effect of mycorrhizal networking and its importance relative to root competition.

Seeds for the experiment were collected as open-pollinated families from three different native source populations of Monterey pine, including our study site at the Swanton Pacific Ranch in Santa Cruz County, California ($n = 4$ open-pollinated families); a site in San Luis Obispo County, California (the University of California's Kenneth S. Norris Ranch Marino Reserve; $35^{\circ}32'15''$ N, $121^{\circ}04'59''$ W; $n = 4$ open-pollinated families); and a site on Guadalupe Island, Mexico ($29^{\circ}9'36''$ N, $118^{\circ}17'60''$ W; $n = 3$ open-pollinated families). Seeds were germinated in sterile peat and grown in the laboratory for twelve weeks, before seedlings of approximately uniform size were chosen and planted into the field experiment in a non-mycorrhizal condition. Replicate plots were distributed among 12 experimental zones ($25\text{--}50\text{ m}^2$ in size) and were completely randomized with respect to treatment, with at least 1 m of space between plots. During the first growing season, seedlings were protected from herbivory by an open-topped cylindrical mesh (6 mm) cage. Herbaceous vegetation was removed from each plot by clipping twice during each growing season.

Data collection

After out-planting non-mycorrhizal seedlings into each plot in January 2006, we assessed height growth and survival of the seedlings for two years, including two growing seasons (~January–June) and two dry seasons (~July–December). During one dry season (October 2006) and one growing season (February 2007) we estimated maximum net photosynthetic rates of seedlings at saturating irradiance in Experiment 1 using a portable photosynthesis system (LI-COR-6400; LI-COR, Lincoln, Nebraska, USA). At harvest (December 2007), seedlings were removed from the field plots, their roots washed free of soil, and roots examined for mycorrhizal colonization. As is typical in pine forests, virtually all root tips of every networked and nonnetworked seedling were observed to be ectomycorrhizal with the exception of newly formed tips. Aboveground biomass (both experiments) and belowground biomass (Experiment 1 only) were dried at 55°C for 48 hours and then weighed.

To test the hypothesis that CMNs reduced seedling water limitation, we compared foliar $\delta^{13}\text{C}$ of networked and nonnetworked seedlings in Experiment 1. Foliar $\delta^{13}\text{C}$ provides a reliable measure of plant water use (stomatal conductance), relative to the net amount of carbon fixed in photosynthesis, integrated over leaf lifespans (Farquhar et al. 1989). Subsamples of the dried leaf tissue were finely pulverized in a ball mill (Mini Beadbeater 96; Biospec Products, Bartlesville, Oklahoma, USA) for foliar tissue chemistry (percentage N and $\delta^{13}\text{C}$) analyses, which were conducted by the University of California, Davis Stable Isotope Facility, using a PDZ Europa ANCA-GSL elemental analyzer interfaced

to a PDZ Europa 20-20 isotope ratio mass spectrometer (Sercon, Cheshire, UK).

Data analysis

Separate statistical analyses were used to compare the effects of treatments in the two experiments, since the two experiments contained the only two pairwise treatment comparisons of interest (Experiment 1, CMNs only vs. no CMNs; Experiment 2, no roots/no CMNs vs. roots and CMNs), using $\alpha = 0.05$ for determination of significance of effects. Final seedling aboveground biomass, total biomass, root:shoot ratio, foliar %N, foliar $\delta^{13}\text{C}$, and maximum net photosynthetic rate were compared between treatments using restricted maximum likelihood analysis of mixed models (PROC MIXED in SAS version 9.1 [SAS Institute 2003]) in which treatment, plant population, zone, and the treatment \times plant population interaction were fixed factors, and plant open-pollinated family nested within plant population was a random factor. To account for the possibility that plant $\delta^{13}\text{C}$ differences between treatments in Experiment 1 were associated with changes in root:shoot allocation, we included final root:shoot ratio as a covariate in the initial statistical model for $\delta^{13}\text{C}$ as a response variable. Final seedling survivorship was compared between treatments using logistic regression models (PROC LOGISTIC in SAS [SAS Institute 2003]) in which treatment, plant population, zone, and the treatment \times plant population interaction were categorical explanatory variables. Seedling height growth was compared between treatments using a mixed-model repeated-measures analysis (PROC MIXED in SAS), with treatment, plant population, zone, and the treatment \times plant population interaction as fixed between-subjects factors, and day and the treatment \times day interaction as within-subjects factors. When the treatment \times day interaction was found to be significant, treatment effects on plant height were tested within each sampling date using the SLICE option within SAS PROC MIXED. All analyses in SAS PROC MIXED assumed normality of residuals, which was confirmed by inspection of histograms of residuals from each analysis. The effect of overstory tree roots on seedling survival (separate from the CMN effect) was calculated from the results of Experiments 1 and 2 as described in Appendix B.

RESULTS

Experiment 1

In Experiment 1, which compared the growth and survival of pine seedlings that were allowed to join overstory common mycorrhizal networks (CMNs) with seedlings that were prevented from joining these networks, networked seedlings survived significantly better throughout the experiment and, at the time of harvest, had 56% higher survivorship than nonnetworked seedlings ($P = 0.0036$; Fig. 1c). Networked and nonnetworked seedlings had nearly identical height growth ($P = 0.824$; Fig. 1a), and at the end of the

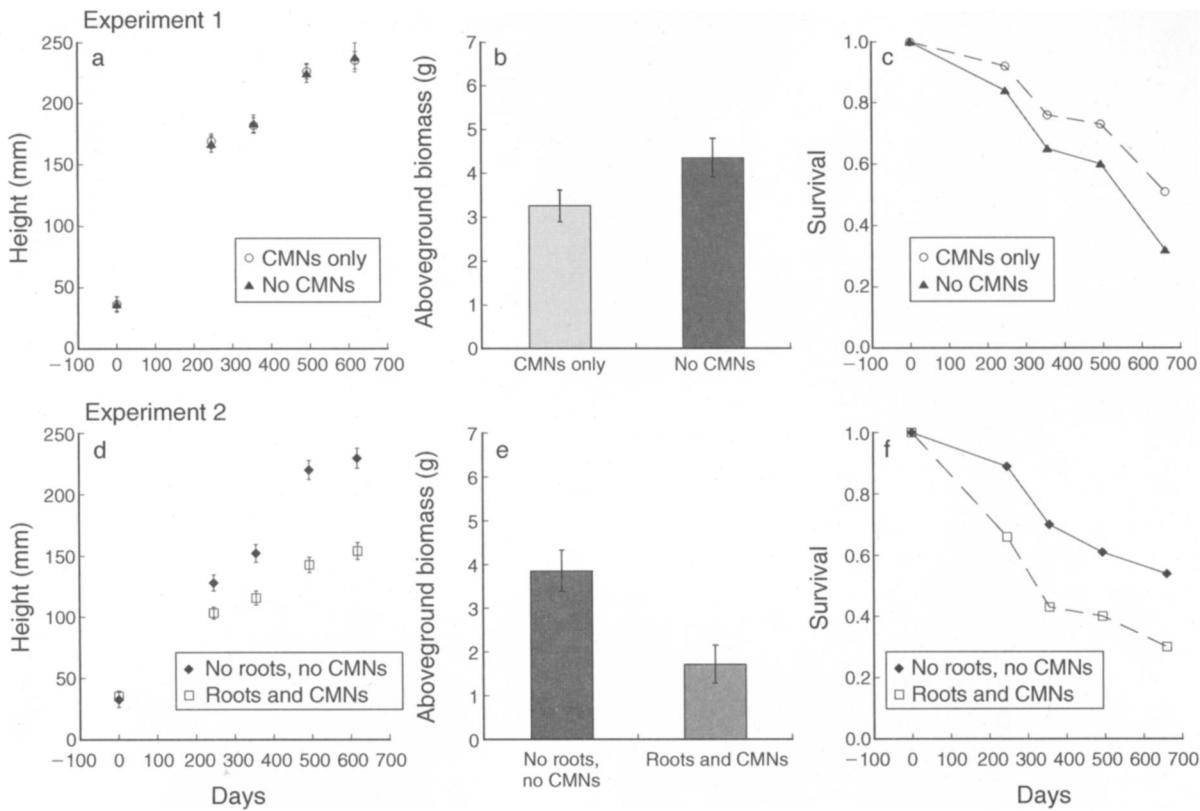


FIG. 1. Performance of Monterey pine (*Pinus radiata*) seedlings in two field experiments in Monterey pine forest (California, USA). Data are means \pm SE. The disjunction between positive effects of common mycorrhizal networks (CMNs) on survival and non-effects on growth and biomass are typical of low-productivity systems that experience periodic pulses of soil moisture. Seedling performance measures tested were (a, d) seedling height growth, (b, e) final aboveground biomass, and (c, f) survival. The net negative effects of roots on seedling growth and survival (as shown in Experiment 2) reveal the greater influence of root competition vs. facilitation by CMNs in this system. Height and survivorship were first measured in February 2006, shortly after planting in January, and were measured subsequently in October 2006, February 2007, July 2007, and November 2007, before harvest in December 2007. At the start of Experiment 1, $n = 97$ for “CMNs only” and $n = 94$ for “no CMNs.” At the simultaneous start of Experiment 2, $n = 44$ for “no roots and no CMNs” and $n = 104$ for “roots and CMNs.”

second year, networked and nonnetworked seedlings had similar aboveground biomass ($P = 0.250$; Fig. 1b) and total biomass ($P = 0.431$). The final ratio of root biomass to shoot biomass was 39% greater for networked seedlings than nonnetworked seedlings ($P = 0.007$, Fig. 2d). Networked seedlings were 0.58% depleted in ^{13}C relative to nonnetworked seedlings ($P = 0.005$; Fig. 2b), and the foliar N concentrations ($P = 0.914$; Fig. 2a) and maximum rates of photosynthesis in these two treatments were approximately equal (2006 dry season $P = 0.9955$; 2007 wet season $P = 0.7218$; Fig. 2c). No measured variables in Experiment 1 differed significantly among seedling source populations, or were influenced by an interaction between treatment and source population. Full results of all statistical analyses can be found in Appendix C.

Experiment 2

In Experiment 2, which assessed the net effect of overstory roots and CMNs together on seedling growth

and survival, we compared seedlings that were planted in the natural forest floor with others that were planted inside conical slit trenches where they were isolated from both overstory roots and mycorrhizal networks. After two years, seedlings grown in direct contact with overstory tree roots and mycorrhizas (non-trenched seedlings) had 34% less height growth ($P < 0.0001$; Fig. 1d) and 69% less aboveground biomass ($P = 0.0002$; Fig. 1e) than those that were isolated from both overstory roots and CMNs. Non-trenched seedlings also had 44% lower survivorship ($P = 0.0029$; Fig. 1f). Although height growth and aboveground biomass differed significantly among seedling source populations (Appendix C), no measured variables in Experiment 2 were significantly influenced by an interaction between treatment- and source population. Assuming that the separate effects of roots and CMNs on seedling survival were additive, and using both the CMN effect observed in Experiment 1 along with the net effect of roots and CMNs together in Experiment 2, we calculated that the

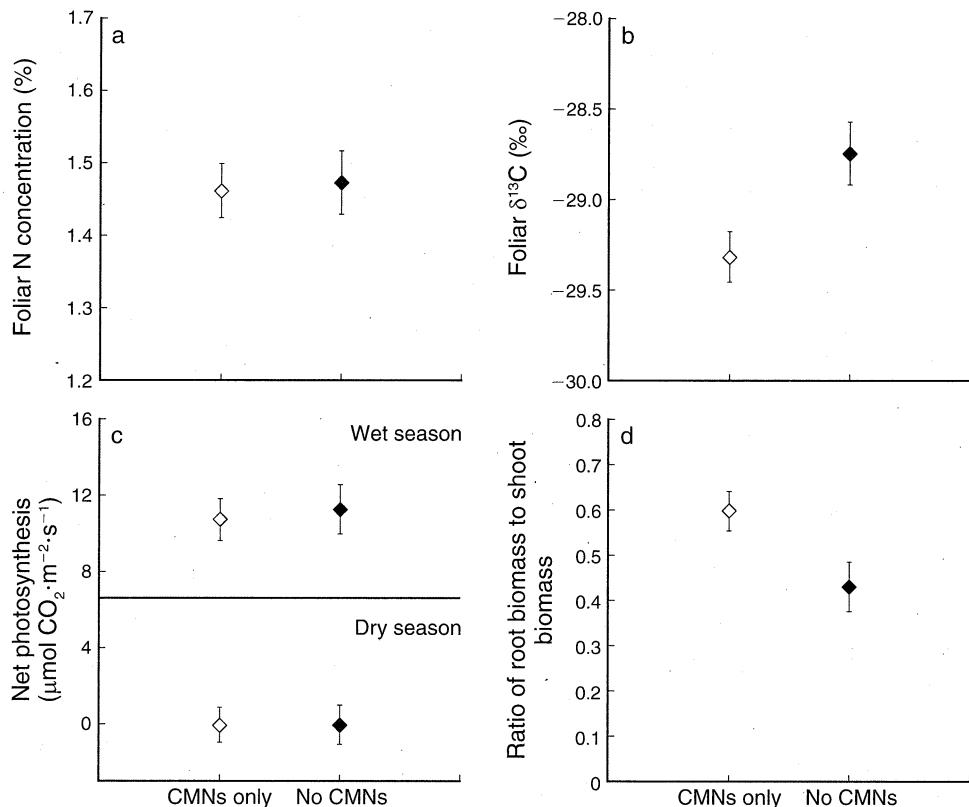


FIG. 2. Foliar chemistry, photosynthesis, and root : shoot partitioning of Monterey pine (*Pinus radiata*) seedlings in Experiment 1. Data are means \pm SE. The lack of differences in foliar nitrogen and photosynthesis between the treatments, along with significant depletion of foliar ^{13}C in the CMN treatment, suggests that CMNs provided seedlings with additional water and not with additional N or overstory C. (a) Final foliar N concentration (CMNs only, $n = 40$; no CMNs, $n = 25$). (b) Final foliar $\delta^{13}\text{C}$ (CMNs only, $n = 40$; no CMNs, $n = 25$). (c) Net maximum photosynthetic rate in a dry season, October 2006 (CMNs only, $n = 66$; no CMNs, $n = 59$) and a growing (wet) season, February 2007 (CMNs only, $n = 58$; no CMNs, $n = 54$). (d) Final ratio of root biomass to shoot biomass (CMNs only, $n = 44$; no CMNs, $n = 25$).

effect of roots alone on seedling survival in Experiment 2 was -65% (Appendix B).

DISCUSSION

These results show that overstory trees influence understory seedlings through multiple, simultaneous belowground pathways: negatively through resource competition and positively by the provision of established mycorrhizal networks (Appendix D). An earlier study, in which we developed our experimental approach, showed positive growth effects of common mycorrhizal networks (CMNs) on *Pinus strobus* seedlings over a growing season but did not continue long enough for any of the seedlings to experience mortality (Booth 2004). The present study revealed large, positive effects of CMNs on survival of Monterey pine seedlings (Fig. 1c). Mycorrhizal network effects did not, however, vary among three native-source populations of the seedlings, suggesting that the considerable genetic differentiation that has evolved among these populations (Wu et al. 1998, Rogers 2004) has not increased the ability of local seedling genotypes at our study site to benefit from access to CMNs.

The magnitude of the observed CMN effect on seedling survival, especially relative to the negative effect of overstory competition, suggests that CMNs between trees and seedlings may have a large influence on the age structure and rate of overstory replacement in this forest (Shugart 1984, Oliver and Larson 1996). This possibility highlights a key uncertainty in our understanding of how natural forests regenerate. Estimates of seedling shade tolerance, light availability, and soil resource supply may be insufficient to predict the outcomes of plant interactions in forests, and thus the short-term survival and long-term importance of tree seedlings growing in forest understories. Given the ubiquity of mycorrhizal network connections among trees in forest ecosystems (Newman 1988, Simard and Durall 2004, Selosse et al. 2006), the degree to which different seedlings are compatible with and benefit from CMNs may explain a large amount of variation in the establishment of different plant species in forests.

The contrast between the large, positive effect of CMNs on seedling survival (+56%) and the lack of a CMN effect on seedling height growth and biomass in this study is consistent with the disjunction of growth



PLATE 1. (A) A field assistant hauls networking cylinders up to the Monterey pine forest stand in Santa Cruz County, California, USA; (B) a cylinder laid out on the forest floor before installation; (C) a cylinder installed with an intact soil core; (D) a networked pine seedling growing with a sporocarp in a cylinder. Photo credits: M. G. Booth and J. D. Hoeksema.

and survival observed in low-productivity ecosystems that experience periodic pulses of soil water (Noy-Meir 1973). In such systems, growth typically occurs when water availability is high and uniformly distributed. Mortality occurs principally when soil water is less available and relatively patchy. Thus, the traits and conditions that favor plant growth may not be the same as those that favor plant survival, and ecological dynamics during the dry season, not the growing season, may be most important for species persistence (Goldberg and Novoplansky 1997). As in other mediterranean ecosystems characterized by alternating cool-wet and warm-dry seasons, seedling survival in native Monterey pine forests is likely water limited and not nitrogen (N) limited (Zavala et al. 2000). This assumption is supported by the observation that seedlings in our

experiments generally had higher growth rates during wet months and higher mortality rates during dry months (Fig. 1). Finally, the observations that networked seedlings were 0.58% depleted in ^{13}C relative to nonnetworked seedlings (Fig. 2b), and that maximum rates of photosynthesis between the treatments were approximately equal (Fig. 2c), suggest that networked pines received and transpired more water than nonnetworked pines (Farquhar et al. 1989, Fotelli et al. 2003, Querejeta et al. 2006). The higher allocation to roots by networked seedlings (Fig. 2d) also supports the notion that networked pines were better able to acquire water during the dry seasons (Hutchings and Dekroon 1994). Dry season (July to December) precipitation was below average in both years of our study (2006, 137 mm; 2007, 126 mm; 1898–2007 average = 185 mm, SD = 86

mm; National Oceanic and Atmospheric Administration, Central Coast Drainage Climate Division), and it is possible that CMNs would be less important for seedling survival in above-average rainfall years. The observed difference in $\delta^{13}\text{C}$ between networked and nonnetworked seedlings (Fig. 2b) is the opposite of the pattern expected if the leaves of networked seedlings contained a substantial proportion of carbon transferred through the CMN from overstory trees (Högberg et al. 1999).

There are at least two hypothetical mechanisms by which CMNs could reduce seedling water limitation and increase seedling survival in this system. Previous studies, including one in a monodominant pine forest (Warren et al. 2008), have shown that mycorrhizal mycelia can act as conduits of water lifted hydraulically from deep soil layers by their host plants, distributing it evenly throughout their masses to multiple plants connected to the same CMN (Egerton-Warburton et al. 2007, Warren et al. 2008). Such redistribution within a CMN could deliver water to seedlings that otherwise would not have access to it. An alternative explanation of the benefits of CMNs for seedlings might be found in the types of mycorrhizal fungi that colonize networked and nonnetworked seedlings. Seedlings that have been isolated from tree roots in forest understories (i.e., nonnetworked seedlings) have been shown to host species of mycorrhizal fungi that are common in more disturbed environments, while networked seedlings host mostly fungi that associate with their overstories (Deacon and Fleming 1992, Jones et al. 2003). Little is known of the relative costs and benefits to seedlings of associating with particular species or groups of fungi that typify early and late-successional environments. Nevertheless, the life-history traits that cause some fungi to associate exclusively with networked or nonnetworked seedlings may influence the terms of resource exchange in symbiosis. By either mechanism, greater profitability of the mycorrhizal symbioses in CMNs might cause seedling hosts to increase belowground C allocation as networked pines did in this study.

Prior to the results presented here, five other field manipulation experiments of which we are aware compared seedling performance among different belowground treatments designed to estimate the effects of established CMNs on tree seedlings (Booth 2004, Kranabetter 2005, McGuire 2007, Teste and Simard 2008, Teste et al. 2009). The results of these experiments varied widely, but it is not clear whether this variation resulted from inherent differences in effects of CMNs among different ecosystems, or from differences in methodology used among the five studies. For example, McGuire (2007) found that seedlings of *Dicymbium corymbosum* growing in the shaded understory of a monodominant rain forest grew larger and survived better when they had access to CMNs alone (without overstory roots) in 20- μm mesh pots compared to when they interacted with neither overstory roots nor CMNs in 0.45- μm mesh pots. Such results are consistent with

the findings reported here for survival and by Booth (2004) for seedling growth and survival. The experiment by Booth (2004) had the unique feature of showing that arbuscular-mycorrhizal (AM) seedlings not compatible with a common ectomycorrhizal (EM) network did not benefit from the CMN, while EM seedlings did benefit from it.

In contrast to these results, Kranabetter (2005) found no difference in performance of hybrid spruce seedlings grown in a shaded forest understory with or without access to CMNs in the absence of roots (250- μm vs. 4- μm mesh cylinders). In that study, experimental cylinders had open bottoms at 20 cm depth, making it likely that both CMNs and overstory roots were present in both treatments, thereby masking distinct CMN effects. Teste and Simard (2008) and Teste et al. (2009) also found no difference in *Pseudotsuga menziesii* seedling performance between treatments with no CMNs or overstory roots (0.5- μm mesh bags) compared to treatments with CMNs only (35- μm and 250- μm mesh bags). These authors did, however, find increases in seedling performance between treatments with both roots and CMNs (no mesh barrier) vs. treatments with no roots or CMNs (0.5- μm mesh bags), suggesting that tree roots, themselves, may have facilitated seedlings independently of CMNs.

One explanation for the different results in the latter two studies and others might be variation in the density of overstory trees. The experiment described here and the experiments of Booth (2004) and McGuire (2007) were set in the understories of mature forests in which light levels were low and in which root density and the mass of mycorrhizal networks available to seedlings were probably high. Experiments performed by Teste and colleagues were conducted in recently logged stands that retained a low density of either mature trees (Teste and Simard 2008) or advanced regeneration saplings (Teste et al. 2009). Under these conditions, where light was abundant and overstory demand for soil resources was presumably less, established vegetation may have facilitated seedlings directly by hydraulic lift of water (Caldwell et al. 1998, Horton and Hart 1998) or by protecting seedlings otherwise from resource limitation or harsh exposure. Indeed, Schoonmaker et al. (2007) estimated that hydraulic redistribution from retention trees in similarly logged *Pseudotsuga menziesii* stands accounted for 21.6% of seedling water, and, the authors noted, there was no difference in the amount of water distributed to seedlings via soil pathways alone and via soil pathways combined with a CMN pathway. Differences in the conditions and results of CMN manipulation experiments suggest that the role of CMNs in mediating overstory-understory interactions may be greater and/or involve different mechanisms depending on the structure of the established cohort.

Ecologists now recognize the importance of incorporating facilitation into general theories of how species interactions contribute to the organization of commu-

nities (Callaway and Walker 1997, Bruno et al. 2003). In plant ecology, specific hypotheses have been proposed that include the influences of simultaneous positive and negative interactions on community dynamics (Dickie et al. 2002, 2005, Lortie et al. 2004, Brooker et al. 2008, Maestre et al. 2009). Our results show that forest trees can influence subsequent cohorts negatively, through belowground competition, and positively by the provision of mycorrhizal networks. The size of the positive CMN effect on seedling survival in this study, compared to the net negative effect of tree roots, suggests CMNs influence the trajectory and rate of stand development in this forest. We suggest that, as plant ecologists refine and test theories of species interactions and community assembly, they consider the potential of common mycorrhizal networks as agents of facilitation between established vegetation and later generations.

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APPENDIX A

Summary of soil characteristics at the study site in native Monterey pine forest at Swanton Pacific Ranch, Santa Cruz County, California, USA (*Ecological Archives* E091-163-A1).

APPENDIX B

Description of how the separate effects of roots vs. common mycorrhizal networks (CMNs) on seedling survival were calculated, using data from both Experiment 1 and Experiment 2 (*Ecological Archives* E091-163-A2).

APPENDIX C

Full results of all statistical analyses (*Ecological Archives* E091-163-A3).

APPENDIX D

Conceptual model for the simultaneous belowground mechanisms by which overstory trees influence understory seedlings in forests (*Ecological Archives* E091-163-A4).