DOES THE PRESENCE OF *WYETHIA MOLLIS* AFFECT GROWTH OF *PINUS JEFFREYI* SEEDLINGS?

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ABSTRACT.—Regeneration of *Pinus jeffreyi* in the Sierra Nevada is often limited on sites dominated by *Wyethia mollis*. Allelopathic chemicals and competition for soil moisture have been suggested as possible mechanisms for limiting regeneration. We tested the hypothesis that soil chemical and microbial properties from sites in different stages of succession influence seedling growth of *Pinus jeffreyi*. Soil was collected from an early-seral site dominated by *Wyethia mollis*, a mid-seral site dominated by the shrubs *Arctostaphylos patula*, *Ceanothus prostratus*, *C. velutinus*, and *Purshia tridentata*, and a late-seral site dominated by mature *Pinus*. These sites were compared for nutrient content, *Pinus* seedling growth capacity, and microbial population size. Soil (0–33 cm) from the early-seral site had the lowest C, microbial biomass, and fungal and bacterial populations. There were no consistent trends in soil nutrient content among sites. The early-seral site had the lowest soil Ca and Mg contents but also had a lower C/N ratio and more than twofold greater P content than either the mid- or late-seral site. *Pinus* seedling growth and foliar nutrient concentrations were compared at 3 harvest dates (220, 314, and 417 days after germination) in a greenhouse bioassay. The treatment design was a 3 × 2 factorial with soil from each of the 3 sites either with or without *Pinus* seedlings. Pots without seedlings were used as controls to assess the effects of seedlings on microbial biomass. Seedling growth in the early-seral soil was initially suppressed in comparison to growth in the mid-seral soil, but by the final harvest total seedling weight was similar between these 2 treatments. The most obvious treatment effect was a reduction in growth for seedlings planted in late-seral soil, probably due to a nutrient imbalance in the soil. Seedlings grown in late-seral soil had Fe and Al levels that were nearly twice those of seedlings grown in early- and mid-seral soils. Microbial biomass followed a temporal pattern similar to that found for seedling growth. Differences in microbial biomass between the early- and mid-seral soils, although initially large, were not detected by the final harvest. We interpret these results to indicate that allelopathy or soil nutrient deficiencies resulting from the presence of *Wyethia* are unlikely to be responsible for limited growth of *Pinus* seedlings in *Wyethia*-dominated stands.

Key words: allelopathy, Wyethia mollis, Pinus jeffreyi, succession, forest regeneration, seedling growth, foliar nutrients, soil chemical, soil microbial biomass.

In the eastern slopes of the Sierra Nevada, regeneration of *Pinus jeffreyi* is often limited on sites dominated by *Wyethia mollis* (Yoder-Williams and Parker 1987, Parker and Yoder-Williams 1989, Williams 1995). *Wyethia* occurs as an understory species of pine forests (*P. jeffreyi, P. ponderosa,* and *P. washoensis*) east of the Cascade Range from south central Oregon and northeastern California through the eastern Sierra Nevada of California and Nevada (Hopkins 1979, Riegel 1982, Smith 1994, Sawyer and Keeler-Wolf 1995), as an associate species in the montane and subalpine *Artemisia* steppe throughout this range (Cronquist 1994), and as isolated populations in central Nevada (Kartesz 1988). An herbaceous, disturbance-adapted, long-lived perennial (>50 years) with a deep taproot and fleshy resinous foliage, *Wyethia* was most likely an early successional plant with low frequency and cover prior to European arrival (Young and Evans 1979, Williams 1995). Effective suppression and exclusion of fire since the early 1930s have resulted in increased fuel loading and higher forest stand densities (Agee 1993). As a result, wildfires now burn with greater severity and intensity than the frequent, presuppression fires (3–20 years) and provide *Wyethia* with a competitive edge for expansion and dominance (Rundel et al. 1977, Agee 1993, Barbour and Minnich 2000). Heavy livestock grazing of palatable plants from 1860 to the 1930s, primarily by sheep, also resulted in increased *Wyethia* abundance (Coville 1898, Kennedy and Doten 1901, Leiberg 1902, USDA Forest Service 1937, Olmstead 1957). By the

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end of the 19th century, Coville (1897) described *Wyethia* as being "abundant" in south central Oregon where the Klamath Indians had traditionally used the mashed roots as a poultice to control swelling. *Wyethia* continues to dominate sites in Lassen National Park that have been excluded from livestock grazing for more than 80 years (Oswald et al. 1995).

Though there are many pathways succession can take following disturbance in pine forests, a generalized summary includes (1) early seral: *Wyethia* dominance with associated herbaceous species and some conifer regeneration; (2) mid-seral: *Arctostaphylos patula, Ceanothus prostratus, C. velutinus,* and *Purshia tridentata* shrub dominance with pole-sized conifers; (3) and late seral: conifer dominance with some shade-tolerant herbs and shrubs. *Wyethia*-dominated sites may suppress succession to mid-seral conifer regeneration for up to 100 years (Kennedy and Doten 1901, Evanko 1951, Parker and Yoder-Williams 1989) and can persist in the understory of mid- and late-seral *Pinus jeffreyi* overstories as an infrequent, low-cover (<5%) associate (Smith 1994). Shrub dominance in the mid-seral phase should increase the soil nutrient capital with time between fire and logging disturbance intervals that temporarily reduce the cover of N-fixing shrubs, *Ceanothus* and *Purshia* (Conard et al. 1985, Johnson 1995, Busse et al. 1996, Busse 2000a, 2000b).

Parker and Yoder-Williams (1989) proposed 2 hypotheses to explain the dominance of *Wyethia:* (1) allelopathy and (2) competition for water. However, the precise mechanism(s) responsible for poor regeneration on sites dominated by *Wyethia* remains unclear. After a forest is removed through fire or logging, the developing shrub and herbaceous understory often suppresses conifer regeneration as a result of competition for soil moisture (Conard and Radosevich 1981, 1982, McDonald 1983a, Lanini and Radosevich 1986, Shainsky and Radosevich 1986, Parker and Yoder-Williams 1989). Water-extractable allelochemicals from *Wyethia* have also been suggested as an additional factor suppressing *Pinus jeffreyi* regeneration (Heisey and Delwiche 1983, Yoder-Williams and Parker 1987, Parker and Yoder-Williams 1989, Williams 1995). Yoder-Williams and Parker (1987) hypothesized that *Wyethia*

tissue leachates exhibited an allelopathic effect on *Pinus* seedlings. In a field bioassay *Wyethia* litter inhibited germination and reduced radicle elongation of *Pinus* seedlings. Allelopathy is thought to depress germination and early growth by inhibiting cell division and elongation, disrupting membrane regulation, nutrient uptake, respiration, and photosynthesis (Rice 1984). However, allelopathy can be difficult to demonstrate and remains controversial (Harper 1977, Radosevich and Holt 1984, Kelsey and Everett 1995, Dakshini et al. 1999). Depression of biomass production may be caused by leaf extracts other than allelochemicals. For example, plant litter high in labile C, particularly carbohydrates, may enhance soil microbial growth and result in increased nutrient immobilization and decreased plant growth (Michelsen et al. 1995).

In this study we tested the hypothesis that *Pinus jeffreyi* seedling growth is limited due to the presence of *Wyethia* in early-seral sites. Soils and litter were collected from early-seral (*Wyethia*-dominated), mid-seral (shrub-dominated), and late-seral (depauperate understory with mature *Pinus* overstory) sites and used as growth media for greenhouse-grown *Pinus* seedlings.

METHODS

Study Area

The study area (39°53'N, 120°25'W) is 9.7 km northeast of Beckwourth, Plumas County, in the northern Sierra Nevada of California. Precipitation at nearby Clover Valley averaged 903 mm per year ($s = 342$) from 1959 through 1991, approximately 85% of which was in the form of snow falling in the months of November through March (State of California, Resources Agency, Department of Water Resources, personal communication, 1993). Soils are loamy-skeletal, mixed, frigid Ultic Agrixerolls (Sattley Family). They are moderately to well drained and formed from weathered andesitic tuff breccia (USDA Forest Service 1988). Horizon development includes a gravelly loam A1 (0–7.6 cm), a very cobbly loam A2 (7.6–20.3 cm), a cobbly loam Bt1 (20.3–33.0 cm), and a very cobbly clay loam Bt2 (33.0–117 cm) above bedrock.

We selected 3 sites within a *Pinus jeffreyi* forest type (Sawyer and Keeler-Wolf 1995): (1) early-seral site with *Wyethia*-dominated

understory, (2) mid-seral, shrub-dominated stand, and (3) late-seral, *Pinus*-dominated stand. The sites have similar elevation (1829 m) and slope (0–3%), and are within 2.4 km of each other in Crocker Creek drainage. *Wyethia* is found in or adjacent to every site with a *Pinus* overstory. The early-seral site is dominated by *Wyethia* with scattered *Artemisia tridentata* ssp. *vaseyana, Lithophragma parviflorum, Collinsia parviflora,* and *Paeonia brownii;* the mid-seral shrub site is dominated by *Arctostaphylos patula, Ceanothus prostratus, C. velutinus, Purshia tridentata, Elymus elymoides (= Sitanion hystrix), Poa fendleriana,* and *Lupinus* spp.; and the late-seral site has a very depauperate understory with scattered *Symphoricarpos rotundifolius, Rosa woodsii, Hydrophyllum capitatum, Vicia americana,* and *Sarcodes sanguinea,* an obligate mycotroph. *Pinus* dominates the overstory at all sites with increasing amounts of *Calocedrus decurrens* and *Abies concolor* found at the mid- and late-seral sites. Vascular plant nomenclature and taxonomy follow Hickman (1993).

Pinus diameter and age distribution by site within a 0.10-ha plot were as follows: early-seral mean DBH = 41.6 cm, $s_{\bar{x}}$ = 6.93 cm, age <50 years (n = 9); mid-seral mean DBH = 59.0 cm, $s_{\bar{x}}$ = 6.55 cm, age 60–100 years (n = 9); and late-seral mean DBH = 88.3 cm, $s_{\bar{x}}$ = 16.95 cm, and age 100–226 years (n = 4). Ocular estimates of *Wyethia* canopy cover by site within the 0.10-ha plot were early-seral 40%, mid-seral 10%, and late-seral 5%.

Soil Properties

Four replicate soil samples were collected randomly at each site on 29 May 1990 from the A1, A2, and Bt1 horizons, transported to greenhouses in Reno, Nevada, and sieved (<2 mm) prior to nutrient analyses. Organic matter was determined by weight loss on ignition (Nelson and Sommers 1982) and converted to organic C by dividing weight loss by 1.742. The maximum furnace temperature was 400°C to avoid loss of clay-bound H_2O. Total N was determined with a continuous flow analyzer following Kjeldahl digestion (Bremner and Mulvaney 1982). Extractable P was determined by dilute acid fluoride extraction (Olsen and Sommers 1982). Microbial biomass was determined by the chloroform-fumigation-incubation method (Jenkinson and Powlson 1976). Additional soil samples (triplicate) were col-

lected from the A1 horizon at each site on 8 June 1990, and culturable bacteria (grown on 1/10 strength tryptic soy agar) and fungi (grown on malt extract agar) were enumerated within 48 hours of collection.

Greenhouse Experiment

The experiment was a completely randomized block design with 6 treatments, 3 harvest dates, and 10 replications. Treatment design was a 3 × 2 factorial, with soil from the 3 sites in combination with 2 vegetation treatments: (1) soil from early-seral, *Wyethia*-dominated site + *Pinus;* (2) soil from mid-seral, shrub-dominated site + *Pinus;* (3) soil from late-seral, *Pinus*-dominated site + *Pinus;* (4) soil from *Wyethia*-dominated site without *Pinus;* (5) soil from mid-seral, shrub-dominated site without *Pinus;* and (6) soil from late-seral, *Pinus*-dominated site without *Pinus.* Treatments receiving no seedlings were included as controls to determine whether *Pinus* root proliferation counteracts the effect of *Wyethia* allelochemicals on soil biota. To avoid potential microclimatic effects, we placed the pots on the greenhouse bench and randomly assigned them to each replication by block.

The 4 replicate A1 horizons were bulked by site (early-, mid-, and late-seral), and 4 *Pinus jeffreyi* seeds were placed in each pot filled on 19 June 1990. *Pinus* seeds, which were collected in the Toiyabe National Forest at 1982 m in 1988, were stored dry (8% humidity) prior to a 30-day stratification at 1°C (R. Walker, University of Nevada, Reno). Pot volume was 2625 cm^3 and height was 15 cm.

Pots were watered until the first sign of drainage: daily the 1st week, every other day the 2nd week, and twice weekly thereafter. Plants were thinned to 2 per pot on day 56 (13 August) and eventually to 1 per pot on day 71 (28 August).

We collected current-year litter from each site, and 10 g (dry weight equivalent which approximated the volume/weight ratio found in the field) was placed on the soil surface in each pot on day 71 (28 August) after germination. Litter at each site varied in species composition and spatial distribution. *Wyethia* litter was collected from the early-seral site; *Arctostaphylos, Ceanothus velutinus,* and *Purshia* litter from the mid-seral site; and *Pinus* litter from the late-seral site. Throughout the experiment we added new litter monthly (~2.5 g) to

TABLE 1. Selected chemical and microbial properties of mineral soil from the early-, mid-, and late-seral field sites. error ($n = 4$) except for viable counts ($n = 3$). Different lowercase letters within a column and horizon indicate signifi-

Horizon	Site	pH	C	N	C/N	P	K	Ca	Mg
			----- g kg⁻¹ -----			mg kg⁻¹	--------- cmol$_c$ kg⁻¹ -------		
A1	Early-seral	6.3 (0.1)[a]	33 (2)[b]	1.7 (0.1)[a]	19 (1)[b]	83 (11)[a]	1.8 (0.1)[a]	16 (1)[b]	3.0 (0.2)[c]
	Mid-seral	6.4 (0.2)[a]	47 (3)[a]	1.5 (0.2)[a]	34 (5)[a]	35 (8)[b]	2.3 (0.2)[a]	25 (1)[a]	4.8 (0.3)[b]
	Late-seral	6.5 (0.1)[a]	38 (4)[ab]	1.4 (0.2)[a]	27 (1)[ab]	33 (3)[b]	2.3 (0.2)[a]	18 (1)[b]	5.8 (0.2)[a]
A2	Early-seral	6.2 (0.1)[b]	28 (2)[a]	1.5 (0.1)[a]	19 (1)[c]	60 (12)[a]	1.9 (0.1)[a]	17 (1)[b]	2.9 (0.1)[c]
	Mid-seral	6.4 (0.1)[ab]	30 (2)[a]	0.8 (0.1)[c]	37 (1)[a]	17 (2)[b]	1.5 (0.1)[b]	22 (1)[a]	4.5 (0.3)[b]
	Late-seral	6.5 (0.2)[b]	31 (2)[a]	1.1 (0.1)[b]	29 (1)[b]	19 (2)[b]	2.0 (0.1)[a]	19 (2)[b]	5.4 (0.3)[a]
Bt1	Early-seral	6.2 (0.1)[a]	26 (1)[a]	1.2 (0.1)[a]	21 (1)[c]	26 (8)[a]	2.0 (0.2)[a]	18 (1)[b]	3.1 (0.1)[b]
	Mid-seral	6.3 (0.1)[a]	31 (3)[a]	0.7 (0.1)[b]	45 (2)[a]	12 (1)[b]	1.2 (0.1)[b]	22 (1)[a]	4.6 (0.2)[a]
	Late-seral	6.5 (0.1)[a]	34 (1)[a]	1.1 (0.1)[a]	31 (1)[b]	10 (4)[b]	1.9 (0.1)[a]	19 (1)[b]	5.2 (0.2)[a]

each pot. Plants were harvested on 25 January, 24 May, and 4 September 1991.

Mineral soil (50 g) was collected at each harvest and tested for microbial biomass. After washing soil from the *Pinus* seedlings, we separated them into aboveground and belowground components. The samples were dried at 60°C for 48 hours and weighed. Needles were collected from the aboveground fraction and analyzed for nutrient content. Nitrogen content was measured by Kjeldahl analysis (Bremner and Mulvaney 1982), and other nutrients were measured with an ICP spectrophotometer (Issac and Johnson 1985) using a nitric acid digest (Zarcinas et al. 1987).

Data were analyzed with the General Linear Models procedure in SAS (SAS Institute Inc. 1990). Mean separations were conducted with the Waller-Duncan test at the $P \leq 0.05$ level.

RESULTS

Soil Properties

Several differences in soil organic matter, nutrient status, and microbial characteristics were found between sites. The early-seral site had the shallowest mean O horizon depth (early-seral = 1.85 cm ($s_{\bar{x}} = 0.47$ cm); mid-seral = 3.78 cm ($s_{\bar{x}} = 0.72$ cm); late-seral = 4.30 cm ($s_{\bar{x}} = 0.71$ cm), in addition to the lowest C, Ca, and Mg contents at each mineral soil depth (Table 1). The trend of lower nutrient status at the *Wyethia*-dominated, early-seral site was not consistent, however. Phosphorus content was two- to threefold greater,

and C/N ratios were lower at the early-seral site compared to the mid- or late-seral sites. Microbial biomass and viable bacterial and fungal populations in the upper horizon were lowest at the early-seral site and highest at the shrub-dominated, mid-seral site. Despite a smaller biomass, the microbial population at the early-seral site had the highest C utilization efficiency among the 3 sites, as shown by lower qCO$_2$ values (CO$_2$ respired per unit biomass; Table 1).

Greenhouse Study

PLANT BIOMASS.—There were no significant differences in *Pinus* germination between treatments, as nearly 95% of all seeds germinated. *Pinus* seedlings accumulated more total mass when grown in soil from the mid-seral site than from the late-seral site, but were not greater in total mass than the early-seral, *Wyethia*-dominated site at the 1st harvest (Table 2). By the 2nd harvest the ranking for total plant mass was mid-seral > early-seral > late-seral. There was no difference in total weight of *Pinus* seedlings at the final harvest between mid-seral and early-seral treatments, but both were greater than the late-seral treatment. Root and shoot weights followed similar trends (Table 2).

FOLIAR NUTRIENT CONCENTRATION.—There were no significant differences between treatment sites in foliar N, P, K, or Na (Table 3). Micronutrients were generally found in lowest concentrations in foliage from the mid-seral treatment (Table 3). Seedlings grown in soil from the late-seral site had higher levels of

Depth of the upper horizons: A1 = 0–7.6 cm; A2 = 7.6–20 cm; Bt1 = 20–33 cm. Values in parentheses are standard cant differences ($P < 0.05$) between sites.

Microbial biomass	C_{micro}/C_{total}	qCO_2	Viable counts	
			Bacteria	Fungi
mg kg^{-1}	%		-------- $\times 10^6$ g^{-1} --------	
688 (76)[b]	2.1 (0.2)[a]	0.21 (0.03)[b]	9.6 (0.1)[c]	0.3 (0.1)[c]
1217 (93)[a]	2.6 (0.3)[a]	0.38 (0.02)[a]	18.3 (3.0)[a]	2.0 (0.3)[a]
888 (64)[b]	2.4 (0.2)[a]	0.35 (0.02)[a]	12.0 (0.1)[b]	1.3 (0.1)[b]
545 (98)[a]	1.9 (0.4)[a]	0.23 (0.01)[b]		
740 (90)[a]	2.3 (0.2)[a]	0.31 (0.06)[a]		
691 (41)[a]	2.1 (0.1)[a]	0.37 (0.03)[a]		
406 (86)[a]	1.6 (0.3)[a]	0.22 (0.02)[b]		
557 (81)[a]	1.9 (0.3)[a]	0.31 (0.03)[a]		
603 (17)[a]	1.8 (0.1)[a]	0.31 (0.08)[a]		

Mg, Fe, and Al than the other treatments. Iron and Al concentrations were nearly twice those of the early- and mid-seral treatments.

SOIL MICROBIAL BIOMASS.—The temporal response of microbial biomass was similar to the pattern for total seedling growth. Microbial biomass was greatest for the mid-seral treatment at the 1st and 2nd harvest (Table 4). By the final harvest, the early- and mid-seral treatments had comparable microbial biomass, and both were significantly greater than the late-seral treatment. Root proliferation had little effect on microbial biomass; soils with or without seedlings had the same microbial biomass at the 2nd harvest. A slight increase was found for soils without seedlings by the final harvest, although the relative trend between sites (mid-seral = early-seral > late-seral) was consistent, regardless of presence or absence of seedlings.

DISCUSSION

We initially hypothesized that *Pinus jeffreyi* seedlings grown in soil from early-seral sites dominated by *Wyethia mollis* would produce less total plant mass than seedlings grown in soil from the shrub-dominated, mid-seral site. The basis for this hypothesis was potential allelopathic effects of *Wyethia* (Yoder-Williams and Parker 1987) and superior nutrient status at the mid-seral site due to the beneficial effects of nitrogen-fixing shrubs (Conard et al. 1985, Johnson 1995, Busse et al. 1996, Busse 2000a, 2000b) and other microbial processes purportedly associated with shrub-dominated sites (Amaranthus and Perry 1987, 1989, Perry

et al. 1989, 1992, Amaranthus et al. 1990, Borchers and Perry 1990). Results from this greenhouse study showed that seedling growth in soil from the early-seral site was indeed lower than that of the mid-seral site during the 1st and 2nd harvests. By the 3rd harvest, however, differences between the 2 treatments were not significant. Thus, any nutrient limitations or allelopathic effects of *Wyethia* occurred only during the initial growth phase following germination and had no long-term effect on seedling growth.

Leaves of *Wyethia* contain both water-extractable and volatile allelochemicals when collected soon after total leaf expansion in mid- to late spring (Heisey and Delwiche 1983). These compounds include linoleic acid, isoflavones, lanostene-type triterpenes, and some *n*-alkalines (Waddell et al. 1982). Soil and litter were collected for the greenhouse study in late spring specifically for this reason, to approximate the period of maximum leaf expansion and allelochemical concentrations in *Wyethia* leaves. Below-average precipitation in the year prior to soil collection may have also contributed to higher concentrations of allelochemicals due to limited leaching losses. Given these optimum conditions for allelochemicals, the lack of a sustained reduction in *Pinus* seedling growth in the *Wyethia*-dominated soil suggests an unlikely role of allelopathy by *Wyethia* in the early growth of *Pinus*, at least via allelochemicals stored in the soils. Other bioassays have shown that fresh *Wyethia* leaves are inhibitory to germination of *Bromus mollis*, *Hordeum vulgare*, and *Lactuca* spp. (Heisey

TABLE 2. Total, shoot, and root dry mass of *Pinus jeffreyi* seedlings grown in soils from early-, mid-, and late-seral sites. Different lowercase letters within a row indicate significant differences ($P < 0.05$) between sites.

Days from sowing	Total plant mass (g)			Shoot mass (g)			Root mass (g)		
	Early-	Mid-	Late-	Early-	Mid-	Late-	Early-	Mid-	Late-
220	2.01[ab]	2.45[a]	1.68[b]	0.68[ab]	0.82[a]	0.61[b]	1.33[ab]	1.63[a]	1.10[b]
314	7.80[b]	9.71[a]	6.31[c]	4.38[a]	4.77[a]	3.46[b]	3.42[b]	4.94[a]	2.85[c]
417	15.07[a]	15.72[a]	11.36[b]	6.52[a]	6.14[a]	4.43[b]	8.55[a]	9.58[a]	6.93[b]

TABLE 3. Foliar nutrient concentration of *Pinus jeffreyi* seedlings at 417 days after sowing. The seedlings were grown in soils from early-, mid-, or late-seral sites. Different lowercase letters within a column indicate significant differences ($P < 0.05$) between sites.

Site	N	P	K	Ca	Mg	Mn	Fe	Zn	Al	B	Cu	Na
			- - mg g^{-1} - -					- - - - μg g^{-1} - - - -				
Early-seral	8.56	1.93	6.94	3.83[b]	1.47[b]	160[a]	33[b]	96[a]	60[b]	31[b]	4.9[a]	37
Mid-seral	10.02	1.87	6.77	4.96[a]	1.53[b]	88[b]	33[b]	71[b]	45[b]	33[ab]	4.3[b]	50
Late-seral	8.78	1.75	7.10	4.05[b]	1.88[a]	137[a]	60[a]	77[b]	104[a]	37[a]	4.7[ab]	34

TABLE 4. Microbial biomass (mg C kg^{-1} soil) from the early-, mid-, and late-seral sites. Values are compared for pots with and without seedlings in order to separate the influence of seedling roots from site effects. Different lowercase letters within a row for with seedlings or without seedlings indicate significant differences ($P < 0.05$) between sites.

Days from sowing	With seedlings			Without seedlings		
	Early-	Mid-	Late-	Early-	Mid-	Late-
220	675[b]	913[a]	591[b]	348[b]	504[a]	392[b]
314	732[b]	875[a]	517[c]	740[b]	859[a]	513[c]
417	783[a]	799[a]	523[b]	858[ab]	928[a]	637[b]

and Delwiche 1983, Yoder-Williams and Parker 1987). Yoder-Williams and Parker (1987) found reduced germination of *Pinus jeffreyi* seeds that overwintered beneath *Wyethia* litter. In contrast, we found no evidence of allelopathic inhibition of seed germination in soil from the *Wyethia*-dominated site. Germination of *Pinus* seeds was near 95% in our pot study, regardless of soil origin. Williams (1995) suggested that direct interference by *Wyethia* is observed as an allelopathic effect on germinating *Pinus* seed in spring and in soil moisture utilization in late summer. These interacting processes may be moderated during years of above-average precipitation by leaching allelochemicals and increased soil moisture availability.

Allelopathic inhibition from water-soluble leachates of *Arctostaphylos* and *Ceanothus velutinus* leaf litter has also been implicated in laboratory bioassays. Reduced radicle elongation of *Bromus tectorum*, *Hordeum vulgare* (del Moral and Cates 1971), *Cucumis sativus*, *Pseudotsuga menziesii* (Tinnin and Kirkpatrick 1985), and *Abies concolor* (Conard 1985) was reported in soil-free bioassays. However, no inhibitory effect of litter leachates was found by Tinnin and Kirkpatrick (1985) when soil was used as the growth medium. It is doubtful, therefore, that litter from these shrubs suppressed growth of *Pinus* in the mid-seral treatment of our experiment.

We had anticipated an improved soil nutrient status, particularly soil N, at the shrub-dominated site (mid-seral) compared to the early-seral site due to the N fixation from actinorhizal symbionts associated with *Ceanothus prostratus*, *C. velutinus* (Delwiche et al. 1965, Conard et al. 1985, Busse 2000a, 2000b), and *Purshia tridentata* (Webster et al. 1967, Busse 2000a, 2000b). Annual N fixation in forests east of the Sierra Nevada and Cascade Range crest varies from 5 to 15 kg ha^{-1} for *C. velutinus* and 1 kg ha^{-1} for *C. prostratus* and *Purshia tridentata* (Busse 2000a, 2000b). Busse et al. (1996) found increased soil C, N, and microbial biomass in the upper horizon of a ponderosa

pine forest due to long-term retention of shrubs. The shrub species in their study were the same as those found at the mid-seral site (*Ceanothus velutinus, Purshia tridentata*, and *Arctostaphylos patula*). Johnson (1995) also found improved soil N status at pine sites with a dominance of *C. velutinus*. Results from our study indicate that the shrub-dominated site had greater soil C and microbial biomass, but slightly lower N than the early-seral site. Site differences in soil P, Ca, Mg, and microbial characteristics had no measurable effect on seedling growth in the greenhouse bioassay, however. Without knowing cover, density, and duration of site occupancy of N-fixing shrubs, it is difficult to interpret and compare the results between other studies and ours.

The most striking effect we measured was a reduction in seedling mass for the *Pinus*-dominated, late-seral treatment. Although the mechanism(s) responsible for this observation is unclear, high Al and Fe concentrations in foliage from the late-seral treatment suggest either potential metal toxicity or nutrient imbalance. *Pinus* litter has also been shown to have allelopathic effects on plant growth (Lodhi and Killingbeck 1980, 1982). It is not uncommon to find the area below large *Pinus* trees to have few species with low density and aboveground biomass. There is often a thick needle layer below large *Pinus* trees that may serve as a physical barrier to seed/soil contact for many species. *Pinus* litter generally has a high C/N value (Hart et al. 1992), which can limit N availability. We found no evidence to support this mechanism, however. Soil from the late-seral site had a lower C/N ratio than soil from the mid-seral site, and no significant differences in foliar N concentrations were found between treatments in the greenhouse bioassay. Bever (1994) suggested that repeated planting of a single species (analogous to the limited species diversity of the late-seral site) may suppress plant growth due to negative feedback from soil biota or accumulations of specific pathogens, and/or change in composition of the community of mutalists. Root pathogens were not observed in our short-term bioassay or in our field-collection sites. However, *Wyethia*-dominated understories in *Pinus ponderosa* in south central Oregon are associated with higher rates of mortality of 10- to 30-year *Pinus* regeneration from *Heterobasidium annosum* root disease centered around old

(late-seral) *Pinus* stumps (Hopkins et al. 1988).

The discrepancy between our findings and observations from the field suggests that factors other than allelochemicals or soil nutrients per se limit regeneration of *Pinus* on *Wyethia*-dominated sites. Succession may be influenced by competition for soil moisture and changes in microclimate (Tilman and Wedin 1991, De Pietri 1992). Previous research has shown that soil moisture availability on shrub-dominated sites can be significantly greater than on *Wyethia*-dominated sites during the latter part of the growing season (Williams 1995). The shrub species associated with the mid-seral stage compete with establishing conifers for light, water, and nutrients (Conard and Radosevich 1981, 1982, McDonald 1983a, 1983b, Roy 1983, Radosevich 1984, Shainsky and Radosevich 1986). A shrub canopy also provides a shaded microenvironment, however, that reduces evaporative demand and improves the water balance of trees, which may be less physiologically stressful than competition for soil resources on hot, dry sites (Conard and Radosevich 1982, Lanini and Radosevich 1986).

In conclusion, we could find only limited evidence that allelochemicals from *Wyethia* might suppress growth of *Pinus* seedlings. There was an initial reduction in growth of seedlings in early- versus mid-seral soils, but after 417 days of growth the 2 treatments were similar. The more obvious response was the reduction in seedling growth associated with the late-seral, *Pinus*-dominated soil. We suggest that research into the relationship of microclimate and plant-soil water relations, and an examination of the community composition of soil biota, may aid in explaining the pattern of regeneration of *Pinus jeffreyi*. This information, coupled with an increased understanding of larger scale climate effects (e.g., increased atmospheric temperature and drought cycles), may further our knowledge of the complex interactions involved in succession of *Wyethia*-dominated seral stages within western pine forests.

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