Mycorrhiza (1996) 6:137–143

ORIGINAL PAPER

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The effects of *Laccaria proxima* and fibrous pulp waste on the growth of nine container-grown conifer seedling species

Accepted: 30 August 1995

Abstract Japanese larch (*Larix kaempferi*), white spruce (*Picea glauca*), black spruce (*Picea mariana*), red spruce (*Picea rubens*), jack pine (*Pinus banksiana*), mugo pine (*Pinus mugo*), red pine (*Pinus resinosa*), Japanese black pine (*Pinus thunbergii*) and Douglas-fir (*Pseudotsuga menziesii* var. *menziesii*), were inoculated to test the effective host range of the ectomycorrhizal fungus *Laccaria proxima* and the possibility of utilizing pulp waste as a potting medium for containerized seedling production. *Laccaria proxima* tended to improve the container growth of Japanese black pine and white spruce, and significantly improved that of jack pine, mugo pine, black spruce, red spruce and Douglasfir. The growth of red pine and Japanese larch were only slightly improved with *L. proxima.* Pulp waste (33% by volume) had negative effects on tree seedling growth, except for Douglasfir (no significant effect). The interactions of *Laccaria proxima* and pulp waste varied; the hosts were significantly positive ($P < 0.01$) in the case of jack pine and black spruce, but there was no significant effect for the rest. Negative effects were found with Japanese black pine. Use of pulp waste in seedling production of jack pine, black spruce, mugo pine, red spruce and Douglasfir inoculated with *L. proxima* and of Japanese black pine both with and without *L. proxima* is feasible, but further research is necessary to determine the optimal percentage of pulp waste that can be utilized in seedling production of tree species and the field performance of these seedlings.

Key words Conifers · Containerized seedlings · Ectomycorrhiza · Fibrous pulp waste · *Laccaria proxima*

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Introduction

Laccaria proxima (Boud.) Pat. is a native North American ectomycorrhizal fungus (Singer 1977; Aquirre-Acosta and Perez-Silva 1978; Danielson et al. 1984; Mueller 1992) occurring as a pioneer with many coniferous species (Mason et al. 1987; Mueller 1992). It has a broad host range and an extensive geographical distribution. Worldwide, the fungus is reported to occur in Australia (May et al. 1987), Britain (Last et al. 1984), China (Ying et al. 1982), Italy (Giovanni 1980; Ballero and Contu 1987), Japan (Sagara 1981), Korea (Lee and Yang 1987), New Zealand (McNabb 1972), Norway (Høiland 1976), and Romania (Salageanu and Stefureac 1972). The fungus is found associated with many gymnosperm and angiosperm hosts, including Fraser fir (*Abies fraseri*) (Kenerley et al. 1984), paper birch (*Betula papyrifera*) (Jones and Hutchinson 1986), European white birch (*B. pendula, B. pubescens*) (Last et al. 1984; Pelhain et al. 1988), Japanese chestnut (*Castanea crenata*) (Lee and Yang 1987), tamarack (*Larix laricina*) (Zhu 1985), white spruce (*Picea glauca*), black spruce (*Picea mariana*), Sitka spruce (*Picea sitchensis*), jack pine (*Pinus banksiana*), lodgepole pine (*Pinus contorta*), and Scotch pine (*Pinus sylvestris*) (Kampert and Strzelczyk 1978; Danielson 1984; Mason et al. 1987; Cote and Thiboult 1988; Browning and Whitney 1991). There is limited information about its host specificity and its effects, especially on the growth of economically important tree species.

In most wood pulping processes, only about one half of the original wood is converted to usable products (Dolar et al. 1972). Producing one ton of wood pulp consumes 4.37 m^3 of wood (Canadian Pulp and Paper Association 1989). The leftover fiber and by-products of the pulping process are largely disposed of as waste (Dolar et al. 1972). In northwestern Ontario alone, thousands of tons of pulp and fibrous pulp waste are produced each day (D'Arcy 1988). The liquid and solid wastes pose a serious problem to a society that is becoming increasingly aware of the detrimental influences of pollution on the environment and the need for recycling and conservation. Paper mills dispose of approximately three quarters of their pulp waste sludge in landfills (Miner and Marshall 1976) while the other quarter is combusted in a power boiler or incinerator (Edde 1984). However, increasing concern over potential leachates from landfills, limited landfill capacity, increasing transportation costs and negative impact on the environment have increased interest in other disposal methods (Dolar et al. 1972; Edde 1984; Sawhney and Kozloski 1984; D'Arcy 1988). With limited loading, the waste can be successfully incorporated into the soil due to the high organic matter content (Gehm 1973). A feasible approach is to utilize fibrous pulp waste as a potting medium component in the production of containerized tree seedlings. Since fibrous pulp waste is reported to have a negative effect on jack pine seedling growth (D'Arcy 1988), *L. proxima* (Boud.) Pat. might be utilized to negate the harmful effect on jack pine and other tree seedlings. The objective of the present study was to determine the interactive influence of *L. proxima* and pulp waste (33% v/v) on the growth of nine conifer species.

Materials and methods

Laccaria proxima (isolate 79191C) was obtained from the forest pathology laboratory, Lakehead University. This isolate originally came from the University of Toronto and was collected under jack pine growing in sandy soil. This fungus was chosen according to three criteria: (1) it is a pioneer ectomycorrhizal species in reforestation sites, (2) it is a native species in North America and northern China, and (3) it has a broad host range (Singer 1977; Ying et al. 1982; Mason et al. 1987; Mueller 1992). Seven tree species economically important in Canada and North America were established from the following Lakehead University seed lots: white spruce [*Picea glauca* (Moench) Voss], seed lot 48.400 S81086; black spruce [*Picea mariana* (Mill.) B.S.P.], seed lot 48.600 S83032; red spruce (*Picea rubens* Sarg.), seed lot 48.700 S86031; jack pine (*Pinus banksiana* Lamb.), seed lot 50.200 S83034; mugo pine (*Pinus mugo* Turra), seed lot 50.600 S80270; red pine (*Pinus resinosa* Ait.), seed lot 50.700 S83053; and Douglasfir [*Pseudotsuga menziesii* var. *menziesii* (Mirbel) Fr.], seed lot 55.150 S82007. Seeds of two important forest species in northern China, Japanese larch [*Larix kaempferi* (Lamb.) Carr.] and Japanese black pine (*Pinus thunbergii* Parl.), were obtained from the Qingdao Forestry Bureau, Shandong, People's Republic of China. Fibrous pulp waste was obtained from Canadian Pacific Forest Products, Thunder Bay, Ont.

The inoculum was in the form of *L. proxima* colonized rye grain. Aliquots of rye grain (180 g, 250 ml with 100 ml water) were placed in ten 500-ml fruit jars and autoclaved at 121 °C for 20 min. After cooling, each jar was inoculated with about 20 ml of prepared rye grain inoculum and stirred to assure mixing of rye grain and *L. proxima*. The jars were incubated at 25 °C for ca. 15 days for mycelial establishment.

A 2^2 factorial design was used in the experiment, the two factors being *L. proxima* and pulp waste and the two levels, absence and presence of the two factors.

The potting medium was a mixture of *Sphagnum* peat moss (Sunshine), vermiculite (Terra-Lite) and pulp waste autoclaved at 121 °C for 20 min before use. The control medium was equal parts by volume of peat moss and vermiculite (Riffle and Tinus 1982; Hung and Molina 1986). The pulp waste treatment was pulp waste, peat moss and vermiculite in equal amounts by volume. For the *L. proxima* treatment, peat moss and vermiculite in equal amounts by volume (total of 1500 ml) was inoculated with 20 ml of *L. proxima* rye seed inoculum. In the *L. proxima* plus pulp waste treatment, the medium composed of pulp waste, peat moss and vermiculite in equal proportions by volume (total of 1500 ml) was inoculated with 20 ml of *L. proxima* rye seed inoculum. The seedlings were grown in Can-Am multipots (M-2), 22 × 35 cm with 67 cavities of 3.3 × 12.0 cm and 67 ml. The cavity diameter : height ratio was 1 : 3.6 with 3.5 cm between cavities. The number of replications ranged from 6 to 19. Tree species and treatments were randomly assigned to multipots.

After complete mixing and wetting with tap water, the media were placed in the multipots. The seeds were surface sterilized with 1% sodium hypochlorite for 20 min and rinsed in water. Two to three seeds were sown into each multipot cavity and covered with vermiculite to a depth of 5 mm. After watering to saturation, the multipots were covered with black polyethylene for 3 days to maintain moisture and to induce seed germination. When germination was complete and seed coats shed, seedlings were thinned to one seedling per cavity. The seedlings were watered for 10 s every 16 min by an automatic sprinkling system until August 1988, when they were watered twice each day with an automatic overhead irrigation system. The seedlings were fertilized once a week a TERR-LIFE Siphon Mixer at 300 ppm of 20-20-20 (N-P-K) from a stock solution of 7500 ppm. The fertilizer solution was allowed to drip freely through the drain holes of multipots. The temperature range was 17–32 °C in winter and 19–38 °C in summer. Relative humidity varied from about 80 to 90%. The seedlings were exposed to natural sunlight in room 1 of the greenhouse at Lakehead University. The day length in Thunder Bay varies from 8 h 20 min to 16 h 19 min. Jack pine seedlings were grown in room 2 of the greenhouse under daylight and 16 h high pressure sodium lamps (400 W). To eliminate position effects, the multipots were systematically rotated in position weekly throughout the experiments. Jack pine was grown from 6 March to 2 May 1988, and Japanese black pine and larch were grown from 10 July to 16 December 1988. The other species were grown from 20 May to 21 December 1988.

After the seedlings were harvested, the lengths and dry weights of shoots and roots were determined. Dry weights were measured after oven-drying seedlings at 102 ± 2 °C for 24 h. In order to verify the formation of ectomycorrhiza by *L. proxima* on the conifer species, 7-μm paraffin sections were prepared from ultimate laterals following the procedure used by Bakowsky (1988). *L. proxima* formed intact fungal mantles and Hartig nets on the nine conifer species.

SPSS-X software on the Lakehead University Microvax computer was used for statistical analysis. Statistical comparisons were made between (1) mycorrhizal and non-mycorrhizal plants (effects of *L. proxima*), (2) seedling growth in media with and without pulp waste (effects of pulp waste) and (3) seedling growth in media with pulp waste and with or without mycorrhiza (interaction of *L. proxima* and pulp waste). When the interaction reached a significant level, multiple comparison analysis (Duncan) between four treatments was conducted.

Seedling dry weight was considered a more important parameter than seedling length for evaluating the results because it more accurately reflects the photosynthetic activities of the seedlings.

Results

White spruce

Laccaria proxima had no significant effects on shoot, root or total lengths of white spruce. The addition of pulp waste to the peat/vermiculite had significantly negative effects on shoot and total lengths but not root length (Tables 1, 2). *L. proxima* and pulp waste had no significant interaction on the shoot and the total lengths of white spruce, but had a significantly negative interaction on root length. There were no significant differences in root length among the four treatments (Tables 1, 2).

Laccaria proxima had a significantly positive effect on root dry weight, but not on shoot and total dry weights. Pulp waste without *L. proxima* had significantly negative effects on shoot, root and total dry weights. *L. proxima* with pulp waste showed no interactive effects on the dry weights (Tables 1, 2).

Black spruce

Laccaria proxima had significant positive and negative effects, respectively, on shoot lengths and root lengths of black spruce, and there was no significant effect on total length (Tables 1, 2). Pulp waste had significantly negative effects on the shoot and total lengths but not the root length. Although *L. proxima* and pulp waste had no interactive effects on the shoot and total lengths, there was a negative interaction on the root length (Tables 1, 2). Black spruce seedlings grown in the control treatment, *L. proxima,* and pulp waste produced significantly longer roots than those in *L. proxima* plus pulp waste, but there were no significant differences among the *L. proxima,* pulp waste and control treatments.

While *L. proxima* significantly increased dry weights (Tables 1, 2), the overall effects of pulp waste alone were negative. Interactions of *L. proxima* and pulp waste on black spruce dry weights were significantly positive (Tables 1, 2).

Red spruce

Laccaria proxima significantly increased the root length of red spruce, but not the shoot and total lengths (Tables 1, 2). Pulp waste had significantly negative effects on the shoot and total lengths, but not on the root length. *L. proxima* and pulp waste had no interactive effects on the lengths of red spruce (Tables 1, 2).

Laccaria proxima significantly increased root and total dry weights, but not shoot dry weight. Pulp waste had significantly negative effects on dry weights. *L. proxima* and pulp waste had no interaction on red spruce seedling dry weights (Tables 1, 2).

Jack pine

Laccaria proxima significantly increased the shoot and total lengths of jack pine seedlings, but not the root length (Tables 1, 2). Pulp waste without *L. proxima* was markedly detrimental to root and shoot lengths, but there were significantly positive interactive effects of *L. proxima* and pulp waste on shoot and total lengths, though not on root length (Tables 1, 2). Shoot lengths of jack pine seedlings grown in the control, *L. proxima,* and *L. proxima* plus pulp waste treatments were significantly greater than in the pulp waste treatment. Total lengths of jack pine seedlings grown in the control and *L. proxima* treatments were significantly greater than in the pulp waste and *L. proxima* plus pulp waste treatments, and seedlings grown in the *L. proxima* plus pulp waste treatment had significantly greater total length than those grown in pulp waste alone (Tables 1, 2).

Both *L. proxima* and pulp waste had significant effects, positive and negative respectively, on jack pine seedling dry weights (Tables 1, 2), and *L. proxima* and pulp waste had significant interactive effects on dry weight (Tables 1, 2). The jack pine seedlings were smaller than those of other species tested because they were grown for a 3- to 5-months shorter period.

Mugo pine

Laccaria proxima and pulp waste had no individual or interactive effects on lengths of mugo pine seedlings (Tables 1, 2). Both *L. proxima* and pulp waste had significant effects, positive and negative respectively, on dry weights of mugo pine seedlings, and there were no interactive effects (Tables 1, 2).

Red pine

Both *L. proxima* and pulp waste had no significant effects on lengths of red pine seedlings. *L. proxima* plus pulp waste had a significantly negative interaction on root length, but not on shoot and total lengths (Tables 1, 2). Root lengths of red pine grown in pulp waste were significantly greater than those in the control, *L. proxima* and *L. proxima* plus pulp waste. There were no significant differences among these latter three treatments.

Laccaria proxima had no significant effects on dry weight of red pine, and there was no significant interaction of *L. proxima* and pulp waste (Tables 1, 2). Pulp waste had a significant negative effect on dry weights.

Douglas-fir

Laccaria proxima had significant effects on shoot length of Douglasfir but not on root and total lengths. Pulp waste had significantly negative effects on shoot

Table 1 Average length and dry weight (*dry wt*) of seedlings of nine container grown conifers with or without *Laccaria proxima* and fibrous pulp waste (*Pw*). Data followed by different letters within a group of four values (Pw 0% or 33%, *L. proxima* absent or present) are significantly different

Tree species	Growth parameter	Pw (%)	*Laccaria proxima*					
			Shoot		Root		Total	
			Absent	Present	Absent	Present	Absent	Present
Larix kaempferi								
	Length (mm)	0	175 ± 5	173 ± 5	124 ± 3	123 ± 3	299 ± 3	296 ± 7
	± SE	33	156 ± 9[1]	153 ± 6	114 ± 6[1]	111 ± 1	270 ± 12[1]	265 ± 6
	Dry wt (mg)	0	383 ± 31	406 ± 25	69 ± 7	80 ± 7	452 ± 35	486 ± 31
	± SE	33	289 ± 39[1]	318 ± 55	112 ± 7	127 ± 10	401 ± 49	445 ± 70
Picea glauca								
	Length (mm)	0	81 ± 3	79 ± 4	123 ± 2a*	128 ± 3a	204 ± 4	207 ± 5
	± SE	33	68 ± 4[1]	67 ± 5	127 ± 2a	121 ± 2a[2]	194 ± 6[1]	188 ± 5
	Dry wt (mg)	0	259 ± 27	262 ± 27	174 ± 22	196 ± 21[3]	433 ± 46	458 ± 44
	± SE	33	114 ± 15[1]	165 ± 24	84 ± 18[1]	144 ± 19	198 ± 31	308 ± 41
Picea mariana								
	Length (mm)	0	134 ± 3	143 ± 5[3]	126 ± 2a	127 ± 2a[3]	260 ± 4	270 ± 5
	± SE	33	109 ± 7[1]	131 ± 8	131 ± 2a	119 ± 6b[2]	240 ± 7[1]	250 ± 7
	Dry wt (mg)	0	439 ± 21a	471 ± 18a[3]	266 ± 21a	290 ± 18a[3]	706 ± 38a	761 ± 27a[3]
	± SE	33	163 ± 18c[1]	318 ± 33b[2]	84 ± 18c[1]	144 ± 19b[2]	256 ± 35c[1]	517 ± 45b[2]
Picea rubens								
	Length (mm)	0	174 ± 6	165 ± 6	119 ± 2	128 ± 3[3]	293 ± 6	293 ± 6
	± SE	33	130 ± 6[1]	144 ± 6	121 ± 2	125 ± 2	251 ± 6[1]	269 ± 7
	Dry wt (mg)	0	604 ± 23	588 ± 31	226 ± 17	269 ± 22[3]	830 ± 31	857 ± 44[3]
	± SE	33	284 ± 34[1]	383 ± 32	117 ± 18[1]	198 ± 20	401 ± 50[1]	581 ± 48
Pinus banksiana								
	Length (mm)	0	79 ± 2a	78 ± 3a[3]	112 ± 3	115 ± 3	191 ± 3a	193 ± 3a[3]
	± SE	33	52 ± 2b[1]	76 ± 3a[2]	106 ± 2[1]	107 ± 02	158 ± 3c[1]	183 ± 3b[2]
	Dry wt (mg)	0	53 ± 2b	82 ± 8a[3]	26 ± 1b	36 ± 4a[3]	79 ± 3b	117 ± 11a[3]
	± SE	33	14 ± 1c[1]	77 ± 10a[2]	11 ± 1c[1]	38 ± 4a[2]	25 ± 2c[1]	114 ± 13a[2]
Pinus mugo								
	Length (mm)	0	130 ± 5	132 ± 6	120 ± 2	123 ± 2	249 ± 5	255 ± 6
	± SE	33	127 ± 6	128 ± 3	122 ± 3	128 ± 6	248 ± 6	256 ± 9
	Dry wt (mg)	0	488 ± 44	640 ± 47[3]	223 ± 27	342 ± 25[3]	711 ± 67	982 ± 67[3]
	± SE	33	202 ± 25[1]	446 ± 31	80 ± 7[1]	201 ± 20	281 ± 31[1]	647 ± 49
Pinus resinosa								
	Length (mm)	0	154 ± 5	165 ± 6	118 ± 1a	119 ± 2a	273 ± 6	284 ± 6
	± SE	33	155 ± 8	152 ± 9	127 ± 4b	118 ± 3a[2]	282 ± 7	270 ± 7
	Dry wt (mg)	0	510 ± 52	583 ± 53	335 ± 21	332 ± 37	845 ± 66	916 ± 82
	± SE	33	323 ± 57[1]	397 ± 80	179 ± 31[1]	262 ± 46	502 ± 86[1]	657 ± 125
Pinus thunbergii								
	Length (mm)	0	160 ± 3b	170 ± 3a	120 ± 2	126 ± 3[3]	280 ± 4b	296 ± 4a
	± SE	33	159 ± 3b[1]	147 ± 4c[2]	118 ± 2	126 ± 2	278 ± 3b[1]	273 ± 4b[2]
	Dry wt (mg)	0	518 ± 25b	617 ± 38a	92 ± 9b	147 ± 14a[3]	610 ± 31b	763 ± 47a
	± SE	33	512 ± 16b[1]	453 ± 28b[2]	112 ± 7b	127 ± 10a[2]	624 ± 21b[1]	580 ± 37b[2]
Pseudotsuga menziesii								
	Length (mm)	0	155 ± 7	189 ± 13[3]	109 ± 1	109 ± 1	264 ± 8	298 ± 14
	± SE	33	137 ± 7[1]	149 ± 9	110 ± 7	109 ± 4	247 ± 11[1]	258 ± 10
	Dry wt (mg)	0	364 ± 32	506 ± 55[3]	277 ± 36	384 ± 41[3]	640 ± 53	890 ± 96[3]
	± SE	33	297 ± 47	416 ± 42	260 ± 30	361 ± 32	558 ± 75	777 ± 62

[1] Significant effect of pulp waste
[2] Significant interaction of *L. proxima* and pulp waste
[3] Significant effect of *L. proxima*

and total lengths but not on root length. *L. proxima* and pulp waste had no statistically significant interactive effects on Douglasfir seedling lengths (Tables 1, 2).

Laccaria proxima significantly increased the dry weight of Douglasfir seedlings, but there was no significant effect of pulp waste, and no interaction of *L. proxima* with pulp waste (Tables 1, 2).

Table 2 *P* values of the effects and interactions of *Laccaria proxima* (*Lp*), pulp waste (*Pw*) and their combination (L_p-P_w) on the growth of nine conifer species [(+) significant positive effect or interaction, (−) significant negative effect or interaction]

Tree species	Seedling part	df	Length			Dry weight		
			Lp	Pw	Lp-Pw	Lp	Pw	Lp-Pw
Larix kaempferi								
	Shoot	47	0.743	0.009 (−)	0.960	0.512	0.025 (−)	0.950
	Root	47	0.599	0.010 (−)	0.825	0.266	0.736	0.943
	Total	47	0.555	0.002 (−)	0.824	0.441	0.058 (−)	0.948
Picea glauca								
	Shoot	45	0.771	0.003 (−)	0.954	0.195	0.001 (−)	0.307
	Root	45	0.687	0.615	0.032 (−)	0.036 (+)	0.001 (−)	0.362
	Total	45	0.664	0.007 (−)	0.327	0.075 (+)	0.001 (−)	0.301
Picea mariana								
	Shoot	55	0.019 (+)	0.005 (−)	0.323	0.001 (+)	0.001 (−)	0.012 (+)
	Root	55	0.009 (−)	0.287	0.001 (−)	0.001 (+)	0.001 (−)	0.031 (+)
	Total	55	0.105	0.001 (−)	0.957	0.001 (+)	0.001 (−)	0.007 (+)
Picea rubens								
	Shoot	68	0.729	0.001 (−)	0.057	0.216	0.001 (−)	0.056
	Root	68	0.012 (+)	0.750	0.363	0.002 (+)	0.001 (−)	0.335
	Total	68	0.183	0.001 (−)	0.132	0.027 (+)	0.001 (−)	0.081
Pinus banksiana								
	Shoot	85	0.001 (+)	0.001 (−)	0.001 (+)	0.001 (+)	0.001 (−)	0.008 (+)
	Root	85	0.500	0.005 (−)	0.683	0.001 (+)	0.007 (−)	0.002 (+)
	Total	85	0.001 (+)	0.001 (−)	0.001 (+)	0.001 (+)	0.001 (−)	0.007 (+)
Pinus mugo								
	Shoot	65	0.709	0.501	0.922	0.001 (+)	0.001 (−)	0.239
	Root	65	0.059	0.155	0.596	0.001 (+)	0.001 (−)	0.939
	Total	65	0.124	0.648	0.509	0.001 (+)	0.001 (−)	0.400
Pinus resinosa								
	Shoot	38	0.416	0.426	0.389	0.225	0.005 (−)	0.997
	Root	38	0.227	0.127	0.039 (−)	0.404	0.002 (−)	0.227
	Total	38	0.636	0.736	0.099	0.247	0.002 (−)	0.635
Pinus thunbergii								
	Shoot	64	0.721	0.002 (−)	0.001 (−)	0.451	0.007 (−)	0.005 (−)
	Root	64	0.003 (+)	0.731	0.704	0.001 (+)	0.767	0.034 (−)
	Total	64	0.109	0.005 (−)	0.009 (−)	0.106	0.035 (−)	0.005 (−)
Pseudotsuga menziesii								
	Shoot	24	0.028 (+)	0.007 (−)	0.232	0.010 (+)	0.108	0.799
	Root	24	0.974	0.788	0.799	0.009 (+)	0.589	0.924
	Total	24	0.088	0.033 (−)	0.323	0.007 (+)	0.227	0.846

Japanese black pine

Laccaria proxima had a significant effect on root length of Japanese black pine seedlings but not on shoot or total lengths. Pulp waste and *L. proxima* plus pulp waste had significantly negative individual and interactive effects, respectively, on shoot length and total length but not significant effects on root length (Tables 1, 2). The shoot lengths of Japanese black pine seedlings grown in the *L. proxima* treatment were significantly greater than in the other three treatments. Among the other three treatments, shoot lengths in the control and the pulp waste treatments were significantly greater than in the *L. proxima* plus pulp waste treatment. The total lengths of Japanese black pine seedlings grown in the *L. proxima* treatment were significantly greater than in the other three treatments, amongst which there were no significant differences.

Laccaria proxima significantly increased root dry weight of Japanese black pine, but not shoot or total dry weights. Pulp waste had significantly negative effects on the shoot and total dry weights but not on root dry weight, and *L. proxima* plus pulp waste had significantly negative interactions on all dry weights (Tables 1, 2). Shoot and total dry weights of Japanese black pine grown in the *L. proxima* treatment were significantly higher than in the other three treatments. In the *L. proxima* plus pulp waste treatment, root dry weight was significantly higher than in the pulp waste treatment but not higher than in the control and *L. proxima* treatment.

Japanese larch

Laccaria proxima had no significant effects on seedling length of Japanese larch. Pulp waste had significantly negative effects on seedling length, and the presence of *L. proxima* with the pulp waste failed to improve the seedlings (Tables 1, 2).

Laccaria proxima had no significant effects on seedling dry weight of Japanese larch. Pulp waste had a significantly negative effect on shoot dry weight, but not on root and total dry weights. There were no significant interactions of *L. proxima* with pulp waste on seedling dry weights (Tables 1, 2).

Discussion

Laccaria proxima significantly increased the seedling weights and lengths of most of the tree species. *L. proxima* had no significant effects on Japanese larch and red pine growth (Tables 1, 2). Pulp waste significantly decreased the seedling growth of all tree species except Douglasfir (Tables 1, 2).

The interaction of *L. proxima* plus pulp waste on the tree species was quite different. There were highly positive significant interactions in the case of black spruce and jack pine, positive significant interactions with red spruce, no significant interactions with white spruce, mugo pine, red pine, Douglasfir and Japanese larch, and highly negative interactions with Japanese black pine (Tables 1, 2). In other words, the apparent detrimental effect of the pulp waste is at least partially negated by the mycorrhizal fungus.

Preliminary experiments conducted by D'Arcy (1988) showed that pulp waste as a partial potting medium produced a negative effect on jack pine seedling growth. Our results verified this result.

For pulp waste to be successfully used as a potting medium component for growing seedlings inoculated with *L. proxima, L. proxima* must establish ectomycorrhizae and improve seedling growth, the interaction of *L. proxima* with pulp waste must be positive, and the harmful effects of pulp waste on seedling growth must be partially negated.

The utilization of 33% v/v pulp waste appeared feasible for containerized seedling production of jack pine, black spruce, mugo pine, Douglasfir and red pine, with jack pine and black spruce showing the most promise. However, it is now crucial to find out the optimal proportions of pulp waste and *L. proxima* necessary to minimize the negative effects on the seedling growth.

In the conventional procedure of ectomycorrhizal fungi evaluation, only optimal conditions were adopted (Valdés 1986, Gagnon et al. 1987). This procedure does not permit evaluation of how mycorrhizae perform under adverse environmental conditions. Mycorrhizae that improved seedling growth in the greenhouse failed to improve growth performance in the field. One reason could have been the impact of the changed environmental conditions and difficulties in adapting. To reduce possible shock, the change should be as small as possible and seedlings should be resistant to adverse conditions of the new habitat (Mikola 1973). Browning and Whitney (1992) reported that seedlings of black spruce and jack pine inoculated with *L. proxima* had significantly better shoot growth than uninoculated seedlings over a 2-year period on reforestation sites. *L. proxima* mycorrhizae also enhanced the drought tolerance of jack pine after outplanting (Browning and Whitney 1993). It would be interesting to evaluate field performance on some adverse sites, such as coal spoils. Nevertheless, Browning and Whitney's results suggest that the seedlings produced in our study would perform well or even better after outplanting, but only further research will verify this. *L. proxima* did not stimulate the growth of black spruce unless pulp waste was introduced, which resulted in a significant increase in growth over the nonmycorrhizal control. Ectomycorrhizal fungi vary in their resistance to adverse conditions such as drought and waterlogging (Trappe 1977), and certain species of fungal symbionts are more beneficial than others to pines on certain sites, especially on adverse sites (Marx and Bryan 1971). For example, loblolly pine seedlings with *Pisolithus tinctorius* grew as well at 40 °C as they did at 24 °C, but seedlings with *Thelephora terrestris* or without ectomycorrhiza did not grow at 40 °C (Marx and Bryan 1971). In nurseries, the performance of pine with both *Pisolithus tinctorius* and *Thelephora terrestris* was very good. On adverse sites, such as coal spoils, severely eroded sites, borrow pits, prairie soil and clay spoils, pine seedlings with *Pisolithus tinctorius* performed much better than those with *Thelephora terrestris* or other naturally occurring ectomycorrhizae (Marx 1977; Marx et al. 1984). Soil and site characteristics markedly influence the types of mycorrhizal fungi that develop on tree roots (Mason et al. 1987). The most adaptable species should be used to colonize seedlings prior to planting (Marx 1977).

Acknowledgements Thanks go to Dr. E. C. Setliff, in whose laboratory this work was undertaken, also for his advice and discussion. I gratefully acknowledge Drs. K. M. Brown and G. Hazenberg and Mr. J. C. Liu for help in statistical analyses, Mr. L. Sevean for technical help in the laboratory and Ms. M. Maley for making mycorrhizal root sections. I would like to express my appreciation to Mrs. N. Luckai for providing greenhouse assistance and tree seeds, to the Qingdao Forestry Bureau, China for providing some tree seeds and to Mr. P. Wong of Canadian Pacific Forest Products Ltd., Thunder Bay for providing pulp waste for the study. A Graduate Student Award from the Centre for Northern Studies is also acknowledged. This paper is based on a portion of a thesis by the author in partial fulfilment of the requirements for an MScF degree at Lakehead University.

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