CHLOROPICRIN, EPTC, AND PLANT GROWTH PROMOTING RHIZOBACTERIA FOR MANAGING SOILBORNE PESTS IN PINE NURSERIES

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Abstract

The effects of preplant soil treatments and seed treatment on seedling production and soilborne pests were evaluated on loblolly pine (Pines taeda) at three forest nurseries. Treatments were applied in 1998 at the Flint River Nursery (Byromville, GA) and at the Hauss Nursery (Atmore, AL). In 1999, treatments were applied at the Carter Nursery (Chatsworth, GA) and continued at Flint River Nursery. Soil treatments included 67% methyl bromide/33% chloropicrin at 350 lb/ac (MC33), EPTC (Eptam® 7-E), chloropicrin at 150 and 300 lb/ac (CH150 and CH300) and in combination with EPTC (CH1 50E and CH300E). At the Carter Nursery, a soil treatment of metam sodium at 80 lb/ac and chloropicrin at 150 lb/ac was added (M80/CH150). A seed treatment with the plant growth-promoting rhizobacteria (PGPR) Paenibacillus macerans was also evaluated with each soil treatment. In 1999, the EPTC herbicide treatment and PGPR seed treatment were reapplied to plots in the second-year crop at the Flint River Nursery. Funigation and EPTC treatments did not significantly affect seedling density at the three nurseries by the end of the growing season. At the Flint River Nurserv in 1998, seedling root collar diameter was greater in the CH300 and CHI 50E treatments, and seedling top weight was greater in the CH300 and CHI 50 treatments. No other differences in seedling size were observed among treatments. Seedling density at the Hauss Nursery was greater in plots with the seed treatment compared with untreated seed. At the Carter Nursery, there were fewer seedlings (2/ft) in the seed treatment plots, and at the Flint River Nursery, the seed treatment reduced seedling height in 1998 and 1999. No observed disease or insect problems occurred in any of the nurseries.

The effect of fumigation with MC33 on soilborne *Pythium* and *Fusarium spp.* varied among the nurseries. In general, fumigation reduced populations of these fungi. Parasitic plant nematodes were reduced by all fumigants following fumigation at Carter Nursery. Nematodes were rarely observed in soil samples at the other nurseries. Nutsedge was seldom found in the fumigated plots at Flint River Nursery in 1998. By 1999 only the CH300 treatment had less nutsedge than the controls. EPTC was not effective for nutsedge control at Flint River Nursery. Populations of nutsedge at the Carter and Hauss Nurseries were very low.

Key Words

Fumigation, biological control, bareroot nursery

Forest tree nursery managers across the United States use soil fumigation to control fungi, insects, and weeds. Most forest nurseries fumigate with methyl bromide that contains either 2% or 33% chloropicrin (Smith and Fraedrich 1993). Alternatives to methyl bromide fumigation are necessary, as nursery managers confront the current phaseout of this chemical. Fumigation with 100% chloropicrin has been shown to be a promising alternative to MC33 (67% methyl bromide/33% chloropicrin) for control of soilborne fungi (Enebak and others 1990) as well as nematodes (Harris 1991) and insects (Breakey and others 1945). However, chloropicrin is not considered as effective as methyl bromide for the control of weeds such as nutsedge (South and others 1997). Although the herbicide EPTC (Eptam[®] 7-E) is registered for control of nutsedge in pine nurseries, few southern nurseries use this herbicide (South 1986). Damping-off is the most common disease problem cited by nursery managers in the South (Cram and Fraedrich 1996). Plant growth-promoting rhizobacteria (PGPR) are used in agricultural crops to promote plant growth (Ryder and others 1994), and are an emerging technology for tree production (Chanway 1997). PGPR seed treatment has been found to decrease damping-off of loblolly pine (Pinus taeda) seedlings (Enebak and others 1998), and to increase seedling emergence and growth (Enebak and others 1998; Holl and Chanway 1992; O'Niell and others 1992).

Pest management programs in the future will likely have to integrate various strategies to achieve the broad-spectrum control of pests that is currently provided by fumigation with MC33. This project examined combinations of EPTC, chloropicrin, and PGPR seed treatments as potential alternatives to MC33 for the production of loblolly pine (*P. taeda*).

METHODS

The effects of preplant soil treatments on seedling production and soilborne pest problems were evaluated at the Flint River Nursery (Byromville, GA) and the E. A. Hauss Nursery (Atmore, AL) in 1998. Soil treatments consisted of MC33 at 350 lb/ac, EPTC at 7 pt/ac (E), 100% chloropicrin at 150 and 300 lb/ac (CH150 and CH300), and chloropicrin at both rates with EPTC (CH150E and CH300E). Tarps were used with all fumigants. Whole plots were split and a PGPR seed treatment of *Paenibacillus maceran* was applied at the rate of approximately 10³ cfu/100 lb seed. In 1999, the study was continued at the Flint River Nursery, and a second study site was established at the Carter Nursery (Chatsworth, GA). An additional soil treatment of metam sodium at 80 lb/ac with chloropicrin at 150 lb/ac (M80/CH 150) was included at the Carter Nursery. A composite soil sample was obtained from each plot that consisted of 5 to 10 soil cores to a 6-inch depth. Soil samples were taken after fumigation and before lifting to assess nematodes and fungi. The presence of fungi in the soil was determined using various selective media: Komada's (Komada 1975), PARP (Kannwischer and Mitchell 1981), TMR (Trichoderma, pink medium) (Elad and others 1981) and PDA-T. Three permanent history plots (1 x 4 ft) were

established in each split-plot, and seedling counts were performed weekly for 5 to 6 weeks after sowing, at mid-season, and again prior to lifting. The presence of nutsedge and other weeds were documented within the first 6 weeks. At lifting, 45 seedlings per history plot were collected for assessment of seedling height (if not pruned), root collar diameter (RCD), and dry weight. All tests of significance were carried out at a =0.10. The Tukey's studentized range test was used for testing all multiple comparisons.

RESULTS

Flint River Nursery

Fumigation and EPTC treatments did not affect seedling density (Table 1 and Table 2). In the first year. seedling RCD was greater in the CH300 and CH 150E plots than the controls, and seedling top weight was greater in the CH300 and CH 150 plots. In both years, seedling heights were lower with the PGPR seed treatment than without treatment (Table 3 and Table 4). In 1999, a seed count at the time of sowing showed that one extra seed was sown per square foot; therefore, the greater seedling density with the seed treatment was discounted. All fumigated plots had less nutsedge than the controls in 1998, but only treatments with CH300 were significantly lower than the control plots in 1999 (Table 5). Fusarium and *Pythium spp.* were rarely isolated from fumigated soil immediately following application in 1998. Plant nematodes were rarely found in any of the treatment plots.

 Table 1. Mean seedling density, size, and dry weight by soil treatment for

 loblolly pine at Flint River Nursery (Bryomville, GA) in November 1998

Treatments*	Density (ft²) [†]	Root collar diameter (in) [‡]	Height (in) [†]	Root weight (oz) [†]	Top weight (oz) [‡]
CH300	22.0	0.180a	9.02	0.027	0.133a
CH150 E	21.5	0.175ab	7.69	0.027	0.107 abc
CH300 E	21.0	0.173abc	7.73	0.026	0.110abc
CH150	22.2	0.172abc	8.83	0.026	0.121 ab
MC33	22.5	0.163abc	7.67	0.023	0.103abc
E	22.6	0.158bc	8.10	0.023	0.092bc
Control	22.5	0.154c	7.57	0.022	0.082c

*CH300 = 100% chloropicrin at 300 lb/ac; CH150 = 100% chloropicrin at 150 lb/ac; E = EPTC (Eptam 7-E); MC33 = methyl bromide 67%/chloropicrin 33%.

[†]Means do not differ significantly ($P \le 0.10$) from one another.

[‡]Means followed by the same letter do not differ significantly ($P \le 0.10$) according to Tukey's studentized range test.

Table 2. Mean seedling density, size, and dry weight by soil treatment for loblolly pine at Flint River Nursery (Bryomville, GA) in January 2000

Treatments*	Density (ft ²) [†]	Root collar diameter (in) [†]	Height (in) [†]	Root weight (oz) [†]	Top weight (oz) [†]
CH300	22.5	0.141	9.55	0.024	0.094
CH300 E	22.3	0.139	9.69	0.024	0.092
CH150	21.7	0.146	10.08	0.025	0.099
CH150 E	21.1	0.145	9.70	0.025	0.095
MC33	22.5	0.139	9.75	0.021	0.094
Е	22.9	0.140	9.90	0.024	0.089
Control	22.5	0.141	9.85	0.026	0.093

*CH300 = 100% chloropicrin at 300 lb/ac; CH150 = 100% chloropicrin at 150 lb/ac; E = EPTC (Eptam 7-E); MC33 = methyl bromide 67%/chloropicrin 33%.

[†]Means do not differ significantly ($P \le 0.10$) from one another.

Table 3. Mean seedling density, size, and dry weight by seed treatment with *Paenibacillus macerans* for loblolly pine at Flint River Nursery (Bryomville, GA) in November 1998

Seed Treatment	Density (ft ²) *	Root collar diameter (in) *	Height (in) [†]	Root weight (oz) *	Top weight (oz) *
No	21.4	0.170	8.41a	0.024	0.109
Yes	21.7	0.166	7.76 b	0.025	0.105

*Means do not differ significantly ($P \le 0.10$) from one another.

[†]Means differ significantly ($P \le 0.10$) according to Tukey's studentized range test.

Table 4. Mean seedling density, size, and dry weight by seedtreatment with *Paenibacillus macerans* for loblolly pine at FlintRiver Nursery (Bryomville, GA) in January 2000

Seed Treatment	Density (ft ²) *	Root collar diameter (in) [†]	Height (in) *	Root weight (oz) [†]	Top weight (oz) [†]
No	21.7 a	0.143	9.94 a	0.025	0.095
Yes	22.7 b	0.141	9.63 b	0.023	0.092

*Means differ significantly (P \leq 0.10) according to Tukey's studentized range test.

^TMeans do not differ significantly ($P \le 0.10$) from one another.

Table 5. Nutsedge plants per ft² atFlint River Nursery (Bryomville, GA)

Treatments*	1998 [†]	1999 [†]
Control	1.75a	1.98a
E	0.69ab	1.73ab
CH150	0.02b	0.35ab
CH150 E	0.04b	1.00ab
CH300	0.00b	0.08b
CH300 E	0.00b	0.13b
MC33	0.00b	0.25ab

*CH300 = 100% chloropicrin at 300 lb/ac; CH150 = 100% chloropicrin at 150 lb/ac; E = EPTC (Eptam 7-E); MC33 = methyl bromide 67%/chloropicrin 33%.

[†]Means followed by the same letter do not differ significantly ($P \le 0.10$) according to Tukey's studentized range test.

E. A. Hauss Nursery

Funigation and EPTC soil treatments did not affect seedling density or seedling size (Table 6). Seedling density was higher in plots with the PGPR seed treatment, but seedling size was significantly lower with the seed treatment

(Table 7).

The population density of *Fusarium spp.* within the soil was less initially in the CH150 lb/ac and MC33 treatments; however, at the end of the year, only the CH300E treatment had a lower density of *Fusarium* spp. than the controls. The populations of *Pythium* spp. within the soil were lower initially in most fumigation treatments; however, at the end of the year, only MC33 was significantly lower than the controls. Plant nematodes were rarely found in any plots. The population of nutsedge plants was very low; however, there were no nutsedge plants observed in plots treated with MC33 or EPTC.

Table 6. Mean seedling density, size, and weight by
soil treatment for loblolly pine at E. A. Hauss Nursery
(Atmore, GA) in November 1998

Treatments*	Density (ft ²) [†]	Root collar diameter (in) [†]	Root weight (oz) [†]	Top weight (oz) [†]
CH300	22.2	0.188	0.030	0.111
CH300 + E	23.5	0.175	0.024	0.089
CH150 + E	23.1	0.178	0.027	0.095
CH150	23.3	0.180	0.024	0.105
MC33	23.0	0.180	0.027	0.098
E	23.8	0.172	0.028	0.092
Control	23.0	0.181	0.028	0.100

*CH300 = 100% chloropicrin at 300 lb/ac; CH150 = 100% chloropicrin at 150 lb/ac; E = EPTC (Eptam 7-E); MC33 = methyl bromide 67%/chloropicrin 33%.

^TMeans do not differ significantly ($P \le 0.10$) from one another.

Table 7. Mean seedling density, size, and weight byseed treatment with Paenibacillus maceransfor loblolly pine at E. A. Hauss Nursery (Atmore, GA) inNovember 1998

Seed Treatment	Density (ft ²) *	Root collar diameter (in) [†]	Root weight (oz) [†]	Top weight (oz) *
No	22.5 b	0.181	0.028	0.103 a
Yes	23.8 a	0.177	0.026	0.095 b

*Means differ significantly ($P \le 0.10$) according to Tukey's studentized range test.

^TMeans do not differ significantly ($P \le 0.10$) from one another.

Carter Nursery

Seedling density was initially greater in the plots fumigated with M80/CH 150 than the control plots; however, by the end of the year these differences were no longer significant for either seedling density or size (Table 8). Seedling density was significantly less in plots with PGPR seed treatment compared to untreated seed (Table 9). There was a corresponding increase in seedling size with the reduction in seedling density in the seed treatments plots.

Table 8.Mean seedling density, size, and dry weight by soiltreatment for loblolly pine at Carter Nursery (Chatsworth, GA)in November of 1999

Treatments*	Density (ft ²) [†]	Root collar diameter (in) [†]	Root weight (oz) [†]	Top weight (oz) [†]
CH300	20.3	0.201	0.034	0.123
CH150 E	19.4	0.197	0.033	0.109
CH300 E	20.5	0.201	0.031	0.120
CH150	19.6	0.201	0.036	0.116
M80/CH150	22.5	0.197	0.029	0.102
MC33	21.1	0.205	0.037	0.131
E	20.3	0.197	0.029	0.109
Control	20.5	0.197	0.032	0.109

*CH300 = 100% chloropicrin at 300 lb/ac; CH150 = 100% chloropicrin at 150 lb/ac; E = EPTC (Eptam 7-E); M80 = metam sodium at 80 lb/ac; MC33 = methyl bromide 67%/chloropicrin 33%.

Table 9.Mean seedling density, size, and dry weight byPGPR seed treatment with Paenibacillus macerans forloblolly pine at Carter Nursery (Chatsworth, GA) inNovember 1999

Seed Treatment	Density (ft ²) *	Root collar diameter (in) [†]	Root weight (oz) *	Top weight (oz) *
No	21.6 a	0.195	0.031 b	0.109 b
Yes	19.4 b	0.203	0.034 a	0.120 a

*Means followed by the same letter do not differ significantly (P ≤ 0.10) according to Tukey's studentized range test.

^TMeans do not differ significantly ($P \le 0.10$) from one another.

The population density of *Fusarium spp.* was significantly lower in CH300 plots than the unfumigated EPTC plots after fumigation. The population density of *Pythium spp.* were significantly less in the CH300, M80/CH150, and MC33 treatments than the controls immediately following fumigation; however, only the M80/CH150 treatment had significantly lower levels by the end of the year. Parasitic plantnematodes were reduced by all fumigants, but by November 1999 there were no significant differences among treatments. Nutsedge plants were rare at Carter Nursery.

DISCUSSION

The various fumigation treatments had little influence on seedling bed densities, and differences in seedling size only occurred at the Flint River Nursery, where seedling size was greater in some of the chloropicrin treatments than the controls in the first year. This lack of consistent differences among treatments was most likely due to the absence of insect and disease problems at any of the nurseries. Although fumigation has primarily been used for pest control in forest-tree nurseries, the practice can also affect soil nutrient availability. Fumigation has been shown to influence the availability of nitrogen (Hansen and others 1990), manganese (Alexander 1967), and phosphorus (Ingestad and Molin 1960). In addition, fumigation can also change the soil microbiota (Hansen and others 1990) and favor the presence of fungi that may be beneficial for seedling growth (Ingestad and Nilsson 1964). The increase in seedling quality in the first year following fumigation at the Flint River Nursery may have been related more to changes that occurred to nontarget soil microbiota and soil chemical factors than the actual control of pests. The PGPR seed treatments can be beneficial to loblolly pine seedling emergence and growth as well as reduce seedling growth (Enebak and others 1998). The complex interactions of PGPR with the seedling and the soil environment have led to variable results (Enebak and others 1998). In the current study, the PGPR seed treatments in the three nurseries resulted in inconsistent effects on seedling density and quality. While the PGPR seed treatments increased seedling density at the Hauss Nursery, the treatments had the opposite effect at the Carter nurserv and no affect at the Flint River Nurserv. The effect of seed treatment on seedling size was largely confounded by seedling density, except at the Flint River Nursery where seedling height was significantly less in the PGPR treatments. A better understanding is needed of the mechanisms by which PGPR seed treatments affect seed germination and seedling growth in southern pine nurseries if this treatment is to be operationally applied. Most weeds, with the exception of nutsedge. were effectively managed through the operational weed control programs used at the nurseries. Populations of nutsedge varied greatly among the nurseries, with the Flint River Nursery having the greatest population and the Carter Nursery having almost none. The effectiveness of

fumigants and EPTC to control nutsedge may be tied to soil texture. In the sandy-textured soil of the Flint River Nursery, chloropicrin provided good control of nutsedge, while EPTC did not. The opposite trend occurred in the heavier textured soils of the Hauss Nursery, where nutsedge was observed in plots treated with chloropicrin and not in lots treated with EPTC. The success of fumigation in reducing soilborne fungi may also be tied to soil texture. The population densities of Fusarium and Pythium spp. were initially low in all fumigated plots of the Flint River Nursery. In the heavier soils at the Hauss and Carter nurseries, the success of fumigation in reducing the population of Fusarium spp. was variable. Lower populations of *Pythium spp.* were obtained by most of the fumigants initially in the nurseries with heavier soils; however, by the end of the growing season only MC33 of M80/CH150 maintained a significant difference. Chloropicrin, alone and in combination with metam sodium, reduced soilborne pests as well as MC33 in most cases. The amount and type of fumigants needed for control of soilborne pests vary among nurseries depending on their soil condition and pest problems. In this study, chloropicrin at the lower rate was adequate for reducing soilborne fungi and nutsedge in the sandier soils at the Flint River Nursery. In the heavier textured soils of the Carter Nursery, the higher rate of chloropicrin improved the control of soilborne fungi. Evaluations of the treatments are continuing for a third year at the Flint River Nursery and for a second year at the Carter Nursery to determine the value of these treatments over 2 or 3 vears.

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