Evaluation of Systemic Fungicides for Control of *Cronartium quercuum* f. sp. *fusiforme* on Loblolly Pine Seedlings

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ABSTRACT

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Four experimental systemic fungicides applied individually as preplant incorporated soil treatments and as foliar sprays were evaluated for controlling fusiform rust on loblolly pine (*Pinus taeda*) seedlings. Seedlings were inoculated with *Cronartium quercuum* f. sp. *fusiforme* and were evaluated for gall formation after 36 wk. Of the four fungicides tested, only triadimefon (Bayleton) effectively controlled fusiform rust. No stem galls developed on seedlings in soil in which triadimefon had been incorporated as a preplant treatment at a rate of 2 kg a.i./ha. Triadimefon applied as a foliar spray at a rate of 0.56 kg a.i./ha eradicated rust infections that occurred up to 7 days before treatment and provided protection against the pathogen for 14–21 days after application. No phytotoxic effect of triadimefon on seedling growth was observed, and no significant differences were observed between the fungicide-treated seedlings and the controls concerning the presence of basidiocarps of the mycorrhizal symbiont *Thelephora terrestris*.

Fusiform rust, caused by *Cronartium quercuum* (Berk.) Miyabe ex Shirai f. sp. *fusiforme* Burdsall & Snow, has been controlled in forest nurseries in the southern United States for many years with the contact fungicide ferbam (1). Although effective in preventing infection of pine seedlings by *C. quercuum* f. sp. *fusiforme*, ferbam requires frequent applications (often 20–50) during the spring spore-flight season. The number of applications depends on the weather; during rainy periods, the fungicide is applied more frequently than the normal twice-weekly sprays (1). Because ferbam has no eradicative action, it must be present on the foliage at the time spores land to prevent infection.

Labor and equipment costs of applying ferbam to control fusiform rust in nurseries and the damage to nursery beds from maneuvering spray equipment through the nursery during wet weather are major concerns of nursery managers. This paper reports results of preliminary screening tests with four experimental systemic fungicides to identify alternatives to ferbam for control of fusiform rust. A preliminary report has been published (3).

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MATERIALS AND METHODS

The USDA Forest Service Rust Testing Center near Asheville, NC, supplied loblolly pine (*Pinus taeda*) seed from a family (WV 11-23) known to be highly susceptible to *C. quercuum* f. sp. *fusiforme* and standard pasteurized soil mix used at the Center to grow seedlings for rust resistance tests.

Establishment of seedlings. Seeds were soaked for 2 days in tap water, stored moist at 5 C for 6 wk, soaked for 10 min in 20% (v/v) hydrogen peroxide, and then planted in greenhouse flats containing autoclaved vermiculite. As seedlings emerged, they were transplanted into plastic planter trays (34 × 13 × 10 cm; #135 Planter, Ball Seed Co., West

Chicago, IL) each containing 4 kg of soil mix. Twenty seedlings (2 rows of 10 each) were transplanted into each tray with a multiprobe template to ensure equal spacing between seedlings.

Test design and application of fungicides. The test was arranged in completely randomized blocks with five replicates (trays) per treatment. Fungicides tested were: triadimefon (Bayleton 50 WP, Mobay Chemical Corp.); B-([1,1'-Biphenyl]-4-yloxy)-α-(1,1-dimethyl-ethyl)-1H-1,2,4-triazole-1-ethanol (Bay-Kwg 0599 25WP, Mobay Chemical Corp.); α-(2-Chlorophenyl)-α-(4-fluorophenyl)-5-pyrimidinemethanol (EL-228 0.75 EC, Elanco Products Co.); and 1-[2-(2,4-Dichlorophenyl)-4-ethyl-1,3-dioxolan-2-yl]methyl]1H-1,2,4-triazole (CGA-64251 21.5WP, Ciba-Geigy).

Treatments 1 and 2 were untreated controls. Treatments 3–10 were triadimefon, Bay-Kwg, EL-228, and CGA-64251 incorporated in the soil before transplants at rates of 2 and 5 kg a.i./ha. Treatments 11–20 were triadimefon and Bay-Kwg foliar sprays applied at 0.56 kg a.i./ha, and treatments 21–30 were EL-228 and CGA-64251 foliar sprays applied at 0.15 kg a.i./ha, each applied once either 21, 14, or 7 days before inoculation or 2 or 7 days after inoculation.

Foliar sprays were applied with a laboratory belt-type sprayer calibrated to deliver a volume of 247 L/ha (26.4 gal/A). No spreader-sticker was used with any fungicide. For fungicides incorporated into the soil, 150 ml of a suspension or emulsion containing sufficient fungicide to provide a rate of 0.89 or 2.23 μg a.i./g of soil (equivalent to 2 and 5 kg/ha, respectively) was hand-mixed into the 4 kg of soil from each tray. No more than 3 days elapsed between incorporating the fungicide into the soil and transplanting the seedlings.

Inoculation of seedlings. Seedlings were brought to the Rust Testing Center 39 days after transplantation to be inoculated with *C. quercuum* f. sp. *fusiforme* on the 40th day. The inoculum was prepared as previously described (4), with a virulent isolate (L-6) of *C. quercuum* f. sp. *fusiforme*. The aqueous inoculum contained 35,000 basidiospores/ml. Inoculation was accomplished on a moving belt line calibrated to deliver 10 ml of the spore suspension to each tray of seedlings through two spray nozzles. Immediately after inoculation, the trays were placed in an incubation chamber at 20–22 C for 24 hr at relative humidity above 95%. Upon removal from the incubation chamber, trays with seedlings that had been treated with the fungicides before inoculation and one set of untreated controls were placed in a greenhouse at the Rust Testing Center. The second set of untreated controls and trays of seedlings scheduled for post-inoculation treatment with fungicides were brought to a greenhouse at Auburn University, where they were treated and maintained.

Seedling evaluation. Each seedling was examined individually for fusiform rust galls 36 wk after inoculation; only definite stem swellings were counted as galls. The height of each seedling was measured and recorded to determine whether the fungicide treatments inhibited seedling growth. The presence or absence of basidiocarps of the mycorrhizal symbiont *Thelephora terrestris* was recorded for each tray of seedlings to indicate the test fungicides' effects on the development of mycorrhizae on the seedlings.

All data were subjected to analysis of variance, and means were compared for significant differences at the 1% level of probability by Duncan's multiple range test.

Table 1. Effect of systemic fungicides on the incidence of fusiform rust galls on greenhouse-grown loblolly pine seedlings 36 wk after inoculation

Treatment[x]	Rate (kg a.i./ha)	Application of foliar spray[y]	Seedlings with galls[z] (%)
Seedlings treated before inoculation			
Control	...	...	32 a
Triadimefon PPI	2.00	...	0 b
Triadimefon PPI	5.00	...	0 b
Triadimefon FS	0.56	−7	0 b
Triadimefon FS	0.56	−14	0 b
Triadimefon FS	0.56	−21	1 b
Bay-Kwg PPI	2.00	...	35 a
Bay-Kwg PPI	5.00	...	26 a
Bay-Kwg FS	0.56	−7	29 a
Bay-Kwg FS	0.56	−14	34 a
Bay-Kwg FS	0.56	−21	23 a
EL-228 PPI	2.00	...	43 a
EL-228 PPI	5.00	...	36 a
EL-228 FS	0.15	−7	37 a
EL-228 FS	0.15	−14	26 a
EL-228 FS	0.15	−21	32 a
CGA-64251 PPI	2.00	...	21 a
CGA-64251 PPI	5.00	...	35 a
CGA-64251 FS	0.15	−7	29 a
CGA-64251 FS	0.15	−14	22 a
CGA-64251 FS	0.15	−21	24 a
Seedlings treated after inoculation			
Control	...	...	31 a
Triadimefon FS	0.56	+2	0 b
Triadimefon FS	0.56	+7	0 b
Bay-Kwg FS	0.56	+2	22 a
Bay-Kwg FS	0.56	+7	32 a
EL-228 FS	0.15	+2	32 a
EL-228 FS	0.15	+7	37 a
CGA-64251 FS	0.15	+2	23 a
CGA-64251 FS	0.15	+7	34 a

[x] PPI = preplant soil incorporated; FS = foliar spray.
[y] Days before (−) or after (+) inoculation.
[z] Average of five trays of seedlings. Means followed by the same letter do not differ according to Duncan's multiple range test ($P = 0.01$).

RESULTS

Seedlings treated with triadimefon before inoculation with *C. quercuum* f. sp. *fusiforme* were free of galls after 36 wk in all cases except where triadimefon was applied as a foliar spray 21 days before inoculation (1% galled) (Table 1, top). No significant differences were observed between controls and seedlings treated with the other three test fungicides. No galls were observed on seedlings treated with triadimefon after inoculation, and no significant differences were observed between controls and seedlings treated with the other fungicides after inoculation (Table 1, bottom).

No phytotoxic effects of the test fungicides on seedlings and no significant differences in seedling height were observed among the various treatments (data not shown). Overall average seedling height was 23.7 cm after 36 wk. Although the distribution of fruiting bodies of *T. terrestris* was not uniform, no significant differences were observed among the various treatments after 36 wk (data not shown).

DISCUSSION

Results of this study indicate that triadimefon is effective against *C. quercuum* f. sp. *fusiforme* and that Bay-Kwg, EL-228, and CGA-64251 are not effective at the rates tested.

Triadimefon applied as a preplant incorporated soil treatment (40 days

before inoculation) provided complete protection against *C. quercuum* f. sp. *fusiforme* even at the lowest rate tested (2 kg/ha). These results suggest that nursery pine seedlings may be protected against fusiform rust with a single preplant incorporated soil application of triadimefon, thus eliminating foliar sprays altogether. Studies are currently under way at three Alabama forest nurseries to test this hypothesis.

A single foliar spray application of triadimefon at a rate of 0.56 kg/ha was effective in eradicating fusiform rust infections that occurred up to 7 days before application of the fungicide. In addition, seedlings were protected from infection by *C. quercuum* f. sp. *fusiforme* for 14–21 days after application of the triadimefon spray; even after 21 days, only 1% of the seedlings became infected. Greenhouse and field tests do not always agree (5); however, if triadimefon performs as well in the field as in the greenhouse, foliar applications of triadimefon every 4 wk at a rate of 0.56 kg/ha may be sufficient to protect pine seedlings from fusiform rust. Compared with 20–50 applications of ferbam at a rate of 2.24 kg/ha, this procedure would significantly decrease the amount of fungicide used annually in forest nurseries. The savings in labor and equipment usage would also be substantial. Further studies are being conducted to determine if even lower rates of triadimefon are effective in protecting pine seedlings from *C. quercuum* f. sp. *fusiforme*. In any case, the effective rates of triadimefon reported in this study are substantially lower than those previously reported (2) for the experimental systemic fungicide benodanil.

The distribution of fruiting bodies of *T. terrestris* among the treatments at the end of the study does not provide any information concerning the fungitoxic or fungistatic properties of triadimefon toward this mycorrhizal symbiont. However, the fact that fruiting bodies were present at the end of the study and that no significant differences were observed in their distribution among treatments indicates that any effect triadimefon may have on *T. terrestris* was not evident after 36 wk.

Triadimefon is currently registered for use in control of azalea blight (*Ovulinia azaleae*), and the prospect of obtaining registration for control of fusiform rust is promising.

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