Fungicide Trials for Control of Phomopsis Canker of Douglas-Fir at a Northern California Nursery

John Kliejunas

Plant pathologist, USDA Forest Service, Pacific Southwest Region, State and Private Forestry, San Francisco, CA

Based on in vitro *evaluation of sixteen fungicides, five were field-tested for efficacy against phomopsis canker of 2 + 0 Douglas-fir (*Pseudotsuga menziesii *(Mirb.) Franco). All gave significantly better control than no treatment. Benomyl, the fungicide registered for phomopsis control, was more efficacious than mancozeb and imazalil but was not as effective as diniconazole or chlorothalonil.* Tree Planters' Notes 39(2):26-29; 1988.

Phomopsis canker of 2+0 Douglas-fir, caused by *Phomopsis occulta*, usually results in scattered and infrequent losses at Humboldt Nursery (USDA Forest Service) in coastal northern California. About 5% of the crop is lost each year. However, incidence of the disease has increased the last few growing seasons, Benomyl is registered and used for control of *Phomopsis juniperovora*-caused diseases (1-3), but the effectiveness of the fungicide against *P. occulta* had not been previously tested. Humboldt Nursery currently uses benomyl, but the efficacy of the treatment has been questionable.

The increase in disease, along with the cost of benomyl and recommended frequency of application, caused the Nursery to request an evaluation of alternative fungicide treatments. The objective was to evaluate alternative fungicides for control of Phomopsis canker of Douglas-fir at Humboldt Nursery, through *in vitro* laboratory screening and field trials.

Methods

Laboratory screening. An isolate of *P. occulta* from 2+0 Douglas-fir seedlings at Humboldt Nursery was used. The isolate was morphologically identical to other Douglas-fir isolates and to isolates obtained from western hemlock.

Sixteen fungicides were initially selected (table 1) to determine their effect on spore germination. The fungicides were recommended and supplied by Dr. A.H. McCain, extension plant pathologist, University of California at Berkeley. Three concentrations (1, 10, and 100 ppm; active ingredient) of each fungicide were incorporated into 2% water agar (2 g agar/100 ml water). Spores were obtained by adding 10 ml of sterile water to each sporulating culture and brushing gently with a brush to dislodge spores. One milliliter of the spore suspension was placed onto the surface of each plate containing 2% water agar and incorporated fungicide. Four replications were made for each fungicide concentration and for a control (2% water agar without fungicide). After 24 hours' incubation, the percentage of germination was determined for each treatment. Germ tube development was followed over an 11-day period.

Table 1—*Fungicides evaluated* in vitro *for control of Phomopsis canker of 2+ 0 Douglas-fir*

Common name	Trade name
Benodanil	BAS 3170F 50W
Benomyl	Benlate
Captan	Captan 50W
Chlorothalonil	Bravo W-75
Chlorothalonil	Bravo 500
Diniconazole	Spotless (Chevron 779 12.5% W)
Dithianon	Delan 75 WP
Fenarimol	Rubigan 50W
Imazalil	Fungaflor 20% EC
Iprodione	Chipco 26019
Mancozeb	Dithane M 45
Penconazole	Topas (CGA 71818 10W)
Prochloraz Mn complex	Sportak 50W
Triadimefon	Bayleton 50WP
Tri-basic copper sulfate	Tri-Basic
Vinclozolin	Ornalin 50W

The seven fungicides that were most effective in inhibiting or reducing germination compared to the control were selected to determine their effect on mycelial growth. Three concentrations (1, 10, and 100 ppm active ingredient) of each were incorporated into potato dextrose agar (PDA). A 0.5-mm plug of mycelium of *P. occulta* was removed from the perimeter of an actively growing colony of

PDA and placed on the fungicide-amended PDA in petri plate. Four replications were made for each fungicide concentration and for a control (PDA without fungicide). Radial growth of mycelium from each plug was measured after 3 days' incubation.

Field trials. Five of the most effective fungicides from the laboratory trial were tested in field trials in 1985 and again in 1986. A randomized complete block design with six treatments—control; benomyl at 1 lb product/ acre; imazalil at 20 ounces product/acre; diniconazole at 0.2 lb product/acre; chlorothalonil (Bravo 500) at 5 pints product/ acre; and mancozeb at 2 lbs product/acre—was used. The six treatments were replicated 10 times down a bed of 2+0 Douglas-fir in 1985, and replicated 8 times in 1986. Each replicate covered 24 feet of bed (six 4 by 4 ft plots), with each treatment assigned randomly in each replicate.

The 1985 trial was initiated in June. Fungicide treatments continued at monthly intervals through September (four applications). In 1986, monthly fungicide treatments began in May and continued through August (four applications). Efficacy of treatments was determined by counting cankered seedlings monthly. After the final count, the total number of seedlings in each plot was counted, and percentage of seedlings cankered in each treatment determined.

Results and Discussion

Laboratory screening. Spore germination percentages are presented in table 2. Spores with emerging germ tubes after 24 hours' incubation were considered germinated. With some treatments (imazalil, prochloraz, diniconazole, penconazole and benomyl in particular), the germ tubes had limited development or became distorted.

Results of the effect of 7 fungicides on mycelial growth

Table 2—*Germination percentage of* Phomopsis occulta *spores after 24 hours on fungicide-amended media*

Fungicide	Percent germination at 3 concentrations		
	1 ppm	10 ppm	100 ppm
Chlorothalonil (Bravo 500)	0	0	0
Chlorothalonil (Bravo W-75)	71	0	0
Imazalil	41*	43*	0
Mancoxzeb	94	29	0
Captan	92	89	0
Dithianon	84	87	0
Prochloraz	80	62*	31*
Diniconazole	71	48*	28*
Penconazole	87*	89*	67*
Benomyl	95	88*	88*
Benodanil	82	80	27*
Tribasic copper sulfate	94	95	84*
Fenarimol	90	88	86*
Iprodione	91	90	89
Vinclozolin	92	91	88
Triadimefon	95	95	93

Control value = 98% germination.
* Germ tube inhibition or distortion.

Table 3—*Average radial growth of* Phomopsis occulta *after 3 days on fungicide-amended potato dextrose agar*

Fungicide	Radial Growth (mm) at 3 concentrations		
	1 ppm	10 ppm	100 ppm
Benomyl	0	0	0
Imazalil	0	0	0
Prochloraz	0	0	0
Diniconazole	1.1*	0	0
Chlorothalonil (Bravo 500)	2.2	0	0
Chlorothalonil (Bravo W-75)	15.5	1.7	0
Mancozeb	24.0	2.7	0

*Average of four replications.

are presented in table 3. After 11 days, no growth had occurred on benomyl-amended PDA at 1, 10, and 100 ppm, on imazalil-amended and prochloraz-amended PDA at 10 and 100 ppm, and on diniconazole and the Bravo 500 formulation of chlorothalonil-amended media at 10 and 100 ppm.

To determine if the fungicides were fungitoxic or fungistatic, plugs where no mycelial growth had occurred into the fungicide-amended PDA were removed, placed on PDA, and examined after 7 days (table 4). Benomyl and imazalil were fungitoxic at all concentrations. Prochloraz and diniconazole were not fungitoxic at 1 ppm, but were at 10 and 100 ppm, with mycelial growth occurring after the plug was removed from the fungicide-amended media. Chlorothalonil was fungitoxic only at 100 ppm.

Field trials. Based on the laboratory assays, 5 fungicides were selected for field trials. Levels of disease incidence during the 1985 trial were not sufficient to compare treatments as cankered seedlings in plots ranged from less than 1% to about 3%.

Results of the 1986 trial are presented in table 5. All 5 fungicides gave significantly better control of phomopsis canker than no treatment. Benomyl was more efficacious than mancozeb or imazalil, but was not as effective as diniconazole or chlorathalonil.

Table 4—*Mycelial growth of* Phomopsis occulta *on potato dextrose agar after 11 days on fungicide-amended potato dextrose agar*

Fungicide	Mycelial growth at 3 concentrations		
	1 ppm	10 ppm	100 ppm
Benomyl	–	–	–
Imazalil	–	–	–
Prochloraz	+	–	–
Diniconazole	+	–	–
Chlorothalonil (Bravo 500)	+	+	–

+ = Growth, – = no growth.

Table 5—*Percentage of 2+0 Douglas-fir seedlings cankered as a result of* Phomopsis occulta *infection at the Humboldt Nursery*

Treatment	Percent cankered
Diniconazole	1.9 a
Chlorothalonil	2.1 a
Benomyl	4.0 b
Mancozeb	5.1 c
Imazalil	5.3 c
Control	9.1 d

Means followed by the same letter are not significantly different at the 1% level, according to Duncan's multiple range test. Means are the average number of seedlings cankered in eight replicate plots.

Conclusions

Benomyl, the fungicide registered for control for *Phomopsis* spp., was efficacious under the conditions for the study. The label for benomyl suggests treatment at 10- to 14-day intervals. Because we used 28- to 30-day intervals, the fungicide may be even more effective if used according to label recommendations.

Chlorothalonil and diniconazole were more effective than benomyl. Chlorothalonil is registered for use on conifers in nurseries and could be used (5 pints product/acre, applied at monthly intervals throughout the growing season) as a replacement for benomyl for control of phomopsis canker. Diniconazole, although not currently registered for use in nurseries in California, has some systemic properties and therefore may be more effective than chlorothalonil or benomyl in preventing secondary infections during rainy periods.

Although fungicide treatments for control of phomopsis canker were effective under the conditions of this trial, results should be interpreted with caution because of the low incidence of disease during the two seasons of field testing. Incidence of phomopsis canker in nontreated portions of the test bed in the 1986 trial was low (9.1%), with little variation among the 8 replications. During years more favorable for disease development, the relative efficacy of the

fungicides evaluated in relation to each other may vary.

Most of the cankers during these trials had caused dieback of lateral tips, with only about 10% of these infections resulting in death of the seedlings or cull due to killing of terminals. This low level of disease severity, along with the low levels of overall disease in test beds and production beds, suggests that no fungicide treatment the past two growing seasons may have been more economical than treatment with preventive fungicides. Observations in 1986 suggested that disease levels may have been lower than in previous years because seedlings were not top-pruned, a practice that creates wounds for infection.

Literature Cited

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