Fungicide Trials for Control of Cypress Canker on Port-Orford-cedar

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Introduction

In November 2000, Cypress canker was identified on Port-Orford-cedar seedlings at the Dorena Genetic Resource Center in Cottage Grove, Oregon. This disease is caused by the fungus *Seiridium cardinale* (W. Wagener) Sutton & I. Gibson. Initial symptoms of the disease included dieback of branches and copious resin flow from sunken cankers on the boles. In many cases the infection court appeared to be at the point where small branchlets joined the bole. Some of the cankers eventually became large and swollen, with broken bark. Mortality occurred in some severely infected trees due to mechanical failure of the bole at the canker site.

Cypress canker caused by *Seiridium cardinale* was first identified on planted Monterey cypress in Palo Alto, California in 1928 (Wagener 1939). It has since become widespread in Monterey cypress planted on dry inland sites (Scharpf 1993). The disease has caused widespread damage to *Cupressus sempervirens* in the Mediterranean region of Europe and to planted cypresses and cedars, including Port-Orford-cedar, in New Zealand (Graniti 1998). A similar species *Seiridium unicorne,* causes symptoms that are indistinguishable from those caused by *S. cardinale* and is also known as Cypress canker. It is very damaging in plantations of Leyland cypress grown for Christmas trees in the southeastern United States (personal communication, Dr. Jean Woodward, Plant Pathologist, University of Georgia).

The first outbreak of cypress canker at Dorena apparently occurred in 1996, although the causal agent was not identified at that time (Sandquist 1996). It is not clear whether *S. cardinale* is native to the area around Dorena or was imported on seedlings or cuttings. In both 1996 and 2000 the infected seedlings had been kept outdoors at Dorena for a period of time during the winter. After *S. cardinale* was identified following the outbreak in 2000, several large incense cedars and one Leyland cypress on the lawn at the Center were found to be infected, as were Port-Orford-cedar saplings imported as seedlings from California in 1996 and planted in outdoor raised beds. Cypress canker has been reported but not confirmed on Port-Orford-cedar in natural stands in the vicinity of Hiouchi, California; and on incense cedar in the Willamette Valley, Oregon (personal communication, Jack Marshall, California Department of Forestry and Fire Protection and Alan Kanaskie, Oregon Department of Forestry).

All visibly infected seedlings in the greenhouses were destroyed in Fall 2000. Each year since then a small number of new infections have appeared on Port-Orford-cedar seedlings, both in the greenhouse and outdoors. Until the source of inoculum can be determined and eliminated, chemical treatments will be needed to control this disease.

Objectives

The objective of these trials was to identify fungicides that provide the best control of cypress canker on Port-Orford-cedar, and provide data for fungicide labels and Special Local Needs (SLN) registration, if necessary.

Materials and Methods

Research by McCain (1984) was used as the basis for selecting the fungicides we tested (Table 1). McCain found that chlorothalonil and benomyl provided a high degree of control of cypress canker on Leyland and Monterey cypress. Tribasic copper sulfate (53% Cu) was not effective in controlling spore germination, mycelial growth or formation of cankers. We chose thiophanate methyl to replace benomyl because benomyl is no longer registered. Both benomyl and thiophanate methyl are converted by plants into the same active fungicidal compound, carbendazim (Pscheidt and Ocamb 2001). Although tribasic copper was not recommended by McCain, we included copper hydroxide in the trials because fixed copper was the only chemical registered in Oregon for control of cypress canker when we started (Pscheidt and Ocamb 2001), and because it is effective in controlling other canker diseases on conifer seedlings. In each trial we applied the fungicides at the mid-point rate recommended on the manufacturer's label (1x) and at twice the recommended rate (2x).

Table 1. Fungicides used in Trials 1 and 2

Treatment	Fungicide	Mode	Active ingredient	Rate	Dosage (a.i.)
1	Cleary 3336F	systemic	thiophanate methyl	1x	0.9 gm/l
2	Cleary 3336F	"	"	2x	1.8 gm/l
3	Daconil Weather Stik	protectant	chlorothalonil	1x	1.9 gm/l
4	Daconil Weather Stik	"	"	2x	3.8 gm/l
5	Champ Formula 2	protectant	copper hydroxide	1x	0.45 gm/l
6	Champ Formula 2	"	"	2x	0.9 gm/l
7	Control			-	-

In the first trial, conducted in November 2001, 224 Port-Orford-cedar seedlings were treated with the fungicides, held for 24 hours, and then exposed to a natural source of inoculum by placing them underneath the infected Port-Orford-cedar saplings in the outdoor raised beds at Dorena. The fungicides were reapplied at the minimum interval on the manufacturer's label. Each treatment and control was replicated twice. Each replication contained 16 seedlings in Ray Leach #10 tubes held together in a rack. The seedlings were exposed until mid-December, and then incubated in a protected location until May 2002.

In the second trial, conducted in May 2003, 224 larger Port-Orford-cedar seedlings were used. Each treatment and the control were replicated twice. Each replication contained 16 seedlings in one-gallon pots held together in a rack. The seedlings were wounded, treated with the fungicides, held for 24 hours, and then artificially inoculated with *S. cardinale*. The control trees were wounded and inoculated but were not treated with fungicides. To ensure that the inoculation would be successful, each seedling was inoculated in three places on the stem using three different inoculation methods (Table 2).

Table 2. Inoculation methods used in Trial 2

Inoculation	Wound method	Fungus application
1.	wound	mycelial plug
2.	no wound	spore suspension
3.	wound	spore suspension

The seedlings were wounded by exposing a 2 millimeter square area of cambium using a sterile scalpel. They were then sprayed to wet with the fungicides. After inoculation, the sites were covered with strips of sterile moist paper towels and Para-film to maintain 100 percent humidity. The seedlings were incubated in a greenhouse for 90 days. The paper towel/Para-film coverings were removed after

two weeks. After 90 days the seedlings were examined for evidence of canker formation. Isolations were made from cankers on a subsample of seedlings to determine whether they had been caused by *S. cardinale*.

Results and Discussion

Trial 1. The seedlings were examined in May, June, July and November 2002. No evidence of cankers were found on any of the seedlings. Either fruiting bodies were not produced by the cankers on the trees serving as the inoculum source, or the weather was not favorable for infection during the time the seedlings were exposed. It is also possible that the seedlings were too small.

Trial 2. After 90 days each seedling was examined for evidence of canker formation. The inoculations with wounds and plugs of mycelium produced cankers consistently (Table 3). Ninety-four percent of the control trees inoculated using this method showed evidence of cankers, compared to less than 25 percent with cankers using spore inoculation methods, with or without wounding.

Table 3. Number of seedlings with cankers by treatment and inoculation method

Treatment	**Rate**	**Number seedlings**	**Number of seedlings with cankers by inoculation method**		
			wound, mycelial plug	no wound, spores	wound, spores
Cleary 3336F	1x	32	1 a[1]	0	0
Cleary 3336F	2x	32	0 a	0	1
Daconil	1x	32	14 b	0	0
Daconil	2x	32	4 a	0	0
Champ	1x	32	30 c	0	0
Champ	2x	32	29 c	0	0
Control		32	30 c	1	7

1. Treatments with the same letter were not statistically different

Statistical analysis using Chi-squared tests of independence in two-by-two tables (Ramsey and Schafer 1997) showed there were significant differences among the fungicide treatments for seedlings inoculated with wounds and mycelial plugs (Table 3). The differences were considered statistically significant when p-values were less than 0.05.

Only one seedling treated with Cleary at the 1x rate had a canker. This was significantly fewer cankers than seedlings treated with Daconil 1x, Champ or the control seedlings. Seedlings treated with Daconil 1x had significantly fewer cankers than those treated with Champ or the control seedlings. Almost all the seedlings treated with Champ and the control seedlings had cankers. There was no significant difference between the number of cankers on seedlings treated with Champ and the control seedlings.

Seedlings treated with Daconil 2x had significantly fewer cankers than seedlings treated with Daconil at the 1x rate. There was no significant difference in the number of cankers between seedlings treated with Cleary 1x and Cleary 2x, or between Cleary 1x and Daconil 2x. However, there were slightly fewer cankers on seedlings treated with Cleary 2x than with Daconil 2x, although the difference was of borderline significance.

Seedlings inoculated with spores, either with or without wounding, and treated with fungicides had fewer cankers than the control seedlings, so it is possible that all three fungicides gave some measure of protection from development of cankers from spore infections. However, so few cankers developed in either the control or treated seedlings that it was not possible to determine whether the lack of cankers in the treated seedlings was due to the fungicides or to unsuccessful inoculations.

Each seedling was also examined for evidence of possible phytotoxicity caused by the fungicide treatments (Table 4). Seedlings with discolored foliage, lesions on the foliage, cracks in the bark, or dead tips were rated as having evidence of phytotoxicity. A total of thirteen seedlings showed damage that may have been the result of phytotoxicity. Six of the damaged seedlings had been treated with Cleary 3336F at the 2x rate and five had been treated with Daconil at the 2x rate. Two had been treated with Champ at the 1x rate. The damage to three of the seedlings treated with Cleary consisted of longitudinal cracks in the bark on the stems. This may have been due to heat rather than the effects of the Cleary, as these seedlings were right next to the greenhouse wall where it was very hot when the sun shone directly on the plastic.

Table 4. Seedlings with evidence of possible phytotoxicity

Treatment	Rate	Number seedlings	Number of seedlings with evidence of phytotoxicity
Cleary 3336F	1x	32	0
Cleary 3336F	2x	32	6
Daconil	1x	32	0
Daconil	2x	32	5
Champ	1x	32	2
Champ	2x	32	0
Control		32	0

Conclusion

Cleary 3336F applied at the rate recommended on the manufacturer's label was very effective at preventing development of cypress canker and with no evidence of phytotoxicity. It was significantly more effective than Daconil or Champ applied at the recommended rates. Daconil was as effective as Cleary when applied at twice the recommended rate, but at this rate damage was observed that may have been due to phytotoxicity. Champ did not appear to provide any protection from cypress canker even when applied at twice the rate recommended on the label.

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