SEED-TREATMENT FUNGICIDES FOR CONTROL OF CONIFER DAMPING-OFF: LABORATORY AND GREENHOUSE TESTS, 1967

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Abstract

Sixty-nine seed treatment chemicals were tested in laboratory bioassays, 13 in laboratory germination tests, and 9 in greenhouse damping-off control tests. Damping-off of red pine seedlings was effectively controlled with thiram (2.0 g/g of seed, and 0.5 g/g of seed), captan (2.0 g/g of seed), and Polyram (0.5 g/g of seed). Damping-off of jack pine and white spruce seedlings was also controlled; however, the seed treatment chemicals giving control were also phytotoxic.

Introduction

The main species of conifers grown in prairie nurseries are white spruce (<u>Picea glauca</u> (Moench) Voss), Colorado spruce (<u>P. pungens Engelm.</u>), Scots pine (<u>Pinus sylvestris L.</u>), jack pine (<u>P. banksiana</u> Lamb.) and red pine (<u>P. resinosa</u> Ait.). Dampingoff has been a problem on seedlings of all species of conifers at one time or another (1, 3). The most important pathogens involved are <u>Rhizoctonia solani</u> Kühn, <u>Pythium debaryanum</u> Hesse, <u>P. ultimum</u> Trow, <u>Phytophthora cactorum</u> (Leb. & Cohn) Schroet., and several species of <u>Fusarium</u> and <u>Cylindrocarpon</u> (3).

Numerous attempts to control conifer dampingoff with chemicals have been made, but only a few seed-treatment chemicals, such as captan and thiram, have proved useful (2), and even these do not completely control the disease. The present screening program of seed-treatment chemicals is designed to evaluate by laboratory, greenhouse, and field tests the activity of these chemicals against three major pathogens, <u>Pythium, Rhizoctonia</u> and <u>Fusarium</u>. This report presents results of preliminary tests of the effect of a number of seed treatment chemicals on the in vitro growth of mycelium and germination of conifer seed, and on dampingoff control in natural soil in the greenhouse.

Materials and methods

Laboratory bioassay Sixty-nine seed-treatment chemicals (Table 1)* were tested for inhibition of mycelium growth of isolates of <u>Pythium</u>, <u>a</u> **&**, and <u>Rhizoctonia</u> known to cause damping-off of conifer seedlings. A 5-mm disc of actively growing mycelium was placed at the center of a petri dish containing 25 ml of malt agar (30 g malt extract and 20 g Difco Bacto-agar in 1 liter of distilled water). Seed-treatment chemicals were suspended in acetone at rates of 10,200; 2,560; 640; 160; and $40 \,\mu g$ active chemical per, ml. Sterile 10-mm discs of Whatman No. 1 filter paper were infiltrated with the chemicals and placed on the agar 18-20 mm from the inoculum. Three discs were used for each petri dish. The presence and amount of inhibition were recorded after 3 days at room temperature for the <u>Pythium</u> and <u>Rhizoctonia</u> cultures, and after 5 days for the <u>Fusarium</u> cultures. Each chemical was tested twice and there were four replications per test.

Laboratory germination tests - Seedsof jack pine, white spruce, and red pine were pelleted with seed-treatment chemicals at rates of 0.25 and 1.0 g chemical per gram of seed. Dow Latex 512R was used as a binder at a rate of lg of a 10% (V/V) solution per 3 grams of seed. This material was used because preliminary tests showed that it inhibited germination less than methyl cellulose. In each treatment, 100 air-dried seeds of each tree species were placed on filter paper and incubated at 100% relative humidity in a germinator that provided alternating 8-hr periods of darkness at 20 C and 16-hr periods of light (70 ft-c, fluorescent) at 30 C. The germinated seeds were counted after 14 days. Each test was repeated at least once, and some were repeated three times.

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Chemicals were supplied by Stauffer Chemical Co. Ltd., Vancouver; Diamond Alkali Co., Painesville, Ohio; Naugatuck Chemicals, Elmira, Ont.; Niagara Brand Chemicals, Burlington, Ont.; DuPont of Canada Ltd., Montreal, Que.; Chemagro Corp., Kansas City, Mo.; American Cyanamid, New York, N. Y.; Sherwin-Williams Co. of Canada Ltd. (Green Cross Products), Montreal, Que.; Morton Chemical Co., Woodstock, Ill.; Chipman Chemical Ltd., N. Hamilton, Ont.; Interprovincial Cooperatives Ltd., Winnipeg, Man.; Dow Chemical Co., Midland, Mich.; American Hoechst Crop., North Hollywood, California.

Treatment	Source	Product and formulation	Chemical name or active ingredient
1	Stauffer	Captan 50% WP	captan
2	Diamond	Daconil 2787	-
	Alkali	75% WP	tetrachloroisophthalonitrile
3	Diamond	Daconil 2787	
	Alkali	& Captan (35-35)	
4	Naugatuck	Spergon 95%	chloranil
5	Naugatuck	Plantvax 75% (F461)	2,3-dihydro-5-carboxoanilido-6 methyl-1,4- oxathiin-4,4 dioxide
6	Naugatuck	Vitavax 75% (D735)	2,3-dihydro-5-carboxoanilido-6-methyl-1,4- oxathiin
7-9	Naugatuck	Numbered compounds	identity not available
10	Niagara	Phygon 50%	dichlone
11	Niagara	Polyram 80%	zinc activated polyethylenethiuram disulfide
12	Niagara	C. O. C. S. 55%	copper oxychloride sulfate
13	Niagara	Polyram 7D	zinc activated polyethylenethiuram disulfide (7% mixture)
14	Niagara	Polyram ZMCS 80	identity not available
15	Dupont	Arasan 75%	thiram
16	Dupont	Manzate D 80%	maneb
17	Dupont	Parzate C 75%	zineb
18	Dupont	Fermate 76%	ferbam
19	Dupont	Demosan 65%	1,4 dichloro-2, 5-dimethoxybenzene
20	Chemagro	4497 50%	bis (1, 2, 2, -trichloroethyl) sulfide
21	Chemagro	Dyrene 50%	2, 4-dichloro-6-(0-chloroanalino)-S-triazine
22	Chemagro	Dexon 50%	p-dimethylaminobenzenediazo sodium sulfo ⁻ nate
23	Chemagro	Bay 47531	dichlofluanid
24	Cyanamid	Cyprex 65%	dodine (n-dodecylguanidine acetate)
25	Green Cross	Duter 20%	triphenyl tin hydroxide
26-43	Green Cross	Numbered compounds	identity not available
44-59	Morton	"EP"-compounds	identity not available
60-66	Chipman	Numbered compounds	identity not available
67	co-op	Hexa	identity not available
68	Dow	Dowicil 100 95%	1-(3chloroallyl)-3, 5, 7-triaza-1-azoniaada - mantane chloride
69,70	Hoechst	Numbered compounds	identity not available
71	Niagara	Polyram seed Protectant	zinc activated polyethylenethiuram disulfide

Table 1.	Source an	d identity	of	seed	treatment	materials	

Greenhouse damping-off control tests – Seeds of jack pine, white spruce and red pine were pelleted with nine fungicides at rates of 0.5 and 2.0 g chemical per gram of seed and were germinated in soil from the Pineland Nursery at Hadashville, Manitoba. Dow Latex 512R was used as a binder as previously described. Seeding was done as soon after treatment as possible, but in some cases it was delayed as much as 10 days after treatment. The experimental plots contained 100 seeds per 8 inch x 9 inch plot and were arranged in a randomized block design with five replications for each treatment. Damping-off was recorded weekly from the beginning of emergence until 3 months after seeding. Supplementary lighting in the greenhouse was used from 8 AM to 8 PM and the temperature controls were set at 72° F (22.2 C).

Results and discussion

<u>Bioassay of seed treatment chemicals</u> – Data on the lowest concentration of seed-treatment chemica 1s that inhibited growth of <u>Pythium</u>, <u>Fusarium</u>, and <u>Rhizoctonia</u> are shown in Table 2. Thirty-one of the 69 chemicals tested showed a high level of activity (inhibitory at concentrations e qual to or less than 631 μ g/ml) against all three fungi. Nine others were effective against <u>Rhizoctonia</u> and, <u>Pythium</u> only, and one was active against <u>Pythium</u> alone. High activity against <u>Rhizoctonia</u> was shown

Treatment number	Product and formulation	<u>Lowest inhibi</u> <u>Rhizoctonia</u>	<u>torv concentrat</u> <u>Fusarium</u>	<u>ion(ug/ml)</u> Pythium
1	Captan 50 WP	158	200	158
2	Daconil 2787 WP	3,400	316	40
3	Daconil 2787 & Captan (35-35)	79	158	631
4	Spergon 95%	631	3,981	631
5	Plantvax 75%	10,000	10,000	5,012
6	Vitavax 75%	40	NIa	631
7	6638	316	NI	316
9	D-735-10D	316	2,512	2,512
10	Phygon 50%	631	1,259	158
11	Polyram 80%	317	40	158
12	C. O. C. S. 55%	NI	NI	5,012
13	Polyram 7D	NI	317	NI
14	Polyram ZMCS 80%	40	158	NI
15	Arasan 75%	1,259	1,259	40
16	Manzate D 80%	63	40	199
17	Parzate C 75% Fermate 76%	2,512 158	398 79	5,012
18		40	40	40
19	Demosan 65% 4497 50%	40 40	40 40	100
20	Dyrene 50%	40 40	100	158
21	-	158	2,512	1,000
22 23	Dexon 50% Bay 47531	40	40	100 40
23 24	Cyprex 65%	631	5,012	40 631
24 25	Duter 20%	158	158	40
25 26	RD 8684 15%	158	10,000	10,000
26 27	RD 8684 & Cyprex	631	5,012	
28	3944X	158	398	631 40
28 29	Drillbox Lindasan	79	10,000	40 79
29 30	MHC 223	40	40	2,512
31	TMHC 175 (2)	40	158	1,259
32	TMHC 2222	79	158	2,512
34	Dual purpose Bunt No More 50%	79	631	158
35	RD 8684 & Maneb 50%	79	40	631
36	RD 8684 & Captan 50%	79	631	158
37	KHC 324	79	40	40
38	MHC 324	40	158	1,259
39	PHC 324	40	79	79
40	XHC 324	79	79	1,259
41	BHC 324	79	158	1;259
42	DHC 324	40	40	79
43	THC 324	79	40	631
44	EP 277 50%	40	316	631
45	EP 277A liquid	40	631	2,512
46	EP 279 50%	158	2,512	10,000
47	EP 279A liquid	63	40	1,000
48	EP 293 50%	100	40	631
49	E P 294 50%	63	40	126
50	EP 301B 50%	398	251	40

Table 2. The lowest concentration of seed treatment chemical that inhibited the growth of three damping-off fungi on malt agar

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Table 2 (Continued)

Treatment	Product and	Lowest inhibitory concentration(µg/ml)				
number	formulation	Rhizoctonia	Fusarium	Pythium		
51	EP 301C	631	158	158		
52	EP 301C	631	316	631		
53	EP 301E	398	63	631		
54	E P 302B	398	63	1,000		
55	E P 302C	100	631	398		
56	E P 302D	251	100	10,000		
57	E P 305	79	316	10,000		
58	E P 306 75%	40	317	2,512		
59	E P 308	40	40	2,512		
60	65-S-1	317	5,012	5,012		
61	65-S-7	158	158	5,012 NI		
62	66-S-1	5,012	1,259	10,000		
63	66-S-2	158	40	5,012		
64	66-S-3	40	40	10,000		
65	66-S-4	40	40	40		
66	66-S-5	40	158	79		
67	Hexa	398	158	158		
68	Dowicil 100 95%	40	NI	40		
69	2844	40 79	79	40 79		
70	2874	316	631	79 40		
71	Polyram seed protectant	40	631	40 NI		

^aNI indicates no inhibition at highest concentration used.

by 61 chemicals and against <u>Fusarium</u> by 52 chemicals. Fifteen of the 40 chemicals that showed high activity against <u>Rhizoctonia</u> and <u>Pythium</u> were more effective than the standard captan treatment.

Laboratory germination tests – Results of the seed germination tests are shown in Table 3. All 13 chemicals caused some inhibition of germination. The least inhibitory were copper sulfate (COCS) and zineb (Parzate C). Captan inhibited germination of red pine and white spruce more than jack pine; Chemagro 4497 inhibited jack pine and red pine more than white spruce: and ferbam (Fermate) inhibited white spruce more than jack pine and red pine.

(Greenhouse damping-off control tests – The incidence of damping-off was greater in the greenhouse than is usually observed in the field. In the check plot, jack pine was most severely affected by postemergence damping-off (Table 4), but emergence of white spruce and red pine was reduced by apparent preemergence damping-off. Polyram, thiram, and captan showed the most promise for control of postemergence damping-off and were most effective on red pine. All seeds treated with Spergon emerged more rapidly than those that had been treated with other chemicals, and initial stands of red pine and jack pine were good: however, damping-off became quite severe 3 to 4 weeks after emergence. There was no evidence that preemergence damping-off was controlled by any of the chemicals.

Phytotoxic effects of the chemicals were severe at the higher rate of application. Duter caused the greatest reduction in emergence in all three conifer species. The percentage emergence for seed treated with Plantvax and Vitavax was inversely proportional to the amount of chemical applied. Thiram appeared to be the least injurious of the effective chemicals. Captan and polyram were phytotoxic to white spruce at both concentrations, but germination of jack pine and red pine seeds was reduced by polyram only at the higher concentration.

Seed treatment chemicals that reduced the amount of damping-off in the greenhouse tests were generally highly active against <u>Pythium</u> in the bioassay tests. It is possible that combinations of these chemicals would give a broader range of activity. However, control of damping-off with seedtreatment chemicals alone appears to be quite difficult to achieve at the present time, because many chemicals are effective against only part of the damping-off complex, and those that are effective against all damping-off fungi are often phytotoxic. An acceptable level of phytotoxicy might be established but only after an acceptable level of damp-

	Jack pine		White s	pruce	Red pine	
reatment No. and product	heavy ^a	lightb	heavy	light	heavy	light
1 Captan	63''	76	38''	55''	24 ^{II}	31*
2 Daconil	47"	86	1''	43''	35*	17"
4 Spergon	72	89	21''	80	29''	52*
11 Polyram	32''	67''	40 "	40 *	5''	30 *
12 C. O. C. S.	78	77	55''	70	70	94
15 Arasan	80	78	37''	50 *	7"	58''
16 Manzate D	37''	60 *	0"	15''	12"	19'
17 Parzate C	71	66 "	52''	74	83	88
18 Fermate	55''	75	1*	14"	5*	43''
20 Chemagro 4497	2''	0"	19''	46*	0"	0*
21 Dyrene	13''	19*	4 *	2''	7"	6''
23 Bay 47531	17"	50 *	1''	1*	0"	1*
25 Duter	6*	4"	0 ''	0 ''	0*	0 "

Table 3. Germination in seed germinator of conifer seeds pelleted with two amounts of seed-treatment chemicals

a heavy = 1 g chemical/gram of seed

b light = 0.25 g chemical/gram of seed

* Statistically significant from the check at the 5% level.

ing-off control is obtained. Our data indicate that 31 of the chemicals tested have a high degree of activity against isolates of the three main damping-off pathogens, <u>Pythium, Rhizoctonia</u>, and <u>Fusarium</u>. Of the three chemicals found effective against damp ing-off of red pine in greenhouse tests, thiram and captan were also found useful by Vaartaja and Wilner in controlling damping-off of Scots pine (4). The prospect of finding a satisfactory seed treatment for control of conifer damping-off **s**ee m**s** promising, and additional laboratory and field tests are planned.

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fers in Saskatchewan. Can. Dep. Agr., Div. Forest Biol., Progress Rep. 9(5):2.

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	Grams of	Emergence (Yo)			Damping-off (Y ₀)		
Treatment no. and product	chemical per gram of seed	Jack pine	Red pine	White spruce	Jack pine	Red pine	White spruce
and product	gram or seed	pine	pine	spruce	pine	pine	spruce
1 Captan	2.0	62.0	40.5	13.6*	68.2	34.3"	20.8"
	0.5	60.9	49.0	13.1*	71.5	44.5	37.9
2 Daconil	2.0	40. 2*	15.4"	4.7"	88.3	85.2	35.4
	0.5	54.2"	19.1"	3.9"	95.7	62.5	33.6
4 Spergon	2.0	77.5	63.1	17.6"	65.9	52.7	42.9
	0.5	76.7	64.2	17.1"	74.9	61.6	53.6
5 Plantvax	2.0	8.0*	6.8*	0.2"	53.4"	3.4"	20.0*
	0.5	41.5*	45.5	11.7"	90.0	66. 3	25.9"
	0.1	71.9	60.6	29.9	96.2	95.3	50.2
6 Vitavax	2.0	0.8*	3.0"	0.2"	20.0"	44.8	20.0"
	0.5	32.5"	41.7	13.9"	94.6	78.9	47.7
	0.1	71.1	72.5	22.6*	87.5	89.9	64.9
10 Phygon	2.0	23.2*	1.4"	13.6"	82.2	20. 0''	61.6
	0.5	32.6*	11.8*	17.2"	88.8	60.0	55.2
11 Polyram	2.0	45.5*	29.0*	17.5"	38.9"	12.1"	26.2"
	0.5	67.7	40.7	19.9"	62.3	20.2"	24.4*
15 Arasan	2.0	78.0	45.3	28.6	79.4	26.7"	33.5
	0.5	77.6	61.6	30. 1	76.7	37.0"	35.2
25 Duter	2.0	8.8*	0.3*	1.0*	45.1"	6.7 [*]	3.3"
	0.5	21.5*	2.5"	1.1"	74.9	24.8*	10.5"
Check		71.6	58.4	45.1	86.3	87.3	52.6

 Table 4. Effect of seed treatments on preemergence and postemergence damping-off of conifer seedlings in natural soil in the greenhouse

* Statistically significant from the check at the 5% level.