

### Prevention of Ascocarp Formation in *Claviceps purpurea* by Fungicides Applied over Sclerotia at the Soil Surface

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Cooperative investigations, Plant Science Research Division and the Oregon Agricultural Experiment Station. Published with approval of the Director as Technical Paper No. 3100, Oregon Agricultural Experiment Station.

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Accepted for publication 6 January 1972.

#### ABSTRACT

Twenty-seven systemic and nine protectant fungicides were evaluated for suppression of ascocarp formation in *Claviceps purpurea*. The chemicals were applied once over sclerotia from *Lolium perenne* at the soil surface. In one or two separate tests, complete or nearly complete suppression of ascocarps was obtained by application to 92 cm<sup>2</sup> of soil surface of benomyl at 1 to 4 mg, triarimol at 0.5 to 2 mg, cadmium chloride at 1 mg, 1,2-bis(3-methoxycarbonyl-2-thioureido)benzene at 2 and 4 mg, and parinol at 2 and 4 mg. These dosages failed

to suppress ascocarps in a third test with sclerotia with greater capacity for ascocarp production. Phenyl-5,6-dichloro-2-trifluoro-methyl-1-benzimidazole-carboxylate (Lovoal) consistently suppressed ascocarp development in all tests at 2 mg/92 cm<sup>2</sup> of soil surface, and thus showed the strongest activity of the organic chemicals evaluated. Cadmium chloride may provide an economic chemical control if acceptable for registration.

Phytopathology 62:609-611.

*Additional key words:* ergot control.

Practical control of ergot, *Claviceps purpurea* (Fr.) Tul., on a large scale was first obtained in perennial ryegrass, *Lolium perenne* L., fields grown for seed in western Oregon during 1949 by destruction of sclerotia by postharvest burning of straw and stubble during the previous summer (1). The incidence of ergot dropped sharply after the practice of burning was generally adopted, and the disease has been held at low levels in recent years. Among the 1970 perennial ryegrass seed samples entered for certification, 63% had no ergot and 30% had only a trace infestation, based on spore recovery tests on the cleaned seed (5).

Ergot has been controlled by annual burning in fields of other perennial grasses, including: tall fescue, *Festuca arundinacea*; colonial bentgrass, *Agrostis tenuis*; Kentucky bluegrass, *Poa pratensis*; red and chewing fescue, *F. rubra*; and orchard grass, *Dactylis glomerata* (2, 3, 4). Postharvest burning has controlled ergot in annual ryegrass, *L. multiflorum*, coincident with the adoption of the practice of reseeding with rangeland seeders in burned over but unplowed soil.

Although field burning is the most valuable cultural method now practiced in grass seed production in Oregon (3), open burning may be banned to reduce air pollution. Development of substitute control methods, therefore, is urgently needed. The objective of the present study was to evaluate systemic and protectant fungicides for suppression of ascocarp formation.

**MATERIALS AND METHODS.**—Sclerotia of *C. purpurea* from *L. perenne* were placed on the surface of a sandy loam, pH 5.8, soil 8.5 cm deep in 10-cm square plastic pots (about 100 sclerotia/pot) with four bottom drainage holes. The pots were held outdoors overwinter or at 5 C in a constant temperature chamber for 30 to 90 days to condition the sclerotia for ascocarp production.

The pots were placed in a greenhouse to force ascocarp formation. Just before chemical treatment, the soil was moistened and pressed firmly to provide a flat surface area of 92 cm<sup>2</sup> and to prevent the chemical solution or suspension from running down the innerwalls of the pot. The chemicals were applied once in a suspension or solution with sufficient water

to aid distribution of chemical in a uniform layer over the soil surface after the water was absorbed. Dosages are all expressed as active ingredient.

We maintained the soil surface continuously moist by holding the pots in plastic saucers constantly supplied with water. Results from three pots treated with each dosage rate were obtained by counting the perithecial heads and removing these mature ascocarps with attached sclerotia at weekly intervals starting 4 or 5 weeks after chemical application as compared with three untreated pots. Effectiveness of the compound was measured by the reduction in numbers of ascocarps as compared with the untreated pots.

**RESULTS.**—None of a wide variety of systemic chemicals applied at 2, 4, and 10 mg/92 cm<sup>2</sup> of soil surface provided adequate suppression of ascocarps, including: N-tridecyl-2,6-dimethylmorpholine (NIA9211 = BAS2203F); 3-(2-methylpiperidino)propyl 3,4-dichlorobenzoate (EL211); oxycarboxin; carboxin; 2,4-dimethyl-5-carboxanilido thiazole (G696); 5,6-dihydro-2,2',3'-trimethyl-1,4-oxathiazin-3-carboxanilide (F827); 1,1,1-trichloro-3-nitro-2-propanol (TCNP); 1-methyl-3-(6-methoxy-3-pyridyl)urea (LCS761); 2-methylbenzanilide (BAS3050F); 2-methyl-5,6-dihydro-4H-pyran-3-carboxylic acid anilide (HOE2989); 5-n-butyl-2-ethylamino-4-hydroxy-6-methylpyrimidine (PP149); 5-n-butyl-2-dimethylamino-4-hydroxy-6-methylpyrimidine (PP675); P-(2-ethylimidazol-2-yl)-P-imidazol-1-yl-N,N-dipropyl (PTA); 1-imidazolylphenylpiperidine (PS); 4-n-butyl-1,2,4-triazole (RH124); chloroneb; and 4-amino-6-chloro-2-(methylthio)pyrimidine (U8342). Other systemics were unsatisfactory at 1, 2, and 4 mg including: symmetrical dichlorotetrafluoroacetone (DCTFA); 2,4-dimethyl-5-N-(2-methylphenyl)carboxamidothiazole (H115); 2-(4-thiazolyl)benzimidazole (Thiabendazole); and parinol. The numbers of ascocarps were reduced slightly at 10 mg by NIA9211 and by piperazin-1,4-diyl-bis[1-(2,2,2-trichloroethyl)formamide] (W524). Complete or nearly complete suppression of ascocarps was obtained by 1,2-bis(3-ethoxycarbonyl-2-thioureido)benzene (thiophanate) at 10 mg; 1,2-bis(3-methoxycarbonyl-2-thioureido)benzene (thiophanate-methyl) at 4 and 10 mg; benomyl at 2, 4, and 6 mg; and triarimol at 2, 4, and 6 mg.

Nine protectant fungicides that have shown activity against *Gloeotinia temulenta* (6) were tested. The selected protectant fungicides that have shown unsatisfactory control of *C. purpurea* ascocarps at several dosage rates are: 2-(thiocyanomethylsulfanyl)benzothiazole (TCMTOB) at 1, 2, and 4 mg; 2-(thiocyanomethylthio)benzothiazole (TCMTB) at 2, 4, and 6 mg; triphenyltin acetate (TPTA) at 1 and 2 mg; triphenyltin hydroxide (TPTH) at 1 and 2 mg; a mixture of ammoniates of [ethylenebis(dithiocarbamate)] zinc with ethylenebis[dithiocarbamic acid] bimolecular and trimolecular cyclic anhydrosulfides and disulfides (Polyram) at 2, 4, and 6 mg; and monosodium salts of 2,2'-methylenebis(2,4,6-

trichlorophenol) (Isobac 20) at 1 and 2 mg.

In a separate test with both systemic and protectant fungicides, ascocarp formation was well suppressed for 7 weeks by benomyl at 1 mg; triarimol, at 0.2 to 1 mg; cadmium succinate, at 2 and 4 mg; cadmium chloride, at 1 mg; thiophanate-methyl, at 2 and 4 mg; parinol, at 2 and 4 mg; and phenyl-5,6-dichloro-2-trifluoromethyl-1-benzimidazolecarboxylate (Lovoal), at 1 or 2 mg/92 cm<sup>2</sup> of soil surface. Poor control was obtained with Thiabendazole at 1, 2, and 4 mg; thiophanate, at 1, 2, and 4 mg; TCMTOB, at 1, 2, and 4 mg; TCMTB, at 2, 4, and 6 mg; TPTA, at 1 and 2 mg; and TPTH, at 1 and 2 mg. Fair activity but incomplete control resulted from TPTH at 4 mg, Isobac 20 at 4 mg, and TPTA at 4 mg.

The eight most active chemicals were evaluated again in a third test in which well conditioned sclerotia produced larger numbers of ascocarps than in previous tests. Under these conditions, highly favorable for ascocarp production, seven of the eight chemicals gave unsatisfactory control at dosages that were effective previously (Table 1). Poor control resulted from benomyl at 1, 2, and 4 mg; triarimol, at 0.5, 1, and 2 mg; parinol, at 1, 2, and 4 mg; cadmium succinate, at 1, 2, and 4 mg; cadmium chloride, at 0.2, 0.5, and 1 mg; Lovoal, at 1 mg; thiophanate, at 1, 2, and 4 mg; and thiophanate-methyl, at 1, 2, and 4 mg/92 cm<sup>2</sup> of soil surface. Since complete control was obtained by only 1 mg of cadmium chloride in the previous test, 2 to 4 mg might have been adequate. Even in this extreme test, Lovoal completely suppressed ascocarps at 4 mg and gave a very high degree of control at 2 mg. Lovoal was the only chemical that gave comparable suppression of ascocarp development at the dosage (2 mg) effective in previous tests.

**DISCUSSION.**—Diverse methods have been suggested for ergot control, including burial of sclerotia by deep plowing; mowing grasses before formation of sclerotia; planting nonsusceptible crops; eradication or prevention of heading in fence rows or other adjacent waste areas; and planting ergot-free or 2-year-old seed. These methods are not feasible in established seed fields of perennial grasses or in continuous culture of annual ryegrass by the nonplowing method. Furthermore, cultivars resistant to *C. purpurea* are not available in susceptible grass species, and resistance is not being sought in most grass breeding programs.

By killing the fungus within sclerotia left in fields after harvest, the burning of straw and stubble has furnished good to excellent control of *C. purpurea* in grasses in Oregon. The burning of grass fields, unfortunately, contributes to air pollution, and open burning probably will be banned or severely restricted eventually by legislation in most areas of grass seed production.

Elimination of ascospore inoculum is an attractive approach to ergot control in perennial grasses, if safe fungicides can be found that will suppress ascocarp formation at a reasonable cost. Although a few chemicals reduced the number of

TABLE 1. Prevention of ascocarp formation in *Claviceps purpurea* by chemicals applied over sclerotia from *Lolium perenne* at the soil surface

Chemical <sup>a</sup>	mg/ 92 cm <sup>2</sup>	No. ascocarps/attached sclerotia removed Weeks after chemical application					
		4	5	6	7	8	9-12
Benomyl	1.0	1/1	8/7	17/13	11/8	1/1	7/3
	2.0	0/0	9/8	3/3	10/7	0/0	4/4
	4.0	0/0	10/8	3/3	14/4	0/0	6/3
Triarimol	0.5	9/7	5/4	8/5	1/1	3/3	2/1
	1.0	13/8	14/11	6/6	0/0	0/0	4/4
	2.0	3/3	3/3	12/9	0/0	0/0	1/1
Parinol	1.0	57/38	29/25	21/12	5/5	2/2	15/7
	2.0	42/33	16/16	9/9	8/6	3/3	17/8
	4.0	10/9	15/13	6/6	3/3	4/4	9/5
Cadmium succinate	1.0	19/16	8/6	5/5	4/2	0/0	4/3
	2.0	15/11	4/4	1/1	3/1	0/0	0/0
	4.0	2/2	8/7	4/4	2/2	0/0	1/1
Cadmium chloride	0.2	38/29	17/12	7/4	5/4	0/0	0/0
	0.5	25/20	22/18	6/4	5/4	0/0	2/1
	1.0	18/16	16/16	5/3	2/1	1/1	6/4
Lovoal	1.0	11/8	25/22	11/7	1/1	4/1	16/9
	2.0	2/2	3/3	0/0	0/0	0/0	0/0
	4.0	0/0	0/0	0/0	0/0	0/0	0/0
Thiophanate-methyl	1.0	26/15	26/22	18/13	7/6	5/3	5/5
	2.0	0/0	21/12	12/9	5/5	1/1	7/6
	4.0	0/0	2/2	3/3	4/3	2/2	1/1
Thiophanate	1.0	10/10	32/27	17/14	12/10	0/0	8/8
	2.0	1/1	9/9	17/13	4/4	0/0	4/2
	4.0	4/3	5/5	10/7	12/7	4/4	19/12
None		43/27	60/45	17/16	12/8	8/7	13/9

<sup>a</sup> Triarimol = *a*-(2,4-dichlorophenyl)-*a*-phenyl-5-pyrimidinemethanol. Parinol = *a,a*-bis(*p*-chlorophenyl)-3-pyridinemethanol. Lovoal = phenyl-5,6-dichloro-2-trifluoromethyl-1-benzimidazolecarboxylate. Thiophanate-methyl = 1,2-bis(3-methoxycarbonyl-2-thioureido)benzene. Thiophanate = 1,2-bis(3-ethoxycarbonyl-2-thioureido)benzene.

ascocarps after application of 10 mg/92 cm<sup>2</sup> (about 10 lb./acre), this dosage would be too costly for most organic compounds with complex structures.

Certain chemicals, e.g., LCS761, W524, and RH124, showed activity at 10 mg/92 cm<sup>2</sup> soil surface that temporarily suppressed ascocarp development up to 5 or 6 weeks after application. Such temporary control by excessive dosages is not promising for immediate field use, but the limited activity may indicate possible potential value of other derivatives of these molecular structures. Activity of Lovoal is encouraging, but use of this compound will probably be delayed by its high cost. Thiophanate-methyl may be usable if cost does not exclude its application at effective dosages. Cadmium chloride may represent a practical chemical because of expected lower cost, if it can be registered for this purpose.

The present results indicate that suppression of ascocarp formation by an available chemical at a reasonable dosage and at the low cost needed for ergot control in low value grass seed crops has not yet been demonstrated. Because of the pressing need for control methods to substitute for open burning of

grass fields, tests with additional fungicides are in progress.

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