Control of Dry Seed Decay of Wheat

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ABSTRACT

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Soils collected from four areas of Montana where wheat is grown under semiarid conditions were adjusted to moisture levels near or below those necessary for seed germination. Pondera spring wheat seed was treated with five commercial fungicides that had previously been screened for activity against a *Penicillium* sp. isolated from a seed showing dry seed decay. Treated seed was placed into soil and stored for 5 wk at 17 C. After incubation, some of the seed was retrieved and evaluated for visible colonization by *Penicillium*. Germination in soil after addition of moisture was also determined. Seed treatment with any of the materials reduced the level of *Penicillium* and increased germination. The addition of *Penicillium* spores to the soil resulted in the recovery of higher levels of *Penicillium*-colonized seed and reduced levels of germination. Treatment with imazalil provided the best control of dry seed decay.

In semiarid regions of the world, wheat (Triticum aestivum L.) seed is often planted when the soil is too dry to allow germination and emergence. Growers plant under such conditions with the hope that precipitation will provide the moisture necessary for germination. In some cases, however, the seed may remain in the soil several weeks to several months without germinating. Even though the soil may be too dry to allow the seed to imbibe enough water to germinate, the soil water potential may be sufficient to allow fungi to colonize the seed. This can lead to so-called dry seed decay, usually caused by one or more species of Penicillium (1,8). Wallace (7) determined that various factors affect the subsequent germination of cereal seed sown in soils of subgermination moisture content, including chemical seed treatment and injuries to the seed coat.

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With the advent of systemic fungicides highly inhibitory to a variety of fungi, including *Penicillium* spp. (2,6), our objectives were to determine: 1) the impact of these seed treatments on the development of dry seed decay, caused by *Penicillium* sp., when treated seed was sown in soil too dry for seed germination and 2) subsequent emergence of wheat seedlings when sufficient moisture for germination was applied. A preliminary report of this work has appeared (5).

MATERIALS AND METHODS

Soils were collected from four fields in Montana that had supported the production of small grain cereals for a number of years under frequent drought conditions. Growers in these areas often plant wheat seed into soil too dry for immediate seed germination. All soils were screened through a 2-mm mesh and air-dried for 48 hr before use.

The soil moisture content at which permanent wilting would occur (-1.5 MPa) was determined for each soil by a pressure membrane technique (4) and was as follows: Folkvord, 10.6%; Menli, 8.2%; McIntosh, 6.0%; and Big Sandy, 4.0%. Soil moisture content was adjusted

to be near or below the permanent wilting percentage by atomizing water onto soil tumbling in a mixing chamber. Actual moisture content of the adjusted soils was then determined gravimetrically immediately before use.

Bioassay tests. Seed of spring wheat cultivar Pondera was treated with the following at the indicated formulated product rates per kilogram: carboxin + thiram (Vitavax 200), 17 + 17% a.i. at 2.60 ml; imazalil (Flo Pro IMZ FL), 32% a.i. at 0.33 ml; captan (Captan 400D). 39.1% a.i. at 1.3 ml; maneb + lindane (DB Green), 50 + 18.75% a.i. at 2.08 g; and Vitavax 200 + Flo Pro IMZ FL at 2.60 + 0.33 ml. An untreated control was included for comparison. The treated seed was bioassayed for activity against an unidentified Penicillium sp. that had been isolated from a wheat seed showing dry seed decay. Plates of cornmeal agar were inoculated by dusting the surface with spores of this fungus. After a 7-hr incubation period, four seeds from each treatment group were placed equal distances apart on each of two plates and incubated for 72 hr at 22 C. The zone of inhibition of *Penicillium* was measured.

A second bioassay test was conducted using the same methods with varying rates of carboxin + thiram or imazalil. The following formulated product rates per kilogram were tested: carboxin + thiram at 5.21 and 2.60 ml and imazalil at 0.33, 0.16, 0.07, and 0.03 ml.

Soil incubation tests. Each soil was adjusted to three moisture levels at or below the permanent wilting point of each soil (with the exception of the 8.9% level with Menli). The levels were: Folkvord, 6.7, 8.5, and 8.9%; Menli, 7.0, 8.2, and 8.9%; McIntosh, 2.5, 5.2, and 5.7%; and Big Sandy, 1.9, 2.9, and 3.8%. Soil was added to glass petri dishes (100

 \times 15 mm) to a depth of 6 mm and firmed by pressing. Fifty seeds of each treatment were added to each dish and covered with 6 mm of soil, which was also pressed down. The dish covers were then replaced. Six replications of each treatment were prepared. The petri dishes were placed in pressed-fiber boxes along with moist paper towels to prevent the soil moisture from changing rapidly over the incubation period. The covered boxes were placed at 17 C for 5 wk, at which time, without the soil moisture levels being rechecked, three of the dishes from each treatment received 17 ml of tap water each to allow the seeds to germinate. This amount of water was sufficient to moisten the soil layer to the bottom of the dish. The uncovered dishes were placed in a growth chamber programmed at 21/15 C under a 12/12 hr light/dark cycle. Water was added as needed to maintain optimum conditions for germination, which were determined after 6 days.

The seed was removed from the other three dishes by dry-sieving the soil through a 2.4-mm mesh wire screen. The seed was placed on two layers of filter paper, 2 ml of distilled water was added, and the plates were incubated at 25 C for 4 days, after which the number of seeds with obvious *Penicillium* growth was recorded.

A second test was conducted with two moisture levels: Folkvord, 9.0 and 9.5%; Menli, 6.8 and 7.9%; McIntosh, 4.7 and 5.5%; and Big Sandy, 2.7 and 3.7%. One-

half of the soil was infested with conidia of the Penicillium sp. used in the bioassay tests. This inoculum was developed by incubating the fungus on autoclaved oat kernels (150 g of oats + 100 ml of distilled water in 1-L canning jars) for 2 wk. The conidia were collected by dry sieving, and 8.5 g of spores was added to 9 kg of dry soil and mixed in a tumbling vessel for 5 min. Each treatment was set up as in the first test and incubated at 17 C for 5 wk, at which time one-half of the plates were watered to encourage germination. The seed from the remaining plates was recovered as before and incubated on moist filter paper to encourage growth of Penicillium sp.

The data collected from each soil were analyzed (one-way ANOVA with LSDs for mean separations) as separate experiments. This was necessary because of differences in adjusted soil moisture levels across the four soil locations, which negated combining the data for a common statistical analysis.

RESULTS

Bioassay tests. Mean inhibition zone diameters around the seed were as follows: carboxin + thiram, 0 mm; imazalil, 16.8 mm; carboxin + thiram + imazalil, 18.1 mm; captan, 9.9 mm; maneb + lindane, 3.5 mm; and untreated, 0 mm; the experimental LSD (P=0.05) was 1.3. Results in the second bioassay test were: carboxin + thiram at 5.21 ml, 1 mm; carboxin + thiram at 2.60 ml, 0.7 mm; and imazalil at 0.33, 0.16, 0.07, and 0.03

ml, 27.8, 21.2, 15.1, and 14.3 mm, respectively. Untreated seed showed no zone of inhibition. The experimental LSD (P = 0.05) for the second bioassay test was 2.0.

Soil incubation tests. When treated seed was placed in soil under conditions too dry for germination, the subsequent growth of Penicillium sp. varied with the soil used (Tables 1 and 2). In general, the amount of dry seed decay that developed did not vary across the differing moisture levels, as indicated by a lack of significant treatment × moisture level interactions. For this reason, the data reported in the tables for both tests have been averaged across moisture levels. The maximum amount of visible seed colonization that could be detected in the first soil test (Table 1) was 6.6% with untreated seed in the Folkvord soil. Treatment with any of the materials significantly (P = 0.05) decreased the amount of colonization, i.e., dry seed decay. In the first test, growth of Penicillium sp. was never observed on seed treated with imazalil or carboxin + thiram + imazalil.

In the second test where a portion of the soil was infested with *Penicillium* sp. to augment that occurring naturally, the levels of seed decay were much higher than in the first test and significantly (P = 0.05) higher than where the soil was not infested. The highest level observed was for untreated seed in Folkvord soil with 31.3% colonization (Table 2). When the various seed treatments were com-

Table 1. Percent seeds with visible *Penicillium* colonization and percent germination of seeds of spring wheat cultivar Pondera treated with five fungicides and stored for 5 wk at 17 C in four natural soils

Seed treatment		Penic	rillium (%)ª		Germination (%) ^a					
	Folkvord	Menli	McIntosh	Big Sandy	Folkvord	Menli	McIntosh	Big Sandy		
	6.6	5.6	3.6	0.8	66.8	68.2	88.2	89.6		
Untreated	0.4	0.0	0.0	0.0	92.2	85.4	92.6	92.2		
Carboxin + thiram	0.4	0.0	0.0	0.0	91.2	87.6	92.2	91.2		
Imazalil Carboxin + thiram + imazalil	0.0	0.0	0.0	0.0	82.2	84.0	86.2	85.8		
	0.0	1.1	0.2	0.0	95.2	79.2	94.4	95.8		
Captan Maneb	1.3	1.1	0.0	0.0	90.8	87.4	95.6	96.2		
LSD $(P = 0.05)^b$	2.6	1.4	1.5	0.5	5.9	5.9	5.3	4.3		

^a Means are averaged over three moisture levels that, with the exception of one level of the Menli soil, were each less than -1.5 MPa.

Table 2. Percent seeds with visible *Penicillium* colonization and percent germination of seeds of spring wheat cultivar Pondera treated with five fungicides and stored for 5 wk at 17 C in four natural soils infested (I) or uninfested (U) with a *Penicillium* sp.^a

	Penicillium (%) ^b						Germination (%) ^b									
Seed treatment	Folkvord		Menli		McIntosh		Big Sandy		Folkvord		Menli		McIntosh		Big Sandy	
	I	U	I	U	I	U	I	U	I	U	I	U	I	U	I	U
	31.3	19.0	30.6	13.0	24.0	7.3	13.3	0.0	51.6	55.6	59.0	59.4	68.6	83.4	83.6	86.6
Untreated	7.7	0.0	7.0	1.0	5.4	0.4	0.0	0.0	79.6	89.0	77.6	86.4	83.6	90.6	86.4	90.6
Carboxin + thiram	0.4	0.0	0.0	1.4	0.0	0.4	0.0	0.0	88.4	85.4	89.4	81.4	83.4	87.6	91.6	88.6
Imazalil		0.0	0.0	0.0	0.0	0.0	0.0	0.0	84.0	87.4	79.6	82.0	84.0	84.6	89.4	85.6
Carboxin + thiram + imazalil	0.0	0.0	9.0	1.4	1.6	1.0	2.0	0.0	77.0	89.4	53.0	69.4	85.6	89.4	92.6	95.0
Captan	6.4			1.6	14.3	1.6	0.0	0.0	85.4	80.0	69.6	82.6	85.0	86.0	91.0	93.6
Maneb	14.3	3.0	21.4	1.0	14.3	1.0	0.0	0.0	05.7	00.0						
LSD $(P = 0.05)^{c}$	2.9		2	2.5		.1	0.7		4.9		7.2		5.3		4.9	

^a Penicillium spores (8.5 g/9 kg of dry soil) were added to one-half of each soil.

^bFor comparison of data within soil locations.

^bMeans are averaged over two moisture levels that were each less than -1.5 MPa.

^cFor comparison of data within soil locations.

pared across all the soils, moisture contents, and infested vs. noninfested, imazalil (0.1% colonized seeds), either alone or combined with carboxin + thiram (0.0% colonized seeds), was superior (P=0.05) to the other treatments in controlling *Penicillium* sp. infection (carboxin + thiram, 2.6%; captan, 2.8%; maneb, 7.0%; and untreated, 17.2% colonized seeds). Although the other treatments did reduce the amount of seed decay, performance was not consistent under all of the conditions.

The germination of wheat seed incubated in dry soil for 5 wk varied between soils and seed treatment (Tables 1 and 2). In the first test, untreated seed incubated in Big Sandy and McIntosh soils had germination rates of 89.6 and 88.2%, respectively, vs. 66.8% for Folkvord and 68.2% for Menli. However, Big Sandy and McIntosh soils also had the lowest amount of dry seed decay, 0.8 and 3.6%colonized seeds, respectively, vs. 5.6% for Menli and 6.6% for Folkvord. The greatest germination difference between treatments occurred in Folkvord soil, where 66.8% of untreated seed germinated compared with 95.2% of seed treated with captan. In the second test, the addition of Penicillium spores to the soil resulted in a mean increase in untreated seeds with dry seed decay of 15 percentage points and a corresponding mean reduction in germination of 5.4 percentage points. Results of the combined carboxin + thiram + imazalil

treatment in the germination tests were erratic. With few exceptions, the untreated seed had the poorest germination.

DISCUSSION

In in vitro tests, imazalil was very effective at inhibiting the growth of the *Penicillium* sp. that causes dry seed decay of wheat seed, even at rates one-tenth that of the recommended 0.33 ml formulation. The other materials tested were less effective in inhibiting the growth of this pathogen.

Carboxin + thiram failed to inhibit Penicillium in the plate assay test but resulted in excellent control of dry seed decay in the soil assay tests. The active ingredients in this material are reported by the manufacturer to be dispersible in water. The inability to control Penicillium in vitro, however, may be due in part to solubility or migration problems of the active ingredient through agar.

Under conditions where seed is planted in soil too dry to allow for immediate germination and emergence, the use of seed treatment can be very effective in preventing seed decay caused by *Penicillium* and perhaps related fungi. Thus, when moisture is sufficient to allow germination, the grower is assured of getting the best stand possible. Most of the treatments tested significantly (P = 0.05) increased the germination level of seed placed in soil. In the test with soil amended with *Penicillium* sp., the imazalil treatment was superior to the

other treatments tested. In addition to controlling common root rot (3), the use of imazalil can be beneficial when seed is planted in dry soil.

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