Effect of Fungicide Seed Treatments and Seed Quality on Seedling Diseases and Yield of Cotton

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ABSTRACT

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In greenhouse tests using soils artificially infested with *Rhizoctonia solani* and *Trichocladium* (= *Thielaviopsis*) *basicola,* treatment of cottonseed with carboxin 34 F + benomyl or technical carboxin + benomyl increased plant stands. In field tests using soil naturally infested with *R. solani, T. basicola, Fusarium* spp., *Pythium* spp., and *Meloidogyne incognita,* treatment of cottonseed with captafol, technical carboxin + captafol, thiram, chloroneb, and carboxin 34 F increased the number of surviving seedlings. Lint yields were higher but not significantly for all fungicide treatments than for the control. Application of fungicides to mechanically damaged seed increased the performance of Acala SJ-2 more than that of Stoneville 213. Captafol, thiram, carboxin 34 F, and technical carboxin + captafol increased seedling survival. Captafol appeared to reduce the adverse effects (phytotoxicity) associated with the application of technical carboxin in acetone.

Seedling diseases, which are ubiquitous in cotton (*Gossypium hirsutum* L.) production, are caused by one or more pathogens acting singly or in combination (3,6). Preemergence and postemergence diseases must be controlled to obtain uniform stands of vigorous plants. Seedlings with diseased roots may be inefficient in absorbing moisture and nutrients and more susceptible to pathogens occurring later in the season, and plants may mature later when compared with those with healthy roots. Soil temperature and moisture during the first few weeks after planting cottonseed are important in establishing stands of healthy plants because these conditions can affect the activity of both the host and pathogens (6,13).

Control of cotton disease pathogens depends to some extent on planting high-quality seeds that are properly treated with fungicides (1,7,8). Seed quality may be affected during production, harvesting, or ginning (2,4,5,12). During processing of cottonseed for planting, some damaged and light (immature) seeds are removed. Nevertheless, variations in planting seed intensify seed and seedling diseases and their control. All internally seedborne pathogens are not controlled by fungicides registered for coating of cottonseed. An effective fungicide for use on cottonseed must be applied by a method that controls both seedborne and soilborne pathogens.

The objectives of this study were to determine the effects of methods of applying fungicides to cottonseed on seedling survival and cotton yield and the effects of seed coat damage on the performance of fungicides.

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MATERIALS AND METHODS

Fungicides. The treatments and the concentration (grams of active ingredient [a.i.] per kilogram of seed) of the fungicides evaluated were: benomyl 50 WP [methyl 1-(butylcarbamoyl)-2-benzimidazolecarbamate, E. I. du Pont de Nemours & Co.], 1.2 g; chloroneb 65 WP (1,4-dichloro-2,5-dimethoxybenzene, E. I. du Pont de Nemours & Co.), 3.1 g; thiram 70 WP [bis(dimethylthiocarbamoyl)disulfide, E. I. du Pont de Nemours & Co.], 1.6 g; captafol 65 SP (*cis-N*-[(1,1,2,2-tetrachloroethyl)thio]-4-cyclohexene-1,2-dicarboximide, Chevron Chemical Co.), 1.9 g; carboxin 34 F (5,6-dihydro-2-methyl-1,4-oxathiin-3-carboxanilide, Uniroyal, Inc.), 2.1 g; carboxin 34 F + benomyl 50 WP, 2.1 g + 1.2 g; carboxin technical 2.0% in 70% acetone in 1977 and 100% acetone in 1978; 2.0% technical carboxin in 100% acetone + benomyl 50 WP, 1.2 g; technical carboxin + captafol 65 SP, 1.0 g. Untreated seeds were used as controls.

Captafol 65 SP and carboxin 34 F were mixed with water and then applied directly to the seed. Benomyl 50 WP and thiram 70 WP were mixed with graphite (2 mg of graphite:1 g of seed) and applied directly to moist seed. The treated seeds were shaken for 10 min and then air-dried. Only technical carboxin was applied by the organic solvent infusion technique (9,11). In combination treatments, technical carboxin was applied and the seeds were then dried before application of formulated fungicides. All treated seeds were stored 2 mo at room temperature before planting.

Seeds. Seeds of cotton cultivars Acala SJ-2 and Stoneville 213 were grown and acid-delinted in California and Mississippi, respectively. Seeds of Acala SJ-2 were used in 1977 and 1978, and those of Stoneville 213 were used in 1977 only.

In 1977, the treated seeds were separated into those with and without visible (naked eye) mechanical damage to the seed coat. Although both classes of seed were evaluated in 1977, major emphasis was placed on those without damage. In 1978, the damaged seeds (about 1%) were not separated out.

Greenhouse tests. The 1977 greenhouse tests at Beltsville, MD, were conducted with a sandy loam soil (pH 6.2). Soil in one bench was infested with mycelium of *Rhizoctonia solani* Kühn (isolates R-5, R-35, R-85, anastomosis group 4). Soil in another bench was infested with endoconidia of *Trichocladium basicola* (Berkeley & Broome) Carmichael (= *Thielaviopsis basicola* (Berk. & Br.) Ferr., isolates Tb-3 and Tb-20). The *R. solani* inoculum was grown for 4 wk in a sand-cornmeal mixture (97% sand and 3% cornmeal by weight, water to 20% moisture, v/w) and added to the soil at a rate of 2.0% by volume. The *T. basicola* inoculum was produced on V-8 juice agar. Infested soil in each bench was planted several successive times with Stoneville 213 seed to maintain high soil inoculum. These infested soils were mixed thoroughly (1:1 ratio) before 1 kg was placed in 20-cm-diameter plastic pots.

These experiments were conducted in growth chambers with a 14-hr photoperiod at approximately 3,600 lux in a greenhouse. Percentage of plant stand was determined 8 and 26 days after planting. Immediately after planting, 10 acid-delinted cottonseeds per pot, six pots per treatment, were placed in a growth chamber at 22 C for 26 days. Another set of pots was placed in a growth chamber at 28 C for 8 days and then moved to 22 C for an additional 18

Table 1. Effect of fungicides applied to acid-delinted Stoneville 213 cottonseed directly or with acetone on control of seedling disease in plant growth chambers caused by *Rhizoctonia solani* + *Trichocladium* (*Thielaviopsis*) *basicola* at Beltsville, MD, 1977[x]

Treatment	Constant 22 C		At 28 C (8 days) and 22 C (18 days)	
	Plant stand (%) days after planting		Plant stand (%) days after planting	
	8	26	8	26
None (control)	18 c[y]	2 e	34 e	4 d
Benomyl[z]	18 c	18 cde	52 cde	44 bc
Chloroneb	40 b	38 abc	46 de	46 abc
Thiram	54 ab	24 cd	68 abcd	26 cd
Captafol	58 ab	28 bcd	56 bcde	10 d
Carboxin	70 a	58 a	80 a	68 a
Carboxin + benomyl	66 a	48 ab	76 ab	66 ab
Carboxin (tech.) 2.0% in acetone	50 ab	50 a	58 abcd	46 abc
Carboxin (tech.) 2.0% in acetone + benomyl	40 b	50 a	72 abc	67 a
Carboxin (tech.) 2.0% in acetone + captafol	70 a	14 de	60 abcd	14 d

[x] Soils containing the two pathogens were mixed at the rate of 1:1; plant growth chambers in a greenhouse had 14-hr photoperiods at approximate 3,600 lux.

[y] In each column, values followed by the same letter do not differ significantly ($P = 0.05$) according to Duncan's multiple range test.

[z] Direct application to cottonseed at grams (a.i.) per kilogram of seed: benomyl = 1.2 g, chloroneb = 3.1 g, thiram = 1.6 g, captafol = 1.9 g, and carboxin = 2.1 g.

days to permit disease development caused by *T. basicola*. The isolate of *T. basicola* used was the same one previously used by Papavizas and Lewis (10) that did not attack cotton at temperatures above 24 C.

The greenhouse tests at Lubbock, TX, were conducted in plant benches filled with Amarillo loam soil (a member of the fine loamy, mixed thermic Aridic Paleustalfs) naturally infested with *R. solani* and *Fusarium* spp. The pathogenicity of the *Fusarium* sp. was not determined. Low-quality, untreated cottonseed was planted several times during the previous 4 yr in the soil under greenhouse conditions of 20–30 C and natural light to maintain high populations of these fungi. The experimental design was a randomized complete block replicated four times. Each plot consisted of 40 cottonseed planted in 100-cm rows, 10 cm apart. Percentages of seedling emergence and survival were determined 14 and 40 days after planting the seed, respectively.

Field tests. The 1977 and 1978 field tests were performed at Lubbock in Amarillo fine sandy loam soil naturally infested with *R. solani, T. basicola, Fusarium* spp., *Pythium* spp., and *Meloidogyne incognita* (Kofoid & White) Chitwood. Randomized complete blocks replicated six times were used. Each plot consisted of two rows, 1 m apart and 15 m long, with 250 seeds per row. Undamaged and a mixture of damaged and undamaged seeds were planted in 1977 and 1978, respectively.

In 1977, the performance of seeds with only damaged seed coats was also determined in field tests. Randomized complete blocks replicated three times were used. One row 6 m long comprised a plot, and each row was planted with 50 seeds.

Stand counts were made periodically, and only those (percentage of the seed planted) obtained 40 days after planting are given. A 2-cm section of diseased radicle (1 cm above and 1 cm below the junction of the hypocotyl-root tissues) from all tests at Lubbock was washed in running tap water for 20 min. These tissues were placed on 1.5% water agar and incubated at 27 C for 7 days. The organisms growing from the tissues were identified. Lint yield was determined at the end of the season. Data were analyzed statistically, and Duncan's multiple range test was used to identify significant differences among treatment means.

RESULTS AND DISCUSSION

Greenhouse tests. At constant 22 C for 8 days, all fungicide treatments, except benomyl, resulted in higher plant stands than that of the untreated control in plant growth chambers at Beltsville (Table 1). After 26 continuous days at 22 C, a temperature conducive to *T. basicola* infection (10), all fungicide treatments again, except benomyl and technical carboxin + captafol, resulted in significantly higher stand counts than that of the control. Carboxin (applied to seed directly or by acetone infusion), carboxin + benomyl, and chloroneb gave high final plant stands.

After exposure to 28 C for 8 days, a temperature conducive to *R. solani* but not to *T. basicola* infection, high percentages of surviving seedlings were obtained with thiram, carboxin (direct application or acetone infusion), carboxin + benomyl, and technical carboxin + captafol (Table 1). After 8 days at 28 C and 18 days at 22 C, chloroneb, benomyl, carboxin, and carboxin + benomyl had higher plant counts than the control (Table 1). These effects were caused by little or no decrease in the number of surviving seedlings for these fungicide treatments in comparison with a significant reduction in stands for the control after 26 days. Also, postemergence damping-off increased from the 8th to the 26th day for seed treated with thiram, captafol, and technical carboxin in acetone + captafol.

In plant benches of field soil naturally infested with *R. solani* and *Fusarium* spp. at Lubbock during 1977, for *R. solani* alone, carboxin was again the best fungicide, and its effectiveness was not improved by the addition of either benomyl or captafol (Table 2). Only carboxin and carboxin + benomyl gave seedling stands significantly higher than the stands for the control 40 days after planting the seed. In 1978, all fungicide treatments reduced damping-off, with technical carboxin with and without captafol and carboxin + benomyl being the most effective treatments. Higher percentages of surviving seedlings were obtained with all treatments in 1978 than in 1977 except with the control. The differences in the performance of technical carboxin in acetone + captafol during 1977 and 1978 appeared to be related to lower pathogen pressure in 1978. In general, the percentages of surviving seedlings at 14 and 40 days after planting followed similar trends among the treatments for each respective year. Isolates of *R. solani* and *Fusarium* spp. were cultured from diseased seedlings 40 days after planting. The *R. solani* isolate was obtained most frequently and appeared to be the primary pathogen involved in the damping-off of seedlings. Similar to the results in Beltsville, treatments containing carboxin were the most effective in reducing damping-off in 1978.

Field tests. The percentages of surviving seedlings among the fungicide treatments applied to undamaged seed of Acala SJ-2 planted at Lubbock in 1977 (Table 3) indicated that treatments with

chloroneb, thiram, captafol, carboxin with and without benomyl, and technical carboxin + captafol significantly reduced damping-off in the field compared with the control and other fungicide treatments. Although similar trends were followed with both Acala SJ-2 and Stoneville 213, a higher percentage of surviving seedlings occurred for carboxin treatments applied to seed of Acala SJ-2 than to Stoneville 213.

The percentages of surviving seedlings were lower for all treatments applied to seed with than without damaged seed coats (Table 3). The low percentages of surviving seedlings obtained with technical carboxin alone and in combination with benomyl appeared to be related to chemical injury of the embryo of the damaged seed. Also, some of the treatments may have predisposed the damaged seed to seed decay or to preemergence damping-off. The percentages of surviving seedlings were similar for technical carboxin + captafol and direct application of captafol, carboxin, and thiram. Apparently, captafol reduced the adverse effect associated with application of technical carboxin.

Both *R. solani* and *T. basicola* were cultured frequently from the roots of diseased seedlings from the field tests 40 days after planting and appeared to cause the major disease losses. *Fusarium* spp., *Pythium* spp., and *M. incognita* were detected only occasionally and were considered to cause minor losses.

Differences in fungicide effects on damping-off were also observed with the cultivars. In some instances, improvement in the relative performance for the fungicide treatments vs. control was obtained with seed of Acala SJ-2 but not with seed of Stoneville 213. The overall percentages of surviving seedlings for the treatments applied to undamaged and damaged seed of both cultivars showed that high-quality, fungicide-treated cottonseed should be planted to obtain the maximum number of surviving cotton seedlings.

Methods of fungicide applications in relation to seed integrity are important considerations in cotton culture. In several instances, final stands were similar when carboxin was applied directly or by the solvent-infusion method. Reduced stands obtained from damaged seeds treated with fungicides in acetone were probably caused by the direct contact of the chemicals with the embryo, because stands were not adversely affected when these fungicides were applied to undamaged seeds by both the solvent-infusion and direct application methods. Fungicides applied to cottonseed by acetone infusion gave higher stands than the control, but this technique must be further developed to improve its effectiveness. Perhaps other solvents may not be as phytotoxic as acetone and at the same time may improve the performance of the fungicide.

The trends in yield of lint among the treatments were similar for all tests; only the 1977 data for Acala SJ-2 are presented (Table 3). Lint yields for Acala SJ-2 grown from undamaged seed were about 75% higher in 1977 than in 1978. Lint yields in 1977 were about 15% higher for Stoneville 213 than for Acala SJ-2. These responses were apparently related to environmental variations between years and to the inherited production potential of the cultivars. The lack of significant differences in lint yield among the treatments applied to undamaged seed of Acala SJ-2 may be related to the ability of the cotton plants to compensate in terms of lint yield for the narrow range in plant populations. There were significant differences in lint yield among the treatments applied to undamaged seed of Stoneville 213. Only the yield for benomyl was different (lower) from that for the control. The overall average lint yield across varieties and years was significantly higher for all fungicide treatments except benomyl than for the control.

Lint yields for both cultivars were higher for each fungicide treatment applied to seed with undamaged than with damaged seed coats (Table 3). Lint yields differed among the fungicide treatments applied to seed with damaged seed coats and were directly related to plant populations. The major responses were increased yields when carboxin was applied directly to seed of Stoneville 213 and reduced yields when technical carboxin was applied to seed of both cultivars in comparison with those of the control.

Table 2. Effect of fungicides applied to acid-delinted Acala SJ-2 cottonseed directly or with acetone on seedling disease in the greenhouse caused by *Rhizoctonia solani* in plant benches of naturally infested soil at Lubbock, TX, 1977

	Seedling survival (%) 40 days after planting	
Treatment	**1977**	**1978**
None (control)	18 cd[y]	15 e
Benomyl[z]	20 cd	28 d
Chloroneb	32 abc	41 bc
Thiram	22 cd	29 d
Captafol	15 d	31 cd
Carboxin	45 a	42 bc
Carboxin + benomyl	41 ab	50 ab
Carboxin (tech.) 2.0% in acetone	28 bcd	53 ab
Carboxin (tech.) 2.0% in acetone + benomyl	25 cd	44 b
Carboxin (tech.) 2.0% in acetone + captafol	15 d	59 a

[y] In each column, values followed by the same letter do not differ significantly ($P = 0.05$) according to Duncan's multiple range test.

[z] Direct application to cottonseed at grams (a.i.) per kilogram of seed: benomyl = 1.2 g, chloroneb = 3.1 g, thiram = 1.6 g, captafol = 1.9 g, and carboxin = 2.1 g.

Table 3. Effects of fungicides applied to undamaged and damaged seed coats of seed of Acala SJ-2 on seedling survival and lint yields in field tests, Lubbock, TX, 1977

	Surviving seedlings (%) by seed coat type		Lint yield (kg/ha) by seed coat type	
Treatment	**Undamaged**	**Damaged**	**Undamaged**	**Damaged**
None (control)	72 c[y]	40 cd	1,327 a	1,202 ab
Benomyl[z]	33 d	28 e	1,390 a	1,254 ab
Chloroneb	82 ab	37 de	1,520 a	1,413 ab
Thiram	81 ab	52 ab	1,460 a	1,208 ab
Captafol	86 ab	48 abc	1,499 a	1,404 a
Carboxin	86 ab	50 abc	1,541 a	1,510 a
Carboxin + benomyl	90 a	44 bcd	1,555 a	1,190 ab
Carboxin (tech.) 2% in acetone	76 c	10 f	1,494 a	579 c
Carboxin (tech.) 2% in acetone + benomyl	80 bc	16 f	1,483 a	887 bc
Carboxin (tech.) 2% in acetone + captafol	86 ab	56 a	1,494 a	1,143 ab

[y] In each column, numbers followed by the same letter do not differ significantly ($P = 0.05$) according to Duncan's multiple range test.

[z] Direct application to cottonseed at grams (a.i.) per kilogram of seed: benomyl = 1.2 g, chloroneb = 3.1 g, thiram = 1.6 g, captafol = 1.9 g, and carboxin = 2.1 g.

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