

Control of Seed Transmission of *Cylindrocladium parasiticum* in Peanut with Seed Treatment Fungicides

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ABSTRACT

Seeds of peanut cultivars VA 98R and NC-V 11 with speckled testae were treated with fungicide and assayed on a selective medium to determine the viability of *Cylindrocladium parasiticum*. The fungus was isolated from 78 and 90% of the untreated speckled seed of the respective cultivars. Seed treatment with captan + pcnb + carboxin, fludioxonil, captan, and thiophanate methyl significantly reduced recovery of the pathogen in both cultivars. Speckled seed of VA 98R and NC-V 11 were treated with fungicides and planted in greenhouse and/or field trials in Suffolk, VA. Plants became infected with *C. parasiticum* after treated and untreated speckled seed were planted in steam-treated soil in the greenhouse. In one trial, seed treatment with fludioxonil, tebuconazole, and LS 176 significantly reduced taproot colonization by *C. parasiticum* compared to the untreated check. In a second greenhouse trial, only fludioxonil provided significant suppression of disease. In the field, treatment with fludioxonil, thiram, and tebuconazole significantly lowered *Cylindrocladium* black rot (CBR) incidence compared to the untreated check. Thiram significantly reduced taproot colonization compared to all treatments except fludioxonil. Based on the present study, the addition of thiram and/or fludioxonil to the standard treatment of captan + pcnb + carboxin may offer the best protection against seed transmission of *C. parasiticum*.

Key Words: *Cylindrocladium* black rot, peanut diseases.

Cylindrocladium black rot (CBR) of peanut (*Arachis hypogaea* L.) is a disease of economic importance to growers in Virginia and North Carolina, where the disease is widespread. CBR caused an estimated yield loss of 6.7 million dollars for Virginia in 2000 (8). The causal fungus, *Cylindrocladium parasiticum* Crous, Wingfield and Alfenas (teleomorph = *Calonectria ilicicola* Boedijn and Reitsma), overwinters in soil and plant debris as multicel-

lular, thick-walled microsclerotia (1, 2, 16). During the growing season, root exudates from peanut are thought to stimulate the germination of microsclerotia and subsequent taproot infection is favored when soil temperatures are 20 to 25 C and soil moisture is near field capacity (6, 10). Taproot colonization by *C. parasiticum* results in decay and eventually plant death (1, 3).

In addition to taproots, the fungus can infect hypocotyls, pegs, pods, and seed (1). A speckled testae is a symptom of seed infection by *C. parasiticum* (14, 15). Colonization and survival of the fungus in speckled seed during winter storage makes them likely agents for the dissemination of *C. parasiticum* (5, 14). No other fungal pathogen has been consistently isolated from speckled seed. Asymptomatic seed may also be infected at very low frequencies.

Greenhouse and field trials in Virginia and North Carolina have provided evidence for transmission of *C. parasiticum* after planting speckled seed of some seed lots (4, 14, 15). Plants grown from speckled seed planted in steam-treated soil in the greenhouse became infected by *C. parasiticum*. In the field, the occurrence of seed transmission was associated with the level of viable inoculum in seed lots at the time of planting. When speckled seed with a high recovery rate of the pathogen were planted in field plots, CBR incidence was correlated to the number of speckled seeds planted.

The most effective strategy for prevention of seed transmission of CBR probably will include the integration of several practices. A 3-yr rotation with non-leguminous crops and soil fumigation with metam sodium 2 wk prior to planting can reduce populations of *C. parasiticum* in seed production fields (7, 9, 11). Soil fumigation is currently practiced in ca. 75% of the peanut acreage in Virginia. The establishment of thresholds for CBR incidence in fields where seed peanuts are produced may reduce the number of infected seeds in commercial seed lots. However, plants with no apparent aboveground disease symptoms but with infected pods and seed below ground may go unnoticed during field inspections. Additional efforts to remove speckled seed from commercial seed during routine electronic sorting procedures can reduce seed transmission of the fungus. Application of seed treatment fungicides also may reduce seed transmission of *C. parasiticum*. Chemical treatment of all commercial seed may provide the additional protection necessary for speckled seed that escape other management

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practices. Application of seed treatment fungicides is a well-established practice in the Virginia seed industry.

Previous research on the role of seed treatment fungicides in the eradication of seedborne *C. parasiticum* has produced conflicting results. Porter *et al.* (13) were unable to isolate the pathogen from seed after treatment with dicloran + captan, carboxin + captan + dicloran, captan + maneb + pentachloronitrobenzene (pcnb) + etridiazole, thiram + pcnb, and carboxin + thiram + dicloran (12, 13). These studies were conducted before the symptom of speckled testa now associated with seed infection by *C. parasiticum* had been thoroughly described by Randall-Schadel (14, 15). More recently, Randall-Schadel demonstrated seed transmission in a field with no history of peanut cropping after planting speckled seed treated with either captan + carboxin + dicloran or captan + pcnb + carboxin (14, 15). Greenhouse and field trials in Virginia have confirmed that transmission of *C. parasiticum* can occur when speckled seed treated with captan + pcnb + carboxin, the commercial standard seed treatment fungicide of the industry, are planted (4).

The objective of this study was to identify seed treatment fungicides with increased activity against *C. parasiticum* compared to the commercial standard. Fungicides were evaluated for their ability to eradicate *C. parasiticum* from speckled seed in the laboratory and to prevent seed transmission of the pathogen in greenhouse and field trials.

Materials and Methods

Seed Collection. Seed used in all trials were produced in Virginia in 1998 and 1999. Speckled seed were handpicked from seed lots at commercial shelling plants after standard shelling, sizing, and sorting but before treatment with fungicides. Collected seed were packaged in polyethylene bags (4 mil). Bags were stapled and stored at 15 C in the dark.

Laboratory and Greenhouse Trials. Fungicides were evaluated in the laboratory and greenhouse for activity against *C. parasiticum*. Speckled seed of VA 98R and NC-V 11 were separated into 80-g batches and treated with the following fungicides: captan (Captan 400, Gustafson, Plano, TX), trifloxystrobin (Flint, Syngenta Crop Protection, Greensboro, NC), fludioxonil (Maxim 4FS, Syngenta), azoxystrobin (Abound 2.08F, Syngenta), tebuconazole (Raxil 2.6F, Gustafson), thiophanate methyl (Topsin M 70W, Elf Atochem, Philadelphia, PA), thiabendazole (Mertect 340F, Syngenta), difenoconazole + mefenoxam (Dividend XL, Syngenta), LS 176 (Gustafson), and captan + pentachloronitrobenzene (pcnb) + carboxin (Vitavax PC, Gustafson). Rates (g ai/kg seed) of fungicides tested are listed in Table 1. The rate of experimental compound LS 176 was expressed as the mass of formulated product per kg seed. Fungicides were applied while seed rotated in the drum of a Gustafson laboratory seed treater. Liquid and wettable formulations were diluted with water and applied to seed as a mist at a spray volume of 7 mL/kg seed using an airbrush. Ethephon dust at 1.25 g/kg seed was applied to all seed in order to break seed dormancy. Seed were rotated in the

seed treater for 5 min after fungicide or ethephon application to allow for uniform coverage of the material. The drum was cleaned with detergent and rinsed with warm water between fungicide treatments.

Treated speckled seed of VA 98R and NC-V 11 were stored for 3 wk in polyethylene bags (4 mil) at 15 C in the dark and assayed on a medium selective for *C. parasiticum*. The medium consisted of sterile potato dextrose agar base (Difco, Becton Dickenson and Company, Sparks, MD) amended with chlortetracycline (100 mg/L), chloramphenicol (100 mg/L), pcnb (52.5 mg/L), thiabendazole (2.3 mg/L), and dicloran (2 mg/L). Media was poured into 10 x 2.5 cm plastic petri plates and allowed to congeal. For each cultivar, 50 seeds per treatment were each placed in a single layer in small plastic dishes and covered with distilled water (not sterile) for 1 hr to promote some fungicide absorption. Seed with intact testae were rinsed with water and cut latitudinally (each cotyledon cut in half). Both cut ends were placed in contact with the medium. Five seeds were assayed per petri plate. Plates were incubated at 25 C and observed for growth of *C. parasiticum* for 14 d. The percentage of seed from which the pathogen was recovered was calculated.

Two greenhouse trials were conducted in 2000. Treated and untreated speckled seed of VA 98R in one trial and NC-V 11 in a second trial at a different location were planted in a complete block design with five replications. Seed were planted in a soil mixture containing 2:2:1 parts steamed soil, peat moss, and vermiculite (VA 98R trial) or 2:1 parts commercial potting soil and sand (NC-V 11 trial). The soil was a fine sandy loam from a field with a history of peanut/corn rotation. *Rhizobium* inoculant (Nitro-fix, Trace Chemical, LLC, Pekin, IL) was added to potting mixtures before seed were planted. Seed were planted at a depth of 3.8 cm (radicle down) in 15.2-cm clay pots (VA 98R) or plastic pots (NC-V 11). Five seeds were planted per pot. Each pot was a replication. Plants were watered to maintain adequate soil moisture. Foliar applications of acephate were applied as needed to control thrips. Air temperatures during the VA 98R and NC-V 11 trials ranged from 18 to 33 C and 9 to 40 C, respectively. Experiments were initiated in Feb. and terminated in May. Artificial lighting increased day lengths to ca. 15 hr.

The number of emerged seedlings was counted ca. 24 d after planting. Each week, plants were visually rated for aboveground disease symptoms using a zero to three scale (0 = healthy, 1 = stunted, 2 = wilted, 3 = dead). Plants were removed from soil approximately 11 wk after planting and root systems were rinsed free of debris. Each taproot was evaluated for severity of decay using a zero to five scale (0 = healthy, 5 = completely decayed). Five thin cross-sections (ca. 4 mm long) from each taproot were excised, disinfested in a 0.526% sodium hypochlorite solution for 2 min, and immediately placed on the previously described medium. Petri plates were incubated at 25 C and observed for growth of *C. parasiticum* for 14 d. Fresh weight of total plant tissue and root tissue were recorded. Non-emerged seedlings were rated a 3 for aboveground disease severity, 5 for taproot decay, and positive for the taproot assay. This was done to more accurately compare treatments with large differences in plant emergence.

Field Trial. Treated and untreated speckled seed of NC-V 11 were planted in four randomized complete blocks in Suffolk, VA in 2000. Plots consisted of two 7.6-m rows. Fungicide treatments were: captan, thiram (42-S Thiram, Gustafson), pcnb (RTU PCNB, Gustafson), trifloxystrobin, fludioxonil, azoxystrobin, tebuconazole, difenoconazole + mefenoxam, and captan + pcnb + carboxin. Rates (g ai/kg seed) of fungicides tested are listed in Table 2.

All field plots were treated twice with metam sodium at 36 kg ai/ha (Metam 42%, Taminco Inc., Smyrna, GA) to destroy soilborne inoculum of *C. parasiticum*. The fumigant was applied as a row treatment 20 cm below the soil surface using a coulter with a trailing shank. Rows were shaped to form beds (61 cm wide x 10 cm high) as the fumigant was applied. Several days after the first application, metam sodium was applied again to ensure even application of the chemical. Both applications of metam sodium were made at least 2 wk prior to planting. Seed were planted in the center of beds using KMC planters (Kelly Manufacturing Co., Tifton, GA). The seeding rate was ca. 10 to 13 seed/m and placement was ca. 5 cm deep. Thereafter, standard practices for peanut production in Virginia were followed.

Plant populations were recorded at 2 and 4 wk after planting. Plots were scouted for CBR incidence every 2 wk and the numbers of symptomatic and/or dead plants per plot were recorded. Areas under the disease progress curve (AUDPC) were calculated to compare treatments over an entire season. Immediately after inverting field plots on 25 Oct., taproots were randomly selected from each plot and assayed on the selective medium for *C. parasiticum*. One section from each root was excised, disinfested, and assayed as previously described. Petri dishes were incubated at 25 C and percentages of taproots colonized by *C. parasiticum* per plot were recorded. Pods were harvested in October with a two-row Amadas machine (Amadas Industries, Inc., Suffolk, VA) modified for plot research. Whole pods were dried with forced air and supplemental heat as recommended in the region until seed had 8 to 10% moisture content.

Laboratory Assay of Seed Planted in Field Trials.

The recovery rate for *C. parasiticum* in untreated speckled seed was determined for seed of NC-V 11 planted in the field trial. Fifty speckled seeds were assayed on the seed isolation medium in June 2000. Seed were rinsed 1 min under running water, cut latitudinally, and surface disinfested in a 0.26% sodium hypochlorite solution for 1 min. The cut ends were placed in contact with the medium and five seeds were assayed per petri plate. Petri plates were incubated at 25 C and observed for growth of *C. parasiticum* for 14 d.

Statistical Analysis. Analyses were performed using SAS (SAS Institute, Cary, NC). Treatment means were compared using Duncan's new multiple range test ($P = 0.05$).

Results

Laboratory Assay of Fungicide-treated Seed.

Cylindrocladium parasiticum was isolated from 78% of

untreated speckled seed of VA 98R (Fig. 1A). All fungicides significantly reduced pathogen recovery compared to untreated VA 98R seed. Treatment with captan + pcnb + carboxin, captan, fludioxonil, or the higher rate of thiophanate methyl reduced recovery to less than 15%. No fungicides were significantly better than the commercial standard, captan + pcnb + carboxin. In NC-V 11, *C. parasiticum* was recovered from 90% of untreated speckled seed and only captan + pcnb + carboxin, captan, trifloxystrobin, fludioxonil, and thiophanate methyl significantly reduced recovery compared to untreated seed (Fig. 1B). Fludioxonil at 0.1 g/kg seed lowered pathogen recovery to 34% and was more effective than the standard. Fungal colonies growing from seed treated with fludioxonil were small and did not exhibit the cottony mycelial growth typical of *C. parasiticum*. Seed treatment with LS 176 and tebuconazole did not result in significant reductions in pathogen recovery but did prevent mycelial growth on the seed testa.

Greenhouse Trials. Only 52% of the untreated speckled seed of VA 98R produced seedlings by 19 d after planting (Table 1). Seedling emergence was greatest (84

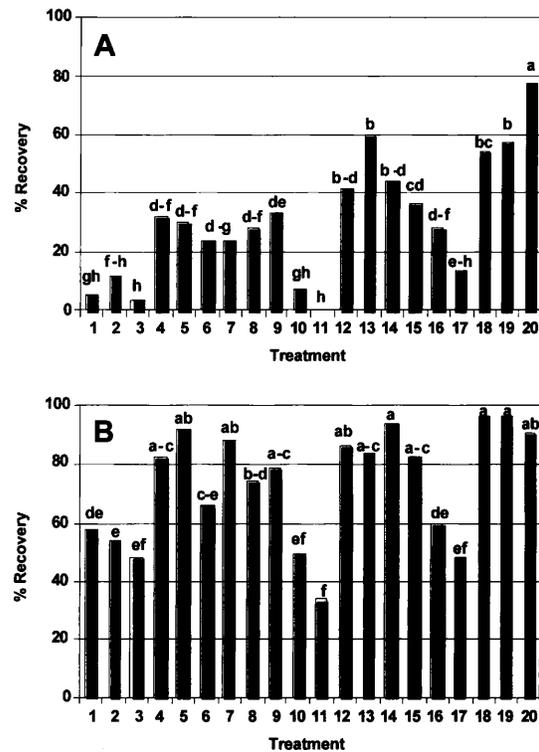


Fig. 1. Recovery of *Cylindrocladium parasiticum* from fungicide-treated speckled peanut seed of A) VA 98R and B) NC-V 11. Treatments followed by the same letter(s) are not significantly different according to Duncan's new multiple range test ($P = 0.05$). Treatments and rates (g ai/kg seed): 1. captan 1.13 + pcnb 0.38 + carboxin 0.25, 2. captan 0.94, 3. captan 1.88, 4. LS 176 0.22, 5. LS 176 0.44, 6. trifloxystrobin 0.2, 7. trifloxystrobin 0.4, 8. difenoconazole 0.12 + mefenoxam 0.01, 9. difenoconazole 0.24 + mefenoxam 0.02, 10. fludioxonil 0.05, 11. fludioxonil 0.1, 12. azoxystrobin 0.16, 13. azoxystrobin 0.33, 14. tebuconazole 0.05, 15. tebuconazole 0.1, 16. thiophanate methyl 0.22, 17. thiophanate methyl 0.44, 18. thiabendazole 0.05, 19. thiabendazole 0.09, 20. untreated check.

to 100%) from seed treated with captan + pcnb + carboxin, captan (0.94 g/kg seed), LS 176 (0.22 g/kg seed), fludioxonil (0.05 g/kg seed), azoxystrobin (0.33 g/kg seed), and tebuconazole (0.05 and 0.1 g/kg seed). Treatments with thiophanate methyl and thiabendazole had fewer emerged plants than the untreated check, but differences were not significant.

LS 176, fludioxonil, and the high rate of tebuconazole significantly lowered both severity of aboveground symptoms and taproot decay compared to the untreated check (Table 1). Root assays confirmed that these fungicides provided the best protection against taproot colonization by *C. parasiticum*. The pathogen was recovered from 80% of the plants grown from untreated seed. Significantly lower recovery rates ($\leq 36\%$) were from LS 176, fludioxonil, and tebuconazole treatments. All 25 plants from the tebuconazole (0.1 g/kg seed) treatment remained healthy throughout the study and the pathogen was not isolated from any of these plants. Fresh weight of root and total plant tissue was significantly increased when seed was treated with captan + pcnb + carboxin, the low rate of captan, LS 176, or the high rate of tebuconazole compared to the untreated check.

In NC-V 11, 84% of the untreated speckled seed germinated. Germination of seed treated with fungicides was

similar (data not shown). Aboveground symptoms of CBR were minimal in plants grown from untreated seed. No treatment was significantly different from the untreated check (data not shown). Seed treatment with fludioxonil (0.05 g/kg seed) significantly lowered the severity of taproot decay compared to the untreated check. *Cylindrocladium parasiticum* was recovered from more than 80% of taproots in all treatments except fludioxonil. Taproot colonization was reduced to 52 and 48% in plants grown from seed treated with the low and high rates of fludioxonil. None of the fungicides significantly increased fresh weight of total plant tissue compared to the untreated check.

Field Trial. *Cylindrocladium parasiticum* was recovered from 75% of the subsamples of NC-V 11 speckled seed. Treatment with captan + pcnb + carboxin, thiram, pcnb, and fludioxonil resulted in a significant increase of seedling emergence on 13 June compared to the untreated check (Table 2). Seed treatment with tebuconazole (0.05, 0.1 g/kg seed) delayed emergence and stunted plant growth. By 4 wk after planting, plant stands for seed treated with the low rate of tebuconazole increased to levels comparable with other treatments but populations remained among the lowest for seed treated with the high rate of tebuconazole. Tebuconazole also resulted in a sig-

Table 1. Efficacy of seed treatment fungicides in preventing seed transmission of *Cylindrocladium parasiticum* in speckled peanut seed of VA 98R in the greenhouse.^a

Treatment and rate g ai/kg seed	Emergence % ^b	CBR 0-3 ^c	Root decay 0-5 ^d	Root assay % positive ^e	Fresh weight	
					Roots	Total
Captan 1.13, pcnb 0.38, carboxin 0.25	88 ab	0.9 d-f	2.4 d-g	52 b-f	8.9 ab	64.6 a
Captan 0.94	96 ab	0.5 ef	2.5 d-g	44 c-f	8.8 ab	71.4 a
Captan 1.88	76 a-d	1.0 c-f	3.0 c-f	52 b-f	8.2 a-c	66.6 a
LS 176 0.22	88 ab	0.5 ef	1.4 gh	28 fg	8.7 ab	69.6 a
LS 176 0.44	80 a-c	0.6 ef	1.7 f-h	32 e-g	9.8 ab	70.0 a
Trifloxystrobin 0.2	76 a-d	1.0 c-f	2.9 c-f	40 c-g	6.6 a-d	51.1 ab
Trifloxystrobin 0.4	76 a-d	0.8 d-f	2.8 d-g	40 c-g	7.2 a-c	61.5 ab
Difenoconazole 0.12, mefenoxam 0.01	72 a-e	1.2 b-e	3.4 a-e	72 a-e	5.5 b-d	49.7 ab
Difenoconazole 0.24, mefenoxam 0.02	68 b-e	1.1 c-f	3.2 b-e	68 a-f	5.6 b-d	47.8 ab
Fludioxonil 0.05	84 ab	0.9 d-f	2.3 e-h	32 e-g	7.7 a-c	60.2 ab
Fludioxonil 0.1	68 b-e	1.0 c-f	2.2 e-h	28 fg	8.1 a-c	56.9 ab
Azoxystrobin 0.16	80 a-c	1.2 b-e	2.7 d-g	68 a-f	5.8 b-d	49.5 ab
Azoxystrobin 0.33	92 ab	1.0 c-f	3.1 b-f	64 a-f	6.6 a-d	53.1 ab
Tebuconazole 0.05	84 ab	0.9 d-f	2.4 d-g	36 d-g	8.1 a-c	58.9 ab
Tebuconazole 0.1	100 a	0.1 f	0.9 h	0 g	10.6 a	75.9 a
Thiophanate methyl 0.22	36 f	2.0 a-c	4.2 a-c	96 a	4.1 c-e	32.0 bc
Thiophanate methyl 0.44	48 d-f	1.7 a-d	3.5 a-e	76 a-d	6.3 a-d	48.2 ab
Thiabendazole 0.05	44 ef	2.4 a	4.4 ab	96 a	2.3 de	15.8 c
Thiabendazole 0.09	48 d-f	2.6 a	4.8 a	92 ab	1.1 e	9.3 c
Untreated check	52 c-f	2.1 ab	3.8 a-d	80 a-c	4.2 c-e	34.0 bc

^aMeans followed by the same letter(s) are not significantly different according to Duncan's new multiple range test ($P = 0.05$).

^bPlants were counted at 19 d after planting (DAP).

^cScale: 0 = healthy, 1 = stunted, 2 = wilted, 3 = dead. Non-emerged plants were rated 3. Ratings were taken at 74 DAP.

^dScale: 0 = healthy, 5 = completely decayed. Non-emerged plants were rated 5.

^eRoots were assayed on a selective medium. Percentages are based on the number of roots positive for *C. parasiticum* of a potential 25 plants. Non-emerged plants were considered positive.

Table 2. Plant populations and growth of peanuts in field plots planted in 2000 with fungicide-treated speckled seed of NC-V 11.^a

Treatment and rate	Plants/m of row ^b		Canopy width
	30 May	13 June	7 July ^c
g ai/kg seed	no.		cm ^c
Untreated check	4.66 b-d	7.81 e-g	37.1 ab
Captan 1.13, pcnb 0.38, carboxin 0.25.	5.74 a	10.63 a	37.1 ab
Captan 0.94	4.63 b-d	8.66 c-e	35.3 b-d
Captan 1.88	4.33 b-d	8.43 d-f	38.1 a
Thiram 0.94	4.72 b-d	9.22 b-d	36.3 a-c
Thiram 1.88	5.18 ab	9.28 b-d	38.1 a
pcnb 0.52	4.82 b-d	9.12 b-d	38.9 a
Trifloxystrobin 0.20	4.27 cd	8.17 d-g	35.1 b-d
Trifloxystrobin 0.40	4.66 b-d	8.56 c-f	36.3 a-c
Difenoconazole 0.12, mfenoxam 0.01	4.13 d	7.41 fg	35.1 b-d
Difenoconazole 0.24, mfenoxam 0.02	4.36 b-d	7.68 e-g	34.0 cd
Fludioxonil 0.05	4.86 b-d	9.65 a-c	38.9 a
Fludioxonil 0.10	5.05 a-c	10.24 ab	36.8 ab
Azoxystrobin 0.16	4.76 b-d	8.40 d-f	35.3 b-d
Azoxystrobin 0.33	5.12 ab	8.40 d-f	33.8 cd
Tebuconazole 0.05	1.38 e	7.38 fg	32.8 d
Tebuconazole 0.10	0.46 f	7.12 g	28.7 e

^aMeans followed by the same letter(s) are not significantly different according to Duncan's new multiple range test ($P = 0.05$).

^bDetermined from counts of two 7.6-m rows per plot. Planting date was 16 May.

^cData are the mean of six plants per plot.

Table 3. Incidence of CBR, area under the disease progress curve (AUDPC), percent of taproots colonized by *Cylindrocladium parasiticum*, and pod yield of peanuts in field plots planted in 2000 with fungicide-treated speckled seed of NC-V 11.^a

Treatment and rate	CBR ^b			AUDPC	Taproot	Pod yield
	31 July	31 Aug.	24 Sept.		colonization	
g ai/kg seed	no.				% ^c	kg/ha
Untreated check	4.5 a	23.8 a	53.3 ab	1347 a	76 ab	1696 e
Captan 1.13, pcnb 0.38, carboxin 0.25	2.3 ab	17.3 a-d	49.3 a-c	1130 ab	55 c-e	2734 a-d
Captan 0.94	2.0 ab	16.0 a-d	42.7 a-d	991 a-d	—	2324 c-e
Captan 1.88	4.3 a	12.3 b-e	50.3 a-c	1058 a-c	56 c-e	2636 a-d
Thiram 0.94	1.3 ab	9.3 c-e	32.5 cd	625 de	35 g	3105 ab
Thiram 1.88	1.0 ab	11.8 b-e	33.5 b-d	665 c-e	38 fg	3267 a
pcnb 0.52	1.3 ab	11.5 b-e	56.3 a	1027 a-d	62 a-d	2348 b-e
Trifloxystrobin 0.20	1.0 ab	14.3 b-d	37.0 a-d	897 b-d	—	2494 b-d
Trifloxystrobin 0.40	2.0 ab	18.0 a-c	43.0 a-d	1097 ab	61 b-e	2288 c-e
Difenoconazole 0.12, mfenoxam 0.01	0.0 b	14.3 b-d	36.3 b-d	853 b-d	—	2296 c-e
Difenoconazole 0.24, mfenoxam 0.02	1.0 ab	19.0 ab	53.3 ab	1098 ab	69 a-d	2143 de
Fludioxonil 0.05	0.8 ab	8.8 de	37.0 a-d	734 b-e	53 d-f	2983 a-c
Fludioxonil 0.10	1.3 ab	5.0 e	23.8 d	446 e	45 e-g	3348 a
Azoxystrobin 0.16	2.3 ab	17.3 a-d	51.5 a-c	1126 ab	—	2395 b-e
Azoxystrobin 0.33	3.8 ab	17.0 a-d	42.5 a-d	1016 a-d	71 a-c	2161 de
Tebuconazole 0.05	2.8 ab	12.0 b-e	32.3 cd	778 b-e	78 a	2427 b-e
Tebuconazole 0.10	1.3 ab	11.5 b-e	33.5 b-d	687 c-e	59 c-e	2291 c-e

^aMeans followed by the same letter(s) are not significantly different according to Duncan's new multiple range test ($P = 0.05$).

^bData represent number of symptomatic and/or dead plants per two-row plot.

^cData are percent recovery of *C. parasiticum* from 25 taproots selected at random from plants in each plot. "—" denotes treatments not assayed.

nificant reduction in plant growth compared to the untreated check based on canopy width measurements on 7 July (Table 2).

Statistical analyses of disease incidence and AUDPC revealed significant differences among treatments (Table 3). By 24 Sept., plots planted with untreated speckled seed contained 53 symptomatic plants. CBR incidence was significantly lower when seed were treated with thiram (0.94 g/kg seed), fludioxonil (0.10 g/kg seed), or tebuconazole (0.05 g/kg seed). AUDPC levels were lowered when seed were treated with two rates of these fungicides as well as the low rate of trifloxystrobin and difenoconazole + mfenoxam compared to the untreated check. Percentages of taproots colonized by *C. parasiticum* were lowest ($\leq 45\%$) for seed treated with thiram and the high rate of fludioxonil (Table 3). The commercial standard, captan (1.88 g/kg seed), thiram, and fludioxonil all significantly increased pod yield compared to untreated seed (Table 3).

Discussion

In order to provide increased protection against seed transmission of *C. parasiticum*, one or more fungicides should be added to the standard treatment of captan + pcnb + carboxin. Several fungicides have been identified with increased activity against the pathogen. Further research is aimed at evaluating fungicide mixtures.

Captan + pcnb + carboxin (Vitavax PC) is the standard

seed treatment fungicide of the peanut industry in Virginia. The fungicide has both protectant and systemic activity and is registered for control of seed rot and damping off diseases caused by *Rhizoctonia solani* Kühn, *Rhizopus* spp., and *Aspergillus* spp. in peanut (17). In this study, seed treated with the industry standard significantly reduced the recovery of *C. parasiticum* from speckled seed in laboratory assays. This treatment also resulted in some of the highest plant populations in both greenhouse and field trials. However, in agreement with previous studies, the standard seed treatment did not prevent transmission of *C. parasiticum* from speckled peanut seed in the greenhouse or field. Other fungicides were active against *C. parasiticum* based on laboratory assays of treated seed. Captan, fludioxonil, and thiophanate methyl significantly reduced pathogen recovery in speckled seed of VA 98R and NC-V 11. Although treatment with LS 176 and tebuconazole did not reduce pathogen recovery, these fungicides showed activity against the pathogen and prevented mycelial growth on the testae of seed.

In the greenhouse and field, several fungicides were superior to the untreated check. Seed treated with fludioxonil, tebuconazole, thiram, and LS 176 gave the greatest level of protection against seed transmission. Although tebuconazole showed activity against the fungus, treatment with this fungicide delayed emergence and stunted plant growth. LS 176 is an experimental compound. Thiram and fludioxonil are registered as seed treatments for use on peanut. Thiram was not tested in the laboratory or greenhouse, but seed treated with this fungicide resulted in significant disease suppression in the field compared to all other treatments except fludioxonil. Thiram significantly reduced CBR incidence and taproot colonization compared to the standard of captan + pcnb + carboxin. Fludioxonil consistently suppressed seed transmission of *C. parasiticum* in laboratory, greenhouse, and field trials. This fungicide also resulted in some of the highest plant populations and pod yields. A fungicide mixture containing thiram and/or fludioxonil with captan + pcnb + carboxin may provide better protection against seed transmission of *C. parasiticum* than the commercial standard alone.

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