REPORT OF THE COTTONSEED TREATMENT COMMITTEE - 2000

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Introduction

The 2000 National Cottonseed Treatment Program evaluated cotton seedling survival for a number of fungicide seed treatment combinations over diverse environmental conditions and levels and types of cotton seedling pathogens. Fifteen fungicide seed treatments were nominated by chemical industry representatives for evaluation in the 2000 National Cottonseed Treatment Program. A standard fungicide treatment, Vitavax-PCNB + Allegiance , and a nontreated control were included to assess efficacy of the nominations and seedling disease pressure. In addition, the fungicide treatments Allegiance or PCNB were included to aid in determining the importance of *Pythium* spp. or *Rhizoctonia solani*, respectively. Disease ratings and pathogen isolations for seedlings and soil populations of selected soilborne fungi were conducted by collecting seedlings and soil from the nontreated control plots at each location. Soil temperature and plant development data also were collected for sites for the 2000 National Cottonseed Treatment Program.

Materials and Methods

Fungicide Treatment

Acid-delinted seed of *Gossypium hirsutum* L., 'Deltapine 50' or 'Paymaster HS26', were provided by Delta and Pine Land Company, Scott, MS. Deltapine 50 was planted at all locations, with the exception of locations in Oklahoma and the College Station and Lubbock sites in Texas, where the cultivar Paymaster HS26 was planted. Fungicide treatments and dye (DayGlo EPX seed colorant, Gustafson Inc.) were mixed with water at a rate of 2% water to seed weight (v/w). Water and dye also were applied to the nontreated seed treatment at the same rate. Treatments were applied to the cottonseed while the seed tumbled in a rotating drum. When two or more fungicides were applied, the fungicides were mixed and applied in a single application. The technical information for the fungicides is given in Table 1. Seed germination was evaluated for all treated and nontreated seed by rolling seed in moistened germination paper and incubating at 28ºC.

Field Experiments

Seventeen field experiments were conducted by 16 cooperators across the U.S. Cotton Belt (Table 2). Each location utilized a randomized complete block experimental design, with the number of replications ranging from 4 to 10 (Table 2). The stand counts used in the analyses were taken from 18 to 32 days after planting, average 29 days, depending on the location. A soil sample and seedling samples from plots containing nontreated seed were taken from 28 to 38 days after planting, average 30 days, depending on the location. Soil and seedlings were placed in insulated packages with refrigerated cool packs and mailed overnight to the University of Arkansas for processing. A subsample of soil was sent to T. L. Kirkpatrick, Southeast Research and Extension Center, Hope, Arkansas, for determination of populations of plant parasitic nematodes. Soil temperature was monitored by burying a temperature sensor, tidbit (Onset Computer Corp, Pocasset MA), 10 cm. deep at planting.

Seedlings were evaluated for growth by recording the number of nodes from five arbitrarily selected seedlings and then the aboveground portions of all seedlings were removed and discarded. Approximately 50 seedlings per location were then rinsed for 20 minutes in running tap water and rated for disease symptoms. The hypocotyl disease severity index was 1=no symptoms, 2=few pinpoint lesions or diffuse discolored areas, 3=distinct necrotic lesion, 4=girdling lesion, and 5=seedling dead. The root disease index was 1=no symptoms, 2=1-10% of the root system discolored, 3=11-25% of the root system discolored, 4=26-50% of the root system discolored, and 5>50% of the root system discolored. Seedlings were surface disinfested by immersion for 1.5 min in 0.5% NaClO, blotted dry on a paper towel, and plated on water agar (2%) amended with 10 mg and 250 mg of the antibiotics rifampicin and ampicillin, respectively, and 0.5 ml of the miticide Danitol (Valent Chemical Co.) per liter. Resulting colonies were transferred to PDA and identified to genus. Seedlings were subsequently transferred to the Thielaviopsis selective medium TB-CEN (Specht and Griffin, 1985), which was modified by adding Penicillin G (60 mg/L), to determine isolation frequency for *Thielaviopsis basicola*.

Soil samples were assayed for populations of *Rhizoctonia* species by using the multiple-pellet soil method (Henis et. al., 1978), and Rhizoctonia populations were quantified on a modified Ko and Hora medium (Ko and Hora, 1971). Soil populations of *Pythium* species and *T. basicola* were detected by diluting 25 g of soil in 0.1% water agar to a total volume of 250 ml and placing on a wrist action shaker for 20 minutes. *Pythium* spp. were quantified by the spread plate method on the selective medium P_5ARP (Jeffers and Martin, 1986), and *T. basicola* populations were quantified using the pour plate method with the selective medium TB-CEN.

Statistics

Data were analyzed by the GLM procedure using SAS (SAS Institute Inc., Cary NC). Percent stand was analyzed over locations and by location. Treatment means were separated by using a protected LSD at P=0.05. The Pearson-product correlation method was used to examine the relationship among soil temperature, percent stand, plant growth, disease, pathogen isolation frequency, and soil populations over locations.

Results and Discussion

After the seed were treated with the fungicide treatments, seed germination ranged from 90% to 96% for Deltapine 50, with an average germination of 94%. Seed germination ranged from 94% to 99% for Paymaster HS26, with an average germination of 97%, after the seed were treated with the fungicide treatments. Several of the treatment combinations increased germination slightly but significantly compared to the nontreated control.

For the 17 locations reporting data for 2000 there was a significant location, treatment, and location x treatment effect (Table 3), indicating that the treatment response was dependent on the environmental or pathogen pressures for a particular location. A significant difference between treatments was found for 12 of the 17 locations (Table 4). In all of these experiments, at least one of the fungicide treatments performed better than the nontreated control. The Allegiance treatment increased stands compared to the nontreated control in 4 of the 12 experiments having a significant response compared to the nontreated control (AR2, LA1, TX1, and VA), indicating the importance of *Pythium* spp. in stand establishment at these sites. In 3 of these 12 experiments (AR2, MS1, and OK2), the PCNB treatment increased stands over the nontreated control, indicating the importance of *Rhizoctonia solani* in stand establishment at these sites. The Vitavax-PCNB + Allegiance standard fungicide treatment increased stands compared to the nontreated control in 5 of 12 experiments (AR2, LA1, MS1, OK2, and OK3). The nominated treatments increased stands over the nontreated control from 58% of the sites (7 of 12 sites) to 92% the sites (11 of 12 sites) depending on the treatment. Treatments giving increases in stand compared to the nontreated control in 11 of the 12 sites were RTU Baytan-Thiram + Allegiance, Baytan + Ascend + Allegiance, and LS1006 + Ascend. Selected nominated fungicide treatments performed significantly better than the standard fungicide treatment, Vitavax-PCNB + Allegiance, at 5 locations (AL, AR2, LA1, TN, and VA). Treatments increasing stand above the standard fungicide treatment for 3 of the 12 sites where a

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response was found were Baytan + Ascend + Allegiance, LS1006 + Ascend, RTU Baytan-Thiram + Allegiance + Ascend + L0125 + L0241, and Apron XL-LS + Maxim 4FS + Azoxystrobin. The number of fungicide treatments significantly increasing stands over the nontreated control ranged from 2 of the 15 nominated treatments for the AL site to all of the nominated treatments tested for 4 sites (AR2, LA1, MS1, and OK2). The mean stand for a location was not related to locations where stands were increased by fungicide treatments.

Hypocotyl disease indices ranged from 2.0 at OK1, OK2, and TN to 3.4 at LA1, average 2.4 (Table 5). Root disease indices ranged from 2.2 at MS1 to 4.1 at AR2, average 3.0. *R. solani* was isolated from seedlings from the nontreated plots at 15 of 17 locations (Table 5). *R. solani* was isolated from over 20% of the seedlings at 5 locations (AL, LA1, LA2, MS1, and MS2). *Pythium* spp. were isolated from seedlings all locations (Table 5). Isolation frequencies for *Pythium* spp. were 20% or greater for 7 sites (AR2, GA, LA1, MS2, OK2, TX1, and TX3). *Thielaviopsis basicola* was isolated from seedlings at 5 of the 17 locations on the modified TB-CEN medium (AL, AR3, OK2, TN, and TX2) (Table 5). *Fusarium* spp. were isolated from seedlings at all locations (Table 5). Isolation frequencies for *Fusarium* spp. ranged from 42% to 100%.

Soil populations of *R. solani* were detected at 7 of the 17 sites, range 3 to 50 CFU/100 g of soil (Table 6). *Pythium* spp. were detected in soils at all 17 sites, range 7 to 417 CFU/g of soil. *T. basicola* was detected in 3 of the 17 soils assayed, range 3 to 74 CFU/g soil.

The mean percent stand for the locations was negatively correlated, –0.58 (P=0.02), with isolation frequency of *R. solani* from seedlings. The hypocotyl disease index was positively correlated with soil populations of *R. solani,* 0.67 (P=0.005), and weakly positively correlated with isolation frequency of *R. solani,* 0.45 (P=0.08). *T. basicola* recovery from seedlings was positively correlated with soil populations of *T. basicola,* 0.93 (P=0.0001). Isolation frequency of Fusarium spp. was positively correlated with mean plant stand for the locations, 0.55 (P=0.03).

Conclusions

The results from the 17 locations in the 2000 National Cottonseed Treatment Program indicated that seed treatment fungicides consistently improved stands of cotton compared to a nontreated control. Most of the nominated fungicide combinations improved stands over the nontreated seed at most of the sites where a response was found. PCNB alone increased stands for 3 sites, indicating an important role for *R. solani* in these tests. This is supported by the negative correlation between mean stand and *R. solani* isolation frequency and the positive correlation between hypocotyl disease index and isolation frequency from seedlings and soil populations of *R. solani*. *T. basicola* isolation frequencies from seedlings were positively correlation with soil populations of the pathogen. *Rhizoctonia solani*, *Pythium* spp., and *Fusarium* spp. were isolated from seedlings over all or most locations. *Thielaviopsis basicola* was an important seedling disease pathogen in only a few locations in 2000.

Disclaimer

This paper reports the results of research only. Mention of a pesticide in this paper does not constitute a recommendation by the University of Arkansas, Department of Plant Pathology, nor does it imply registration under FIFRA.

References

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Table 1. Fungicides, formulations and the active ingredients included in the 2000 National Cottonseed Treatment Program.

Common or Registered Name[1]	Formulation	Active Ingredient (%)
ALLEGIANCE (Metalaxyl)	Flowable	35% *N*-(2,6-dimethylphenyl)-*N*-(methoxyacetyl) alanine methyl ester
APRON XL-TL (Mefenoxam)	Liquid	33.3% (R,S)-2-[(2,6-dimethylphenyl)-methoxyacetylamino]-propionic acid methyl ester
APRON XL-LS (Mefenoxam)	Liquid	33.3% (R,S)-2-[(2,6-dimethylphenyl)-methoxyacetylamino]-propionic acid methyl ester
ASCEND (TCMTB)	Flowable	30% 2-(thiocyanomethylthio)benzothiazole
Azoxystrobin 100FS	Flowable	9.2% Methyl (E)-2-{2-[6-(2-cyanophenoxy)pyrimidin-4-yloxy]phenyl}-3-methoxyacrylate
BAYTAN 30 (Triadimenol)	Flowable	30% Beta-(4-Chlorophenoxy)-alpha-(1,1-dimethylethyl)-1*H*-1,2,4-triazole-1-ethanol
DIVIDEND 3MG (Difenoconazole)	Flowable	32.8% 1-{2-[4-(chlorophenoxy)-2-chlorophenyl-(4-methyl-1,3-dioxolan-2-yl)-methyl]}-1*H*-1,2,4-triazole
HM-9906		Helena Chemical Company
HM-9906-A		Helena Chemical Company
HM-9703		Helena Chemical Company
L0125		Gustafson Incorporated
L0241		Gustafson Incorporated
LS 1006-A2		Gustafson Incorporated
MAXIM 4FS (Fludioxonil)	Flowable	42% 4-(2,2-difluoro-1,3-benzdioxol-4-yl)-1*H*-pyrrole-3-carbonitrile
NU-FLOW M-HF (Myclobutanil)	Emusifiable conc.	25.1% A-butyl-a-(4-chlorophenyl)-1*H*-1,2,4-triazole-1-propanenitrile
NU-FLOW M (Myclobutanil)	Wettable powder.	40% A-butyl-a-(4-chlorophenyl)-1*H*-1,2,4-triazole-1-propanenitrile
NU-FLOW ND (Chloroneb & TCMTB)	Flowable	23.5% 1,4-dichloro-2,5-dimethoxy-benzene 9.0% 2-(thiocyanomethylthio)benzothiazole
NU-FLOW T (TCMTB)	Liquid	30% 2-(thiocyanomethylthio)benzothiazole
RTU-PCNB	Flowable	24% Pentachloronitrobenzene
RTU-BAYTAN-Thiram	Flowable	5% Beta-(4-Chlorophenoxy)-alpha-(1,1-dimethylethyl)-1*H*-1,2,4-triazole-1-ethanol, 15.3% Tetramethylthiuram disulfide
VITAVAX (Carboxin) - PCNB	Flowable	17% 5,6-dihydro-2-methyl-N-phenyl-1,4-oxathiin-3-carboxamide, 17% Pentachloronitrobenzene
WE-143		Wilbur-Ellis Company
WE-144		Wilbur-Ellis Company

[1]Registered chemical name, all capital letters.

Table 2. List of cooperators and procedures used in the 2000 National Cottonseed Treatment Program.

			Date				Row Length Counted		
Cooperator	Location		Planted	Sampled	Counted	Reps.	(ft)	Seed Planted	Soil Temperature[1]
K. McLean	Auburn, AL	(AL)	4/20	5/18	5/17	6	25	125	NA
R. Benson	Keiser, AR	(AR2)	4/21	5/22	5/22	6	40	180	21(19)
C. S. Rothrock	Clarkedale, AR	(AR3)	5/1	5/30	5/30	8	50	250	23(19)
R. H. Garber	Shafer, CA	(CA)	4/25	5/23	5/23	8	20	125	24(20)
B. Cochran	Tifton, GA	(GA)	4/10	5/10	5/10	5	35	105	24(21)
P. D. Colyer	Bossier City, LA	(LA1)	4/18	5/17	5/16	5	25	100	21(17)
B. Padgett	Winnsboro, LA	(LA2)	4/11	5/11	5/11	4	25	100	17(15)
W. E. Batson Jr.	Mississippi State, MS	(MS1)	4/25	5/23	5/23	5	83	240	20(15)
G. L. Sciumbato	Stoneville, MS	(MS2)	4/18	5/18	5/18	4	40	200	22(18)
L. Verhalen &	Tipton, OK	(OK1)	5/5	6/6	6/6	4	22	100	25(21)
B. E. Greenhagen	Altus, OK	(OK2)	5/9	6/6	6/6	4	22	100	25(21)
	Perkins, OK	(OK3)	5/16	6/15	6/15	4	22	100	22(20)
A. Y. Chambers	Jackson, TN	(TN)	4/28	5/30	5/30	10	25	100	18(15)
P. M. Thaxton	College Station, TX	(TX1)	4/7	5/11	5/9	8	30	100	18(14)
H. W. Kaufman	Lubbock, TX	(TX2)	5/5	6/2	6/2	4	37	185	NA
T. S. Isakeit	Weslaco, TX	(TX3)	3/31	5/8	4/18	4	18	100	20(17)
P. M. Phipps	Suffolk, VA	(VA)	5/3	6/5	6/2	4	60	180	19(13)

[1]Mean (Minimum) soil temperature; 3-day average following planting.

Table 3. Mean squares for combined analysis of variance across locations, 2000 National Cottonseed Treatment Program.

Source	Degrees of Freedom	Mean squares
Location	16	18319**[1]
Replication(Location)	76	248**
Treatment	18	1267**
Location*treatment	288	110**
Error	1360	71

[1] ** = significant *F*-test, *P*=0.0001.

Table 4. Cotton seedling stands for locations of the 2000 National Cottonseed Treatment Program.

		Plant Stand (%)																	
Treatment	**Rate (oz/cwt)**	**AL**	**AR2**	**AR3**	**CA**	**GA**	**LA1**	**LA2**	**MS1**	**MS2**	**OK1**	**OK2**	**OK3**	**TN**	**TX1**	**TX2**	**TX3**	**VA**	**Mean**
Baytan + Ascend + Allegiance	0.5 + 1.5 + 0.75	60	67	74	81	77	62	30	71	76	82	74	71	88	71	84	46	66	69
RTU Baytan-Thiram + Allegiance + Ascend + L0125 + L0241	3..0 + 1.0 +1.5 + 0.25 + 7.0	63	79	68	72	66	66	28	72	81	88	78	72	86	74	76	44	66	69
Apron XL-LS + Maxim 4FS + Azoxystrobin	0.32 + 0.08 + 3.07	58	66	68	78	76	63	38	74	74	84	74	61	89	65	84	45	65	68
Nu-Flow T + Nu-Flow M 40WP+ Apron XL-TL	2.25 + 0.79 + 1.0	52	68	70	75	71	59	34	64	70	85	80	70	86	68	85	47	69	68
HM 9906	12.0	64	73	68	76	74	61	35	71	77	86	67	62	88	65	75	52	60	67
Apron XL-LS + Maxim 4FS + Dividend 3FS	0.32 + 0.08 + 1.0	52	67	70	71	70	57	30	72	72	81	73	64	85	67	79	46	70	66
HM 9906-A	12.0	55	74	66	74	67	58	30	65	67	83	79	75	86	69	74	49	66	67
LS 1006 + Ascend	0.6 + 1.5	53	74	73	76	68	60	28	71	70	76	66	64	88	66	79	38	70	66
Nu-Flow T + Nu-Flow M 40WP+ Apron XL-TL + WE-143 + WE-144	2.25 + 0.79 + 1.0 + 0.035 + 0.035	60	75	69	76	64	55	36	70	67	85	75	72	86	64	76	36	63	66
RTU Baytan Thiram + Allegiance	3.0 + 0.75	58	70	73	78	70	56	29	67	76	81	73	65	85	74	74	39	69	66
Apron XL-LS + Maxim 4FS + Nu-Flow M-HF	0.32 + 0.08 + 1.25	46	71	68	72	71	60	34	67	80	85	69	56	86	70	82	42	66	65
Apron XL-LS + Maxim 4FS + Nu-Flow M-HF	0.32 + 0.08 + 1.75	54	70	71	71	71	58	30	68	77	83	74	40	88	71	85	42	71	65
HM 9703	12.0	52	72	68	76	62	61	24	63	70	81	78	60	84	66	80	48	63	65
Nu-Flow ND + Nu-Flow M 40WP + Apron XL-TL	7.5 + 0.79 + 1.0	55	70	60	71	69	62	24	71	78	87	72	61	87	66	81	44	62	65
Vitavax PCNB + Ascend + Allegiance	6.0 + 1.5 + 0.75	59	73	63	72	60	64	25	65	74	80	76	48	87	72	77	53	65	65
Vitavax-PCNB + Allegiance	6.0 + 0.75	45	65	66	75	64	52	25	66	81	84	72	61	84	65	80	48	58	6
RTU-PCNB	14.5	49	58	63	74	66	44	15	65	77	80	73	53	82	57	77	41	60	60
Allegiance	1.5	45	54	62	71	61	52	14	53	72	75	58	51	80	67	73	42	63	58
Nontreated	---	51	42	63	68	64	36	12	51	70	57	47	42	81	56	74	46	53	53
Location average		54	68	68	74	68	57	27	67	74	81	71	60	86	67	79	45	64	65
Coefficient of Variation (%)		18	11	12	9	15	14	36	10	13	13	12	18	5	15	11	24	10	
LSD (*P*=0.05)		11.5	8.3	8.2	7.1	NS	9.8	14.0	8.8	NS	NS	12.5	15.8	3.5	9.6	NS	NS	9.3	

Table 5. Disease ratings and isolation frequency of seedling pathogen groups for the 2000 National Cottonseed Treatment Program locations.

		Disease Index		Isolation Frequency (%)			
Location	**Nodes[2]**	**Hyp.[3]**	**Root[4]**	***Rhizoctonia solani***	***Pythium* spp.**	***Thielaviopsis basicola***	***Fusarium* spp.**
AL	1.8	2.5	3.3	36	18	38	66
AR2	3.2	3.0	4.1	2	20	0	90
AR3	4.0	2.7	3.5	18	10	80	72
CA	2.0	2.4	2.3	0	8	0	100
GA	3.4	2.2	3.7	6	22	0	66
LA1	2.8	3.4	3.6	22	44	0	74
LA2	3.6	2.7	3.7	30	8	0	42
MS1	3.6	2.5	2.2	32	6	0	44
MS2	4.2	2.5	2.4	40	32	0	70
OK1	6.6	2.0	2.4	2	12	0	88
OK2	3.0	2.0	3.1	2	24	12	80
OK3	5.0	2.4	2.9	6	12	0	82
TN	4.4	2.0	2.8	6	16	10	98
TX1	4.5	2.1	2.3	0	32	0	90
TX2	-	2.4	2.7	2	14	78	90
TX3	2.0	2.2	2.4	8	44	0	84
VA	4.4	2.4	3.1	14	12	0	94

[1]Isolation frequency is based on approximately 50 seedlings per location.

[2]Nodes based on five seedlings per location.

[3]Hypocotyl disease severity index; 1=no symptoms, 2=few pinpoint lesions or diffuse discolored areas, 3=distinct necrotic lesion, 4=girdling lesion, and 5=seedling dead.

[4] Root disease index; 1=no symptoms, 2=1-10% of the root system discolored, 3=11-25% of the root system discolored, 4=26-50% of the root system discolored, and 5>50% of the root system discolored.

Table 6. Soil populations of selected soilborne fungi from sites in the 2000 National Cottonseed Treatment Program.

Location	*Rhizoctonia solani*	*Pythium* spp.	*Thielaviopsis basicola*
	CFU[1]/100g	CFU/g	CFU/g
AL	ND[2]	17	0
AR2	ND	33	0
AR3	14	83	74
CA	ND	217	0
GA	14	33	0
LA1	48	200	0
LA2	ND	233	0
MS1	8	67	0
MS2	50	69	0
OK1	ND	33	0
OK2	ND	17	3
OK3	ND	30	0
TN	ND	133	0
TX1	ND	417	0
TX2	7	17	57
TX3	3	7	0
VA	ND	17	0

[1]Colony forming units.

[2]Populations were not detected in the soil sample; less than approximately 3 CFU/100 g of soil for *Rhizoctonia solani.*