

REPORT OF THE COTTONSEED TREATMENT COMMITTEE

FOR 1996

Compiled by C.S. Rothrock
Department of Plant Pathology,
University of Arkansas, Fayetteville, AR

Introduction

The 1996 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. Fourteen fungicide seed treatments were nominated by chemical industry representatives for evaluation in the 1996 National Cottonseed Treatment Program. A standard fungicide treatment Vitavax-PCNB + Apron and a nontreated control were included to assess efficacy of the nominations and seedling disease pressure. In addition, the fungicide treatments Apron 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 populations of selected soilborne fungi in soil also were included in the 1996 National Cottonseed Treatment Program by collecting seedlings and soil from the nontreated control plots at each location.

Materials and Methods

Fungicide treatment

Acid-delinted seed of *Gossypium hirsutum* L., 'Deltapine 50' or 'Paymaster HS26' (Delta and Pine Land Company, Scott, MS), were planted at all locations. 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 were mixed with water at a rate of 2% water to seed weight (v/w). Water also was 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.

Field experiments

Twenty field experiments were conducted by 17 cooperators across the U.S. Cotton belt (Table 2). However, seedling disease or soil population data was collected from only 18 sites in 1996. Soils were naturally infested with seedling

disease pathogens for all experiments. 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 28 to 55 days after planting, average 33 days, depending on the location. A soil sample and seedling samples from plots containing nontreated seed were taken from 28 to 62 days after planting, average 34 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 K. S. McLean, Northeast Louisiana University, for determination of populations of plant parasitic nematodes.

Seedling disease and pathogen isolation

Approximately 47 seedlings (range 26 to 52 seedlings) per location were 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 µl 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 (4), 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 (1), and *Rhizoctonia* populations were quantified on a modified Ko and Hora medium (3). 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 P₅ARP (2), 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). Analysis of percent stand over locations indicated a significant location by treatment interaction (Table 3), thus subsequent analyses were done by location. Treatment means for a location were separated by using a protected LSD at $P=0.05$. The Pearson-product correlation method was used to examine the relationship

among stand, disease, pathogen isolation frequency, and soil populations over locations.

Results and Discussion

Seed germination for nontreated seed was 95% and 74% for Deltapine 50 and Paymaster HS26, respectively. After the seed were treated with the fungicide treatments, seed germination ranged from 86% to 98% for Deltapine 50, with an average germination of 95%. Seed germination ranged from 74% to 94% for Paymaster HS26, with an average germination of 83%, after the seed were treated with the fungicide treatments. There was no consistent effect of any of the treatment combinations on germination.

There was a significant location, treatment, and location x treatment effect when cotton stands were analyzed over locations (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 20 experiments (Table 4). In all of these experiments at least one of the fungicide treatments performed better than the nontreated control. In 3 of these 12 experiments (AR2, OK2, and TN) the Apron treatment increased stands compared to the nontreated control. In 5 of the 12 experiments (OK1, OK2, OK3, TN, TX2) the PCNB treatment increased stands over the nontreated control. The number of fungicide treatments significantly increasing stands over the nontreated control ranged from 1 of the 14 treatments for the CA site to 14 of the 14 nominated treatments for two sites (OK2 and TN). The Vitavax-PCNB + Apron FL standard fungicide treatment increased stands compared to the nontreated control in 10 of the 12 experiments where significant stand differences were found among treatments. The nominated treatments increased stands over the nontreated control for 5 to 11 experiments depending on the treatment. The treatment RTU Baytan-Thiram + Apron FL + Thiram 42-S (3.0 + 0.75 + 1.0) increased stands at 11 sites, while the treatments Apron 350FS + Dividend + Maxim 4FS (0.9 + 1.25 + 0.08) and Thiram 75WDG + Nu-Flow M + Apron TL (2.7 + 1.25 + 2.0) increased stands at 10 of the 12 sites. Selected nominated fungicide treatments performed significantly better than the standard fungicide treatment at four locations (AR3, CA, FL, OK3). The mean stand for a location was not related to locations where stands were increased by fungicide treatments.

Hypocotyl disease indices ranged from 1.5 at TX1 to 2.8 at TN, average 2.1 (Table 5). Root disease indices ranged from 1.4 at OK1 to 4.3 at FL, average 2.5. Some discoloration of seedlings appeared to have occurred during shipping for the AR1 and FL locations. *R. solani* was isolated from seedlings in the nontreated plots at 15 of 18 locations (Table 5). *R. solani* was isolated from 68% of the seedlings at the MS1 site, and 4 locations had isolation frequencies greater than 25%. *Pythium* spp. were isolated from seedlings at 18 of 18 locations (Table 5). Eight

locations had isolation frequencies for *Pythium* spp. of 25% or greater. *Thielaviopsis basicola* was isolated from seedlings at four locations on the modified TB-CEN medium (Table 5). The greatest frequency of isolation of this pathogen from seedlings was 85% and 76% at AR3 and OK3, respectively. *Fusarium* spp. were isolated from seedlings at all locations (Table 5). Isolation frequencies for *Fusarium* spp. ranged from 36% to 98%. *Macrophomina phaseolina* was isolated from seedlings at 14 locations. The sites AL, CA, FL, GA, LA2, MS1, and OK1 had isolation frequencies above 10%. Sixteen sites were analyzed for nematode populations; only the LA1 site had a population considered great enough to be a problem, with 258 root-knot nematodes/500 cc of soil.

Soil populations of *R. solani* were detected at 3 of the 17 sites assayed, range 6 to 15 CFU/100 g of soil (Table 6). *Pythium* was detected in soils from all sites assayed, range 33 to 375 CFU/g of soil. *T. basicola* was detected in 3 of the 17 soils assayed, range 1 to 54 CFU/g soil.

The hypocotyl and root disease indices were positively correlated with each other, 0.66 ($P=0.01$). The mean stand for the locations was weakly negatively correlated with population of *R. solani*, -0.47 ($P=0.06$). The soil populations and isolation frequency of *T. basicola* were positively correlated, 0.94 ($P=0.0001$). Similarly, the soil populations and isolation frequency of *R. solani* were positively correlated, 0.56 ($P=0.05$).

Conclusions

The results from 20 locations in the 1996 National Cottonseed Treatment Program indicated that seed treatment fungicides consistently improved stands of cotton compared to a nontreated control. A number of fungicide combinations gave increased plant stands, however, some fungicide combinations gave more consistent stand responses than other fungicide combinations among the locations. Differences in disease severity and the frequency of pathogen isolation over locations may partially explain the variation in cotton seedling survival observed among the fungicide treatments. *Rhizoctonia solani*, *Pythium* spp., and *Fusarium* spp. were isolated frequently from seedlings over all or most locations. *Thielaviopsis basicola* was isolated from seedlings from a few locations. The soil populations and isolation frequency for the pathogens *T. basicola* and *R. solani* were positively correlated.

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 1996 National Cottonseed Treatment Program.

Common or registered name ¹	Formulation	Active ingredient (%)
APRON FL (Metalaxyl)	Flowable	28.4% N-(2,6-dimethylphenyl)-N-(methoxyacetyl) alanine methyl ester
APRON 3FS (Metalaxyl)	Flowable	33.3% N-(2,6-dimethylphenyl)-N-(methoxyacetyl) alanine methyl ester
APRON TL (Metalaxyl)	Liquid	11.5% N-(2,6-dimethylphenyl)-N-(methoxyacetyl) alanine methyl ester
BAYTAN 30 (Triadimenol)	Flowable	30% Beta-(4-chlorophenoxy)-alpha-(1,1-dimethylethyl)-1H-1,2,4-triazole-1-ethanol
Captan 4000	Flowable	38.5% N-(trichloromethylthio)-4-cyclohexene-1,2-dicarboximide
DIVIDEND (Difenoconazole)	Flowable	32.8% 1-{2-[4-(4-chlorophenoxy)-2-chlorophenyl-(4-methyl-1,3-dioxolan-2-yl)-methyl]}-1H-1,2,4-triazole
FB23		Gustafson Incorporated
MAXIM 4FS	Flowable	42% 4-(2,2-difluoro-1,3-benzodioxol-4-yl)-1H-pyrrole-3-carbonitrile
NU-FLOW M (Myclobutanil)	Emusifiable conc.	25% Alpha-n-butyl-alpha-(4-chlorophenyl)-1H-1,2,4-triazole-1-propanenitrile
NU-FLOW ND (Chloroneb & TCMTB)	Flowable	23.5% 1,4-dichloro-2,5-dimethoxybenzene, 9.0% 2-(thiocyanomethylthio)benzothiazole
RTU-PCNB	Flowable	24% Pentachloronitrobenzene
RTU-BAYTAN-Thiram	Flowable	5% Beta-(4-chlorophenoxy)-alpha-(1,1-dimethylethyl)-1H-1,2,4-triazole-1-ethanol, 15.3% Tetramethylthiuram disulfide
Thiram 42-S	Flowable	42% Tetramethylthiuram disulfide
VITAVAX (Carboxin) - PCNB	Flowable	17% 5,6-dihydro-2-methyl-1,4-oxathiin-3-carboxanilide, 17% Pentachloronitrobenzene
Thiram 75WDG	Wettable granule	75% Tetramethylthiuram disulfide
WE 120C		Wilbur Ellis Company

Table 2. List of cooperators and procedures used in the 1996 National Cottonseed Treatment Program.

Cooperator	Location		Date			Reps	Row length	Seed
			Planted	Sampled	Counted		counted	planted
W. S. Gazaway	Auburn, AL	(AL)	4/12	5/13	5/13	6	10	67
T. L. Kirkpatrick	Hope, AR	(AR1)	4/24	5/22	5/22	5	40	200
G. Palmer	Keiser, AR	(AR2)	4/18	6/19	6/6	8	25	150
C. S. Rothrock	Clarkedale, AR	(AR3)	5/3	5/31	5/31	7	50	250
M. R. Davis	Shafter, CA	(CA)	4/18	5/16	5/16	5	25	120
F. M. Shokes	Quincy, FL	(FL)	4/8	5/15	5/13	6	23	100
D. R. Sumner	Tifton, GA	(GA)	4/10	5/13	5/8	5	25	100
P. D. Colyer	Bossier City, LA	(LA1)	4/19	5/20	5/17	6	25	100
K. S. McLean	Monroe, LA	(LA2)	4/24	5/22	5/22	5	40	240
W. E. Batson	Mississippi State, MS	(MS1)	4/29	5/30	5/30	5	80	240
G. L. Sciumbato	Stoneville, MS	(MS2)	4/11	-	5/17	5	40	200
L. Verhalen &	Tipton, OK	(OK1)	5/2	6/3	6/3	4	20	100
	Altus, OK	(OK2)	5/2	6/19	6/19	4	20	100
B. E. Greenhagen	Perkins, OK	(OK3)	5/15	6/17	6/17	4	20	100
J. D. Mueller	Edisto, SC	(SC)	5/13	-	6/27	4	26	105
A. Y. Chambers	Jackson, TN	(TN)	4/26	5/29	5/28	10	20	100
P. M. Thaxton	College Station, TX	(TX1)	4/9	5/13	5/8	8	30	100
H. W. Kaufman	Lubbock, TX	(TX2)	5/7	6/4	6/4	4	37	185
T. S. Isakeit	Weslaco, TX	(TX3a)	2/23	3/25	3/24	4	20	100
T. S. Isakeit	Weslaco, TX	(TX3b)	3/15	4/12	4/12	4	20	100

Table 3. Mean squares for combined analysis of variance across locations, 1996 National Cottonseed Treatment Program.

Source	Degrees of freedom	Mean squares
Location	19	17,080 ^{*1}
Replication(Location)	88	320 [*]
Treatment	17	1,237 [*]
Location*treatment	323	121 [*]
Error	1504	68

¹* = significant F-test, P=0.0001.

Table 4. Cotton seedling stands for locations of the 1996 National Cottonseed Treatment Program.

Treatment	Rate (fl oz. form/cwt)	Plant stand (%)																			
		AL	ARI	AR2	AR3	CA	FL	GA	LAI	LA2	LA3	MS1	MS2	OK1	OK2	OK3	SC	Tn	TX1	TX2	TX3a
APRON 3FS + DIVIDEND + MAXIM 4FS	0.9 + 1.0 + 0.08	64	64	72	79	87	60	71	68	74	59	73	70	65	58	40	85	43	41	48	64
APRON 3FS + DIVIDEND + MAXIM 4FS	0.9 + 1.25 + 0.08	67	59	72	74	83	59	81	64	70	50	68	70	67	81	38	85	51	41	56	80
APRON 3FS + NU-FLOW M + MAXIM 4FS	0.9 + 1.5 + 0.08	67	51	69	78	87	63	73	63	77	61	75	72	63	75	31	84	55	42	59	73
BAYTAN 30 FL + FB23 + APRON FL	0.5 + 2.0 + 0.75	76	62	70	76	90	61	72	68	70	50	70	76	67	89	36	90	53	47	63	78
BAYTAN 30 FL + FB23 + APRON FL	1.0 + 2.0 + 0.75	77	53	76	79	92	60	72	71	71	49	79	72	68	81	32	88	40	47	57	72
BAYTAN 30 FL + Thiram 42S + APRON FL	1.0 + 2.0 + 0.75	69	61	74	79	86	62	65	62	83	52	79	68	71	89	33	85	52	51	60	70
Captan 4000 + NU-FLOW M + APRON TL	2.5 + 1.25 + 2.0	67	61	68	79	87	60	74	68	74	62	80	77	72	75	37	87	46	37	53	78
NU-FLOW ND + NU-FLOW M + APRON TL	7.5 + 1.75 + 2.0	79	62	68	76	84	60	65	66	76	56	79	74	66	84	36	89	53	46	59	74
RTU BAYTAN-Thiram + APRON FL + Thiram 42-S	3.0 + 0.75 + 1.0	76	65	77	78	88	70	76	71	76	54	75	63	68	79	33	86	47	40	58	76
RTU BAYTAN-Thiram + APRON FL + Thiram 42-S	3.0 + 1.0 + 1.0	75	64	71	77	89	58	80	66	80	63	73	51	65	88	35	88	47	42	61	73
Thiram 75WDG + NU-FLOW M + APRON TL	2.7 + 1.25 + 2.0	74	60	70	74	86	51	77	70	78	60	74	65	73	85	35	88	49	40	53	80
WE 120C	1.92	60	58	61	68	85	52	66	63	71	60	76	66	63	78	34	79	41	40	58	73
WE 120C + NU-FLOW M	1.92 + 1.75	71	63	61	72	78	57	66	60	77	51	73	75	67	81	36	80	37	34	46	78
WE 120C + NU-FLOW M + APRON TL	1.92 + 1.75 + 2.0	60	58	63	75	84	67	71	71	75	54	75	75	64	73	35	89	50	47	66	71
RTU-PCNB	14.5	67	58	59	67	79	56	70	59	80	60	77	62	65	74	35	82	42	45	63	71
APRON FL	1.5	67	58	66	66	82	51	71	50	75	53	70	47	69	57	36	82	43	31	53	62
Nontreated	---	62	54	57	63	80	56	65	49	72	47	72	42	35	57	24	70	38	28	52	68
VITAVAX-PCNB + APRON FL	6.0 + 0.75	73	55	72	70	78	58	77	67	73	55	71	60	66	72	31	90	48	44	52	74
Location average		69	59	68	74	84	59	72	64	75	55	74	66	65	76	34	85	46	41	56	73
LSD 0.05		NS ¹	NS	8	6	10	10	10	13	NS	NS	NS	17	11	10	NS	4	7	9	NS	NS

Table 5. Disease ratings and isolation frequency of seedling pathogen groups for the 1996 National Cottonseed Treatment Program locations.

Location	Disease Index		Isolation frequency (%) ¹			
	Hypocotyl ²	Root ³	R. Pythium solani spp		T. Fusarium basicola spp	
			solani	spp	basicola	spp
AL	2.1	3.2	14	2	12	71
AR1	2.4	3.9 ⁴	35	19	0	92
AR2	1.8	1.5	4	26	0	74
AR3	2.5	3.2	32	46	85	70
CA	1.9	2.1	2	8	0	98
FL	2.1	4.3 ⁴	18	10	0	94
GA	1.9	2.0	14	4	0	69
LA1	2.2	2.8	18	44	0	96
LA2	1.9	2.3	10	10	0	37
MS1	1.9	1.8	68	36	0	36
OK1	1.8	1.4	6	38	0	86
OK2	2.3	2.6	0	22	2	98
OK3	2.1	2.4	0	20	76	94
TN	2.8	2.9	32	42	0	70
TX1	1.5	1.6	6	12	0	90
TX2	1.9	1.5	6	33	0	83
TX3a	2.0	2.0	0	26	0	78
TX3b	2.6	3.0	9	20	0	78

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

² 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.

³ 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.

⁴ Discoloration associated with shipping.

Table 6. Soil populations of selected soilborne fungi from sites in the 1996 National Cottonseed Treatment Program.

Location	Rhizoctonia solani	Pythium spp.	Thielaviopsis basicola
	CFU/100g	CFU/g	CFU/g
AL	ND ²	50	0
AR1	ND	217	0
AR2	7	200	0
AR3	ND	375	31
CA	ND	125	0
FL	ND	75	0
GA	ND	250	0
LA1	ND	250	0
LA2	ND	167	0
MS1	15	300	0
OK1	ND	33	0
OK2	ND	50	1
OK3	ND	50	54
TN	ND	83	0
TX1	6	158	0
TX3a	ND	58	0

TX3b	ND	33	0
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¹ Colony forming units.

² Populations were not detected in the soil sample, less than approximately 3 CFU/100 g of soil.