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Multiple Cottonseed Treatments: Effect of Sequence of Application of Pesticides on Germination, Seedling Growth, and Survival[1]

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ABSTRACT

This study evaluates the effect of the sequence of application of pesticides used as seed treatments on germination, seedling growth, and field survival of cotton (*Gossypium hirsutum* L.). In separate tests three protectant fungicides, 2-(thiocyanomethylthio)benzothiazole (Busan 72), pentachloronitrobenzene + 5-ethoxy-3-trichloromethyl-1,2,4-thiadiazole (Terracoat L21), or bis(dimethylthiocarbamyl)disulfide (thiram) were used with the systematic fungicide 1,4-dichloro-2,5-dimethoxybenzene (chloroneb) and the systemic insecticide *0,0*-diethyl *S* [2-(ethylthio)ethyl] phosphorodithioate (disulfoton), and were applied in all feasible combinations of the various pesticides. Evaluations included germination tests at standard conditions, at constant 20 C, and a constant 30 C. Dry weight transfer (DWT) determinations were made at 20 and 30 C, and seedling survival in the field at Stoneville, Miss. 42 days after planting.

In all three studies the results indicated that the sequence of pesticide treatment affected subsequent performance of cottonseed. In the case of Terracoat L21, only seedling survival in the field was not significantly affected by the sequence of pesticide application. 'Traditional' sequence of application — protectant fungicide followed by the systemic fungicide and then by the systemic insecticide — was the best sequence with these pesticides. When Busan 72 was used, alternative sequences proved to be more effective. When thiram was used, application of disulfoton first, irrespective of the application sequence of the other pesticides, reduced seedling survival and germination values at 20 C. Our results indicate that any change in pesticide usage, where multiple pesticides are used for seed treatment, may require a change in the sequence of pesticide application to obtain optimum results. In some instances, germination values may be affected to the point that treated seed would be rejected for certification and possible sale because of substandard germination, whereas untreated seed would have been acceptable.

Additional index words: Fungicides, Thiram, PCNB, Chloroneb, Systemic insecticides, Disulfoton, Chemical interactions, *Gossypium hirsutum* L.

IN the U. S. essentially all of the cottonseed (*Gossypium hirsutum* L.) used for planting purposes is treated with at least a protectant fungicide. Much of the planting seed is also treated with a systemic fungicide, a systemic insecticide, or both. By the mid-1950's, the alkyl-mercury fungicides were the major seed-protectant fungicides used for cottonseed treatment (3). Systemic fungicides and systemic insecticides were developed for and used in combination with the alkyl-mercury seed protectants. In development testing and actual practice, the seed protectant was applied first, the systemic fungicide was applied next, with the systemic insecticide applied last. A major reason for the development of this "traditional" sequence was the nature of the pesticides. The seed protectant was either an aqueous solution, an emulsifiable concentrate, or a wettable powder; the systemic fungicide was a wettable powder that required a larger amount of water for effective coverage. The systemic insecticides, however, were an oil or an oil-based liquid. In 1971 the commercial usage of the alkyl-mercury fungicides ceased and a number of different seed-protectant fungicides replaced the mercury materials. These materials were equally if not more effective than the alkyl mercuries (3) and were compatible with the systemic fungicides and systemic insecticides (2, 4, 5). The seed-protectant fungicides now used present a spectrum of pesticide types: emulsifiable concentrates, wettable powders, and oil-based formulations. With the availability of these formulations the use sequence developed with the alkyl mercuries could be, and perhaps has been, changed. The purpose of this study was to determine if the sequence of pesticide application of three different protectant fungicides, a systemic fungicide, and a systemic insecticide affected seed germination, seedling growth, and seedling survival of cotton.

MATERIALS AND METHODS

Seeds used in these studies were acid delinted 'Stoneville 213' (85% germination), heavily infested with fungi (4). Evaluation of the seed lot indicated 15% of the seeds were nicked or cracked during harvesting and processing, only 38% of these damaged seeds produced normal seedlings. Chemicals were applied with a nasal-atomizer onto seeds tumbling in a rotating drum-mixer. The following pesticides and rates were used.

Seed-Protectant Fungicides

Busan 72[1], emulsifiable concentrate: 60% 2-(thiocyanomethylthio) benzothiazole, 1.96 ml/kg (3 fl oz/cwt).

Terracoat L21[1], oil base: 22.9% pentachloronitrobenzene + 11.4% 5-ethoxy-3-trichloromethyl-1,2,4-thiadiazole, 7.8 ml/kg (12 fl oz/cwt).

Thiram, wettable powder: 70% bis(dimethylthiocarbamyl) disulfide, 1.88 g/kg (3 oz/cwt).

Systemic Fungicide

Chloroneb, wettable powder: 65% 1,4-dichloro-2,5-dimethoxybenzene, 6.25 g/kg (10 oz/cwt).

Systemic Insecticides

Disulfoton, oil base: 96% *O,O*-diethyl *S*-[2-(ethylthio)ethyl] phosphorodithioate, 4.5 ml/kg (6.9 fl oz/cwt).

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[1] Mention of a trademark or proprietary product does not constitute a guarantee or warranty of the product by the U.S. Department of Agriculture and does not imply its approval to the exclusion of other products that may also be suitable. Received May 19, 1972.

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Materials were applied in the sequence indicated in Tables 1, 2, or 3. Treatments where pesticides are enclosed by a ( ) were mixed together and the appropriate amount of the mixture was applied in a single application. Materials were applied immediately one after another. Seeds were tumbled in the mixer until surface "moisture" disappeared. Upon completion of pesticide application, the treated seeds were packaged in tightly closed glass jars and were stored in the laboratory for 14 days before testing was started. Jars were opened only to withdraw samples.

Germination studies were conducted by placing 50 seeds between two 30.5- by 45.7-cm (12- by 18-inch) germination papers that were wet with distilled water, made into a roll, covered with wax paper, and held in a vertical position. Each treatment was replicated six times. Three different temperature regimes were used to evaluate effects on germination: (i) Standard germination test conditions (6), 16 hours at 20 C, and 8 hours at 30 C alternating for 12 days. After 4 days, germinated seeds were counted and removed from the seed-rolls and the rolls were returned to the germinator for final evaluation at 12 days. (ii) Continuous 20 C germination was evaluated after 7 days. (iii) Continuous 30 C germination was evaluated after 4 days. Percent germination was recorded for each seed-roll. A seed was considered to have germinated if the emerging radicle was normal and at least 8 mm long.

The effects of the various treatments on seedling growth were assayed by a modification of the dry weight transfer (DWT) method of Christiansen (1). Twenty-five seeds were placed in seed-rolls as above; at the end of the respective growth periods, the most uniform 20 seedlings were selected from each seed-roll. The cotyledons were separated from the plant axis. The tissues were separately oven-dried at 65 C for 24 hours, cooled in a desiccator, and weighed. The DWT method yields a measure of seedling growth in terms of the amount of cotyledon reserves transferred to the axial parts of the developing plant. It is expressed as follows:

$$\mathrm{DWT} = \frac{\text{Dry weight of axial plant tissue}}{\text{Dry weight of total plant}} \times 100$$

DWT determinations were made in two temperature-time regimes: 20 C for 7 days, and 30 C for 4 days. Each treatment was replicated six times.

A field evaluation of these three tests was possible through the cooperation of Dr. Robert G. Davis at the Delta Branch of the Mississippi Agricultural Experiment Station at Stoneville, Miss. A random block design was used with six replications. A single plot consisted of 150 seeds planted 3.8 cm deep in a 9-m row with a belt planter. Stand counts were made 42 days after planting on April 27, 1971.

RESULTS

The results of these three studies are summarized in Tables 1, 2, and 3. Because there were different time intervals between treatment and evaluation of germination and DWT, comparisons between tables are not valid. In these studies the time period from seed treatment to evaluation of germination and DWT varied from 2 weeks for Terracoat L21 to 9 weeks for thiram. Field survival studies were made at the same time but in different positions within the same immediate area.

The study evaluating the sequence of pesticide application using Terracoat L21 as the seed protectant fungicide is summarized in Table 1. In this study the treatments using the "traditional" sequence of pesticide application, treatments 2 through 5, gave results similar to those reported previously (2, 4, 5) in that there were no significant adverse effects on germination or DWT, and there were significant improvements in seedling survival in the field.

In order to evaluate possible significant effects of the sequence of pesticide applications, nine single degrees of freedom comparisons were made for each of the variables measured. Four of these comparisons proved to show significance in one or more instances and are identified as A, B, C, and D in Tables 1, 2, and 3.

Comparison A compares the results from "traditional" sequence of application of all three pesticides with the mean of the results of the other seven application sequences that were tested. In Table 1 the treatment with the traditional sequence of application of pesticides exhibited a lower DWT at 20 C than the mean of the seven other sequence-of-application treatments.

Comparison B compares the mean of the three treatments receiving the seed-protectant fungicide first and including chloroneb, with the mean of the three treatments receiving chloroneb first and including the seed-protectant fungicide. In Table 1 the application of Terracoat L21 first resulted in improved DWT at 30 C in contrast to the application of chloroneb first.

Comparison C compares the mean of the three treatments receiving the seed-protectant fungicide first and including disulfoton, with the mean of the three treatments receiving disulfoton first and including the seed-protectant fungicide. In Table 1 this com-

Table 1. Effects of sequence of application of Terracoat L21, chloroneb, and disulfoton to cottonseed on germination, seedling dry weight transfer, and seedling survival in the field.

Seed treatment and sequence of application		Germination, %†			Dry weight transfer at†		% seedling survival in field†
No.	Pesticides used‡	30C	20C	STD	30C	20C	
1	None (check)	79.3 abc	75.7 a-e	84.3 ab	32.8 a-d	30.8 bcd	48.5 ef
2	Terracoat L21	85.7 a	80.7 abc	85.3 ab	33.9 ab	32.6 b	77.5 a-d
3	Terracoat L21 + chloroneb	80.0 abc	79.3 a-e	78.0 bc	29.4 e-h	27.5 efg	85.2 a
4	Terracoat L21 + disulfoton	83.0 ab	81.7 ab	84.0 abc	32.1 bcd	29.0 def	69.3 bcd
5	Terracoat L21 + chloroneb + disulfoton	77.7 a-d	75.0 a-e	81.0 abc	28.5 fgh	26.3 gh	80.7 ab
6	Terracoat L21 + disulfoton + chloroneb	79.0 abc	82.7 a	80.0 abc	30.7 def	27.6 efg	75.0 a-d
7	Chloroneb	61.7 e	76.3 a-e	82.7 abc	25.0 i	25.3 gh	68.3 cd
8	Chloroneb + Terracoat L21	77.0 bcd	77.0 a-e	80.0 abc	27.1 hi	24.9 h	77.0 a-d
9	Chloroneb + disulfoton	79.0 abc	76.7 a-e	77.7 bc	28.2 gh	26.5 gh	55.2 e
10	Chloroneb + Terracoat L21 + disulfoton	80.0 abc	73.7 b-e	80.0 abc	28.2 gh	29.1 de	75.8 a-d
11	Chloroneb + disulfoton + Terracoat L21	77.3 bcd	73.3 b-e	75.7 c	27.9 gh	27.8 efg	72.5 bcd
12	Disulfoton	83.0 ab	80.0 a-d	88.0 a	34.7 a	35.4 a	41.2 f
13	Disulfoton + Terracoat L21	75.0 bcd	79.7 a-e	78.3 bc	33.3 abc	32.2 bc	70.7 bcd
14	Disulfoton + chloroneb	82.0 ab	76.3 a-e	81.7 abc	32.2 bcd	32.2 bc	66.5 d
15	Disulfoton + Terracoat L21 + chloroneb	77.3 bcd	73.3 b-e	77.0 bc	31.0 cde	30.5 bcd	74.8 a-d
16	Disulfoton + chloroneb + Terracoat L21	73.3 cd	72.3 cde	79.7 bc	29.6 efg	31.3 bcd	78.3 abc
17	(Terracoat L21 + disulfoton) + chloroneb	70.0 d	70.7 e	78.7 bc	31.2 cde	30.2 cd	84.2 a
18	Chloroneb + (Terracoat L21 + disulfoton)	70.3 d	71.3 de	75.7 c	28.0 gh	26.6 fgh	76.5 a-d
Coefficient of variation		7.7	8.4	7.5	6.3	6.4	11.9
Comparisons:	A. T5 vs. (T6, 10, 11, 15, 16, 17, 18)/7	-	-	-	-	*	-
	B. (T3, 5, 6)/3 vs. (T8, 10, 11)/3	-	-	-	*	-	-
	C. (T4, 5, 6)/3 vs. (T13, 15, 16)/3	*	-	-	-	*	-
	D. (T8, 10, 11)/3 vs. (T13, 15, 16)/3	-	-	-	*	*	-

* Significant single degree of freedom comparisons at the 5% level. † Means not followed by the same letter are significantly different at the 5% level as measured by Duncan's New Multiple Range Test. ‡ Pesticides enclosed by ( ) were mixed together and the appropriate amount of the mixture applied in a single application.

Table 2. Effects of sequence of application of Busan 72, chloroneb, and disulfoton to cottonseed on germination, seedling dry weight transfer, and seedling survival in the field.

Seed treatment and sequence of application		Germination, %†			Dry weight transfer at†		% seedling survival in field†
No.	Pesticides used†	30C	20C	STD	30C	20C	
1	None (check)	84.3 ab	77.7 abc	85.7 abc	34.8 ab	32.4 b	37.2 e
2	Busan 72	85.3 a	81.7 ab	87.7 ab	32.6 bc	30.7 b-e	68.5 bcd
3	Busan 72 + chloroneb	79.0 abc	78.6 ab	84.3 a-d	32.4 bc	30.0 c-f	82.3 a
4	Busan 72 + disulfoton	80.6 abc	79.6 ab	82.3 b-e	31.9 cd	28.8 e-h	59.7 d
5	Busan 72 + chloroneb + disulfoton	79.3 abc	83.3 a	83.0 a-e	31.3 cd	27.3 hi	79.3 ab
6	Busan 72 + disulfoton + chloroenb	78.0 abc	81.3 ab	77.3 de	27.9 e	26.7 i	80.7 ab
7	Chloroneb	79.7 abc	81.0 ab	84.3 a-d	32.8 bc	27.4 hi	68.8 bcd
8	Chloroneb + Busan 72	78.6 abc	76.3 abc	80.3 b-e	29.6 de	27.5 hi	75.0 abc
9	Chloroneb + disulfoton	78.3 abc	78.6 ab	81.0 b-e	32.9 bc	30.5 cde	73.7 abc
10	Chloroneb + Busan 72 + disulfoton	83.0 ab	81.7 ab	83.7 a-d	28.4 e	28.5 f-i	83.5 a
11	Chloroneb + disulfoton + Busan 72	73.0 c	80.3 ab	75.7 e	31.2 cd	27.9 ghi	85.5 a
12	Disulfoton	81.3 abc	81.3 ab	90.0 a	36.1 a	35.8 a	34.8 e
13	Disulfoton + Busan 72	77.6 abc	77.0 abc	81.3 b-e	32.9 bc	30.6 b-e	63.0 cd
14	Disulfoton + chloroneb	79.6 abc	70.0 c	82.0 b-e	33.1 bc	30.5 cde	64.5 cd
15	Disulfoton + Busan 72 + chloroneb	81.0 abc	77.7 abc	83.3 a-d	32.2 c	29.4 d-g	72.8 abc
16	Disulfoton + chloroneb + Busan 72	76.0 bc	79.3 ab	80.6 b-e	31.5 cd	28.9 e-h	82.8 a
17	(Busan 72 + chloroneb) + disulfoton	72.6 c	80.0 ab	82.0 b-e	31.4 cd	31.3 bcd	78.3 ab
18	Disulfoton + (Busan 72 + chloroneb)	74.0 c	73.7 bc	79.7 cde	32.6 bc	31.4 bc	82.0 a
Coefficient of variation		7.9	8.0	6.7	5.8	4.9	13.4
Comparisons:	A. T5 vs. (T6, 10, 11, 15, 16, 17, 18)/7	-	-	-	-	*	-
	B. (T3, 5, 6)/3 vs. (T8, 10, 11)/3	-	-	-	-	-	-
	C. (T4, 5, 6)/3 vs. (T13, 15, 16)/3	-	-	-	*	*	-
	D. (T8, 10, 11)/3 vs. (T13, 15, 16)/3	-	-	-	*	*	*

* Significant single degree of freedom comparisons at the 5% level. † Means not followed by the same letter are significantly different at the 5% level as measured by Duncan's New Multiple Range Test. † Pesticides enclosed by ( ) were mixed together and the appropriate amount of the mixture applied in a single application.

Table 3. Effects of sequence of application of thiram, chloroned, and disulfoton to cottonseed on germination, seedling dry weight transfer, and seedling survival in the field.

Seed treatment and sequence of application		Germination, %†			Dry weight transfer at†		% seedling survival in field†
No.	Pesticides used‡	30C	20C	STD	30C	20C	
1	None (check)	77.0 b	84.0 abc	85.7 a	33.4 bcd	33.3 a	41.8 e
2	Thiram	86.7 a	83.3 a-d	84.7 ab	34.6 abc	30.8 bc	61.8 bc
3	Thiram + chloroneb	82.3 ab	83.7 abc	83.0 a-d	31.5 d-g	28.4 def	76.2 a
4	Thiram + disulfoton	79.7 ab	84.0 abc	84.0 abc	33.3 bcd	28.9 cde	47.7 de
5	Thiram + chloroneb + disulfoton	76.7 b	85.3 a	83.7 abc	29.7 ghi	25.3 gh	71.7 ab
6	Thiram + disulfoton + chloroneb	77.0 b	84.7 ab	84.0 abc	30.9 e-h	26.6 fgh	75.0 a
7	Chloroneb	76.7 b	85.3 a	83.7 abc	34.5 abc	29.6 bcd	62.5 bc
8	Chloroneb + thiram	81.0 ab	81.3 a-d	82.3 a-c	35.1 ab	28.7 c-f	76.8 a
9	Chloroneb + disulfoton	80.0 ab	84.7 ab	88.3 a	33.0 b-e	31.2 b	54.0 cd
10	Chloroneb + thiram + disulfoton	83.3 ab	78.7 b-e	76.7 de	30.4 f-i	26.6 fgh	71.0 ab
11	Chloroneb + disulfoton + thiram	77.7 b	79.0 a-e	77.7 cde	29.2 hi	27.5 def	68.5 ab
12	Disulfoton	83.0 ab	83.0 a-d	87.3 a	36.4 a	35.0 a	27.3 f
13	Disulfoton + thiram	78.7 ab	82.0 a-d	83.3 abc	31.2 d-h	27.4 d-g	47.2 de
14	Disulfoton + chloroneb	77.3 b	78.3 a-e	76.0 e	32.2 def	29.1 b-e	51.3 de
15	Disulfoton + thiram + chloroneb	78.7 ab	76.3 cde	78.0 bcde	28.4 i	25.1 h	70.3 ab
16	Disulfoton + chloroneb + thiram	79.0 ab	75.7 de	85.3 a	33.0 b-e	27.6 def	72.7 ab
17	(Thiram + chloroneb) + disulfoton	75.3 b	73.3 e	77.3 cde	30.6 fgh	27.3 efg	71.0 ab
18	Disulfoton + (Thiram + chloroneb)	78.0 ab	83.0 a-d	81.7 a-e	32.5 c-f	28.0 def	77.0 a
Coefficient of variation		8.2	7.2	6.0	5.3	5.8	13.7
Comparisons:	A. T5 vs. (T6, 10, 11, 15, 16, 17, 18)/7	-	*	-	-	*	-
	B. (T3, 5, 6)/3 vs. (T8, 10, 11)/3	-	-	-	-	-	-
	C. (T4, 5, 6)/3 vs. (T13, 15, 16)/3	-	*	*	-	-	*
	D. (T8, 10, 11)/3 vs. (T13, 15, 16)/3	-	*	-	-	-	*

* Significant single degree of freedom comparisons at the 5% level. † Means not followed by the same letter are significantly different at the 5% level as measured by Duncan's New Multiple Range Test. ‡ Pesticides enclosed by ( ) were mixed together and the appropriate amount of the mixture applied in a single application.

parison indicated significant differences in the 30 C germination data and the 20-C DWT data. Application of Terracoat L21 first in the sequence significantly improved germination values but reduced DWT values at 20 C.

Comparison D compares the mean of the three treatments receiving chloroneb first and including the seed-protectant fungicide, with the mean of the three treatments receiving disulfoton first and including the seed protectant fungicide. In Table 1 this comparison indicated significant differences in DWT values at both 20 and 30 C. Application of chloroneb first resulted in a significant reduction in DWT at both 20 and 30 C.

In regard to survival in the field (Table 1), the sequence of application of pesticides did not significantly affect the results. With the exception of disulfoton alone or the chloroneb-disulfoton treatment all of the treatments significantly improved seedling survival.

The study evaluating the sequence of pesticide application using Busan 72 as the seed-protectant fungicide is summarized in Table 2. In this study the treatments using the "traditional" sequence of pesticide application, treatments 2 through 5, gave results similar to those reported previously (4), including a reduction of DWT values at 20 and 30 C associated with the treatment combination of Busan 72 and disulfoton.

The single degree of freedom comparison A (Table 2) indicated that a significant difference in response occurred in the 20-C DWT data. The traditional application sequence of all three pesticides retarded DWT in contrast to the mean of the seven other possible application sequences tested. Results indicate that either Busan 72 or chloroneb could be applied first without significantly affecting any of the measurements made in this study, comparison B. However, significant differences in DWT data at 20 and 30 C were evident. Comparison C indicated that the application of disulfoton first resulted in significantly higher DWT values than when Busan 72 was applied first.

Comparison D was significant for three of the variables measured. When disulfoton was applied first, in contrast to applying chloroneb first, there was a significant improvement of DWT values at 20 and 30 C. In regard to seedling survival, however, the opposite was true. The mean survival of seedlings receiving treatments with chloroneb applied first in the sequence of application was significantly higher than the mean seedling survival from treatments where disulfoton was applied first.

The study evaluating the sequence of pesticide application using thiram as the seed-protectant fungicide is summarized in Table 3. In this study the treatments using the "traditional" sequence of pesticide application, treatments 2 through 5, gave results similar to those reported previously (2, 4, 5).

The single degree of freedom comparison A (Table 3) indicates significant differences in the germination and DWT data at 20 C. In the case of the 20-C germination data, the "traditional" sequence of application of all three pesticides resulted in a higher germination value than the mean of the other seven sequences of application. In the case of the DWT at 20-C the reverse was true; the mean DWT of the seven other possible sequences of application was greater than the DWT values obtained with the "traditional" sequence of application. Comparison B indicated that either thiram or chloroneb could be applied first without significantly affecting the variables measured. Significant differences in 20-C and standard germination data, however, and in seedling survival were evident in comparison C. Results of this comparison indicated that the application of thiram first resulted in significantly higher values for each of these three variables than when disulfoton was applied first. Comparison D was also significant for the 20-C germination and seed-survival data. When chloroneb was applied first, rather than disulfoton, the mean values for the chloroneb-first applications were significantly higher.

In all three of these studies the treatment with disulfoton alone presented some interesting results. At all temperatures the seeds treated with only disulfoton consistently had excellent DWT values and high germination values, in many instances the highest values. In regard to seedling survival, the exact opposite was true, with survival values either equal to or significantly lower than the untreated seeds. In all instances, the inclusion of a seed-protectant fungicide, or a seed protectant and chloroneb significantly "safened" this deleterious effect of disulfoton on field seedling survival.

In statistical analysis of these data, all possible correlations between the germination and DWT data were made with the seedling-survival data. Only standard germination was found to be significantly correlated (1% level), —0.5187, and even so, only could be assumed to account for 25% of the variability in seedling survival. Undoubtedly, by removing the "opposite effect" of the disulfoton-only treatment, this value could be raised, but at best any of these measures are poor estimates of field performance of cottonseed.

DISCUSSION

In initiating this study our objective was to clarify the role that the sequence of application of pesticides to cottonseed has on their subsequent performance. While these studies may be of help in clarifying the question, they certainly failed to yield a simple answer.

Our results indicate that any change in pesticide usage, where multiple pesticides are used for seed treatment, may require a change in the sequence of pesticide application to obtain optimum results from the multiple treatment. With the elimination of alkylmercury fungicides, numerous other fungicides are being used as seed-protectants, and in this study we have used only three of the many available. In the U. S. there are two other systemic insecticides registered for use on cottonseed, *O,O*-diethyl *S*-[(ethylthio) methyl] phosphorodithioate (phorate) and 3-hydroxy-N-methyl-cis-crotonamide dimethyl phosphate (monocrotophos). Also, there is another systemic fungicide registered for cottonseed use, 5,6-dihydro-2-methyl-1,4-oxathiin-3-carboxanilide (carboxin). Consequently, the possible combinations of treatments that are available are numerous. If the possible sequence of application is also considered, the possible multiple treatments exceed 500, considering only six seed protectant fungicides.

Consequently, seed processors should evaluate not only the materials they are using, but also the sequence of application that is used. The importance of this is apparent in evaluating the standard germination data in our tables. With each seed protectant we evaluated there were some application sequences that reduced germination levels of the untreated seed (85%) to well below 80%. In many states this reduction in germination would be sufficient to bar the sale of these seeds as certified or registered seeds because of a low germination value.

In our studies the "traditional" sequence of application of pesticides (seed-protectant fungicide followed by systemic fungicide and then by systemic insecticide) gave acceptable values for both germination and seedling survival. Some of the other sequences of application gave higher germination and seedling survival values than the "traditional" sequence in this study. In most instances, however, a change in the application sequence resulted in reduced values. It should be noted that the "traditional" sequences of cottonseed treatment with these pesticides have proven to be effective in a wide range of environmental conditions (2, 3, 4). Consequently, any change in the sequence of treatment should also be evaluated under a wide range of conditions.

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