Influence of Low Temperature During Cottonseed Germination on Growth and Disease Susceptibility

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ABSTRACT

Low-temperature (10 C) treatments of germinating cottonseed that subsequently were planted in the field resulted in increased severity of pre-emergence damping-off and in decreased yields. Generally, seedling survival decreased as the period of low temperature was lengthened; seedling mortality was highest for seed held for 24 hr at 30 C, then chilled for 4 or 6 days. Survival of seedlings from chilled seed generally increased as temperatures associated with later planting dates increased. In greenhouse tests, chilling of seed decreased shoot and root growth of seedlings grown in fumigated soil, and increased severity of damping-off in field soil and in fumigated soil artificially infested with *Pythium irregulare* and *Rhizoctonia solani*. Phytopathology 61: 1426-1429.

Additional key words: Gossypium hirsutum.

Low temperatures during the early stages of germination of cottonseed influence the growth and morphological development of the seedlings (2) and their subsequent field performance (4). Christiansen (1) reported that chilling of germinating seed at 10 C sometimes resulted in injury to the radicle and hypocotyl. Such damage is apparently associated with changes in cell permeability and high solute loss from the radicles (3). Reports that chilling decreases the rate of seedling emergence and vigor, increases solute loss, and causes root damage suggested to us that disease susceptibility may also be influenced. We conducted greenhouse and field studies to determine the influence of low temperature during germination of cottonseed on growth and on susceptibility of the seedlings to certain seedling pathogens. A preliminary report on a portion of this work has been published (6).

MATERIALS AND METHODS.—*Field studies.*—Plots were established in the Lower Coastal Plain, Tifton, and in the Piedmont region, Athens, Ga., during 1970. Acid-delinted cotton (*Gossypium hirsutum* L. 'Coker 201')-seed treated with Ceresan L (2.8% Methylmercury 2,3-dihydroxypropylmercaptide and 0.62% methylmercury acetate) and chloroneb (Demosan 65W) (1.3 and 6.2 g/kg, respectively) were soaked in tap water for 1 hr. Cracked and floating seeds then were discarded, and the remaining seeds rolled in wet paper towels, wrapped in aluminum foil, and placed at 30 C for 24 hr to initiate germination. Germinating seed with radicles about 2 cm long were planted directly (check), or were subjected to 10 C (hereafter referred to as chilling) while still in the paper towels for 2, 4, or 6 days prior to planting. All treatments were planted simultaneously in the field. Seed were hand-planted 2.5 cm deep, two seed/hill, with hills and rows 15 cm and 1 m apart, respectively. Plots, 7.6 m in length, were replicated 6 times in a randomized complete block design. Three seeding dates were used to provide early, intermediate, and late plantings comparable to those used by growers at each location. Plating dates were 20 April, 11 May, and 1 June at Athens; and 10 April, 1 May, and 22 May at Tifton.

Irrigation was used as needed to provide optimum moisture conditions for emergence and growth. The plots were maintained according to standard grower practices for the area. Data on emergence, survival, and relative vigor of plants were taken at each location. At the Athens location, yield of seed cotton was determined by hand picking.

Greenhouse studies.—Atlas 67 or Coker 201 cottonseed treated with Ceresan L (1.3 g/kg) were handled as described for the field studies, except that in initial tests prior to rolling the seeds in paper towels, the testae were removed to insure uniform radicle emergence (2). This step was eliminated in later tests; instead, extra seed were germinated, and only those with uniform radicle growth selected. Chilled (held at 10 C for 2, 3, 4, or 6 days) or nonchilled (check) seed were planted in field soil, in fumigated soil, and in fumigated soil artificially infested with *Pythium irregulare* Buis. or *Rhizoctonia solani* Kuehn. The field soil, which had high populations of *Pythium* spp. and *R. solani* and was obtained from the same area near Athens where the field studies were made, was mixed with vermiculite (3:1, v/v); *P. irregulare*, isolated from cotton roots, was grown in a moist sand-cornmeal (90:10, v/v) medium in 1-liter jars or in 2.8-liter Fernbach flasks for 14-18 days at room temperature. Cultures were mixed thoroughly, added to a fumigated (methyl bromide, 454 g/m^3) soil-sand-vermiculite (3:1:1, v/v) mix at the rate of 55 g/kg mix, and mixed for 3 min in a cement mixer. *Rhizoctonia solani* was grown in 100 ml potato-dextrose broth in 250-ml flasks on a wrist-action shaker for 7 days at room temperature. The mycelium was rinsed in sterile water, fragmented in distilled water in a Waring Blendor for ca. 5 sec, and mixed with soil at the rate of 0.3 g wet wt mycelium/kg soil.

Tests were conducted either on a greenhouse bench, where the temperature ranged from 14 to 28 C (mean 20 C), or in water-bath tanks where the

temperature was regulated to increase gradually from 15 to 27 C (mean 19 C) during the day and decrease to 15 C during the night. For tests that involved the use of the temperature tanks, 20 seed were planted in 4-liter cans filled with fumigated soil that either was not infested or was infested with *Pythium*. When the greenhouse bench was used, 20 seed of each treatment (0- and 3-days chilling) were planted in rows in wood flats (50 x 35 x 8 cm) filled with field soil, fumigated soil, or fumigated soil artificially infested with *Pythium* or *Rhizoctonia*. Seed in all tests were planted ca. 1.8 cm deep. Treatments were replicated 5 times, and tests were run at least twice. Data on emergence and survival were taken 7 and 14 days after planting.

RESULTS.—*Field studies.*—Soil temperature and moisture conditions during 1970 were near optimum for the emergence and growth of cotton plants. Despite these generally favorable conditions, stands were drastically reduced when seed were subjected to low temperatures during germination (Table 1). Generally, stands decreased as the period of chilling increased; the greatest stand reduction occurred with seed chilled for 4 or 6 days. At Tifton, stands resulting from chilled seed increased as the planting date was delayed. At Athens, stands from chilled seed increased from the early to intermediate dates of planting, but generally decreased again in the late planting. Mean air temperatures were 19, 21, and 24 C at Tifton, and 20, 23, and 22 C at Athens, during the 7 days following the early, intermediate, and late dates of plantings, respectively. Stand reductions with chilled seed at both locations were mainly due to seed rot and pre-emergence damping-off. Postemergence losses, though greater in the early plantings at both locations, were low in all cases and were the same for plants from chilled and from nonchilled seed. Plants from chilled seed were initially less vigorous than the controls, but this difference was not detectable by midseason. However, the reduction in stand affected by chilling seed for 4 or 6 days resulted in a significantly lower yield of seed cotton regardless of planting date (Table 1). Yields were similar for the April and May plantings, and lowest for the 1 June planting.

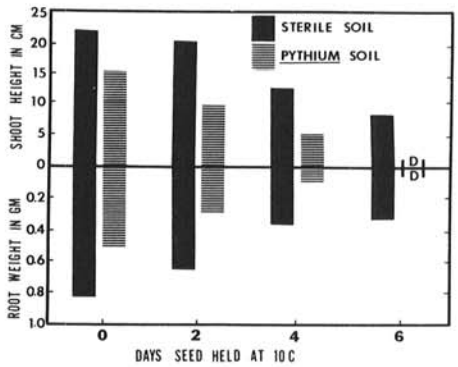

Fig. 1. Growth of cotton from seed germinated at 30 C for 24 hr, held for various periods at 10 C, then planted in fumigated or *Pythium irregulare*-infested soil at alternating temperatures of 15 and 27 C.

Greenhouse studies.—The low-temperature treatments given to germinating cottonseed resulted in decreased growth of both shoots and roots of plants in fumigated soil (Fig. 1) and in increased disease susceptibility of plants in field soil and in artificially infested soil (Fig. 1, Tables 2, 3). Plant growth

TABLE 1. Field performance of cottonseed subjected to 10 C during germination and planted at two locations in Georgia

Days seed held at 10 C[b]	Planting date[a]					
	Early		Intermediate		Late	
	Survival[c]	Yield[c]	Survival	Yield	Survival	Yield
Planted at Athens	%	*kg/hectare*	%	*kg/hectare*	%	*kg/hectare*
0	63.5 w[d]	3,362 w	67.5 w	3,423 w	54.2 w	2,528 w
2	40.2 x	3,380 w	55.5 x	3,175 w	35.2 x	2,412 w
4	15.2 y	2,031 x	33.5 y	2,371 x	22.7 y	1,355 x
6	2.3 z	562 y	12.2 z	1,276 y	13.2 y	853 y
Planted at Tifton						
0	52.8 w		42.7 w		80.2 w	
2	21.8 x		41.3 w		72.2 wx	
4	1.7 y		22.7 x		64.6 wx	
6	2.7 y		6.0 y		39.2 y	

[a] Planting dates were 20 April, 11 May, and 1 June at Athens, and 10 April, 1 May, and 22 May at Tifton.

[b] Seeds were germinated in rag dolls at 30 C for 24 hr, then planted directly (0 days), or were subjected to 10 C for 2-6 days prior to planting.

[c] Survival refers to final stand expressed as per cent of seed planted. Each value is based on six replications of 100 seeds each. Yield values are for seed cotton picked by hand.

[d] Column means not followed by the same letter are significantly different ($P = .05$) as determined by Duncan's multiple range test.

TABLE 2. Emergence and survival of seedlings from cottonseed held first at 10 C, then planted in soil infested with *Pythium irregulare*

Days seed held at 10 C[a]	Fumigated soil		*Pythium*-infested soil	
	Emergence[b]	Survival[b]	Emergence	Survival
	%	%	%	%
0	94 w[c]	93 w	93 w	64 x
2	90 w	90 w	91 w	52 x
4	89 w	89 w	69 x	15 y
6	49 x	49 x	19 y	1 z

[a] Seed were germinated in rag dolls at 30 C for 24 hr, then planted directly (0 days) or subjected to 10 C for 2-6 days prior to planting them in temperature tanks at 15-27 C (mean 19 C).

[b] Each value is based on five replications of 20 seed each. Data were taken 2 weeks after planting.

[c] Column and line means not followed by the same letter are significantly different ($P = .05$) as determined by Duncan's multiple range test.

TABLE 3. Emergence and survival of seedlings from chilled and nonchilled cottonseed planted in soil and grown in the greenhouse

	% Emergence		% Survival	
	Days seed held at 10 C[a]			
Field soil	0	3	0	3
Not fumigated	56.1[b]	7.5	33.7	3.3
Fumigated, *Pythium*-infested	61.0	28.0	54.0	17.8
Fumigated, *Rhizoctonia*-infested	23.0	2.0	2.0	0.0
Fumigated	98.0	93.0	95.0	90.0

[a] Seed were germinated in rag dolls at 30 C for 24 hr, then planted directly (0 days) or subjected to 10 C for 3 days prior to planting them in the greenhouse at 14 to 28 C (mean 20 C).

[b] Each value is based on five replications of 20 seed each. Emergence and survival data were taken 8 and 14 days after planting, respectively. All 0- and 3-day comparisons except those for sterile soil are significantly different ($P = .05$) according to Duncan's multiple range test.

[c] Field soil prepared as a soil-sand-vermiculite mixture (3:1:1, v/v) was fumigated with methyl bromide (454 g/m^3 soil), and either not infested or infested with *Pythium irregulare* or *Rhizoctonia solani*.

decreased as the period of chilling was increased (Fig. 1). Emergence of plants in fumigated soil was lower than that of control plants only when seed were subjected to 6 days of chilling (Table 2). Tap root deterioration and lesions on the hypocotyl similar to those described by Christiansen (2) were observed on plants subjected to the 4- and 6-day chilling treatments and grown in fumigated soil. Chilling also increased the incidence of damping-off in *Pythium*-infested soil in the temperature tanks (Table 2) and in natural field soil, or in *Pythium*- and *Rhizoctonia*-infested soil in the greenhouse bench tests (Table 3). Most of the loss associated with plants from chilled seed was due to seed rot, pre-emergence damping-off, or very early postemergence damping-off (Fig. 2).

DISCUSSION.—These results corroborate Christiansen's findings (1, 2) that low temperature during germination reduces subsequent growth of cotton and causes tap root and hypocotyl damage. Our results also show that chilling increases the susceptibility of germinating seed to certain soil-borne pathogens, and results in marked increases in seed rot and pre-emergence damping-off. Similar results were reported by Hayman (5), who found that holding cottonseed in field soil at 12 C for 3 days prior to incubation at 24 C increased damping-off caused by *R. solani*, as compared with seeds incubated in the same soil at 24 C without the initial 12-C treatment. Schulz & Bateman (8) also reported that low temperature (5 C) during the first 24 hr of germination increases susceptibility of several plant species other than cotton to attack by *R. solani*. In our tests, differences between chilled and nonchilled seed in their susceptibility to pathogens were most evident under marginal conditions for disease development, such as when the inoculum level and

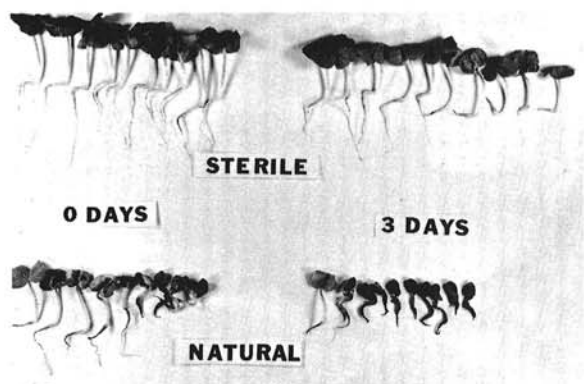

Fig. 2. Cotton plants from chilled (3 days at 10 C, right) and nonchilled (left) seed 5 days after planting the seed in field soil (natural) containing high populations of *Rhizoctonia solani* and *Pythium* spp., and in methyl bromide-fumigated soil (sterile).

temperature were suboptimum for the pathogen. Under optimum conditions for the pathogen (high infestation and optimum temperature), seedlings from both chilled and nonchilled seed succumb readily. How chilling effects a change in seedling susceptibility was not determined. However, large losses of solutes from radicles (3) may be involved, because plant exudates stimulate certain root-infecting fungi (7). Hayman (5) showed that increased seed exudation at low temperature is responsible for increased pre-emergence damping-off of cotton seedlings by *R. solani*. Changes in host vigor and the presence of necrotic tissue on the radicles of chilled seed are other possible explanations for our results.

In field tests, Christiansen & Thomas (4) found that the reduced seedling vigor resulting from chilling resulted in reduced plant height throughout the growing season, delayed fruit maturity, and reduced fiber quality, but did not influence yield. Early in the season we observed some growth retardation of plants from chilled seed, but this effect was not generally detectable by midseason. In our tests, plant stands from chilled seed were usually depleted by damping-off organisms; this allowed the remaining plants to develop with less competition. Christiansen & Thomas (4) obtained uniform stands by starting plants in sterile soil in the greenhouse and transplanting them to the field when growing conditions were favorable.

In the field tests, we tried to simulate a grower situation where seed are planted under favorable conditions for germination, but the soil temperature drops after radicle emergence. However, our tests differed from the field in that the seed were not exposed to pathogens during the low-temperature period, and some seed exudates remained in the paper towels after the seed were planted. The probability that a temperature change similar to that used in our study will occur is highest for an early planting, and decreases for later plantings.

Differences in plant survival from chilled seed planted on different dates apparently are related to changes in temperature. At Tifton, each delay in date of planting was associated with an increase in the number of surviving plants and higher temperatures. At Athens, plant survival was higher for the intermediate than the early planting, but lower in the late planting, and these differences were associated with corresponding changes in temperature. The decreased yield in the late planting was due primarily to the failure of many bolls to open prior to freezing.

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