

## Relation of Air Temperature to Development of *Verticillium* Wilt on Cotton in the Field

R. H. Garber and John T. Presley

Plant Pathologists, Crops Research Division, ARS, USDA, Shafter, California 93263, and Beltsville, Maryland 20705, respectively.

Accepted for publication 23 September 1970.

### ABSTRACT

Two cotton strains grown in two seasons had different levels of field tolerance to *Verticillium* wilt. Neither Cal 7-8, the more susceptible, nor Acala 4-42-77, the more tolerant, were seriously affected by the disease in a warm summer. In a cool summer, the relative disease tolerances of the strains were readily separated. In a warm summer, *V. albo-atrum* was isolated from plants with symptoms in early summer and fall. A decline in isolations in midsum-

mer was attributed to a prolonged hot period from late June through August. In the cool summer, the fungus was isolated from most plants throughout the summer. In both seasons, the organism was present in many symptomless plants. High temperatures can mask or obscure the influence of pathogen population and virulence in determining disease severity. *Phytopathology* 61:204-207.

*Verticillium albo-atrum* Reinke & Berth. may be present from root tips to leaf tips in cotton (*Gossypium hirsutum* L.) plants with no symptoms of the *Verticillium* wilt disease (3). Four days following inoculation of roots, Garber & Houston (3) isolated the organism from leaves. Five additional days elapsed before chlorosis appeared (4).

Optimum temp for growth of the causal organism, depending on the isolate, is usually 22 to 23 C (6). Isolates vary considerably in their ability to grow between 27 and 34 C, the maximum temp range.

All cotton varieties tested by Bell & Presley (1) were susceptible at 22 C and resistant at 32 C. Susceptible varieties developed symptoms, but resistant varieties were symptomless at 25 to 29 C. Tolerant varieties, however, were susceptible at 25 C, tolerant at 27 C, and resistant at 29 C. Halisky et al. (6) observed that incidence and severity of wilt were reduced as soil temp were increased above 25 C. Brinkerhoff et al. (2) found that three varieties of cotton grown under a 12-hr diurnal cycle of 28 C (day) and 18 C (night), following inoculation with *V. albo-atrum*, were severely diseased. When the daytime temp was raised to 36 C, the varieties remained symptomless. When the three varieties were held at 36 C day temp and 18 C night temp for 7 days, then transferred to a day temp of 28 C, one was resistant, one tolerant, and the other susceptible. Bell & Presley (1) observed, in the greenhouse, that diseased tolerant and resistant cotton strains recover completely from *Verticillium* wilt when grown at 27 to 30 C for several months. Leyendecker (7) reduced the incidence of disease by raising the height of seedbeds which increased soil temp several degrees. In California we observed that cotton plants which exhibit disease symptoms in late May or June usually recover if extended periods of warm days and nights occurred in late June and July.

In our investigations, disease observations were compared with min and max air temp to determine the relationship between air temp, the presence or absence of the organism in the host, and disease symptoms in plants grown in the field.

**MATERIALS AND METHODS.**—Plants of Cal 7-8, a

susceptible cotton strain (S); and Acala 4-42-77, strain (T) with the highest wilt tolerance of commercial varieties in California; were grown in six-row plots 43 m in length, replicated four times. The strains were planted in the same plots of soil each season. Approximately 280 plants in each replication were observed for disease symptoms in the first true leaf stage in May and throughout the growing season, until the plants were chemically defoliated just prior to harvest in October. Diseased and symptomless plants were tagged and observed throughout the growing season. Leaves with and without chlorotic symptoms were picked from plants with symptoms. Corresponding mature lower and upper leaves were picked from adjacent plants without symptoms. Isolations were made from 60 leaf parts of each category collected at intervals throughout the summer. Plant parts used for isolations were washed in running water for 3 hr and surface-disinfested in 0.5% aq sodium hypochlorite. A portion of each leaf blade, petiole leaf-blade juncture, petiole, and lower and upper stem in some instances, were placed on the surface of water agar in petri plates, incubated at 22 C for 5 days, and examined for presence of *V. albo-atrum*. Foliar and vascular disease ratings were made throughout each season.

**RESULTS.**—More days had temp above 35 C in 1967 than in 1966 (Table 1). The average maximum temp of the days above 35 C was also higher in 1967 than in 1966. The minimum temp were also higher in 1967 than in 1966, with very few days below 18 C. In addition, the average minimum temp of days below 18 C was higher in 1967.

In June 1966, 3 times as many S plants as T plants showed chlorotic leaf symptoms of *Verticillium* wilt (Fig. 1). The number of diseased S plants doubled as the summer progressed, and in September nearly all showed leaf symptoms. Less than 1 in 10 of the T plants showed foliar symptoms through August, but in September almost one-half had symptoms.

Less disease occurred in 1967 than in 1966. For every five T plants, ten S plants had chlorotic leaf symptoms. Some plants affected in June recovered. New leaves forming on these plants were free of disease damage

TABLE 1. Comparison of maximum and minimum air temp, Shafter, California, 1966-67

Observation period	Year	Max temp above 35 C	Max temp of days above 35 C	Min temp below 18 C	Min temp of days below 18 C	Infected plants
		days	avg	days	avg	%
20-30 June	1966	4	37.1	9	15.8	88
	1967	7	37.8	4	17.5	90
1-31 July	1966	14	37.3	19	17.6	100
	1967	26	37.4	4	17.4	30
1-31 Aug.	1966	23	36.8	11	15.2	100
	1967	23	37.3	5	17.6	30
<b>Totals</b>						
20 June-31 Aug.	1966	41	36.9	39	15.5	
	1967	56	37.4	13	17.5	

until late in the summer, when the number of S plants with symptoms approx doubled, increasing from 1 to 2 plants in 10.

*Cultures from diseased plants.*—In June, July, and August 1966, *V. albo-atrum* was isolated from most leaves of both S and T plants with disease symptoms (Fig. 2-A, B). The fungus was present in fewer leaves in September than in October 1966. In June 1967, most plants with symptoms yielded *V. albo-atrum* when cultured (Fig. 2-C, D). The percentage of infected plants dropped sharply in July and August 1967, then climbed rapidly in September and October, when the fungus was present in most plants.

*Cultures from symptomless plants.*—In mid-May 1966, when plants were in the first true leaf stage, *V. albo-atrum* was isolated from approx 1 plant in 10 of both S and T symptomless plants. The number of plants yielding the fungus increased in June, and in July approx 50% of the plants were infected (Fig. 2-A, B). The percentage of infected plants remained about the same or decreased slightly in August and September, but by October almost every plant was infected.

In 1967, more plants were also infected in July than in June (Fig. 2-C, D). The percentage of infected S plants decreased in August but increased sharply in September. Fewer T plants were infected in August,

and by September most plants were apparently free of the fungus. The fungus was isolated from 9 out of 10 symptomless plants in October.

*Fungus infection and vascular discoloration in plants after harvest.*—*Verticillium albo-atrum* was isolated from some portion of most plants after harvest. The fungus was isolated from most plants and plant parts with vascular discoloration. Plants with foliar symptoms had slightly more vascular discoloration than plants without symptoms. Some vascular tissue of almost every plant part examined was discolored. All S plants with external disease symptoms had vascular discoloration. A higher level of vascular discoloration, based on a scale of 0 to 15, was present in S than in T plants.

**DISCUSSION.**—Severity of *Verticillium* wilt of cotton is a function of fungus concn and activity at the site of disease symptoms in the leaves  $\times$  the number of leaves affected, according to Garber & Houston (4). Inoculum potential of the pathogen (5), as well as resistance level of the host, are of prime importance in determining disease severity. Our experiments, however, indicated that effects of these factors, as important as they are, can be masked or obscured by temp, and agree with data obtained by Brinkerhoff et al. (2) in the greenhouse. Bell & Presley (1) concluded that the temp at which plants are grown must be known if the phenotypic expression of resistance is to be meaningful. Neither cotton in our tests, Cal 7-8 (S) with low tolerance, nor Acala 4-42-77 (T) with higher tolerance, was seriously affected by *Verticillium* wilt in 1967. The susceptible and resistant strains were clearly separated in 1966. Temperature data reveal that 1967 had higher maximum temp and lower minimum temp than did 1966 (Table 1). Losses due to *Verticillium* wilt were the highest in a decade in California in 1965 (8), and the average maximum and minimum temp that year were even lower than in 1966. The cotton strains were planted in the same soil areas each year, so the inoculum level in 1967 might be presumed to be high following severe disease years in 1965 and 1966. Moisture and nutritive factors both years were similar; however, disease incidence was markedly lower in 1967. The number of plants containing the organism decreased sharply following a period of hot days and nights, 18 July to 20 August 1966. We believe the decline in posi-

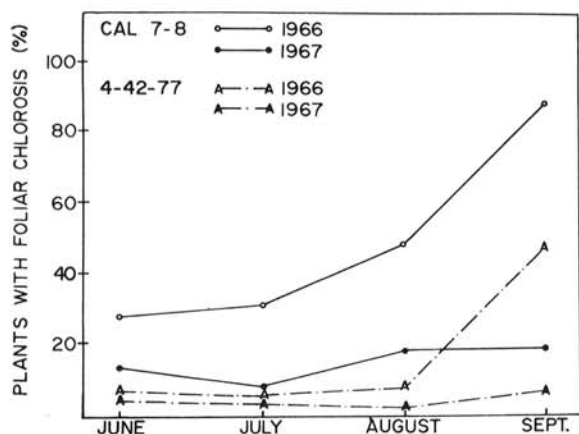


Fig. 1. Effects of a warm summer (1967) compared to a cool summer (1966) on the percentage of cotton plants with foliar chlorosis symptoms of *Verticillium* wilt.

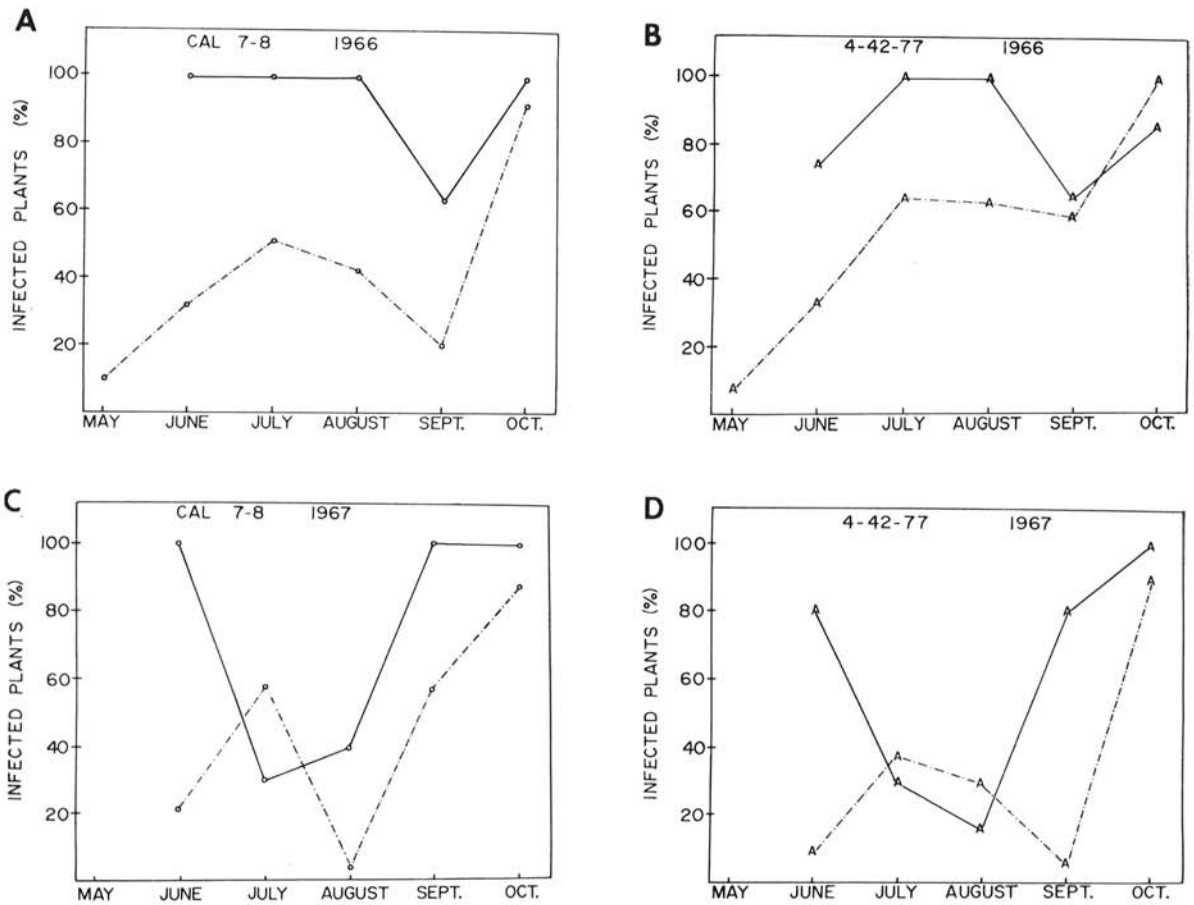


Fig. 2. Effects of a cool summer, 1966 (A, B), compared to a warm summer, 1967 (C, D) on the percentage of Cal 7-8 (susceptible) and Acala 4-42-77 (tolerant) cotton plants infected with *Verticillium albo-atrum*. Plants with disease symptoms, solid lines; symptomless plants, broken lines.

tive isolations which occurred earlier in 1967 (Fig. 2) was associated with a prolonged hot period between 24 June and late August. We question the dependability of disease evaluations based on recovery of the pathogen, as the organism was present in many plants that were symptomless throughout the season. Decrease in positive isolations from leaves in mid-season suggests that the organism is actually killed during prolonged hot periods, but may appear later in the season as a result of new infections. We assume that the reappearance of the organism after a temp decline results from re-infection of the leaves, perhaps from new root infections. Conidia produced in previously infected xylem vessels of roots, moving into the upper portions of the plants (4, 9), however, may explain this observation.

Lack of symptom expression in some infected plants may be due to the fact that some strains of the fungus are not very virulent, or that these plants may have a higher level of resistance to fungus development than plants with symptoms. Bell & Presley (1) suggested that speed of the host response following infection may help explain differences in resistance between cotton strains. Cal 7-8 (S) is heterogeneous, and variation in

disease expression between plants of this strain might be explained on a genetic basis. Acala 4-42-77 (T), on the other hand, is quite homogeneous (10); and most plants of this strain possess the same potential for disease resistance, yet some infected plants are symptomless. We suggest that plants without disease symptoms are influenced by undetermined, subtle micro-environmental factors that we have not measured in studies thus far.

#### LITERATURE CITED

- BELL, A. A., & J. T. PRESLEY. 1969. Temperature effects on resistance and phytoalexin synthesis in cotton inoculated with *Verticillium albo-atrum*. *Phytopathology* 59:1141-1146.
- BRINKERHOFF, L. A., E. SAMAYOA, & J. C. MURRAY. 1967. *Verticillium* wilt reactions in cotton as affected by post-inoculation temperatures. *Phytopathology* 57:805 (Abstr.).
- GARBER, R. H., & B. R. HOUSTON. 1966. Penetration and development of *Verticillium albo-atrum* in the cotton plant. *Phytopathology* 56:1121-1126.
- GARBER, R. H., & B. R. HOUSTON. 1967. Nature of *Verticillium* wilt resistance in cotton. *Phytopathology* 57:885-888.

5. GARRETT, S. D. 1956. Biology of root-infecting fungi. Cambridge Univ. Press, London & N. Y. 292 p.
6. HALISKY, P. M., R. H. GARBER, & W. C. SCHNATHORST. 1959. Influence of soil temperature on *Verticillium hadromycosis* of cotton in California. *Plant Dis. Repr.* 43:584-587.
7. LEYENDECKER, P. J., JR. 1950. Effects of certain cultural practices on *Verticillium* wilt of cotton in New Mexico. *New Mexico Agr. Exp. Sta. Bull.* 356. 28 p.
8. MCDANIEL, M. C. 1966. Reduction in yield of cotton caused by diseases in 1965. *Plant Dis. Repr.* 50:350.
9. PRESLEY, J. T., H. R. CARNS, E. E. TAYLOR, & W. C. SCHNATHORST. 1966. Movement of conidia of *Verticillium albo-atrum* in cotton plants. *Phytopathology* 56:375.
10. TURNER, J. H. 1963. Breeding methods in maintenance and improvement of the Acala 4-42 variety of cotton. *USDA Bull.* 34-51.