Epidemiology of Verticillium Wilt of Cotton: Effects of Disease Development on Plant Phenology and Lint Yield

G. S. Pullman and J. E. DeVay

Postgraduate research plant pathologist and professor, respectively, Department of Plant Pathology, University of California, Davis 95616.

The authors gratefully acknowledge the help of E. J. Butterfield, G. E. Friebertshauser, D. P. Matherson, J. J. Stapleton, and R. J. Wakeman in the collection of field data.

We also thank the California Planting Cotton Seed Distributors of Bakersfield, CA, for financial support.

This study was funded in part by EPA Grant L800151.

Accepted for publication 13 August 1981.

ABSTRACT

Pullman, G. S., and DeVay, J. E. 1982. Epidemiology of Verticillium wilt of cotton: Effects of disease development on plant phenology and lint yield. Phytopathology 72:554-559.

The major effect of Verticillium wilt on cotton plants was the reduction of growth and development, which resulted in reduced plant height, lateral branching, and dry matter accumulation in leaves, stems, roots, squares (flower buds), and bolls. The number of nodes was not greatly reduced in diseased plants and plant stunting was mainly due to decreased internode elongation. One of the first visible disease symptoms was a reduction in growth rate that could be detected approximately 2 wk prior to the appearance of foliar symptoms. Squares were shed before or after foliar symptoms appeared and rarely developed to flowering size on diseased plants unless high air temperatures occurred. Set bolls, however, generally remained attached to the plant and eventually opened, even when defoliation occurred. At the time foliar symptoms appeared, the dry weight of taproots and main lateral roots was greater in plants with foliar symptoms than in those that appeared to be healthy. Cotton lint yields and the number of open bolls per plant were directly related to the growth period before foliar symptoms appeared. During periods of high air temperatures, the effects of Verticillium wilt were reduced while plant growth and dry matter accumulation were resumed. Isolates of *Verticillium dahliae* from diseased cotton leaves in the field under study ranged from nondefoliating to defoliating pathotypes. Approximately 19% of the isolates tested were classified as nondefoliating, 72% as intermediate types, and 9% as defoliating.

Additional key words: *Gossypium hirsutum*, soilborne pathogen.

Verticillium wilt, caused by *Verticillium dahliae* Kleb., is the major disease of cotton (*Gossypium hirsutum* L.) grown in California. Estimates of the overall yearly loss range from 7.6% in 1976 to 2.4% in 1978 (6,10). Verticillium wilt is currently managed by the use of tolerant Acala cotton cultivars, crop rotation, and irrigation and fertilization practices. Highly virulent strains of *V. dahliae*, however, are widespread and frequently cause significant reductions of lint yields in fields planted to Acala cotton cultivars (24).

Among the main factors involved in the epidemiology of Verticillium wilt in cotton are: inoculum density; the strain or pathotype of the fungus and its corresponding virulence; cotton cultivar response to the pathogen and environmental factors (eg, high air and soil temperatures) that slow or arrest disease development; soil moisture; and planting density (4,13,21). Inoculum density is closely related to the final percentage of plant infection determined by the presence of vascular discoloration (2,7). However, final disease incidence, determined by foliar symptoms, was not related to inoculum density when many fields were compared. This discrepancy appears to be due to the variability of environmental conditions and pathogenicity among the fungal pathotypes isolated from field to field (7,11). In a 7-yr

0031-949X/82/05055406/$03.00/0

study by Pullman and DeVay (21) this variability was reduced by concentrating the experimental work within a single field. In their studies, final disease incidence based on foliar symptoms and the rate of disease appearance were related to inoculum density. Friebertshauser and DeVay (12) further demonstrated the importance of pathotypes on cotton lint yields and plant response in field-grown plants inoculated with either a defoliating or nondefoliating isolate of *V. dahliae*. The defoliating isolate had a greater effect on all plant growth parameters at all times of inoculation when compared to the nondefoliating isolate. Other factors having major effects on the development of Verticillium wilt are cross-protection (18,23) and the varying levels of tolerance to *V. dahliae* (1) among cotton cultivars.

The effect of Verticillium wilt on the phenology of cotton plants is poorly understood, although changes in plant phenology have been reported for Verticillium wilt in other hosts. Tomato seedlings root-inoculated with *V. albo-atrum* had less stem growth, reduced leaf area, and decreased dry weights of leaf, stem, and root tissues compared to uninoculated seedlings (25). Leaf area and photosynthetic efficiency also were reduced due to the failure of leaves to fully expand. Potato plants showed reduced leaf area and function following infection with *V. albo-atrum* or *V. dahliae* due to reduced leaf development, premature leaf abscission, and leaf chlorosis (16,17). Reductions in stem growth and lateral shoot development were associated with the reductions in green leaf area. Mint plants also show reduced internode elongation after infection with *V. dahliae* (20).

The objective of this study was to determine the effects of disease on plant phenology and cotton lint yields by weekly plant growth analyses. To minimize the variation in effects due to different pathotypes of *V. dahliae*, two adjacent fields were studied over several successive years. The fungal pathotypes were mainly nondefoliating. Results of this study will be used to model Verticillium wilt of cotton based on existing models for cotton plant growth and development (15) and for cotton defoliation and fruit attack caused by plant herbivores (26).

MATERIALS AND METHODS

General field description. Experimental fields were located near Five Points, CA. The soil consisted of a deep deposit of Panoche clay loam. One field (2.3 ha) had a history of Verticillium wilt in areas cropped continuously to cotton and contained approximately 55–80 propagules of *V. dahliae* per gram of soil. The second and adjacent field (0.5 ha) had a variable crop history of cotton, melons and grain, and contained 7–15 propagules per gram (p/g) of soil.

Cotton cultivar Acala SJ-2 was used thoughout this study and was planted in rows 1 m apart with approximately four plants per meter of row. Cultivation, fertilization, and irrigation practices common to the area were used each year. All crops were furrow irrigated except the 1979 crop, which was sprinkler irrigated.

Field experiments, 1976–1977. A 3.1-m section of row was marked off in each of six continuous cotton plots (high inoculum density) and also in six cotton plots following paddy rice (low inoculum density) (9). Consecutive plants in these 3.1-m sections were examined weekly for plant height and foliar symptoms of Verticillium wilt from the time of first square (flower bud) formation until 20 August 1976 and finally in mid-September, 1977. At the time of harvest, the number of open bolls present on each plant was recorded.

During 1977, three 3.1-m sections of row also were marked off in the adjacent 0.5-ha field. Height, foliar symptoms of Verticillium wilt, and number of open bolls at harvest time were monitored as indicated for the larger field.

Field experiments, 1978–1979. To study a possible relationship between symptom development and plant growth, 80 m of row containing approximately 350 plants, was monitored weekly within the 2.3-ha field in each of six continuous cotton plots from the time of first square formation until mid-September. Individual plants were tagged weekly to identify when foliar symptoms first appeared. From the time of emergence until late September, five healthy-appearing plants (without foliar symptoms) were pulled by hand at 1- to 3-wk intervals from each of the six continuous cotton plots. Five plants tagged for early-, middle-, and late-season appearance of foliar symptoms were also sampled at 1- to 3-wk intervals from late June through September. Plant height, number of main stem nodes, squares, bolls, and the presence or absence of vascular discoloration were also recorded. When counting the number of nodes, the cotyledonal node was counted as "0". Squares smaller than 3–4 mm were not counted. To determine dry matter accumulation, plants were divided into squares, bolls, leaves (with petiole attached), stems, and roots. All components were dried in forced-air ovens at approximately 50 C and then weighed. At harvest 10 plants for each time of foliar symptom appearance and 10 plants without foliar symptoms in mid-September were randomly collected from each plot. The number of open bolls were counted and averaged and the seed cotton was hand picked from each group of plants.

Field experiments, 1980. Plants in 61 m of row were monitored and tagged weekly in a manner similar to previous years within each of three cotton plots in the 0.5-ha field. At the time of harvest, five plants for each time of foliar symptom appearance and five plants without foliar symptoms in mid-September were collected for each replication. The seed cotton was handpicked and open bolls were counted for each plant.

Physiological time. Maximum and minimum air temperatures were obtained from a weather station on the University of California West Side Field Station, located approximately 0.5 km from the plots. Physiological time was determined as an accumulation of temperatures above 53.5 F (11.9 C) and expressed in units of Fahrenheit degree days (D°). Degree days were calculated by using a computer program that integrated the area under sine curves passing through the daily maximum and minimum air temperatures and above the developmental threshold for cotton (15). Degree days more adequately reflect the growth of cotton plants than Julian days and have been used in predicting developmental times for cotton (15). Physiological time calculated in Fahrenheit degree days can be converted to centigrade degree days by multiplying by 0.555.

RESULTS

Effects of disease on plant height and main stem node production. Figure 1A and B and 1C and D shows the effects of Verticillium wilt on plant height and main stem node production during 1978 and 1979. During both growing seasons, reduced plant growth, epinasty, and a slight change in leaf color were the first noticeable symptoms of Verticillium wilt. These symptoms occurred before the development of interveinal chlorosis and leaf necrosis. Graphs of plant height versus physiological time (Fig. 1A and B) for both healthy and diseased plants show that plants stopped increasing height approximately 2 wk (250 degree days) prior to the development of foliar symptoms. Main stem node number, which is a linear function of physiological time (15), however, was not greatly altered indicating that changes in height are caused by altered rates of internode elongation. Plant height reduction was similar during 1976–1980; the earlier foliar symptoms appeared, the greater was the reduction in plant height.

Effect of disease on square formation and boll set. Figure 1E and F and 1G and H shows the effects of Verticillium wilt on the number of squares and bolls formed during 1978 and 1979. Square production was greatly altered by the development of Verticillium wilt; the main effects were the shedding of squares already formed and the inhibition of further square production. By the time foliar symptoms appeared, most squares had already been shed. Square formation did not resume its normal rate unless air temperatures rose to levels that inhibited Verticillium wilt, such as those that occurred during August, 1979.

Once set, bolls usually remained attached to the plant, continued to accumulate dry matter, and eventually opened, although plants diseased early in the season usually bore small bolls with less cotton. Severely defoliated plants were frequently observed with full loads of green bolls attached.

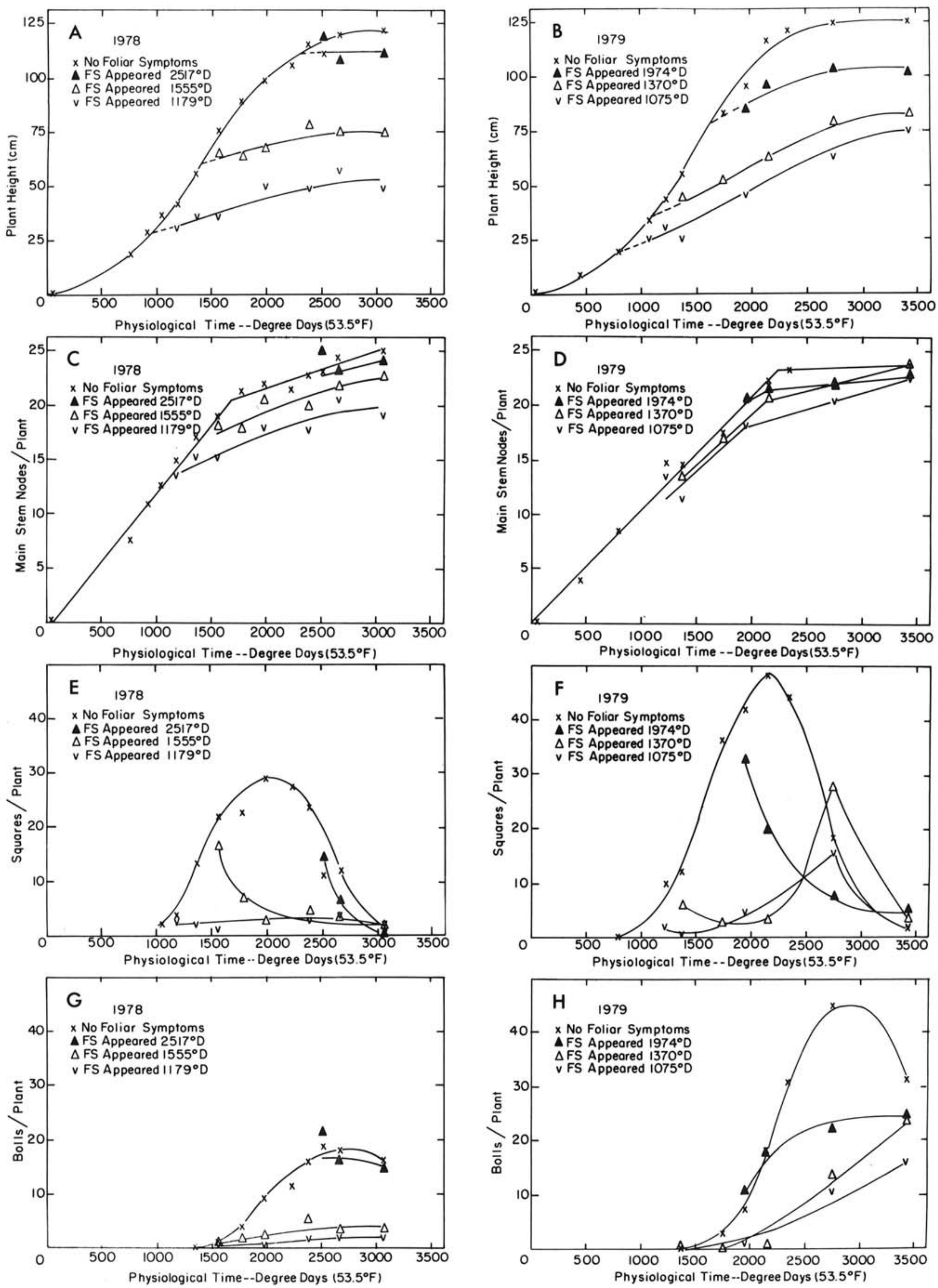

Fig. 1. The effect of physiological time of foliar symptom appearance on the plant growth of cotton plants affected by Verticillium wilt during 1978 and 1979. The first data point for each curve indicates the time when foliar symptoms (FS) first appeared. **A and B,** Effects on plant height; **C and D,** effects on plant nodes; **E and F,** effects on the number of squares per plant; and **G and H,** effects on the number of bolls per plant.

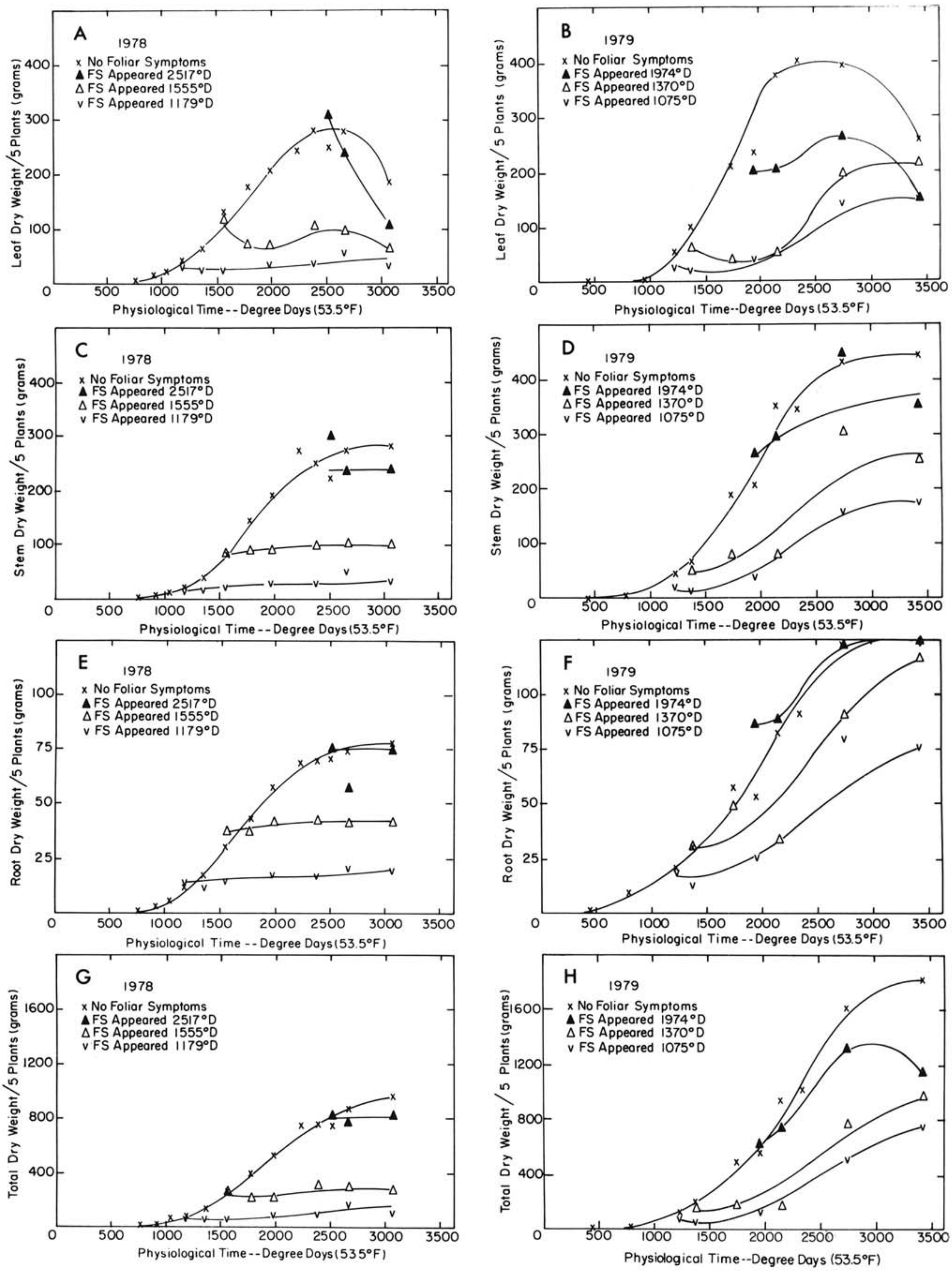

Fig. 2. The effect of physiological time of foliar symptom appearance on the dry weight accumulation of cotton plants affected by Verticillium wilt during 1978 and 1979. The first data point for each curve indicates the time when foliar symptoms (FS) first appeared. **A and B,** Effects on leaf dry weight; **C and D,** effects on the stem dry weight; **E and F,** effects on root dry weight of hand-pulled plants; and **G and H,** effects on total plant dry weight (leaf, stem, root, squares [flower buds], and bolls).

Effects of disease on leaf, stem, root, and total dry matter accumulation. With the appearance of foliar symptoms, at least three changes took place that altered the accumulation of leaf dry matter (Fig. 2A and B) and indirectly altered the production of photosynthates: areas within the leaf wilted, became chlorotic, and eventually necrotic; leaves often abscised, resulting in partial-to-complete defoliation; and an associated reduction in branching resulted in fewer apical growing points and fewer leaves. Leaf lamina (minus petioles) comprised approximately 83% of the leaf dry weight.

Stem production also was reduced in diseased plants (Fig. 2C and D). Both a decrease in internode elongation and a decrease in the development of lateral and fruiting branches contributed to the loss of stem dry weight in diseased plants. Inhibition of branching resulted in the reduction of both leaf and fruit production.

Differences in the dry weight of taproots from healthy and diseased plants are shown in Fig. 2E and F. Plants collected within 7 days of the first appearance of foliar symptoms contained more dry matter in their taproots than did healthy plants. After the appearance of foliar symptoms, root dry matter accumulation either stopped (1978, Fig. 2E) or was less (1979, Fig. 2F) in diseased plants.

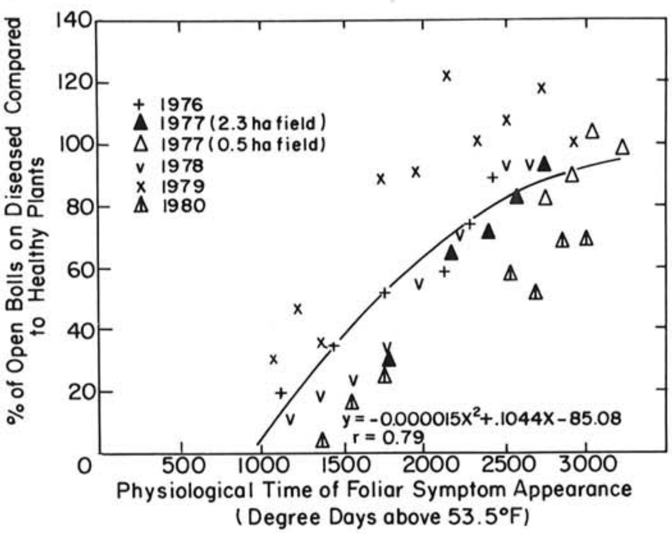

Fig. 3. The effect of physiological time of foliar symptom appearance on open bolls at the time of harvest. For comparison, open bolls from diseased plants are plotted as percentages of those on plants without foliar symptoms at the last disease rating in mid-September.

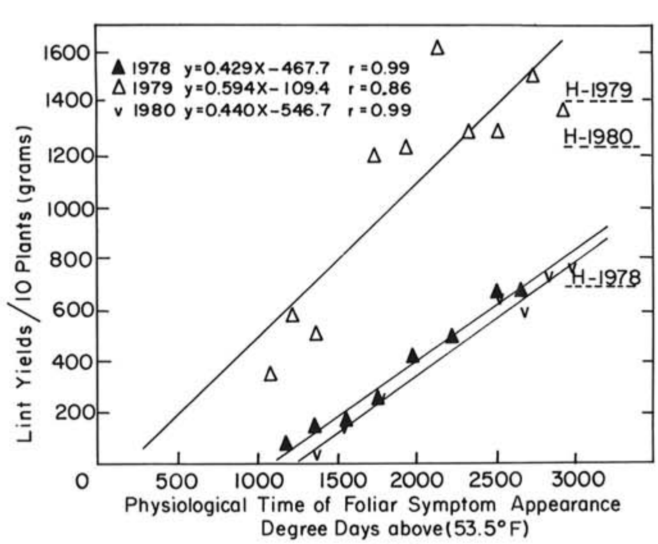

Fig. 4. The effect of physiological time of foliar symptom appearance on cotton lint yields. H = cotton lint yields from plants that appeared healthy in mid-September.

Total dry matter accumulation, based on the sum of leaf, stem, root, square, and boll dry weights, was greatly reduced in diseased plants when foliar symptoms appeared (Fig. 2G and H). However, during 1979, the occurrence of high day temperatures (⩾38 C) caused regrowth and increased dry weights of diseased plants.

Effect of disease on cotton lint yields. Figure 3 shows the general relationship between open bolls and time of foliar symptom appearance from 1976 to 1980. The number of open bolls for each time of disease appearance was plotted as a percentage of open bolls found at harvest on plants that appeared healthy at the last disease rating in mid-September. In 1977, all plants were diseased by mid-September; therefore, an average number of open bolls for healthy plants in other years was used as the basis for calculating percentages. The average number of open bolls increased as foliar symptoms developed later in the growing season; however, after approximately 2,500 degree days, foliar symptom development did not greatly affect the number of open bolls at harvest.

Figure 4 shows the relationships between cotton lint yields and physiological time of foliar symptom appearance for 1978 to 1980. As foliar symptoms developed later in the season, yields increased arithmetically with physiological time until approximately 2,500 degree days.

Characterization of plant isolates of *V. dahliae*. Ninety-six leaf isolates of *V. dahliae* were tested for pathogenicity in two cotton cultivars, Acala SJ-2 and 70-110 (21). The mixture of naturally occurring pathotypes present in the experimental field site ranged from the nondefoliating to the defoliating pathotypes. Approximately 9% of the isolates tested were classified as defoliating, 67–77% as intermediate types, and the remainder as nondefoliating types (21).

DISCUSSION

The major effect of Verticillium wilt of cotton was the inhibition of plant growth and development. As a result, total dry matter accumulation, boll set and development, internode elongation, and lateral and fruiting branch development were reduced or stopped. Young squares were often shed and total leaf area was reduced, often through the premature loss of leaves. Most of these changes began approximately 2 wk prior to the appearance of foliar symptoms, probably due to water stress (14).

The time lag between foliar symptom appearance and a reduction in growth rate also was reported by Butterfield and DeVay (8) to be 2 wk in cotton plants naturally infected with *V. dahliae*. Selman and Pegg (25) noted similar reductions in height increase and dry matter production 2 wk before the appearance of foliar symptoms in tomato seedlings, which were root inoculated with *V. albo-atrum*. In addition, during 1978, when cool weather occurred following a period of high air temperatures that had completely suppressed new foliar symptom development, 2 wk passed before new foliar symptoms began to appear.

High air temperatures (⩾28 C) not only restrict in vivo growth of *V. dahliae*, but also decrease the susceptibility of cotton to wilt (5,13,27). Studies have shown that germination and growth of *V. dahliae* are inhibited at temperatures above 30–33 C (4,27), and that death of fungal structures may occur at temperatures above 36 C if these are maintained for long periods (22). Recovery of diseased cotton plants or "heat therapy" has been reported during periods of high air temperatures (5). During both years of the present study, periods of high air temperatures occurred, which inhibited the development of foliar symptoms of Verticillium wilt (21).

As shown by plant growth analysis in this study, cotton lint yields and the number of open bolls per plant were directly related to the time of foliar symptom appearance. Bassett (3) also found that ratings of Verticillium wilt, based on foliar symptoms in August, accounted for the yield variations among the 28–35 upland cotton cultivars that were tested. Past attempts, however, to relate cotton lint yields to inoculum density or percentage plants with vascular discoloration have been unsuccessful (1,3). Relationships have been found between lint yields and end of season defoliation due to Verticillium wilt (1) and also between pathotypes of *V. dahliae* and lint yields (12). Because field-to-field variations in pathotypes of *V. dahliae* and environmental conditions apparently contribute to low correlations between cotton lint yields and inoculum density or vascular discoloration (1,3,7), the present study was made in adjoining fields characterized by similar inoculum densities and pathotypes of *V. dahliae*.

Cotton lint reductions due to Verticillium wilt were small when foliar symptoms appeared after mid-August (approximately 2,500 degree days). Fruit-load stress that develops when photosynthate demand exceeds supply often occurs at this time due to the simultaneous growth of many squares and bolls. As a result, new growth of leaf, stem, and root tissues stops and young squares and bolls less than 10 days of age are often shed (15). Plants with foliar symptoms of Verticillium wilt apparently behave similarly to those undergoing fruit-load stress, probably due to the occlusion of vascular tissue (19) and the resulting water stress (14). Therefore, when foliar symptoms develop in plants undergoing fruit stress (approximately 2,500 degree days), growth has already slowed and the effects of Verticillium wilt are often less severe.

The main results of this study show that plant growth and cotton lint yields are related to the period of cotton growth before foliar symptoms of Verticillium wilt appear and that growth reductions can be first observed about 2 wk prior to the appearance of foliar symptoms.

LITERATURE CITED

1. Ashworth, L. J., Jr., Huisman, O. C., Harper, D. N., Stromberg, L. K., and Bassett, D. M. 1979. Verticillium wilt disease of cotton: Influence of inoculum density in the field. Phytopathology 69:483-489.
2. Ashworth, L. J., Jr., McCutcheon, O. D., and George, A. G. 1972. *Verticillium albo-atrum*: The quantitative relationship between inoculum density and infection of cotton. Phytopathology 62:901-903.
3. Bassett, D. M. 1974. Resistance of cotton cultivars to Verticillium wilt and its relationship to yield. Crop Sci. 14:864-867.
4. Bell, A. A. 1973. Nature of disease resistance. In: Verticillium Wilt of Cotton. U.S. Dep. Agric., Agricultural Research, Southern Region 14:47-62.
5. Bell, A. A., and Presley, J. T. 1969. Heat-inhibited or heat-killed conidia of *Verticillium albo-atrum* induce disease resistance and phytoalexin synthesis in cotton. Phytopathology 59:1147-1151.
6. Berry, R. W. 1979. Cotton disease loss estimate committee report—1978. Page 2 in: Proc. Beltwide Cotton Production Research Conferences, 7–11 January 1979. Phoenix, AZ. National Cotton Council, Memphis, TN. 314 pp.
7. Butterfield, E. J. 1975. Effects of cultural practices on the ecology of *Verticillium dahliae* and the epidemiology of Verticillium wilt of cotton. Ph.D. dissertation. University of California, Davis. 71 pp.
8. Butterfield, E. J., and DeVay, J. E. 1977. The effect of Verticillium wilt on the growth and development of cotton. (Abstr.) Proc. Am. Phytopathol. Soc. 4:173.
9. Butterfield, E. J., DeVay, J. E., and Garber, R. H. 1978. The influence of several crop sequences on the incidence of Verticillium wilt of cotton and on the populations of *Verticillium dahliae* in field soil. Phytopathology 68:1217-1220.
10. Crawford, J. L. 1977. Cotton disease loss estimate committee report—1976. Page 3 in: Proc. Beltwide Cotton Production Research Conferences, 10–12 January 1976. Atlanta, GA. National Cotton Council, Memphis, TN. 258 pp.
11. DeVay, J. E., Forrester, L. L., Garber, R. H., and Butterfield, E. J. 1974. Characteristics and concentration of propagules of *Verticillium dahliae* in air-dried field soils in relation to the prevalence of Verticillium wilt in cotton. Phytopathology 64:22-29.
12. Friebertshauser, G. E., and DeVay, J. E. 1982. Differential effects of the defoliating and nondefoliating pathotypes of *Verticillium dahliae* upon the growth and development of *Gossypium hirsutum*. Phytopathology 72:(In press).
13. Garber, R. H., and Presley, J. T. 1971. Relation of air temperature to development of Verticillium wilt on cotton in the field. Phytopathology 61:204-207.
14. Grimes, D. W., and Yamada, H. 1982. Relations of cotton growth and yield to minimum leaf water potential. Crop Sci. 22:(In press).
15. Gutierrez, A. P., Falcon, L. A., Leow, W., Leipzig, P. A., and Van Den Bosch, R. 1975. An analysis of cotton production in California: A model for Acala cotton and the effects of defoliators on its yields. Environ. Entomol. 4:125-136.
16. Harrison, J. A. C., and Isaac, I. 1968. Leaf-area development in King Edward potato plants inoculated with *Verticillium albo-atrum* and *V. dahliae*. Ann. Appl. Biol. 61:217-230.
17. Harrison, J. A. C., and Isaac, I. 1969. Effect of main stem number and lateral stem development in potato plants infected with *Verticillium albo-atrum* and *V. dahliae*. Ann. Appl. Biol. 63:379-387.
18. Melouk, H. A., and Horner, C. E. 1975. Cross protection in mints by *Verticillium nigrescens* against *Verticillium dahliae*. Phytopathology 65:767-769.
19. Misaghi, I. J., DeVay, J. E., and Duniway, J. M. 1978. Relationship between occlusion of xylem elements and disease symptoms in leaves of cotton plants infected with *Verticillium dahliae*. Can. J. Bot. 56:339-342.
20. Nelson, R. 1950. Verticillium wilt of peppermint. East Lansing, Mich. Agric. Exp. Stn. Tech. Bull. 221. 259 pp.
21. Pullman, G. S., and DeVay, J. E. 1982. Epidemiology of Verticillium wilt of cotton: A relationship between propagule density and disease progression. Phytopathology 72:549-554.
22. Pullman, G. S., DeVay, J. E., and Garber, R. H. 1981. Thermal death: A logarithmic relationship between time and temperature for four soilborne plant pathogens. Phytopathology 71:959-964.
23. Schnathorst, W. C., and Mathre, D. E. 1966. Cross-protection in cotton with strains of *Verticillium albo-atrum*. Phytopathology 56:1204-1209.
24. Schnathorst, W. C., and Mathre, D. E. 1966. Host range and differentiation of a severe form of *Verticillium albo-atrum* in cotton. Phytopathology 56:1155-1161.
25. Selman, I. W., and Pegg, G. F. 1957. An analysis of the growth response of young tomato plants to infection by *Verticillium albo-atrum*. Ann. Appl. Biol. 45:674-681.
26. Wang, Y., Gutierrez, A. P., Oster, G., and Daxl, R. 1977. A population model for plant growth and development: Coupling cotton-herbivore interaction. Can. Entomol. 109:1359-1374.
27. Wyllie, T. D., and DeVay, J. E. 1970. Growth characteristics of several isolates of *Verticillium albo-atrum* and *Verticillium nigrescens* from cotton. Phytopathology 60:907-910.