Selection of Pathogenic Strains of *Verticillium dahliae* and Their Influence on the Useful Life of Cotton Cultivars in the Field

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ABSTRACT

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Three cotton cultivars were grown in the same rows at the University of California West Side Field Station for five successive years, 1976–1980. The cultivars were 70-110, an Acala-type cotton used locally as a susceptible comparison in breeding trials; Acala SJ-2, a cultivar with moderate tolerance to Verticillium wilt; and Acala SJ-5, which is more tolerant of Verticillium wilt than either 70-110 or Acala SJ-2. In the beginning (1976) all cultivars were exposed to identical inoculum densities (ID), 2, 4, 15, and 21 microsclerotia per gram of soil. The rate of buildup of new inoculum at each original ID was greatest in 70-110 blocks, followed by Acala SJ-2 blocks, and it was least in Acala SJ-5 blocks. Within-cultivar differences in ID of *Verticillium dahliae* were not distinguishable after 2 yr, but differences occurred between blocks of 70-110 and Acala SJ-2 versus blocks of Acala SJ-5 each year. Sixty to 90% of plants of the most susceptible cultivar, 70-110, were defoliated each year by the time of harvest (a severe expression of Verticillium wilt), but the percentage of defoliated plants of the more tolerant cultivars increased in successive years. This observation suggested that differentially tolerant cultivars induced increases of soilborne inoculum of more aggressive strains of *V. dahliae* when planted year after year in the same infested soil. The data provide a quantitative basis for understanding the demise of cotton cultivars grown in fields infested with *V. dahliae*.

Additional key words: cotton genotype effects on inoculum of *V. dahliae*.

Cotton, *Gossypium hirsutum* L., has by law (1) been grown under essentially monoculture conditions in the San Joaquin Valley (SJV) of California since 1925 and in large areas of Russia for 60 or more years (15). The negative influence of monoculture (11), according to Kasyanenko et al (15) resulted in six changes of cotton cultivars in Russia due to Verticillium wilt during 1921–1970. In both California (17) and Russia (15) researchers suggested that the demise of Verticillium wilt-tolerant cultivars was caused by the buildup in soil of indigenous highly aggressive strains of *Verticillium dahliae* Kleb. in response to host selection pressure. Quantitative data on this subject, however, are lacking, although Ashworth (4) recently reported that isolates of *V. dahliae* from Verticillium wilt-tolerant cultivars generally were more aggressive than isolates from less tolerant cultivars. Furthermore, isolates of the fungus from plants defoliated by Verticillium wilt (a severe expression of the disease [8,16]) were more aggressive than isolates from randomly selected infected plants.

The purpose of this paper is to report results of experiments made to determine the influence of differentially tolerant cotton cultivars grown in monoculture on development of new inoculum of *V. dahliae* and on the aggressiveness of new inoculum under field conditions. The findings are related to observations made on tenure and yield of cotton cultivars in the SJV.

MATERIALS AND METHODS

The field plots used for this test were described earlier for a study of the influence of inoculum density (ID) of *V. dahliae* on disease severity and cotton yield (6), thus methods will be briefly described here. A block of land (~1.6 ha) was fumigated with methyl bromide-chloropicrin. Then 32-row blocks, ~12 m long, were differentially infested with *V. dahliae* by distributing over the surface, then mixing into the soil four different amounts of dry bits of infested tomato plant residue. There were five replications of four IDs determined (13) in midsummer of 1975 to be 0.5, 1.0, 7.7, and 22.5 microsclerotia (MS) per gram of soil (6).

Only cultivar Acala SJ-2 was grown in the blocks described above in 1975; thereafter, through 1980, the performance of four cultivars was compared in eight-row blocks. The cultivars used were: 70-110, an Acala cultivar used locally as a Verticillium wilt-susceptible comparison in breeding trials; Acala SJ-2, moderately tolerant to Verticillium wilt; and Acala SJ-4 and Acala SJ-5, both more highly tolerant to Verticillium wilt. Each cultivar succeeded itself in blocks each year, with one exception. Cultivar 70-110 was deleted during 1 yr (1977) and cultivar Acala SJ-4 was planted in its place. Besides annual ID determinations made during midsummer (13), the percentage of plants defoliated by wilt disease was determined on the two center rows of each plot, each year near the time of harvest. Data for cultivar Acala SJ-4, which is closely related to Acala SJ-5, were excluded to simplify this report. Yield data for experiments described here were reported earlier (6). Yield data for the SJV are from official records (2) and planting seed data were supplied by California Planting Cotton Seed Distributors (T. Cherry, *personal communication*).

RESULTS

The influence of differentially tolerant cotton cultivars on development of new inoculum of *Verticillium dahliae* under field conditions. Only Acala SJ-2 was grown in differentially infested plots in 1975. ID increased from 0.5, 1.0, 7.7, and 22.5 MS per gram of soil to, respectively, 2, 4, 15, and 21 MS per gram of soil in 1976. In that year, cultivars 70-110, Acala SJ-2, and Acala SJ-5 were grown for the first time. Differential increases in ID, both within and between cultivars, occurred in 1977 following the first year the three cultivars were grown. More inoculum was produced in residues of cultivar 70-110 plants grown in 1976 than in residues of the other cultivars. And, plant residues of Acala SJ-2 produced more inoculum than did plant residues of Acala SJ-5 (Table 1).

Following 1977, intracultivar differences in ID were not detected regardless of whether original IDs were relatively low or high

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(Table 1). However, the ID of cultivar 70-110 blocks was always greater than the ID of cultivar SJ-2 blocks, and both were greater than those observed in blocks of cultivar Acala SJ-5.

The influence of host selection pressure on the development of inoculum with increased aggressiveness. Most of the plants (63–92%) of the least tolerant cultivar, 70-110, were defoliated in response to Verticillium wilt each year it was grown during 1976–1980 (Fig. 1). On the other hand, except for 1977, each year there was an increase in the percentage of plants of cultivar Acala SJ-2 defoliated by Verticillium wilt, but the amount of defoliation for cultivar Acala SJ-2 was always less than that for cultivar 70-110. The most tolerant cultivar, Acala SJ-5, also suffered greater amounts of defoliation each year, but annual increases for it were always less than for cultivar Acala SJ-2 (Fig. 1).

Relationship of Verticillium wilt with yield and tenure of cotton cultivars in the SJV in California. A Verticillium wilt-tolerant cultivar of cotton, Acala 4-42, was first grown throughout the SJV in 1954 as a result of a breeding program begun in 1934 (19). It was a composite cultivar, at first made up of seed from eight closely related single-plant selections, but thereafter its makeup varied from year to year (Fig. 2A). Cotton yields rose steadily in the SJV following the release of cultivar Acala 4-42 and were about 150% greater during 1958–1959 than during 1949–1953 (Fig. 2A); but the average yield for the SJV in 1966, the last year cultivar Acala 4-42 was grown, was 12% less than in the previous 2-yr period (Fig. 2A). In 1967, cultivar SJ-1 succeeded cultivar Acala 4-42, which had been grown for 11 yr. Depressed yields continued during the 7-yr tenure of cultivar Acala SJ-1 (Fig. 2A) but they were not uniformly depressed in the six counties of the SJV. Cotton yields in Kings County were reduced by only 4%, while those in Fresno, Kern, and Tulare counties were reduced 13–20% below the peak yield years of 1958–1965 (Fig. 2B).

Four cotton cultivars, Acala SJ-2, SJ-3, SJ-4, and SJ-5 were released during 1974–1979. Two of these, Acala SJ-3 (not shown Fig. 2A) and Acala SJ-4, were grown for, respectively, 1 and 2 yr. Thus, they had little impact on yield increases observed during 1974–1981 (Fig. 2A). After 8 yr, cultivar Acala SJ-2 still was used on 80–90% of lands planted with cotton, while a more Verticillium wilt-tolerant cultivar, Acala SJ-5, was used on about 20% of lands planted with cotton since its release in 1979. The average cotton lint yields of the SJV returned to pre-1966 levels during the ongoing tenure of SJ-2 and SJ-5 (Fig. 2A).

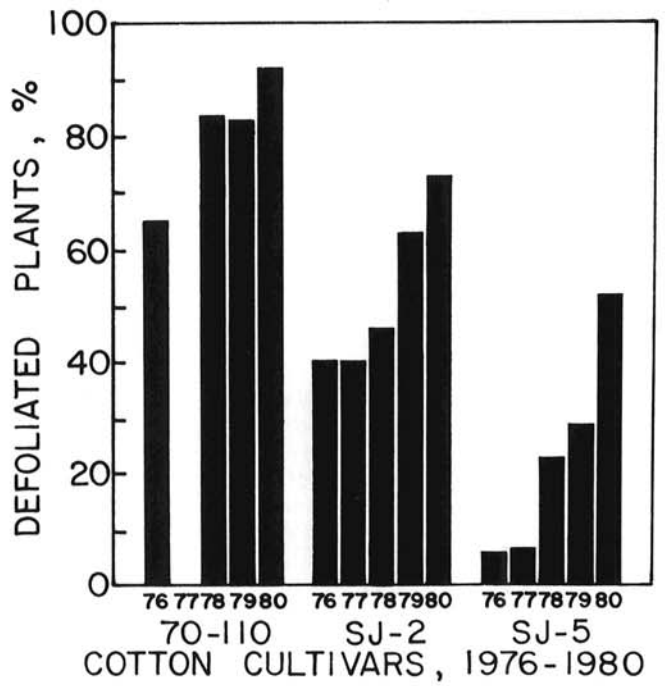

Fig. 1. Percentage of plants of three cotton cultivars defoliated by Verticillium wilt during the five successive years they were grown in the same rows.

TABLE 1. The influence of differentially tolerant cotton cultivars on development of new inoculum (microsclerotia [MS]) of *Verticillium dahliae* under field conditions

Cotton cultivars	Inoculum density (MS per gram of soil)					
	1975	1976	1977	1978	1979	1980
70-110	···	2	42	15	54	32
	···	4	83	25	47	31
	···	15	92	12	72	36
	···	21	100	27	54	35
LSD, $P = 0.05$	···	7	52	NS[a]	NS	NS
Cultivar mean	···	···	79	20	57	34
Acala SJ-2	0.5	2	23	17	25	27
	1.0	4	35	14	32	28
	7.7	15	30	7	43	26
	22.5	21	69	19	26	28
LSD, $P = 0.05$	6.7	7	35	NS	NS	NS
Cultivar mean	···	···	39	14	32	28
Acala SJ-5	···	2	11	9	17	22
	···	4	17	7	27	16
	···	15	31	2	22	14
	···	21	43	13	25	17
LSD, $P = 0.05$	···	7	18	NS	NS	NS
Cultivar mean	···	···	26	8	23	17
Mean for cultivars, LSD, $P = 0.05$		···	16	4	5	7

[a] Differences not significant $P = 0.05$.

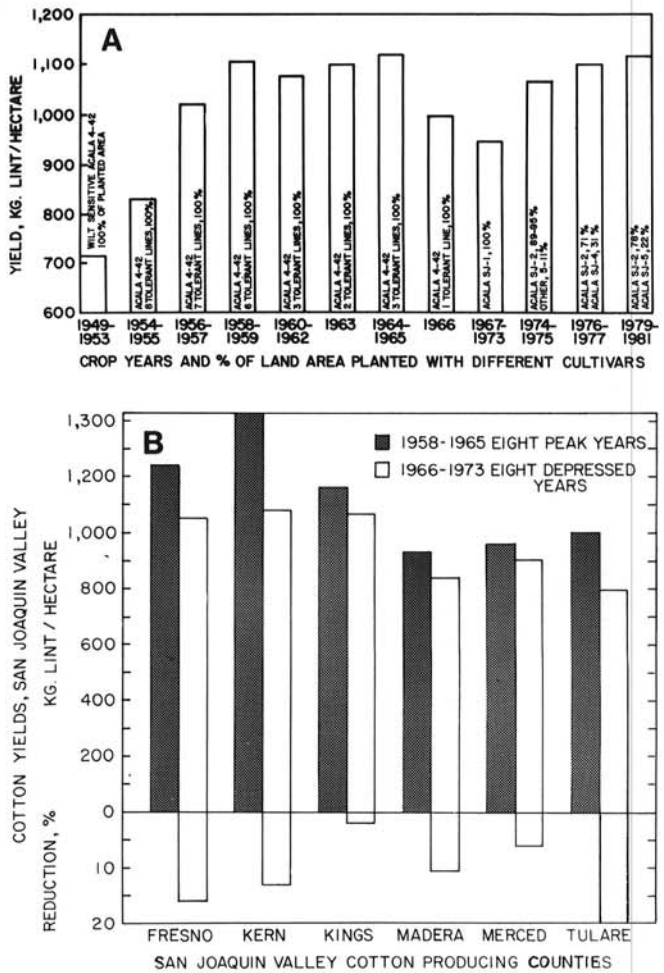

Fig. 2. A, The average yield of cotton in the San Joaquin Valley of California (SJV) during 1949–1981 and the percentage of the area planted with various cultivars. **B,** The average yield of cotton in the counties of the SJV during eight peak and eight depressed years and the percentage reduction in yields during depressed years.

DISCUSSION

Microsclerotia of *V. dahliae* were produced only in moribund tissues of cotton plants in studies made by Evans et al (9), although vegetative hyphae persisted in dry tissues for at least 21 mo and regrew to produce MS under suitable conditions (18). We observed greater production of new MS in debris of a cotton cultivar having low tolerance to Verticillium wilt (70-110) than in debris of the more tolerant cultivars (Acala SJ-2 and Acala SJ-5) in the field over a 5-yr period (Table 1). Reductions in ID during continuous cropping of cotton, as occurred in 1978 (Table 1) appears paradoxical. Nevertheless, such reductions, while not explained, commonly occur in the field (14).

The production of inoculum in moribund tissues of differentially tolerant cotton cultivars in the field agreed with observations made on vegetative development of the fungus in live cotton tissues. Garber and Houston observed less hyphal and conidial development within tissue sections of a tolerant cultivar than in those of a nontolerant cultivar (10), as did Ashworth who determined ID of conidia in lamina and petiole tissues of tolerant and nontolerant cultivars (4). Thus, the tolerance of live cotton tissues appeared to extend to moribund tissues.

Defoliation of cotton by Verticillium wilt is a severe expression of the disease (8,16), often leading to premature death (9). While defoliation in the field is, in part, related to ID of *V. dahliae* in the soil (6), in greenhouse tests (4) some isolates of the fungus always induced defoliation, regardless of field tolerance of cultivars. Ashworth found that isolates of *V. dahliae* from field-grown plants of tolerant cultivars generally were more aggressive than isolates from less tolerant cultivars, suggesting that tolerant cultivars differentially select for aggressiveness of the pathogen (4). Results of his experiments indicated presence in soil of a continuum of strains of *V. dahliae* which were expressed under host selection pressure, as suggested by others to explain the demise of once-tolerant cotton cultivars in California (17) and in Russia (15). Results of the field experiments reported here support the concept of a continuum of strains of *V. dahliae* for cotton as suggested for tomato by Grogan et al (12) and in results of other tests with cotton made by Ashworth (4), since we observed increases in defoliation among differentially tolerant cultivars grown in monoculture during 5 yr (Fig. 1).

Interpretation of yield data and tenure of cotton cultivars in the SJV were possible, based on results of experiments described above. The first Verticillium wilt-tolerant cultivar, Acala 4-42, persisted for 11 yr (Fig. 2A), although significant numbers of failures were observed 2 yr earlier (3). A 7-yr period of depressed yields continued during the tenure of its successor, Acala SJ-1 (Fig. 2A). While it had a higher yield potential in absence of Verticillium wilt than cultivar Acala 4-42 (19), it did not prove to have greater tolerance to Verticillium wilt. An increase in tolerance to Verticillium wilt was essential to improving cotton production in the SJV, where the average ID of *V. dahliae*, measured in 1971 and 1976, was 5–7 MS per gram of soil (5) but where 100 MS per gram of soil may now be found (Table 1).

Cotton yields in the SJV improved following release of cultivar Acala SJ-2 in 1974 and persisted through 1981, following release in 1979 of an even more tolerant cultivar, Acala SJ-5 (Fig. 2A). Cultivar Acala SJ-5 is considerably more tolerant than cultivar Acala SJ-2 to Verticillium wilt, outyielding cultivar Acala SJ-2 in 1982 by 19% during a cool growing season, at a *V. dahliae* ID of 19 MS per gram of soil (*unpublished*). On the other hand, Acala SJ-2 commonly outyields cultivar Acala SJ-5 under unfavorable conditions for Verticillium wilt development, regardless of ID of *V. dahliae*, as in 1981, when it outyielded cultivar Acala SJ-5 by 12% at an ID of 45 MS per gram of soil (*unpublished*). Results of other experiments during several years (5,6) showed that yield of cultivar Acala SJ-2 was essentially unaffected by Verticillium wilt, regardless of environmental conditions, at the average ID of *V. dahliae* observed in the SJV (5–7 MS per gram of soil) as determined in 1971 and 1976 (5). This was true even though essentially all plants were infected before the end of the growing season wherever ID was 3–4 MS per gram of soil (7). These observations appear to explain why cultivar Acala SJ-2 was favored over cultivar Acala SJ-5 during 1979–1981 (Fig. 2A).

Observations reported here also suggest a program for controlling Verticillium wilt of cotton in lieu of seeking even more tolerant cultivars which may be expected to preferentially select aggressiveness of strains of *V. dahliae* in soil (4) (Fig. 1). Results of surveys made in 1971 and 1976 showed that 75% of SJV soils had <6 MS *V. dahliae* per gram of soil (5) and were safe for a moderately Verticillium wilt-tolerant cultivar (eg, Acala SJ-2) and that short-term rotations with crops that did not contribute new inoculum of *V. dahliae* would result in maintenance of a low level of inoculum (5). Short-term rotations, however, were not successful where high levels on inoculum existed when rotations were begun (14). This is the situation of about 25% of soils of the SJV, where only a highly tolerant cultivar such as Acala SJ-5 will perform well under severe conditions. But, continued use of cultivar Acala SJ-5 would be expected to lead to its demise as occurred for the other once-tolerant cultivars grown in monoculture in California (17) and Russia (15).

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