Cultural Control of ... a three-

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Photo left, shows four-times magnification of ½ gm of microsclerotia of Verticillium albo-atrum, recovered from 50 diseased cotton plants killed in the field by natural infection. Many millions of microsclerotia are shown; one alone is able to infect a plant.

COTTON YIELDS in certain areas of the San Joaquin Valley have been decreasing for several years. An estimated 76% of the Tulare County cotton acreage (involving 91% of the growers) has been affected, and between 1962 and 1965 the county lost 1,200 cotton growers—in part at least, because of the low yields. Evidence points to *Verticillium* wilt as the major cause of crop losses, which, in turn, coincide with intensification of cultivation in the affected areas. Only on alkaline and very sandy soils has the disease not been a serious problem.

Control of soil-borne fungus diseases of plants often revolves around three basic principles of plant pathology: the use of disease-free seed; planting on land relatively free of the pathogen; and crop sanitation. A continuing program for reducing losses (based on these three principles) is suggested in this article. Beneficial results may not come immediately, especially in areas of heavily infested soil, but the recommendations are sound and should bring relief ultimately. More research is needed to determine how best to take practical advantage of these principles.

Our research shows that the microsclerotia (see photo, right) of Verticillium albo-atrum provide the principal inoculum for infection of the cotton plant. The microsclerotia are produced in the dead tissues of the diseased cotton plant: the roots, the pith and cortex of the stem, the petioles and blades of the leaves, and the branches to the very top of the wilted plant. When the dead tissues are broken, some of the microsclerotia are freed, and these contaminate the seed and equipment or fall to the ground where they survive until the next planting season. Woody crop refuse contains large numbers of the microsclerotia which may live for several years in the soil. Information on such contamination is needed before a control program can be developed.

Infested seed is one way the fungus can be introduced into a field of cotton. As many as 100 live microsclerotia have been found on a single seed that had been contaminated at harvest time. One microsclerotium is enough to initiate infection of a plant, and high populations of the fungus increase the severity of the disease. The cotton plant is susceptible to *Verticillium* wilt at any stage of growth from seedling to mature plant.

Tests made of infested seed after treatment with Panogen and Ceresan showed the microsclerotia were killed. Thus the fungicidal treatment designed to protect the seedling against damping-off diseases also rids the seed of *Verticillium* inoculum. Acid delinting also destroys the microsclerotia. Cultures made from seed parts failed to show the presence of the fungus internally. Thus there is no danger in the use of seed from contaminated fields so long as it is acid-delinted, or fungicide-treated.

In fields already infested, the inoculum level must be kept low. Repeated cultivation of cotton increases the number of

Verticillium in Cotton point approach

Seed treatment, soil manipulation, and crop sanitation can help cotton growers of the San Joaquin Valley avoid severe losses from Verticillium wilt, according to recent studies. A sustained, well-coordinated combination of cultural practices is suggested as a basis for cultural control of Verticillium wilt in cotton.

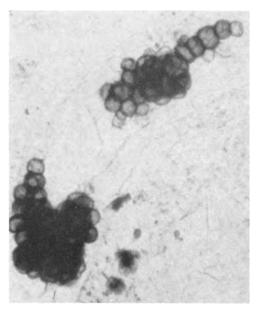
microsclerotia to as high as 500 to 1,000 per gram in the top few inches of soil. This means that an abundance of microsclerotia will be in contact with the underground parts of the cotton plant, permitting multiple infections of susceptible tissues when conditions are right. Studies on the vertical distribution of the *Verticillium* fungus in soil show that while the top few inches may carry high populations of the fungus, the count drops rapidly with depth, and at the 1-ft level there may be only a small fraction of what it is in the top few inches, and at 2 to 3 ft the amount is even lower.

Of a total of 1,320 soil samples collected from three cotton fields throughout the 1964 season (4-inch increments to a depth of 24 inches), 58% of the samples from the upper soil produced infection in the cotton plant in contrast to 2.5% and 1.5% in soil collected at the 16-inch depth and below. Infection in the cotton plant was initiated from soil collected at 8 to 12- and 12 to 16-inch depths in only 23% and 11% of the samples, respectively. Verticillium inoculum is evidently concentrated in the surface soil, and it is in these layers also that the highest number of cotton root infections were detected.

One of three methods available to reduce inoculum in the soil is to rotate cotton with a cereal such as barley, corn, sorghum, or other *Verticillium* nonhost crops. Microsclerotia gradually die in the absence of a susceptible crop or in the presence of such immune crops as represented by the cereals. (Even when cotton is being grown, the count of viable microsclerotia drops in one season to less than 50% of what it was during the period of land preparation and preplant irrigation in March.) In fields with low levels of infestation, short-term rotation may be enough to maintain that status, but in heavily infested fields long-term rotation is indicated.

Since the high concentration of microsclerotia is in the top, cultivated layer of the soil, the grower may bury this layer on some lands by deep, or rigolen plowing. Rigolen plowing means to plow in such a way that the soil layers are inverted, and earlier studies have shown that inverting the top 2 ft of soil submerges the inoculum of Verticillium and replaces the surface soil with deeper soil relatively free from infestation. A moldboard plow, plowing to $2\frac{1}{2}$ to 3 ft, equipped with a scraper which directs the surface foot of soil into the bottom of the furrow, is required to do this job. Since microsclerotia gradually die in soil, those buried would not be expected to survive.

Soil fumigation may be another effective method for controlling Verticillium wilt of cotton. In the strawberry and market-tomato industries, fumigation with a mixture (by weight) of 55%chloropicrin and 45% methyl bromide at rates of 250-350 lbs per acre will control Verticillium wilt. Tests have shown that



Two microsclerotia of Verticillium albo-atrum formed in a cotton rootlet in the field, enlarged 300 times to show the multicellular nature of the individual resting structure of the fungus, the form surviving in crop refuse and in soil.

control of *Verticillium* wilt by soil fumigation is possible in cotton, but stunting sufficient to cause severe reductions in yield has occurred. Bromine from methyl bromide is the primary cause of the stunting, but the exact mechanism of the action is not understood. Although soil fumigation is expensive, and has created a new problem, it may yet prove to be economical for the cotton grower, if the cost can be spread over 3 to 4 years and the stunting overcome.

More research is needed on the practices of removing diseased plants, of fumigation, and of Rigolen plowing before they can be recommended for the control of *Verticillium* wilt of cotton.

As already observed, the microsclerotial population in soil tends to decline, probably through the natural processes of microbial competition. This advantage is offset, however, when refuse of a diseased crop is shredded and spread over the land. One small cotton plant affected with Verticillium wilt produced (by actual count) a quarter of a million microsclerotia in its dead tissue (see photos). This figure could easily reach one million in a large plant in the field. Returning the refuse of such plants to the seedbed just prior to sowing a new crop adds a tremendous new quantity of inoculum. In one test using diseased cotton stems, roots, and leaves separately to infest soil, 100% of cotton plants succumbed to Verticillium wilt where stem pieces were used, in contrast to much lower percentages from roots or leaves. Infested refuse should be removed and burned to kill the microsclerotia, or disposed of in other ways.

As is usually true of programs for the control of crop diseases, a sustained, wellcoordinated combination of practices is most effective. In the case of Verticillium wilt of cotton, grower losses may be reduced by adherence to the basic principles of disease control: (1) Continue to use seed freed of Verticillium by acid delinting, or by mercury (Panogen-Ceresan) treatment. (2) Rotate nonsusceptible, irrigated crops with cotton to decrease populations of the fungus already present in infested land. Soil fumigation or deep-inversion plowing offers possible additional means of lowering the inoculum load of microsclerotia in soil. (3) Practice sanitation to avoid the buildup of Verticillium in the soil. Avoid adding to the land large populations of the fungus that may be present in diseased cotton refuse.

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Testing effects of

LOW- VS. HIGH-LEVEL PROTEIN CONCENTRATE **MIXES FOR DAIRY** MILK PRODUCTION

S. E. BISHOP • D. L. BATH

T IS COMMON PRACTICE for dairymen in southern California to feed freechoice alfalfa hay containing 18 to 22% crude protein, supplemented with concentrate mixes containing 14 to 18% crude protein (90% dry matter basis). These rations provide more protein than recommended allowances for even exceptionally high producing cows. Even when part of the alfalfa hay is replaced by corn silage or cereal green chop, recommended allowances for crude protein are exceeded. With feed costs now amounting to about 55% of yearly expenses, and economic conditions demanding increased efficiency, the possibility of using lower-cost concentrate mixes should be considered.

Prices of the more common highprotein feeds used for dairy cattle, such as coconut oil meal and cottonseed meal, are usually higher than those for lowprotein grains such as barley and milo which are high in energy. Under these conditions, partial replacement of highprotein feeds with grains will reduce feed costs. If the energy level is not lowered and the protein allowance is fulfilled, milk and milk fat production levels should be maintained. A feeding trial was conducted at La Sierra College Dairy, Riverside County, to evaluate possibilities for commercial application of the low-protein concept.

The milking herd was divided randomly into two groups. Twenty cows from each group were paired according to: (1) previous DHIA production or, in the case of first-calf heifers, predicted production from previous DHIA test-day data; (2) number of previous lactations; (3) number of days elapsed in present lactation; (4) last test-day production. All production data (see table 1) were adjusted to a 305-day mature equivalent basis and expressed as pounds of 4% fat-corrected milk (4% FCM).

The control group received a 17% crude protein concentrate mix which had been used regularly by the dairy, and the test group was fed a mix containing 12% crude protein. Both mixes contained the same ten ingredients but amounts of four ingredients were adjusted to obtain a lower protein level in the experimental mix (table 2).

Each group was fed its concentrate mix in pelleted form in an elevated parlor barn. The parlor was divided into two one-sided units, each with four stalls. The cows on high protein concentrate were milked on one side, those on the test feed on the other. All cows were offered concentrates free choice while in the milking parlor.

Both groups received identical forage allowances. Alfalfa hay was fed throughout the trial. When available, corn silage, oat silage, barley green chop and alfalfa green chop were fed free choice in corral mangers in addition to alfalfa hay. At any given time the amount of alfalfa hay was varied according to the amount of silage or green chop available. Core samples of hay and grab samples of silage, green chop, and the concentrate mixes were obtained periodically for proximate analyses.