

*Variations in Aphid Transmission
of Tristeza Virus*

VARIATION in efficiency of aphids as vectors of tristeza virus has been noted among species reported as vectors in different infected areas. The brown citrus aphid [*Toxoptera citricidus* (Kirk.)] is considered an efficient vector in certain areas (1), but is not known to be present in the United States. The melon aphid [*Aphis gossypii* (Glov.)] was shown by Dickson to transmit quick-decline (tristeza) virus in California, but was judged to be an inefficient vector (2). Tests in Florida (4, 5) have shown that the spirea aphid (*Aphis spiraecola* Patch) and the black citrus aphid [*Toxoptera aurantii* (Fonsc.)] as well as the melon aphid are relatively inefficient vectors of the tristeza virus.

The present studies were undertaken to obtain information as to whether source plant varieties and variations in clones within an aphid species could influence vector efficiency.

Materials and Methods

Alate spirea and melon aphids and apterous black citrus aphids were reared on Temple orange trees growing in 10-gallon crocks. Apterous melon aphids from Orlando and Belle Glade were reared on kenaf (*Hibiscus cannabinus* L.). The melon aphids from Belle Glade were obtained from John N. Simons, Everglades Experiment Station. This species had been found to be an efficient vector of southern cucumber mosaic virus at Belle Glade (6). The Orlando clone was from a colony used in studies on transmission of tristeza at the Orlando laboratory of the U.S. Department of Agriculture (5).

Alate aphids were collected from the sources of inoculum with an aspirator and placed on Key lime test plants in darkened cages inside the insectary. At the end of each test, aphids were sent to the taxonomists of the Entomology Research Division of the U.S. Department of Agriculture for confirmation of identification.

The three strains of tristeza virus used in these studies have been previously described (4, 5). Sources of inoculum for the mild strain (T_1 and T_2) were Temple orange seedlings selected for uniformity and inoculated by means of leaf pieces from aphid-infected Key lime test plants. Sources of inoculum for the severe strain T_3 were young sweet orange seedlings grown in the greenhouse. Each of these plants had been inoculated by inserting 2 leaf pieces from Key lime test plants into the bark. Seedlings were cut back to single 14-inch stems to stimulate new growth. Leaf pieces from each of the sweet orange sources of inoculum were inserted into the trunks of healthy Key lime indicator plants to verify the presence of tristeza virus. Only inoculum sources which had proved to be positive for the virus were used.

Initial tests for aphid transmission were made 160 days after the carrier plants had been inoculated. Sources of inoculum and test plants selected were in a flush of growth with optimum conditions for aphid feeding. A minimum feeding period of 24 hours was allowed on the sources of inoculum. Leaves and shoots infested with aphids were then removed and placed on healthy Key lime indicator plants cut back to 12 inches to stimulate growth. Aphids were allowed to move voluntarily to the test plants. The feeding period was measured from the time the majority began to feed on the indicator plants. Melon and spirea aphids fed for 6 hours and black citrus aphids for 12 hours. Following the experimental transmission attempt, plants were sprayed with nicotine sulfate and returned to the greenhouse to prevent contamination by other insects.

Results

All 3 strains of virus were transmitted experimentally by the Orlando clone of melon aphids. They infected 5 of 6 Key lime plants with the T_1 strain, 6 of 6 test plants with the T_2 strain, and 3 of 12 test plants with the T_3 strain. The T_3 sources of inoculum were 5 Hamlin, 2 Bedman, 1 Sanguina Grosse Ronde, 2 Maltese Oval, 1 Mediterranean Blood, and 1 Valencia orange. Two of the transmissions of the T_3 strain were from Hamlin and the third from Maltese Oval.

PROCEEDINGS of the IOCV

Vein-clearing symptoms typical of each strain of virus appeared in test plants 40 to 55 days after the test, but became masked and less distinct by the middle of the summer. In July, stems were examined for pits. There were 4 pits per inch average in plants with the T₁ strain, 13 per inch for the T₂ strain, and 80 per inch for the T₃ strain. By June, 1960, one of the T₃-strain test plants had died and another had severe dieback. The average numbers of pits at this time were 4 for the T₁ strain, 10 for the T₂ strain, and 49 for the T₃ strain.

Alate melon aphids of the Orlando clone were allowed to feed on McIlhenny, Princess Early, and Hamlin sweet orange seedlings that had been inoculated with the T₃ strain and were then transferred to healthy Key lime indicator plants. In 15 tests, there was one positive transmission by 50 alate melon aphids, this being from a Hamlin seedling.

Comparisons of transmission of T₃ inoculum by Belle Glade and Orlando clones of melon aphids are summarized in Table 1. In the

TABLE 1. TRANSMISSION OF THE T₃ SEVERE STRAIN OF TRISTEZA BY APTEROUS MELON APHIDS OF TWO CLONAL ORIGINS

Date	Number of aphids	Belle Glade clone		Orlando clone	
		number of plants tested	percentage of plants infected	number of plants tested	percentage of plants infected
Sept. 1958	100	6	66	6	33
May 1959	300	5	20	5	0
Aug. 1959	100 - 300	10	10	10	0

first series of tests (September, 1958), all sources of inoculum were Hamlin seedlings except one, a Florida sweet orange. All transmissions were from Hamlin sources. In a second series (May, 1959), sources of inoculum were Hamlin and Pineapple orange seedlings for the Belle Glade clone, and Hamlin, Selecta, and Joppa orange for the Orlando clone. The lone transmission with the Belle Glade clone was from the Hamlin orange. In the third series (August, 1959), sweet orange sources of inoculum were Valencia, Precose de Valence, Joppa, St. Michael, Morocco, Princess Early, and Hamlin. Except for the one transmission with Belle Glade aphids fed on Precose de Valence orange, all other transmissions in the 3 series of tests were from Hamlin orange.

To determine whether the black citrus aphids were capable of trans-

mitting the T_3 strain, 102 transmissions were attempted. Of these, only 3 were positive, made in February, 1958. Seedling sources of inoculum included the sweet orange varieties Mediterranean Blood, Precose de Valence, Cuba Sweet, and St. Michael, all tissue-inoculated. In the 3 transmissions, Precose de Valence and St. Michael were sources of inoculum, with 100 aphids per test plant.

Twenty-five tests were made with alate spirea aphids to determine whether this species would transmit the T_3 strain. Several varieties of inoculated sweet orange seedlings were used, with 100 aphids on each plant. Only a single positive transmission was obtained, and the source was a Hamlin orange seedling.

Surveys for aphids on Temple and Valencia orange trees on sour orange rootstock were made in 1953 and again in 1960. A spring survey in 1953 showed that 31 per cent of the Temple orange trees were lightly infested and 9 per cent moderately infested with spirea aphids. Twenty per cent of the Valencia block was lightly infested. Surveys during the summer and fall months showed a decline in the aphid population. In April 1960, all the Temple trees were heavily infested with spirea aphids, and were infested with a few colonies of melon and black citrus aphids. In the Valencia block, the infestation rate was as follows: light, 33 per cent; moderate, 31 per cent; and heavy, 7 per cent. Six of the Valencia trees had side branches of Temple orange grafted on them. Two of these Temple side branches were infested with spirea aphids, but the Valencia portion of each tree was not infested. The Temple branches on the Valencias had much more succulent growth than the rest of the trees at the time of examination.

The spread of tristeza in this grove was shown by Florida State Plant Board inspectors to be greater in the Temple trees. In August, 1955, 20 per cent of the Valencias and 30 per cent of the Temples were diseased. In December, 1958, 35 per cent of the Valencias and 73 per cent of the Temples were infected.

Discussion

Tests with tristeza virus of citrus suggest that the rate of transmission is influenced by the variety of citrus serving as the source of inoculum. Temple orange, which was associated with the earliest discoveries of tristeza-infected groves in Florida, has proved to be a good source of inoculum for transmission by aphids under laboratory conditions. A Temple orange containing the T_3 strain of tristeza was not available at

the time the present study was made; therefore transmissions by aphids of the mild and severe strains from this variety could not be compared. Hamlin orange has appeared to be a good source for transmission of the standard T₃ strain.

Simons (6), working with aphid vectors of southern cucumber mosaic virus, found that the plant species providing the virus could influence transmission. Kirkpatrick and Ross (3), using the green peach aphid (*Myzus persicae* (Sulz.)) as a vector of potato leafroll virus, found Florida groundcherry (*Physalis floridana*) to be a better source of inoculum and cutleaf groundcherry (*P. angulata*) a poorer source than Jimsonweed (*Datura stramonium*). All these were better sources than potato (*Solanum tuberosum*). Sylvester (8), working with the green peach aphid as a vector of lettuce mosaic virus, found evidence to indicate that the host plant on which the aphids were reared could influence virus transmission.

Simons (7) found differences in the ability of several clones of the cotton (melon) aphid to transmit southern cucumber mosaic virus. Among aphids which he used to transmit this virus, the clone used in the tristeza tests was considered by Simons to be the least efficient.

Variations in vector efficiency also have been reported for the green peach aphid by workers investigating other vegetable viruses. Williams and Ross (9) found marked differences, apparently because of inherited characters, in the ability of green peach aphid individuals to transmit potato leafroll virus. Progeny of individuals that were good transmitters were efficient vectors, whereas progeny of poor transmitters were inefficient. A clone of melon aphids obtained from noncitrus hosts produced slightly more transmissions than aphids obtained from citrus hosts.

The black citrus aphid in earlier experiments (4) transmitted the mild strain of tristeza virus when an infected Valencia orange scion on Key lime rootstock was used as the source of inoculum. In the present work, Precose de Valence, an early variety of Valencia, was the source of inoculum for one of the transmissions. These transmissions show that although the black citrus aphid is an inefficient vector of both the mild and severe strains of tristeza virus under laboratory conditions, it must be considered a potential vector under field conditions.

Work in Brazil (1) and later in Florida (4) indicated that higher rates of transmission could be obtained with large numbers of aphids (100 to 500). Apterous aphids were used in most of these experiments

to facilitate handling. Spread of the virus in the field presumably is by winged forms of aphids. Alate and apterous forms of both the melon and spirea aphids were found to be capable of transmitting the T₃ strain of tristeza under experimental conditions.

Literature Cited

1. COSTA, A. S., and T. J. GRANT. 1951. Studies on transmission of the tristeza virus by the vector *Aphis citricidus*. *Phytopathology* 41: 105-113.
2. DICKSON, R. C., R. A. FLOCK, and M. M. JOHNSON. 1951. Insect transmission of citrus quick-decline virus. *J. Econ. Entomol.* 44: 172.
3. KIRKPATRICK, H. C., and A. F. ROSS. 1952. Aphid-transmission of potato leafroll virus to solanaceous species. *Phytopathology* 42: 540-546.
4. NORMAN, P. A., and T. J. GRANT. 1956. Transmission of tristeza virus by aphids in Florida. *Proc. Florida State Hort. Soc.* 69: 38-42.
5. NORMAN, P. A., and T. J. GRANT. 1959. Transmission of T₂ severe strain of tristeza virus of citrus by the melon aphid. *J. Econ. Entomol.* 52: 632-634.
6. SIMONS, J. N. 1955. Some plant-vector-virus relationships of southern cucumber mosaic virus. *Phytopathology* 45: 217-219.
7. SIMONS, J. N. 1959. Variation in efficiency of aphid transmission of southern cucumber mosaic virus and potato virus Y in pepper. *Virology* 9: 612-623.
8. SYLVESTER, E. S. 1955. Lettuce mosaic virus transmission by the green peach aphid. *Phytopathology* 45: 357-369.
9. WILLIAMS, W. L., and A. F. ROSS. 1957. Aphid transmission of potato leafroll virus as affected by the feeding of nonviruliferous aphids on the test plants and by vector variability. *Phytopathology* 47: 538.