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Epidemiology of the Tristeza Virus Complex under South African Conditions

THE SPREAD OF TRISTEZA has been studied in relation to the flight behavior of the aphid vector by Dickson *et al.* (1) in California where the vectors, *Aphis gossypii* Glover and *A. spiraecola* Patch, are inefficient transmitters. The present work reports on the flight behavior of the tropical citrus aphid [*Toxoptera citricidus* (Kirkaldy)], the most efficient vector of tristeza, and how it correlates with spread of the tristeza virus complex in South Africa. Information is also included on the flight behavior of other aphid species, found on citrus, that might be concerned with the transmission of tristeza virus.

Materials and Methods

Yellow water traps similar to those described by Moericke (4) but provided with a refilling device were exposed to aphid flight at Nelspruit (altitude 665 m.) in September, 1961, and at Buffelspoort (altitude 1295 m.) in January, 1962. At Nelspruit ten traps were set at various places inside the orchards of the Research Institute and at Buffelspoort nine were installed.

The yellow water traps were suspended five feet from the ground and held by means of iron rings attached to a pole.

A group of four healthy glasshouse-raised West Indian lime [*Citrus aurantifolia* (Christm.) Swing.] seedlings (bait plants) were likewise exposed at each site. The seedlings were in pots painted to reduce evaporation, and were also suspended five feet from the ground. The seedlings served as traps for any aphid-transmitted virus that might be carried by the winged aphids feeding on them. To attract aphids to the bait plants, two artificial leaves made from aluminum foil painted yellow

were attached to each plant about an inch above the soil level. The artificial leaves served as an attractive landing platform from which the aphids crawled to the seedling and eventually to its tip.

The bait plants were exposed for four weeks and then withdrawn for observation in the glasshouse. Those bait plants developing vein clearing and wood pitting were grafted each onto one sour orange (*C. aurantium* L.) and one lemon [*C. limon* (L.) Burm. f.] seedling to see whether the virus picked up by the bait plant would cause a yellows reaction. At some sites, the unit of one water trap and four bait plants was adjacent to an orchard consisting of all sweet orange [*C. sinensis* (L.) Osbeck], all lemon, or all grapefruit (*C. paradisi* Macf.) plants. At other sites, the adjacent trees were a mixture of different species of citrus.

An experiment was done to determine whether or not the artificial yellow leaves attracted more aphids to a seedling. A unit of four bait plants with yellow leaves attached and four plants without yellow leaves was set up at each of five sites, a distance of 20 feet being maintained between the two sets of plants in the unit to prevent interaction. Twice a week the two groups of plants at each site were switched in order to cancel out any advantage either position might have in attracting winged aphids. A yellow water trap was also positioned at each site.

Results

Flight behavior of the vector species.—In Nelspruit, the most common citrus aphid is *Toxoptera citricidus*. In summer 1961-62 the flight activity of this species was about six times as great as in summer 1962-63. At Buffelspoort, where the over-all flight activity was about one-third that at Nelspruit, there was hardly any flight in the summer 1962-63. At both places the flight activity of *T. citricidus* is closely correlated with the flush cycles of the citrus trees. Data from single traps reveal very clearly the correlation between the trappings and the flush cycles of the species of citrus next to the trap. Whereas the flight peaks of *Aphis gossypii*, the most common aphid on citrus, correlate well with those of *T. citricidus* at Buffelspoort, they do not at Nelspruit. It is supposed that the polyphageous *A. gossypii* trapped at Buffelspoort came mainly from citrus, whereas those at Nelspruit came from hosts other than citrus, especially from April to August, 1961, a period in which there is hardly any flush on citrus and no aphid colonies were found on citrus. Only single specimens of the aphid species *Toxoptera aurantii* (Boper de Fonscolombe) and *Aphis craccivora* (Koch) were found in the traps.

Taking the number of *T. citricidus* collected twice weekly from the plants with yellow leaves as 100 per cent, the percentages of those collected from the plants without yellow leaves were 12 per cent in the November, 1961, flight peak, 107 per cent in the April, 1962, peak, and 44 per cent in the January, 1963, flight peak. On the average 1.56 times as many aphids were collected from the bait plants with yellow leaves as from those without them. These results may be explained by a seasonal variation in the color reaction of *T. citricidus.*

VIRUS INFECTION OF WEST INDIAN LIME BAIT PLANTS BY WINGED APHID VECTORS.—Figure 1 shows the correlation between water trappings of *T. citricidus,* the number of *T. citricidus* collected from ten groups of four West Indian lime bait plants, and the number (out of 40) of bait plants infected with strains of tristeza virus at Nelspruit and at Buffels-

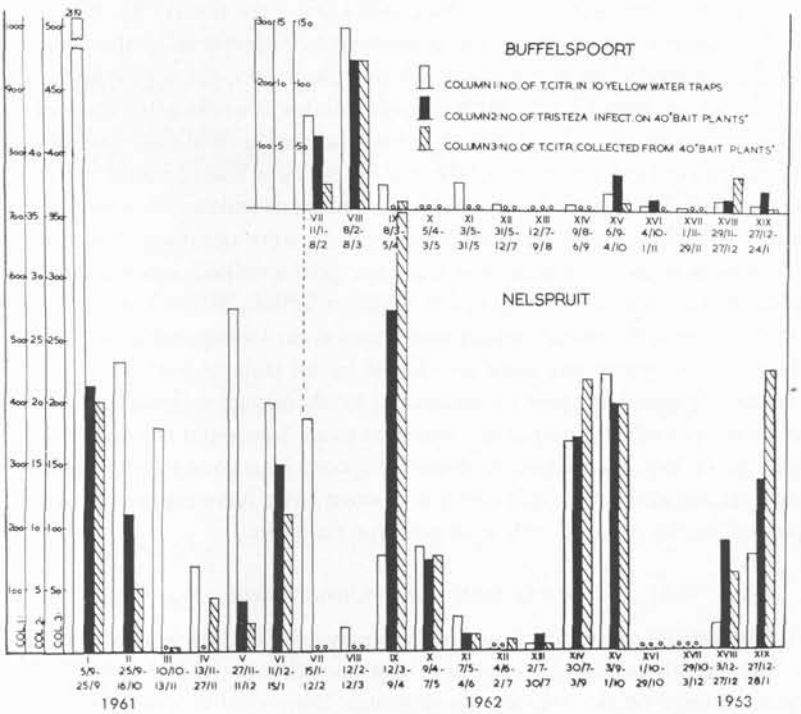

FIGURE 1. *Comparison of the numbers of T. citricidus trapped in 10 water traps, the numbers of T. citricidus collected from 40 bait plants, and the numbers of 40 bait plants infected by tristeza virus during similar exposure periods throughout the year at Nelspruit and at Buffelspoort.*

poort. The exposure time was usually four weeks. The correlation coefficient between the yellow water trap data and tristeza virus infection is 0.44. The correlation coefficient between the number of *T. citricidus* collected from the bait plants and the tristeza virus infections is, however, significant at the 5 per cent level; the correlation coefficient is 0.93. This suggests that the alighting and staying-leaving behavior of *T. citricidus* as reflected by the number of aphids collected from the bait plants gives a better indication of the degree of infection than that reflected by the yellow water trappings. The infection at Buffelspoort, due to a weaker flight of *T. citricidus,* is lower than that at Nelspruit. In view of the higher proportion of *A. gossypii* present at Buffelspoort, single infections might have been caused by this species. Because of the considerably lower vector efficiency of this species, however, its influence seems to be negligible.

Strain variation in tristeza isolates.—As shown by the West Indian lime reaction, there are considerable differences in the severity of the trapped virus strains. At both trappings sites, the severe and very severe strains prevail. Of 180 Nelspruit isolates, 8 weeks after the end of their exposure period, 16 per cent had induced a medium reaction on West Indian lime test plants, whereas 84 per cent had induced severe to very severe symptoms. By retesting the Nelspruit isolates on sour orange and lemon, the following preliminary results were obtained. Not one of 12 isolates with a medium lime reaction gave a yellows reaction. Of 41 isolates with a severe to very severe lime reaction, 30 gave a clear, 5 a doubtful, and the others no yellows reaction on lemon and sour orange. The yellows symptoms were developed by all isolates with severe lime reaction trapped at three sites adjacent to old orange orchards, two next to a variety orchard, and at one site next to an 8-year-old lemon orchard. Groups of bait plants that had been exposed in a young grove planted with lemon and grapefruit did not in most cases have the yellows component in the isolates with a severe lime reaction.

Discussion and Conclusion

The results show that even in a country with an efficient vector and a high incidence of tristeza there are considerable differences in the epidemiology of the tristeza virus group from year to year, region to region, and from grove to grove. The relative abundance of *T. citricidus* and that of *A. gossypii* can be different at various places. This raises the question whether there are ecological factors that limit the presence

of *T. citricidus* in some areas of South Africa or, on a larger scale, prevent this species from becoming endemic in some citrus-growing countries. The answer to this question is most important to the Mediterranean countries (2).

Whereas a high infection by severe strains of tristeza virus could take place at Nelspruit within a few weeks after the flight of *T. citricidus* has commenced; at Buffelspoort a seedling could be exposed for years without becoming infected. This is even more true for a seedling in the close stand of a nursery; because in the case of bait plants, standing much less crowded at a height of five feet, they are favored infinitely more by visitations of flying aphids. McClean (3) presented evidence that seedling yellows virus, which is present in the tissues of a rough lemon stock, has an immediate stunting effect on the lemon or grapefruit scion. He proposed a spraying program in nurseries especially for stock seedlings which are to be budded to grapefruit and lemon. This, in the light of the findings, seems to be a practical approach.

It appears that the value of investigations as set out above would be considerably increased if the experiments would be replicated in various countries. This would facilitate the analysis of the local results and would make forecasts on the spread of the virus easier. Furthermore it would facilitate forecasts as to the possibility of introducing the tristeza complex to countries where tristeza is not yet present.

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