## The Distribution of *Xylella fastidiosa* in Citrus Variegated Chlorosis-Affected Trees

## M. J. G. Beretta, R. Harakava, K. S. Derrick, R. F. Lee, and F. F. Laranjeira

ABSTRACT. The distribution of *Xylella fastidiosa* in citrus variegated chlorosis (CVC)-affected trees was examined in a grove in north São Paulo State. The study was conducted over a 2-yr period, starting in 1993 when the trees were 3 yrs old. Tissue extracts were assayed for the bacterium by dot immunobinding assays and western blots. Positive results were obtained from young and mature leaves with disease symptoms and occasionally from asymptomatic leaves from affected branches. The bacterium was also present in branches that had symptomatic leaves up to 2 cm in diameter, but was not detected in leaves or branches with no symptoms on affected trees. In mature fruit taken from symptomatic branches, the bacterium was found in the abscission zone, peduncle and albedo. There was no indication that the CVC-causing bacterium was present in roots of various sizes, old limbs, flowers, seeds or juice. These observations on the uneven distribution of the bacterium in affected trees led to the development of a very effective scheme for controlling CVC based on removal of all symptomatic branches.

Citrus variegated chlorosis (CVC) is caused by a pathovar of Xylella fastidiosa (3, 7, 9). Studies on other diseases caused by X. fastidiosa have shown that these bacteria are readily transmitted by xylem-feeding leafhoppers and sharpshoooters and by vegetative propagation. The pathogen has a wide host range, and is frequently found in weeds (4, 5, 8). CVC has been shown to be transmitted by budding and appears to have an aerial vector, although experimental transmission by vectors has not been demonstrated. Trees in established groves that were previously free of CVC frequently develop symptoms on only one branch. This indicates vector transmission; at this time it is not known if the source of inoculum is citrus or weeds. CVC appears to be more severe where weed and sharpshooter populations are high. Observations of the uneven distribution of symptoms of CVC in grove trees led to experiments reported herein on the distribution of X. fastidiosa in infected trees.

A large commercial planting on Rangpur lime rootstock in Macaubal, located in northern São Paulo State, was used in these experiments. The trees received standard cultural practices with no irrigation. At the beginning of the experiment only a few leaves on affected plants had symptoms. Over the course of the 2 yr experiment, symptoms developed over entire trees and only small fruit were produced.

Ten trees were selected and sampled beginning in March, 1992. Samples were collected in March, July and November; the last samples were taken in July, 1994. Samples were taken from symptomatic and asymptomatic limbs (up to 2 cm in diameter), from trunks (above and below the budunion) and from young and old leaves. Samples were also taken from roots of various sizes, fruit (peduncle, albedo and abscission layers), flowers, seeds, and juice. Serological tests were done by dot immunobinding and western blot assays (2, 6).

X. fastidiosa was always found to be present in symptomatic young or mature leaves and in limbs up to 2 cm in diameter that had symptomatic leaves. The bacteria were not detected in leaves or branches with no symptoms or in limbs larger than 2 cm. The bacteria were found in mature fruit taken from symptomatic branches in the abscission layer, peduncle and fruit albedo. All assays of various sizes roots, flowers, seeds and juice were negative.

In CVC-affected trees, there is a good correlation between symptoms and the presence of disease-causing bacteria. The bacteria move very slowly to adjacent branches, suggesting the possibility that the disease might be controlled by timely removal of symptomatic limbs. These findings have been used to develop an effective strategy for controlling CVC (1).

## LITERATURE CITED

- Beretta, M. J. G., V. Rodas, A. Garcia Junior, and K. S. Derrick 1996. Control of citrus variegated chlorosis by pruning. p. xx-xx. *In:* Proc. 13th Conf. IOCV, IOCV, Riverside.
- Harakava, R., M. J. G. Beretta, R. F. Lee, K. S. Derrick, and C. B. deJesus 1994. Detection of *Xylella fastidiosa*, causal agent of citrus variegated chlorosis, by western-blotting. Summa Phytopathogica 20: 53.
- Hartung, J. S., J. Beretta, R. H. Brlansky, J. Spisso, and R. F. Lee 1994. Citrus variegated chlorosis bacterium: Axenic culture, pathogenicity and serological relationships with other strains of *Xylella fastidiosa*. Phytopathology 84: 591-597.
- Hill, B. L. and A. H. Purcell 1995. Acquisition and retention of *Xylella fastidiosa* by an efficient vector, *Graphocephala atropunctata*. Phytopathology 85: 209-212.
- Hopkins, D. L. and W. C. Adlerz 1988. Natural hosts of Xylella fastidiosa in Florida. Plant Disease 72: 429-431.
- Lee, R. F., M. J. G. Beretta, K. S. Derrick, and M. E. Hooker 1992. Development of a serological assay for citrus variegated chlorosis. A new disease of citrus in Brazil. Proc. Fla. State Hort. Soc. 105: 32-34.

7. Lee, R. F., M. J. G. Beretta, J. S. Hartung, M. E. Hooker, and K. S. Derrick

- 1993. Citrus variegated chlorosis: confirmation of a *Xylella fastidiosa* as the causal agent. Summa Phytopathologica 19: 123-125.
- 8. Tebar, L. R., L. Beretta, C. B. de Jesus, A. P. C. Alba, M. J. G. Beretta, R. F. Lee, and K. S. Derrick

1994. Detection of *Xylella fastidiosa* antigens in weeds collected from citrus orchards affected by citrus variegated chlorosis - CVC. Fitopatol. Bras. 19: 319.

9. Rossetti, V., M. Garnier, J. M. Bové, M. J. G. Beretta, A. R. R., Teixeira, J. A. Quaggio, and J. D. De Negri

1990. Présence de bacteriés dans le xyléme d'orangers atteints de chlorose variégée, une nouvelle maladie des agrumes au Brazil. C. R. Acad. Sci. Paris, Serie III 310: 345-349.