

## Preliminary Evaluation of Citrus Tristeza Virus Isolates from the Isla de la Juventud, Cuba

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**ABSTRACT.** Several citrus tristeza virus (CTV) isolates from the Isla de la Juventud, Cuba, were selected for characterization. The field isolates were indexed in Mexican lime, sour orange, and Valencia sweet orange on sour orange, and the results suggested that all isolates were mild. CTV was detected in field trees by ELISA surveys conducted at two times of the year. The reaction of isolates to a panel of five monoclonal antibodies enabled differentiation of the isolates into serogroups. Some differences were observed between healthy and virus-infected trees in the field when fruit quality and growth parameters were analyzed.

During an earlier survey carried out in Cuba, a low incidence of citrus tristeza virus (CTV) was found. All plants were asymptomatic although they were on sour orange rootstock. The highest CTV incidence was found on the Isla de la Juventud. Characterization of the CTV isolates in Cuba suggest the presence of mild isolates (1). Due to the importance of isolate characterization in defining control strategies, the present study was conducted to characterize some of the isolates from the Isla de la Juventud citrus region.

Isolates were collected from five trees, designated as 1-2, 1-12, 1-13, 1-18, and 1-24, in a 20-yr-old grove of Marsh grapefruit on sour orange rootstock. For ELISA assay, four young shoots with leaves were collected from different parts of the

canopy at two different times of the year, December and June, 1996. For CTV detection, DAS ELISA was conducted using a mixture of 3DF1 + 3CA5 monoclonal antibodies (Mabs) (2). It was determined that CTV could be detected with high OD values from leaf and flush samples during both sample periods. The assay results were consistent for leaf and flush samples for three of the isolates (1-12, 1-13, and 1-18), but for isolates 1-2 and 1-24 there were differences between the leaf and flush samples. Analysis of the five isolates using a panel of five different Mabs, 3C1F10, 3DF1, 3CA5, 4B1-3, 310E3, demonstrated that the isolates could be differentiated into three serogroups (Table 1).

The length of individual flushes and number of leaves from each infected field tree were analyzed

TABLE 1  
BIOLOGICAL ACTIVITY OF CITRUS TRISTEZA VIRUS ISOLATES AND REACTIVITY AGAINST A PANEL OF FIVE MONOCLONAL ANTIBODIES

Isolates	Symptoms <sup>a</sup>		Monoclonal antibodies					Serogroup
	Chlorosis	Stem pitting	3C1F10	3DF1	3CA5	4B1-3	310E3	
1-2	ND	ND	+	+	+	+	+	1
1-12	—	+	+	—	+	+	+	2
1-13	+	+	+	+	+	+	—	3
1-18	+	—	+	+	+	+	—	3
1-24	ND	ND	+	+	+	+	+	1

<sup>a</sup>Symptoms on sweet orange grafted onto sour orange indicator plants. ND = not determined.

TABLE 2  
EFFECT OF CITRUS TRISTEZA VIRUS (CTV) INFECTION ON AVERAGE SHOOT LENGTH  
AND LEAVES PER SHOOT

Plants	Average shoot length			Average leaves per shoot		
	1995	1996	1997	1995	1996	1997
CTV infected	7.2	10.2	8.3	6.6	8.8	6.9
Healthy	8.1	12.6	12.6	6.8	8.9	9.5
CV (%)	3.2	4.6	4.9	1.3	2.1	1.5
SE	0.25**	0.53**	0.51**	0.03ns	0.06**	0.04**

\*\*Indicates means are significantly different at  $P \leq 0.01$ . ns = means are not statistically different.

during the spring flushes and compared to five healthy trees that were included as controls. Differences were found in both parameters between infected and control plants during each of the three years of the study with the exception of the number of leaves in 1995 (Table 2). To evaluate fruit difference, five fruit from each tree were weighed and their diameter, degree of acidity, and maturity determined during 1995 and 1996. No differences were found between fruit from healthy and CTV-infected trees.

Three of the isolates were included in a biological characterization assay carried out in the greenhouse with temperatures ranging from 20° to 25°C. The indicator plants used were Mexican

lime, sour orange, and Valencia sweet orange on sour orange. Different intensities of vein clearing and leaf cupping were detected for all isolates in Mexican lime, but the reactions were all considered to be mild. Seedling yellows was not observed on the sour orange, nor did decline occur on the sweet orange on sour orange combination.

The few phenotypic and immunological differences observed among the isolates combined with symptoms expression in the host plant bioassays suggest that they are predominantly mild isolates. Nevertheless, considering that the field isolates are a complex of CTV strains, it is necessary to continue their characterization to more fully define their composition and severity.

#### LITERATURE CITED

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