

Incidence of Tristeza and Other Citrus Diseases in Bolivia

L. W. TIMMER, Associate Professor, Institute of Food and Agricultural Sciences, Agricultural Research and Education Center, University of Florida, Lake Alfred, 33850; RALPH SCORZA, Assistant Research Scientist, Department of Fruit Crops, University of Florida, with the U.S. Agency for International Development, La Paz, Bolivia; and R. F. LEE, Assistant Professor, Institute of Food and Agricultural Sciences, Agricultural Research and Education Center, University of Florida, Lake Alfred

ABSTRACT

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Citrus tristeza virus was detected serologically in more than 75% of the citrus trees tested in the Chapare and Santa Cruz areas of Bolivia, in more than 40% of the trees tested in the Yungas, and was also detected in the Alto Beni. *Toxoptera citricida*, an efficient aphid vector of the disease, was observed in the first three areas. Symptoms on naturally infected limes were generally mild. Phytophthora foot and root rot was the most serious disease in the Chapare and in the Yungas. Pink disease and Septoria leaf spot were important in the Yungas, and areolate leaf spot was serious in the Chapare. Melanose, greasy spot, scab, lime anthracnose, exocortis, xyloporosis, and psorosis were observed but were not of economic importance. Bacterial canker was not observed in the Yungas or in the Chapare.

Although Bolivia produces more than 100,000 tons of citrus fruit a year, the only published information on the occurrence and severity of citrus diseases there is a brief mimeographed report (4). Most citrus in Bolivia is grown in the Yungas, a mountain valley (altitude of 1,500–2,000 m) northeast of La Paz with distinct wet and dry seasons, and in the Chapare, a

lowland (300 m), tropical area east of Cochabamba that has 1.5–4.5 m of rainfall annually (Fig. 1). Some citrus is grown in the vicinity of Santa Cruz, a drier lowland area, with minor amounts in the Alto Beni and other localities. Most citrus species are nonbudded, seedling trees that are slow to bear and are susceptible to soilborne diseases. Citrus is usually intercropped with coffee, banana, coca, corn, yuca, or other crops.

Bolivian growers are being encouraged to use budded trees and, where citrus tristeza virus (CTV) is present, to avoid susceptible rootstocks. This study surveyed important citrus producing areas for CTV and surveyed the Yungas and the Chapare for other diseases.

MATERIALS AND METHODS

Survey. We examined commercial and experimental citrus plantings for symptoms of fungal, bacterial, and viral diseases of citrus (3, 5). In the Chapare we

examined commercial citrus in the vicinity of Villa Tunari, Paractí, Ibuelo, Villa 14 de Septiembre, and Chimoré and experimental plantings on the Chipiriri and La Jota experiment stations. In the Yungas we observed commercial citrus groves in the vicinity of Coroico, Cruz Loma, Coripata, Huancané, Chulumani, Ocobaya, Chicaloma, and Irupana and experimental plantings at San Pedro, Chulumani, and Irupana. Except for tristeza, all diagnoses were based on symptomatology alone.

Serologic tests for CTV. Bark samples were collected in the areas of the Yungas and the Chapare surveyed for disease symptoms, along the main road between Santa Cruz and Cochabamba (17 to 35 km from Santa Cruz), and at Sapecho in the Alto Beni. Bark was collected from the base of three young shoots from each tree. About 0.5 g fresh weight of bark was placed in a 10-ml serum vial partially filled with calcium chloride to dry the samples. Air and moisture were evacuated from the rubber-stoppered vials with a syringe, and the vials were sent to Florida for serologic tests.

Dried bark was ground in a mortar and pestle at a 1:5 (w/v) dilution in 0.05 M of tris(hydroxymethyl)aminomethane buffer, pH 8.0, for sodium dodecyl sulfate (SDS)-immunodiffusion tests (2); it was further diluted to 1:20 in the same buffer for enzyme-linked immunosorbent assay (ELISA) (1). Antisera were prepared to the severe T3 strain and to the moderate T4 strain of CTV (2), both of which

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Second author is now research horticulturist, Appalachian Fruit Research Station, Agricultural Research, Science and Education Administration, U.S. Department of Agriculture, Kearneysville, WV 25430.

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reacted with all strains of CTV tested (1).

All samples were tested by SDS-immunodiffusion using antisera prepared to whole, unfixed virus of the T3 strain and to SDS-degraded virus of the T4 strain. Two antisera were used for ELISA: one prepared to SDS-degraded virus of the T3 strain and the other prepared to whole, unfixed virus of the T4 strain. All samples were run in four ELISA tests, using each antiserum for coating and each as the conjugate antiserum in all possible combinations. All ELISA tests were scored visually and read in a spectrophotometer at 405 nm.

RESULTS

Tristeza and other viral diseases. We observed mild tristeza vein-clearing, leaf

cupping, and stem pitting in Mexican lime (*Citrus aurantifolia* (Christm.) Swingle) at several locations in the Yungas and the Chapare. Stem pitting was mild or absent on all Mexican limes and was observed on only one grapefruit (*C. paradisi* Macf.) in the Chapare. No stem pitting was observed on other varieties. Sweet orange (*C. sinensis* (L.) Osbeck) and tangerine (*C. reticulata* Blanco) on sour orange (*C. aurantium* L.) rootstock at Chulumani were in severe decline and had typical tristeza honeycombing symptoms at the bud union. *Toxoptera citricida* Kirk., the highly efficient aphid vector of CTV, was commonly observed in the Chapare, Yungas, and Santa Cruz areas.

Results of the SDS-immunodiffusion and ELISA tests using the various antisera were in good agreement; most samples rated positive had absorbance values of 0.18 to 0.65 in all tests. A few questionable samples were considered positive if they gave a precipitin line with

either antiserum and had an absorbance at 405 nm of more than double the background level of 0.05 in two ELISA tests.

A high percentage of trees tested in the Chapare (86%) and Santa Cruz areas (78%) was infected with CTV, but less than half (41%) of the trees tested in the Yungas were infected (Table 1). Samples positive for CTV occurred in all localities in the Yungas. Samples from the Alto Beni arrived in poor condition, but several were positive for CTV in tests using SDS-immunodiffusion and ELISA. No discernible difference was found in the amount of infection in different citrus species and hybrids. Young seedlings in the seedbed or nursery were generally not infected. Of eight young seedlings of several citrus species included in the survey, only a single Cleopatra mandarin (*C. reticulata* Blanco) was found to be infected.

Because most of the trees in commercial plantings were of seedling origin, the common graft-transmissible viral diseases were rare in Bolivia. Symptoms of exocortis were seen on trees budded on trifoliolate orange (*Poncirus trifoliata* (L.) Raf.) rootstock, and xyloporosis symptoms were observed on grafted trees of a tangerine hybrid. Psorosislike symptoms and concave gum symptoms were observed on budded grapefruit and tangerine trees, respectively. These diseases were observed only on budded trees of foreign origin at experiment stations and not in commercial plantings.

Fungal and bacterial diseases. The severity of fungal diseases in the Yungas and the Chapare is indicated in Table 2. Phytophthora foot and root rot was the most serious disease. Because seedling sweet orange, tangerine, and grapefruit trees are highly susceptible to the disease, many young trees were killed and older trees were weak and unproductive. The high rainfall and fine-textured soils create ideal conditions for *Phytophthora* spp. A bark rot, known as pink disease, was particularly serious on tangerine trees in the Yungas.

Greasy spot and melanose, foliar diseases of concern in humid, subtropical areas such as Florida, were absent or unimportant in the tropical areas of Bolivia. In the Chapare, areolate leaf spot was the most serious foliar disease, causing severe necrosis and leaf drop on grapefruit and less damage on oranges and tangerines. Cleopatra mandarin seedlings in seedbeds were severely defoliated, but extension agents reported good control with benomyl. In the cooler, drier Yungas, Septoria leaf spot was the most common foliar disease. Like areolate leaf spot, it was most severe on grapefruit and less so on oranges and tangerines. Lime anthracnose was relatively severe on Mexican limes in the Yungas.

Bacterial canker, which is caused by

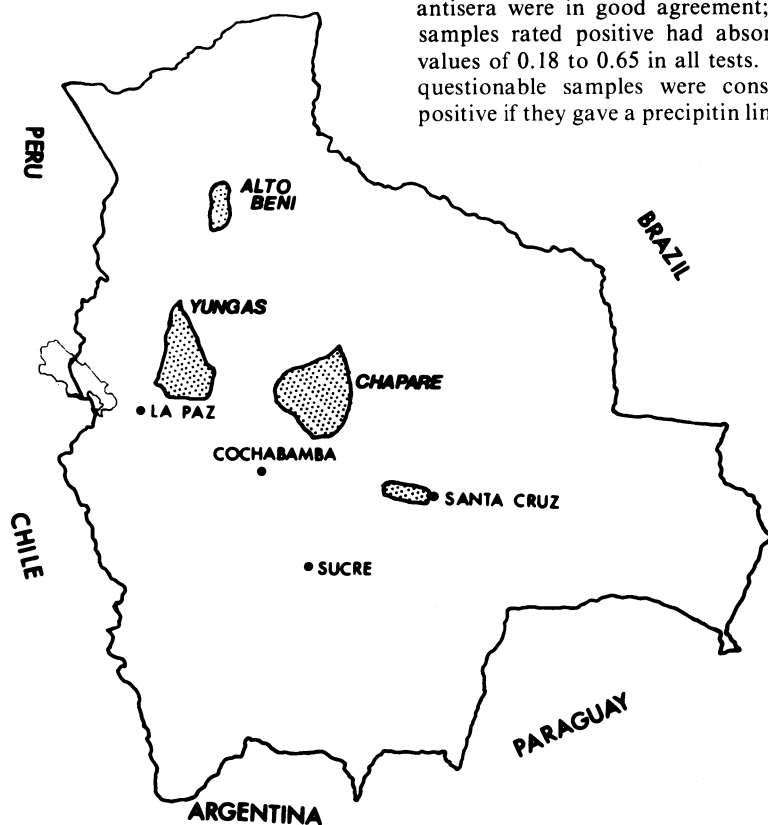


Fig. 1. Citrus producing areas of Bolivia that were surveyed for disease.

Table 1. Citrus tristeza virus in citrus species and hybrids in different areas of Bolivia

Species or hybrid	Number of plants (positive/sampled)			
	Yungas	Chapare	Santa Cruz	Total
Sweet orange (<i>Citrus sinensis</i>)	13/23	18/20	4/6	35/49
Mexican lime (<i>C. aurantifolia</i>)	3/7	7/7	...	10/14
Grapefruit (<i>C. paradisi</i>)	2/3	6/6	3/4	11/13
Tangerine (<i>C. reticulata</i>)	0/5	1/2	7/8	8/15
Tangelo (<i>C. reticulata</i> × <i>C. paradisi</i>)	...	1/1	...	1/1
Lemon (<i>C. limon</i>)	1/2	1/2
Sweet lime (<i>C. limettioides</i>)	1/5	1/1	2/2	4/8
Rough lemon (<i>C. jambhiri</i>)	2/2	...	1/1	3/3
Cleopatra mandarin (<i>C. reticulata</i>)	0/2	1/3	...	1/5
Sour orange (<i>C. aurantium</i>)	0/2	1/2	...	1/4
Volkamer lemon (<i>C. volkameriana</i>)	0/1	0/1
Swingle citrumelo (<i>Poncirus trifoliata</i> × <i>C. paradisi</i>)	0/1	0/1
Total	21/51	36/42	18/23	75/116

Xanthomonas citri (Hasse) Dowson, was not observed on citrus in the Yungas or the Chapare.

DISCUSSION

Citrus tristeza virus was found to be widespread in the major citrus areas of Bolivia. With the high populations of *T. citricida*, nearly all the citrus in the country could become infected soon. Although CTV and the vector are common in the Yungas, many trees remain free of the virus. The virus probably has spread more slowly there because citrus groves are widely separated on mountainsides and in valleys. In the Chapare and in Santa Cruz, where CTV is more abundant, groves and dooryard plantings cover much of the area.

Because citrus areas of Bolivia are not contiguous with production areas in neighboring countries, the virus was probably introduced with imported citrus trees or budwood many years ago. Tristeza now occurs throughout South America, with the exception of some areas of Chile (5).

CTV and other viruses have caused little damage to citrus production in Bolivia. Tristeza decline, such as that affecting CTV-infected sweet orange on sour orange rootstock, has not occurred in Bolivia because most of the trees are unbudded seedlings. Waite (4) correctly presumed that tristeza was widespread but attributed much of the decline, defoliation, and root rot to CTV and the bark scaling at the base of the trunk to exocortis. We believe that most of those symptoms are caused by *Phytophthora* foot and root rot.

Stem pitting caused by CTV was absent or mild even in highly susceptible Mexican lime, grapefruit, and Pera sweet orange. Warm temperatures, which suppress tristeza symptom development, occur all year in the Chapare, but temperatures are sometimes more favorable in the Yungas. Because severe

Table 2. Severity of fungal diseases of citrus in two areas of Bolivia

Disease	Causal organism	Disease severity ^a	
		Yungas	Chapare
Foot and root rot	<i>Phytophthora</i> spp.	+++	+++
Pink disease	<i>Corticium salmonicolor</i> Berk. & Br.	++	--
Areolate leaf spot	<i>Pellicularia filamentosa</i> (Pat.) Rogers	--	++
Septoria leaf spot	<i>Septoria citri</i> Pass.	++	--
Scab	<i>Elsinöe fawcetti</i> Bitanc. & Jenkins	+	++
Melanose	<i>Diaporthe citri</i> (Fawc.) Wolf.	+	--
Greasy spot	<i>Mycosphaerella citri</i> Whiteside	+	--
Lime anthracnose	<i>Gloeosporium limetticolum</i> Clausen	++	+

^a+++ = severe damage; ++ = damage of economic importance on susceptible varieties; + = symptoms observed, but of little economic importance; -- = no symptoms observed.

pitting occurs in Brazil and Peru under warm conditions (G. Müller, *personal communication*), its absence in Bolivia may be ascribed to mild, nonpitting strains rather than to unfavorable environmental conditions.

Our observations on fungal diseases agree in general with those of Waite (4), although he believed *Septoria* leaf spot to be of minor importance. The A strain of the bacterial canker pathogen spread recently from Brazil into northern Argentina and Paraguay, but it apparently has not yet reached the major Bolivian citrus areas.

Growers in Bolivia are being encouraged to use budded citrus. Rootstocks should be selected for resistance to *Phytophthora* root rot and tristeza. Budwood source trees infected with viruses other than CTV should be destroyed, and propagations should be made only from selected imported or indexed local material.

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