
New Whitefly-Vectored Closterovirus of Tomato in Florida 96-05G. W. Simone, R. C. Hochmuth, G. C. Wisler, J. E. Duffus, H. Y. Liu, and R. H. Li¹**Introduction**

A new viral-caused disease of tomato was identified in January 1996 from the greenhouse-grown tomato industry in north central Florida. This virus represents a previously undescribed member of the Closterovirus genus of viruses and has been designated Chlorosis Virus (ToCV) (10). This diagnosis has finally provided an answer for the long-standing malady known as 'yellow leaf disorder' that has existed in greenhouse-tomato production sites within Florida since as early as 1989 (unpublished data, G.W. Simone).

The greenhouse vegetable industry comprised some 66 acres of production in 1991, 32% of which was tomato production (7). These sites are scattered throughout Florida from Escambia County in the northwest to Dade County in the southeast. The appearance of 'yellow leaf disorder' between 1985-1995 was generally correlated to use of a contaminated fungicide (or its residual action) by most growers. Observation of 'yellow leaf disorder', however, in field tomato production in north Florida and in new greenhouse production sites without a history of the suspect fungicide seems to negate the toxic fungicide explanation for this malady. No examination of nutrient levels by direct analysis or through plant tissue analysis revealed any significant macro- or minor element imbalances. The potential occurrence of autogenous necrosis in particular tomato cultivars (8) was pursued and discounted. Examination of plant samples for plant pathogens was also repeatedly negative. Such techniques as plant virus inclusion examination by light microscopy, electron microscopy, serology and mechanical transmission to bioassay host plants yielded no evidence of plant viruses.

The active research by USDA-ARS scientists at Salinas CA on a new Closterovirus of tomato (Tomato Infectious Chlorosis Virus - TICV) prompted submission of symptomatic plant samples from Florida to the USDA-ARS staff in January 1996. Utilizing the greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood), successful transmission of the unknown agent into *Physalis wrightii* and *Nicotiana clevelandii* spp. was obtained (10). These species proved to be superior hosts for the virus, allowing detection of long flexuous rods conforming to the size range of a Closterovirus.

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Symptomatology

Onset of widespread ToCV symptoms in greenhouse grown tomatoes seems linked to late winter and early spring (Feb - Apr). Affected plants develop a progressive interveinal chlorosis. First symptoms in Florida greenhouse tomatoes often appear on leaves in the mid-section of the plant. The oldest leaves are pruned off of the plant, giving the appearance the symptoms initially started on the oldest leaves. The interveinal chlorosis is not uniform on either side of the leaflet midrib or consistent among leaflets on a leaf. Symptom expression develops over a period of months, yet continues to lag 1-2 leaves behind the meristem. Chlorotic zones may develop maroon-to-brown necrotic flecks. Virus impact on greenhouse tomato cultivars is most serious in loss of photosynthetic area due to the pronounced foliar yellowing. Plants enter a long period of general decline. Observations indicate neither flower abortion, fruit abnormalities, nor irregular ripening occurs with greenhouse tomato cultivars. Fruit size is reduced as symptoms expression becomes severe at the end of the season. Although development of viral symptoms in greenhouse tomato is slow, virus spread within the greenhouse proceeds effectively in the presence of the whitefly vector. Infection of all plants in a greenhouse is not uncommon.

Tomato Chlorosis Closterovirus

The Closterovirus and Luteovirus genera are the two genera responsible for causing "yellow-type" diseases across many crops. Florida tomato producers have encountered one such virus with the 1977-79 outbreak of Tomato Yellow Top Luteovirus (11) (Luteovirus). A Closterovirus from tomato has been reported by McGovern et al (9) but the symptoms appear distinct from ToCV.

Closteroviruses are long, flexuous rods with a range in size from 700-2,000 nm in length. Definitive members of the Closterovirus group possess a single stranded RNA and represent viruses with poor or no mechanical transmissibility. Vector insect species that transmit Closteroviruses include aphids, pseudococcid mealy bugs, and whiteflies in a semi-persistent manner. Closteroviruses known to occur in Florida (1) include citrus tristeza virus (aphid vectored), alligator weed stunting virus, carnation necrotic fleck virus, Dendrobium necrotic vein virus, and Nandina stem pitting virus.

The ToCV addition to the Closterovirus genus is unique. This virus is bipartite, with an 850 nm particle length, and single-stranded RNA. This virus does not appear related to other Florida closteroviruses based upon the vector and particle morphology. ToCV seems most closely related to tomato infectious chlorosis virus (3,4) and lettuce infectious yellows virus (2) from California based on the bicomponent genome.

Transmission

As with the majority of the members of the virus genus, *Closterovirus*, ToCV has not proven to be mechanically transmissible to an indicator host plant. Transmission of ToCV in the field and greenhouse is reliant upon the whitefly vector. Initial studies (10) indicated the greenhouse whitefly (*Trialeurodes vaporariorum* (Westwood)) was the vector for ToCV. Since this initial research, this new virus has proven quite unique among Closteroviruses by being effectively vectored by four species of whitefly (10): the greenhouse whitefly, the sweet potato whitefly (*Bemisia tabaci* (Gennadius)), the silver whitefly (*B. argentifolii*, Bellows and Perring) and the banded-wing whitefly (*Trialeurodes abutilonea* (Halderman)). These vectors transmit ToCV in a semi-persistent manner retaining the virus for 1-2 days. Vector acquisition, transmission, efficiency and transmission threshold studies are in process. All four vector species of whitefly occur in either field or greenhouse sites in Florida.

Distribution and Host Range

Distribution of ToCV in Florida is largely based upon conjecture. Prior to February 1996, reports of this problem were confirmed by negative diagnostic assay data and obvious symptoms known widely as 'Yellow Leaf Disorder'. Plant disease clinic reports indicate the earliest likely ToCV reports from Columbia and Suwannee counties in 1989. Frequency and incidence of these suspected ToCV infected samples are summarized in Table 2. The high incidence of these suspected ToCV infected samples in 1991-1993 reflects the intensive search for the cause of the symptoms. Low numbers in 1994 and 1995 reflect the unsuccessful history of detecting the cause through traditional pathological techniques.

Host range investigation of ToCv has only just begun. Studies with one or more of the known whitefly vectors have produced an initial host range listed in Table 2(10). Known hosts include representatives of the *Amarantaceae*, *Chenopodiaceae* and *Solanaceae* families. Representatives of the *Asteraceae* (*Compositae*, *Cucurbitaceae* and *Malvaceae* were negative thus far. Two hosts of importance are tomato and tobacco. Based upon symptomatology, the cultivars Apollo, Belmondo, Bounty, Caruso, Cobra, Correct, Floramerica, Jumbo, LM-200, Laura, Match, Medallion, Panther, Trend, Tropic, Trust, and Walter used in greenhouse tomato production between 1989-1996 are susceptible. The impact of ToCV on field tomato cultivars like Sunny (~16% of Florida's 1994-1995 acreage (6)) is yet to be determined. To date, no symptoms have been observed on cucumber and bell pepper grown as companion crops with greenhouse tomatoes exhibiting severe ToCV symptoms.

ToCV Impact on Florida

To date, the economic impact of ToCV has been limited to the greenhouse tomato industry. Distribution seems limited to the north central Florida area but this is

probably due to the present lack of diagnostic technology to identify this virus (when present) in plant tissue. The potential of ToCV is considerable. This Closterovirus is quite unique in possessing tremendous vector flexibility; utilizing the sweet potato, silver leaf, banded-wing, and greenhouse whiteflies for dissemination. The limited host range information to date suggests a small host range (*Solanaceae*, *Chenopodiaceae*, and *Amaranthaceae*) but this may change rapidly with future collaborative research between Florida and California.

The existing occurrence of ToCV resides in the midst of the 7,200+ A of tobacco (5) and contiguous to the 3,650 A of field tomatoes in the west, north, and north central Florida production zone (1994-1995) (6). The greenhouse tomato crop is initiated 4-6 weeks after tobacco harvest is complete in this area and simultaneous to the fall tomato crop acreage in north Florida. Although tobacco harvest is complete by mid summer, stripped stalks may persist in fields through August or even September. The persistence of both virus and its vectors seems assured (even without weed hosts) in the north central area through the overlap of the greenhouse tomato crop with both the spring and fall field tomato crops as well as the overlap of tobacco (crop or stubble) between greenhouse production cycles. In addition, this geographical zone (5) is host to an increase in cotton acreage (~109,000 A, 1995) which may alter the incidence of the banded-wing whitefly in the presence of a preferred host - cotton.

Present efforts are directed to define weed hosts of importance in north central Florida, further define the economically important host range, evaluate tomato cultivar reaction to this virus, and develop a diagnostic method to quickly and accurately define the incidence of ToCV.

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Table 1. Distribution and incidence of Tomato Chlorosis Virus (ToCV) in Florida based upon symptomatic samples received at the Florida Extension Plant Disease Clinic between 1989-1996.

County	Year of Occurrence							
	1989	1990	1991	1992	1993	1994	1995	1996 ¹
Alachua	--	1	--	8	--	--	--	--
Baker	--	1	1	2	1	1	--	--
Columbia	2	--	15	--	--	1	--	--
Marion	--	--	--	--	--	--	--	3
Suwannee	3	1	1	8	9	--	--	7
TOTAL	5	3	17	18	10	2	0	10
8 year total = 65 samples								
¹ 1996 samples were vectored with ToCV by whitefly transmission to bioassay host plants.								

Table 2. Present Known Host Range for Tomato Chlorosis Virus (ToCV).

<u>Hosts</u>	<u>Host Reaction</u>
Capsicum annuum 'Jalapeno'	-
Chenopodium capitatum	+
Cucumis melo 'Top Mark'	-
Cucumis sativus	-
Cucurbita pepo	-
Datura stramonium	-
Gomphrena globosa	+
Lactuca sativa	-
Lycopersicon esculentum	+
(Celebrity, Cherry, Jackpot, Moneymaker, Peto-19, Sunny, Trust, Valerie)	
Malva sp.	-
Nicotiana benthamiana	+
N. clevelandii	+
N. glutinosa	+
N. megalasiphon	+
N. tobacum (Burley 21)	+
<u>Physalis wrightii</u>	+

¹Host reaction: (+) = susceptible, (-) = immune.