Plant Health Progress

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PDF version for printing

Accepted for publication 17 May 2010. Published 7 July 2010.

Impact Statement

First Report of *Groundnut ringspot virus* Infecting Tomato in South Florida

Craig G. Webster, USDA-ARS, United States Horticultural Research Laboratory, Fort Pierce, FL 34945; **Keith L. Perry** and **Xiaoyun Lu,** Department of Plant Pathology and Plant-Microbe Biology, Cornell University, Ithaca, NY 14853; **Loren Horsman, Galen Frantz,** and **Charles Mellinger,** Glades Crop Care Inc., Jupiter, FL 33458; and **Scott Adkins,** USDA-ARS, United States Horticultural Research Laboratory, Fort Pierce, FL 34945

Corresponding author: Scott Adkins. scott.adkins@ars.usda.gov

Webster, C. G., Perry, K. L., Lu, X., Horsman, L., Frantz, G., Mellinger, C., and Adkins, S. 2010. First report of *Groundnut ringspot virus* infecting tomato in south Florida. Online. Plant Health Progress doi:10.1094/PHP-2010-0707-01-BR.

For about a decade, symptoms typical of those induced by tospoviruses have been sporadically observed in fresh market tomatoes in south Florida. During this period of time, *Tomato spotted wilt virus* (TSWV) infections have been confirmed in some but not all samples from such symptomatic tomatoes. From November 2009 through February 2010 these tospovirus-like symptoms were again seen in tomatoes in and around Homestead, FL. Foliar symptoms included necrotic flecks and spots, irregular chlorotic areas and deformation of leaflets (Fig. 1). Necrotic lesions on the epidermal tissue of stem and petioles were also observed (Fig. 2).

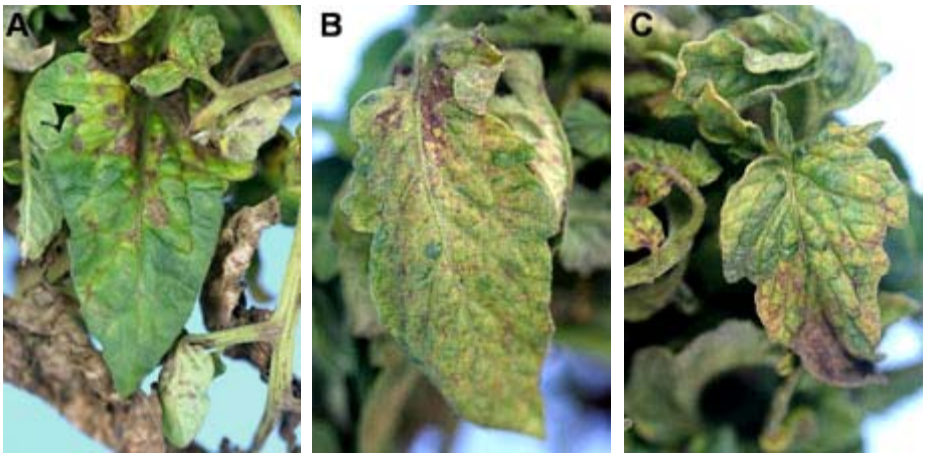

Fig. 1. Typical tospovirus symptoms observed on leaflets of tomato plants infected with *Groundnut ringspot virus* included necrotic spots (**A**) and flecks (**B**), irregular chlorotic areas (**A**, **B**, and **C**), and deformation (**C**).

Fig. 2. Necrotic lesions on epidermal tissue of the stem (**A**) or petioles (**B**) of tomato plants infected with *Groundnut ringspot virus*. Leaf deformation (inward rolling) of leaflets (**B**) was also seen.

Nine, two, and thirteen samples were collected from tomato plants showing these typical tospovirus symptoms in December 2009, January 2010, and February 2010, respectively, for subsequent testing. Reverse transcription-polymerase chain reaction (RT-PCR) using primers TSWV722 and TSWV723 (1) amplified a fragment of the expected size from total nucleic acid [extracted using Trizol Reagent (Invitrogen Life Sciences, Carslbad, CA)] from one February sample. The sequence of a portion of the nucleocapsid (N) gene from this fragment showed > 95% nucleotide identity to TSWV N-gene sequences in GenBank demonstrating the presence of TSWV in this sample. No product was amplified with these primers from the December or remaining February samples.

However, the presence of a tospovirus was indicated in many samples from December and February using commercially available double antibody sandwich-enzyme linked immunosorbent assay (DAS-ELISA) reagents (BIOREBA, Reinach, Switzerland). Four February samples reacted with commercially available DAS-ELISA reagents (Agdia, Elkhart, IN) which detect both *Groundnut ringspot virus* (GRSV) and *Tomato chlorotic spot virus* (TCSV). *Iris yellow spot virus*, *Impatiens necrotic spot virus* (INSV), *Groundnut bud necrosis virus*, and *Watermelon silver mottle virus* were not detected by additional DAS-ELISA testing using commercially available reagents (Agdia).

Similarly, the presence of a tospovirus was indicated in both January samples using an oligonucleotide macroarray designed for the detection of solanaceous plant pathogens. This is an expanded version of an array for detection of potato viruses (2). The macroarray positive January samples yielded hybridization signals for probes with sequences specific for TCSV, INSV, and TSWV, with the majority of positive signals coming from TCSV sequences. The array did not contain GRSV specific sequences.

RT-PCR using primers J13 and UHP for broad spectrum tospovirus detection (5) amplified several fragments from each February sample. A 697 bp fragment of the N-gene was sequenced from two February samples and showed 93.7 to 96.7% nucleotide and 97.4 to 98.7% amino acid identity to GRSV sequences in GenBank confirming the presence of GRSV. Lower nucleotide (81.1 to 83.4%) and amino acid (85.3 to 90.1%) identity was observed with TCSV sequences in GenBank. Comparable results were obtained from one of the January samples using these same primers further supporting the presence of GRSV. The single TSWV infected February sample described above was found to also be infected with GRSV by this same approach.

GRSV was originally described from peanut (groundnut) in South Africa and tomato in Brazil but has more recently been reported infecting peanut in Argentina and soybean in South Africa [reviewed in (7)], and sweet pepper (4), cocona [*Solanum sessiliflorum*, (3)] and coriander (6) in Brazil. The relatively

narrow reported host range of GRSV is in contrast to the extremely wide host range of TSWV.

GRSV is transmitted exclusively by thrips including the western flower thrips (*Frankliniella occidentalis* Pergande), *F. schultzei* Trybom, and *F. gemina* Bagnall [reviewed in (7)]. The virus must be acquired by larval thrips for subsequent transmission as adults. Both *F. occidentalis* and *F. schultzei* were observed in December and February on tomato plants in the same fields from which symptomatic plants were collected. However, it is not yet known which species of thrips are responsible for GRSV transmission in Florida.

To the best of our knowledge this is the first report of GRSV in the United States in any host although TSWV has been present in Florida for many years. Although the detection of GRSV in Florida tomatoes is cause for concern, the close relationship of GRSV and TSWV may allow successful adaptation of the integrated disease management strategies currently in use for TSWV in tomatoes for management of GRSV.

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