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Disease Notes

First Report of *Tomato yellow leaf curl virus* in Tomato in Costa Rica

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One of the most important invasive and harmful members of the genus*Begomovirus* (family *Geminiviridae*) is the monopartite *Tomato yellow leaf curl virus* (TYLCV), which is widespread over the world associated with tomato yellow leaf curl disease (TYLCD). Tomato (*Solanum lycopersicum*) plants infected with TYLCV show upward leaf curling and yellowing. In Latin America, isolates of TYLCV have been reported from Cuba, the Dominican Republic, Mexico and Puerto Rico (1), Guatemala (GenBank Accession No. GU355941), and Venezuela (partial genome sequence DQ302033). In Costa Rica, only isolates of the bipartite begomoviruses *Tomato leaf curl Sinaloa virus* (TLCSiV) (3) and Tomato yellow mottle virus (KC176780, KC176781) have been reported infecting tomatoes. During a survey conducted in 2012, similar begomovirus-like symptoms (leaf yellowing and upward leaf curling) were observed in tomato plants of five commercial growing areas in the Central Valley (Grecia region) of Costa Rica. In total, 65 tomato samples were randomly collected, 14 from greenhouses and 41 from open fields. Symptoms of upward leaf curling and yellowing were observed in three samples. Total DNA was extracted from collected samples and tested by dot blot hybridization using a probe to the coat protein (CP) gene of a Guatemalan isolate of *Bean golden yellow mosaic virus* (3). Only the three symptomatic samples tested positive, which represents an incidence of 14% (2 samples) in greenhouses and 2.4% (1 sample) in open field crops. These samples were subjected to rolling circle amplification (RCA) for viral circular genome amplification (2). The amplified products were then digested with *Msp*I restriction endonuclease, which resulted into DNA fragments of 2,320 and 458 bp for all three samples. This suggested infection with a monopartite begomovirus. In order to obtain the full-length clone, the RCA product of two samples (5240 and 5241) was digested with *Bam*HI, and the ~2.8 kb DNA fragment was cloned into pBluescript II SK(+) (Stratagene, La Jolla, CA) vector. After transformation of *Escherichia coli* DH5α, recombinant plasmids with inserts of expected size were selected and the insert was sequenced by primer walking (Macrogen Inc., Korea). The inserts of three clones from the two samples (CR:5240-16:2012, CR:5240-17:2012, and CR:5241-14:2012) were sequenced (deposited in GenBank as KF533855, KF533856, and KF533857, respectively). Sequences were all 2,781 nt long and shared 100% identity between themselves (1-nt mismatch between CR:5240-16:2012 and CR:5240-17:2012, and CR:5240-16:2012 and CR:5241-14:2012; and 2-nt mismatches between CR:5240-17:2012 and CR:5241-14:2012) and 99% with the sequence of *Tomato yellow leaf curl virus*-Israel[Japan:Haruno:2005] (TYLCV-IL[JR:Har:05]) (AB192966). These sequences represented full length genomes of isolates of the monopartite begomovirus TYLCV-IL and grouped in a phylogenetic clade (4) that comprised TYLCV-IL isolates reported from Asia (China and Japan) and from Mexico, while more distantly related to the clade comprising TYLCV-IL isolates reported from Central America (Cuba, Guatemala, Puerto Rico) and the United States, suggesting a distinct introduction event in Costa Rica. This is the first report of the presence of TYLCV in Costa Rica, therefore it is imperative to study the incidence and geographical spread of this virus in the country as well as its genetic diversity, since TYLCV infections might lead to significant yield losses, as reported in other countries.

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