Disease Note

Diseases Caused by Viruses

Q:1 **First Report of Cucurbit Chlorotic Yellows Virus Infecting Cucumber and Zucchini in Algeria**

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Q:3 Cucurbit chlorotic yellows virus (CCYV, *Crinivirus*, *Closteroviridae*) is a single-stranded, positive-sense plant RNA virus composed of RNA1 and RNA2. It is transmitted by whitefly *Bemisia tabaci* biotypes MEAM1 and MED in a semipersistent manner (Li et al. 2016). CCYV was first reported on melon plants in Japan in 2004 (Gyoutoku et al. 2009). Later, it was reported on many other cucurbits and several noncucurbit species in countries including China, Taiwan, Sudan, Lebanon, Iran, Greece, Turkey, Egypt, and Saudi Arabia (Kawazu et al. 2018). Recently, CCYV was reported in Israel (Luria et al. 2019) and in the New World, concretely infecting melon in California (Wintermantel et al. 2019). In 2018 and 2019, zucchini (*Cucurbita pepo*), cucumber (*Cucumis sativus*), and melon (*Cucumis melo*) plants showing virus-like symptoms were observed in northeast Algeria. Whiteflies were observed in all investigated fields. The observed symptoms, including foliar yellowing and vein clearing, were similar to those caused by whitefly-transmitted viruses such as the begomovirus (*Geminiviridae*) tomato leaf curl New Delhi virus (ToLCNDV), the ipomovirus (*Potyviridae*) cucumber vein yellowing virus (CVYV), and the criniviruses (*Closteroviridae*) CCYV and cucurbit yellow stunting disorder virus (CYSDV). To investigate the etiology, 69 symptomatic leaf samples (zucchini, 43; cucumber, 18; melon, 8) collected in field crops from four agricultural areas in Biskra and its surroundings (Biskra, El Ghrous, Sidi Okba, and Zribat el Oued) and 13 asymptomatic samples from pumpkin (*Cucurbita maxima*) collected from Sidi Okba were analyzed by tissue printing (Aparicio et al. 2009). Freshly cut leaf petioles were directly pressed onto nylon membranes and hybridized with individual riboprobes corresponding to the complete coat protein (CP) gene of these four whitefly-transmitted Q:4 viruses. Of 69 symptomatic samples, 58 were positive for ToLCNDV (zucchini, 38; cucumber, 12; melon, 8) and 5 for CCYV (zucchini, 3; cucumber, 2); CVYV and CYSDV were not detected. CCYV was detected only in mixed infections with ToLCNDV, which was recently first reported in this area (Kheireddine et al. 2019). Mixed CCYV-ToLCNDV infection occurred in 1 sample from El Ghrous (zucchini) and 4 from Sidi Okba (zucchini, 2; cucumber, 2). To confirm CCYV, total RNA extracts were obtained with TRIzol reagent (Thermo Fisher Scientific) from the original zucchini (3) and cucumber (2) samples and from asymptomatic samples (2) and reverse transcribed using random primers. Based on the complete CCYV sequence (AB523788 and AB523789 for RNA1 and RNA2, respectively) (Okuda et al. 2010), three pairs of primers were designed. RdRp-up (5′-CCTAATATTGGAGCT TATGAGTAC-3′)/RdRp-do (5′-CATACACTTTAAACACAACCCCCT -3′) amplified a portion of the RNA dependent RNA polymerase (RdRp) Q:5 region (754 bp) of RNA1, and Hsp-up (5′-TGCGTATGTCAATGGTGT TATG-3′)/Hsp-do (5′-ATCCTTCGCAGTCAAAAACC-3′) and CP-up (5′-ATGGAGAAGACTGACAATAAAC-3′)/CP-do (5′-TTATTTACTA CAACCTCCCGGTGC-3′) amplified, respectively, a portion of the heat shock protein 70 homolog (Hsp70h) region (462 bp) and the complete CP gene (753 bp) of RNA2. PCR products of the expected sizes were obtained from symptomatic samples but not asymptomatic controls. PCR fragments were directly sequenced in both directions by Sanger sequencing at the Instituto de Biología Molecular y Celular de Plantas (UPV, Valencia, Spain). BLAST analysis indicated that symptomatic samples shared identical amplicon sequences; therefore, only the sequences of a CCYV isolate from zucchini (CCYV-Sidi) were deposited in GenBank as MN529558 (RdRp), MN529559 (Hsp70h), and MN529560 (CP). The RdRp, Hsp70h, and CP sequences demonstrated >99% nucleotide identity with the respective genes of CCYV isolates from Greece, Asia, Africa, and California. This is the first report of CCYV in Algeria. CCYV could represent a serious threat for valuable cucurbit crops in the Mediterranean basin. It is necessary to implement efficient control measures to prevent further spread and minimize yield losses.

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The author(s) declare no conflict of interest.

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