Diseases Caused by Viruses

First Report of Tomato Black Ring Virus on Tomato (*Solanum lycopersicum*) in Saudi Arabia

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Tomato is a popular vegetable crop that is cultivated worldwide. It is also one of the most important crops in Saudi Arabia. In 2017, the area in which tomato was grown in Saudi Arabia was estimated to be 13,317 ha and produced 306,389 t. Al Kharj Governorate in Riyadh region contributes the highest production of greenhouse tomatoes in Saudi Arabia (Ministry of Environment, Water and Agriculture 2017). In fall 2015, striking virus-like symptoms (mottling, leaf rolling, yellowing, and deformation, black strip on the stem, cracking on fruits, deformation, mottling, and mummification with severe yield losses) were observed on greenhouse tomato plants in several farms in Al Kharj Governorate. Samples were collected within the period of fall 2015 and the summer of 2017. The collected samples were tested serologically using enzyme linked immunosorbent assay (ELISA) for identification of the causal agent(s) using kits and protocols from AC Diagnostics (Fayetteville, AR). Out of 18 common tomato viruses tested, 14 viruses were detected in tomato plants in the region. The greatest concern was the presence of tomato black ring virus (TBRV), because this was the first detection in Saudi Arabia and displayed the highest frequency of detection among all other detected viruses. Seventy-one out of the 135 tested samples were positive for TBRV. To confirm the presence of TBRV in the infected tomato samples, total RNA was extracted from positive samples and tested by RT-PCR with the newly designed primer pair F-TBRV (5'-GCAAAC-CAACGCTCTATGTTGT-3')/R-TBRV (5'-AGAGCCAACTGGAATGG-TAGG-3'), which is specific to the CP gene of TBRV. RT-PCR products of 978 bp in length were successfully obtained from the naturally infected tomato

plants. One of the detected isolates was used to inoculate *Chenopodium amaranticolor* with the aim of obtaining a pure isolate from single local lesions that could later be used for propagation and maintenance in *Nicotiana tabacum*. A host range experiment was conducted using mechanical inoculation with the single-lesion isolate of TBRV on four replicates of 14 different plant species in parallel with healthy controls (Brunt et al. 1996). Three weeks postinoculation, varying reactions and symptoms ranging from local lesion to plant death, depending on host species, were observed on the tested plants. Host range results were largely similar to those reported in previous studies (Rymelska et al. 2013; Sneideris et al. 2012). The presence of TBRV was confirmed both by ELISA and RT-PCR. Nucleotide sequences obtained from PCR products of selected samples were submitted to GenBank and assigned the following accession numbers: MT274656, MT274657, and MT274658. Saudi isolates of TBRV were found to share 99 to 100% of their nucleotide sequences. They had the highest similarity of 98% with the Polish isolates (MG458221 and KX977561) and the lowest similarity of 85% with isolates from Lithuania (KF678369 and KF678370). To the best of our knowledge, this is the first report of occurrence of TBRV in Saudi Arabia. Because this virus is transmitted by seeds, it may have entered through imported seeds and spread in greenhouses through mechanical means. A survey of the different agricultural regions is encouraged to determine the incidence, distribution, and damage induced by this virus in Saudi Arabia.

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