Molecular characterisation and first complete genome sequence of Tomato yellow leaf curl virus (TYLCV) infecting tomato in Iraq

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Tomato yellow leaf curl disease (TYLCD, genus Begomovirus, family Geminiviridae) is one of the factors that severely limits tomato production in Iraq, as it attacks tomato cultivated in both protected and open fields (Al-Fadhal, 2012). In Iraq, TYLCD incidence may reach up to 100% and can cause economic losses between 50-90%, especially when the virus infects tomato plants in the early growing stages (Al-Ani et al., 2011). In April 2010 leaf curling, yellowing and a reduction in leaf size symptoms were observed on tomato in fields in Baghdad, Iraq. A total of 49 tomato samples were collected and total nucleic acids were extracted from samples using the CTAB protocol described by Abashi et al. (2010). PCR amplifications of extracted nucleic acids were performed using Red Hot DNA polymerase (Thermo Scientific Inc., USA) and begomovirus specific primers (Deng et al., 1994). Rolling circle amplification (RCA) was performed using an Illustra Templiphi 100 Amplification Kit (GE Healthcare Limited, UK) according to the manufacturer’s protocol. Restriction digestion of RCA products with NcoI was performed to release the full-length fragment of the amplified viral DNAs, which were gel purified, cloned and sequenced (Bioscience Gene Service, UK). Based on PCR and sequence analyses using MEGA 5 software (Tamura et al., 2011), six out of 49 tomato samples (12%) were found to be infected by TYLCV. MEGA BLAST search of sequences amplified by Deng primers revealed that sequences isolated showed 98% maximum identity with Tomato yellow leaf curl virus GenBank sequences.

The full length sequence referred to as TYLCV IRQ is 2780 bp in length (GenBank Accession No. JQ354991). Sequence comparison of full length TYLCV-Iraq using MEGA BLAST search was performed against available GenBank sequences. TYLCV IRQ shared 99% identity with TYLCV isolates Mauritius (HM448447) from Mauritius, RE4 (AM440920) from Reunion Island and Almeria (AJ489258) from Spain. The primer pair ‘TYLCV-F/TYLCV-R’ (TYLCV-F, 5’-CAAGATAACAGAACAC AGCCA-3’) and TYLCV-R, 5’-GGATAAGCACATGGAGATGTG-3’ was designed from the TYLCV genome regions between 1394-2463 nucleotide positions. This primer could detect TYLCV IRQ isolate to amplify a 1.7 kb fragment from the TYLCV genome using different techniques for extraction and purification of Tomato yellow leaf curl virus (TYLCV). Journal of Baghdad for Science 8, 447-452 [In Arabic, English abstract].

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