

The Complete Nucleotide Sequence of a Taiwan Strain of Papaya Leaf Distortion Mosaic Virus

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ABSTRACT

The complete nucleotide (nt) and deduced amino acid (aa) sequences of Taiwan strain of Papaya Leaf Distortion Mosaic Virus – TW DL (PLDMV-TW DL) were determined. A total of seventeen upstream and downstream primers specific to PLDMV was designed according to the published full length PLDMV sequence of Japan isolate (PLDMV-J 56P, Accession No.AB088221) and the previously determined 1927 nucleotides of 3' region of Taiwan isolate. Reverse transcription- polymerase chain reactions (RT-PCR) were performed to amplify different regions of PLDMV-TW DL viral RNA. Eight overlapping cDNA clones that covered the whole genome of PLDMV-TW DL were obtained. The complete genomic RNA is 10153 nucleotides in length while the 5' leader sequence is 134 nucleotides and the 3' untranslated region (UTR) is 208 nt, excluding the 3' poly(A) tail. It contains one large open reading frame which encoded a polyprotein of 3269 amino acids with an M(r) of 372.895 kDa which is the fourth largest polyprotein in the reported Potyvirus genus. The nucleotide sequence and the deduced amino acid were compared with 40 different isolates of the genus Potyvirus. PLDMV-TW DL shows 94% of nucleotide identity to PLDMV Japan isolate and 53-57% of nucleotide identity to the other 39 potyviruses when compared with the complete potyviral sequences. Furthermore, NIb is the most conserved gene among the potyviruses while P1 and 3'UTR are the most variable ones. Phylogenetic trees of the 41 potyvirus isolates, including PLDMV-TW DL, were constructed using the aa sequences of polyprotein, NIb, CP and P1, respectively. It was found that PLDMV-TW DL and PLDMV-J 56P were closely related. In the host range determinative tests, PLDMV-TW DL was able to induce mosaic and leaf distortion symptoms on papaya plants while the other three species of Cucurbitaceae plants 【*Cucumis metuliferus*(Acc.2459), *Cucurbita pepo* (Diner), *Cucumis sativus* (Sagami-hansiro)】 were symptomless. Indirect enzyme-linked immunosorbent assay (Indirect-ELISA) and reverse transcription- polymerase chain reaction (RT-PCR) were further performed to confirm the latent infection in *C. metuliferus*(Acc.2459) and *C. pepo* (Diner). Virus particles were purified from the PLDMV-TW DL inoculated papaya plants and examined under the transmission electron microscope (TEM). Filamentous viral particles in about 780 nm long resemble to the size of potyvirus were observed. From the nucleotide and amino acid sequence comparison, phylogenetic analyses and host range determination, we conclude that PLDMV- TW DL is a new pathotype of PLDMV species and different from the Japanese isolate PLDMV-J 56P.

Keywords : Papaya leaf distortion mosaic virus (PLDMV) ; reverse transcription-polymerase chain reaction ; open reading frame ; Potyvirus ; phylogenetic analysis ; Indirect enzyme-linked immunosorbent assay ; taiwan ; Japan ; transmission electron microscope

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