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New record of *Papaya leaf curl virus* and Ageratum leaf curl beta satellite associated with yellow vein disease of aster in India

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An outbreak of yellow vein disease of aster (Kalimeris indica, family Asteraceae) was observed at Lucknow, India in the winters of 2010 and 2011. During a survey in winter 2012, 23% (56/234) of aster plants exhibited yellow vein disease symptoms at NBRI gardens in Lucknow and a higher disease incidence of 34% (125/367) was found in other gardens at Lucknow. All 181 infected plants exhibited highly similar yellow vein symptoms (Fig. 1) as compared to healthy ones. Presence of whiteflies in the vicinity and the typical yellow vein symptoms suggested a begomovirus infection. For diagnosis, total DNA was extracted from samples of five plants showing symptoms (collected from two locations) and from one symptomless plant. PCR was then performed using PALIc1960/PALIv722 begomovirus specific primer pair (Rojas et al., 1993) and beta-01/beta-02 beta satellite specific primer pair (Briddon et al., 2003). By contrast to the DNA sample of the symptomless aster plant, all the five samples from diseased plants showed PCR products of expected sizes for a begomovirus (~1.2 kb; Fig. 2a) and a beta satellite (~1.3 kb; Fig. 2b), which indicates their association with the disease.

For identification of the begomovirus, Φ -29 DNA polymerase-based rolling circle amplification (RCA)(TempliPhi kit, GE healthcare, USA) was performed followed by restriction digestions with *Bam*HI enzyme. A representative DNA sample of an infected aster was used as a template.The resulting fragment of ~2.8 kbwas cloned and sequenced (JQ954859). Analysis of the sequence data revealed 90-92% sequence identity with *Papaya leaf curl virus* (PLCV) strains (Fig. 3a) leading to identification of the isolate as a strain of PLCV. The ~1.3 kb fragment obtained with the beta satellite specific primers was also cloned and sequenced (JQ408217). This sequence showed the highest identity (93%) and closest phylogenetic relationships with Ageratum leaf curl beta C1

(AgLCB) strain (Fig. 3b). Several attempts to detect a DNA-B genome by PCR using its specific primers (Padidam *et al.*, 1995) or by restriction analyses of RCA products failed, which indicates the begomovirus as monopartite. A literature search revealed that *Potato virus Y* is the only reported virus infection so far in *K. indica* (Wang *et al.*, 2012). Thus, the association of PLCV and a beta satellite with yellow vein disease of *K. indica* is a new record. The disease is considered as significant, since aster plants are grown in gardens for ornamental purposes and the infected ornamental plants may serve as a virus reservoir for other hosts.

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Figure 1

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