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Occurrence of a DNA β-containing begomovirus associated with leaf curl disease of kenaf (*Hibiscus cannabinus* L.) in India

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Abstract. The association of a begomovirus, which has satellite DNA β, with leaf curl disease of kenaf has been detected for the first time.

Kenaf (*Hibiscus cannabinus* L.) is a potentially valuable industrial crop due to its fibre content, medicinal value and effective use in the paper industry (Duke 1983). The USDA recognises kenaf as the best non-woody paper-making plant. The crop is attacked by several viral diseases (Brunt *et al*. 1996; Jones and Behncken 1980). In recent years a disease causing leaf curl symptoms on kenaf has been observed in different parts of India. The infected plants showed curly leaves at early stages, and then gradually became distorted and puckered (Fig. 1). The height of the

Fig. 1. Leaf curl disease-infected kenaf in the field with a close view of symptoms in leaves (inset).

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infected plants was also progressively reduced. A survey was carried out at three villages located in the Bahraich district of Uttar Pradesh, India, and records were taken at 100 days after sowing. Incidence, disease severity (proportion of affected leaves) and plant height reduction were recorded. Leaves from plants showing typical symptoms were collected for molecular analysis.

Disease incidence, disease severity and height reduction averaged 22, 66, and 25%, respectively from a total 1203 plant surveyed. Transmission electron microscopy of typical symptomatic leaves of kenaf using 2% uranyl acetate revealed the presence of geminate particles measuring 18×27 nm. Total DNA obtained from infected leaves gave a strong Southern hybridisation signal after hybridisation with a *Cotton leaf curl Rajasthan virus* DNA A (GenBank accession number NC_003199) probe after stringent washing (three washes) with $2 \times$ SSC and 0.1% SDS at 65°C. The healthy samples did not give a hybridisation signal. Using PCR with universal DNA β (Briddon *et al*. 2002) and coat protein primers (Jose and Usha 2000), a ~1.3 kb fragment corresponding to DNA β and a ~0.77 kb fragment corresponding to the coat protein gene of DNA A were amplified from DNA samples obtained from 20 randomly selected plants from ten different fields showing leaf curl symptoms. This is the first record of a begomovirus, which has DNA A along with a satellite DNA β, being associated with leaf curl disease of kenaf in India.

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