First report of Pepper mottle virus infecting chilli pepper in India

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Chilli pepper (Capsicum annuum) is an important vegetable and spice crop in India. Leaf samples from plants exhibiting stunting, mottling, crinkling and misshapen leaves or malformed and discoloured fruits were collected from pepper-growing districts of the Punjab, India in 2013 and 2014. Crude sap was extracted from symptom-bearing leaves using 0.1 M phosphate buffer (pH 7), and used to mechanically inoculate five plants each of seven different plant species. Symptoms developed on C. annuum cv. ‘California Wonder’ (mild mosaic and leaf deformation), Solanum lycopersicum cv. ‘Punjab Upna’ (mild mosaic with slight yellowing), Nicotiana benthamiana, N. glutinosa, N. tabacum cvs. ‘White Burley’ and ‘Xanthi’ (mild mosaic symptoms), whereas no symptoms were observed on S. tuberosum.

The symptom-bearing leaf samples collected from different pepper-growing areas were analysed using DAS-ELISA kits (Agdia, Elkhart, USA) with commercial antisera specific for commonly occurring pepper viruses: Pepper mild mottle virus (PepMMoV), Pepper mottle virus (PepMoV), Potato virus Y (PVY O/C, N) and Cucumber mosaic virus (CMV; subgroups I and II). Of the 115 leaf samples, 34% tested positive with PVY O/C antisera, 8% with PepMoV antisera and 4% with CMV subgroup I and II antisera. The presence of the potyvirus was confirmed by RT-PCR using the P9502/CPUP (van der Vlugt et al., 1999) and the Sprimer/M4T (Chen et al., 2001) primer sets. Amplified products of the expected size were cloned and sequenced and the sequences obtained were deposited in GenBank (Accession Nos. KJ564302 and KJ744259). Multiple sequence alignment using Clustal Omega (http://www.ebi.ac.uk/Tools) showed that sequences KJ564302 and KJ744259 shared 95.1-97.8% and 94.87-99.47% nucleotide sequence identity respectively with published sequences of PepMoV (AF501591, AB126033, EU586121 EU586125 and EU 586124). Based on these results, we concluded that the virus isolated from the diseased pepper plants was PepMoV. Mottle disease of chili was characterised using the micro-precipitin test by Sandhu & Chohan (1979). This is the first time that two sensitive assays, ELISA and RT-PCR based partial sequence characterisation have been used to identify and confirm the occurrence of PepMoV in pepper from India. Further studies are required to fully characterise this isolate and determine its distribution and incidence.

References

Chen J, Chen J, Adams MJ, 2001. A universal PCR primer to detect members of the Potyviridae and its use to examine the taxonomic status of several members of the family. Archives of Virology 146, 757-766.
