

# Identification of *Peanut bud necrosis virus* and *Tomato spotted wilt virus* in Indonesia for the first time

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In surveys conducted in May 2008 in Warung Kondang, Cianjur, West Java, tomato plants (*Solanum lycopersicum*) growing in farmers' fields showed stunting and leaf symptoms of either bronzing or general chlorosis with vein-banding. In addition, chilli peppers (*Capsicum* spp.) were observed with chlorosis and vein-banding symptoms in Salabintana, Sukabumi, West Java. Finally, peanut plants (*Arachis hypogaea*) showing chlorotic rings and necrosis on leaves were observed in a field near to the Bogor Agricultural University Campus, Darmaga Bogor. All these symptoms suggested the possible involvement of tospoviruses.

To identify the viruses involved, leaves with symptoms were pressed onto FTA classic cards [FTA<sup>®</sup> Classic Card, Whatman International Ltd, UK] and shipped to Washington State University, with the appropriate permit from USDA-APHIS-PPQ. Nucleic acids were recovered from the cards using a sample extraction protocol (Alabi *et al.*, 2008) and tested by one-tube, single-step reverse transcription-polymerase chain reaction (RT-PCR) using degenerate 'universal' tospovirus primers [Upstream primer: 5'-CCTTTAACAGT(A/T/G)GAAACAT-3' and Downstream primer: 5'-CAT(A/T/G)GC(A/G)CAAGA(A/G)TG(A/G)TA(A/G)ACAGA-3'] conserved in the L RNA segment of tospoviruses (Chu *et al.*, 2001). A DNA fragment of approximately 800 base pairs was amplified from the tomato, pepper and peanut samples. Each was cloned separately into pCR 2.1-TOPO vector (Invitrogen Corp, Carlsbad, CA, USA). Three independent clones from each sample were sequenced in both directions. A comparison of the nucleotide sequences obtained from the tomato (GenBank Accession No. FJ 177301) and chilli pepper samples (FJ 177302) was made with corresponding sequences of tospovirus L-RNA deposited in GenBank. This analysis showed 96% and 97% nucleotide sequence identity with *Tomato spotted wilt virus* (TSWV) isolates from Hawaii, USA (AY070218) and Japan (AB198742), respectively. The sequences obtained from tomato and chilli shared only 93% identity between them, indicating that the Indonesian TSWV isolates from tomato and pepper may be distinct strains. An analysis of the nucleotide sequence obtained from the peanut sample (FJ177300) showed 94% sequence identity with corresponding L RNA sequence of a *Peanut bud necrosis virus* (PBNV) isolate from India (AF025538).

These results confirm for the first time that two distinct tospoviruses, TSWV and PBNV, occur in Indonesia.

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