

## Amazon lily: a new natural host of *Tomato spotted wilt virus*

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The Amazon lily (Eucharis grandiflora, Amaryllidaceae) is found in tropical rain forests and has been widely cultivated as an ornamental plant. Although viral infections do not usually kill these plants, Amazon lily mosaic virus, Hippeastrum mosaic virus, Nerine latent virus, Cucumber mosaic virus, Impatiens necrotic spot virus (INSV) and a member of the Anulavirus genus, tentatively designated as Amazon lily mild mottle virus, have been reported in Amazon lilies (Alexandre et al., 2011; Fuji et al., 2013). In the case of INSV, it is worth noting that the virus was originally identified in this host was described as the impatiens strain of TSWV (TSWV-I). This was subsequently reclassified as INSV and recognised by ICTV (International Committee on Taxonomy) as a distinct species (Murphy et al., 1995). Chlorotic rings and spots were observed on E. grandiflora plants from a commercial nursery in Atibaia county, São Paulo State, Brazil (Fig. 1). Disease symptom incidence was estimated at 60%. In order to identify the causal agent, transmission tests, electron microscopy (EM), RT-PCR and sequencing were carried out.

Symptoms were induced following mechanical inoculation on *Penunia hybrida*, *Chenopodium amaranticolor* and *Datura stramonium*. Up to four months after inoculation, *Eucharis* and *Hippeastrum* remained symptomless and the virus could not be recovered from these hosts. EM observations of negatively stained leaf extract from naturally infected *E. grandiflora* revealed the presence of a large number of quasispherical particles, approximately 100 nm in diameter. EM of thin sections of the leaf lesions also identified enveloped particles in the lumen of the endoplasmic reticulum (Fig. 2). Total RNA from *E. grandiflorum* and *D. stramonium* (inoculated host plants) was separately extracted and an approximately 450 bp fragment, corresponding to the non-translated region and partial nucleocapsid protein, was amplified by RT-PCR, using the pair of primers BR60 and BR65 (Eiras *et al.*, 2001). The amplicon was sequenced and the

sequence deposited in GenBank (Accession No. KC894720). Analysis indicated that this sequence shared the highest identity (96.5%) with isolates of *Tomato spotted wilt virus* (TSWV). Further analysis was conducted by construction of rooted trees (PAUP software) with different TSWV isolate nucleotide sequences from multiple regions of the world, including Brazil, and different hosts. These revealed that the *Eucharis* TSWV sequence (KC894720) shared a common ancestor with the Brazilian and Italian isolates (Fig. 3) but the TSWV isolates could not be separated by geographical origin and host. This is the first report of Amazon lily as a natural host to TSWV.

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Figure 1

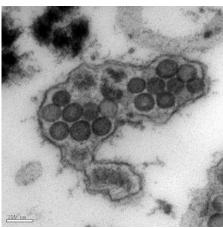


Figure 2

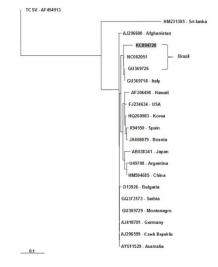


Figure 3

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