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Report of *Tomato yellow spot virus* **Infecting***Leonurus sibiricus* **in Paraguay and Within Tomato Fields in Brazil**

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Leonurus sibiricus L. (Lamiaceae) is a subtropical weed frequently found with golden mosaic symptoms. Leonurus mosaic virus (LeMV) was the first begomovirus reported on L. sibiricus in Brazil (3). Later, a new bipartite species (Tomato yellow spot virus, ToYSV) was reported affecting tomatoes, beans, and also *L. sibiricus* (1,2). A survey of begomovirus isolates was conducted within tomato fields also displaying high incidence of plants with begomovirus-induced symptoms. Thirty L. sibiricus and 33 tomato samples were collected (2007 to 2012) in nine districts in Paraná State, Brazil. Two L. sibiricus isolates were also obtained within citrus orchards in Major Otaño, Itapúa, Paraguay. Total DNA was extracted from all 65 isolates and PCR assays were conducted with primers for conserved DNA-A (PAL1v1978/PAR1c496) and DNA-B (PBL1v2040/PCRc1) regions (3). Nucleotide sequence identity of the 1,193-bp DNA-A amplicons of our L. sibiricus isolates ranged from 93.4 to 98.2% with LeMV (GenBank Accession No. U925321) and from 92.4 to 94.8% with ToYSV isolates from tomato (DQ336350.1) and bean (FJ538207). None of the 33 tomato samples was found to be infected by ToYSV, with all having high nucleotide sequence identity (92 to 99%) only with Tomato severe rugose virus (GU358449). Complete DNA-A genome sequence was obtained via a rolling circle amplification-based strategy for one Brazilian L.

sibiricus isolate (PR-088) and one isolate from Paraguay (PAR-07). The entire DNA-A genome of PR-088 (JQ429791) had 96.8% nucleotide sequence identity with PAR-07 (KC683374) and ranged from 95.6 to 96.3% with ToYSV isolates from bean, tomato, and L. sibiricus(JX513952). The nucleotide sequence identity of the 487-bp DNA-B amplicon ranged from 87 to 92% among PR-088 (KC 683374); PAR-07 (KC740619) and ToYSV isolates from tomato (DQ336351.1) and L. sibiricus (JX513953.1). Leonurus cuttings infected with the ToYSV (PR-088) were caged together with healthy L. sibiricus and tomato 'Alambra' seedlings. Hybridization assays with ToYSV-specific probes (2) and sequencing of PCR amplicons indicated that *Bemisia tabaci* biotype B adults were able to transmit ToYSV to both hosts as reported (1). Our results suggest that L. sibiricus is the main ToYSV reservoir under natural conditions and tomato seems to be an occasional alternative host. In fact, ToYSV has not often detected in tomatoes as observed in a number of extensive surveys (4). So far, the complete LeMV genome is not available for comparison (3). However, our analyses with a DNA-A segment indicated that LeMV and ToYSV isolates might represent strains of single virus at the current threshold of 89% nucleotide sequence identity for Begomovirus species discrimination (4). Thus, a reappraisal of the taxonomic status of ToYSV is necessary to clarify its genetic relationship with LeMV. This is the first report of ToYSV on *L. sibiricus* in Paraguay.

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