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NEW DISEASE REPORT

First report of *Verbena latent virus* infecting giant goldenrod in Belgium

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Giant goldenrod (*Solidago gigantea*, *Asteraceae*) is a rhizomatous herbaceous perennial native to North America. Introduced to Europe in the 18th century as an ornamental, *S. gigantea* is now considered as highly invasive, exerting negative impacts on native plant communities (Weber & Jakobs, 2005; Pal et al., 2015).

In summer 2023, a survey was done in Belgium to better characterise the viruses infecting giant goldenrod. Fifty asymptomatic plants were collected from ten sites at the roadside and garden edges in the centre of Belgium (the Walloon and Flemish Brabant regions; five plants were collected per site) and pooled for molecular analysis by high throughput sequencing (HTS). Virus particles were purified, and virion-associated nucleic acids were extracted following the protocol described in Maclot et al. (2021). Library preparation was performed at GIGA (University of Liege, Belgium) with the NEBNext Ultra II DNA library prep kit (New England BioLabs, USA). The Illumina Novaseq platform (2×150 nt) was used for sequencing. A total of 6,263,188 trimmed reads (Bioproject PRJNA1088467) were *de novo* assembled using rnaviralSPAdes on the Galaxy.org server and viral contigs were identified by BLASTn and BLASTx against the GenBank database. One contig (8,571bp, average coverage 887) had 94.82% identity with a *Verbena latent virus* (VeLV, *Carlavirus*, *Betaflexiviridae*) isolate from Israel (GenBank Accession No. AF271218). VeLV has been detected in *Gynura aurantiaca*, *Tropaeolum majus* and *Verbena hybrida* but its genome has not been sequenced (Cohen et al., 2003; Lebas et al., 2005; Ochoa-Corona et al., 2010). The genome generated in this study (PP502869) is the first including all known six ORFs from carlaviruses. HTS found two other novel plant-associated virus-like sequences in the sample (*Genomoviridae* family, PP502870-PP502871), and small sequences of unknown mycoviruses (*Partitiviridae*, *Totiviridae*).

To confirm VeLV infection, total RNA extracted from the pooled sample was tested by RT-PCR using existing primers for amplification of the Triple Gene Block 1 (Ochoa-Corona et al., 2010), and newly designed primers for the viral replicase (Forward: 5′-CTCTCAAGCCTGTTGTTTAGGG-3′ / Reverse: 5′-ACCTGCATGAACCAACTTCC-3′; 639 bp amplicon) and coat protein (Forward: 5′- TCAATGATGCACAATCTGCGAA-3′ / Reverse: 5′-GCAGCCTCATTCTCGACATA-3′; 844 bp amplicon). Sequence analysis showed 99.9% nt identity between the amplicons and the HTS contig, and the amplicons matched the sequence of VeLV (96.2-97.4% nt identities with AF271218). We also tested the leaf samples individually and VeLV infection was detected by RT-PCR in only one site (four of five plants tested positive) of the 10 sites surveyed in 2023.

Because of VeLV's wide host range, the broad transmission profile of carlaviruses (via aphids, whiteflies, seeds or mechanically) and their potential pathogenicity in single or mixed infections, including synergistic effects (Untiveros et al., 2007)), it is important to better characterise the biology and symptomatology of VeLV. To our knowledge, this is the first report of VeLV infecting *S. gigantea* worldwide. Giant goldenrod is widespread in Belgium and Europe, and may represent a significant asymptomatic reservoir for VeLV.

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