New Disease Reports

First report of Mint vein banding-associated virus infecting *Mentha* × *gracilis* in Germany

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Mint has been used in traditional medicine for thousands of years and dried or fresh leaves are the source of mint flavour in cooking and teas. More recently, mint has also become established as a perennial bedding plant. In August 2016, symptomatic *Mentha* × *gracilis* (ginger mint) plants were offered for sale as potted herbs (Fig. 1) in a supermarket in Braunschweig, having been produced in a nursery in Papenburg (both Lower Saxony, Germany). Symptoms observed consisted of vein banding/clearing and chlorotic spots (Fig. 2) on many leaves. Mechanical transmission of a putative causal virus to herbaceous indicator hosts (*Chenopodium quinoa*, *Nicotiana benthamiana*, *N. hesperis*, *N. occidentalis* 37b, *N. occidentalis* P1 and *N. tabacum* Xanthi nc) was not successful. Leaves of one symptomatic mint plant served as template for dsRNA extraction, resulting in a high molecular weight dsRNA. The dsRNA was significantly larger than the largest marker fragment of 10 kb as observed after gel electrophoresis, indicating the presence of a clostero- or endornavirus. Viruses belonging to these genera are known to have the largest RNA genomes amongst plant-infecting viruses. Random RT-PCR, cloning and sequencing resulted in two viral sequences (651 and 1349 bp) showing the highest nt sequence identities to Mint vein banding-associated virus (MVBaV; GenBank Accession No. KJ572575). MVBaV is an unassigned species in the family *Closteroviridae*, of which only one partial genomic sequence is available. One sequence was located in the polymerase gene (position 3849-4500 nt) and had 80% nt / 88% aa sequence identity to MVBaV. The second sequence (position 11014-12366 nt) partially covered ORF4 (putative minor CP) and the entire ORF5 (encoding CP), to which it showed 87% nt / 95% aa sequence identity.

The sequence identity values are above the species demarcation threshold of 75% for aa sequences of relevant genes (CP, HSP70, polymerase), identifying this virus as a deviating isolate of MVBaV. This virus species was discovered in a survey in the USA more than ten years ago (Tzanetakis *et al.*, 2005). It was found in several different mint species and hybrids exhibiting similar foliage symptoms. Our report is the first for MVBaV outside the USA.

Based on the polymerase gene sequence obtained in this study, a primer pair (Mint-f 5′-CAAACCTGTTGGGTGTTCAGA-3′; Mint-r 5′-TTCCTCGTTAAACATATTTAGGA-3′) was designed to specifically test six individually sampled symptomatic plants (same propagation batch) as well as the plant originally used for dsRNA extraction. Amplicons of the expected size (586 bp) were obtained from all samples. Subsequent direct sequencing revealed that all were almost identical to the sequences originally obtained (>99.5% nt identity). The MVBaV isolate is available at the DSMZ plant virus collection under accession no. W16-112 and the two sequences obtained by shotgun cloning in this study were deposited in GenBank (KY381598-99).

References

Tzanetakis IE, Postman JD, Martin RR, 2005. A member of the *Closteroviridae* from mint with similarities to all three genera of the family. *Plant Disease* **89**, 654-658. http://dx.doi.org/10.1094/PD-89-0654

Figure 1

Figure 2

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