New Disease Reports

First record of *Ranunculus white mottle virus* from Australia

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In 2004, a vein yellowing disease of soil-grown greenhouse capsicum was observed in southern Australia (Fig. 1). Symptoms were similar to those of pepper yellow vein disease, a suspected viral disease of capsicum transmitted by soilborne *Olpidium* species (Fletcher *et al*., 1987). Disease symptoms were apparent on young plants and faded as the plant matured. In 2008, the disease was again detected in this region and was graft-transmitted to four capsicum plants which developed vein-yellowing symptoms. This isolate (2155) and a sample lyophilised in 2004 (isolate 1631) were tested by RT-PCR using the OP1 and OP2 primers specific for RNA-1 of the genus *Ophiovirus* (Varia *et al*., 2003). RT-PCR amplicons of the expected size (136 bp) were obtained from the suspected virus-infected samples but not from healthy capsicum.

A larger amplicon was amplified using the OP2 primer and a newly designed degenerate genus-specific primer OP3 (5´-TCDCAAACHCAAGTACAAATGGAAG-3´) in RT-PCR. The product was amplified from isolate 1631 and the amplicons cloned. Four clones were sequenced, and all were identical (GenBank Accesssion No. MN128532). This sequence was 97.5% identical to that of *Ranunculus white mottle virus* (RWMV, AF335429) using a pairwise nucleotide sequence alignment. ICTV criteria for classifying *Ophiovirus* species are based on coat protein sequence (http://www.ictv.global/report/aspiviridae; Garcia *et al*., 2017; 2018), however, there are no RWMV coat protein sequences available for comparison. Based on available sequence, the virus is identified as RWMV.

Specific primers, RWMVF1 (5´-CGAACATTCCATCTACGCCT-3´) and RWMVR1 (5´-GATAGACAATGCCGCAACAA-3´) were developed and used in RT-PCR to screen field samples. The expected 383 bp amplicons were obtained from the original two symptomatic field samples collected from southern Australian and samples from the four graft-transmitted plants. In 2018, disease symptoms were seen on capsicum plants in seven greenhouse crops inspected in geographically separated areas of the growing region. The disease incidence was between 1-5% and infection by RWMV was confirmed using the specific RT-PCR in samples collected from these greenhouses. No incidence data is available for the earlier detections.

In Italy, RWMV was reported to infect ranunculus (*Ranunculus asiaticus* hybrids) (Vaira *et al*., 1997) and anemone (*Anemone coronaria*) (Vaira *et al*., 2000) and was mechanically transmitted from *Nicotiana benthamiana* to both *N. clevelandii* and *N. megalosiphon* where it elicited systemic infection (Vaira *et al*., 1997). There is no evidence that RWMV causes significant economic loss in capsicum crops, but symptoms can be confused with those caused by the more damaging *Tomato spotted wilt virus* which can lead to inappropriate management choices.

This is the first report of RWMV in Australia, and of the virus infecting capsicum. It is, however, likely that RWMV is the cause of the earlier reports of pepper yellow vein disease in this host (Fletcher *et al*., 1987).

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Figure 1

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