

## First record of *Turnip mosaic virus* in *Pachycladon* spp. (Brassicaceae): an endangered native plant species in New Zealand

J. D. Fletcher<sup>A,C</sup>, R. A. Lister<sup>A</sup>, S. R. Bulman<sup>A</sup> and P. B. Heenan<sup>B</sup>

<sup>A</sup>The New Zealand Institute for Plant & Food Research Limited, Private Bag 4704, Christchurch, New Zealand.

<sup>B</sup>Manaaki Whenua-Landcare Research, PO Box 40, Lincoln 7640, New Zealand.

<sup>C</sup>Corresponding author. Email: john.fletcher@plantandfood.co.nz

**Abstract.** *Pachycladon* comprises nine native species from the South Island, New Zealand, including several threatened species. Virus-like disease symptoms were observed in a *Pachycladon* species collection. This is the first record of a virus in *Pachycladon* and a further report of *Turnip mosaic virus* (TuMV) infections in New Zealand native hosts. This may be the first record of association between the aphid species *Aulacorthum solani* and *Pachycladon*.

*Pachycladon* comprises nine species endemic to the South Island, New Zealand, with a single species occurring in Tasmania, Australia. Most of the species occur in the alpine zone of the Southern Alps where they grow on shaded, south-facing bluffs. The genus has been the subject of taxonomic and evolutionary studies for over 14 years (e.g. Heenan 1999; Heenan *et al.* 2002; Heenan and Mitchell 2003), and for this research an extensive collection of wild-sourced plants has been cultivated in glasshouses at Landcare Research, Lincoln. Several species have restricted distributions and are classified as threatened or uncommon (de Lange *et al.* 2009).

Five *Pachycladon* spp. exhibiting virus-like symptoms in a glasshouse at Manaaki Whenua-Landcare Research, Lincoln, were submitted for diagnosis in November 2008 (Table 1, Fig. 1). The affected plants were infested by *Aulacorthum solani* (Kaltenbach), the potato or fox glove aphid (CM Till, pers. comm.).

Crushed leaf sap from the five plants tested positive for *Turnip mosaic virus* (TuMV) (*Potyviridae*) (mean of controls + 3 × s.d.) using double antibody sandwich enzyme-linked immunosorbent assay (DAS-ELISA) (AS0132, DSMZ, Braunschweig, Germany). TuMV infection was further confirmed by reverse transcriptase polymerase chain reaction (RT-PCR) essentially as

in Fletcher *et al.* (2009). RNA was extracted from ~1 cm<sup>2</sup> leaf discs of five TuMV-infected plants using the RNeasy Plant Mini Kit (Qiagen, Germantown, MD, USA). RT-PCRs were carried out using the Qiagen OneStep RT-PCR kit (Qiagen) using the TuMVF1 and TuMVR1 primers (Fletcher *et al.* 2009). The expected ~290 bp DNA fragment was amplified from all five samples, while RT-negative and H<sub>2</sub>O-negative PCR reactions did not give DNA amplification (Fletcher *et al.* 2009). The PCR products from the five samples were then sequenced and were confirmed by DNA sequence alignment to be from a single TuMV isolate.

Leaf tissue from the five symptomatic plants was used to mechanically inoculate appropriate herbaceous indicator plants and the TuMV isolate found to infect *Brassica rapa* ‘Striker’, *Brassica pekinensis* ‘Wong bok’ and ‘Pak choi’, *Chenopodium quinoa*, *C. amaranticolor*, *Raphanus sativus* ‘Gentle Giant’, *R. sativus* ‘French Breakfast’ and *Sinapsis alba*. Symptoms on all hosts appeared characteristic of those for TuMV infection as summarised in Provvidenti (1996).

*Turnip mosaic virus* is found in New Zealand in a wide range of hosts (Pearson *et al.* 2006; Fletcher *et al.* 2009). In Canterbury, large areas of *Brassica rapa* (turnip) and *Brassica napus* var. *napobrassica* (swede) have been grown regularly for sheep and

**Table 1.** Five *Pachycladon* spp. exhibiting virus-like symptoms, collected from a glasshouse at Manaaki Whenua-Landcare Research, Lincoln, November 2008

Species	Accession <sup>A</sup>	Location of original collection	Symptoms
<i>Pachycladon enysii</i>	408 TuMV	Turks Head, Yeo Stream, Marlborough, 17/06	Yellow vein banding, leaf mottle mosaic, leaf distortion
<i>P. wallii</i>	409 TuMV	Eyre Creek, Eyre Mountains, Southland, 19/66	Yellow vein banding, mottling and leaf margin yellowing
<i>P. enysii</i> × <i>P. fastigiatum</i>	410 TuMV	Artificial hybrid, 76/08	Yellow vein banding, leaf mottle and mosaic
<i>P. fastigiatum</i>	411 TuMV	Mt Hodgkinson, Ben Ohau Range, Canterbury, 133/06	Yellow vein banding, leaf mottle, mosaic margin yellowing and leaf distortion
<i>P. fastigiatum</i>	412 TuMV	Mt Sutton, Barrier Range, Canterbury	Yellow vein banding, leaf mottle and cupping

<sup>A</sup>Accession reference code for samples deposited in the Crop and Food Research Plant Virus Collection located in Christchurch, New Zealand.



**Fig. 1.** (a) *Pachycladon ensyia* × *P. fastigiatum* 76/08 with yellow vein banding, leaf mottle and mosaic symptoms of Turnip mosaic virus (TuMV). (b) *Pachycladon wallii* 'Eyre Creek' with yellow vein banding mottling and leaf margin yellowing symptoms of Turnip mosaic virus (TuMV).

cattle fodder. This is the first record of a virus in *Pachycladon* spp. and a further report of a TuMV infection in a New Zealand native host (Fletcher *et al.* 2009).

The aphid species *Myzus persicae* and *Brevicoryne brassicae* are the two most common vectors of TuMV in cultivated brassicas (Karl 1971). In 2006, *B. brassicae* was identified on *Pachycladon fastigiatum* from Mt. Sutton (Barrier Range, Canterbury) and *Myzus cymbalariae* on *Pachycladon stellatum* from Yeo Stream, Marlborough (D.A.J. Teulon, pers. comm.). *Myzus cymbalariae* is not recorded as a vector of TuMV but is known to vector *Shallot yellow stripe potyvirus* (Van Dijk 1993). *Aulacorthum solani* has also been recorded as a vector of TuMV (Kennedy *et al.* 1962) but has not been recorded as a coloniser of *Pachycladon*. As such, this also appears to be the first record of an association between *A. solani* and *Pachycladon*.

The three *Pachycladon* species, *P. cheesemanii*, *P. exile*, and *P. stellatum* are classified as threatened (de Lange *et al.* 2009). Interestingly, specimens of these three species were grown alongside the infected plants examined in this study but did not exhibit disease symptoms, suggesting that they may be resistant to TuMV infection. We believe TuMV infection probably occurred in the glasshouse and although most species of *Pachycladon* occur in the alpine zone, well away from crop plants, *P. cheesemanii* and *P. exile* occur in lowland sites and may be at risk from nearby crops that are potential hosts to both aphids and TuMV. However, it is also of concern that *B. brassicae*, one of the main vectors of TuMV, has been collected from *P. fastigiatum* in the alpine zone (~1600 m) at Mt. Sutton, Barrier Range (Canterbury). This raises the possibility of alpine species of *Pachycladon* that occur some distance from lowland *Brassica* crops and associated aphid vectors of TuMV also being infected with the virus. If infected with TuMV these endangered plants may either die directly as a result or indirectly from secondary fungal, bacterial or insect attack. There is also a further risk of infection to other native plants from such incursions.

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