Occurrence, taxonomy, biology and pathogenicity of aphelenchid nematodes associated with conifers in south-eastern Australia

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Statement

This work contains no material which has been accepted for the award of any other degree or diploma in any university or other tertiary institutions and, to the best of my knowledge and belief, contains no material previously published or written by another person, except where due reference has been made in the text.

I give consent to this copy of my thesis, when deposited in the university library, being available for loan and photocopying.

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Abstract

Australia has large plantations of exotic *Pinus radiata* conifers. This species is highly susceptible to *Bursaphelenchus xylophilus*, the pine wilt nematode, which is not found in Australia. Potentially pathogenic nematodes were isolated from several dead *Pinus* trees in Williamstown, Heidelberg and Knoxfield, suburbs of Melbourne, Victoria in 2000-2002. A survey of the above-ground nematode fauna of *Pinus* and other conifers in south-eastern Australia was undertaken. Stands of *Pinus* were surveyed in the Kuipto Forest and the South-East Region of SA; the south-west and the Gippsland region of Victoria; and the Hume region in NSW; and native *Callitris preissii* was sampled in the Murray Mallee. A total of 1140 samples from *P. radiata*, 50 from *P. pinaster* and 40 from *C. preissii* were examined. No nematodes were found in wood or young shoots of conifers except in the wood samples from diseased trees at Knoxfield and Heidelberg in Victoria. In contrast, nematodes were common in the bark samples of healthy trees.

Morphologically, extracted nematodes were classified into five trophic groups, including: aphelenchida (plant, fungal and lichen feeders), rhabditids and areolaimids (bacterial feeding), *Macrolaimus* spp. (saprophagus), tylenchids (plant feeding), and dorylaimids (bacterial and algal feeders). Aphelenchids were the most commonly found trophic group. Three genera and twelve morphospecies of aphelenchids were identified. Eight species of *Laimaphelenchus* and one putative species of *Acugutturus* appear to be new records for Australia. Descriptions of two new species, *L. preissii* and *L. australis* have been published. Three species of *Aphelenchoides* were also found. No *Bursaphelenchus* spp. were found.

Molecular studies included sequencing of the ITS region of *Laimaphelenchus preissii*, morphospecies Aphelenchid K1, and Aphelenchid H1; D2D3 fragments of 28S and 18S of *L. preissii*, morphospecies Aphelenchid K1, Aphelenchid K2, and Aphelenchid H1, *Laimaphelenchus australis*, and *Laimaphelenchus* Heidelberg; and COI of three aphelenchid morphospecies *L. preissii*, *Laimaphelenchus* Heidelberg and Aphelenchid K1. Phylogenetic analyses confirmed that *Laimaphelenchus* spp.

are new species and that the unknown aphelenchids are close to *Aphelenchoides*. None of the six isolates studied from Australia was close to *Bursaphelenchus*.

Population growth and mean doubling time of *L. preissii*, Aphelenchid K1 and Aphelenchid H1 were studied at different temperatures and on different food resources. The different species had markedly different population growth rates, which were significantly affected by temperature and food.

A study on desiccation was carried out with *L. preissii* and morphospecies Aphelenchid K1, Aphelenchid K2, Aphelenchid H1 and *Laimaphelenchus* Heidelberg. Ability to survive desiccation varied between species, and the recovery rate of the different species was significantly different.

A pathogenicity study was performed using young *P. radiata* trees in a shadehouse. No symptoms were observed following inoculation with Aphelenchid K1, Aphelenchid K2, Aphelenchid H1 and *Laimaphelenchu*s Heidelberg isolated from diseased *P. radiata* in Victoria, or *L. preissii* from native *Callitris* in South Australia.