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## Nematodes from galls on Myrtaceae. *V. Fergusobia* from large multilocular shoot bud galls from *Angophora* and *Eucalyptus* in Australia, with descriptions of six new species

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### Abstract

Six new species of *Fergusobia*, from large multilocular shoot bud galls on two species of *Angophora* and four species of *Eucalyptus* from both subgenera *Eucalyptus* and *Symphyomyrtus*, are described. *Fergusobia cosmophyllae* Davies n. sp. is characterized by the combination of a C-shaped parthenogenetic female with a short arcuate conoid tail, a broad (small *a* ratio) arcuate infective female with an hemispherical tail tip, and an arcuate to J-shaped male with broad, angular spicules and short bursa. *Fergusobia delegatensae* Davies n. sp. has an open C-shaped parthenogenetic female with a broadly conoid tail, an infective female of variable shape with an hemispherical tail tip, and a male of open C-shape with a crenate bursa that arises 40–70% along the length of the body from the tail tip and terminates just anterior to the cloaca. *Fergusobia diversifoliae* Davies n. sp. has a C-shaped parthenogenetic female with a conoid tail, an arcuate infective female with a hemispherical tail tip, and an arcuate, C- or J-shaped male with angular spicule and a long peloderan bursa. *Fergusobia*

*floribundae* Davies **n. sp.** has a C-shaped parthenogenetic female with a narrow, arcuate, conoid tail, an arcuate infective female with a hemispherical tail tip, and an arcuate or J-shaped male with an angular spicule and a short to mid-body length peloderan bursa. *Fergusobia minimus* Lisnawita **n. sp.** has a C-shaped parthenogenetic female with a conoid tail, an arcuate to open C-shaped infective female with a hemispherical tail tip, and an arcuate to open C-shaped male with an angular spicule and a peloderan bursa arising at about 10–30% of body length. *Fergusobia pimpamensis* Davies **n. sp.** has an open C to C-shaped parthenogenetic female with a narrow conoid tail, an arcuate to open C-shaped infective female with a hemispherical tail tip, and an arcuate to C-shaped male with an arcuate spicule and a long, crenate, peloderan bursa. An inventory of all known *Fergusobia*/*Fergusonina* associations from terminal shoot bud galls is presented. The larval shield morphology of the associated mutualistic *Fergusonina* species is discussed where known. Analyses of DNA sequences of D2/D3 and COI suggested that the six new species are distributed between three clades of *Fergusobia*.

**Key words:** Myrtaceae, *Angophora*, *Eucalyptus*, Neotylenchidae, *Fergusonina*, flies, Australia, taxonomy, DNA sequencing, molecular phylogeny

## Introduction

Multilocular shoot bud galls are the largest, most easily recognised form of gall induced by the *Fergusobia* Currie 1937 (Christie 1941)/*Fergusonina* Malloch 1924 mutualism, and hence have been widely collected (Taylor *et al.* 2005). *Fergusonina turneri* Taylor, 2004 and its mutualist *Fergusobia quinquenerviae* Davies and Giblin-Davis 2004 form multilocular shoot bud galls on *Melaleuca quinquenervia* (Cav.) S.T. Blake, an invasive woody weed in the Florida Everglades, and were investigated as potential biological control agents for the host (Goolsby *et al.* 2000). As part of these investigations, the development of shoot bud galls on *M. quinquenervia* was examined (Giblin-Davis *et al.* 2001), and five other species of *Fergusobia* from similar galls on other species of broad-leaved *Melaleuca* Linnaeus 1767 were described (Davies & Giblin-Davis 2004). The galls of *Fergusonina lockharti* Tonnoir 1937 and its mutualist *Fergusobia brittenae* Davies 2010 were found to support a biologically diverse assemblage of parasitoids, hyperparasitoids and gall inquilines (see *Fn. flavicornis* Malloch 1924, in Taylor *et al.* 1996, Taylor & Davies 2010). The galls of *Fn. turneri* Taylor 2004/*F. quinquenerviae* also support a similar assemblage (Davies *et al.* 2001; Goolsby *et al.* 2001).

Multilocular shoot bud galls, also known as terminal and axial leaf bud galls (TLBG or TLG) (Taylor *et al.* 2005; Davies *et al.* 2010a) or stem tip galls (Currie 1937), have been recorded from *Angophora* Cav. 1797 and both sub-genera of *Eucalyptus* L'Héritier 1788. The Waite Insect and Nematode Collection (WINC) contains multilocular shoot bud gall material and associated fly and/or nematode specimens from 30 species of 'eucalypts' (and one hybrid) from every state and territory in Australia (Table 1). This gall form is a widespread type of *Fergusobia*/*Fergusonina* gall, occurring on a broad range of hosts.

Multilocular shoot bud galls are stalked, the stalk comprising the basal portion of the shoot bud from which the gall develops. On eucalypt species of the subgenus *Eucalyptus* the galls are often longer than wide with a pointed apex (*i.e.*, chilli-shaped) and frequently have leaf tissue growing from the apex (Taylor *et al.* 2005). On *Angophora* and species of the subgenus *Symphyomyrtus* they are usually more globular, and may attain up to 30 mm in diameter, but are occasionally chilli-shaped. Leaf tissue grows from the apices of some. Fewer locules develop in the elongate forms (~ 10–20), but hundreds may be found in the large globular forms (Taylor *et al.* 2005). Small multilocular bud galls with irregular shapes, containing 2–10 locules, have also been collected (Taylor *et al.* 2005) from *Eucalyptus* and *Corymbia* spp., and will be described in a later paper.

In this paper, six new species of *Fergusobia* are described, all from large multilocular shoot bud galls from *Angophora* and *Eucalyptus*. Information is presented on similar gall forms collected from other host species, and on their associated fergusoninid flies where known (Table 1). Molecular and phylogenetic analyses of *Fergusobia* nematodes isolated from various multilocular shoot bud galls were performed (Ye *et al.* 2007; Davies *et al.* 2010a), and are further discussed here.

Clade A: Nematodes from this clade occur on *Angophora* but also include *E. nr. acmenioides*. The distribution of *Angophora* is restricted to eastern Australia, whereas *E. acmenioides* and *A. woodsiana* overlap in coastal northern NSW. These findings suggest that a host switch may have occurred between the two hosts.

Clade B: All the plant species in this clade are placed in subgenus *Eucalyptus*, but from five different sections. Genetically, V's 284 & 7 from *E. diversifolia* appear to be the same with COI sequences, but differ with D2/D3 sequences. This may be an artifact, or there may have been multiple fly conspecific foundresses for this gall form, potentially allowing for horizontal transfer of nematodes between offspring of different females (Davies *et al.* 2010b). This requires clarification, but repeated efforts to re-collect this species have been unsuccessful.

Clade C: Based on similar morphology of nematodes, fly larvae and gall form, it is likely that nematodes from several host plant species, all from subgenus *Symphymyrtus*, belong to the one clade (Davies *et al.* 2010a). This suggests that these *Fergusobia*/*Fergusonina* mutualisms have radiated only within the subgenus, and not switched to hosts outside it. For example, *E. tereticornis* and *E. camaldulensis* are both from Section *Exsertaria* and their distributions overlap. Their associated fly larvae have similar shield types, with sclerotised plates with teeth. Nematodes from galls on each are also of similar morphological type and have similar molecular sequences. Thus there is evidence that the nematode/fly mutualisms and the host plants may have co-evolved and co-specified.

A parallel phylogenetic study of the associated *Fergusonina* flies (S. Scheffer, personal communication) closely matches the inferred phylogeny of the *Fergusobia* nematodes. This supports the hypothesis that the flies and nematodes have co-specified.

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