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(Article begins on next page)
The first record of *Laboulbeniales* (Fungi, Ascomycota) on Ants (Hymenoptera, Formicidae) in The Netherlands

Danny HAELEWATERS
Department of Organismic and Evolutionary Biology, Harvard University
22 Divinity Avenue
Cambridge, Massachusetts 02138, USA
dhaelewaters@fas.harvard.edu

**Summary:** *Laboulbeniales* (Fungi, Ascomycetes) are obligate ectoparasites of arthropods, mostly true insects. 80% of all *Laboulbeniales* parasitize beetles, 10% flies. Also other groups of insects are known to support *Laboulbeniales* infections. This paper gives data and microscopic illustrations on *Rickia wasmannii* from an ant, a *Myrmica scabrinodis* worker, collected in The Netherlands. It appears to be the first Dutch record of a myrmecophilous fungus. Information on specificity and geographical distribution is given.

**Keywords:** Ant-specific fungi, *Rickia wasmannii*, Hymenoptera, Formicidae, ants.

**Introduction**

*Laboulbeniales* (Fungi, Ascomycota) include some 2,050 species in about 140 genera (*Santamaria*, 1998; *Weir* & *Blackwell*, 2005; 146 genera according to *Kirk et al.*, 2008) of obligate ectoparasitic fungi that live associated with arthropods, mostly true insects. Amongst insect-associated fungi they are of excellent use in estimating the diversity of arthropods, mostly true insects. 80% of all *Laboulbeniales* parasitize beetles, 10% flies. Also other groups of insects are known to support *Laboulbeniales* infections. This paper gives data and microscopic illustrations on *Rickia wasmannii* from an ant, a *Myrmica scabrinodis* worker, collected in The Netherlands. It appears to be the first Dutch record of a myrmecophilous fungus. Information on specificity and geographical distribution is given.

Most *Laboulbeniales* are known to be fairly to extreme host-specific (*Thaxter*, 1896; *Scheioske*, 1969; *Tavares*, 1985; *Majewski*, 1994; *De Kezel*, 1996, 1997). Each species seems to have its own host range, i.e. infecting either one host species (stenotopic) or several species (eurytopic). The wider host spectrum is always composed of related taxa (congenic). In case the hosts of a single parasite species are not related, i.e. from a different family, order or subphylum, they always occupy the same micro-habitat (ants nest, termite mound), suggesting that the parasite’s success is affected by this specific environment. This is where the nature of the relationship between host, habitat and parasite is not yet fully understood. Also strong morphological variability within a number of genera such as *Chitonomycetes Peyr.* (*Santamaria*, 2001; *De Kezel* & *Weerbroeck*, 2008; *De Kezel* & *Haelewaters*, 2012) and *Laboulbenia* Mont. & C.P. Robin (*Benjamin* & *Shanor*, 1952; *Rossi* & *Kotrba*, 2004; *De Kezel* & *Van den Neucker*, 2005) and failing classical tools (biometry/allometry and statistics) present many challenges to resolving the group’s both ecology and taxonomy, all this explaining the unrelenting need for molecular tools in addition to the traditional morphological approach.

**Hosts of Laboulbeniales**

*Laboulbeniales* occur almost exclusively on adult hosts, infections of pre-imaginal stages are excessively rare and only observed on particular hosts (cockroaches, termites and ants; *Benjamin*, 1971). The majority of the *Laboulbeniales* parasitize representatives of the subphylum Hexapoda, often Coleoptera (beetles); representatives of ten orders are known as hosts (table 1; *Weir* & *Hammond*, 1997). The greater part of the beetle hosts are members of the two families Carabidae (ground beetles) and Staphylinidae (rove beetles). It is interesting that the diversification of laboulbenialan genera within Coleoptera is greatest within Staphylinidae, i.e. 49 genera, with relatively few species per genus. In contrast, Carabidae host only 15 genera of *Laboulbeniales*, sometimes with hundreds of species in a single genus (*Laboulbenia* (*Tavares*, 1979).

Within the Hymenoptera only ants are known to serve as hosts of these fungi. In this paper the first record for The Netherlands of an ant inhabiting member of the order is des cribed. So far, in Europe, three laboulbenialean species associated with ants (*Hymenoptera*, family *Formicidae*) have been reported: *Rickia wasmannii* Cavara, *Laboulbenia formicarum* Thaxt. and *Laboulbenia camponoti* S.W.T. Batra (*Herraz* & *Espadaier*, 2007).

**Material and methods**

**Host**

The infected ant specimen was collected in ‘Zure Dries’, a nature reserve near the city of Maastricht in the southeastern part of The Netherlands. The specific site is a very small, yet centuries old woodland clearing on a steep south facing slope. Currently the grassland is managed as a pasture and shows a strong vegetation gradient ranging from acid nutrient poor dry grassland (*Violi on caninae*) at the top, through well-developed limestone grassland (*Mesobromion*) in the middle part, to more productive grassland on loamy soil (*Arrhenatherion*) at the basis of the slope. In the 20th century the open area of the clearing has gradually shrunk by wood encroachment, but because of its rare and for Dutch standards thermophilous plant species, the limestone grassland part has always been kept open and in recent years the...
Subphylum Cheliceriformes  
Class Chelicerata  
Subclass Arachnida  
Order Acari

Phylum Arthropoda

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Class Diplopoda  
Subclass Chilognatha  
Order Callipodida  
Order Julida  
Order Sphaerotheriida  
Order Spirostriptida

Subphylum Hexapoda  
Class Pterygota  
Subclass Endopterygota  
Order Hemiptera  
Order Mallophaga  
Order Blattodea  
Order Thysanoptera  
Order Orthoptera  
Order Dermaptera  
Order Isotopera  
Subclass Endopterygota  
Order Hymenoptera  
Order Diptera  
Order Coleoptera

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Subphylum Cheliceriformes  
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Subclass Arachnida  
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Phylum Arthropoda

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<td>Bees, wasps and ants</td>
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Table 1. Distribution of arthropod hosts parasitized by Laboulbeniales (based on Weir & Hammond, 1997). About 80% of the described species of Laboulbeniales parasitize Coleoptera (beetles) (Weir & Blackwell, 2005).

clearing has been enlarged once again (Smits & Schaminée, 2004).
The fungus-infested ant, a Myrmica scabrinodis worker, was sampled by hand in the limestone zone, where according to an earlier ant survey (De Boer, 1993) M. scabrinodis appears to be one of the most abundant ant species. Although M. scabrinodis is somewhat thermophilous (Seifert, 1984), it has a rather broad ecological amplitude (Elmes et al., 1998) and is not characteristic for limestone grasslands (De-Connick et al., 2007). As it is highly tolerant to humidity differences M. scabrinodis is often abundant or even dominant in wet biotopes like bogs (Seifert, 1984).

As the ant was collected as a possible contribution to an ongoing Dutch DNA barcoding project conducted by NCB Naturals (http://science.naturalis.nl/dnabarcoding), the specimen was preserved in 90% ethanol in order to enable DNA analysis. Yet NCB Naturals was not interested in the specimen as fungi do not belong to its target species. Moreover DNA extraction of Laboulbeniales is extremely difficult (Hansson, 1992; Goodwin & Lee, 1993; Haugland et al., 1999; Weir & Blackwell, 2001). Eventually the specimen was sent to the author.

Laboulbeniales

Thalli were removed from the hosts and mounted in permanent microscope slides for identification. Thalli were removed from the host using an entomological pin (size 2 or 3) and immediately embedded in glycerine to which was added a small amount of a saturated alcoholic solution of eosin for permanent mounting (Thaxter, 1896). Cover slips were rinsed with nail varnish.

Microscopic photographs of thalli were taken in the lab of Dr. Alex Weir at the State University of New York College of Environmental Science and Forestry, Syracuse (NY), using a Nikon Eclipse E800 light microscope equipped with differential interference contrast optics and Spot Edition 4.5 imaging software (Digital Camera Systems, Sterling Heights, MI). The microscope slide collection is deposited in FH (Farlow Herbarium, Harvard University Herbaria, Cambridge, MA).

Description

Rickia wasmannii Cavara, Malpighia, 13 : 182, pl. VI (1899) – Fig. 1.

Total length of thallus average 153 µm, maximum 175 µm.

Receptacle comprising a single (non-septate) basal cell surrounded by three series of cells: a posterior series consisting of five to six cells, with secondary appendages or antheridia, terminated by a single primary appendage; an anterior series similar to the posterior, consisting of four to five cells, and ending in a perithecium; and a median series consisting of about six cells. Antheridia 7–15 × 2.5–4.8 µm, compound, subtended by a blackened septum, irregularly disposed, numbers varying in different individuals, flask-shaped, the inflated venters about twice as long as the narrow necks. Perithecium 44–53 × 14–21 µm, hyaline, normally solitary, somewhat asymmetrical, its distal half ending in the somewhat irregular blunt, or truncate, hardly differentiated tip.

Studied material: (female) Myrmica scabrinodis Nylander, 1846 (fig. 2). The Netherlands: Savelsbos, Zure Dries (180.3-312.6), 23.VIII.2011, leg. I. Raemakers, FH-DH40 (fig. 3).

Specificity and geographical distribution

Rickia wasmannii was originally described from Germany by Cavara (1899) on Myrmica rubra (Linnaeus), collected in Linz on the Rhine by the renown formicologist Wasmann (as M. laevinodis), but is known to also appear on several other species of Myrmica (Espadaler & Santamaría, 2012); M. sabuleti Meinert, M. scabrinodis Nylander, M. slovaca Sadil, M. specioides Nylander, M. spinosior Bondroit, and M. vandeli Bondroit. This is the first report of Rickia wasmannii for The Netherlands, the second for the Benelux, where in Luxembourg it was recorded on Myrmica rubra (Linnaeus) by Hulden (1985). Elsewhere in Europe, R. wasmannii has been reported in France, Switzerland, Austria, Slovenia, Spain and Italy (Espadaler & Santamaría, 2012), and more recently in the United Kingdom (Poinin, 2005; «Silfolinia’s AntBlog», 2009), Hungary, Romania (Tartally et al., 2007), Bulgaria (Lapeva-Gjionova & Santamaría, 2011), Czech Republic (Bezděčková & Bezděčka, 2011) and Slovakia (Bezděčka & Bezděčková, 2011). Rickia wasmannii seems likely to be widespread in Europe.

Laboulbeniales

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Laboulbeniales

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**Fig. 1** - *Rickia wasmannii*  

**Fig. 2** - Head of *Myrmica scabrinodis*, heavily infested with *Rickia wasmannii*  
Picture: I. Raemakers.

**Fig. 3** - Site where *Myrmica scabrinodis* with *Rickia wasmannii* has been found in The Nether-
Fig. 4 – *Rickia wasmannii* - SEM
Acknowledgements

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References


