

ORIGINAL RESEARCH



## A mid-Cretaceous fungus, *Philothysanus burmanicus* gen. et sp. nov. (Ascomycota: Spheciophilaceae), on a thrip (Thysanoptera) in Burmese amber

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### ABSTRACT

Male and female thalli of a mid-Cretaceous fungus attached to a thrip (Thysanoptera) in Burmese amber are described as *Philothysanus burmanicus* gen. et sp. nov. in the family Spheciophilaceae (Ascomycota). Both spermatia and ascospores are present. Currently, *Philothysanus burmanicus* gen. et sp. nov., together with the previously described *Spheciophilia adercia* Poinar, represent the oldest known ectoparasitic fungi of insects and their age can be used as a reference point in future phylogenetic studies on the origin of fungi with this life style.

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### Introduction

Ectoparasitic fungi developing on the cuticle of insects are typically members of the Laboulbeniales (Tavares, 1985; Benjamin, 1973; Thaxter, 1895, 1926; Perreau et al., 2021). They obtain nourishment by producing haustoria that enter the cuticle of the host through their embedded holdfasts. They are regarded as commensals, with most causing little or no noticeable damage to their hosts, although some appear to be pathogenic (Haelewaters et al., 2021).

In the mid-Cretaceous, similar types of entomogenous ectoparasitic fungi existed.

One recently described in Burmese amber was *Spheciophilia adercia* Poinar

(Spheciophilaceae) that was developing on the cuticle of a parasitic wasp (Poinar, 2016).

Another is *Philothysanus burmanicus* sp. et gen. nov., that is described in the present paper. The thalli of this fungus are attached to a thrip (Thysanoptera). It is the second member of the family Spheciophilaceae in Burmese amber. These two species currently represent the oldest known ectoparasitic fungi of insects.

### Materials and methods

The thysanopteran fossil was obtained in 2002 from the Noiye Bum Summit Site mine in the Hukawng Valley located southwest of Maingkhwan in Kachin State (26°20'N, 96°36'E) in Myanmar.

Palynomorphs obtained from the amber beds where the fossil piece originated have been assigned to the Upper Albian of the Early Cretaceous (100-110 mya) (Cruickshank & Ko, 2003; Shi et al., 2012). Nuclear magnetic resonance (NMR) spectra of amber samples taken from the same locality as the fossil indicated an araucarian (possibly *Agathis*) source of the amber (Poinar et al., 2007).

Observations, drawings, and photographs were made with a Nikon SMZ-10R stereoscopic microscope and Nikon Optiphot compound microscope with magnifications up to 800X. Helicon Focus Pro X64 was used to stack photos for better depth of field. Views at various levels of the male and female thalli were clarified in Adobe Photoshop CS 5.1 using a range of multichannel settings.

Several variously sized thalli occur on the tip of the abdomen of the thrip. Some of these thalli were originally hidden behind debris and were only recently discovered after the amber was re-polished.

## Results

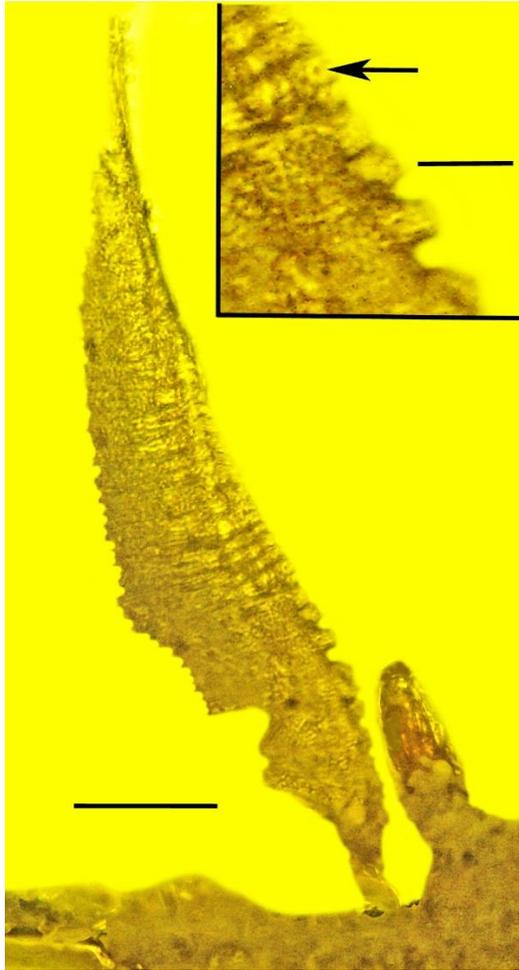
The several adjacent thalli bearing perithecia and antheridia on the abdomen of the thrip are considered to represent variability within a single species (Figs. 1, 2). The large mature antheridium (Fig. 3) represents the holotype specimen.



**Fig. 1.** Thysanopteran host of *Philothysanus burmanicus* showing attached holotype antheridium thallus (arrow) and adjacent thalli in Burmese amber. Scale bar = 180  $\mu$ m.



**Fig. 2.** Tip of tail of thrip host showing different stages of *Philothysanus burmanicus* in Burmese amber. Arrow shows spermatia on an antheridial branch. Scale bar= 12  $\mu$ m.



**Fig. 3.** Extended compound antheridium of holotype of *Philothysanus burmanicus* in Burmese amber. Scale bar = 62  $\mu\text{m}$ . Insert shows a portion of antheridium with emerging spermatia (arrow). Scale bar = 20  $\mu\text{m}$ .

Phylum: Ascomycota (Berk.) Caval.-Sm. 1998

Family: Spheciophilaceae Poinar, 2016

Genus: *Philothysanus* gen. n.

**Mycobank for genus:** MB 841539

**Diagnosis:** Dioecious. Thalli of both sexes with from 5 to 17 receptacle cells. Antheridia either simple, on extended branches from the thallus (Fig. 2), or compound, filling the thallus with

spermatia (Fig. 3). Perithecia short, one (A) is still immature; the second (B), is mature with asci and ascospores. Spores are released from the lower portion of perithecium. Thalli are attached to the abdomen of an adult thrip (Thysanoptera) in mid-Cretaceous Burmese amber.

**Type species:** *Philothysanus burmanicus* gen. et sp. nov.

**Etymology:** The generic name is derived from the Greek “philos” = lover or friend and the Greek “thysanos” = fringe in reference to the wings of the host.

**Mycobank for species:** MB841542

**Etymology:** The specific epithet is based on the geographical origin of the fossil.

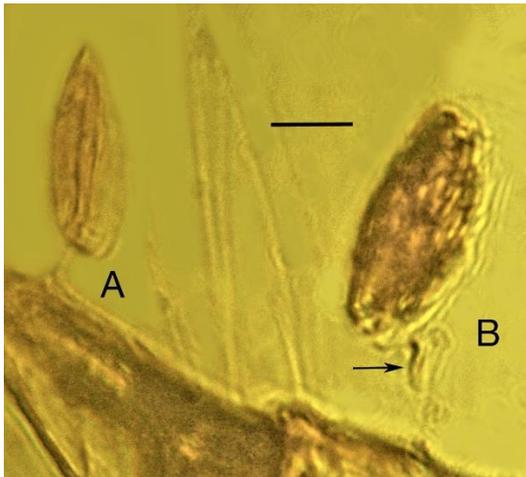
**Type material:** (accession # B-F-11) deposited in the Poinar amber collection maintained at Oregon State University.

**Type locality:** Noije Bum Summit Site mine in the Hukawng Valley located southwest of Maingkhwan in Kachin State (26°20'N, 96°36'E) in Myanmar. The deposits date to the lowermost Cenomanian (98.79  $\pm$  0.62 Ma).

**Diagnosis:** as for genus (by monotypy)

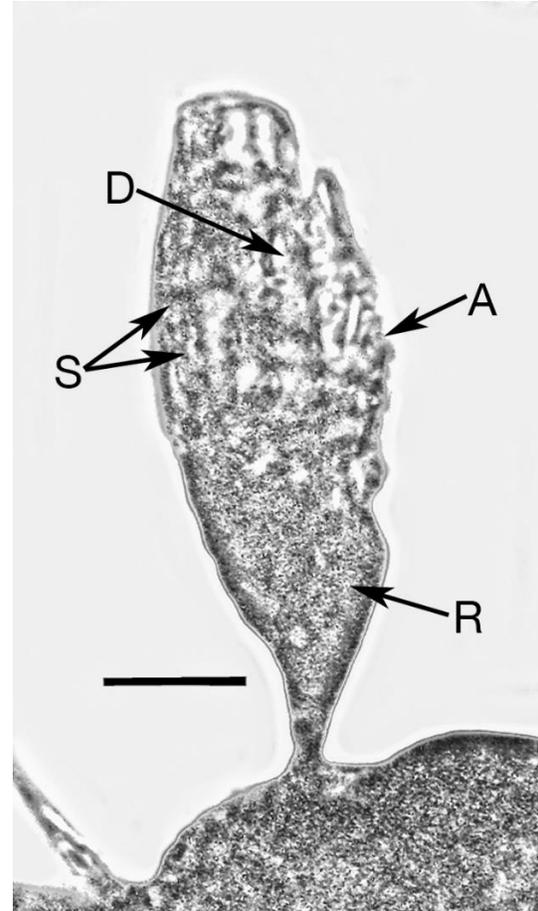
**Description:** (Figs.1-9). Dioecious species. Holotype male thallus compound (Fig. 3), with 8 or more basal receptacle cells and seriate, compound antheridia with superposed cells containing developing spermatia, some of which are emerging from tubular flasks (Fig. 3, insert); length male holotype thallus, 415  $\mu\text{m}$ , greatest width, 70  $\mu\text{m}$ . Second male thallus with extended branches containing antheridia with spermatia (Fig. 2). Female thallus A immature, length, 43  $\mu\text{m}$ , width 15  $\mu\text{m}$ , with short, straight, basal holdfast (Fig.4). Female

thallus B mature, length 48  $\mu\text{m}$ , greatest width, 17  $\mu\text{m}$ , with multiseriate receptacle of 17 oval to elliptical, parenchyma-like cells and adhered lateral trichogone. Asci and ascospores formed in and released from lower portion of perithecium (Figs. 4-9); length released ascospore, 12  $\mu\text{m}$ . Both female thalli with short straight holdfasts. All thalli attached to abdomen of adult thrip (Thysanoptera) in mid-Cretaceous Burmese amber.



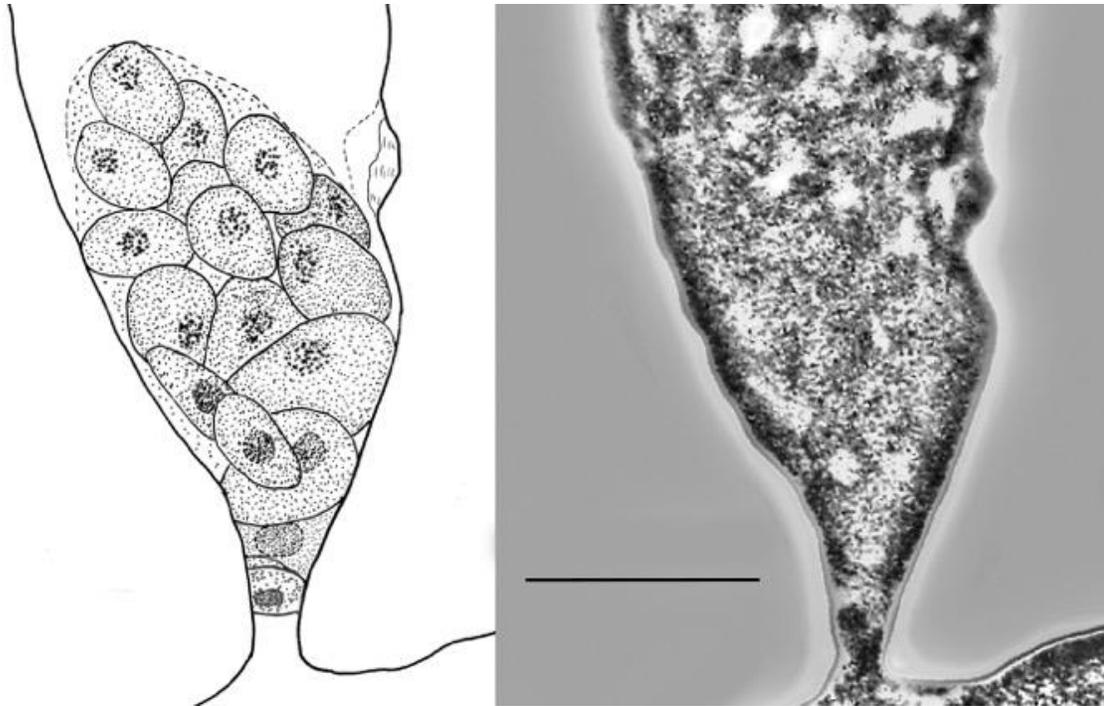
**Fig. 4.** Two female thalli (A and B) of *Philothysanus burmanicus* attached to thrip in Burmese amber. Arrow shows released ascospore. Scale bar = 15  $\mu\text{m}$ .

**Comments:** *Philothysanus burmanicus* sp. nov. represents a dioecious species with thalli producing either spermatia or ascospores. *Philothysanus* is placed in the family Spheciophilaceae based on its multi-celled receptacle, antheridia with spermatia produced in tufted branches and very small ascospores.



**Fig. 5.** Thallus B of *Philothysanus burmanicus* attached to thrip in Burmese amber. A= area of trichogone. D= developing asci. R= multiseriate receptacle. S= septa between outer wall cells. Scale bar = 10  $\mu\text{m}$ .

Several characters distinguish *Philothysanus* from *Spheciophila*, the most important being the dioecious life style of *Philothysanus* and the monoecious status of *Spheciophila*. Also, the thalli of *Philothysanus* are upright, while the thallus of *Spheciophila* is prostrate. The stalk cells of *Spheciophila* are elongate and the terminal portion of the thallus is bent at 90 degrees. The stalk cells of *Philothysanus* are parenchyma-like, clustered in the receptacle and the entire thallus is straight.



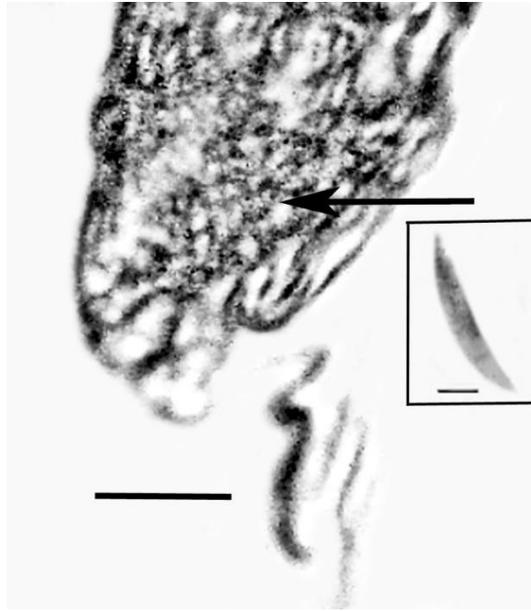
**Fig. 6.** Right photograph shows multiseriate receptacle of thallus B of *Philothysanus burmanicus* in Burmese amber. Using a multichannel setting on Adobe Photoshop revealed 17 oval-elliptical basal cells. These cells are depicted in the drawing on the left. Scale bar = 10  $\mu\text{m}$  (same scale for both figures).



**Fig. 7.** Thallus B of *Philothysanus burmanicus* in Burmese amber showing asci with developing ascospores. Scale bar = 7.4  $\mu\text{m}$ .



**Fig. 8.** Thallus B of *Philothysanus burmanicus* in Burmese amber showing group of ascospores (arrow) in perithecium. Several ascospores have emerged. Scale bar = 10  $\mu\text{m}$ .



**Fig. 9.** Detail of group of ascospores (arrow) in perithecium of thallus B of *Philothysanus burmanicus* in Burmese amber. Several ascospores have been released. Scale bar = 7.5  $\mu\text{m}$ . Insert shows a released ascospore. Bar = 2.7  $\mu\text{m}$ .

**Host:** Thrip (Thysanoptera): Total body length, 0.8 mm. The tubular end of the last abdominal segment places the specimen in the suborder Tubulifera. The forewings are subequal to the hindwings, the wing surfaces lack microtrichia, the fringe cilia are not socketed and the wings overlap. The large bent spine between the terminal tarsomere and arolium prevents placement of the thrip in any known extant or extinct genus. Also unique are the single thick multistriate vein wings. It is not known if these striae represent closely compressed separate veins or just subdivisions of one main vein.

## Discussion

Regarding host selection of the two genera of Spheciophilaceae, *Spheciophila* was attached to the cuticle of a new family (Aptenoperissidae) of parasitic wingless wasps (Rasnitsyn et al.,

2017). There are no other extant or extinct records of ectoparasitic fungi on wasps. This feature alone separates *Spheciophila* from members of the Laboulbeniales, the only other insect ectoparasitic fungal group (Tavares, 1985; Benjamin, 1973).

With *Philothysanus*, ectoparasitic fungi are rarely found on thrips (Tavares, 1985; Benjamin, 1973). Some workers (Perreau et al., 2021) do not even acknowledge the existence of *Dioicomycetes bournieri* Balazuc, the only Laboulbeniales described from a thrip (Balazuc, 1972). This host order has also been omitted in analytical studies of Laboulbeniales (Goldman & Weir, 2018). The single species of *Dioicomycetes* described from a thrip, which was later transferred to the dioecious genus *Dimeromyces* Thaxter (Santameria, 1999) has few structural features in common with *Philothysanus* (Santameria, 2002).

Aside from their respective hosts, both *Philothysanus* and *Spheciophila* differ from the Laboulbeniales by the diminutive size of their ascospores, which range between 9  $\mu\text{m}$  and 12  $\mu\text{m}$  in length. Lengths of ascospores in the Laboulbeniales range from 33  $\mu\text{m}$  to 91  $\mu\text{m}$ . Also the ascospores of *Philothysanus* are not clearly septate as ascospores are in the Laboulbeniales (Tavares, 1985; Benjamin, 1973; Clements & Shear, 1931). In addition, in *Spheciophila* the thallus is prostrate and in *Philothysanus* the asci and ascospores are released from the lower portion of perithecium. These features further separate the two genera from members of the Laboulbeniales (Tavares, 1985; Benjamin, 1973).

Currently, *Philothysanus* and *Spheciophila* represent the oldest known lineages of insect ectoparasitic fungi. Such parasitic relationships could date

back to the Palaeozoic, when many fungal-animal associations first appeared (Harper & Krings, 2021).

### Acknowledgements

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### Conflict of interest statement

The author declares no conflict of interest.

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