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# Putative and novel fossils of insect-associated fungi from Polish Baltic amber

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**ABSTRACT**
Fossil material is very important for better understanding of phylogenetic relationships and evolution of fungi (Beimforde et al. 2014). Unfortunately due to the low preservation potential, only few fossilized fungal structures are found among conservation Lagerstätten (as Rhynie Chert) (Barbee&Taylor 2007) where especially conducive taphonomic conditions occured. A very good source of knowledge about ancient organisms including mycocoenosis, relatively rare is fossil resin, amber. Hitherto only few species of fossil fungi from Baltic amber (dated for Eocene epoch), especially those related with insects, were described (Rossi et al. 2005). Our research focused on light-microscopial of inclusions shared by Museum of The Earth of Polish Academy of Sciences in Warsaw and Museum of Amber Inclusions of University of Gdańsk - especially those containing beetles, flies, ants and other invertebrates. During investigation we found filamentous fungal forms on plant remains, as well as *Mortierella* – like structures associated with rove beetles (Staphylnidae: Pselaphinae).

## INTRODUCTION

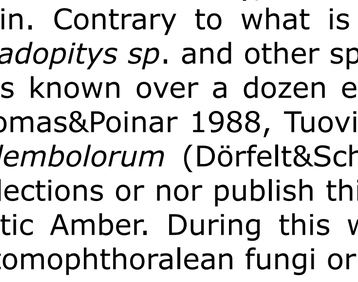
Baltic amber (succinite) is the most popular of all fossil resins. It is dated to the Eocen epoch (about 50 Ma) (Wolfe et al. 2009). It comes from forests growing in the past in the area of the Baltic countries. Chemical composition of this resin is unclear but in contrast to other fossil resins (Matuszewska 2009), baltic amber almost always contains 8 % succinid aicid (Grimaldi 1996). It is not known which tree produced this amount of resin. Contrary to what is generally considered it was not only pine *Pinus succinifera* but probably also *Agatis australis*, *Pseudolarix wehri*, *Sciadopitys sp*. and other species (Wolfe et al. 2009).
It is known over a dozen examples of fungi from amber usually from Dominican and Baltic amber (Rikkinen&Poinar 2000, Schmidt et al. 2013, Thomas&Poinar 1988, Tuovila et al. 2013) but there are only few records of fungus associated with insects from Baltic amber, such as *Aspergillus collembolorum* (Dörfelt&Schmidt 2005) and *Stigmatomyces succini* (Rossi et al. 2005). Hitherto nobody has looked for them deeply in Polish collections or nor publish this data. The main aim of this research was to find and and describe insect-associated fungi from two Polish collection of Baltic Amber. During this work we expected insect-associated fungi such as Laboulbeniales on beetles or flies, *Aegeritella* sp. on ants and a entomophthoralean fungi or an anamorphic form of representative of Clavicipitaceae.

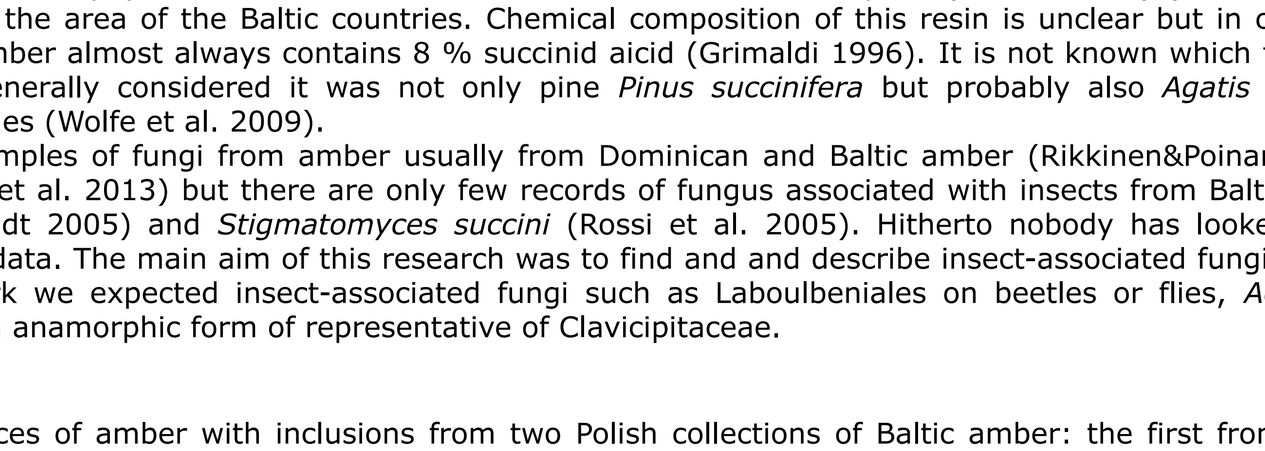
## MATERIAL AND METHODS

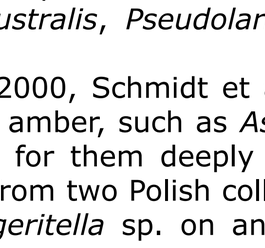
During the research 1230 pieces of amber with inclusions from two Polish collections of Baltic amber: the first from Museum of Earth of Polish Academy of Science and the second from Museum of Amber Inclusions of University of Gdańsk were reviewed and examined using a stereo microscope and light microscope (Nikon SMZ800). Photographs of selected specimens of amber were made using Nikon DX-1200. Selected fragments of amber were cut and polished manually with wet sandpaper to remove external impurities and to minimize light scattering for the investigation.

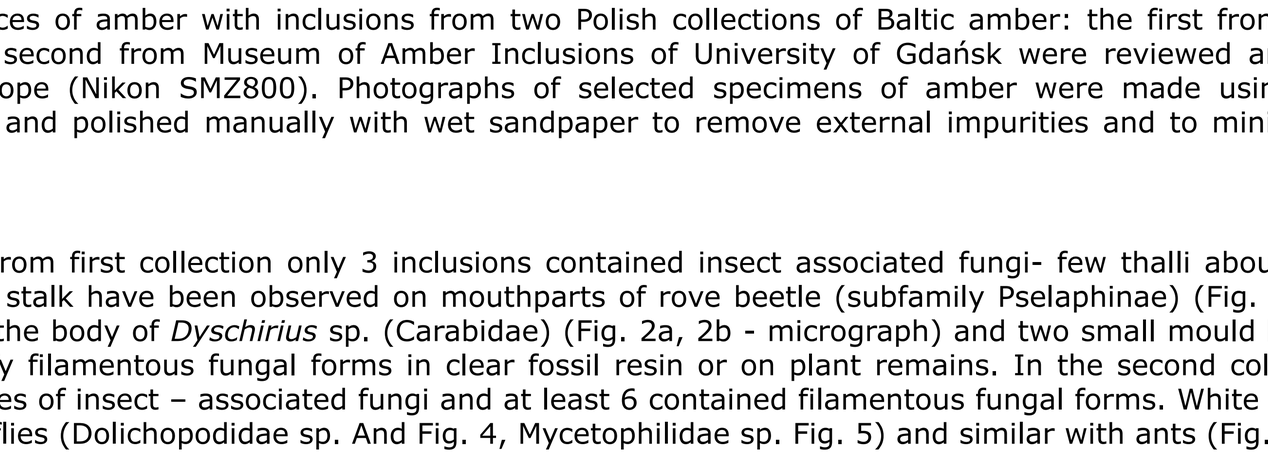
## RESULTS

Among 798 pieces of amber from first collection only 3 inclusions contained insect associated fungi- few thalli about 0,1 mm long with globose sporangium-like structures on stalk have been observed on mouthparts of rove beetle (subfamily Pselaphinae) (Fig. 1a - micrograph, 1b - whole body), traces of mycelium on the body of *Dyschirius* sp. (Carabidae) (Fig. 2a, 2b - micrograph) and two small mould like patches on *Melyridae* sp. (Fig. 3) and 25 with unidentify filamentous fungal forms in clear fossil resin or on plant remains. In the second collection among 441 pieces of amber up to 11 contained traces of insect – associated fungi and at least 6 contained filamentous fungal forms. White hyphae growing between the segments of legs and body of flies (Dolichopodidae sp. And Fig. 4, Mycetophilidae sp. Fig. 5) and similar with ants (Fig. 6).







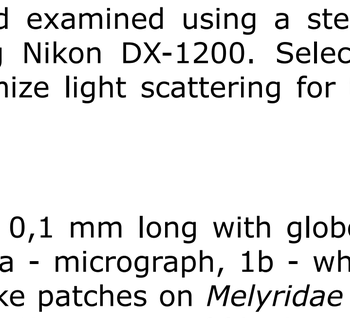



Fig. 1a, 1b. Mouthparts of Pselaphinae sp. (fot. Marta Tischer), on the left side *Mortierella* sp., Fig. 2a Mycelium on the body of *Dyschirius* sp.(fot. Marta Tischer), Fig. 2b. Micrograph of mycelium on the body of *Dyschirius* sp.(fot. Marta Tischer), Fig.3. Gray structure on Melyridae sp.(fot. Marta Tischer) and on right head of ant with *Aegeritella* sp., Fig. 4. Hyphae on Mycetophilidae sp. (fot. Elżbieta Sontag, Museum of Amber Inclusions, University od Gdańsk), Fig. 5. White hyphae on Dolichopodidae sp. (fot. Elżbieta Sontag, Museum of Amber Inclusions, University od Gdańsk) and on right.
Fig. 6. Hyphae and milky impurities on ant Formicidae sp. (fot. Marta Tischer).

## DISCUSSION AND CONCLUSIONS

These new undescribed findings including about 14 putative insects-associated fungi require further and more precise analysis. It is most likely that the fossil insects had been already dead before have been overgrow by fungus. The association was presumably saprotrophs, with the fungus developing on the beetles and flies after they were trapped in resin but still partially exposed to the atmosphere. After the fungus developed, resin subsequent flow covered both organisms (Thomas&Poinar 1988).
There are several possibilities what is the structure on *Merylidae* sp. (Fig. 3.) Firstly, it could be milky impurities which was formed as a result of decay, frequently observed on animal inclusions (Fig. 6) (Mierzejewski 1982). It may also be a initial stage of development of saprotrophic component of the soil microflora. It could be also pathogen of insect for example an anamorphic stage of representative of Clavicipitaceae or an entomophthoralean fungus (Thomas&Poinar 1982). This structure has many morphological similarities to the genus *Aegeritella* which grows only on ants (Bałazy and Wiśniewski 1974). However that should be confirmed by more detailed studies.
It is also possible that the observed structure on mouthparts of rove beetle (Fig. 1a, 1b ) may be a representative of the fungus of the genus *Mortierella* or *Mucor.* Generally, these fungi are not necessarily pathogens, instead they typically represent soil saprotrophs.

## ACKNOWLEDGEMENTS

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