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Communication

New species of gasteroid fungi in Lithuania

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Abstract

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Two new species of Lithuanian gasteroid macromycetes (*Basidiomycota*, *Lycoperdaceae*), *Lycoperdon lambinonii* and *Bovista furfuracea*, collected during field surveys in pine forest and calcareous grassland, respectively, were identified. These species have not been found before due to possible confusion with other species and the specificity of habitats such as calcareous grasslands. Descriptions and photographs of the fungi studied and notes on their ecology and distribution are given.

Keywords: Basidiomycota, gasteromycetes, grasslands, Lycoperdaceae, pine forests.

INTRODUCTION

Gasteroid fungi are fungi of the Basidiomycota division whose spores are formed inside closed, usually round fruitbodies. Because the spores are formed inside the fruitbodies, gasteroid fungi are well adapted to withstand extremes of air, soil temperature and soil dryness, as only the mature fruitbody opens and spores are dispersed. Many of these fungi are saprotrophs, decomposing dead organic matter; some are symbiotrophs, forming mycorrhizal associations with plants. Traditionally, these species have been placed in the artificial class Gasteromycetes based on morphology. Most of them phylogenetically are now placed in the order *Agaricales* and the *Boletales*, *Geastrales* and *Phallales* (Pegler et al., 1995).

According to the latest phylogenetic studies, the genera *Bovista* and *Lycoperdon* belong to the family *Lycoperdacea* (Larsson & Jeppson, 2008). The first gasteroid fungi in Lithuania were mentioned at the beginning of the 19th century. Eighteen species of gasteromycetes were mentioned by Jundziłł (1830); later, in the 1835–1843 manuscript, the same species were described more comprehensively and given Lithuanian names by J.A. Pabrėža (Kutorga, 2015). The most active studies of gasteroid fungi in Lithuania were conducted in the second half of the 20th century (Mazelaitis, 1961; Mazelaitis et al., 1963; Minkevičius & Jovaišienė, 1979; Mazelaitis, 1982), when 70 species of gasteroid fungi belonging to 18 genera were documented. After the revision of herbarium specimens, the list of gasteroid fungi species found in Lithuania was shortened (Iršėnaitė & Urbonaitė, 2024) due to a change in taxonomy and misidentification of specimens. However, new gasteroid fungi species have been found in Lithuania during field work. This article provides information on two new gastroid fungi species, *Lycoperdon lambinonii* Demoulin and *Bovista furfuracea* (J.F. Gmelin) Pers., for Lithuania.

MATERIALS AND METHODS

We carried out field research in Lithuania, the districts of Klaipėda, Lazdijai, Nida, Varėna, Vilnius and Zarasai in summer and autumn of 2023. The material collected during the field work was analysed and identified by routine methods using dissecting microscopy, identification keys, and standard chemicals at the Laboratory of Mycology of the Nature Research Centre. Fungal microstructures were observed using *Nikon ECLIPSE* E200 and *ZEISS Axiolab 5* microscopes, and *Zeiss Axiocam 208* camera. A 5% KOH solution was used to dehydrate the material for microscopy of dried specimens. Some of the collected samples of gasteroid fungi were identified based on morphological characteristics provided in identification keys such as Pegler et al. (1995) and Jeppson (2013, 2018).

Fungal DNA was extracted from dried material and amplified using *Platinum™ Direct PCR Universal Master Mix* (Invitrogen, USA) according to a standard protocol. The complete ITS sequence was amplified using the ITS1F and ITS4 primer pairs (Gardes & Bruns, 1993). Samples for sequencing were purified using *ExoSAP-IT* (Applied Biosystems, USA) according to a standard protocol. The PCR products were sequenced on *BaseClear* (The Netherlands). The same primers were used for sequencing and amplification. Sanger sequences were assembled and manually corrected using *BioEdit* software (Hall, 1999). The aligned sequences were identified using the *NCBI BLAST* (Camacho et al., 2009) and *UNITE* (Kõljalg et al., 2020). Species identification was confirmed when the similarity between the submitted sequence and the sequence in the database exceeded 98%. The collected specimens of the new fungi species were deposited at the Herbarium of the Institute of Botany of the Nature Research Centre (BILAS), Vilnius.

RESULTS AND DISCUSSION

Among the fungi samples collected during the field survey, 24 species of gasteroid fungi belonging to 9 genera were identified. Two identified species, *Lycoperdon lambinonii* and *Bovista furfuracea*, were recorded for the first time in Lithuania.

***Lycoperdon lambinonii* Demoulin**

Fruitbodies grow singly or in small groups, are pear-shaped, 1–3.5 cm wide and 2–5 cm high, ochre-brown, with the top of the fruitbodies darkening later (Fig. 1). The exoperidium is covered with clusters of convergent spines, the endoperidium is less prominent, light ochre, attached to the substrate by hyphae strands, which are densely and sparsely distributed in the lower part of the fruitbody. The basidiospores are rounded, faintly patterned, 3.4–4.5 µm in diameter, with extremely short sterigmata; the capillitium is elastic, 2–7 µm wide, with relatively thick walls (Fig. 2), with sparsely arranged small pores and septa.

Based on its macroscopic characters, the size and colour of the fruiting body, *Lycoperdon lambinonii* can be confused with other fungi of this genus, such as

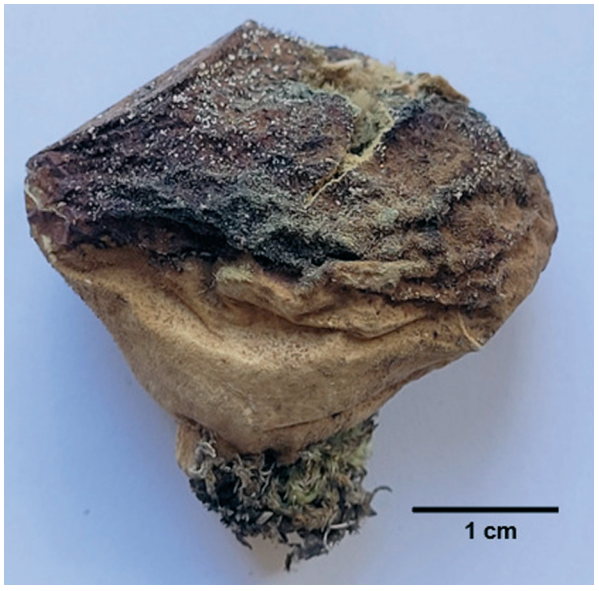

Fig. 1. *Lycoperdon lambinonii* fruit body.

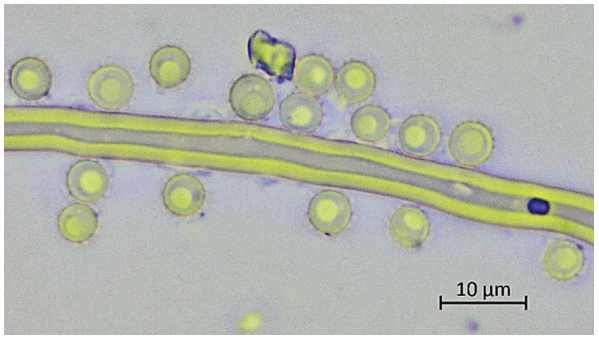

Fig. 2. Microscopic image of *Lycoperdon lambinonii* spores and hyphae.

Lycoperdon molle and *Lycoperdon umbrinum*. Still, it is easier to distinguish based on its microscopic characters. *Lycoperdon lambinonii* is distinguished by its slightly smaller spores, thicker capillitium walls, a denser exoperidium covered with intersecting spines and matte endoperidium surface (Jeppson, 2018).

Ecology and distribution. *Lycoperdon lambinonii* is widespread in Europe, Asia and North America (Larsson & Jeppson, 2008). In Lithuania, it was found in a forest dominated by *Pinus sylvestris* with an admixture of *Picea abies*. The habitat was rich in *Oxalis acetosella*, and 80–90% of the ground surface was covered with moss. This species is mainly found in humus-rich soils under conifers, but can also grow in open areas on calcareous soils (Pegler et al., 1995).

Specimen examined. Lithuania, Zarasai district, Degučiai municipality, near Steponiškės village, Krakynė pine forest, 55.67838° N, 25.99255° E, 15 August 2023, leg. K. Kuzmaitė, det. R. Urbonaitė (BILAS 51826). The sequence result was deposited in the *NCBI BLAST* database (No. PQ570481).

***Bovista furfuracea* (J. F. Gmelin) Pers.**

Fruitbodies are small, 0.8–2 cm wide, globose or subglobose, with a 'root' of hyphae (Fig. 3). The exoperidium of the maturing fruitbody is grey-white, the mature exoperidium may be light brown to grey-brown, covered with white, scale-like clusters of short spines. The mature gleba is olive-brown with no subgleba. The spores are round, small, minimally ornamented, 3.5–4.5 µm, with very short sterigmal remnants, capilitium *Lycoperdon*-type, 4–6 µm in diameter, and small pores (Fig. 4).

The young fruitbodies of *Bovista furfuracea* can be confused with *Bovista aestivalis* (Bates et al., 2009). Still, mature are easier to distinguish because *Bovista furfuracea* is smaller, has a thinner rhizomorph and does not have a subgleba. Microscopically, the spores of *Bovista furfuracea* are larger, and the capillitium is without pores.

Ecology and distribution. *Bovista furfuracea* is widespread throughout Europe and is also found in Asia, North and South America, but has not been recorded in Lithuania or neighbouring Latvia (Jeppson, 2013). In Lithuania, *Bovista furfuracea* was found in a calcareous grassland with 65% grass cover, 17% moss cover, 30% lichen cover and soil pH 5.6. Open, dry and nutrient-poor areas such as dunes or dry calcareous meadows are typical habitats for this species.

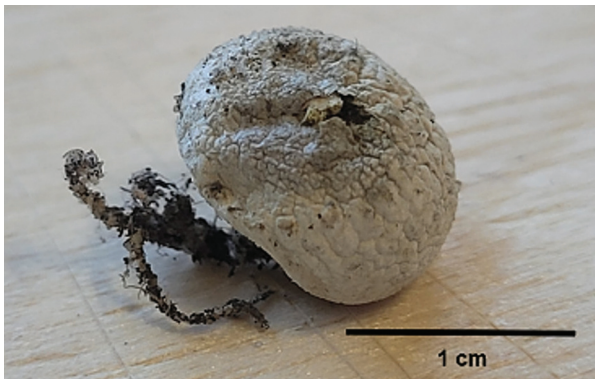

Fig. 3. Fruitbody of *Bovista furfuracea*.

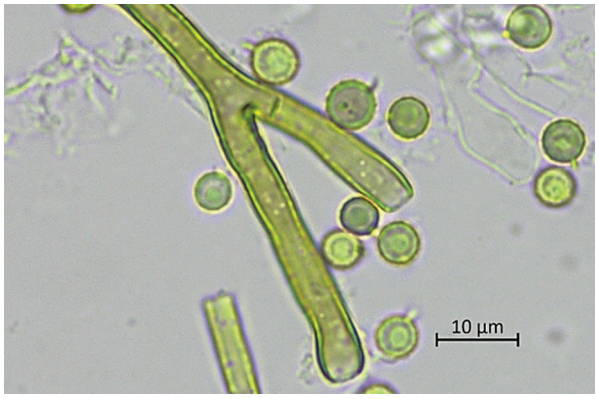

Fig. 4. Microscopic image of *Bovista furfuracea* spores and hyphae.

Specimen examined. Lithuania, Varėna district, in grassland near the village of Mardasavas, 54.142234° N, 24.323893° E, 21 October 2023, leg. et det. R. Urbonaitė (BILAS 51755).

Likely, *Lycoperdon lambinonii* has previously been present in Lithuania, as the country's climate and environment suit the species. It has also been recorded in Scandinavian countries, Finland and Estonia (Jeppson, 2018). This species is often confused with similar species such as *Lycoperdon molle* and *Lycoperdon umbrinum*. To clarify potential misidentifications, specimens of these related species in the Herbarium BILAS were reviewed and compared to confirm that they were correctly identified and did not include *Lycoperdon lambinonii*.

Species of the genus *Bovista*, especially those growing in open habitats, are poorly studied in Lithuania. The newly found *Bovista furfuracea* and other species of the genus *Bovista* are quite rare, as the habitats characteristic for this species are not widespread in Lithuania. However, it may have been

overlooked because of its similarity to *Bovista aestivalis*, or less frequently observed because of its small size. Ongoing fungal surveys in grasslands should reveal a wider distribution of this species.

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