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## **Original research**

# A new addition to the Mycota of Lithuania

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#### Abstract

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Twelve macrofungal species collected in 2012–2020 were identified as new records to the Lithuanian Mycota: Agaricus subperonatus, Desarmillaria ectypa, Entoloma eminens, Hebeloma quercetorum, Hydnellum scabrosum, Hygrophoropsis rufa, Hohenbuehelia unguicularis, Hortiboletus bubalinus, Lactarius mammosus, Lactarius scoticus, Melanogaster broomeanus, Schizophyllum amplum. Descriptions of the fungi studied and notes on their habitats and distribution are provided.

Keywords: agaricoid, Basidiomycota, boletoid, habitat, hydnoid, macrofungi.

#### INTRODUCTION

Basidiomycetous fungi constitute a significant part of terrestrial ecosystems and play an essential role in ecosystems and various aspects of human life (Peay et al., 2016; Sridhar & Deshmukh, 2021; Bahram & Netherway, 2022). They have received much attention due to their role in environmental conservation, atmospheric pollution and forest health monitoring (Sverdrup-Thygeson & Lindenmayer, 2003; Heilmann-Clausen et al., 2015). Some show population decline trends in Europe (Arnolds, 1991). Global conservation assessments of 467 fungi species have recently been published (Mueller et al., 2022). Effective conservation strategies require perfect knowledge about species diversity and their ecology (Runnel et al., 2014; Lõhmus et al., 2018).

The first publication on Lithuanian fungi was published by J. E. Gilibert in 1781, where 35 fungal species were described. Over the next two and a half hundred years, about 1800 basidiomycetous macrofungi species have been described and summarised in publications in Lithuania (Gricius & Matelis, 1996; Mazelaitis, 1976, 1982; Urbonas et al., 1986; Urbonas, 1997, 1999, 2001, 2005; Shiryaev & Iršėnaitė, 2009; Iršėnaitė, 2010). In the last two decades, only a few species of basidiomycetous macrofungi have been published as new to Lithuania (Iršėnaitė, 2004; Iršėnaitė et al., 2013). During this time, species new to Lithuania and Europe have been described, such as *Aureoboletus projectellus* (Motiejūnaitė et al., 2011), formally a North American species, which has been spreading along the Baltic Sea for several decades. *Tricholoma joachimii*, a rare species, has been recorded in Lithuania as a north-eastern European record for this species (Iršėnaitė et al., 2022).

Lithuania belongs to the temperate and hemiboreal zones and, respectively, sections of these zones (Ahti et al., 1968). These zones and the differences in environmental conditions specific to these zones determine the biodiversity, thereby, the diversity of mycobiota in Lithuania.

This study aims to contribute to the addition of the Lithuanian mycobiota and broaden our knowledge about it.

### MATERIALS AND METHODS

The fungal specimens were collected during field trips between 2012 and 2020, mainly in broad-leaved and pine forests in southern and eastern Lithuania, also provided by amateur mycologists. During the field surveys, the macroscopic and ecological characteristics of the specimens were recorded. All fungi were collected for subsequent microscopical identification in the laboratory. The microscopic structures were examined using a Nikon Eclipse Ci-S light microscope. Squashed fungus tissue preparations were mounted in water and 5% KOH solution. Measurements were carried out in water. Identification was based on relevant literature (Heilman-Clausen et al., 1998; Knudsen & Vesterholt, 2012; Kränzlin, 2005; Nitare & Högberg, 2012; Vesterholt, 2005). Descriptions are based on the examined specimens. Voucher specimens are deposited at the Herbarium of the Nature Research Centre (BILAS) in Vilnius. Nomenclature corresponds to MycoBank Fungal Databases (http://www.mycobank.org).

The DNA of the fungal specimens was amplified using Platinum<sup>™</sup> Direct PCR Universal Master Mix (Invitrogen) and purified using UltraClean DNA Purification Kit (MoBio Laboratories, Inc., Carsbad, CA). The target ITS region was amplified using primer pairs ITS1F and ITS4 (Gardes & Bruns, 1993). The PCR products were sequenced on BaseClear (The Netherlands). Chromatograms of the sequences were manually checked and corrected using BioEdit software (Hall, 1999). The sequences obtained in this study were deposited in NCBI Gen-Bank. GenBank numbers were provided for the sequenced specimens.

#### **RESULTS AND DISCUSSION**

Sixteen specimens were assigned to twelve species from eleven genera belonging to *Basidiomycota*, *Agaricales (Agaricus, Desarmillaria, Entoloma, Hebeloma, Hohenbuehelia, Schizophyllum), Boletales* (*Hortiboletus, Hygrophoropsis, Melanogaster), Russulales (Lactarius)* and *Thelephorales (Hydnellum).* We present a list of these species new to Lithuania with their localities, habitats, collection data, collector (*leg.*) and identifier (*det.*) names, specimen number in the Nature Research Centre Herbarium (BILAS) and sequence deposited in GenBank number, description of their morphology, notes on their ecology and distribution. Species are listed in alphabetical order.

#### Agaricus subperonatus (J.E. Lange) Singer

**Description.** Basidiomata agaricoid, cap 60–150 mm, spherical, becoming broadly convex or almost flat, surface breaks into broad brown scales on a pale creamy-white background. It turns red when cut or bruised. Gills are crowded, free, white, turn greyish pink and then dark brown. Stem whitish, smooth cylindrical or slightly clavate,  $50-120 \times 15-30$  mm, flesh firm, pale grey, pink when cut. The ring is ascending, persistent, and double. Basidiospores 6–7.5 × 5–6.5 µm, broadly ellipsoid to ovate, smooth, brownish. Spore print chocolate brown.

**Material examined:** Lithuania, Vilnius district, Nemenčinė township, 54.84762 °N, 25.46766 °E, in a yard under *Tilia cordata*, 13 August 2021, leg. V. Drozdov, det. R. Iršėnaitė (BILAS 51512); GenBank No OM944044.

**Notes.** Agaricus subperonatus grows in open areas, parks, gardens, pastures and cemeteries, prefers fertilised humus-rich soil and compost, and can be found in recently disturbed soil under deciduous trees. Saprotrophic. Widespread, but occasional in temperate and rare in the hemiboreal zone (Knudsen & Vesterholt, 2012). This fungus is also found in western North America (Kibby, 2011).

#### Desarmillaria ectypa (Fr.) R.A. Koch & Aime

**Description.** Basidiomata agaricoid, cap 15– 60 mm, convex to fairly flat, sometimes with a depressed centre, wide, scaly in the centre, yellowish to dark brown, hygrophanous. Gills rare, narrow, decurrent, cream, later light pink to red-brown. Stem  $40-80(100) \times 10-15(20)$  mm, clavate, fibrillose, the same colour as the pileus. Ring on the stipe absent. Flesh thin, white, soft, with a faint smell and mild taste. Hyphae have clamp connections. Basidiospores  $7-9 \times 5.5-6.5 \mu$ m, ellipsoid, smooth. Spore print white.

**Material examined:** Lithuania, Varėna district, the Čepkeliai Strict Nature Reserve, 54.02131 °N, 24.62231 °E, among *Sphagnum* moss in an active relatively undamaged raised bog, 9 September 2012, leg. et det. R. Iršėnaitė (BILAS 51235); GenBank No KY924821. **Notes.** *Desarmillaria ectypa* is confined to various types of wetlands with low or relatively low nitrogen contents and at least localised base-rich micro-habitats (Ainsworth, 2003). Not associated with wood decomposition, rather saprotrophic on decaying peat moss and other bryophytes (Koch et al., 2017; Ainsworth, 2003). A Eurasian species of boreal-mountain range and possibly continental (Ohenoja, 2006). Rare and occasional in the boreal, very rare in temperate hemiboreal zones (Knudsen & Vesterholt, 2012), red-listed in Latvia (Critically Endangered) (Latvijas Daba, 2023), red-listed in Finland (Vulnerable) (Punainen Kirja, 2019), IUCN Global Fungal Red List (Near Threatened) (Svetasheva, 2015).

#### Entoloma eminens Kokkonen

**Description.** Basidiomata tricholomatoid, cap 50–120 mm, conical when young, then applanate, slightly depressed, pale grey-brown, in the centre darker. Gills adnate-emarginate, subventricose, pale greyish, then pink. Stem  $6-120 \times 10-20$  mm wide pale greyish. Ring absent. Flesh greyish in pileus, stem pale grey. Smell indistinct, taste farinaceous. Basidiospores (7)8–10 × (6)7–8 µm, subisodiaetrical with 5–6 angles. Clamp connections are present in all tissues.

**Material examined:** Lithuania, Utena district, Ažuolija Forest, 55.46117 °N, 25.58308 °E, under *Quercus robur* in the broadleaved forest, 25 July 2017, leg. et det. R. Iršėnaitė (BILAS 51441); Gen-Bank No OM630476.

**Notes.** *Entoloma eminens* grows in rich, moist mixed forests on calcareous soils near *Tilia* and *Corylus*. Kokkonen (2015) has reported it from rich, moist, mixed *Picea abies* dominated forests, with scattered deciduous trees such as *Populus tremula* and *Alnus incana*. It is a recently described species and, perhaps for this reason, is unknown in many countries. The finding of this fungus in Lithuania indicates that, although it appears to be rare, its distribution may be underestimated, and the habitat in Lithuania supports the thesis (Noordeloos et al., 2018) that it is generally associated with thermophilous forests and boreal broad-leaved trees. However, the typical habitat for the species is still unknown.

#### Hebeloma quercetorum Quadr.

**Description.** Basidiomata tricholomatoid, cap 30–50 mm, convex to expanded, smooth, slightly

slimy, pinkish buff to clay buff or yellowish brown. Gills crowded, brownish, interspersed with lamellulae. Stem  $30-80 \times 6-13$  mm, cylindrical, base o thickened and bulbous, whitish. Ring absent. Flesh white, darkening from the base. Smell and taste raphanoid. Basidiospores amygdaloid to broadly citriform,  $10-14 \times 6-8$  µm. Cheilocystidia lageniformventricose,  $30-60 \times 6-11$  µm. Spore print chocolate brown.

**Material examined:** Lithuania, Utena district, Ažuolija Forest, 55.46117 °N, 25.58308 °E, under *Quercus robur* in the broadleaved forest, 25 July 2017, leg. et det. R. Iršėnaitė (BILAS 51440); Gen-Bank No OM574971.

**Notes.** *Hebeloma quercetorum* is a thermophilic species, widespread in southern Europe and rare but possibly overlooked in the temperate zone on calcareous soils. Mycorrhizal with deciduous trees such as *Quercus* (Quadraccia, 1993; Vesterholt, 2005) also *Carpinus* and *Fagus*, sometimes *Tilia* and *Corylus*. The finding of *Hebeloma quercetorum* in Lithuania supports the thesis that this fungus is widespread in Northern Europe (Vesterholt, 2005).

#### Hohenbuehelia unguicularis (Fr.) O.K. Mill.

**Description.** Basidiomata pleurotoid, cap 4–15 mm, surface pruinose, strigose towards attachment point, rounded fan-shaped seen from above, bell-shaped from the side, with straight, entire margin, translucently striate, brownish-black to black. Gills distant, relatively narrow, radiating from an eccentric point, with an entire edge, pale brown to black. Stem as a contraction of the cap; smell faintly farinaceous. Basidiospores 7–9(11) × 4–5(6) µm, ellipsoid, cylindrical or phaseoliform; cheilocystidia 11–33 × 4–7.5 µm, subcylindrical, clavate with 1–3 rostra. Spore print white.

**Material examined:** Lithuania, Raseiniai district, Sargeliai village, 55.475664 °N, 23.45880 °E, on the wood of *Populus tremula*, 7 February 2022, leg. B. Gliwa, det. R. Iršėnaitė (BILAS 51631); Gen-Bank No OP026084.

**Notes.** *Hohenbuehelia unguicularis* grows on branches of deciduous trees and shrubs in small groups. Saprotrophic. Rare, but relatively widespread in the European temperate zone. Very rare in the temperate-middle boreal zone (Knudsen & Vesterholt, 2012). Species are red-listed in Denmark (En-

dangered) (Danmarks Svampeatlas, 2016) and rare in Poland (Wojewoda, 2003).

# Hortiboletus bubalinus (Oolbekk. & Duin) L. Albert & Dima

**Description.** Basidiomata boletoid, cap 20–80 mm, hemispherical, later convex to flattened, fine velvety, smooth or very finely cracked, blueing when damaged. Tubes pale yellow to yellow with an olivaceous tint, blueing when injured. Pores are concolorous with the tubes. Stem cylindrical, ventricose or club-shaped, yellowish, blueing when bruised. Flesh whitish in the cap, distinctly pinkish below the cap cuticle, yellowish in the stipe, orange, brown in the stipe base, with few orange-red dots, blueing in the cap. Smell and taste are not distinctive. Basidiospores  $11-15 \times 4.5-5 \mu m$ , subfusiform, smooth. Spore print yellow-olive.

**Material examined:** Lithuania, Vilnius district, Vilnius city, 54.69998 °N, 25.31007 °E, between grass in a lawn under *Betula*, 9 September 2020, leg. et det. R. Iršėnaitė (BILAS 51368).

**Notes.** Fungus grows in urban areas, parks, lawns, and mycorrhizal with poplars (*Populus*) also lime (*Tilia*). The range of mycorrhizal hosts is probably wider and includes *Betula*, *Fagus*, *Picea*, *Carpinus*, *Betula*. *H. bubalinus* is known in several European countries. More widespread than previously thought, but possibly neglected (Assyov & Stoykov, 2011). The fungus habitat in Lithuania seems to be in the eastern part of its known geographic range, and further research is likely to reveal that this fungus is more widespread in Eastern Europe. However, the actual distribution is still not fully understood (Assyov & Stoykov, 2011).

# *Hydnellum scabrosum* (Fr.) E. Larss., K.H. Larss. & Kõljalg

**Description**. Basidiomata hydnoid, cap 12 cm, convex to flattened, brownish, covered with brown scales. It can be tinged with pink at the margins and darken with age. The mushroom has yellow-brown  $5 \times 0.3$  mm spines under the cap. They are decurrent to the stem. Stem  $2-12 \times 1-6$  cm pinkish brown, base greyish green. Flesh whitish to pinkish, grey to black or greenish in the stem base. Hyphae without clamp connections. Odour is not significant, but tastes bitter. Basidiospores  $5-6 \times 4-5 \mu m$ ; irregularly globose to subglobose; nodulose. Spore print brown.

Material examined: Lithuania, Varena district, the Čepkeliai Strict Nature Reserve, 54.03008 °N, 24.55486 °E, on soil in lichen-dominated Pinus sylvestris forest, 17 September 2014, leg. O. Grigaitė, det. R. Iršėnaitė (BILAS 50731); Tauragė district, the Viešvilė Strict Nature Reserve, 55.15381 °N, 22.45799 °E, on soil in Pinus sylvestris forest with Vaccinium, 29 August 2016, leg. A. Uselienė, det. R. Iršėnaitė (BILAS 51233); Švenčionys district, the Girutiškis Strict Nature Reserve, 55.26918 °N, 25.87205 °E, on soil in mixed Pinus sylvestris-Picea forest, 7 September 2018, leg. et det. R. Iršėnaitė (BI-LAS 51302); Švenčionys district, Gelednės Forest, 54.94870 °N, 26.00873 °E, on soil in lichen-dominated Pinus sylvestris forest, 11 September 2018, leg. G. Švitra, det. R. Iršėnaitė (BILAS 51360); Gen-Bank No OM959529.

**Notes**. It grows in lichen-dominated pine forests, on roadsides or small forest paths, next to older pines, mycorrhizal with *Pinus*. The fungus is a signal species indicating habitats of high nature value in coniferous forests (Nitare, 2000). *Hydnellum scabrosum* is known in several European countries, very rare in Denmark (Danmarks Svampeatlas, 2016) and rare in Poland (Wojewoda, 2003).

#### Hygrophoropsis rufa (D. A. Reis) Knudsen

**Description.** Basidiomata boletoid, cap 30–100 mm, dry, convex to shallow funnels, matt to tomentose, margin incurved. Gills orange with a salmon tinge, orange, decurrent, dichotomously forked. Stem  $20-30 \times 5-15$  mm, tapering towards the base, concolourous with the cap. Flesh orange buff, unchanging when exposed to air. Odour and taste are not distinctive. Basidiospores  $5-6.5(7) \times 3-4$  µm, cylindrical or ellipsoid, thick-walled, strongly dextrinoid. Spore print white.

**Material examined:** Lithuania, Marijampolė district, the Žuvintas Strict Nature Reserve, 54.48086 °N 23.67782 °E, on a stump of coniferous tree in spruce forest, 11 October 2019, leg. et det. R. Iršėnaitė (BI-LAS 51463).

**Notes.** *Hygrophoropsis rufa* grows on or near conifer stumps and trunks, on conifer woodchips and sometimes on sawdust. Saprotrophic. Probably widespread in Europe, but its distribution needs to be clarified. Fungus is occasional in hemiboreal-southern boreal, possibly also in middle boreal, rare in the

temperate zone (Knudsen & Vesterholt, 2012), and rare in Poland (Wojewoda, 2003).

#### Lactarius mammosus Fr.

**Description.** Basidiomata agaricoid, cap 25–80 mm, convex to plane with a small umbo, surface finely velvety to finely scaly, greyish brown. Gills crowded, decurrent, white, then pink-ocher. Stem 20–70 × 5–15 mm, cylindrical to slightly club-shaped, dry to velvety, whitish, then greyish or pink, brown, pithy hollow when old. Flesh creamy, reddish, taste mild to slightly bitter or burning, smells sweet, coconut. Latex white, unchanging on exposure, sweet at first, then burning, bitter taste. Basidiospores  $6-9 \times 5-6 \mu m$ , broadly ellipsoid. Spore prints are whitish or creamy.

**Material examined:** Lithuania, Švenčionys district, the Girutiškis Strict Nature Reserve, 55.26918 °N, 25.87205 °E, on dry soil in the mixed forest under *Pinus*, 6 September 2018, leg. et det. R. Iršėnaitė (BILAS 51339).

**Notes.** *Lactarius mammosus* is associated with coniferous and rarely mixed forests on dry, sandy soils among mosses. Fungus forms mycorrhiza with *Pinus, Picea, Betula, Abies, Larix.* It grows at least in Europe (Kränzlin, 2005), is common in hemiboreal–subarctic zone, rare in the temperate zone (Knudsen & Vesterholt, 2012), red-listed in Denmark (Vulnerable) (Danmarks Svampeatlas, 2016), rare in Poland (Wojewoda, 2003).

#### Lactarius scoticus Berk. & Broome

**Description.** Basidiomata agaricoid, cap 14–60 mm, convex to expanded with depressed centre, surface smooth, dry, an increasingly felt-like texture moving towards the margin, pale cream to whitish, later becoming pale pinkish-buff to cream, with a more yellowish to yellowish-brown centre. Gills crowded, adnate to decurrent, whitish to pale pinkish buff, sometimes forked near the stem attachment. Stem 20–70 × 4–10 mm, cylindrical to slightly club-shaped, smooth to finely felty, light cream to pinkish buff. The flesh colour is similar to the outer surfaces. Taste acrid, odour acidic to fruity. Latex is white and has an immediately acrid taste. Basidiospores  $5.5-7 \times 5-5.5 \mu$ m, broadly ellipsoid to ellipsoid. Spore print pale cream.

**Material examined:** Lithuania, Alytus district, the Žuvintas Strict Nature Reserve, 54.48086 °N, 23.67783 °E, under *Betula* in a wet forest near a peat

bog, 10 September 2018, leg. et det. R. Iršėnaitė (BI-LAS 51403); GenBank No OM945717.

**Notes.** Fungus is associated with European peat bogs (very wet *Betula-Salix* carrs), where it grows among mosses in a mycorrhizal association with birch (*Betula*) species. *L. scoticus* is occasional in the temperate-subarctic zone (Knudsen & Vesterholt, 2012), and rare in Latvia (Latvijas Daba, 2023).

#### Melanogaster broomeanus Berk.

**Description.** Basidiomata tuberoid, irregularly globose, up to 40 mm in diameter, 15–30 mm high, initially yellowish brown to dark brownish when mature. Gleba is yellow to blackish, depending on the maturity of basidiomata. Smell intense and fruity. Basidiospores  $6-8(10) \times 3-4.5 \mu m$ , cylindrical, to slightly ellipsoid, smooth, with sterigma remnant at the apex. Hyphae with clamp connections.

**Material examined:** Lithuania, Utena district, Ažuolija Forest, 55.46461 °N, 25.58430 °E, under *Quercus* and *Pinus* in the broad-leaved forest, 14 September 2014, leg. et det. R. Iršėnaitė (BILAS 51666); Kaišiadorys distr., Rumšiškės village, 54.86625 °N, 24.22340 °E, among the grass in the lawn under *Tilia* and *Quercus*, two basidiomata, 10 October 2018, leg. G. Šapranauskaitė, det. R. Iršėnaitė (BILAS 51279); GenBank No OP018413.

**Notes.** *Melanogaster broomeanus* is associated with warm deciduous forests on calcareous soil. Mycorrhizal with deciduous trees such as *Fagus*, *Quercus*, *Tilia*, *Carpinus* and *Corylus*, less frequently grows under conifers and often in gardens. The species is common in the Nordic countries (Knudsen & Vesterholt, 2012), occasional throughout Europe, red-listed in Finland (Critically Endangered) (Punainen Kirja, 2019), and rare in Poland (Wojewoda, 2003).

#### Schizophyllum amplum (Lév.) Nakasone

**Description.** Basidiomata annual, disc-shaped, cap 3–20 mm, cupulate to campanulate, tough, gelatinous, whitish, felty; hymenium smooth or slightly ribbed, pale brown to ochre, attached to the substrate without a stipe. Basidiospores  $7.5-10(11) \times 2.5-3(4) \mu m$ , cylindrical, allantoid, smooth, thin-walled. Hyphae with clamp connections. Odour and taste are not distinctive. Spore print white.

Material examined: Lithuania, Utena district, Žirgės Forest, 55.62219 °N, 25.80585 °E, deciduous

forest, on a fallen branch of *Populus* sp., 24 April 2022, leg. J. Motiejūnaitė, det. R. Iršėnaitė (BILAS 51621).

**Notes**. Fungus grows on dead, mostly rotting twigs, and branches of deciduous trees *Populus* and *Salix* in open places, often 1–2 m above the ground in alluvial and swamp forests. The species is rare in the temperate zone and other parts of Europe (Wojewoda, 2006; Robinson et al., 2011).

It is worth mentioning that more than half of recorded specimens were found in different natureprotected areas of Lithuania: the Čepkeliai Strict Nature Reserve, the Girutiškis Strict Nature Reserve, the Viešvilė Strict Nature Reserve and the Žuvintas Strict Nature Reserve.

This fact and the occurrence of rare species increase the natural value of these nature protection areas and provide grounds for intensifying their mycological study.

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