

## Three new species of myxomycetes from Siberia

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Information is provided on the find of three new species of myxomycetes of the *Perichaena* and *Gulielmina* genera. The description, ecology, known localities, and differences from morphologically similar species are given. A comparative study of the new myxomycete species with similar species was carried out using scanning electron microscopy. Micrographs with relevant details of sporocarps and spores are presented. An analysis of the molecular phylogeny of the new species was carried out and their connections with representatives of bright-spored myxomycetes are shown.

**Key words:** Altai Territory, Asia, DNA, *Gulielmina*, morphology, Novosibirsk Region, *Perichaena*, SEM, slime moulds.

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Vlasenko A.V., Vlasenko V.A. (2023): Tři nové druhy hlenek ze Sibiře. – Czech Mycol. 75(2): 159–177.

Studie přináší informace o objevu tří nových druhů hlenek z rodů *Perichaena* a *Gulielmina*; je uveden jejich popis, ekologie, známé lokality a rozdíly oproti morfologicky podobným druhům. Srovnávací studie nově popsaných druhů s podobnými hlenkami byla provedena s využitím skenovacího elektronového mikroskopu a jsou prezentovány mikrofotografie relevantních detailů sporokarpů a spor. Současně byla provedena analýza molekulární fylogeneze nových druhů, ukazující jejich vazby na další zástupce světlovýtrusých hlenek.

### INTRODUCTION

Siberia is a huge region of Northern Asia with contrasting natural conditions. Siberia stretches from the Ural Mountains in the west to the regions of the Far East in the east. Siberia is washed by the Arctic Ocean in the north and borders with China, Mongolia and Kazakhstan in the south. The area is about 10 million km<sup>2</sup> (Samoilova 2004–2017). The territory of Siberia includes a variety of natural zones from Arctic tundra in the north to steppes and semi-deserts in the south

(Kuminova 1960, Yurkovskaya et Safronova 2019). Siberia is usually divided into Western Siberia, Eastern Siberia, Central Siberia, the Baikal Region, Transbaikalia, Northeastern Siberia, and the mountains of Southern Siberia (Altai, Sayans) (Rakovskaya et Davydova 2001).

Most of the territory located within the West Siberian Plain, the Altai Mountains and the Sayan Mountains is subject to desertification. Desertification occurs due to climate change (Savostyanov 2014, Filandysheva et al. 2015, Trofimov et al. 2015). A study of the influence of aridisation on the state of habitats and species diversity of myxomycetes allowed us to discover bright-spored species of myxomycetes previously unknown to science.

Myxomycetes are a group of free-living protists. Myxomycetes live in the upper layers of the soil, on plant litter, in folds of the bark of living woody plants. Currently, about 1100 species of myxomycetes are known in the world (Lado et Hernández on-line). Although the study of Siberian myxomycetes began in the late 19<sup>th</sup> and early 20<sup>th</sup> centuries, they still remain a poorly studied group in this region (Thümen 1878, 1880a, 1880b, 1881, Saccardo 1880, Karsten 1906, Jaczewski 1907, Lavrov 1927, 1929, 1937, 1938, 1948, 1951).

The large territory and many hard-to-reach places in Siberia make it difficult to study the biodiversity of myxomycetes. We started studying myxomycetes in Siberia in 2007 and have described six species new to science to date (Vlasenko et al. 2018, 2019, 2020a, 2020b, 2022, 2023). In this article, a description and scanning electron microphotographs of three new species of bright-spored myxomycetes of the genera *Perichaena* and *Gulielmina* are presented; their phylogeny is characterised on the basis of the 18S nrDNA gene.

## MATERIAL AND METHODS

**List of abbreviations.** CM – compound microscope; coll. – collected; cult. – cultivated; diam. – diameter; DM – dissecting microscope; ident. – identified; RL – reflected light; SEM – scanning electron microscope; TL – transmitted light.

**Slime mould isolation and morphological examination.** The new species of myxomycetes of the genera *Perichaena* and *Gulielmina* were isolated by the moist chamber method. Substrates (dry leaves of living steppe plants, mosses on living trees) were placed into glass Petri dishes with a diameter of 10 cm. Filter paper was placed on the bottom of the Petri dish to retain water. Distilled water was poured into Petri dishes. The next day, the covers of the wet chamber were opened for 4–5 hours to ensure evaporation of excess water. The cultures were exposed to natural light for at least 8 hours a day. Cultivation was carried out at room temperature (19 to 24 °C) during 3 months. A Carl Zeiss Stemi DV4 stereomicroscope, a Carl Zeiss Axiolab A light microscope, and a Carl Zeiss EVO MA 10 scanning electron microscope were used for morphological analysis. To study them in a scanning electron microscope, the myxomycete samples were air-dried, fixed on aluminum plugs with a double-sided adhesive film, and then sprayed with gold.

Type specimens of new species are stored in the M.G. Popov Herbarium, USU 440537, Novosibirsk (NSK). The nomenclature used corresponds to Lado et Hernández (on-line), taking into account a recent work on the phylogeny of bright-spored myxomycetes (García-Cunchillos et al. 2022).

**DNA extraction and sequencing.** To extract DNA, sporocarp fragments were crushed using aluminum oxide ( $Al_2O_3$ ) and then homogenised in a buffer for lysis with the Phyto-Sorb kit (Synthol, Moscow). The 18S nrDNA regions (SSU) were amplified with standard primers for bright-spored myxomycetes SFATri (AATCTGCGAACGGCTCCGTA) and SR4Bright (TGCTGGCACCAGACTTGT) (Fiore-Donno et al. 2013). HS Taq DNA Polymerase (Evrogen, Moscow) was used. PCR load (50 µl): 35.25 µl of PCR-grade water, 5 µl of 10X Taq PCR Reaction Buffer (Evrogen, Moscow), 2.5 µl of 50 mM  $MgCl_2$  (Evrogen, Moscow), 0.5 µl 20 µM each dNTPs (Evrogen, Moscow), 1 µl 10 ng/µl each primer (Biosset, Novosibirsk), 0.75 µl 5 U/µl SNPdetect HS Taq DNA Polymerase (Evrogen, Moscow) and 4 µl 10 ng/µl DNA, adjusted with ddH<sub>2</sub>O. PCR cycling conditions were as follows: denaturation for 5 min at 94 °C, followed by 36 cycles (15 s at 94 °C, 15 s at 50 °C, 1 min at 72 °C) and a final elongation for 7 min at 72 °C. PCR products were sequenced using the Big Dye terminator cycle sequence kit (ABI) and ABI Prism 3130 sequencer (Perkin-Elmer, Waltham, USA) at SB RAS Genomics Core Facilities (ICBFM SB RAS, Novosibirsk, Russia). All the sequences generated in the study are deposited in GenBank (<https://www.ncbi.nlm.nih.gov/genbank>).

**Phylogenetic analyses.** Sequenograms of the new species were analysed in Chromas version 2.6.6 (<http://technelysium.com.au/wp/chromas/>). New sequences of partial 18S nrDNA (the first ca 500 nucleotides) were compared with the available data in GenBank using the Nucleotide BLAST tool (Altschul et al. 1990, <https://blast.ncbi.nlm.nih.gov/Blast.cgi>). Based on the BLAST search, additional SSU sequences of other related species were incorporated into the analyses. The final dataset included 50 SSU sequences. *Tubifera ferruginosa* (Batsch) J.F. Gmel. was used as an outgroup (García-Cunchillos et al., 2022). Tab. 1 provides an overview of the taxa used, information on the herbarium specimens, GenBank accession numbers and references. Sequences were aligned online in MAFFT (Katoh et al. 2002), version 7 (Katoh et Sandley 2013, <http://mafft.cbrc.jp/alignment/server/>) using the E-INS-I strategy (Katoh et Toh 2008). MEGA X software (Kumar et al. 2018) was used for statistical analysis of nucleotide datasets. The phylogenetic relationships were reconstructed using the Maximum likelihood (ML) method (Felsenstein 1981). Phylogenetic trees were constructed in the online version (Trifinopoulos et al. 2016, <http://iqtree.cibiv.univie.ac.at/>) of the IQ-TREE software (Nguyen et al. 2015). A nonparametric bootstrap with 1000 repetitions as statistical support was used in this analysis. The phylogenetic trees were visualised in Fig Tree version 1.4.4 (Rambaut 2018).

**Tab. 1.** Sequences used for the phylogenetic analyses.

Species	Herbarium voucher/isolate	GenBank accession numbers	Reference
<i>Gulielmina</i> sp. nov. ( <i>Gulielmina</i> group)	NSK 1016035	OQ925673	this study
<i>Perichaena</i> sp. nov. (separate branch)	NSK 1016124	OQ925675	this study
<i>Perichaena</i> sp. nov. ( <i>Perichaena</i> group)	NSK 1016120	OQ925674	this study
<i>Cornuvia serpula</i>	MM29198	JX481285	Fiore-Donno et al. 2013
<i>Gulielmina megaspora</i>	MA:Fungi:82123	MT154026	García-Cunchillos et al. 2022
<i>Gulielmina patagonica</i>	MA:Fungi:91909	MT154035	Ronikier et al. 2020
<i>Gulielmina vermicularis</i>	MA80426	ON713363	García-Cunchillos et al. 2022
<i>Gulielmina vermicularis</i>	MA88424	ON713409	García-Cunchillos et al. 2022
<i>Gulielmina vermicularis</i>	BR<BEL>: 5020025765604	MT154019	Ronikier et al. 2020

Species	Herbarium voucher/isolate	GenBank accession numbers	Reference
<i>Hemitrichia abietina</i>	MA58838	ON713338	García-Cunchillos et al. 2022
<i>Hemitrichia calyculata</i>	MA81807	ON713370	García-Cunchillos et al. 2022
<i>Hemitrichia clavata</i>	MA62018	ON713344	García-Cunchillos et al. 2022
<i>Hemitrichia crassifila</i>	MA:Fungi:91885	MT154031	Ronikier et al. 2020
<i>Hemitrichia decipiens</i>	MA83070	ON713379	García-Cunchillos et al. 2022
<i>Hemitrichia intorta</i>	KR:0022295	MT154021	Ronikier et al. 2020
<i>Hemitrichia lutescens</i>	MA83430	ON713391	García-Cunchillos et al. 2022
<i>Hemitrichia minor</i>	MA80197	ON713361	García-Cunchillos et al. 2022
<i>Hemitrichia pardina</i>	MA80413	ON713362	García-Cunchillos et al. 2022
<i>Hemitrichia serpula</i>	NSK 1031916	ON863805	this study
<i>Hemitrichia serpula</i>	NSK 1030527	OP725706	this study
<i>Metatrachia floriformis</i>	MA83204	ON713382	García-Cunchillos et al. 2022
<i>Metatrachia floripara</i>	Lado25103	ON713324	García-Cunchillos et al. 2022
<i>Metatrachia horrida</i>	MA81778	ON713369	García-Cunchillos et al. 2022
<i>Metatrachia vesparium</i>	MA51719	ON713334	García-Cunchillos et al. 2022
<i>Oligonema affine</i>	MA78975	ON713357	García-Cunchillos et al. 2022
<i>Oligonema favogineum</i>	MA83229	ON713383	García-Cunchillos et al. 2022
<i>Oligonema flavidum</i>	DWM5764	JX481304	Fiore-Donno et al. 2013
<i>Oligonema persimile</i>	–	AY643826	Fiore-Donno et al. 2005
<i>Oligonema schweinitzii</i>	MA85559	ON713399	García-Cunchillos et al. 2022
<i>Oligonema verrucosum</i>	MA83489	ON713394	García-Cunchillos et al. 2022
<i>Ophiotheca calongei</i>	MA78692	ON713353	García-Cunchillos et al. 2022
<i>Ophiotheca chryosperma</i>	MA63754	ON713345	García-Cunchillos et al. 2022
<i>Ophiotheca pedata</i>	MA81941	ON713372	García-Cunchillos et al. 2022
<i>Perichaena agaves</i>	MA50703	ON713332	García-Cunchillos et al. 2022
<i>Perichaena corticalis</i>	AMFD157	JX481306	Fiore-Donno et al. 2013
<i>Perichaena depressa</i>	NSK 1031856	ON863804	this study
<i>Perichaena dictyonema</i>	MA59057	ON713340	García-Cunchillos et al. 2022
<i>Perichaena liceoides</i>	M0073211	ON713328	García-Cunchillos et al. 2022
<i>Perichaena longipes</i>	LMW-2015	KP241120	Walker et al. 2015
<i>Perichaena luteola</i>	DWM4984	JX481308	Fiore-Donno et al. 2013
<i>Perichaena nigra</i>	MA86774	ON713402	García-Cunchillos et al. 2022
<i>Perichaena quadrata</i>	MA88310	ON713406	García-Cunchillos et al. 2022
<i>Perichaena stipitata</i>	MA79150	ON713358	García-Cunchillos et al. 2022
<i>Trichia alpina</i>	MA80534	ON713364	García-Cunchillos et al. 2022
<i>Trichia contorta</i>	sc22513	KT358722	Feng et Schnittler 2017
<i>Trichia erecta</i>	sc22424	KT358724	Feng et Schnittler 2017
<i>Trichia scabra</i>	MA90224	ON713410	García-Cunchillos et al. 2022
<i>Trichia sordida</i>	AMFD 81	EF513182	Fiore-Donno et al. 2010
<i>Trichia varia</i>	NSK 1016006	OP725705	this study
<i>Tubifera ferruginosa</i>	AMFD 196	EF513171	Fiore-Donno et al. 2010



## RESULTS AND DISCUSSION

## TAXONOMY

***Perichaena bryophila* A. Vlasenko, sp. nov.**

Figs 1, 2: A, C, E, G, I

Index Fungorum No.: IF900809

Etymology. Refers to the habitat – living on mosses.

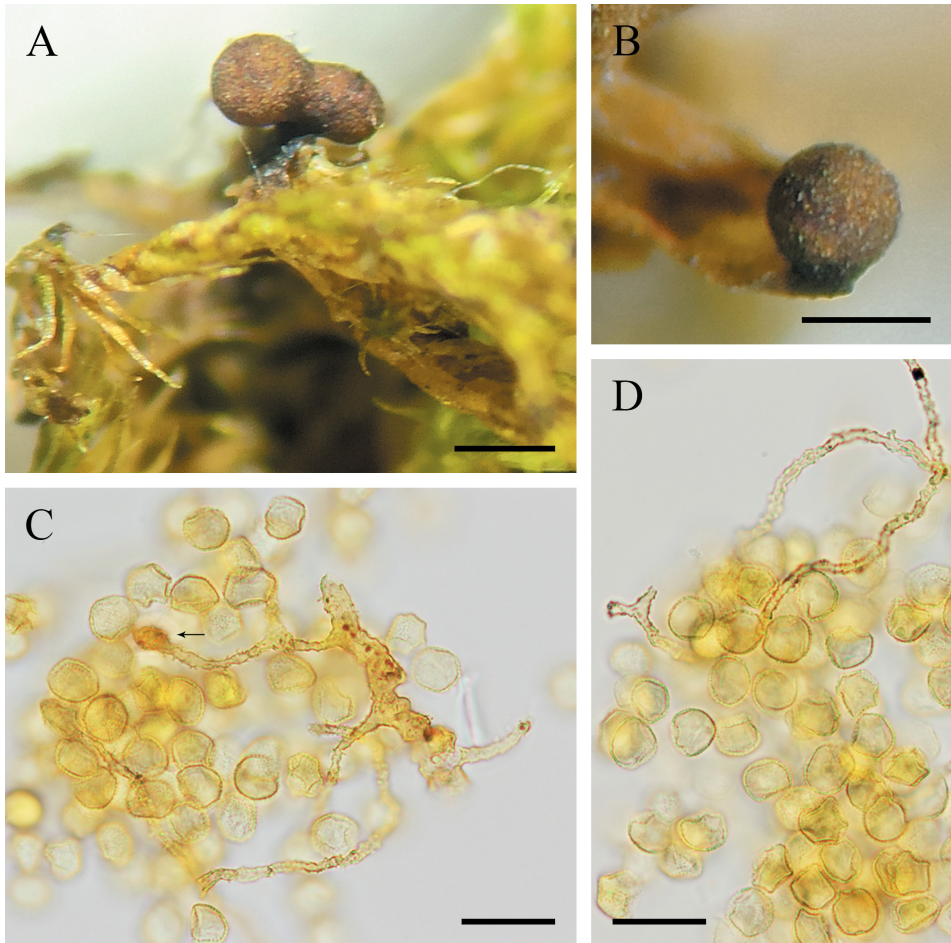
Type. Russia, Novosibirsk Region, near the town of Novosibirsk, floodplain of the Zyryanka River, 54°49'29.0" N, 83°06'10.9" E, 136 m a.s.l., on moss on bark of living *Salix alba* L. collected at a height of 3 m, substrate samples coll. 17 August 2019, A.V. Vlasenko, moist chamber culture 17 August 2019, cult. and ident. by A.V. Vlasenko, NSK 1016124 (holotype), GenBank No.: OQ925675 (18S nrDNA).

Description. Sporocarps scattered, sessile or on short stalks, 0.3–0.7 mm tall, sporotheca 0.3–0.4 mm in diam., yellowish or light brown. Hypothallus undeveloped. Stalk black or dark brown, short, membranous, sometimes resembling a continuation of the hypothallus or completely absent. Peridium single, dense, opaque, dehiscence irregular, yellow, yellow-brown or dark brown in RL. Inner surface of the peridium ornamented with warts merging into lines and an incomplete mesh. Capillitium usually dense, consisting of very long branched threads 2.2–3.5(6.5) µm in diam., ornamented with large spine-like structures and depressions, clearly visible in a CM. In SEM, capillitium ornamentation consisting of depressions of various diameters, large spikes and large elongated structures resembling hollow tubes with funnel-shaped expansion at the top. Spore mass light golden yellow. Spores light yellow, free, spherical, 9.5–10.5 µm in diam. to oval, 9.5 × 10.5 µm, ornamented with irregularly spaced warts.

Ecology. Growing as an epiphyte on moss on the bark of living trees (Fig. 8 G, H).

Comments. Morphologically, the new species is closest to *Ophiotheca pedata* (Lister et G. Lister) García-Cunch., J.C. Zamora et Lado. The ornamentation and size of the spores in these two species are identical (Fig. 2 E, F), but the ornamentation of the capillitial threads differs strongly. The capillitial threads of *Perichaena bryophila* are ornamented with very large spine-like structures and depressions, clearly visible in a CM and a SEM (Fig. 2 G, I), while those of *O. pedata* are covered with small spikes scattered on the surface and small depressions, visible only in SEM (Fig. 2 H, J), and are often almost smooth and elastic. Also, the ecology of these two species is different. *Ophiotheca pedata* is most often found on litter, *P. bryophila* lives on mosses growing on living woody plants.

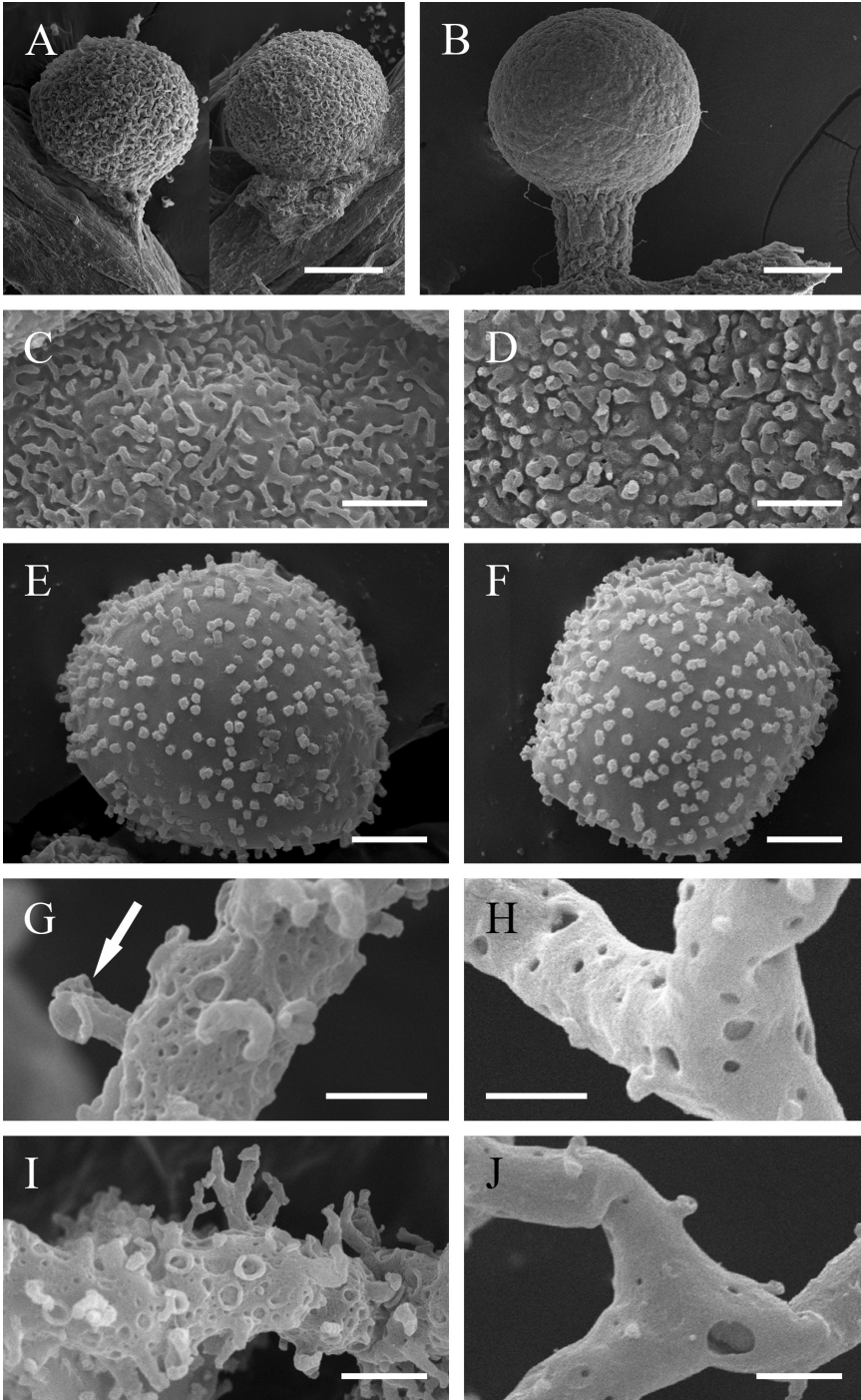
The ML analysis based on the 18S nrDNA region (Fig. 7) showed that the new species of *Perichaena bryophila* is closest to *Trichia contorta* G.H. Oth. The genetic distance of the *P. bryophila* branch on the SSU tree is 0.143, with 54% bootstrap support. *Perichaena bryophila* is not included in a well-supported clade



**Fig. 1.** *Perichaena bryophila* (NSK 1016124, holotype): **A, B** – sporocarps (DM); **C, D** – spores and capillitial threads (CM). The arrow indicates the free end of the capillitial thread. Bars: 300  $\mu\text{m}$  (A, B); 10  $\mu\text{m}$  (C, D). Photos by A. Vlasenko.

**Fig. 2.** *Perichaena bryophila* (NSK 1016124, holotype, SEM): **A** – sporocarps; **C** – ornamentation of the inner part of the peridium; **E** – spore; **G, I** – capillitial threads. The arrow indicates capillitial threads with large elongated structures resembling hollow tubes with funnel-shaped expansion at the top. *Ophiotheca pedata* (NSK 1030018, SEM): **B** – sporocarp; **D** – ornamentation of the inner part of the peridium; **F** – spore; **H, J** – capillitial threads. Bars: 100  $\mu\text{m}$  (A, B); 2  $\mu\text{m}$  (C–F); 1  $\mu\text{m}$  (G, H); 2  $\mu\text{m}$  (I, J). Photos by A. Vlasenko. ▶

with other species, but forms a separate branch among a group of species with unspecified taxonomic status. *Trichia contorta* is the closest species to *P. bryophila*. *Trichia contorta* belongs to the genus *Trichia* by a complex of morphological features, but has no phylogenetic relationships with species of the genus



*Trichia* s. str. with the type species *Trichia varia* (Pers. ex J.F. Gmel.) Pers. The capillitial threads of *T. contorta* are covered with spiral thickenings; *P. bryophila* has capillitium without spiral thickenings. The spores of *T. contorta* are 10–13(14) µm in diam., covered with spines, while the spores of *P. bryophila* are 9.5–10.5 µm in diam., covered with warts. The ecology of these two species is similar. *Trichia contorta* most often lives on the bark of living woody plants.

We have described *P. bryophila* as part of the morphological genus *Perichaena* based on a complex of morphological features. When new taxa are identified/separated and formally described in the scope of the current morphological genera *Perichaena*, *Trichia*, *Metatrachia*, and *Hemitruchia*, the taxonomic position of *P. bryophila* will be clarified by us.

***Perichaena maculosa*** A. Vlasenko et V. Vlasenko, **sp. nov.** Figs 3, 4: A, C, D, G, I

Index Fungorum No.: IF900810

**Etymology.** Refers to the peridium, which has a pattern in the form of golden spots on a dark background.

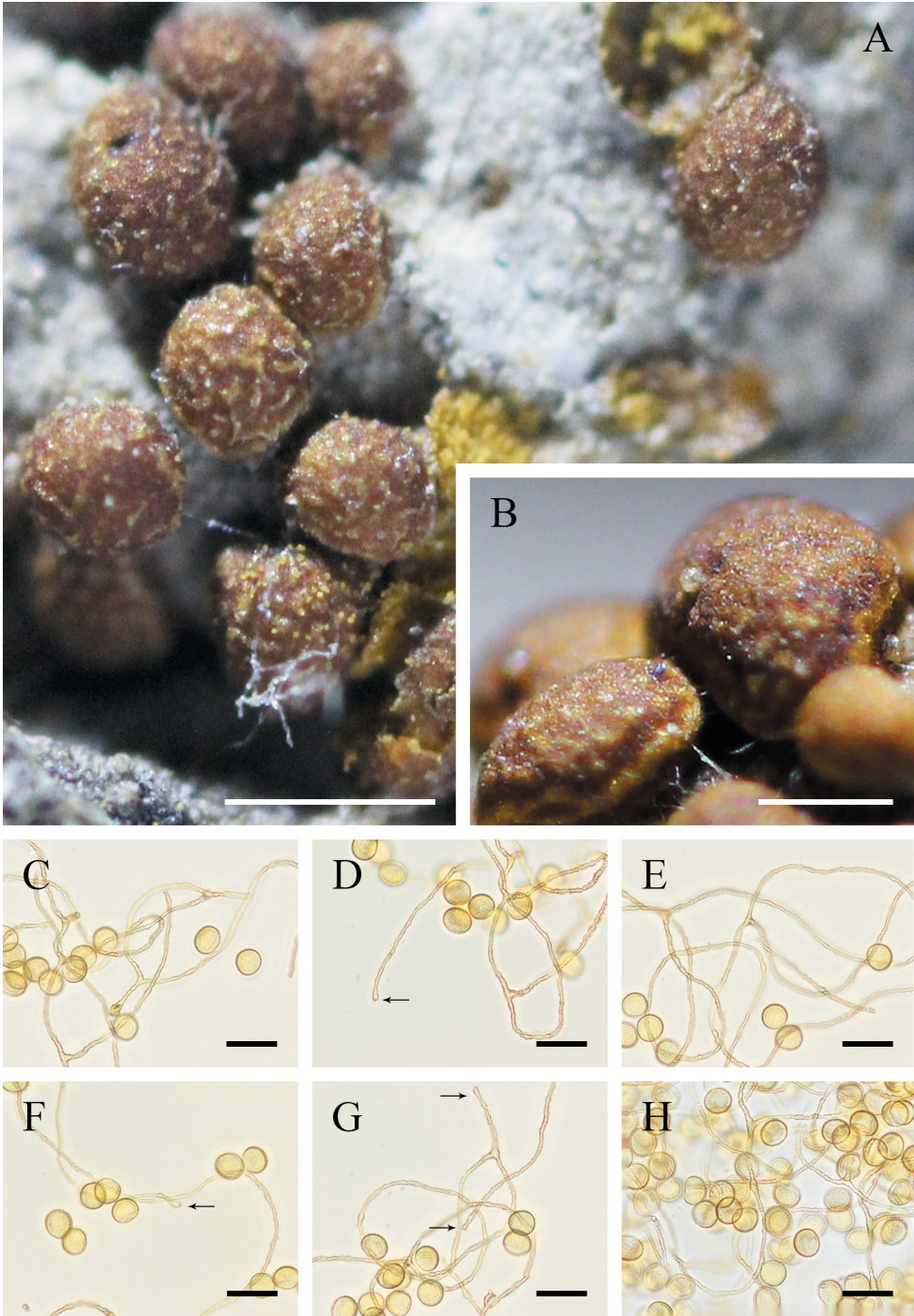
**Type.** Russia, Altai Territory, Kuryinsky District, near the village of Novofirsovo, petrophytic steppe, 51°46'30.1" N, 82°19'34.5" E, 330 m a.s.l., on dry leaves of *Orostachys spinosa* (L.) C.A. Mey., pH 7.65, substrate samples coll. 13 July 2015, V.A. Vlasenko, moist chamber culture 9 July 2017, cult. and ident. A.V. Vlasenko, NSK 1016120 (holotype), GenBank No.: OQ925674 (18S nrDNA).

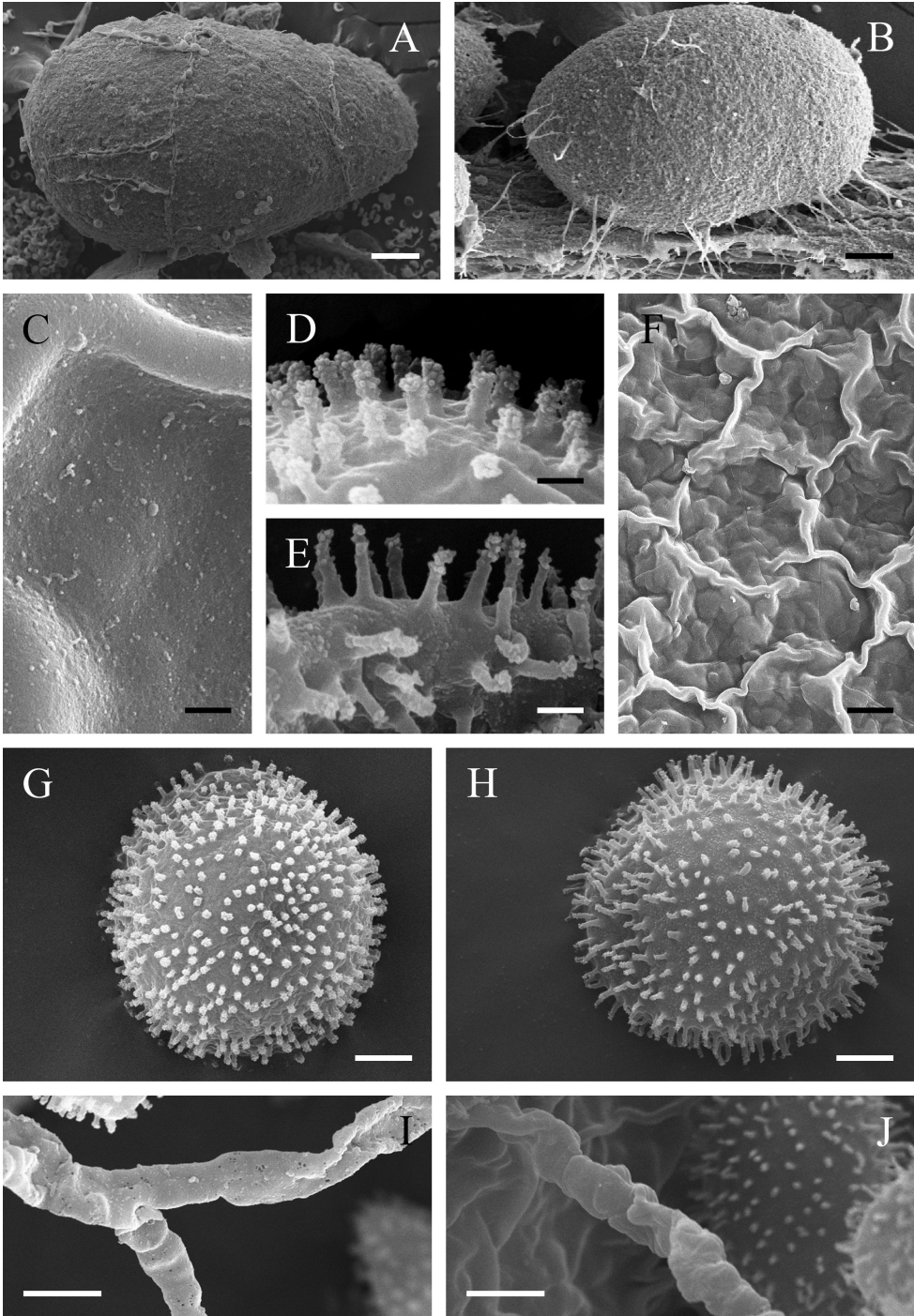
**Other sample examined.** Russia, Altai Territory, Kuryinsky District, near the village of Novofirsovo, petrophytic steppe, 51°46'35.4" N, 82°19'52.7" E, 330 m a.s.l., on dry leaves of *Orostachys spinosa* (L.) C.A. Mey., pH 7.7, substrate samples coll. 13 July 2015, V.A. Vlasenko, moist chamber culture 2 June 2017, cult. and ident. A.V. Vlasenko, NSK 1016125 (paratype).

**Description.** Sporocarps either gregarious to rarely scattered, sessile, clustered, slightly angular, more rarely pillow-like or subglobose, adjacent sporangia walls not fusing, 0.1–0.5 mm in diam., golden brown. Hypothallus inconspicuous. Peridium single, golden brown, yellow in RL, dense, opaque, rough, spotted due to large wart-like structures of a golden colour. Lateral part of sporangium with a light golden line, not being the dehiscence line. Dehiscence occurring along predefined lines in the apical part of the sporocarps (visible only in SEM). Inner surface of the peridium ornamented with a small number of randomly arranged small warts (visible only in SEM). Capillitium well-developed, sometimes attached to the inner surface of the peridium, branched, forming an open network of light yellow, thin, tubular threads 1.2–2 µm in diam., surface ornamented with perforations and warts of different sizes (visible only in SEM). Capillitial threads resembling long tubes with transverse ring-like thickenings in a CM. Spore mass golden yellow to bright orange. Spores yellow, free, spherical,

**Fig. 3.** *Perichaena maculosa* (NSK 1016120, holotype): **A, B** – sporocarps (DM); **C–H** – spores and capillitial threads (CM). The arrows indicate the free ends of the capillitial threads. Bars: 500 µm (A); 200 µm (B); 20 µm (C–H). Photos by A. Vlasenko. ►









◀ **Fig. 4.** *Perichaena maculosa* (NSK 1016120, holotype, SEM): **A** – sporocarp; **C** – ornamentation of the inner part of the peridium; **D** – ornamentation of the spore; **G** – spore; **I** – capillitial thread. *Perichaena liceoides* (NSK 1016469, SEM): **B** – sporocarp; **E** – ornamentation of the spore; **F** – ornamentation of the inner part of the peridium; **H** – spore; **J** – capillitial thread. Bars: 50 µm (A, B); 1 µm (C, F); 500 nm (D, E); 2 µm (G–J). Photos by A. Vlasenko.

10–12 µm in diam. to oval, 9.5 × 11 µm, ornamented with evenly spaced large warts. In SEM, ornamentation consists of dense, abundant, evenly scattered, well-developed coralloid spines.

**Ecology.** Growing on dry leaves and at the base of shoots of drying succulent plants in rocky steppes (Fig. 8 E, F).

**Comments.** Morphologically, the new species is closest to *Perichaena liceoides* Rostaf. The ornamentation of the spores in these two species is very similar. The ornamentation of the spores of *P. maculosa* consists of dense, abundant, evenly scattered, well-developed coralloid spines (Fig. 4 D), while that of *P. liceoides* consists of dense, abundant, evenly scattered, well-developed coralloid spines with small warts scattered between them (Fig. 4 E). The capillitial threads of *P. maculosa* look like long tubes with transverse ring-like thickenings (Fig. 4 I), while those of *P. liceoides* are very short and underdeveloped (Fig. 4 J).

The new species can be easily distinguished from all known species of the genus *Perichaena* by its unique ornamentation of the peridium – spotted due to large wart-like structures of a golden colour.

The ML analysis based on the 18S nrDNA region (Fig. 7) showed that the new species of *Perichaena maculosa* is the sister clade to the species group *Perichaena depressa* Lib., *Perichaena quadrata* T. Macbr. and *Perichaena liceoides* Rostaf. The genetic distance of the *Perichaena maculosa* branch on the SSU tree is 0.0521, with 98% bootstrap support.

Sporocarps of *P. maculosa* have a light golden line on their lateral part, which is not the dehiscence line (Fig. 3 B). Sporocarps of *P. depressa* and *P. quadrata* have a light golden line on their lateral part, which is the dehiscence line. The ornamentation of the spores of *P. depressa* and *P. quadrata* consists of warts.

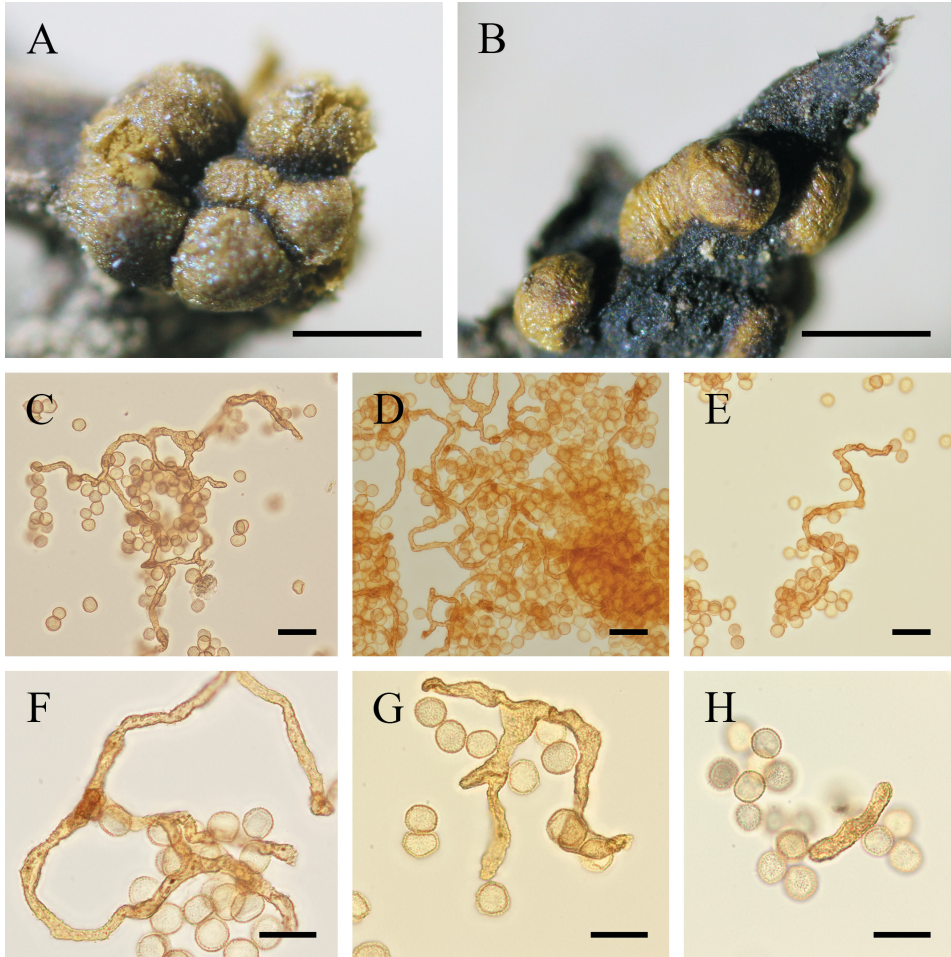
The monophyletic clade to which *P. maculosa* belongs also includes the species *P. depressa*, *P. quadrata* and *P. liceoides*. *Perichaena stipitata* and *P. luteola* are united into a separate clade, joined by the *Perichaena dictyonema* branch. *Perichaena agaves* and *P. corticalis* are united into a separate clade, joined by the *Perichaena nigra* branch. All the above listed species belong to the *Perichaena* s. str. group.



***Gulielmina olivacea*** A. Vlasenko et V. Vlasenko, **sp. nov.** Figs 5, 6: A, C, E, G, H  
Index Fungorum No.: IF900811

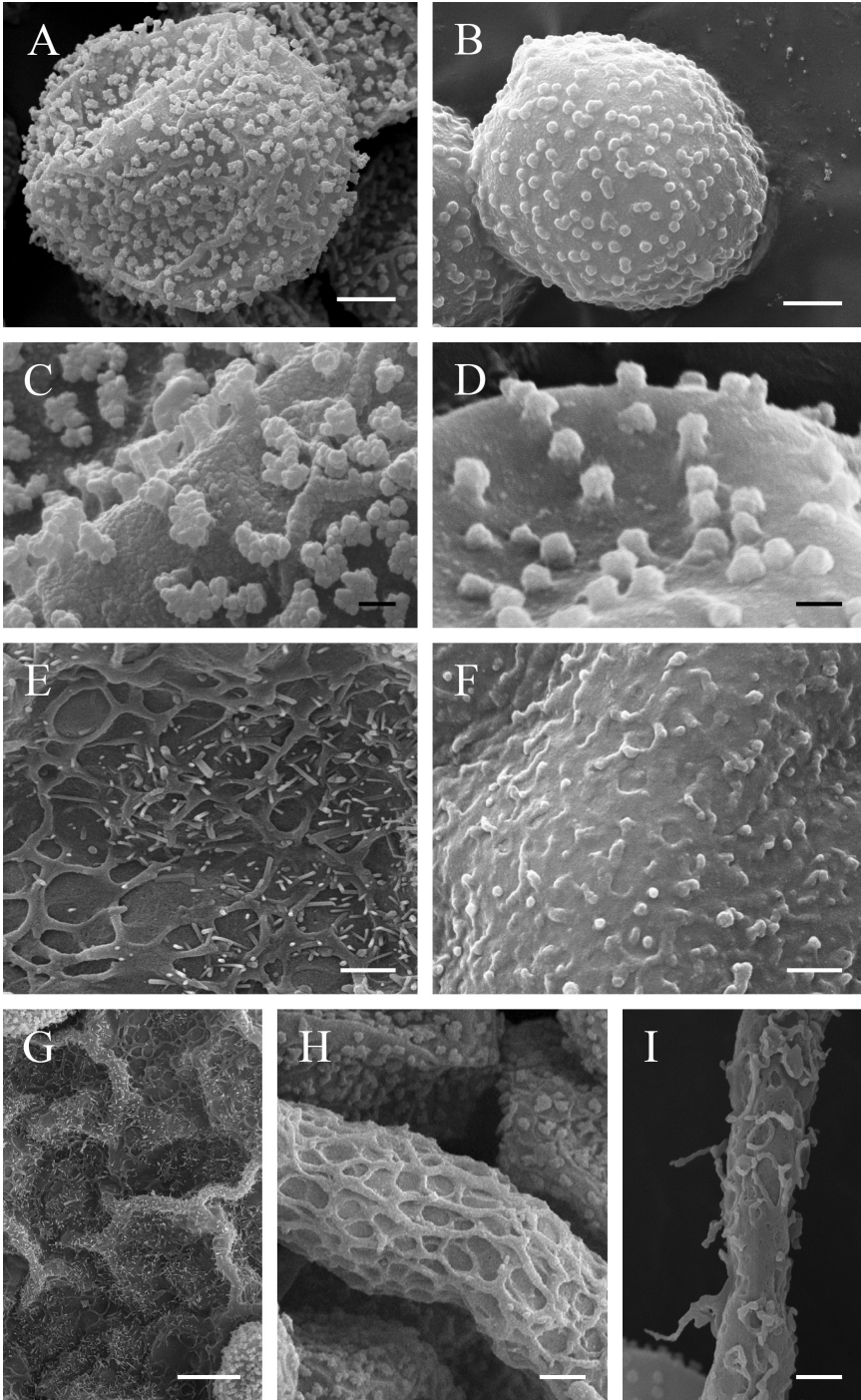
**Etymology.** Referring to the sporocarps and spore mass which have an olive-green colour.

**Type.** Russia, Novosibirsk Region, 12 km southeast of the village of Chistoozernoe, near Lake Chany, 54°40'43.0" N, 76°46'18.5" E, 105 m a.s.l., lake shore, halophyte community with *Salicornia*, on



**Fig. 5.** *Gulielmina olivacea* (NSK 1016035, holotype): **A, B** – sporocarps (DM); **C–H** – spores and capillitial threads (CM). Bars: 200  $\mu$ m (A, B); 30  $\mu$ m (C–E); 20  $\mu$ m (F–H). Photos by A. Vlasenko.

**Fig. 6.** *Gulielmina olivacea* (NSK 1016035, holotype, SEM): **A** – spore; **C** – ornamentation of the spore; **E, G** – ornamentation of the inner part of the peridium; **H** – capillitial thread. *Gulielmina vermicularis* (NSK 1016412, SEM): **B** – spore; **D** – ornamentation of the spore; **F** – ornamentation of the inner part of the peridium; **I** – capillitial thread. Bars: 2  $\mu$ m (A, B); 400 nm (C, D); 1  $\mu$ m (E, F); 5  $\mu$ m (G); 1  $\mu$ m (H, I). Photos by A. Vlasenko. ►



dry leaves of live *Salicornia perennans* Willd., substrate samples coll. 9 August 2014, V.A. Vlasenko, moist chamber culture 9 April 2015, cult. and ident. A.V. Vlasenko, NSK 1016035 (holotype), GenBank No.: OQ925673 (18s nrDNA); NSK 1016023, NSK 1016041 (isotypes).

Other sample examined. Russia, Novosibirsk Region, village of Kanavy, near Lake Chany, 54°41'52.4" N, 77°02'18.5" E, 101 m a.s.l., lake shore, steppe community, on dry dead leaves and at the base of lignified shoots of live *Limonium suffruticosum* L., substrate samples coll. 9 August 2014, V.A. Vlasenko, moist chamber culture 19 May 2015, cult. and ident. A.V. Vlasenko, NSK 1016026, NSK 1016792 (paratypes).

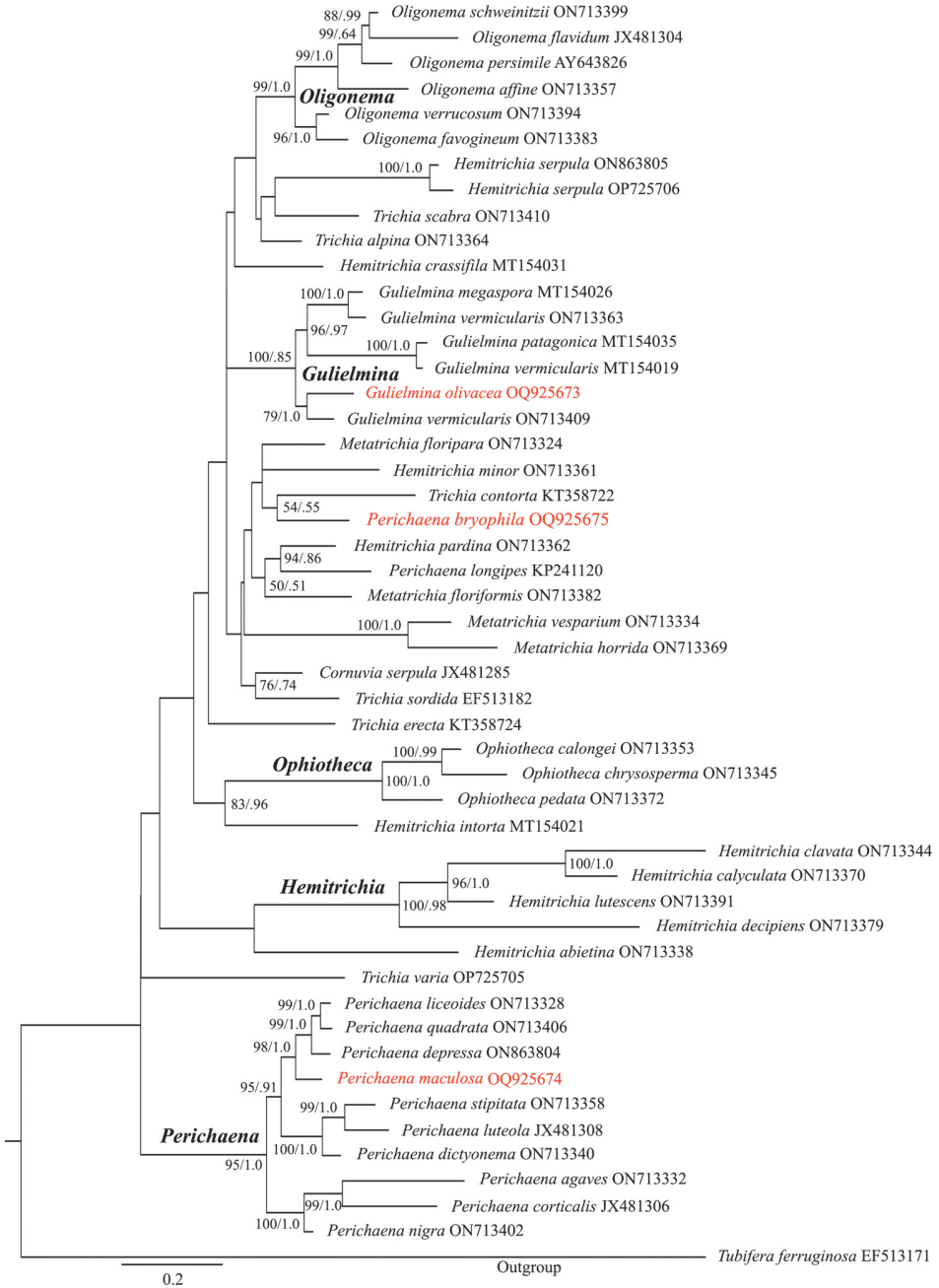
**Description.** Plasmodiocarps gregarious to rarely scattered, 0.2–0.5 mm wide, pillow-like, elongated, spherical, olive-yellow to greenish yellow, yellow-ochre if stored long in a herbarium. Peridium double, thin, the second layer only visible in SEM. Outer peridium layer shiny, smooth; inner peridium ornamented with a reticulum and bacula up to 0.5 µm in length. Capillitium bright yellow through yellow-orange to yellow-brown in TL. Capillitial threads of irregular shape, 4.5–8 µm in diam., with extensions up to 13.5 µm, anastomosed, branched, ornamented with a well-developed reticulum with infrequent small warts. Spore mass olive-yellow. Spores light yellow, spherical, 9.8–12.5 µm in diam. to oval, 10 × 13 µm, ornamented with unevenly spaced warts. When using SEM, warts are visible as low-stalked elements with a coral-like apical part.

**Ecology.** Growing on dry leaves and at the base of lignified halophyte shoots on solonetz soils and on steppe plants in saline steppes (Fig. 8 A, B, C, D).

**Comments.** Morphologically, the new species is closest to *Gulielmina vermicularis* (Schwein.) García-Cunch., J.C. Zamora et Lado. The ornamentation of the spores and the ornamentation of the inner surface of the peridium make it easy to separate these two species. Spores of *G. olivacea* are ornamented with warts with coral-like extensions at the top (Fig. 6 A, C), while the ornamentation of the spores of *G. vermicularis* consists of simple warts without extensions (Fig. 6 B, D). The inner peridium of *G. olivacea* is ornamented with a reticulum and bacula of up to 0.5 µm in length (Fig. 6 E, G), while that of *G. vermicularis* with warts (Fig. 6 F).

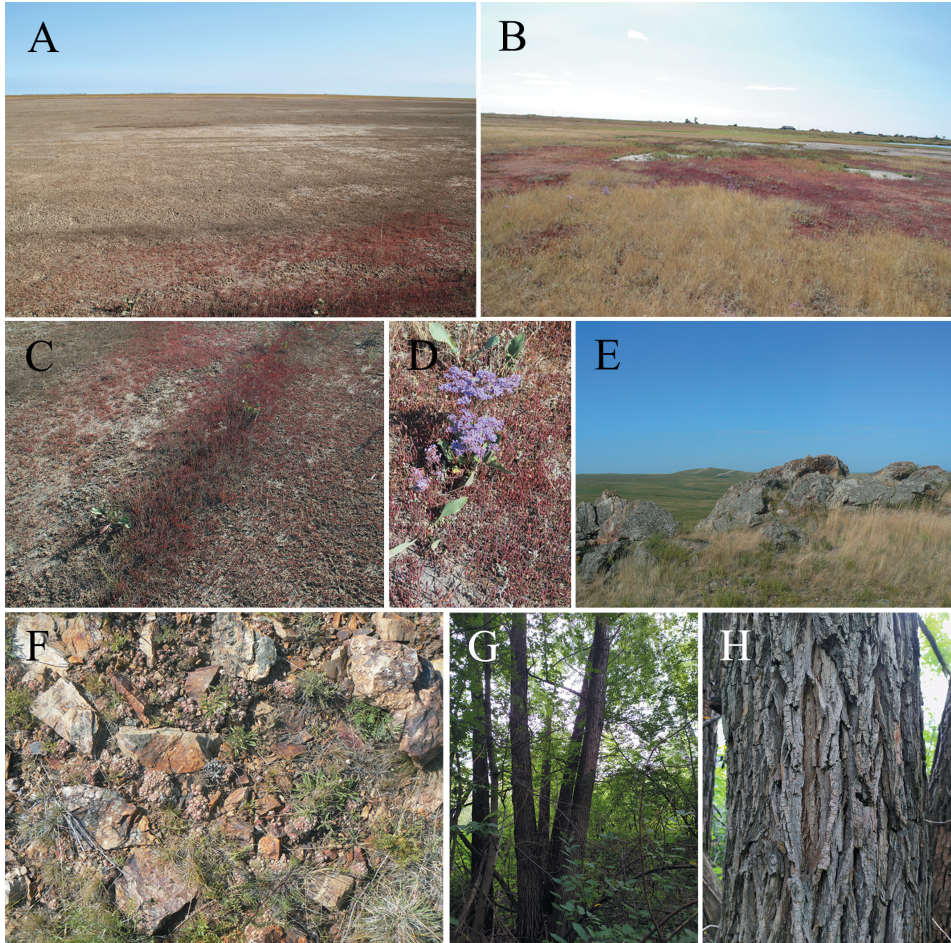
The new species can be easily distinguished from all known species of the genera *Perichaena* and *Gulielmina* by its olivaceous colour and unique ornamentation of the inner peridium consisting of bacula of up to 0.5 µm in length.

The ML analysis based on the 18S nrDNA region (Fig. 7) showed that the new species of *Gulielmina olivacea* is closest to *G. vermicularis* (ON713409). The genetic distance of the *Gulielmina olivacea* branch on the SSU tree is 0.0925, with 79% bootstrap support. Other sequences of *G. vermicularis* are located in a separate clade, along with *G. megaspora* and *G. patagonica*, with 96% bootstrap support. All the above listed species form the *Gulielmina* group. Apparently, the morphological species *G. vermicularis* is a complex of at least three cryptic phylogenetic species.



**Fig. 7.** The maximum likelihood tree based on 18S nrDNA sequences shows the phylogenetic relationships between the three new species and closely related species of bright-spored myxomycetes. BS values with high support and Bayesian posterior probabilities are shown in the branches. GenBank accession numbers are given after the species name.





**Fig. 8.** Habitats and substrates of new species of myxomycetes. **A** – Novosibirsk Region, near Lake Chany, near the village of Chistoozernoe, halophyte community; **B** – *ibid.*, near the village of Kanavy, halophyte community and saline steppe; **C** – *Salicornia perennans*; **D** – *Limonium suffruticosum* and *Salicornia perennans*; **E** – Altai Territory, near the village of Novofirsovo, petrophytic steppe; **F** – *Orostachys spinosa*; **G** – Novosibirsk Region, near the town of Novosibirsk, Zyryanka River, riparian forest; **H** – *Salix alba*. Photos by A. Vlasenko.

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