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The genus *Involucropyrenium* (Verrucariaceae, lichenized Ascomycota) in Russia

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Summary. A review of the lichen genus *Involucropyrenium* in Russia is presented. During the study of the lichen biota of the Magadan Region, *Involucropyrenium waltheri* (Kremp.) Breuss (Verrucariaceae) was identified on the basis of morphological and anatomical data. It is the first record to Russia both as a species and at genus level. A detailed description of the morphology and anatomy of the studied specimens is given. The arctic-alpine distribution of the species has been noted; it is rare in the world, mainly in northern and central Europe, and is rarely mentioned in Asia and North America.

Род *Involucropyrenium* (Verrucariaceae, лихенизированные Ascomycota) в России

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Ключевые слова: Магаданская область, пиренокарпный лишайник, распространение, российский Дальний Восток.

Аннотация. Представлен обзор рода *Involucropyrenium* в России. В ходе изучения лихенобиоты Магаданской обл. на основании морфологии и анатомии были выявлены новые для России род и вид – *Involucropyrenium waltheri* (Kremp.) Breuss, относящийся к семейству Verrucariaceae. Приведено подробное описание морфологии и анатомии изученных образцов. Отмечено арктоальпийское распространение вида, в мире он встречается редко, в основном в Северной и Центральной Европе, единично упоминается для Азии и Северной Америки.

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Introduction

The family Verrucariaceae Eschw. was described by Eschweiler in 1824. This is one of the largest families of lichenized fungi which according to modern classification (Lücking et al., 2017) includes about 940 species. The family is represented by crustose, squamulose, microfruticous and foliose pyrenocarpous lichens, non-lichenized and lichenicolous fungi with biseriate, colourless as well as dark brown, aseptate, septate or muriform ascospores. The species from Verrucariaceae are widespread, prefer rocky substrates, but can also be found on soil, plant debris or tree bark. In the world, they are widespread in mountainous areas, as well as tundra and arid communities. In Russia, 32 genera of the family are known (Urbanavichus, 2014), among which about half of the genera include terricolous species.

The genus *Involucropyrenium* Breuss was separated from the genus *Catapyrenium* Flot. by Othmar Breuss (Breuss, 1996), based on the presence of involucrellum and the position of the perithecia between the squamules. Nowadays, the genus includes 10 species (Breuss, 2010, 2016; Gromakova, Kondratyuk, 2017; Breuss, Türk, 2021). The genus is currently considered as a poorly studied. Some species are still known only from type localities or in no more than a few localities and are probably often confused with species of *Verrucaria* Schrad. (Prieto et al., 2010).

No special phylogenetic studies of the genus *Involucropyrenium* have been carried out. However, some phylogenetic reconstructions (Prieto et al., 2012; Gueidan et al., 2014) using *Involucropyrenium* sequences have confirmed the suggestion by Gueidan et al. (2007) that it belongs to the *Endocarpon* group. Members of the *Endocarpon* group including *Anthracocarpon* Breuss, *Endocarpon* Hedw., *Neocatapyrenium* H. Harada and some *Verrucaria* species are characterized by the presence of *Endocarpon*-type pycnidia (Gueidan et al., 2007; Prieto et al., 2012). According to preliminary analysis of nuLSU sequences from *Involucropyrenium* species, which is not shown in the study by Prieto et al. (2012), the *Involucropyrenium* is nested polyphyletically within the *Endocarpon* group.

The Natural Conditions of the Research Area

The Magadan Region is located in the extreme northeast of Russia and covers an area of 462.4 thousand km^2. The relief is dominated by mountains with a rugged topography, and only in places on the Okhotsk coast, as well as along river valleys, are small lowlands located. The climate in the region is sharply continental and harsh. Winters are long (up to 8 months), summers are cool, and there are frequent fogs. On the coast of the Sea of Okhotsk the climate is milder than in the interior parts of the region. So the average January temperatures on the coast of the Sea of Okhotsk are from –19 to –23 °C, and –38 °C in the interior parts of the region; July respectively +12 °C, +16 °C (Klyukin, 1970).

According to the geobotanical zoning of A. T. Reutt (1970), the study area belongs to the mountainous region of dwarf-pine and larch-birch forests of the Okhotsk coast and to the swamp-tussock-tundra region of the Yamsko-Tauya depression, and to the region of mountain-arctic tundras and lichen woodlands Kolyma Highlands.

Materials and Methods

The specimens were collected in the Magadan Region in 2021. The material was examined by the authors in the Laboratory of Lichenology and Bryology of Komarov Botanical Institute using standard microscopic techniques (Smith et al., 2009; Stepanchikova, Gagarina, 2014). Photographs of the species were taken with a stereoscopic microscope Motic SMZ-171-LED with an attached MotiCam S6 camera and Axio Scope.A1 with Axiocam 506 color camera. The study of ascospores and their measurement were made using the immersion oil under the magnification of 1000×. Results are given as (min.–)($\bar{x}$SD)–$\bar{x}$–($\bar{x}$+SD)(–max.), where min./max. are extremes from all measurements, $\bar{x}$ is the arithmetic mean observed, SD is the standard deviation. The number of measurements (N) is indicated in parentheses after the size of ascospores. Other measurements are presented as min.–max. or the maximum value observed. Geographical coordinates are given in spatial reference system WGS 1984.

The specimens were deposited in the lichen herbaria of the Komarov Botanical Institute of the Russian Academy of Sciences (LE), and Institute of Biological Problems of the North FEB RAS (MAG). The doublet specimens have been deposited in the herbaria of the Botanical Garden-Institute FEB RAS (VBGI) and Altai State University (ALTB).

Results

During the study of the lichen biota in the Magadan Region, we collected the specimens from

family Verrucariaceae which was determined in the laboratory as *Involucropyrenium waltheri* Breuss. It is the first record to Russia both as a species and at genus level. Below we provide a detailed description of the genus *Involucropyrenium* and the species *I. waltheri* indicating characteristic features.

Involucropyrenium Breuss, 1996, Ann. Naturhist. Mus. Wien, Ser. B, Bot. Zool. 98 (Suppl.): 38. – TYPE: *lnvolucropyrenium waltheri* (Kremp.) Breuss

Thallus small-squamulose to almost crustose, squamules thin (50–300 μm), adjacent or imbricate, attached to the substratum by colourless or brown rhizohyphae. Upper surface grey to beige or brown, matt; lower surface pale or dark. Upper cortex rather thin and poorly differentiated from the algal layer. Algal layer occupying almost the entire thallus. Medulla not clearly defined. Lower cortex developed or not. Ascomata perithecia, situated between the squamules, surrounded by an involucrellum (complete, dimidiate or apical), globose to slightly conical or pyriform. Exciple colourless, brown or black. Ostiole inconspicuous to somewhat dented and slightly paler in colour. Hamathecium of periphyses, interascal filaments absent. Asci clavate, thin-walled, non-amyloid, without an ocular chamber, 8-spored, with biseriate ascospores, simple, colourless, 11–27 × 6–13 μm. Photobiont green algae (Prieto et al., 2010; Prieto, 2017; Orange et al., 2023). Pycnidia were observed only in one species (*Involucropyrenium romeanum* (de Lesd.) Breuss) and were described as unilocular (*Endocarpon*-type) (Roux, 2005). Lichen substances detected by TLC were not identified (Orange et al., 2023).

It includes species occurring on calcareous and gypsiferous soils, in rock fissures or directly on limestones, also on artificial substrates as old bricks or mortar. They occur in a wide range of environments from semi-arid to alpine and temperate (Prieto et al., 2010; Prieto, 2017).

Differences between *Involucropyrenium* and others terricolous genera of *Catapyrenium* s. l. are presented in Table 1.

Involucropyrenium waltheri (Kremp.) Breuss, 1996, in Annln naturh. Mus. Wien, Ser. B, Bot. Zool. 98 (Suppl.): 38.

≡ *Verrucaria waltheri* Kremp., 1855, Flora, Regensburg 38: 69.

Type: [Germany], "Bayern, Karwendel, 26 VIII 1850, Krempelhuber" (lecto – M, not seen; iso – WU, not seen, Breuss, 1990, Stapfia 23: 137).

≡ *Catapyrenium waltheri* (Kremp.) Körb., 1855, Syst. lich. germ. (Breslau): 325. ≡ *Endocarpon cinereum* var. *waltheri* (Kremp.) Garov., 1872, Memor. R. Istit. Lombardo, 3 Ser. 3: 278. ≡ *Dermatocarpon waltheri* (Kremp.) Blomb. et Forss., 1880, Enum. Pl. Scand.: 97. ≡ *Involucrocarpon waltheri* (Kremp.) Servít, 1956, in Černohorsky, Nádvorník et Servít, Klic k Urcování Lisejníku ČSR 1: 32.

Description of studied specimens. Thallus squamulose. Squamules irregular, 0.3–1.0 mm in diam., flattened to slightly convex, tightly contiguous, forming a continuous thallus. Squamules margins entire to crenate-lobulate, firmly appressed, surrounded by a dark hypothallus. Upper surface brown to grey, matt. Lower surface dark with dark rhizohyphae. Thallus 80–200 μm thick. Upper cortex paraplectenchymatous, up to 20 μm thick, consists of 1–2 layers of cells. Epinecral layer absent. Algal layer distributed over nearly the entire thallus, with algal cells 6–10 μm in diam. Medulla not clearly differentiated. Lower cortex composed of polygonal cells, dark. Rhizohyphae dark brown, 4–5 μm thick. Perithecia globose, 0.10–0.25 mm in diam., often aggregated, developed on the hypothallus between the squamules, 1/3–2/3-immersed in the hypothallus. Exciple brown to black with an entire carbonaceous involucrellum. Ostiole inconspicuous, rarely visible as a slightly paler dot. Hamathecium formed by short periphyses located in the ostiolar canal, interascal filaments absent. Asci clavate, non-amyloid, 8-spored. Ascospores simple, broadly ovate to elongate, biseriately arranged, (15.0–)15.5–17.4–19.3(–22.0) × (6.0–)6.8–7.4–8.0(–9.0) μm (N = 50). Pycnidia not seen.

Ecology. The species has a typical arctic-alpine distribution pattern. It occurs predominantly on calcareous soil, humus and plant debris (Breuss, 1990; Sun et al., 2008; Øvstedal et al., 2009; Prieto et al., 2010; Roux, 2012). Only in Iran the species is recorded on limestone (Baradaran et al., 2023). Our discovery of the species in the Magadan Region was made in a mountainous region, where the species grew on the soil, that corresponds to its habitat.

Distribution. It is reported from Europe – Svalbard, Norway, Sweden, Great Britain (Scotland), Netherlands, Germany, France, Poland, Czech Republic, Switzerland, Austria, Spain, Italia (Breuss, 1990; Aptroot, Kortselius, 2002; Øvstedal et al., 2009; Prieto et al., 2010; Roux, 2012); Asia – Iran (Baradaran et al., 2023), China (Sun et al., 2008); Greenland (Breuss, Hansen, 1988); North America – USA (Colorado, New Mexico) (Anderson, 1974).

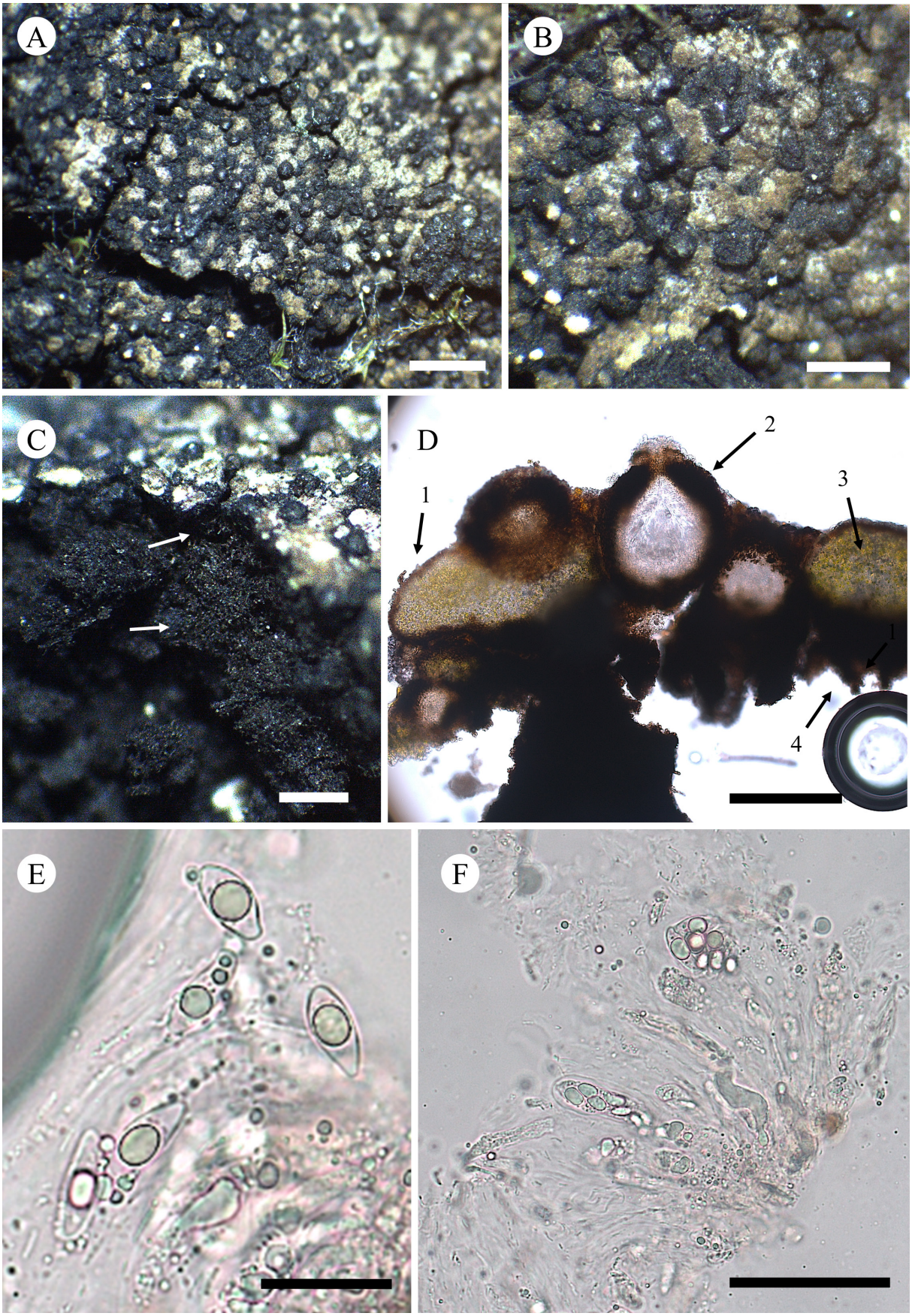

Fig. 1. *Involucropyrenium waltheri* (LE L-24770): A and B – thallus with perithecia growing between the squamules; C – hypothallus with dark rhizohyphae (indicated by arrows); D – section through the thallus and perithecia (1 – squamules; 2 – perithecia with involucrellum; 3 – algal layer, 4 – dark rhizohyphae); E – ascospores; F – asci with ascospores. Scale bar: A = 1 mm; B and C = 0.5 mm; D = 100 μm; E = 20 μm; F = 50 μm

Table 1

Differences between terricolous genera of *Catapyrenium* s. l.

Genus / Features	*Involucropyrenium*	*Anthracocarpon*	*Catapyrenium*	*Clavascidium*	*Heteroplacidium*	*Neocatapyrenium*	*Placidium*
Thallus	small-squamulose to almost crustose	squamulose	squamulose	squamulose	crustose-areolate to small squamulose	squamulose to cushion-like	squamulose
Position of perithecia	between the squamules	within the squamules	within the squamules	within the squamules	within the areoles or squamules	within the squamules	within the squamules
Involucrellum	present	absent	absent	absent	absent	absent	absent
Medulla	not clearly defined	clearly	clearly	clearly	clearly	clearly	clearly
Rhizohyphae	present	present	present	present	present	absent	present
Rhizines	absent	present	absent	present	absent	present	absent
Pycnidia type	*Endocarpon*-type*	*Endocarpon*-type	absent	*Dermato-carpon*-type	*Dermato-carpon*-type	*Endocarpon*-type	*Dermato-carpon*-type

Note: * – data based on the study of pycnidae only in *Involucropyrenium romeanum*.

Specimens examined. RUSSIA. ***Magadan Region***: "Khasynskiy District, Olskoe Plateau, surroundings of the upper reaches of the Tiran Creek, alt. 1094 m. 60°37'43.9"N, 151°31'06.4"E, rocks, on soil. 01 VII 2021. S. V. Chesnokov, L. A. Konoreva" (LE L-24770, VBGI 160864, ALTB); ibid., "vicinity of Yablonevy Pass. 60°42'N, 151°45'E, rocky placers on the slope, on the soil on stones. 01 VII 2021. E. V. Zheludeva" (LE L-24771, MAG Kh-5095).

Discussion

The studied specimens correspond to the description in the protologue (Krempelhuber, 1855) and literature data (Breuss, 1990; Prieto et al., 2010; Prieto, 2017; Orange et al., 2023). However, the sizes of ascospores in our samples are somewhat smaller compared to the literature: 15.5–19.3 × 6.8–8.0 μm according our data, 17.0–21.0 × 8.0–10.0 μm according Breuss (1990), Prieto et al. (2010), Prieto (2017), Orange et al. (2023).

Morphologically, *Involucropyrenium waltheri* may be confused with *I. sbarbaronis* (Servít) Breuss and *I. tremniacense* (A. Massal.) Breuss which also have small squamules, irregularly shaped and perithecia developing from the black hypothallus between the squamules (Table 2). However, *I. waltheri* has a darker thallus color, dark brown rhizohyphae, lower cortex, broadly ovate to elongate spores and globose perithecia with complete involucrellum. While *I. sbarbaronis* and *I. tremniacense* have a beige to pale brown thallus, colourless rhizohyphae and lack of lower cortex (Breuss, 1990). In addition, the perithecia of *I. sbarbaronis* are the same as those of *I. waltheri* with complete involucrellum, but in the later stages of development they become pyriform, and the ascospores are ellipsoid or elongate-ovoid (Breuss, 1990). Perithecia of *I. tremniacense* spherical with apical involucrellum, the latter reaching at most to the center of the perithecia, ascospores ellipsoid (Breuss, 1990; Prieto et al., 2010).

Ecologically, due to its arctic-alpine distribution, *Involucropyrenium waltheri* is also clearly separated from these meditrerranean species (Table 1).

It can resemble *Catapyrenium cinereum* (Pers.) Körb., but it differs by the presence of the involucrellum which fuses with the dark excipulum, and its perithecia develop from a black hypothallus between the squamules. The squamules are small (up to 1.5 mm in diam.), roundish, and densely aggregated to form a thin cartilaginous crust over the substrate. In *C. cinereum*, the perithecia develop within the squamules and lack an involucrellum. The excipulum is initially colourless, changing to blackish-brown at

maturity (Table 2). The squamules of *C. cinereum* are dispersed to continuous, larger than in *Involucropyrenium waltheri* (up to 3 mm in diam.), and more or less distinctly incised (Breuss, 1990; Prieto et al., 2010; Orange et al., 2023).

Table 2

Differences between morphologically similar *Involucropyrenium* and *Catapyrenium* species

Species / Features	*Involucropyrenium waltheri*	*Involucropyrenium sbarbaronis*	*Involucropyrenium tremniacense*	*Catapyrenium cinereum*
Thallus color	brown to grey	beige to pale grey-brown	beige to pale brown	whitish, rarely greenish grey or brownish
Rhizohyphae	dark brown	colourless	colourless	brown to black
Lower cortex	present	absent	absent	present
Perithecia form	globose	pyriform	globose	subglobose
Position of perithecia	between the squamules	between the squamules	between the squamules	within the squamules
Involucrellum	complete	complete	apical	absent
Form of ascospores	broadly ovate to elongate	ellipsoid or elongate-ovoid	ellipsoid	clavate
Distribution	arctic-alpine	mediterranean	mediterranean	arctic-alpine

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