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The second record of the rare freshwater lichen, *Anisomeridium carinthiacum*, to Russia from the south of Far East

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Summary. During the study of the lichen biota of Crylion Cape (Sakhalin Is.), the rare amphibious freshwater lichen growing on siliceous rocks, *Anisomeridium carinthiacum*, was identified on the basis of morphological and anatomical data. This is the second record to Russia after the Caucasus Mts. and the first record for the Russian Far East and Sakhalin Is. The ecology, distribution and differences from the morphologically similar pyrenocarpous freshwater lichen species from East Asia are discussed. The arguments for the conversion of *Anisomeridium japonicum* described from Japan into synonyms of *Anisomeridium carinthiacum* are given.

Вторая находка редкого пресноводного лишайника, *Anisomeridium carinthiacum*, в России с юга Дальнего Востока

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Ключевые слова: гигрофильный вид, о. Сахалин, пиренокарповые лишайники, распространение, флористическая находка.

Аннотация. В ходе изучения лихенобиоты мыса Крильон (о. Сахалин) на основании морфологических и анатомических данных был выявлен редкий пресноводный лишайник, произрастающий на погруженных или периодически затопляемых силикатных камнях – *Anisomeridium carinthiacum*. Это вторая точка в России и первая для Дальнего Востока России и о. Сахалин. Обсуждаются экология, распространение и различия с морфологически близкими видами пиренокарповых пресноводных лишайников из Восточной Азии. Приведены аргументы для сведения *Anisomeridium japonicum*, описанного из Японии, в синонимы *Anisomeridium carinthiacum*.

Introduction

Anisomeridium (Müll. Arg.) M. Choisy is a genus comprising ca. 80 mostly tropical to warm-temperate species (Wijayawardene et al., 2022). The representatives of the genus occur usually on bark but also on lignum, mosses and on calcareous or non-calcareous rocks (Aptroot, 2002; Nimis, 2016; Cannon et al., 2023). The genus combines crustose species associated with a trentepohliod photobiont (some are non-lichenized) and possess thin thalli, black perithecia with the cellular structure of the wall, not containing bark cells, branched and anastomosing pseudoparaphyses, without periphyses, hymenial gel always I–. Asci cylindrical-clavate, K/I–, fissitunicate, thickened above, apex with

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a broad ocular chamber and 1-3-septate, relatively broad, ovoid ascospores without perispore (Harris, 1995; Orange, 2013; Cannon et al., 2023). The genus is represented in Russia by four species – epiphyte *A. biforme* (Borrer) R. C. Harris, *A. macrocarpum* (Körb.) V. Wirth and *A. polypore* (Ellis et Everh.) M. E. Barr., which could also occur on shaded siliceous rocks, as well as saxicolous *A. carinthiacum* (J. Steiner) R. C. Harris, inhabiting deeply shaded, siliceous rocks on the banks of streams and rivers (Urbanavichus, 2010; Urbanavichus, Urbanavichene, 2013, 2015; Himelbrant et al., 2018; Urbanavichus et al., 2020).

Anisomeridium carinthiacum is regarded as extremely rare and is treated as "Heritage of international interest. In danger critical extinction" (Roux et al., 2020). The expert on freshwater lichens, H. Thüs also noted the extremely rarity of the species (pers. comm.; Thüs, Schultz, 2009). In Russia, *A. carinthiacum* was recorded for the first time from Caucasus Mts. (Urbanavichus, Urbanavichene, 2013) and since that no records of this rare amphibious lichen were published. In this paper, *A. carinthiacum* is newly reported for the Russian Far East. The ecology, distribution and differences from the morphologically close species occurring in East Asia are discussed.

Materials and Methods

The specimen was collected by E. A. Davydov in Crylion Cape of Sakhalin Is. in 2023. The specimen was examined using a stereomicroscope (Zeiss Stemi 2000-C) and a compound microscope (Zeiss Axio Lab.A1). Anatomical examination was undertaken using hand cut sections mounted in water with following reagents (R): 10 % KOH (K) and Lugol's solution (I). Measurements of ascospores, perithecia and asci are presented as: (extreme minimum) minimum – maximum (extreme maximum). Measurements of other parameters are given as maximum value observed. The measurements of anatomical structures were made to the nearest 0.5 μm. Number of measurements is given for ascospores and asci in brackets [*n*]. Lichen substances were studied using spot tests with potassium hydroxide solution (K), sodium hypochlorite solution (C) and 1,4-p-phenylendiamine (PD), and by high performance thin-layer chromatography (TLC) with solvent systems A (toluene: 1,4-dioxane: acetic acid, 180 : 45 : 5), B' (hexane: methyl tert-butyl ether: formic acid, 140 : 72 : 18) and C (toluene: acetic acid = 170 : 30) following Orange et al. (2001). The specimen was deposited in the lichen herbaria of Altai State University (ALTB).

Results

The studied sample is fully corresponded to the protologue (Steiner, 1913) and more detailed descriptions of the species (Harada, 1999a; Thüs, Schultz, 2009; Wirth et al., 2013; Liu et al., 2018). Below we provide the description of the species based on studied specimen from the Russian Far East.

Anisomeridium carinthiacum (J. Steiner) R. C. Harris, 1987, in Egan, Bryologist 90(2): 163 (Fig. 1).

≡ *Arthopyrenia carinthiaca* J. Steiner, 1913, Öst. bot. Z. 63: 335.

≡ *Paraphysothele carinthiaca* (J. Steiner) Keissl., 1937, Rabenh. Krypt.-Fl., Edn 2 (Leipzig) 9(1.2): 212.

≡ *Ditremis carinthiaca* (J. Steiner) R. C. Harris, 1990, Some Florida Lichens (New York): 32. – Syntypes: "Austria. Carinthia. On the north side of the Kreuzberg near Klagenfurt, a small forest stream south of the peatlands near St. Primus, on stones (phyllite) in running water. Steiner. 1882; in the pond drain north of the «cold cellar», on stones (phyllite) in running water, Steiner, 1884" (W, not seen).

= *Anisomeridium japonicum* Harada, 1999, Nat. Hist. Res. 5(2): 53. – Holotype: "Japan. Honshu. Chiba-ken, Futtsu-shi, Toyooka, Matsubushi, 130 m alt., on rock, 14 XII 1997, Harada 18687" (CBM-FL-10743, digital photo!).

Description of studied specimens. Thallus crustose, greenish-grey, continuous to rimose, up to 35 μm thick, not subgelatinous when wet, without a distinct prothallus. Perithecia black, 50–125 μm across, 1/2 immersed with exposed uppermost part. Involucrellum present, exciple of cellular hyphae, brown-black in upper part, pale or colourless in lower part; pseudoparaphyses slender, branched and anastomosing, long-celled, ca. 1 μm thick; periphyses absent; hymenial gel I–. Asci 8-spored, (35.0–)37.0–41.0 × 9.5–12.0(–14.0) μm [n = 10], cylindrical, K/I–, fissitunicate, the apical dome with an indistinct ocular chamber, with biseriately arranged ascospores. Ascospores 1-septate, with equal or slightly unequal cells, hyaline, (10.0–)11.0–13.0(–14.5) × 4.0–6.0 μm [n = 25]. Pycnidia unknown. Photobiont trentepohlioid. Spot tests: all negative.

Chemistry: no lichen substances were detected by TLC.

Distribution and ecology. An apparently rare, amphibious freshwater species growing submerged or slightly above water level on siliceous rocks and pebbles, restricted to shaded sites in upland areas (Thüs, Schultz, 2009). It is known from Northern, Central and Western Europe, North America, Australia and New Zealand (Noble, 1982; Galloway, 2007; Thüs, Schultz, 2009; Hafellner, Türk 2016; McCune, 2017; Nimis et al., 2018; Roux et coll., 2020; Westberg et al., 2021). In Asia the species is recorded in Caucasus (Urbanavichus, Urbanavichene, 2013), Taiwan (Aptroot, 2003), Vietnam (Aptroot, Sparrius, 2006), Japan (Harada, 1999a, 2016a; Ohmura, Kashiwadani, 2018, as *A. japonicum*), South Korea (Liu et al., 2018, as *A. japonicum*) and Sakhalin Is. (here). It was collected with an accompany species: *Bacidia inundata* (Fr.) Vězda and *Verrucaria* spp.

Specimen examined: "Russia, Sakhalin Region, Sakhalin Island, Anivsky District, Crylion Cape, Aniva Bay, mouth of Riphlyanka River, rapids on the river, 46°06'07.22" N, 142°12'01.08" E, 1 m a. s. l., siliceous pebble in water, 24 VIII 2023, E. A. Davydov 19606" (ALTB).

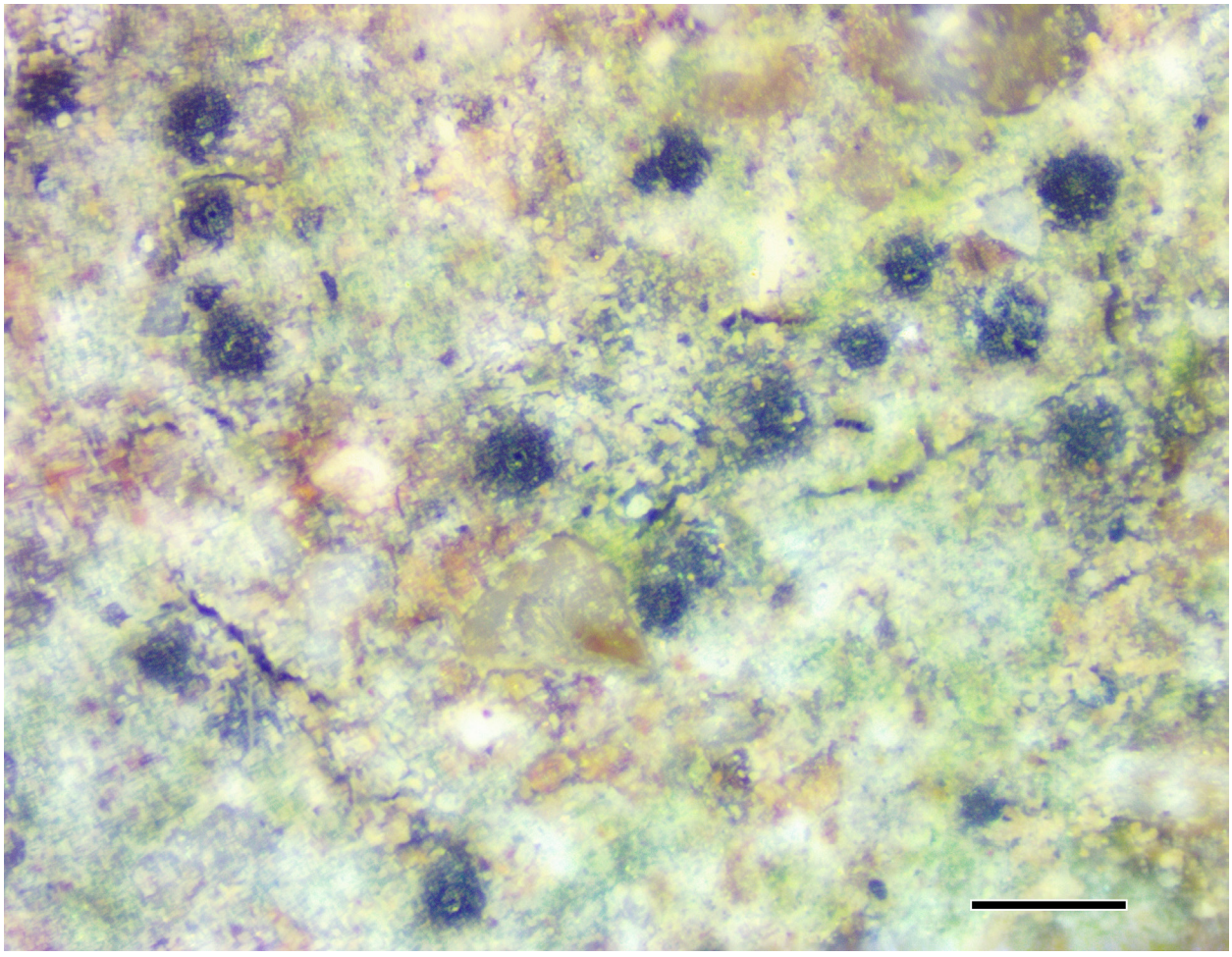

Fig. 1. *Anisomeridium carinthiacum* (ED 19606, ALTB) – thallus and apothecia. Scale bar = 0.2 mm.

Discussion

Anisomeridium carinthiacum was described from Austria (Steiner, 1913) and then was re-described under the name *Anisomeridium japonicum* from Japan (Harada, 1999a). Our comparison of protologues showed that both species have no morphological or ecological differences. Previously, H. Thüs and M. Schultz (2009) came to the same conclusion, in particular, based on the examination of the type material (H. Thüs, pers. comm.), since the range of the ascospores is slightly larger, 9–15 × 4–6 µm (Thüs, Schultz, 2009) compared with the protologue, 9.0–12.0(–13.0) × 4.5–6.0 µm (Steiner,

1913). Describing his new species, H. Harada pointed out that *Anisomeridium japonicum* with ascospores of 9–16 × 4–6 μm differed from *A. carinthiacum* by larger ascospores which had been erroneously compared with North American material (Harris, 1995). In addition to slightly larger ascospores of (11.0–)13.0–18.0 × 5.5–7.0 μm, R. C. Harris (1995) reported "well-developed thallus with often black prothallus", whereas the thallus is thin and without prothallus in material from Europe and East Asia. According to later data from North America (Noble, 1982; McCune, 2017), the ascospores size does not overlap (16–18 × 6–7 μm) and the perithecia are extremely small, less than 0.1 mm in diam., compared to material from Europe and East Asia. It remains an interesting open question whether the data presented for the North American collections of freshwater *Anisomeridium* with their slightly larger ascospores and thallus variability can still be attributed to *A. carinthiacum*, or whether the North American collections represent a different, still undescribed taxon. From the other hand, if the material from Europe is homogenous, since T. Foucard (2001) mentioned a wider range of ascospores 11–18 × 4–7 μm and perithecia (0.3 mm in diam.). Since the name *Anisomeridium japonicum* was used in the current literature from East Asia (Harada, 2016a; Liu et al., 2018; Ohmura, Kashiwadani, 2018), here we provide arguments proving the necessity of reducing the species to synonym of *Anisomeridium carinthiacum*.

Anisomeridium carinthiacum is well distinguished from another freshwater pyrenocarpous species occurring in East Asia, among which Verrucariaceae Eschw. includes a majority of the species. The presence of trentepohlioid photobiont with its yellowish pigmentation (which fades however after long storage) is a constant character for *A. carinthiacum* in opposite to a range of different species of freshwater Verrucariaceae. From the numerous freshwater representatives of *Hydropunctaria* C. Keller et al. and *Verrucaria* Schrad., known from East Asia, *A. carinthiacum*, in addition, is generally distinguished mostly by its septate ascospores and persistent, branched pseudoparaphyses. In contrast to species of *Staurothele* Norman and *Endocarpon* Hedw., *A. carinthiacum* is characterized by the absence of hymenial algae and muriform ascospores. The representatives of *Polyblastia* A. Massal. and *Sporodictyon* A. Massal., in opposite to *A. carinthiacum*, have muriform or more then transversely 1(–3)-septate ascospores. The freshwater species, *Thelidum sinense* H. Harada et Li S. Wang, known from China (Harada, Wang, 2006) and Japan (Harada, 2013b), like *A. carinthiacum*, has a thin thallus 30–50 μm thick, small perithecia 0.15–0.25 mm in diam. and hyaline, 1-septate ascospores, (11–)13–15(–17) × 5–7(–8) μm but unlike of *A. carinthiacum* has a brown granulose thallus, well-developed periphyses and disintegrated paraphyses. Another freshwater *Thelidium*, *T. rehmii* Zschacke known from Japan (Harada, 2013b) and Germany (Thüs, Schultz, 2009), *T. izuense* H. Harada described from Japan (Harada, 2013b) and widespread *T. minutulum* Körb. also known from China (Harada, Wang, 2006) and South Korea (Moon, 2013), are characterized, in addition to the differences of the genus, by larger ascospores, 19–31 × 8–15 μm, 21–23 × 10–11 μm, and (12–)15–25(–32) × (4–)8–10(–15) μm, correspondingly, and the absence of the involucrellum to the latter.

A thin continuous thallus associated with a trentepohlioid photobiont with semi immersed black perithecia up to 0.3 mm in diam. has *Porina chlorotica* (Ach.) Müll. Arg. known from siliceous substrata at shady and humid sites and in the splash zone of streams in Japan (Harada, 2016b), China (Wei, 2020), South Korea (Moon, 2013), in contrast of *A. carinthiacum* has larger 14.0–27.0 × 4.5–7.0 μm, transversely 3-septate ascospores as well as simple paraphyses and unthickened, unitunicate ascus. *Porina ulceratula* Zahlbr. known from Japan, Indonesia and Australia (McCarthy, 1993; Harada, 2015), growing on acidic rocks in the semi-aquatic freshwater habitats, is morphologically distinguished from *A. carinthiacum* by its brownish thallus and concolor with the thallus, larger perithecia (0.25–0.5 mm diam.) as well as anatomically mainly by its transversely 3-septate ascospores (18.0–30.0 × 5.0–6.5 μm) and lack of a distinct involucrellum. Saxicolous species on periodically submerged rocks in mountain streams in Japan (Harada, 2015), *Porina tosaensis* H. Harada, described from Japan (Harada, 2015), is similar to *A. carinthiacum* in having a continuous, very thin, green-brown or brownish green thallus and black perithecia 0.15–0.3 mm in diam. but is distinguished by its larger, 21–27 × 4–6 μm, transversely 5-septate ascospores in addition to the differences of ascus and hamathecium structure. *Porina guentheri* (Flot.) Zahlbr. with olive green to purplish grey continues to finely rimose thallus associated with *Trentepohlia* and black perithecia 0.15–0.4(–0.7) mm in diam. is known on shaded acidic rocks in forests in Japan (Harada, 2016b), Vietnam (Aptroot, Sparrius, 2006), South Korea (Moon, 2013) and China (Wei, 2020), but in Europe

the species has been recorded on stable siliceous boulders in the splash water zone at or in shaded streams (Thüs, Schultz, 2009). The last species is easily distinguished from *A. carinthiacum*, mainly by its larger, 27.0–42.0 × (5.5–)6.0–8.0 μm, transversely 5–7(–8)-septate ascospores (Harada, 2016b).

According to Harada (2016c), *Strigula minutula* P. M. McCarthy, described from Australia (McCarthy, 1995), grows in Japan in forest gorges on periodically inundated acidic rocks with *Anisomeridium* sp. *Strigula minutula* is similar to *A. carinthiacum* in having a thin continuous, green-brown thallus with small black perithecia 0.15–0.25 mm in diam. and hyaline 1-septate ascospores but is distinguished by smaller size of ascospores 9.0–10.0 × ca. 2.5 μm and unbranched paraphyses as well as sometimes perithecia are covered by a thin thalline layer up to the ostiole. *Strigula aquatica* H. Harada (1999c), mentioned in the same habitats as *S. minutula* and *Anisomeridium* sp. (Harada, 2016c), is well distinguished morphologically from *A. carinthiacum* by a brownish thallus, larger perithecia (0.15–)0.25–0.6 mm in diam. and ascospores 26.0–43.0 × 6.0–9.0 μm, transversely (6–)8(–9)-septated (Harada, 2016c). *Strigula australiensis* P. M. McCarthy known from semi-aquatic habitats in Japan (Harada, 2016c) and Australia (McCarthy, 1995) is easily distinguished from *A. carinthiacum* by its submuriform ascospores. *Agonimia deguchii* H. Harada described from semi-aquatic freshwater habitats from Japan (Harada, 2013a) has small black perithecia ca. 0.2 mm in diam., but it is easily distinguished from *A. carinthiacum* by its squamulose thallus and large muriform ascospores.

Cyanopyrenia japonica H. Harada growing in freshwater habitats on siliceous rocks in Japan (Harada, 1995) has a green continuous thallus like *A. carinthiacum* but is easily distinguished by its cyanobacterial photobiont, non-septate ascospores, asci without distinct apical thickening and morphologically by red-brown perithecia. Another freshwater cyanobiont species, *Collemopsidium japonicum* (H. Harada) H. Harada described as *Pyrenocollema japonicum* H. Harada (1999b) is similar to *A. carinthiacum* by its 1-septate ascospores and small black perithecia, 0.15–0.2 mm in diam. but differs, besides the type of the photobiont, by its purple-brown thallus with depressed perithecia at the ostiole and larger ascospores, 15.0–21.0 × 6.0–8.0 μm.

The several species currently accepted under *Anisomeridium*, including *A. carinthiacum*, have been used under *Arthopyrenia* A. Massal. since Harris (1973) pointed out the heterogeneity of the genus. Using the National Handbook of the lichens (Makarevich, 1977) it might be keyed as *Arthopyrenia lomnitzensis* Stein which occurs "on granite rocks irrigated with water, in mountains, on thalli *Ionaspis odora*". A. Orange (2002) indicated the species as lichenicolous on the thallus and hymenium of *Ionaspis odora* (Ach. ex Schaer.) Th. Fr. and *I. lacustris* (With.) Lutzoni and made a combination *Sagediopsis lomnitzensis* (Stein) Orange, although Keissler (1938) and Makarevich (1977) treated the species as lichenized, with thin continuous thallus (C+ violet and K+ yellowish) associated with trentepohlioid photobiont which is however also characteristic to *Jonapsis odora* and *J. lacustris*. *Anisomeridium carinthiacum* might be keyed as *Arthopyrenia inconspicua* J. Lahm in Körber from in the same key (Makarevich, 1977). The species was combined as *Lichenopeltella inconspicua* (J. Lahm) Cl. Roux (Roux et al., 2020) and then as *Lichenopeltella inconspicua* (Nyl.) Cl. Roux (Roux, 2020) since Roux (2020) indicated that *Arthopyrenia inconspicua* J. Lahm. is corrected to *Arthopyrenia inconspicua* (Nyl.) J. Lahm ex Körb. The species is also treated as lichenicolous on the thallus of *Verrucaria muralis* Ach. and probably other calciphilous *Verrucaria* spp., although the species is also connected with the water since in his protologue Nylander provided an important ecological information "on wet stones" (Roux et al., 2020). Using Foucard (2001) it might be keyed as *Arthopyrenula pygmaea* Degel. which is differs from *A. carinthiacum* by indistinct paraphyses and occurrence of apothecia and *A. angermanica* Degel., which differs by its larger ascospores, 17.0–21.5 × 8.5–10.5 μm as well as Nordin (2002) indicated that the species has cyanobacterial photobiont and thus proposed a new combination, *Collemopsidium angermannicum* (Degel) A. Nordin.

The specimen of the very inconspicuous *Anisomeridium carinthiacum* was collected purely by chance. With a high probability, the species can be found in other region within East Asia and the Russian Far East in the cites with suitable ecological conditions.

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