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BIODIVERSITY STUDIES

Towards a study of the genus *Acarospora* A. Massal. (Acarosporales, Ascomycota) in the Magadan Region (Russia)

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ABSTRACT

During the revision of the genus *Acarospora* in the Magadan Region, *A. versicolor* was identified for the Russian Far East for the first time based on morphological, anatomical and molecular studies. *Acarospora glaucocarpa* and *A. sinopica* are reported for the Magadan Region for the first time. Detailed descriptions are given for each species, and distribution data are clarified. Differences from related species are discussed.

Keywords: *Acarospora glaucocarpa*, *Acarospora sinopica*, *Acarospora versicolor*, Asia, biodiversity, distribution, saxicolous lichens, Far East

РЕЗЮМЕ

Панькова В.В., Гагарина Л.В., Желудева Е.В., Чесноков С.В., Конорева Л.А. К изучению рода *Acarospora* A. Massal. (Acarosporales, Ascomycota) в Магаданской области (Россия). В ходе ревизии рода *Acarospora* в Магаданской области впервые для Дальнего Востока России на основе морфологических, анатомических и молекулярных исследований был выявлен вид *A. versicolor*. *Acarospora glaucocarpa* и *A. sinopica* впервые приводятся для Магаданской области. Для каждого вида приведены детальные описания, уточнены данные по распространению. Обсуждаются отличия от близких видов.

Ключевые слова: *Acarospora glaucocarpa*, *Acarospora sinopica*, *Acarospora versicolor*, Азия, биоразнообразие, Дальний Восток, распространение, эпилитные лишайники

The territory of the Magadan Region is characterized by various forms of relief: ridges, highlands, plateaus, depressions and coastal plains (Kolyma Highlands – maximum altitude 1962 m a.s.l., Olskoye Plateau 1400–1600 m a.s.l.). The Magadan Region is located in the tundra and forest-tundra climate zone (subarctic climate zone) (Klyukin 1970). The mean annual temperature in the Magadan Region varies from -2.8 to -11(-13)°C . Geological structures are represented by andesites, basalts, tuffs, as well as sandstones, siltstones and mudstones (Korolkov 1992).

The object of our study is lichens of the genus *Acarospora* in the Magadan Region. The genus *Acarospora* is characterised by numerous, usually tiny, ascospores in the asci, the presence of a thalline margin and K/I- ascus tip. According to Urbanavicius (2010), there are about 60 species of this genus in Russia, 25 of which occur in the Far East. However, the currently available information on *Acarospora* in the Magadan Region is rather scarce. Although, considering the presence of active mountainous relief in Magadan Region (Kolyma Plateau, Bolshoi Anngachag Ridge), it can be assumed that the genus *Acarospora* is insufficiently studied in this region.

MATERIAL AND METHODS

Lichen samples were collected by E.V. Zheludeva, S.V. Chesnokov and L.A. Konoreva. Specimens are stored in the herbaria of the Komarov Botanical Institute RAS (LE) and the Institute of Biological Problems of the North FEB RAS (MAG). Species identification was carried out using standard macro- and microscopic methods (Stepanchikova & Gagarina 2014) in the Laboratory of Lichenology and Bryology of the Komarov Botanical Institute RAS. Specimens were photographed by V.V. Pankova using a Stemi 508 stereoscopic microscope (Carl Zeiss AG, Germany) and an Axio Scope A.1 microscope with an Axiocam 506 camera (Carl Zeiss AG, Germany). Measurements of microscopic characteristics were performed in KOH-free water. The presence or absence of lichen metabolites was determined using potassium hydroxide solution (K), 10 % sodium hypochlorite solution (C) and undiluted Lugol's solution (I). Species descriptions are based on the study of the authors' collections and in some cases supplemented with literature data.

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Each specimen was homogenixed manually, after that DNA extraction was carried out using a commercial Fitosorb kit (Syntol, Russia) in accordance with the production instructions. Amplification of the internal transcribed spacer (ITS) was performed using the following primers: ITS1F (Gardes & Brun 1993) and ITS4 (White et al. 1990). Polymerase chain reaction (PCR) was carried out in accordance with the following program parameters: an initial denaturation at 94°C for 5 min, followed by 5 cycles at 94°C for 30 s, 55°C for 30 s, and 72°C for 60 s, then 30 cycles at 94°C for 30 s, 52°C for 30 s, and 72°C for 60 s, with a final extension 72°C for 5 min (Westberg et al. 2015). The resulting amplicons were purified using the Cleanup Mini kit (Evrogen, Russia). Sequencing was carried out at the BIN RAS. The resulting sequences were checked for consistency and possible contamination using the BLASTn search (Altschul et al. 1990; http://www.ncbi.nlm.nih.gov/BLAST/) and then uploaded into NCBI GenBank database (https://www.ncbi.nlm.nih.gov/).

RESULTS

During the examination of collections from the Magadan Region, *Acarospora versicolor* Bagl. & Car., which is new to the Russian Far East, as well as three species new to the Magadan Region: *A. fuscata* (Schrad.) Arnold, *A. glaucocarpa* (Ach.) Arnold and *A. sinopica* (Wahlenb.) Körb., were identified. Below are detailed descriptions of these species, differences from related species, and additional information on the previously known species of *A. fuscata* (Zheludeva 2023).

Figure 1 Sample collection points. *Acarospora glaucocarpa* (Ach.) Arnold – 2, 3; *A. sinopica* (Wahlenb.) Körb. – 8; *A. versicolor* Bagl. & Car.– 6; *A. fuscata* (Schrad.) Arnold – 1, 4, 5, 7

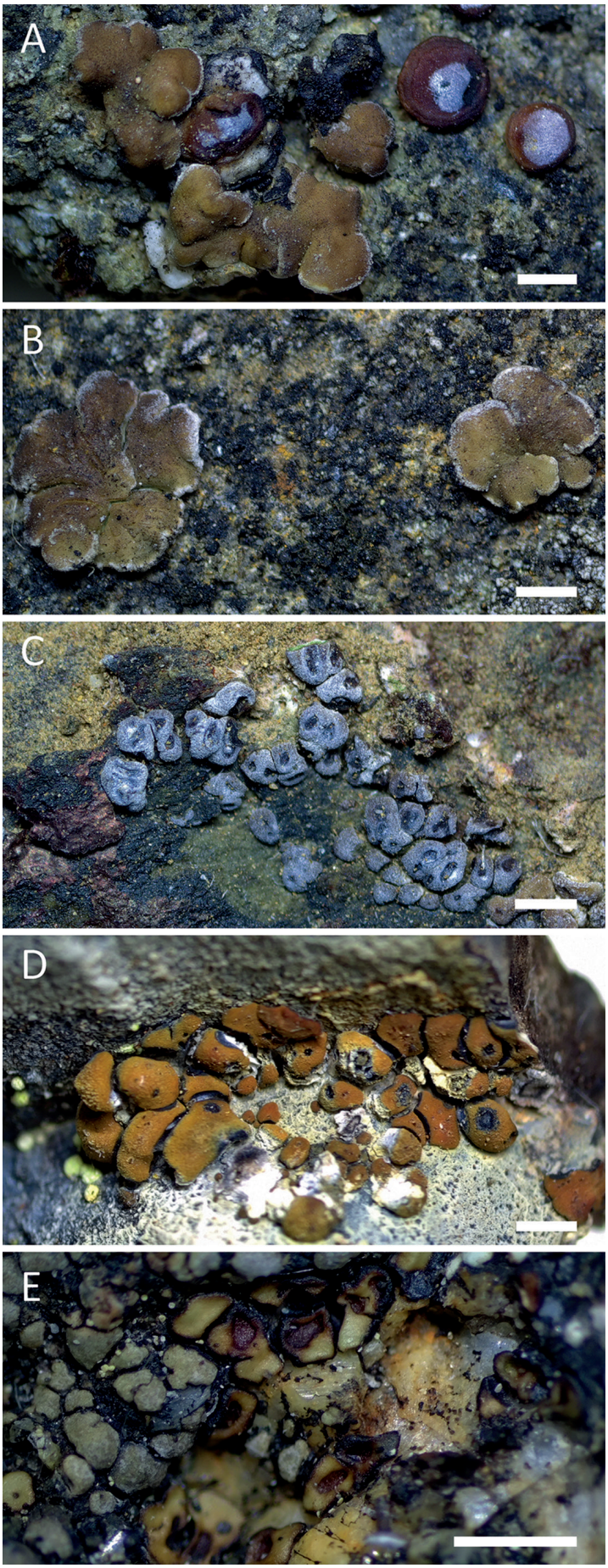

Figure 2 A, B – *Acarospora glaucocarpa*, C – *A. versicolor*, D – *A. sinopica*, F – *A. fuscata*. Scale bar = 1 mm

Table 1. Comparison *Acarospora glaucocarpa* with *A. cervina*

Species	Upper cortex	Apothecia	Spores, µm
A. glaucocarpa	prosoplectenchymatous	expanding with excluding the thalline margine	3.5–6 (7) x 1–2
A. cervina	paraplectenchymatous	expanding with reducing the squamules to a thalline margin	4–8 x 1.5–3

Table 2. Comparison *Acarospora sinopica* with *Myriospora dilatata*

Species	Algal layer	Paraphyses	Hymenium, µm
A. sinopica	uninterrupted	tips not expanding	105–115
M. dilatata	interrupted by hyphal bundles	tips expanding up to 4 µm	130–175

***Acarospora glaucocarpa* (Ach.) Arnold** (Figs 1, 2)

Thallus squamulose, areolate, but sometimes reduced to a margin-like rim around individual apothecia, greenish brown to brown. Areoles up to 3 (4.5) mm diam., dispersed, contiguous or imbricate or often forming extensive patches, often lobed, sometimes have whitish pruinose edges, gomphate or widely attached, lower side white. Epicortex usually 15 µm thick. Upper cortex 40–100 µm thick, prosoplectenchymatous, exterior yellowish-brown. Algal layer 70–150 µm high, irregular, interrupted by hyphal bundles. Apothecia 0.4–3 mm diam., initially immersed, then at the thallus level and sessile, usually one per areole, sometimes reducing it to apothecia. Disk reddish-brown, round or slightly angular, smooth or slightly rough, concave, flat or slightly convex, sometimes covered with bluish pruina. Exciple expanding from 70 to 200 µm thick. Hymenium 75–100 µm high, epihymenium 10 µm thick, conglutinate, yellowish brown. Paraphyses (1.7–) 2–2.5 (–3) µm at mid-level, straight, the apices slightly expanded up to 3–4 µm, brown capitate. Subhymenium 25–45 µm tall. Hypothecium 25–45 µm wide. Asci 50–60 × 13–17 µm, clavate, ascospores about 100. Ascospores 3.5–6 (7) × 1–2 µm, narrowly ellipsoid to cylindrical. Pycnidia not observed. No lichen substances.

Spot tests: hymenium gel dark blue in IKI.

Ecology. On calcareous rocks, schists, sometimes on non-calcareous rocks.

Distribution. *Acarospora glaucocarpa* is a widespread species. Found in Eurasia, North America and Australia. The nearest known locality is in Yakutia and Chukotka (Kristinsson et al. 2010). New for Magadan region.

Note. *Acarospora glaucocarpa* often confused with *A. cervina* A. Massal. Both of them can be covered with pruina and they have similar morphological features. However, these species are distinguished by presence of prosoplectenchymatous upper cortex in *A. glaucocarpa* (Table 1).

Specimens examined: Russia, Magadan Region, Tenkinsky District, Magadan–Nexican highway, Upper Kolyma Highlands, vicinity of the Contact hospital, up the stream Contact, 61°50'26.6"N 147°41'22.1"E, 886 m a.s.l., mossy rock outcrops, 11.07.2021, Zheludeva E.V. (MAG № T-5841); same place, Chesnokov S.V. (LE L-26084); same place, dwarf birch-lichen larch, 11.07.2021, Konoreva L.A. (LE L-26085); same place, 61°50'26.6"N 147°41'22.1"E, 886 m a.s.l., rocky placers on the slope of a hill near the stream, on a stone, 11.07.2021, Zheludeva E.V. (MAG № T-5844); Tenkinsky district, Gavryushka pass, 62°26'52.6"N 147°18'09.9"E, 1076 m a.s.l., stone scattering among dwarf cedar, 10.07.2021, Chesnokov S.V. (LE L-26086); same place (LE L-26087);

Additional specimens examined: Leningrad Region, Priozersk District, S of the bog Torfyanoe (Ervosuo), 60°50'53.5"N 30°22'14.2"E, disturbed area in a pine forest: heaps of slate, concrete, remains of buildings, a landfill, a system of small quarries, 19.09.2020, Himelbrant D.E., Stepanchikova I.S. (LE L-19494); Trans-Baikal Territory, Kalarsky District, Kodar Ridge, Oleniy rog brookvalley, 56°47'54.2"N 117°22'6.2"E, 1705 m a.s.l., *Cadonia–Flavocetraria–Alectoria* tundra on slope near stone rubble on the left bank, on limestone, 18.06.2015, Chesnokov S.V. (LE L-20276).

***Acarospora sinopica* (Wahlenb.) Körb.** (Figs 1, 2)

Thallus squamulose or areolate, rusty shades. Areoles 0.4–1.7 mm, dispersed or contiguous, along the cracks or forming patches 1–2 cm diam., the marginal ones often elongated and weakly radiating, giving the thallus a subplacodioid appearance, sometimes with yellowish pruina, widely attached, lower side pale to dark. Epicortex lacking. Upper cortex 25–30 µm thick, paraplectenchymatous, exterior dark reddish-brown. Algal layer 60–110 µm high, continuous, disappearing near the apothecia. Apothecia punctiform or dilate to 0.2–0.7 mm diam., immersed, usually numerous (up to 8), sometimes 1–2 per areole. Disc blackish brown, black, round or slightly angular, smooth or rough, concave. Exciple 30–40 µm thick, yellowish with dark brown exterior. Hymenium 105–184 µm high, epihymenium 20–30 µm thick, conglutinate, dark reddish-brown. Paraphyses (1.3–) 1.5–1.7 (–2) µm thick at mid-level, thin, convoluted, slightly branching and anastomosing, without apices expanding and colored caps. Subhymenium 60–130 µm tall. Hypothecium 10–15 µm wide. Asci 80–90 × 12–16 µm, clavate, ascospores 100–200. Ascospores 3–4 × 1.5–2 µm, broadly ellipsoidal. Pycnidia 150 × 120 µm, conidia 3 × 1.5–2 µm. No lichen substances.

Spot tests: hymenium gel blue turning green-red in IKI.

Ecology. On iron-rich rocks.

Distribution. Found in Eurasia, North America and Australia. The nearest known locality is in Yakutia (University of Oslo 2024), Wrangel Island (Evdokimov et al. 2022) and Kamchatka Territory (Himelbrant et al. 2021). New for Magadan Region.

Note. *Acarospora sinopica* resembles *Myriospora dilatata* (M. Westb. & Wedin.) K. Knudsen & L. Arcadia. Both of them have a rusty thallus, but *A. sinopica* have a continuous algal layer, non-expanding paraphyses and lower hymenium (Westberg et al. 2011) (Table 2).

Specimens examined: Russia, Magadan Region, Srednekansky District, Kolyma River valley, surroundings of Semchansky section of the Magadansky nature reserve, right bank of the river, Fedorovich stream, 63°44'51.24"N 153°25'28.08"E, 210 m a.s.l., rocky outcrops on the slope of the hill with rare bushes of cedar shale, moss-lichen, 12.07.2020, Zheludeva E.V. (MAG № Ср.-5842).

Additional specimens examined: Tver Region, vicinity of Borodinskaya biological station, Elenkin A.A. (LE L-11079); Murmansk Region, Tersk District, Kandalaksha Reserve, Por'ya Bay, Gorely island, N part, 66°45'52.2"N 33°45'55"E, stones on the seashore on the forest margin, on (weakly) calcareous stone, 23.06.2010, Zhdanov I.S. (LE L-10917).

***Acarospora versicolor* Bagl. & Car.** (Figs 1, 2)

Thallus squamulose or areolate, light brown to dark brown. Areoles 0.6–0.8 mm, dispersed or contiguous, along the cracks or forming patches, sometimes overlapping, often have black margin, with or without heavily white pruina, widely attached, below side pale brown. Epicortex up to 20 µm thick, uneven. Upper cortex 20–30 µm thick, para-

Table 3. Comparison *Acarospora versicolor* with related species

Species	Spores, µm	Hymenium, µm	Pruina	Squamules	Disk
A. versicolor	3–5 x 2.0	70–100 (130)	usually present	convex	blackish-brown
A. cinerascens	6.0–9.0 x 2.5–3.5 (–4.0)	> 130	usually present	convex	black
A. veronensis	3–5 (6) x 1–2 (3)	80–100 (120)	usually absent	more or less flat	reddish-brown

plectenchymatous, exterior dark brown. Algal layer 60–100 µm high, continuous, regular, unterrupted by hyphal bundles. Sometimes have a lower cortex up to 20 µm. Apothecia 0.1–0.4 (0.8) mm diam., immersed or at the thallus level, usually 1, rare 2–3 per areole, sometimes more. Disc blackish brown, black, round or irregular shape, smooth, concave, usually not pruinose. Exciple widened to 25–35 µm thick. Hymenium 70–100 µm high, epihymenium 10–15 µm thick, conglutinate, orange-brown. Paraphyses 2–2.5 µm (according to Knudsen et al. (2015), 1.5–2 µm), thick at mid-level, the apices expanded up to 3–4 (5) with brown caps. Asci 65–90 × 12–17 µm, clavate, ascospores 100–200. Ascospores 3–6 × 1–3 µm, ellipsoidal. Pycnidia not observed (according to Magnusson (1929) – pycnidia immersed, 200 × 100 µm, conidia 2 × 1.5 µm). No lichen substances.

Spot tests: hymenium gel blue turning green-red in IKI.

Ecology. On siliceous and calcareous rocks.

Distribution. The species is commonly found in Eurasia (Westberg et al. 2015). In Russia, it is known from European Russia (Zhdanov & Volosnova, 2012). The nearest known locality is in Mongolia (Golubkova 1981) and South Korea (Park et al. 2023). New for Russian Far East.

Note. *Acarospora versicolor* resembles *A. cinerascens* J. Steiner. Species *A. cinerascens* was not independent and it was considered as a synonym of *A. versicolor* for a long time. However, recent research has shown the existence of two clearly distinguishable species. It is shown that they can be distinguished even at an early stage of development, since *A. cinerascens*, unlike *A. versicolor*, has larger spores 6.0–9.0 × 2.5–3.5 (–4.0) versus 3–5 × 2.0 and higher hymenium (above 130 µm) (Knudsen et al. 2015). Nevertheless, *A. versicolor* is also similar to *A. veronensis*. At an early stage of development it is extremely difficult to distinguish them due to similar anatomical and morphological characteristics, and it can only be done using molecular genetic methods. Developed thallus of *A. versicolor* is usually distinguished by the fact that it often has a whitish pruina (*A. veronensis* usually does not), more convex squamules (*A. veronensis* has more or less flat one) and the disc is often blackish-brown or black (*A. veronensis* has a reddish-brown, brown disc) (Table 3).

Specimens examinated: Russia, Magadan Region, Srednekansky District, Kolyma River valley, tributary of the river Arangas, 63°20'46.19"N 152°40'57.78"E, 197 m a.s.l., rock outcrops on the river bank, 10.08.2021, Zheludeva E.V. (MAG № Sr.-5845).

Additional specimens examinated: Vladimir Region, Gus'-Khrustal'ny District, "Meshchera" National Park, Narma village, 55°38'46.5"N 40°08'31.3"E, old church, on brick stones of old church foundation, 08.07.2011, Zhdanov I.S. (LE L-10920); Sverdlovsk Region, Kamensk District, Noviy Zavod village "Beliy Kameshok", 56°27'11.7"N 61°53'53.1"E, rock, 28.06.1999, Paukov A.G. (LE L-7736).

Acarospora fuscata (Schrad.) Arnold (Figs 1, 2)

Thallus areolate or squamulose-areolate, light to dark brown, shiny. Areoles 0.3–1.0 mm, dispersed or contiguous, often along the cracks or covering large areas, irregular in shape, without pruina, lower side black. Epicortex thin, about 5 µm thick. Upper cortex 20–25 µm thick, paraplectenchymatous, greenish brown. Algal layer 60–80 µm high, regular, uninterrupted by hyphal bundles. Apothecia 0.1–0.2 mm diam., immersed, without margins, usually one per areole. Disk reddish-brown, most often angular or irregular in shape, smooth and shiny, concave. Exciple 10–20 µm thick, inconspicuous. Hymenium 80–115 µm high, epihymenium 10 µm thick, coherent, yellowish brown. Paraphyses 2–2.5 (–3) µm at mid-level, septate, often with oil drops, the apices expanded up to 3–5 µm, without brown caps. Subhymenium 25–30 µm tall. Hypothecium 25–60 µm wide, yellowish (according to Knudsen et al. (2019) – 10–20 µm). Asci 40–60 × 10–16 µm (according to Golubkova (1988) – 60–85 × 15–17), clavate, ascospores 100–200. Ascospores 3–4.5 × 1.5 µm (according to Golubkova (1988) – 4–5.6 × 1.5–1.8), narrowly ellipsoid to ellipsoid. Pycnidia not observed (according to Knudsen et al. (2019) pycnidia 100–110 × 60–70 µm, conidia 1.0–2.0 × 0.5–1.0).

Spot tests: C+ red (gyrophoric acid), hymenium gel greenish red in IKI.

Ecology. On siliceous rocks.

Distribution. Widespread species. Most often, found in Eurasia and North America. The nearest known locality is in Koryakia (Himelbrant et al. 2019).

Note. *Acarospora fuscata* differs mainly from other lichens of this genus by the presence of gyrophoric acid in the cortex. Within the group of acarospores with the C+ red reaction, *A. fuscata* is very similar to *A. gallica* H. Magn. and does not have clear morphological and anatomical differences. Their main difference is in the amyloid reaction: the hymenium of *A. fuscata* is colored in reddish shades, and in *A. gallica* it is blue.

Specimens examinated: Russia, Magadan Region, Olsky District, Magadansky nature reserve, Yama River, Studenaya cordon, about 6 km downstream, right bank of the river, 59°44'N 153°40'E, 72 m a.s.l., rocky dry slope, a few meters from the riverbed, on mossy stones, 23.07.2010, Zheludeva E.V. (MAG № O-3741); Susumansky District, lake Urultun, right bank, 63°46'35.43"N 148°54'35.99"E, 824 m a.s.l., larch woodland with rocky scatterings and shrub-lichen patches, on stones, 20.08.2021, Zheludeva E.V. (MAG № Su.-5839); Srednekansky District, Kolyma River valley, Zamkovaya mountain, 63°21'15.88"N 152°37'54.80"E, 520 m a.s.l., rocky outcrops on the mountainside, 09.08.2021, Zheludeva E.V. (MAG № Sr.-5840); same place, Zheludeva E.V. (MAG № Sr.-5846); Srednekansky District, Kolyma River valley, surroundings Semchansky section of the Magadansky nature reserve, right bank of the river, 63°43'54.73"N 153°23'36.65"E, 182 m a.s.l., rocky placer on the slope of a hill with rare bushes of dwarf cedar lichen, 10.07.2010, Zheludeva E.V. (MAG № Sr.-5843).

Additional specimens examinated: Nenets AD, Nenets Nature Reserve, northeastern part of the Dolgiy Island, extensive pebble spit (old coastal shafts), on pebbles, 31.07.2005, Lavrinenko O.V. (LE L-23280); Leningrad Region, Vyborg District, proposed protected area "Kurenniemi", 60°10'50.1"N 29°01'40"E, boulder in pine forest on seashore, on granite boulder, 03.06.2021, Andreev M.P. (LE L-21058).

Doubtful taxa

Acarospora veronensis A. Massal.

Previously, it was published by Kotlov (1995) for the Upper Kolyma Highlands, but the sample was not found in herbaria LE and MAG. However the herbarium MAG contained samples *A. veronensis* collected by Zheludeva. They

was redefined as *Myriospora smaragdula* (Wahlenb. Ex Ach.) Nägeli ex Uloth. by first two authors.

Specimens examined: Russia, Magadan Region, Olsky District, Olsky estuary, Atargan peninsula (between the "rotten corner" and the beginning of the spit), 59°33'41.68"N 151°28'13.35"E, 88 m a.s.l., slope of a hill, outcrops of rocks with rocky screes, on a stone, 31.03.2016, Zheludeva E.V. (MAG № O-4524).

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