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TAXONOMIC NOVELTIES

Notes on *Usnea barbata* (L.) F.H. Wigg. in Russia, with a new synonym

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ABSTRACT

As a result of studying the lichenological herbarium collections of LE, HERZ, LECB it was revealed that *Usnea samjatnini* Tomin is a new synonym of *Usnea barbata* (L.) F.H. Wigg. A lectotype of *U. samjatnini* Tomin is designated from specimens deposited in LE. The article provides information on the morphological, anatomical and chemotaxonomic features of polymorphic species *U. barbata*.

Keywords: *Usnea barbata*, *Usnea samjatnini*, Russia, new synonym

РЕЗЮМЕ

Гагарина Л.В., Комарницкая Н.А., Прокопьев И.А. Заметки об *Usnea barbata* (L.) F.H. Wigg. в России, с указанием нового синонима. В результате изучения гербарных коллекций лишайников LE, HERZ, LECB установлено, что *Usnea samjatnini* Tomin является новым синонимом *Usnea barbata* (L.) F.H. Wigg. В статье приводится информация о морфологических, анатомических и хемотаксономических особенностях полиморфного вида *U. barbata*.

Ключевые слова: *Usnea barbata*, *Usnea samjatnini*, Россия, новый синоним

The genus *Usnea* Dill. ex Adans. includes macrolichens with recognized beard-like morphology, and may be either shrubby, subpendent or long-pendent. Easily recognized by their elongate, usually pale, yellowish green branches and especially by the presence of a cartilaginous central axis. There are several hundred species widely distributed throughout the world. Species of this genus are predominantly forest species, and are less commonly found on other substrates (Motyka 1936–1938, Golubkova 1996, Halonen 1998, Randlane et al. 2009).

MATERIAL AND METHODS

The study is based on an analysis of literature and examination of specimens in the lichenological herbarium of Komarov Botanical Institute RAS (LE L), herbarium of Herzen State Pedagogical University of Russia (HERZ) and herbarium of Saint-Petersburg State University (LECB). Photographs of specimen were taking using a Stemi 508 stereoscopic microscope (Carl Zeiss AG, Germany) with a Dr. Focal UNISON-12A camera and an Axio Imager A2 microscope (Carl Zeiss AG, Germany) with an E3ISPM12300KPA camera.

The presence of secondary metabolites was determined by color reactions with undiluted Lugol's solution (I), 10 % KOH solution (K) and with a sodium hypochlorite solution (C). TLC was performed according to the standard procedure (Culberson & Ammann 1979, Kranner et al. 2002), using solvent system C.

For the analysis, 5 mg of lichen was ground in a mortar. The resulting powder were extracted twice with 0.5 ml of acetone. The extraction was carried out with constant stirring for 24 hours at a temperature of 20–25°C. The obtained extracts were centrifuged at 15000 g for 5 min and then kept at 4°C until analysis. HPLC-UV analysis was performed with a Shimadzu LC-30 Nexera chromatograph (Japan). For chromatographic separation, a Phenomenex Kinetex C18 column (100 × 2.1 mm, 1.7μm) was used. The mobile phase consisted of (A) water LCMS grade with 0.1 % (v/v) formic acid and (B) acetonitrile LCMS grade with 0.1 % (v/v) formic acid. Analyses were performed at 40°C with a flow rate of 0.3 ml/min in the gradient elution mode, the percentage of B was programmed as follows: 30 % (2 min) – 80 % (15 min) – 100 % (16 min) – 100 % (20 min) – 30 % (20.1 min) – 30 % (25 min, controller stop). The volume of injected sample was 2 μL. Spectra of eluting substances were recorded in UV at 254 nm. To identify lichen substances, we compared of their retention time (Rt) with the authentic standards from the BIN RAS collection.

RESULTS AND DISCUSSION

***Usnea barbata* (L.) F.H. Wigg.**, Primitiae Florae Holsaticae: 91. 1780. ≡ *Lichen barbatus* L., Sp. Pl. 2: 1155. 1753. Type: Table 12, figure 6 Dillenius, Historia Muscorum (lectotype, Jørgensen et al., Bot. J. Linn. Soc. 115: 372. 1994); Sweden, Västmanland, Kila, SO om Granmuren, pa *Malus domestica*, 20.07.1962, A. Nordin 1449 (UPS epitype, Jørgensen et al., Bot. J. Linn. Soc. 115: 372. 1994. G isoepitype). Chemistry: usnic, salazinic acids. % ratio cortex/medulla/central axis = 4.5/33/26; = *Usnea samjatnini* Tomin, Trudy Tiflisskogo Botanicheskogo Instituta 1: 371. 1934. Type: [Georgia], Bakuriani vicinity, mountain Tskho-Tskhara [Tskhratskaro Pass], on branches of *Betula* sp., 01.08.1928, leg. B.N. Samjatnin [Za-

myatnin], det. M.P. Tomin as *U. samjatnini* (TLC/HPLC UZ: usnic, salazinic acids, LE L-17385! lectotype designated here, MBT 10026746; S F210049 isolectotype), **syn. nov.** (Fig. 1).

Thallus pendent, to 30 cm long (rarely to 60 cm), branching anisotomic-dichotomic, branches usually parallel, may be partly divided into irregular segments by annular cracks, green. Base thin, brown to black. Branches not narrowed at point of attachment, often with depressions and/or ridges. Papillae sparse to numerous. Fibrils absent or few to numerous, 2–5 mm long (type figure has numerous fibrils). Soralia punctiform and irregular, developing on top of tubercles, smaller than half the diameter of branches. Isidia few to numerous, often developing on soralia. Apothecia absent to few, usually terminal, subterminal, rarely lateral, 4–8 mm in diam., disc greenish, with marginal fibrils. Ascospores 8–12 × 5–8 μm. Pseudocyphellae absent. Cortex 4–13 %, white. Medulla 24–33 %, loose to dense, white. Axis 24–38 %, white.

Two chemotypes have been recorded: salazinic acid as a main substance and protocetraric acid as an accessory substance in medulla (medulla K+ yellow turning red, P+ orangish yellow, C–, KC–); without medulla substances (K–, P–, C–, KC–). Earlier, we pointed to the chemotype with barbic/diffractaic acid (Gagarina et al. 2017), but further study of this genus showed an erroneous definition of this specimen.

Usnea barbata is a very polymorphic species. The species is closest to *U. dasopoga* (Ach.) Nyl. and there are no clear differences between these species. *Usnea barbata–dasopoga* group is often mentioned in literature (Clerc & Otte 2018). Some authors (e.g. Randlane et al. 2009) suppose that species differ consistency of medulla (loose medulla in *U. barbata*, and dense in *U. dasopoga*). Unfortunately, according to our data, it is impossible to distinguish between species by this feature, since *U. barbata* has loose to dense medulla and *U. dasopoga* has loose (more often) to dense (less often) medulla. Mark et al. (2016) divided *Usnea* species into two groups based on the thickness of the bark and cortex and medulla. The authors placed *U. barbata* in group with thin cortex and thick and more lax medulla. However, as a result of the molecular genetic analysis, specimens of *U. barbata* were divided between these two clades. Other authors (e.g. Motyka 1936–1938) believe that the important feature is the basis. However, both species have a dark base with or without transverse cracks (thalli *U. barbata* without transverse cracks are more common). According to our observations, the presence of transverse cracks for species of the genus *Usnea* with a pendulous thallus is not always a taxonomic feature. *U. dasopoga* always has well-developed fibrils (e.g. Golubkova 1996), while *U. barbata* can have well- or poorly developed fibrils. If *U. barbata* has few or poorly developed fibrils, then it can be distinguished from *U. dasopoga* by this feature. *U. samjatnini* has poorly developed fibrils and can therefore be considered as a synonym of *U. barbata*.

Next, we present the original description of *U. samjatnini* (Tomin 1934). The original description was given in Russian, we present it in English here. "*Thallus yellowish-green, pendent, to 50–60 cm long. Branching almost isotomic-dichotomous, without fibrils, divided by annular cracks, they are densely acutely warty on the surface. Medulla loose, K+ yellow turning red. Central axis medium thickness. Apothecia large, up to 4–8 mm wide, disc pale reddish-green and numerous marginal fibrils. Asci 8-spored, cylindrical-clavate. Ascospores arranged in one or two lines, short ellipsoid, 8–12 × 5–8 μm*". The description does not contain data on the characteristics of the base of the thallus and we did not find it in the sample. The main diagnostic features of *U. samjatnini*, because of which we decided to reduce this species to synonyms, are pendulous thallus, branches divided by annular cracks, absence of fibrils. According to the results of TLC and HPLC-UV, usnic and salazinic acids were identified, which corresponds to one of their *U. barbata* chemotypes.

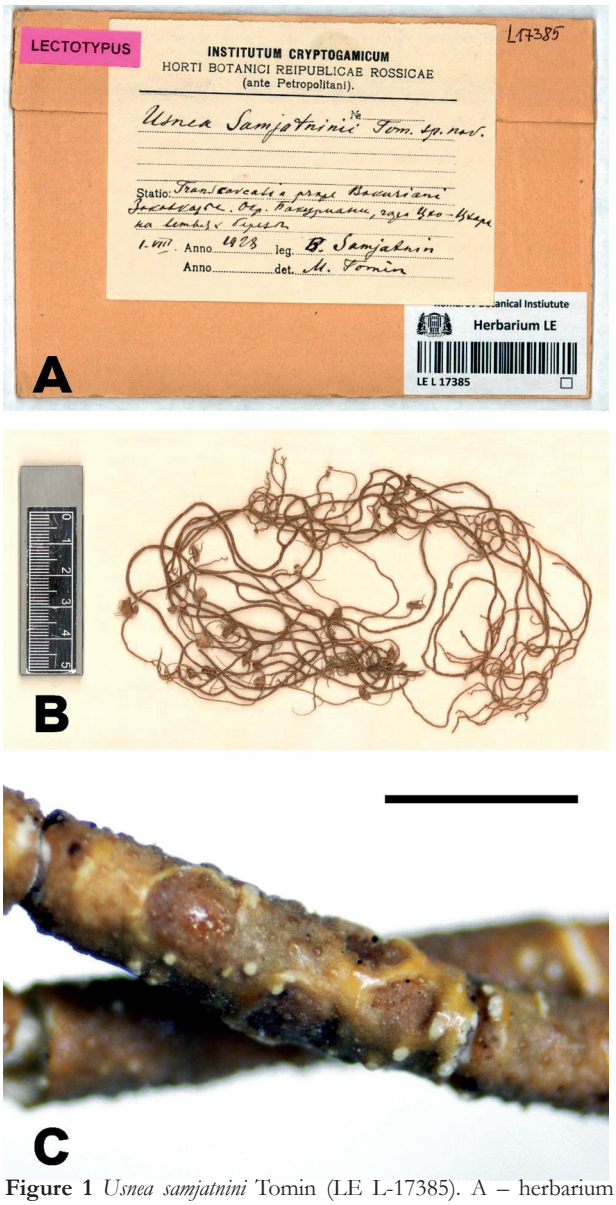

Figure 1 *Usnea samjatnini* Tomin (LE L-17385). A – herbarium label; B – thallus; C – the branch of thallus with annular cracks, papilles and depressions (scale: 1 mm)

Distribution in Russia: According to the literature data, in Russia the species occurs on the territory of Northen part of European Russia, Northen part of Ural Mountains, Russian part of Caucasus, Western Siberia (without mountain territories), Southern Siberia, Southern part of Far East (Urbanavichus & Urbanavichene 2004, Otte 2007, Urbanavichus 2010, Gagarina et al. 2017, Ismailov 2017, Urbanavichene & Urbanavichus 2019, Urbanavichene & Urbanavichus 2023). The species is included in the Red Books of some regions: Bryansk Region (2016), Kirov Region (2014). As a result of the revision of herbarium, almost all samples of *U. barbata* were reidentified. The list of studied samples with chromatography results is given below.

World distribution: holarctic.

Substrate and ecology: corticolous.

Specimen examined:

***Usnea barbata* (L.) F.H. Wigg. –** Country, year, collector unknown, very probably 19th century (TLC UB8: usnic, protocetraric acids) (LE L-23209). Abkhazia. Ritsa Relict National Park, vicinity of the lake Ritsa, fir-spruce-broadleaf

forest, on fallen branches, 11.06.1988, leg. N.S. Golubkova (TLC/HPLC UB2, 3: usnic, salazinic acids) (LE L-29381), on the branches of *Abies*, 11.06.1988, leg. N.S. Golubkova (TLC UB4: usnic, salazinic acids) (LE L-29383), (TLC UB6: usnic, salazinic acids) (LE L-29384), (TLC/HPLC UB5: usnic, salazinic acids) (LE L-29382). Russia. Caucasus [Karachayevo-Circassian Republic], Teberda National Park, on trunk of tree, 05.09.1985, leg. I.N. Drozdova (TLC: usnic, salazinic acids) (HERZ0008298). *U. samjatnini* Tomin. Abkhazia. Ritsa Relict National Park, vicinity of the lake Ritsa, fir-spruce-broadleaf forest, on fallen branches, 11.06.1988, leg. N.S. Golubkova (TLC/HPLC UB1: usnic, salazinic acids) (LE L-29385).

***Usnea ceratina* Ach.** – Germany. leg. J. Flotow (TLC UB17, 18: atranorin, usnic, barbatic, 4-O-demethylbarbatic acids) (LE L-29380).

***Usnea dasopoga* (Ach.) Nyl.** – Russia. [Amur Region] Far Eastern Soviet Socialist Republic, vicinity of Ust'-Norsk, on bark of *Pinus* sp. in forest, 24.07.1956, student brigade no. 7 (TLC: usnic, salazinic acids) (HERZ0007918). Kostroma Region, Manturovo District, Kologriv Forest State Nature Reserve, Manturovskii cluster, 7 sq., 58°03'27.6"N 44°37'15.6"E, valley of the Ivanchikha River, fir forest with linden, on the bark of *Tilia cordata*, 18.05.2022, leg. G.P. Urbanavichus, I.N. Urbanavichene (TLC UB7: usnic, salazinic acids) (LE L-21232). Leningrad Region, Luga District, settlement of Rapti [settlement of Dzerzhinskii], on shore of the Cheremenets Lake, on *Picea abies* and *Pinus sylvestris*, 16.06.1959, leg. Petrova, Vasil'eva (TLC: usnic, salazinic acids) (HERZ0008164); pine forest near Cheremenets Lake, 26.06.1959, leg. Semyashkin (TLC: usnic, salazinic acids) (HERZ0008165); Herzen University Geographic Station "Zhelezo", 17.08.1967, leg. Pavlova, Neusitryva (TLC: usnic, salazinic acids) (HERZ0007920), blueberry spruce forest, 19.08.1967, leg. Murashko (TLC: usnic acid) (HERZ0007921); 1 km from Herzen University Geographic Station "Zhelezo", downstream the Luga River, floodplain forest, on *Betula* sp., 19.06.1969, leg. Sokolova (TLC: usnic, salazinic acids) (HERZ0007922), 0.5 km north-west of the Herzen University Geographic Station "Zhelezo", pine forest, 17.05.2007, leg. A.S. Tartygin, M.N. Mordovtseva (TLC: usnic, salazinic acids) (HERZ0008699), 50 m south of the Herzen University Geographic Station "Zhelezo", on branches of *Pinus sylvestris*, 17.05.2007, leg. I.O. Bortnikov (TLC: usnic, salazinic acids) (HERZ0008701); 50 m south of the Herzen University Geographic Station "Zhelezo", on branches of *Pinus sylvestris*, 17.05.2007, leg. A.G. Perkova, Yu.A. Egorova (TLC: usnic, salazinic acids) (HERZ0008700); [Gatchina District] settlement of Vyritsa, 14.06.1956, leg. Pavlova, Ovcharova (TLC: usnic, salazinic acids) (HERZ0007917); [St. Petersburg], Popovo Forest [Natural monument "Valley of the Popovka River"], on trees, 1922, collector unknown (very probably V.L. Komarov) (TLC: usnic, salazinic acids) (HERZ0007916).

***Usnea florida* (L.) F.H. Wigg.** – Sweden. Uppsala, Uppland, 1852, leg. G. Svanberg (TLC UB10: usnic acid) (LE L-17250). Canada. on trunks near the sea, 1910, leg. J. Macoun (TLC UB16: usnic acid) (LE L-17739). [Georgia]. Caucasus, Borjomi, 1899, leg. A.A. Elenkin (TLC: usnic, squamatic acids) (HERZ0008132).

***Usnea hirta* (L.) F.H. Wigg.** – Germany. J. Flotow (TLC UB19, 21, 22, 23, 24, 25: usnic acid, murolic acid complex) (LE L-17341). Russia. Leningrad Region, Vsevolozhsk District, vicinity of the village of Kirpol'e, 1898, leg. O. Alekseenko (TLC UB28, 29, 30: usnic acid, murolic acid complex) (LE L-17209). [Leningrad Region, Gatchina District], Pavlovsk District, settlement of Mikhailovka, campus "Novaya Zhizn'", student brigade 1-9 (TLC: usnic, diffractic acids) (HERZ0007919).

***Usnea perplexans* Stirt.** Sweden. Bohuslän, Hällesdalen, par Ödsmål, in summon montis, ad rupem, 18.06.1918, leg. H. Magnusson (TLC UB9: usnic, salazinic acids) (LE L-17216).

***Usnea rubicunda* Stirt.** – Schultz, Fl. Gall. Germ. Exs. 782 (TLC UB12: usnic, salazinic acids) (LE L-17334). France. Ex herbario Delise, 1836 (TLC UB14: norstictic, salazinic acids) (LE L-17332).

***Usnea subfloridana* Stirt.** – Austria. Province of Styria, Graz, Eggenberg District, collector and date unknown (TLC UB13: usnic acid, stictic acid complex) (LE L-17743). Czeck Republic. Böhmen, an Nadelhölzern b. K. Podol, on conifers, 1909, leg. V. Kut'ák (TLC UB11: usnic, thamnolic acids) (LE L-17342). Russia. Leningrad Region, Luga District, Herzen University Geographic Station "Zhelezo", on branches and trunks of trees, 28.06.1969, leg. A. Burchuk (TLC: usnic, thamnolic acids) (HERZ0008301).

Questionable sample. *Usnea barbata* (L.) F.H. Wigg. – Russia. Leningrad Region, Lodejnopolsky District, 5 km SW of Lakhta Bay, along the forest road towards the Zubets River, mixed aspen-pine-birch forest, on *Populus tremula*, 05.1999, leg. D.E. Himelbrant 98-9-99 (TLC UB27: usnic, salazinic acids) (LECB). This sample contains a mark of Pekka Halonen (specialist on *Usnea* from Finland) 2002 "*Usnea* cf. *barbata* s. lat.". The sample is small, it is obvious that it is a young thallus; the growth pattern indicates that the thallus will be pendulous. The sample belongs to the group *barbata/dasopoga*. The sample is not fresh, and molecular genetic studies are not possible.

We think that the distribution of this species in Russia has not been studied enough. Most of the published data on the location of this species need to be confirmed. In herbarium collections, more than 90 % are incorrectly identified (samples with pendulous thallus are usually *U. dasopoga*, samples with shrubby or subpendent thallus are usually *U. hirta* or *U. subfloridana*).

Apparently, Motyka (1936–1938) was right when he wrote that the species is reliably known from several regions of Scandinavia and Central Europe. In Russia, the species is most likely distributed in the Caucasus.

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