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FONTINALIS DICHELYMOIDES (FONTINALACEAE, BRYOPHYTA), A NEW SPECIES FOR THE MOSS FLORA OF RUSSIA

FONTINALIS DICHELYMOIDES (FONTINALACEAE, BRYOPHYTA) – НОВЫЙ ВИД ДЛЯ БРИОФЛОРЫ РОССИИ

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Abstract

Fontinalis dichelymoides is newly recorded for the moss flora of Russia; it was found in the course of revising herbarium collections of *Fontinalis squamosa.* The description and illustration of *F. dichelymoides* based on the specimen from Russia are given; key to identification of a group of close species, including *F. dichelymoides, F. squamosa* and *F. dalecarlica,* is provided. The results of molecular studies of *Fontinalis* specimens from Russia are commented.

Резюме

В результате ревизии гербарных образцов *Fontinalis squamosa* был обнаружен новый вид для флоры мхов России *Fontinalis dichelymoides.* Приводится описание вида, рисунок и ключ для различения близких видов, *F. dichelymoides, F. squamosa* и *F. dalecarlica*. Комментируются результаты молекулярных исследований.

KEYWORDS: *Fontinalis dichelymoides*, distribution, Russia

INTRODUCTION

The taxonomy of the genus *Fontinalis* remains poorly known, as these aquatic mosses are extremely variable. In Russia in most publications of 20th century, as well as in the last checklist of mosses, only four species were reported: *F. antipyretica* Hedw., *F. hypnoides* Hartm., *F. squamosa* Hedw. and *F. dalecarlica* Bruch, Schimp. & W. Gümbel (Abramova *et al.*, 1961; Ignatov *et al*., 2006), which is similar to most European countries. At the same time, in older literature, e.g. Limpricht (1896) and Brotherus (1923), more than 10 species of the genus were accepted; however subsequent revision of Welch (1960) reduced most of them to synonymy. Among others, *F. dichelymoides* Lindb. was synonymized with *F. dalecarlica*, and for a certain time remained in oblivion.

Fontinalis dichelymoides was described by Lindberg (1869) from Piojärvi Lake in Finland by collection of Brotherus. It seems to be a rare species, as much later, in "Die Laubmoose Fennoskandias", Brotherus (1923) added to the type locality only one collection from Sweden and three from Finland. Nyholm (1960) thought that *F. dichelymoides* is merely a lake form of *F. dalecarlica,* likely influenced by Welch opinion.

The first authors who returned *F. dichelymoides* as a species were Koponen *et al*. (1977), but the European checklist (Corley *et al*., 1981) and the catalogue of species distribution (Duell, 1984) ignored this, continuing keeping this species as a synonym of *F. dalecarlica*. With time, however, the status of *F. dichelymoides* was accepted by Scandinavian bryologists (Koponen *et al.*, 1995; Söderström, 1996; Ulvinen & Syrjänen, 2009, Hedenäs *et al*., 2014) and in new European checklist (Hill *et al*., 2006). The rarity of the species was confirmed, as Ulvinen &. Syrjänen (2009) reported it from only eight provinces of Finland, from the south to Lapland, and there are only eight dots on the map of its distribution in Sweden (Hedenäs *et al.*, 2014).

To complete the history of the previous studies of *F. dichelymoides*, the species was reported by Welch (1934) in North America; however, in the recent treatment of Allen (2014) it is placed in synonymy of *F. sullivantii* Lindb.

In the course of revision of *Fontinalis* collections in herbarium of Petrozavodsk (PTZ), *F. dichelymoides* was identified from Karelia; it was found among specimens kept under *F. squamosa* Hedw. Along with this first record

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of the species in Russia, we were able to check its status by molecular markers.

MATERIAL AND METHODS

Specimens and DNA markers: as the expanded set of *Fontinalis* is available in GenBank from the study of Shaw & Allen (2000), we used their data, supplemented with several specimens of superficially similar species from Karelia. Nuclear ITS and chloroplastic *trn*L-F markers were used, as they were used by Shaw & Allen (2000), and proved to be useful for pleurocarp phylogeny studies (Gardiner *et al*., 2005). Total genomic DNA was extracted from dry plants using the Nucleospin Plant Extraction Kit (Macherey-Nagel, Germany). The laboratory protocols were essentially the same as in the latter publication.

Phylogenetic analysis. Sequences were aligned by Clustal and modified manually using BioEdit 7.0 (Hall, 1999). Bayesian analysis of the ITS dataset was conducted in MrBayes (Huelsenbeck & Ronquist, 2001) using the GTR+G model. It was run for 10 000 000 generations with sampling every 1000 generations. The first 25% of sampled trees were discarded for the burn-in. Supplementary maximum parsimony analysis was performed in Nona (Goloboff, 1994) in Winclada shell (Nixon, 1999).

RESULTS

ITS sequences were found to be variable enough, and both Bayesian and MP analyses resulted in a resolved phylogenetic tree with some insufficient differences, so only Bayesian tree is shown with MP bootstrap values inserted.

The tree is rooted on *Brachelyma*, and, predictably, *Dichelyma* is the next in a grade to *Fontinalis*. Then *Fontinalis antipyretica* clade (most specimens) [and the only specimen of *F. squamosa* included in our dataset], branches off. The next branch combines *F. hypnoides, F. gracilis* (accepted here as a species), and one specimen of *F. antipyretica.* The terminal clade, called here "*Fontinalis dalecarlica*–clade" (Fig. 1), includes *F. dalecatlica*, many American endemic species, and also *F. dichelymoides*. *Fontinalis reafearnii* keeps the basalmost position, but the clade of *F. reafearnii* + all others members of *"F. dalecarlica*–clade" has support only in Bayesian analysis. The *"F. dalecarlica*–clade" without *F. readfearnii* however received sufficient support in both analyses.

Fontinalis dichelymoides has a sister position to all other species of "*F. dalecarlica*–clade": *F. missourica, F. flaccida, F. sullivantii, F. welchiana, F. novae-angliae* and *F. dalecarlica*. They form polytomy with smaller clades without support, thus their grouping is not worthy a discussion.

Sequences of *trn*L-F were found to be much less variable than ITS, although their phylogenetic analysis divides the genus into two clades: "*F. dalecarlica*–clade" and *F. antipyretica* + *F. hypnoides*–clade (or grade), without support.

Fontinalis dichelymoides obviously is a member of "*F. dalecarlica*–clade" and has no one substitution in *trn*L–F compared with *F. novae-angliae* and *F. dalecarlica*, which, however. markedly differ from *F. dichelymoides* by ITS. The difference in ITS between *F. dalecarlica* (Karelian) and *F. dichelymoides* is much greater (six substitutions and one indel) than between *F. dalecarlica* (Karelian) and *F. novae-angliae* (one substitution) and between *F. dalecarlica* (Karelian) and *F. sullivantii* (two substitutions).

Table 1. Number of substitutions + indels between *F. dichelymoides* and some other *Fontinalis* species

	*trn*L–F	ITS
F. novae-angliae AF191540 / AF192131	0	6+1
F. dalecarlica - / Karelia	-	6+1
F. dalecartica AF191534 / AF192125	0	14.5+1
F. sullivantii AF191521 / AF192112	1	7+1
F. sullivantii AF191511 / AF192102	1	8+1
F. welchiana AF191541 / AF192133	1	7+7
F. missourica AF191515 / AF192106	2	9+2
F. flaccida AF191513 / AF192104	5	15+1
F. redfearnii AF191507 / AF192098	5	22+1
F. squamosa AF191520 / AF192111	10	22
F. hypnoides AF191508 / AF192130	10	18+1
F. antiperetica AF191516 / AF192117	15	23+4

TAXONOMY

The above mentioned differences do not provide support by statistical methods of phylogenetics, but the fact that three specimens of *F. dalecarlica* from Karelia are not variable in ITS indicates that *F. dichelymoides* is better differentiated genetically than some other members of "*F. dalecarlica*–clade" and, along with morphological differences, merits a status of species of its own. Its description based on Russian specimens is provided below.

Fontinalis dichelymoides Lindb., Öfvers. Fin. Vet. Soc. Förhandl. 12: 76. 1869. Fig. 2

Plants soft, glossy, yellowish in distal part and dark-brown below; stems thin, 6–15 cm long, covered with leaves in lower portion or with partially destroyed leaves; subpinnately branched; branches loosely arranged, patent, secund, 4.9–15 cm long, slightly attenuate and curved at tips. Stem leaves loosely arranged, ca. 1 mm apart; straight or curved, firm, narrowly lanceolate, decurrent, slightly concave, narrowly acute, often tubulose-subulate distally, 3.8–5.8 mm long, 0.6–1.0 mm wide, with length: width ratio 5.5–8.7:1; slightly overlapping, forming 20°–30° angle with the stem; margins slightly recurved at places, entire or obscurely denticulate at apex. Median leaf cells linear, 120–210(–260) μm long, 12–15 μm wide, with length: width ratio 11–17.5:1. Alar cells enlarged, hyaline to yellowish or brownish, rectangular or almost hexagonal, slightly inflated, forming clearly differentiated alar group; cells in mid-leaf between alar groups short, thick-walled, with dark or orange-colored cell walls, together with dark alar groups making basal part of leaf darker than its distal part; leaf lamina is often constricted above alar groups. Gametangia and sporophytes absent in collection from Russia.

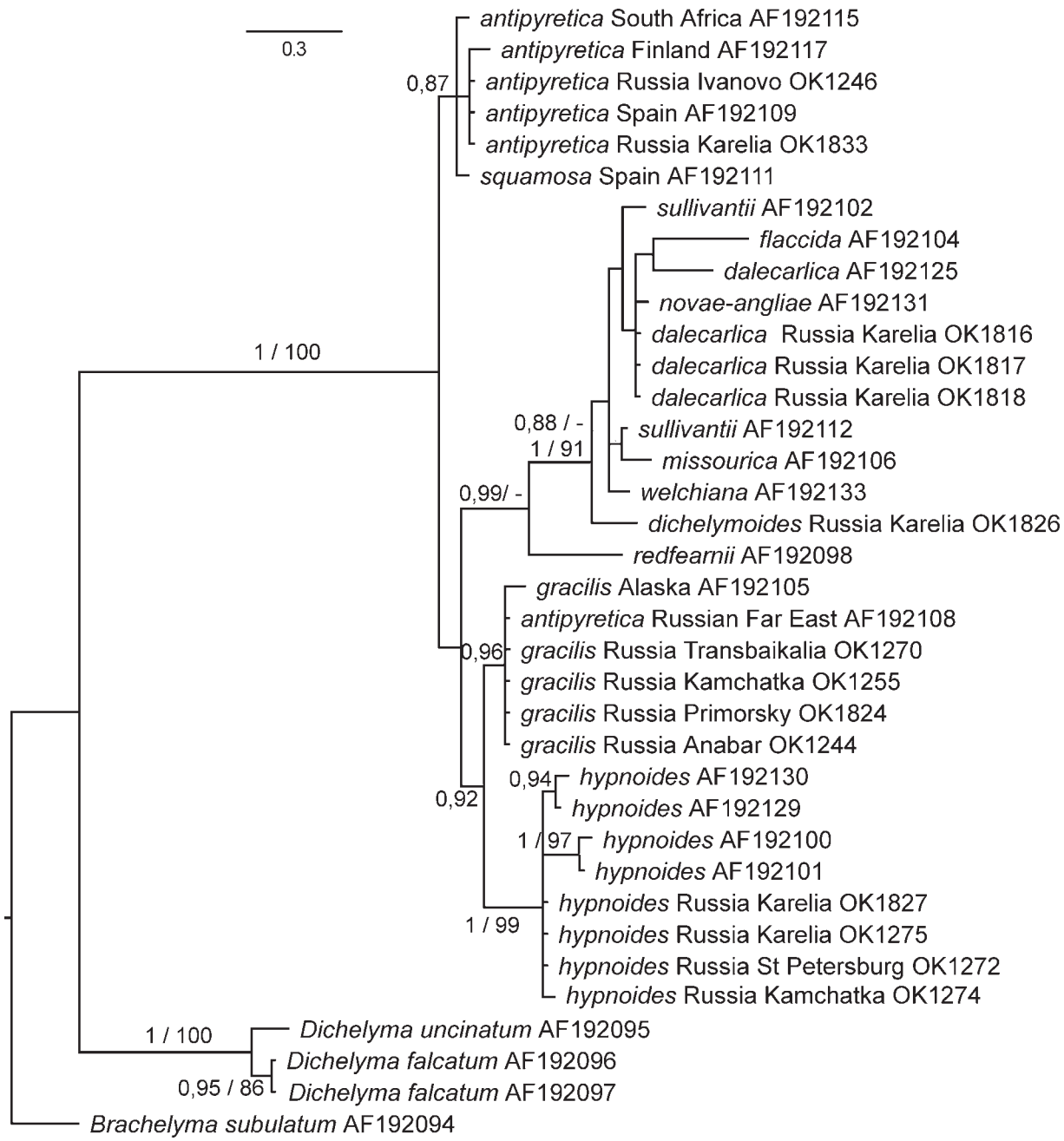

Fig. 1. Bayesian tree, showing position of *Fontinalis dichelymoides* in *F. dalecarlica*–clade. Posterior probabilities above 0.80 are shown at branches. Low posterior probablilites, <0.80 and bootstrap values <70 are not shown.

In Karelia *Fontinalis dichelymoides* was collected in the lake Nizhnee Ladwo, near the shore. Brotherus (1923) also mentions that this species occasionally grows on tree roots in stagnant water.

Differentiation. The habitat of *Fontinalis dichelymoides* is stagnant water, and this alone allows its differentiation in the field from *F. squamosa* and *F. dalecarlica*. Also, *F. dichelymoides* has the relatively longer leaves compared with these two species; their length to width ratio is 5.5–8.7:1, whereas it is 3.6–5.0:1 in *F. squamosa* and 3.2–4.4:1 in *F. dalecarlica*.

In leaves of *F. dichelymoides* marginal border is weakly differentiated or absent (0–1 cells wide), whereas in *F. squamosa* leaves have a clear marginal border 1–2 cells wide and in *F. dalecarlica* even better differentiated, 2–5 cells wide.

KEY TO IDENTIFICATION OF THE SPECIES OF *FONTINALIS* WITH CANALICULATE LEAVES

1. Tufts compact, formed of densely entangled, not secund shoots, often with numerous sporophytes; stem leaves concave, lanceolate, acute, 1.8–2.8 ×0.4–0.6 mm, with lendth: width ratio 3.6–5: 1; differentiated marginal cells in 1–2 rows (best seen in leaf trans-

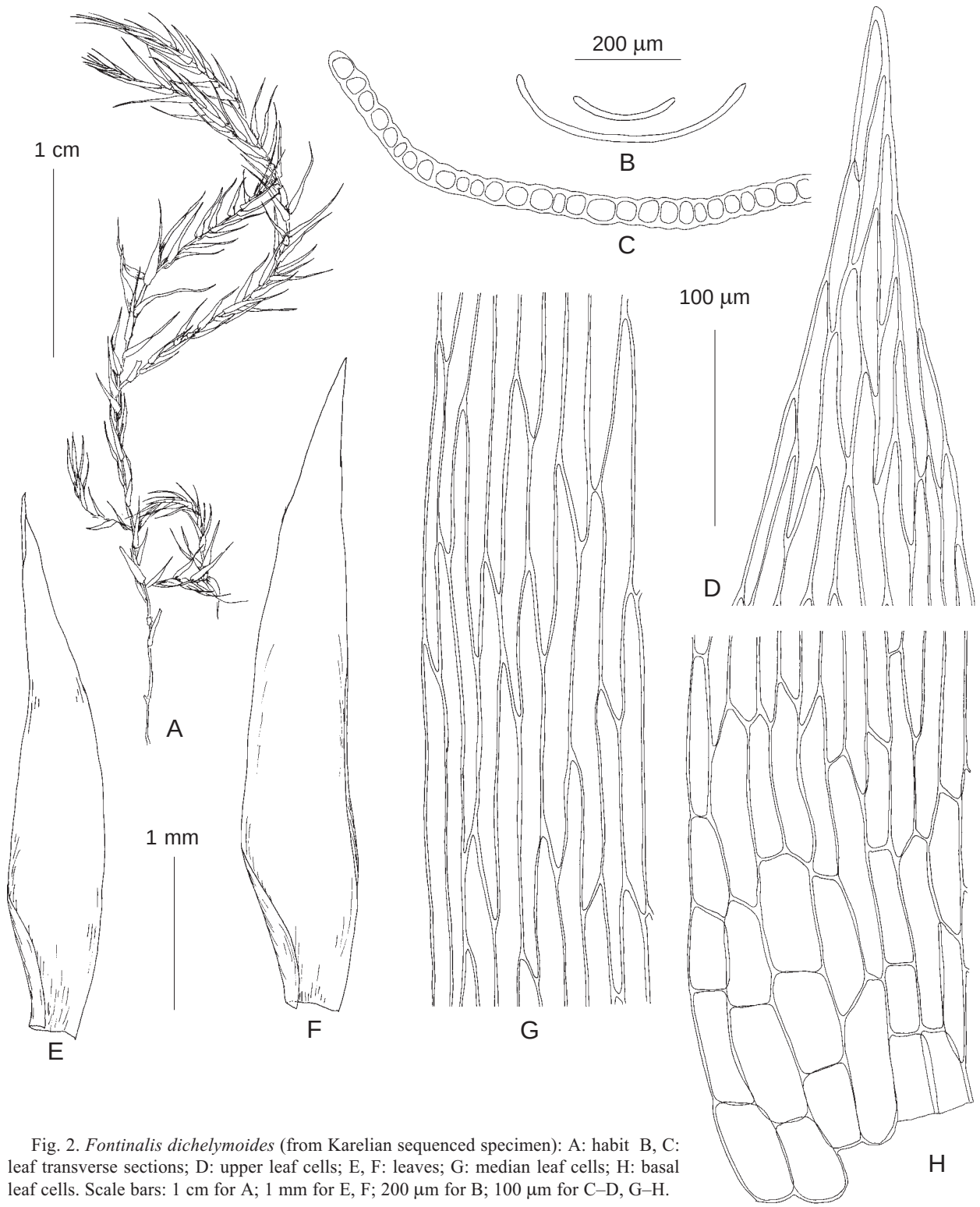

Fig. 2. *Fontinalis dichelymoides* (from Karelian sequenced specimen): A: habit B, C: leaf transverse sections; D: upper leaf cells; E, F: leaves; G: median leaf cells; H: basal leaf cells. Scale bars: 1 cm for A; 1 mm for E, F; 200 µm for B; 100 µm for C–D, G–H.

verse section); median leaf cells 90–135× 11.2–14 µm, 6.8–9.3:1; cell walls weakly thickened .... *F. squamosa*

— Tufts loose, stems and branches secund; sporophytes rare; stem leaves canaliculate; median leaf cells 7–12(–15) µm wide 2

2. Stem leaves narrowly lanceolate to linear, long acuminate, occasionally tubulose at apices, 3.8–5.8 ×0.6–1.0 mm, with length: width ratio 5.5–8.7:1; differentiated marginal cells in 0–1 rows; median leaf cells 129–210×12–15 µm, 11–17.5:1; cell walls weakly thickened ..... *F. dichelymoides*

Table 1. Specimen used in molecular phylogenetic analyses, with GenBank accession numbers.

Species	Vaucher, including herbarium acronym	Extraction	GenBank #	
			ITS	trnL–F
Fontinalis antipyretica	Russia, Ivanovo 4 July 2012 Sorokin & Golubeva sn (MHA)	OK1246	MK330647	
Fontinalis antipyretica	Russia, Karelia, Maksimov (PTZ 2634)	OK1833	MK330649	
Fontinalis dalecarlica	Russia, Karelia, Maksimov (PTZ 2657)	OK1816	MK330638	
Fontinalis dalecarlica	Russia, Karelia Maksimov (PTZ 2664)	OK1817	MK330639	
Fontinalis dalecarlica	Russia, Karelia Maksimov (PTZ 2652)	OK1818	MK330640	
Fontinalis dichelymoides	Russia, Karelia, Boichuk (PTZ 4745)	OK1826	MK330637	MK330933
Fontinalis gracilis	Russia, Zabaikalsky, Bazarova sn 15.IX.2015 (MHA)	OK1270	MK330645	
Fontinalis gracilis	Russia, Kamchatka, 20 July 2004 Czernyasjeva sn (MHA)	OK1255	MK330646	
Fontinalis gracilis	Russia, Primorsky Ignatov 06-3366 (MHA)	OK1824	MK330648	
Fontinalis gracilis	Russia, Anabar Plateau, Fedosov 08-287 (MHA)	OK1244	MK330650	
Fontinalis hypnoides	Russia, Karelia Maksimov (PTZ 24747)	OK1827	MK330641	
Fontinalis hypnoides	Russia, Karelia Boichuk sn, 10.VII.2000)LE)	OK1275	MK330642	
Fontinalis hypnoides	Russia, Leningrad, Leushina sn, 7.VIII.2005 (LE)	OK1272	MK330643	
Fontinalis hypnoides	Russia, Kamchatka, Czernyadjeva, 31.VII.2004 #29 (LE)	OK1274	MK330644	

— Stem leaves_lanceolate, acute, margins incurved in proximal part, 2.9–3.3×0.7–1.0 mm, with length: width ratio 3.2–4.4:1; differentiated marginal cells in 2–5(–6) rows; median leaf cells 90–171×8–14 μm, 7.5–14:1; cell walls strongly thickened .... *F. dalecarlica*

Specimens examined: RUSSIAN EUROPEAN NORTH-WEST: Republic of Karelia: National park "Kaleval'sky", vicinity of Ladvozero Village, Nizhnee (Lower) Ladvozero Lake, an island behind the dam (64°49'N, 29°52'E), in water near the shore, 10.VIII.1997, *Boychuk s.n.* (PTZ 4745); same place, 17.VIII.1997, *Boychuk s.n.* (PTZ 4746).

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