

A NEW *BRACHYTHECIUM* (BRACHYTHECIACEAE, BRYOPHYTA)
FROM THE RUSSIAN FAR EAST

НОВЫЙ ВИД *BRACHYTHECIUM* (BRACHYTHECIACEAE, BRYOPHYTA)
С РОССИЙСКОГО ДАЛЬНЕГО ВОСТОКА

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Abstract

Molecular phylogenetic studies revealed among collections from the Russian Far East an undescribed species of the genus *Brachythecium*. Morphological distinctions of *B. amurense* sp. nov. disagree with any species closely related according to the analysis of the nuclear ITS, the marker explored in most studied species of the genus. The fact that such large and conspicuous plant remained undiscovered indicates still incomplete knowledge of mountain moss flora of the Russian Far East and calls for more efforts for its exploration.

Резюме

В результате молекулярно-филогенетического анализа в коллекциях с российского Дальнего Востока выявлен не описанный вид из рода *Brachythecium*. Морфологические признаки *B. amurense* sp. nov. отличаются от признаков всех видов, которые близки к нему согласно результатам анализа последовательностей ядерного маркера ITS, изученного у большинства видов рода, для которых была исследована ДНК. Тот факт, что такое крупное, хорошо отличающееся растение до сих пор оставалось не описанным, свидетельствует о недостаточной изученности бриофлоры гор Дальнего Востока и о необходимости прилагать больше усилий для ее изучения.

KEYWORDS: mosses, taxonomy, molecular phylogenetic, new species, Russia

INTRODUCTION

The genus *Brachythecium*, even after splitting off *Sciuro-hypnum* and *Brachytheciastrum* (Ignatov & Huttunen, 2002) remains one of the largest genera in the moss flora of temperate regions of Northern Hemisphere (Hill *et al.*, 2006; Iwatsuki, 2004; Noguchi & Iwatsuki, 1991; Takaki, 1955, 1956; Wang, 2008; Ignatov, 2014) in general and moss flora of Russia in particular. In addition to this, recent molecular phylogenetic studies considerably increased the number of species of the genus in Russia. The last check-list of mosses of Russia (Ignatov *et al.*, 2006) included 20 species of *Brachythecium*. Subsequently, the broad screening with molecular markers tracked out the most obvious underdescribed lineages (Ignatov *et al.*, 2008; Ignatov & Milyutina, 2010), thus a preliminary treatment of the genus *Brachythecium* for the Moss Flora of Russia (version 14.IV.2012, <http://http://arctoa.ru/Flora/taxonomy-ru/Brachythecium-text-russian.pdf>) included already 29 species. Subsequent sequencing of enigmatic specimens mostly revealed range extension of some recent-

ly described species (Kuznetsova *et al.*, 2019), but one sample appeared to be conspicuous in morphology, as well as in sequence data, so it is described here as a new species.

The specimen was collected in alpine belt of Tardoki-Yani Mt., a bryological hotspot area. The catalogue of its moss flora was published by Fedosov *et al.* (2016) and this specimen was referred to *B. garovaglioides*, as this species superficially is most similar with this plant. However, more detailed investigation with the help of molecular markers showed that this identification needs reconsideration.

MATERIAL AND METHODS

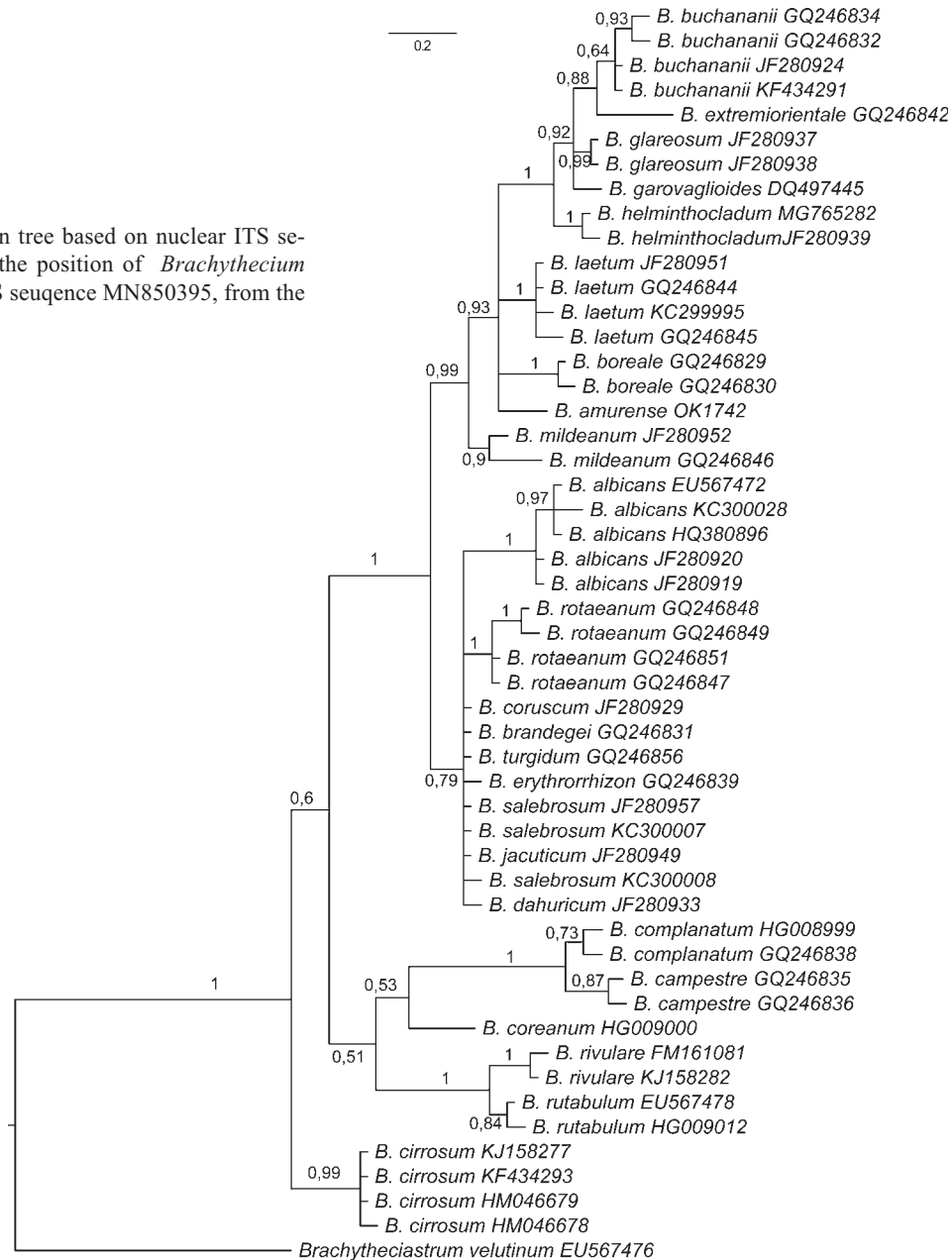
Taxon sampling included 51 accessions with outgroup genus, *Brachytheciastrum*, closely related to *Brachythecium*. The genus *Brachythecium* was represented by 50 accessions from 23 species, evenly covering the diversity of the genus in Russia. This sampling came out from the reduction of larger dataset, over 150 accessions, taken from GenBank by maximum similarity to specimen in question using BLAST search and alignments from our

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Рис. 1. Bayesian tree based on nuclear ITS sequences, showing the position of *Brachythecium amurense* (new ITS sequence MN850395, from the holotype).



previous studies. Preliminary trees inferred from this larger dataset had topology already found in the genus in earlier studies (Huttunen & Ignatov, 2004; Ignatov & Milyutina, 2010), thus numerous repeats of overrepresented species were removed.

Nuclear ITS region was selected, as the most variable and at moment the only suitable at the species level in the genus, where some morphologically distinct species have no one or only one substitution in ITS (cf. Ignatov & Milyutina, 2010). Laboratory protocols and information about PCR primers are given in Huttunen and Ignatov (2010) and Huttunen *et al.* (2008). DNA sequences were aligned by Clustal in Bioedit (Hall, 1999), with subsequent manual editing, in fact very small one. Phylogenetic analysis 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.

RESULTS

The molecular phylogenetic analysis resolved *Brachythecium* with the basal grade of species with rough seta, and two terminal clades of species with smooth setae: (1) *B. salebrosum*–*B. albicans*-clade (PP=0.79) , and (2) *B. mildeanum*–*B. laetum*–*B. b Buchananii*-clade (PP=0.99). In the latter, *B. mildeanum*-clade (PP=0.9) is sister to other species, which clade (PP=0.93) forms a tetratomy of (1) *B. b Buchananii*-clade (PP=1); (2) *B. laetum*-clade (PP=1); (3) *B. boreales*-clade (PP=1) and (4) *Brachythecium* from Tardoki-Yani Mt.

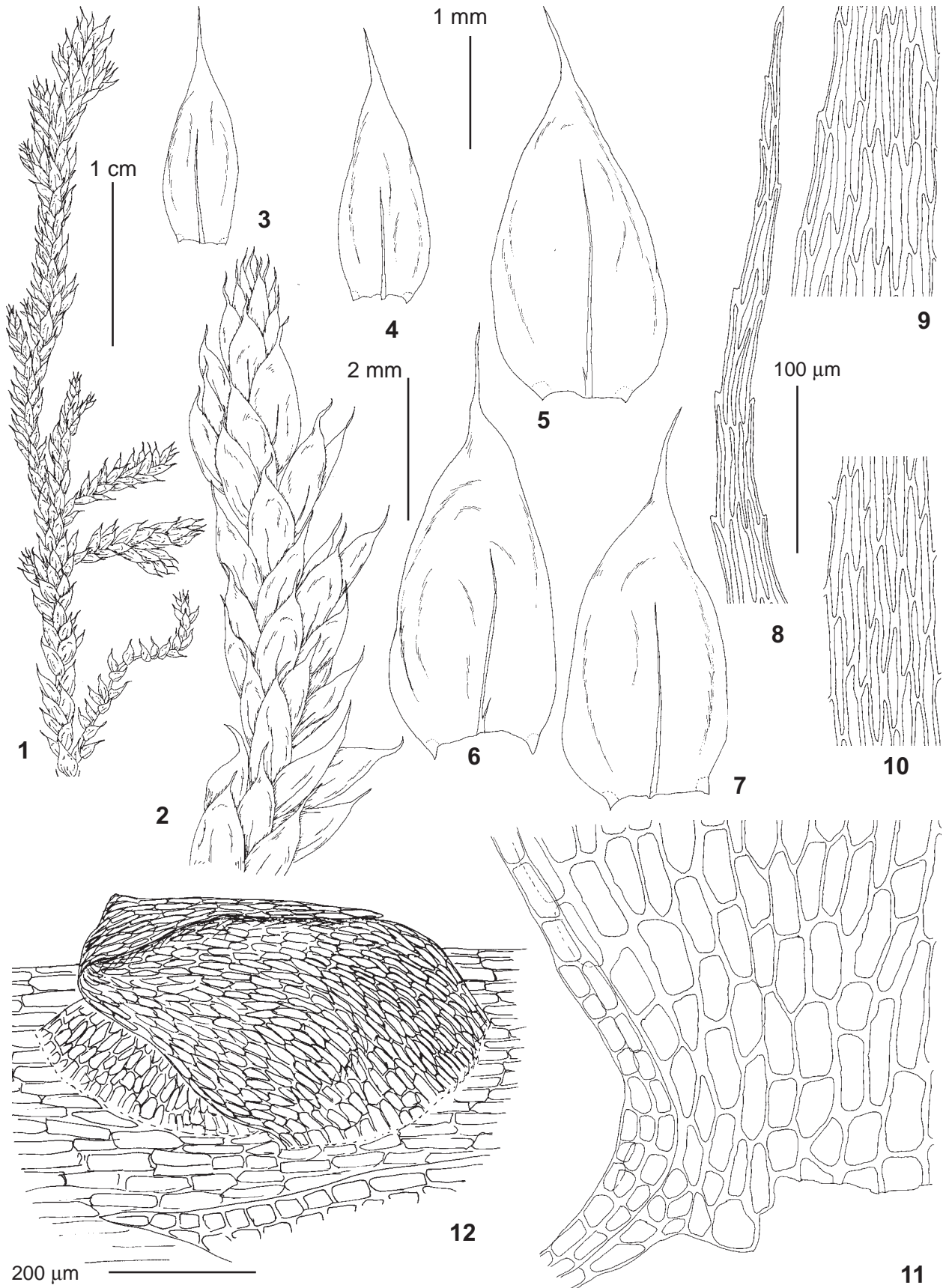


Fig. 2. *Brachythecium amurense* (from holotype). 1, 2 – habit, dry; 3–4 – branch leaves; 5–7 – stem leaves; 8 – upper laminal cells; 9–10 – median laminal cells; 11 – basal laminal cells; 12 – branch primordium (stem apex on the right). Scale bars: 1 cm for 1; 2 mm for 2; 1 mm for 3–7; 200 μm for 12; 100 μm for 8–11.

TAXONOMY

Indefinite phylogenetic position of the specimen in question (Fig. 1), with all nearest groups strongly different in morphology left no other choice than a description of a new species.

Brachythecium amurense Ignatov, sp. nov.

Type: Russia, Khabarovsk Territory, North Sikhote-Alin, ca. 1 km westward of Tardoki-Yani Mt. Peak, N: 48°53'16.9", E: 138°02'52.8", 1940 m, on soil. Coll. A.V. Ermolenko #7-4, 24.VIII.2013 (Holotype MW9112512, Isotype MHA).

Diagnosis. Differs from other species in combination of (1) robust pale-stramineous plants, weakly branched stems, terete foliate shoots; (2) leaves with ovate base, subpiliferous distally, strongly concave, crumple plicate; (3) margins minutely serrulate almost all around; (4) laminal cells 65–70(–90)×7–10 µm, moderately thick-walled, porose; (5) alar cells wider, extending upwards.

Description. Plants large, light yellowish-green, glossy, soft, forming loose tufts; outermost proximal branch leaf in branch primordia in 12 o'clock position, abruptly acuminate and consistently squarrose; axillary hairs with one apical and two basal cells, 45–60 µm long. Stems to 8 cm long, simple, with few short branches or irregularly branched, terete-foliate. Leaves erect-spreading to spreading when dry, crumpled, erect-spreading to loosely appressed when moist, 2–3×1.0–1.3 mm, ovate to ovate-triangular, widest at 1/6 the leaf length, abruptly narrowed into long filiform acumen, strongly concave (forming deep plicae in slides); margins plane or narrowly recurved at base and at the constriction to acumen, weakly serrulate in upper 3/4, subentire below; costa single, weak, quickly narrowing upwards, extending to 1/2–3/4 the leaf length; median laminal cells linear, 65–70(–90)×7–10 µm, slightly porose; basal laminal cells elongate-rectangular, 60–75×15–20 µm, porose; alar group moderately delimited; alar cells in several submarginal rows, not extending to costa, short-rectangular, 35–50×25–30 µm, moderately thick-walled, slightly porose. Apparently dioicous. Gametangia and sporophytes unknown.

Differentiation. Robust habit, crumple plicate leaves, and pattern of basal cell differentiation of *B. amurense* are similar to *B. garovaglioides* and, partly, also to *B. complanatum*. However, both these species have coarsely serrate leaves, and they belong to phylogenetically distant groups (Fig. 1). According to the obtained phylogeny, the closest to *B. amurense* are *B. mildeanum* (differs in entire leaf margins of rather narrowly triangular shape), *B. laetum* (differs in strongly plicate leaves and expanded group of opaque basal cells), and *B. boreale* (differs in medium-sized plants, rather short acumina, dense areolation throughout the leaf, except for a small group of enlarged cells in leaf corners).

Ecology. The species was collected at 1940 m a.s.l., in alpine belt, on soil on moist open slope, with *Philono-*

tis fontana, *Loeskyppnum badium*, *Campylium stellatum*, and *Hylocomiastrum pyrenaicum*.

The finding of such large and conspicuous moss once again illustrates the poor knowledge of high mountain diversity in Far Eastern region.

ACKNOWLEDGEMENTS

We thank support of the molecular study from RFBR 18-04-00574. The work was carried out in accordance with MBG Institutional research project 18-118021490111-5.

LITERATURE CITED

- FEDOSOV, V.E., A.V. ERMOLENKO, D.E. KOLTYSHEVA & V.A. BAKALIN. 2016. On the moss flora of Tardoki-Yani Mountain (North Sikhote-Alin, Russian Far East). – *Arctoa* **25**(2): 420–428.
- HALL, T.A. 1999. BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. – *Nucleic Acids Symposium Series* **41**: 95–98.
- HILL, M.O., N. BELL, M.A. BRUGGEMAN-NANNENGA, M. BRUGUÉS, M.J. CANO, J. ENROTH, K.I. FLATBERG, J.-P. FRAHM, M.T. GALLEGRO, R. GARILLETI, J. GUERRA, L. HEDENAS, D.T. HOLYOAK, J. HYVÖNEN, M.S. IGNATOV, F. LARA, V. MAZIMPAKA, J. MUÑOZ & L. SÖDERSTRÖM. 2006. An annotated checklist of the mosses of Europe and Macaronesia. – *Journal of Bryology* **28**: 198–267.
- HUTTUNEN, S., L. HEDENÄS, M.S. IGNATOV, N. DEVOS & A. VANDERPOORTEN. 2008. Origin and evolution of the Northern Hemisphere disjunctions in the moss genus *Homalothecium* (Brachytheciaceae). – *American Journal of Botany* **95**(6): 720–730.
- HUTTUNEN, S. & M.S. IGNATOV. 2004. Phylogenetic analysis of Brachytheciaceae (Bryophyta) based on morphology, and sequence level data. – *Cladistics* **20**: 151–183.
- HUTTUNEN, S. & M.S. IGNATOV. 2010. Evolution and taxonomy of aquatic species in the genus *Rhynchostegium* (Brachytheciaceae, Bryophyta). – *Taxon* **59**: 791–808.
- HUELSENBECK, J., & F. RONQUIST. 2001. MRBAYES: Bayesian inference of phylogenetic trees. – *Bioinformatics* **17**: 754–755.
- IGNATOV, M.S., O.M. AFONINA, E.A. IGNATOVA et al., 2006. Checklist of mosses of East Europe and North Asia. – *Arctoa* **15**: 1–130.
- IGNATOV, M.S. & S. HUTTUNEN. 2002. Brachytheciaceae (Bryophyta) – a family of sibling genera. – *Arctoa* **11**: 245–296.
- IGNATOV, M.S., I.A. MILYUTINA & V.K. BOBROVA. 2008. Problematic groups of *Brachythecium* and *Eurhynchiastrum* (Brachytheciaceae, Bryophyta) and taxonomic solutions suggested by nrITS sequences. – *Arctoa* **17**: 113–138.
- IGNATOV, M.S. & I.A. MILYUTINA. 2010. The genus *Brachythecium* (Brachytheciaceae, Musci) in Russia: comments on species and key for identification. – *Arctoa* **19**: 1–30.
- IGNATOV, M.S. 2014. *Brachythecium*. – In: *Flora of North America Editorial Committee, eds., Flora of North America North of Mexico. Vol. 28, pp. 414–428. New York and Oxford.*
- IWATSUKI, Z. 2004. New catalog of the mosses of Japan. – *Journal of the Hattori Botanical Laboratory* **96**: 1–182.
- KUZNETSOVA, O.I., E.A. IGNATOVA, V.E. FEDOSOV, D.YA. TUBANOVA & M.S. IGNATOV. 2019. Bryophyte molecular barcoding records. – *Arctoa* **28**: 250–251.
- NOGUCHI, A. & Z. IWATSUKI. 1991. Illustrated moss flora of Japan. Pt. 4. – *Hattori Botanical Laboratory, Nichinan*: 743–1012.
- TAKAKI, N. 1955. Researches on the Brachytheciaceae of Japan and its adjacent areas I. – *Journal of the Hattori Botanical Laboratory* **14**: 1–28.
- TAKAKI, N. 1955. Researches on the Brachytheciaceae of Japan and its adjacent areas II. – *Journal of the Hattori Botanical Laboratory* **15**: 1–69.
- WANG, Y.F. 2008. Brachytheciaceae. – In: *Hu, R.-L., Y.-F. Wang, M.R. Crosby & S. He. 2008. Amblystegiaceae—Plagiotheciaceae. Moss Flora of China. Vol. 7. Science Press & Missouri Botanical Garden, Beijing, New York & St. Louis: viii+ 258 pp.*