

Taxonomic identity of *Chionoloma bombayense* (Pottiaceae, Bryophyta)

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With 4 figures

Abstract: *Chionoloma bombayense* (Müll.Hal.) P.Sollman (Pottiaceae) has a long and complex taxonomic history. Recently it was viewed as a species with over 50 synonyms and considered the only species of the genus *Pseudosymblepharis* Broth., and later as *Chionoloma* Dixon, in Asia and Africa. Some phylogenetic studies have now suggested that more than one species could be recognized within *C. bombayense*. Here, in order to complement these molecular data, a morphological analysis is carried out. The concept of the species is significantly reduced, it is considerably more restricted in distribution and several names previously applied to this taxon need to be resurrected. Therefore, we present a complete description of the species, an updated distribution map and illustrations of the diagnostic characters. A lectotype for *Trichostomum perannulatum* Dixon & P.de la Varde is designated herein.

Key words: Chionoloma, description, distribution, morphology, taxonomy, splitting.

Introduction

Trichostomum bombayense Müll.Hal. is a moss belonging to the family Pottiaceae. It was described by Müller (1851) on the basis of material originating from India. The name was transferred to *Tortula* Hedw. by Mitten (1859) and later to *Didymodon* Hedw. by Jaeger (1873). The species was neglected for many years and resurrected by Townsend (1983). After examination of the type material as well as other collections from India, Townsend treated this taxon under *Trichostomum* Bruch. Sollman (2000) combined it into *Pseudosymblepharis* Broth. as *Pseudosymblepharis bombayensis* (Müll.Hal.) P.Sollman. Eddy (1990) considered the genera *Pseudosymblepharis* and *Chionoloma* Dixon as synonyms, although it was Menzel (1992) who provided the required new combinations in *Chionoloma*. Based on Eddy's work, Sollman

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(2001) transferred P. bombavensis to Chionoloma, Sollman (2000, 2005) carried out the taxonomic revision of *Pseudosymblepharis* (Chionoloma) in Asia and Africa, respectively, and he concluded that the genus consisted of only one species, C. bombayense (Müll.Hal). P.Sollman. A total of 42 names from Asia and 11 from Africa were reduced to synonymy. He pointed out that there were several trends present in Asian and African material. However, he did not give these tendencies any taxonomic status since they were connected by intermediates. *Chionoloma bombayense*, therefore, has been considered the only species remaining in the genus *Pseudosymblepharis*, and later as Chionoloma in these continents for many years (Grundmann et al. 2006, Köckinger et al. 2010, Ignatova et al. 2012). Under this circumscription it has been extensively reported for many countries. In Africa it is known from Cameroon, Comoro Island, Democratic Republic of Congo, Ethiopia, Guinea, Ivory Coast, Kenva, Lesotho, Madagascar, Malawi, Mauritius, Réunion, Rwanda, São Tomé and Príncipe, Sierra Leone, South Africa, Tanzania, Uganda, Yemen and Zimbabwe (O'Shea 2006, Sollman et al. 2016). In Asia, C. bombayense has been reported from Georgia and Russia (Ignatova et al. 2012), Indonesia (Ho et al. 2006), Japan (Iwatsuki 2004), Sri Lanka (O'Shea 2002), United Arab Emirates (Kürschner & Frey 2011) and Vietnam (He & Nguyen 2012). Finally, in Oceania it is known from Australia (Jolley & Milne 2012).

In the last few years, we have focused on a revision of *Chionoloma* (Alonso et al. 2014a, b, Alonso & Long 2014, Alonso et al. 2016a). The morphological and molecular phylogenetic study carried out by Alonso et al. (2016b) was based on three chloroplast loci (*atpB-rbcL*, *trnG* and *trnL*-F) and the nuclear locus ITS. This work included many taxa considered as synonyms of *C. bombayense*, such as *C. angustatum* (Mitt.) M.Menzel, *C. crassicostatus* (D.H.Norris & T.J.Kop.) M.Alonso, M.J.Cano & J.A.Jiménez, *C. dubium* (Thér.) M.Alonso, M.J.Cano & J.A.Jiménez, *C. induratum* Dixon and *C. perlongifolium* (J.Fröhl.) M.Alonso, M.J.Cano & J.A.Jiménez. Unfortunately, neither samples of *C. bombayense* s.str. nor some of its synonyms could be located in sufficiently good condition for DNA study, and therefore, were not included in this work. Even so, the phylogeny suggested that *C. bombayense* could be polyphyletic since specimens identified as *C. bombayense* sensu Sollman were positioned in different clades (Fig. 1).

Based on these previous molecular data (Alonso et al. 2016b) as well as an exhaustive morphological analysis performed here, the concept of *C. bombayense* is revised. A complete description of the species is presented, its distribution updated, and the principal distinctive characters that separate it from the nearest species of *Chionoloma* are analysed and illustrated.

Materials and methods

About 200 collections previously identified as *C. bombayense* from the following herbaria were examined: BM, BOL, BORH, E, EGR, H, JE, KLU, KRAM, L, MO, MUB, NY and S. In addition, more than 50 types including those studied by Sollman (2000, 2005) were compiled and analysed morphologically. The geographic range of the examined samples covered four continents (Africa, America, Asia and Oceania), and the following countries: Australia, Bhutan, Burma, China, Democratic Republic of the Congo, Colombia, Ethiopia, Guinea, India, Indonesia, Japan, Kenya, Lesotho, Malawi, Malaysia, Nepal, Pakistan, Papua New Guinea, Peru, Réunion, Rwanda, Sri Lanka,



Fig. 1. Majority-rule consensus tree of the Bayesian inference analysis for three chloroplast loci (*atpB-rbcL*, *trn*G and *trnL*-F) and the nuclear locus ITS modified from Alonso et al. (2016b). Bayesian posterior probabilities (PP) followed by maximum likelihood bootstrap values (MLBS) are shown above the branches. Branches in bold are supported by both analytical methods (PP \geq 0.95, and MLBS \geq 70). Support values of PP < 0.95 and BS < 70 are not shown. Synonyms of *C. bombayense* sensu Sollman (2000, 2005) are in bold marked with an asterisk (*).

South Africa, Tanzania, Thailand, The Philippines, Uganda, Union of the Comoros, Venezuela and Zambia.

All collections were examined using the typical anatomical and morphological methods applied to the Pottiaceae (Zander 1993). Microscopic examinations and measurements were done with an Olympus-BX41 light microscope, with microphotographs obtained with a Jenoptik ProgRes C7 camera mounted on it. Specimens were examined in 2% potassium hydroxide (KOH). Three shoots were dissected from each examined collection. Leaves were always taken from the upper middle of the stem and stem cross-sections were made in the middle part of stem. Measurements of leaf width were taken at base, mid-leaf and in the distal leaf portion (Fig. 2: 1–3). Cross-sections were made at mid-leaf. Five sorts of basal laminal cells were differentiated according to where they are placed. They are central basal cells, juxtacostal basal cells, cells of the lower parts of basal area, transitional cells and basal marginal cells (Fig. 2: 4–8). Morphological characters examined are detailed in the descriptions. Those considered taxonomically important for *C. bombayense* (Townsend 1983, Sollman 2000, 2005, Aziz & Vohra 2008, Ignatova et al. 2012) were included in the examination.

The International Code of Nomenclature for algae, fungi, and plants (ICBN) (McNeill et al. 2012) and the recommendations presented by McNeill (2014) were strictly followed for the nomenclatural revision.

Results

The previous phylogenetic analysis (Alonso et al. 2016b, Fig. 1) as well as the morphological examination performed here suggests than more than one species can be considered within *C. bombayense*. Consequently, we redefine the concept of this species and update its description, nomenclature and distribution.

Chionoloma bombayense (Müll.Hal.) P.Sollman, Trop. Bryol. 20: 139. 2001.

Trichostomum bombayense Müll.Hal., Syn. Musc. Frond. 2: 628. 1851 = Tortula bombayensis (Müll.Hal.) Mitt., J. Proc. Linn. Soc., Bot., Suppl. 1: 28. 1859 = Didymodon bombayensis (Müll.Hal.) A.Jaeger, Ber. Thätigk. St. Gallischen naturwiss. Ges. 1871–1872: 368. 1873 = Pseudosymblepharis bombayensis (Müll.Hal.) P.Sollman, Trop. Bryol. 12: 2. 1996. TYPE: [India]. Bombay, Kegelianum 10025 [lectotype, designated by Sollman & Een (1996): H-BR No. 4299024!; isolectotypes: JE!, S No. B180402!]. Fig. 3.

= *Trichostomum perannulatum* Dixon & P.de la Varde, Ann. Cryptog. Exot. 1: 280. 1928 ≡ *Oxystegus perannulatum* (Dixon & P.de la Varde) M.N.Aziz & Vohra, Pottiac. India 8: 71. 2008. Type: [India]. [Maharastra]: Mahableshwar, Wester Ghats, 1918, Sedgwick 4726 (lectotype, DESIGNATED HERE: BM barcode 001006322!; isolectotype: PC barcode 0150963!), synonymized by Sollman (2000).

PLANTS 0.2–1.9 cm high, green to dark green. STEMS usually simple; in cross-section round, 140–250 μ m in diameter, sclerodermis weakly developed, with 1 layer of thicker-walled cells, central strand undifferentiated; not tomentose; axillary hairs of 7–21 hyaline cells. RHIZOIDS rarely papillose in the early stage, later smooth. LEAVES appressed and crispate when dry, erect to patent, rarely spreading when moist, lanceolate, rarely obovate, (1.1–)1.7–2.8(–3.2) mm length, 0.26–0.52(–0.61) mm wide at base, 0.17–0.34(–0.45) mm wide at midleaf, 0.09–0.25(–0.30) mm wide at upper; base undifferentiated or weakly differentiated by shoulders, not plicate; lamina yellowish-orange in KOH; margins plane, entire at base, papillose-crenulate, seldom weakly, from near base to apex, not dentate, sinuous, unbordered; apex acute, rarely obtuse and cuspidate; costa (32.5–)50–87.5 μ m wide at base, 20–30 μ m wide at apex, percurrent, rarely shortly excurrent in a mucro, (25–)32.5–75(–87.5) μ m long, ventral



Fig. 2. Areas where the width of the leaf was measured and basal cells were established. 1. Leaf width at upper. 2. Leaf width at midleaf. 3. Leaf width at base. 4. Transitional cells. 5. Juxtacostal basal cells. 6. Basal marginal cells. 7. Central basal cells. 8. Cells of the lower part of basal area.

surface cells quadrate and papillose from near base to apex, vanishing near the apex, dorsal surface cells linear and smooth from base to apex, seldom short-rectangular toward apex, in cross-section at midleaf elliptical, seldom round, with 2–6 guide cells in 1 layer, dorsal stereid band larger, or about the same size as ventral stereid band, (1-)2-4 layers of ventral stereids, 2–4 layers of dorsal stereids, ventral surface cells elliptical to rounded, formed by 2–6(–7) cells, 4–9.6 × (4.8–)6.4–10.4(–11.2) µm, papillose, dorsal surface cells undifferentiated; upper and middle laminal cells quadrate, 6.4–13.6(–17.6) × (5.6–)6.4–12.8 µm, bulging on both sides, with simple or bifid papillae, upper and middle marginal cells undifferentiated; transitional cells entire, smooth; central basal cells rectangular, seldom short-rectangular, 20–82 × 8.8–30 µm, inflated, seldom



Fig. 3. *Chionoloma bombayense*. 1. Leaf. 2. Leaf apex. 3. Leaf base. 4. Juxtacostal basal cells. 5. Cross-section of the costa at midleaf. 6. Cross-section of the stem [1, 2, 5 from Miehe & Miehe 00-86-36 (L); 3, 4, 6, from Townsend 73/1776 (E)].

weakly, hyaline, thin-walled, entire, forming a differentiated area like an inverted "U", covering (11–)14–28% of leaf length; juxtacostal basal cells not differentiated from central basal cells; cells of the lower part of basal area not differentiated from central basal cells; basal marginal cells rectangular, $(12-)15-45(-50) \times (5-)6.3-$



Fig. 4. Map of the updated geographical distribution of Chionoloma bombayense.

11.3 μ m, not inflated, extending up the margins to 11–24% of leaf length, not forming a V-shaped pattern. Specialized asexual reproduction absent. SEXUAL CONDITION dioicous. PERICHAETIAL LEAVES undifferentiated. SETA ca. 10 mm long, twisted to the left, yellowish to brownish. THECA cylindrical, ca. 1.4 × 0.50 mm, orange; exothecial cells 40–60 × 14–18 μ m, thin-walled; annulus of 3 rows of vesiculose cells; peristome of 16 filiform teeth, 87.5–125 μ m long, grey to yellowish, striate; basal membrane weakly developed, smooth. OPERCULUM not seen. CALYPTRAE not seen. Spores 15–17.5 μ m in diameter, brownish, papillose.

HABITAT: Epiphyte or sometimes growing on rocks; altitudinal range from 1200 to 3260 m.

DISTRIBUTION: Bhutan, Democratic Republic of the Congo and India (Fig. 4).

NOMENCLATURAL NOTE: *Trichostomum perannulatum* was described by Dixon and P.de la Varde (Potier de la Varde 1928) based on material collected by L.J.Sedgwick

from South India. Two duplicates of the type collection exist and are housed in the original herbaria of the authors of this name (BM and PC). Both specimens match the protologue, thus the collection from BM is designated as lectotype because it includes more material and exhibits sporophytes.

ADDITIONAL SPECIMENS EXAMINED: BHUTAN. Trashiyangtse: Trashiyangtse Pang La E side Flor-Prov, C 16 (Tashingang), 8 June 2000, Miehe & Miehe 00–86–36 (L). DEMOCRATIC REPUBLIC OF THE CONGO. Katanga: Shaba, Mukuen, Lubumbashi, Haut-Ahaba, env. De Lubumbashi, près de Keyberg, 12 December 1968, Lisowski 4306 (L). INDIA. Maharashtra: western Ghats, on the opposite side of the road from the park, Mahabaleshwar, 14 January 1973, Townsend 73/423 (E); Townsend 73/1776 (E).

Discussion

Fig. 1 displays a modification of the majority-rule consensus tree of the Bayesian inference analysis performed by Alonso et al. (2016b). Only the *Chionoloma* clade and its closer groups are provided in this figure. The six taxa considered co-specific with *C. bombayense* sensu Sollman and included here (*C. angustatum*, *C. crassicostatus*, *C. dubium*, *C. duriuscula*, *C. induratum* and *C. perlongifolium*) are marked with an asterisk (*) and in bold. They were placed in seven different clades. Samples of each taxon formed a distinct lineage, except for specimens of *C. angustatum* which were split in two different clades. To sum up, the data suggested that *C. bombayense* sensu Sollman is polyphyletic and, together with the morphological examination, encouraged us to split *C. bombayense* into at least seven species (*C. angustatum*, *C. bombayense*, *C. crassicostatus*, *C. dubium*, *C. duriuscula*, *C. induratum* and *C. perlongifolium*). Future studies including collections of *C. bombayense* s.str. are still required to clarify the evolutionary position of the species.

According to the morphological analysis, C. bombayense belongs to genus Chionoloma because it presents a stem with hyalodermis and undifferentiated central strand, a costa with two stereid bands and developed ventral surface cells, leaves with well differentiated basal cells and a peristome formed by 16 straight teeth. At species level, it is characterized by unbordered and short leaves, the percurrent leaf costa with the dorsal band larger than the ventral band, and the basal laminal cells forming a differentiated area like an inverted "U", and not extending up along the margins to form a V-shaped transition to the upper cells. During the taxonomic revision of Chionoloma, the 53 names synonymized with C. bombayense by Sollman (2000, 2005) were examined. Among them, only the name *T. perannulatum* is still treated as a synonym of C. bombayense because it is the only one with the character states listed above. Aziz & Vohra (2008) considered this taxon as a single species of Oxystegus (Limpr.) Hilp. in India (O. perannulatum), whereas they placed C. bombayense within Trichostomum. These authors separated both genera (Oxystegus and Trichostomum) by the presence or absence of the central strand and the notched or entire apex. However, no morphological differences have been found between the two type specimens and, therefore, their names are here treated as synonyms.

Only four of the nearly 200 collections previously identified as *C. bombayense* presented the combination of character states observed in the type material. Most of the

samples had been identified as *C. tenuirostre* (Hook. & Taylor) M.Alonso, M.J.Cano & J.A.Jiménez or *C. angustatum* (Mitt.) M.Menzel. *Chionoloma bombayense* is similar to *C. tenuirostre* in the lanceolate leaves and the costa in cross-section with dorsal stereid band larger than the ventral band. However, they differ in the shorter leaves and stems of *C. bombayense*, as well as in the basal laminal cells forming an inverted "U" pattern. *Chionoloma angustatum* and *C. bombayense* have a hyalodermis, a costa with two stereid bands and ventral surface cells differentiated. In addition, both of them can have papillose rhizoids in the early stage. However, in *C. angustatum* the leaf base is differentiated by shoulders, it is usually plicate and the size of the leaf base is usually undifferentiated, not plicate and narrower in size [0.26–0.52(–0.61) mm wide]. The long mucro of *C. angustatum* (100–400 µm long) also readily differentiates this species from *C. bombayense*, whose mucro just reaches up to 75(–87.5) µm in length.

Morphologically, *C. hibernicum* (Mitt.) M.Alonso, M.J.Cano & J.A.Jiménez is most similar to *C. bombayense*, in terms leaf shape and anatomy of the costa, and both of their leaves react similarly to KOH. However, *C. hibernicum* has a wider leaf base (0.43–1.21 mm), stem sclerodermis with 2 or 3 layers of thicker-walled cells and basal marginal cells extending up the margins, forming usually a V-shaped pattern. In *C. bombayense*, leaf base is narrower [0.26–0.52(–0.61) mm], the sclerodermis is formed by 1 layer of thicker-walled cells and the leaf base lacks the V-shaped pattern.

The morphological study performed here suggests that *C. bombayense* is a rarely encountered species. Its distribution therefore has been drastically reduced from earlier reports to two Asian countries (Bhutan and India) and one locality in Africa (Democratic Republic of the Congo). Likely, it could be found in other regions from Central Africa, the Indian subcontinent or the Himalayas, growing as an epiphyte or on rocks between about 1000 and 3000 m. Field trips to these areas would be required to determine the actual distribution of this species.

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