

# ***Chiastocaulon fimbriatum* (Plagiochilaceae) a morphologically variable species new for China, with a description of sporophytes discovered in Yunnan**

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**Abstract** – *Chiastocaulon fimbriatum* is reported new for China based on recent collections from Yunnan. It is a variable species and an updated description is provided with additional details of the newly discovered sporophytes and male plants. *Chiastocaulon fimbriatum* is characterized by the following features: 1) robust plant size with orbicular leaves; 2) cells in the upper part of leaf thin-walled yet with trigones; 3) leaves slightly decurrent dorsally; 4) inner bracts horizontally spreading on ventral side with multiple minute teeth on irregularly-shaped lobes; and 5) large spores. Through different stages of growth, *C. fimbriatum* exhibits a great variety in the shape of both leaf and perianth. The large cylindrical perianth is another important diagnostic character of this species; however, when immature, the perianths are of little taxonomic value since they can be conical or campanulate in shape at different stages of development. Based on newly obtained material, we failed to observe decurrence of leaves on the dorsal side of the stem to be a key diagnostic feature for this species. A key for the genus in China is provided, based on specimens from Yunnan.

**China / *Chiastocaulon fimbriatum* / distribution / Plagiochilaceae / range extension / sporophytes / Yunnan**

## INTRODUCTION

The genus *Chiastocaulon* Carl (Plagiochilaceae) is disproportionately represented by a very limited number of specimens from China. Unlike most other easily overlooked bryophytes, the size, branching pattern and the neat opposite foliage (for almost all species), make *Chiastocaulon* readily recognizable in the field. Nonetheless, it is so under-collected that the lack of both historical specimens and recently collected and silica-dried materials has greatly limited our understanding of this group.

*Chiastocaulon fimbriatum* (Mitt.) S.D.F. Patzak, M.A.M. Renner, Schäf.-Verw. & Heinrichs was initially described by a collection from Khasia Mountains,

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North-east India (Mitten 1861, as *Plagiochila fimbriata*). Due to its opposite leaves, it was later transferred to the genus *Plagiochilion* S.Hatt. after Inoue (1964) had studied the type specimen. Nonetheless, Inoue's updated description was based on only two individual stems, namely all material available for this taxon at that time. The new combination *Chiastocaulon fimbriatum* was proposed by Patzak *et al.*, (2016) based on molecular insights provided by several other taxa previously placed within *Acrochila* and *Plagiochilion*.

*Chiastocaulon fimbriatum* is among the least understood species within the genus in Asia, although it has been reported from Thailand (Inoue, 1967) and Bhutan (Long & Grolle, 1990) outside its type locality in India (Mitten 1861; Majumdar *et al.*, 2013). Because of this, the understanding of this species is largely incomplete as no recent voucher specimens have been studied in detail, and its sporophyte was unknown. The limited information available for this species may have led to its misidentification.

During a bryophyte inventory work for the Fen-Shui-Ling National Nature Reserve, Jin-Ping County in southeast Yunnan in 2015, the first author encountered two fertile specimens of *Plagiochilion* in the field. They were later identified as *Chiastocaulon fimbriatum*, a new liverwort member for the bryoflora of China. A comprehensive survey among the unidentified specimens in both authors' herbaria yielded three more populations within the province. After examination of these newly obtained voucher specimens from Yunnan, *C. fimbriatum* turned out to be morphologically variable and we decided to elaborate the description accompanied by photos of male plants and sporophytic features that were previous unknown for this species.

## METHODS

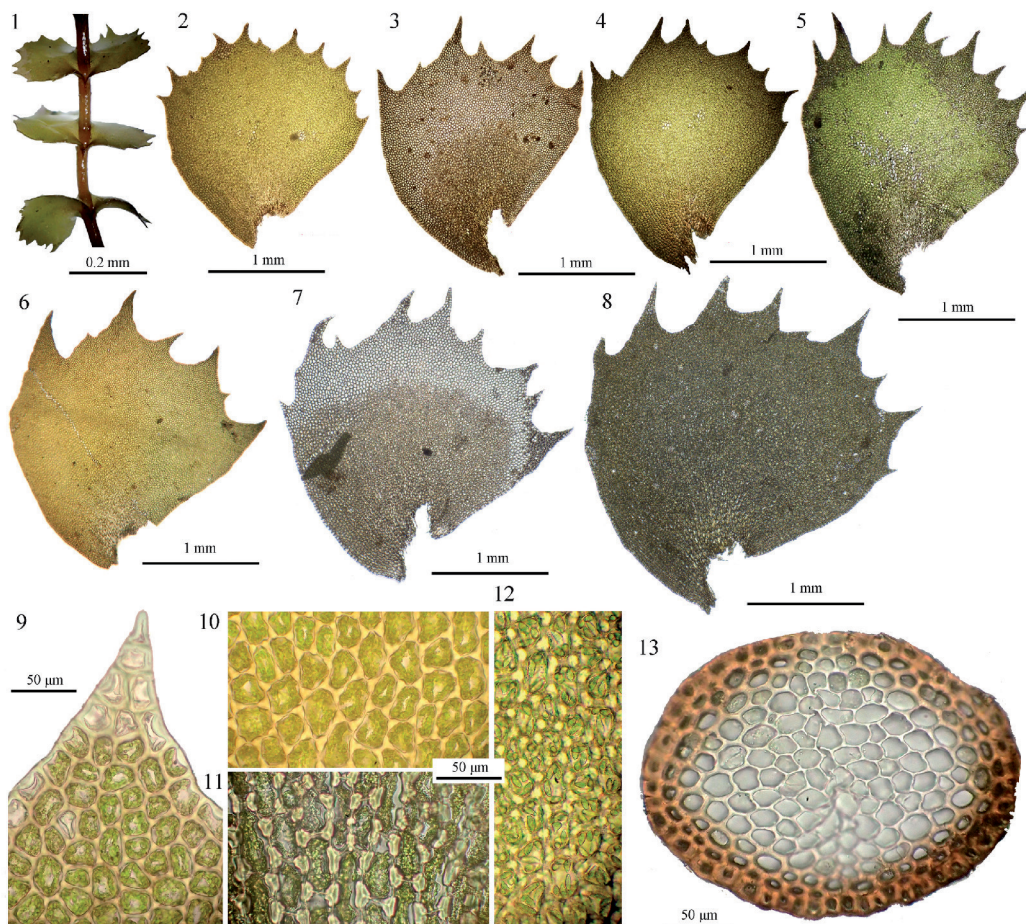
All measurements of leaf and cells sizes were followed by Paton (1999), and oil bodies were counted according to Wagner (2013). For a given specimen, in a particular lamina area, the largest cells were chosen for observation and all its neighboring cells along with the central cell were all measured to generate the data range and mean value for both length and width. Spores and elaters were chosen for measurement randomly. Due to the limited number of perianths and male and female bracts, their measurement were done based on the availability of the material.

## TAXONOMIC DESCRIPTION

***Chiastocaulon fimbriatum*** (Mitt.) S.D.F. Patzak, M.A.M. Renner, Schäf.-Verw. & Heinrichs *Organisms Diversity and Evolution* 16 (3): 492. 2016 **Figs 1-27**

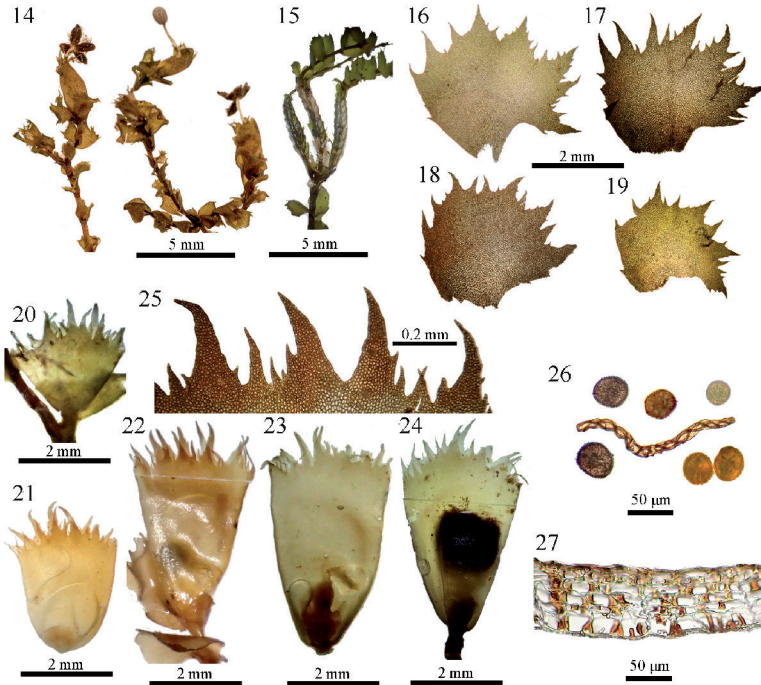
Basionym: *Plagiochila fimbriata* Mitt. (Mitten, 1861).

Plants 2.6-4.8 cm long, 3.3-5.1 mm wide with leaves, green to brownish green, branching frequently, stolons present. Stem elliptical in transverse section, 0.28-0.39 × 0.18-0.27 mm (Fig. 13), with two (or three) layers of thick-walled brownish epidermal cells. Leaves mostly opposite, nearly contiguous when dry,



Figs 1-13. Leaf shape and cells of *Chiastocaulon fimbriatum*. **1.** Dorsal view of plant. **2-8.** Leaves. **9.** Apical cells. **10.** Middle cells. **11.** Middle basal cells. **12.** Oil bodies in middle cells. **13.** Cross section view of stem. (1, 3, 7-11 from *W.Z. Ma 15-7043*; 2, 4, 5 & 12 from *W.Z. Ma 17-8968*; 6 & 13 from *W.Z. Ma 17-8929*).

horizontally spreading when wet, slightly decurrent on dorsal side, united at ventral base, triangularly to orbicular, middle leaves have longer widths than lengths, while upper and lower leaves tend to exhibit longer lengths. Leaves 1.7-2.8 (averaged 2.2) mm long, and 1.8-3.0 (averaged 2.3) mm wide. Marginal teeth 7-11 (-14) in total, among which 5-8 are large and triangular ones accompanied by 1-4 (-6) tiny teeth formed of only a few projecting cells. In the apical region, cells thin-walled, (14.3- ) 18.1-30.0 (-35.8)  $\mu\text{m}$  long, (13.6) 16.0-25.9 wide, trigones clearly visible. In the mid-leaf, cells thin-walled, (16.7-) 24.6-36.8 (-42.4)  $\mu\text{m}$  long and (16.7-) 21.1-28.9  $\mu\text{m}$  wide, trigones become larger, some cells appear nodulose. In the middle base of lamina, cells intensely elongate forming a vitta-like area, (39.3-) 45.7-65.4 (-77.0)  $\mu\text{m}$  long, (17.4-) 22.3-26.1 (-38.4)  $\mu\text{m}$  wide, cell walls moderately thickened, intermediate thickening present, with very large trigones and strongly nodulose (Fig. 11).



Figs 14-27. Sporophytic features of *Chiastocaulon fimbriatum* from Yunnan. **14.** Plants with capsules. **15.** Male plant. **16-19.** Female bracts. **20-24.** Perianths. **25.** Mouth of perianth. **26.** Spores and elater. **27.** Cross-section of exothecium cells. (14-16, 20, 23-24, 26-27 from *W.Z. Ma 15-7043*; 17, 22, 25 from *Long 32672*; 18 & 21 from *W.Z. Ma 14-5441*; 19 from *W.Z. Ma 15-7033*).

Oil body rJung-type (Juslén *et al.*, 2001) (Fig. 12), 4.7-5.3  $\mu\text{m}$  in diameter if globose, and 8.0-9.8  $\mu\text{m} \times 4.4$ -5.2  $\mu\text{m}$  if oblong, 4-6 per cell in the apex and middle of lamina, and 4-8 per cell in the basal area.

Dioicous. Male inflorescences terminal and intercalary, bracts 9-11 pairs, closely imbricate (Fig. 15), the dorsal margin strongly incurved, 3-4 small teeth present at the apex, 1.1-1.3 mm long, and 0.7-0.8 mm wide. Female bracts terminal, mostly in 2 pairs. Inner bracts widely ovate, concave, horizontally spreading on the postical half of bracts (Figs 16-19), 2.8-3.6 mm long and 3.1-3.8 mm wide. Margins 3-5 lobed, lobes irregular in size, triangular, acuminate, often with small additional teeth on ventral half the margin. Outer bracts similar in shape but smaller in size, with less marginal dentations, 1.9-3.4 mm long and 2.6-3.3 mm wide. Mature perianth cylindrical, mouth truncate or slightly arched, irregularly dentate (Fig. 25), 3.7-5.3 mm long and 2.3-2.7 mm wide at the mouth. Archegonia 26-29 in number. Sub-floral innovations (mostly one, occasionally two) frequently appear below perianth. Setae 3-4 mm long at maturity. Capsule oblong, wall composed of 5-6 layers of cells, about 65-80  $\mu\text{m}$  thick (Fig. 27).

Spores spherical to slightly elliptical, finely papillose, 28.3-39.4  $\mu\text{m}$  in diameter, with the longer axis range from 28.4 to 40.4  $\mu\text{m}$ , and the shorter axis range from 28.3 to 39.4  $\mu\text{m}$  (Fig. 26). Elaters brown, bi-spiral, 9.7-12.0  $\mu\text{m}$  wide, 180-260  $\mu\text{m}$  long (Fig. 26).

**Specimens examined:** CHINA. Yunnan Province. Baoshan Region, Long-Ling Co., Nan-Kang Yakou (pass), on mossy tree trunk, 24°49'45"N, 98°45'57"E, 2180 m, *Long 32672* (CAS, E, KUN, MO); Honghe Region, Jin-Ping Co., Zhe-Mi Xiang, Xi-Long-Shan, on trunk, 22°39'44"N, 102°47'59"E, 2200 m, *W.Z. Ma 15-7033* (CAS, KUN); *ibid.*, on trunk, 2170 m, *W.Z. Ma 15-7043* (CAS, KUN); Ma-An-Di Xiang, Wu-Tai-Shan, on trunk, 22°46'0"N, 103°28'58"E, 1480 m, *W.Z. Ma 17-8950* (CAS, KUN); *ibid.*, on trunk, 1580 m, *W.Z. Ma 17-8968* (CAS, KUN); Jin-He Zhen, He-Tou-Da-Shan, on hardwood trunk, 22°50'14"N, 103°16'7"E, 2040 m, *Shevock & W.Z. Ma 50793* (CAS, KUN). Ping-Bian Co., Yu-Ping Zhen, Da-Wei-Shan, Shui-Wei-Cheng Scenic Spot, on trunk about 25 m above ground, 22°54'52"N, 103°42'7"E, 2040 m, *W.Z. Ma 14-5431* (CAS, KUN); Puer Region, Jing-Dong Co., Tai-Zhong Xiang, Ai-Lao-Shan, on trunk, 2490 m, *T. Shen K062* (KUN).

**Key to the species of *Chiastocaulon* in China**

1. Dry leaves appressed to stem, marginal teeth none or not easily seen without hand-lens.....2
1. Dry leaves more or less spreading, marginal teeth large and easily noticeable ....3
  2. Plant dark in color, bluish green to deep brown, leaf border always entire, smooth. Trigones near leaf margin large. Perianth cylindrical ..... *C. braunianum*
  2. Plant lighter in color, yellowish to light green, leaf border with 1-6 small and weakly developed blunt teeth. Trigones near the leaf margin tiny or hardly noticeable. Perianth campanulate ..... *C. mayebarae*
3. Plant small, slender, 1-2 mm wide when hydrated, stem 0.2 mm thick. Cells in the middle of lamina thin-walled, trigones tiny or none ..... *C. oppositum*
3. Plant robust, more than 2.5 mm wide when hydrated, stem 0.3-0.4 mm thick. Cells in the middle of lamina thick-walled, trigones large and conspicuous ....4
  4. Leaves discontinuous or nearly continuous. Middle leaves wider than long, marginal large teeth 5-8, tiny teeth 1-6, mostly borne on the upper 2/3 of the leaf border. Female bracts 3-5 lobed with teeth developed. Perianth long cylindrical, 3-5 mm long..... *C. fimbriatum*
  4. Leaves overlapping each other. Middle leaves longer than wide, marginal large teeth 5-6, tiny teeth many, teeth borne from postical margin to the distal half of antical margin. Female bracts with marginal teeth evenly distributed, without sub-lobes. Perianth cylindrical, 2-3 mm long..... *C. theriotanum*

**DISCUSSION**

In China, four species of *Chiastocaulon* were currently documented in China (Fu, 2008; Gao & Wu 2010, both as *Plagiochilion*), they are *C. braunianum* (Nees) S.D.F. Patzak, M.A.M. Renner, Schäf.-Verw. & Heinrichs, *C. oppositum* (Reinw., Blume & Nees) S.D.F. Patzak, M.A.M. Renner, Schäf.-Verw. & Heinrichs, *C. mayebarae* (S.Hatt.) S.D.F. Patzak, M.A.M. Renner, Schäf.-Verw. & Heinrichs and *C. theriotanum* (Steph.) S.D.F. Patzak, M.A.M. Renner, Schäf.-Verw. & Heinrichs. There is no recent revision of this group in China, probably due to insufficient specimens available for study.

*Chiastocaulon fimbriatum* could be easily mis-identified with *C. oppositum* and *C. theriotanum*. Besides the similarity between *C. fimbriatum* and *C. oppositum* discussed by Inoue (1964) in the thickness of cell walls at the apex and middle,

*C. fimbriatum* is a robust plant usually 3-5 mm wide while only 1-2 mm wide in *C. oppositum*. *Chiastocaulon theriotanum* also shows some morphological resemblance to *C. fimbriatum* in size, but all leaves of *C. theriotanum* are elliptic while the middle leaves of *C. fimbriatum* are more broadly ovate and almost always wider than long.

### Morphological characters consistent with earlier descriptions

**Cell size** – When compared to the description of Inoue (1964, as *Plagiocihlion fimbriatum*), measurements of cells across the lamina of the middle leaves matched precisely. A number of randomly selected cells averaged  $23.7 \times 19.1 \mu\text{m}$  and  $32.6 \times 22.9 \mu\text{m}$  in the apical and middle respectively, both numbers agreeing with the measurements from the type specimen,  $23 \times 17\text{-}20 \mu\text{m}$  and  $32\text{-}40 \times 20\text{-}27 \mu\text{m}$ .

**Leaf size** – The average size of leaves among Yunnan material is  $2.18 \times 2.26 \text{ mm}$ , and are up to  $2.57 \times 2.88 \text{ mm}$  in size, as compared to  $2.0 \times 2.4 \text{ mm}$  described by Inoue (1964). However, when Paton's method (Paton, 1999) was applied to the leaf illustration in Inoue (1964), it is measured as  $2.65 \times 2.72 \text{ mm}$ , which suggests the Yunnan plant matches well with his description.

**Marginal teeth** – The number of large teeth rarely exceeds 8, which is more or less consistent with the protologue. However, the number of total number of teeth along leaf border ranged from 7-14, which we found to be a very variable character that should be treated with caution.

**Trigones** – The distinct trigones near the leaf border were observed in all specimens, and they become larger downwards where they are maximized in size in the basal area (Figs 9-11). Despite the noticeable trigones near the leaf margin which serve as the most reliable character in this species, they are very hard to find on young leaves.

### Variable characters

**Leaf shape** – Leaves from the newly discovered Yunnan populations show a wide range of variation in terms of sizes and shapes, from broadly ovate to oblong and triangularly elliptical. Typically, the biggest-sized of leaf occurs 2-3 pairs below the terminal, and the size of leaves decreases downward. A set of leaves arranged by size shows a great deal of variation (Figs 2-8), which could be quantified by the value changes in the width to length (W/L) ratio. The W/L ratio not only varies dramatically among populations, but also occurs within the same specimen packet, and even among leaves from a single branch. It should be noted that since the type specimen only consists of 2 stems, Inoue may stripped off a few mature leaves so as to avoid damaging the scanty type material. In contrast, the average size of Yunnanese *Chiastocaulon fimbriatum* were slightly smaller than the values in Inoue's description. This is because apart from the middle leaves, upper leaves, which generally appear elliptical and much smaller, were also measured.

According to the analysis of variance on leaves from different positions on the stems, cell widths from the lower parts of the stem are significantly smaller than those sampled above ( $p = 0.042$ ), while no statistical differences observed between the upper and middle cell widths ( $p = 0.357$ ). If leaves from the middle of a branch with a mature capsule were to be considered as fully developed (since they generally appear larger in size), then the upper leaves just below the bracts must also be

assumed to be mature leaves, because cells from both parts have statistically similar widths. Thus the inclusion of upper leaves into the calculation and description is justifiable, although they are very different in size and shape from the middle leaves.

**Bracts** – The orbicular and non-decurrent leaves, 3-5 lobed female bracts, and cells with thin walls and big trigones are characteristic of *C. fimbriatum* as summarized by Inoue (1964). The Yunnan population differs by a distinct horizontal-spreading arrangement on the ventral side. Although being variable in bract shape and size, small teeth scattered on sub-lobes on the ventral side of the bract serve as a stable character for this species (Figs 16-19).

**Perianth** – The cylindrical and large size of the perianth is among the most striking features of *Chiastocaulon fimbriatum*, and quite stable too. But immature perianths could lead to mis-identification as perianths appear so different through varying stages of development (Figs 20-24).

**Decurrence of leaves** – The non-decurrent leaves were thought to be the most diagnostic feature of *Chiastocaulon fimbriatum* by Inoue (1964). According to our observations on numerous leaves (Figs 3, 5, 8), a slight decurrence on the dorsal side is not difficult to see. This decurrence can even be seen in Inoue's (1964) illustration. Young leaves are more likely to retain this feature than old ones because the latter are usually more fragile.

**Capsule & spores** – Capsules in species of Plagiochilaceae are not commonly discovered (So, 2001), therefore the newly found capsule of *Chiastocaulon fimbriatum* may not be of much significance in improving a key to the genus. Yet, it may have the largest spores among all members within the genus with an average diameter of 34.1  $\mu\text{m}$  (ranging from 28.3 to 39.4  $\mu\text{m}$ ), although they may be slightly smaller than some of the biggest spores in *C. braunianum* reported by Majumdar *et al.*, (2013) that ranged from 22.0 to 38.4  $\mu\text{m}$ . The diameter of spores in other *Chiastocaulon* species seldom exceeds 23  $\mu\text{m}$  (Inoue 1964; Kazuhrii 2015).

Herbarium records show that *Chiastocaulon mayebarae* is the best represented species within the genus in Yunnan; however, we expect *C. fimbriatum* to be more widespread than currently known. Because *C. fimbriatum* was typically found on tree trunks at the forest edge, or branches up higher in the middle/outer canopy, they have an advantage over other shade-dwellers when colonizing semi-exposed substrates. In other words, *C. fimbriatum* is possibly widespread since it shows disturbance-tolerant characters.

Being a relatively robust plant and with a relatively broader distribution potential in Yunnan. A possible explanation why *Chiastocaulon fimbriatum* has remained so rarely documented may be due to its close resemblance to some species in *Plagiochila*, a large genus being notoriously difficult in species recognition. This has either led to their exemption from being collected in the field or being mislabeled as "*Plagiochila* sp." or eventually ended up in herbarium backlogs awaiting determination to species level.

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