

The Lichen Genus *Sticta* in South Korea

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Abstract *Sticta* (Schreber.) Ach. is one of the common lichen genera in tropical and subtropical regions, but not in the Korean Peninsula. For almost two decades, no detailed taxonomic or revisionary study has been done on this genus. This study was based on the specimens deposited in the lichen herbarium at the Korean Lichen Research Institute, and the samples were identified on the basis of recent literature. In this revisionary study, a total of eight species of *Sticta*, including a newly recorded one are documented. These species include *Sticta fuliginosa* (Dicks.) Ach., *Sticta gracilis* (Müll. Arg.) Zahlbr., *Sticta limbata* (Sm.) Ach., *Sticta nylanderiana* Zahlbr., *Sticta sublimbata* (J. Steiner) Swinscow & Krog, *Sticta weigeli* (Ach.) Vain., *Sticta wrightii* Tuck., and *Sticta yatabeana* Müll. Arg. Detailed descriptions of *S. nylanderiana*, *S. sublimbata*, *S. weigeli*, and *S. yatabeana* with their morphological, anatomical, and chemical characteristics are provided. A key description of all known *Sticta* species of the Korean Peninsula is also presented.

Keywords Foliose, Lobariaceae, *Nostoc*, South Korea, *Sticta*

The genus *Sticta* (Schreber.) Ach. belongs to family Lobariaceae [1]. It is one of the commonest foliose lichens, characterized by the presence of cyphellae on their lower surface [2]. This genus is superficially similar to two genera, namely, *Lobaria* (Schreber) Hoffm. and *Pseudocyphellaria* Vain., but is distinguished by the presence of cyphellae [3, 4]. In addition to the presence of cyphellae, this genus is mainly characterized by a lobate thallus, spreading or stalked with an erect monophyllous to polyphyllous frond. The lobes are irregularly branched, rounded to imbricate to variously incised, often lacerate-notched, with or without isidia, soredia, or phyllidia [5].

Although the genus *Sticta* is primarily tropical to subtropical in distribution, its species exist as far north as Norway [6] and as far south as the southern tip of South America [7]. Although Moncada *et al.* [8] stated that the actual number of existing species is unknown, 190 species have been identified thus far [9]. Monographic revisionary

work on the genus *Sticta* has been carried out in different regions in the world [4, 7, 8, 10-13].

In a recent monographic study, Takahashi [14] has reported 11 *Sticta* species from the South Asian region including China and Japan. In South Korea, no comprehensive work on *Sticta* has been done, and very little information is available on most of the known South Korean species. Park [15] has made a key description of only two *Sticta* species, namely, *S. nylanderiana* and *S. yatabeana*. To date, seven species have been reported from South Korea [16]. These include *Sticta fuliginosa* (Dicks.) Ach., *Sticta gracilis* (Müll. Arg.) Zahlbr., *Sticta limbata* (Sm.) Ach., *Sticta nylanderiana* Zahlbr., *Sticta weigeli* (Ach.) Vain., *Sticta wrightii* Tuck., and *Sticta yatabeana* Müll. Arg. Therefore, our aim in this study was to identify the presence of other *Sticta* species in the South Korean lichen species collection, which is deposited in the Korean Lichen Research Institute (KoLRI), by using taxonomic, chemical, and molecular analyses.

MATERIALS AND METHODS

The study was based on the specimens deposited in the KoLRI. The lichen samples were identified using stereo and light microscopes: a dissecting microscope (SMZ645; Nikon, Tokyo, Japan) was used for examining the thallus morphology, reproductive structures, color, size, and shape, and a compound microscope (ZEISS Scope, A1; Zeiss, Oberkochen, Germany) was used for studying the anatomy of thalli. Spot test reactions were carried out on thalli under a compound microscope.

Chemicals were extracted in analytical-grade acetone using 0.1 g of the thallus in a 1-mL Eppendorf tube. Then,

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thin layer chromatography (TLC) was done using a glass plate coated with TLC Silica gel 60, in solvent system A (toluene : dioxin : acetic acid = 180 : 45 : 5) [17]. All examined localities of specimens were mapped using open source GIS software Quantum GIS 1.7.0 (<http://www.qgis.org>). Voucher specimens have been deposited in the herbarium of the Lichen & Allied Bio-resource Centre at the KoLRI, Sunchon National University, South Korea.

Total DNA was extracted directly from the thalli of the selected specimens (KoLRI000321-KF730790, KoLRI000449-KF730791, KoLRI000460-KF840681, KoLRI000468-KF730792, and KoLRI008817-KF730793) [18] and was purified using the DNeasy Plant Mini Kit (Qiagen, Hilden, Germany). The nuclear ribosomal RNA gene region, including the internal transcribed spacers (ITSs) 1 and 2 and the 5.8S subunit (ITS), was amplified using the primers ITS1F [19] and LR5 [20]. Amplification was performed on a TaKaRa JP/TP600 PCR machine (TaKaRa Bio Inc., Otsu, Japan). PCR products were then sent to the sequencing facilities of GenoTech Corp. (Seoul, Korea) for cleaning and sequencing.

Alignment was done using BioEdit software [21]. Seventeen additional ITS sequences of *Sticta* were recovered from GenBank. Ambiguous regions were delimited [22] and excluded from the alignment. Four species—*Pseudocyphellaria neglecta* (Müll. Arg.) H. Magn., *P. crocata* (L.) Vain., *Lobaria retigera* (Bory) Trevis., and *Cladonia macilenta* Hoffm)—were selected as out-groups on the basis of BLAST comparison in GenBank. Phylogenetic relationships between taxa were investigated using MEGA 5.2 software [23]. The data set was analyzed using the neighbor joining (NJ) method, and support values were obtained using a bootstrap analysis of 1,000 pseudoreplicates.

RESULTS AND DISCUSSION

Phylogenetic analysis. The ITS matrix included 382 characters, 132 of which were variable. The NJ tree (Fig. 1) shows that all *S. nylanderiana* and *S. yatabeana* are clustered within the well-supported clade (bootstrap, 84%). All species within this clade do not contain any isidia or soredia on their thallus. *S. yatabeana* is closely related to *S. nylanderiana* (bootstrap, 100%) in that it does not have gyrophoric acid in the medulla. *S. weigelii* is closely related to *S. fuliginosa* (bootstrap, 92%). Both these species are characterized by the presence of esorediate isidia. *S. weigelii* from Japan (AB245124) and Finland (AF524905) are clustered in a different clade, with a 49% bootstrap value. Because some specimens were a little old, it was difficult to extract the total DNA from the thallus. Therefore, only a small number of thalli were used to extract DNA in this study.

Key to the South Korean species of *Sticta*

- 1. Thallus stalked, isidiosoredia absent, thallus glabrous at lobe apices *S. gracilis*
- 1a. Thallus non-stalked 2

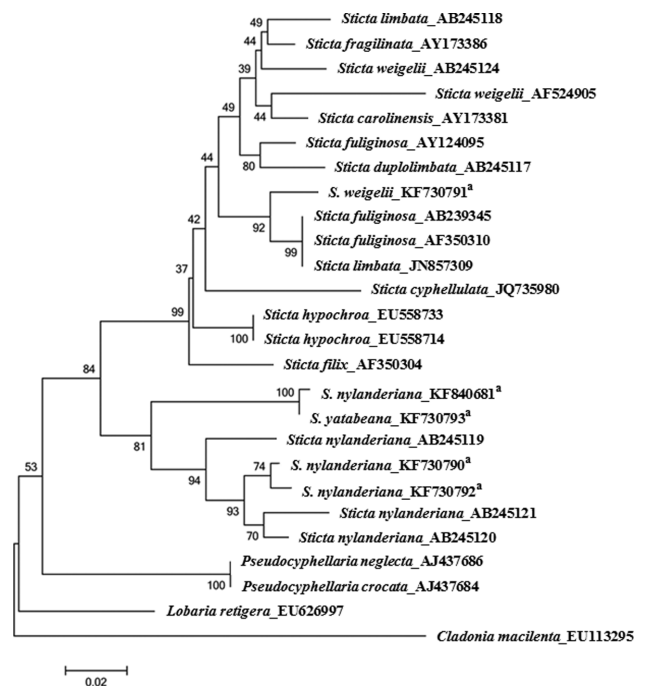


Fig. 1. Phylogenetic relationships between species of *Sticta* for which molecular data are currently available in the public database GenBank. This phylogeny was obtained using the marker internal transcribed spacer and an neighbor joining criterion using software MEGA5. ^aSpecimens selected for the current study.

- 2. Lobes isidiate or sorediate 3
- 2a. Lobes without isidia and soredia 6
- 3. Sorediate, monophyllous *S. limbata*
- 3a. Thallus isidiate 4
- 4. Isidia sorediate, medulla K- *S. sublimbata*
- 4a. Isidia esorediate 5
- 5. Thallus monophyllous, isidia granular to coralloid, medulla K- *S. fuliginosa*
- 5a. Thallus polyphyllous, isidia cylindrical to coralloid, medulla K+ (yellow) *S. weigelii*
- 6. Upper surface glabrous, medulla KC+ (rose-red), gyrophoric acid present *S. nylanderiana*
- 6a. Upper surface hirsute, medulla KC-, gyrophoric acid absent 7
- 7. Cyphellae rounded, shallowly excavate *S. wrightii*
- 7a. Cyphellae irregular, scarcely excavate *S. yatabeana*

Species description.

Sticta nylanderiana Zahlbr., Zahlbruckner's Cat. Lich. Univ. 3: 356 (1925).

Thallus lobate, green to grayish-green, non-stipitate, polyphyllous frond, loosely attached, 5~20 cm wide. Lobes irregularly branching, 0.5 to 1.5 cm wide, rounded to truncate apex, without isidia, soredia, or phyllidia. Upper surface smooth, somewhat glossy, without maculae. Medulla white, 60~200 μm thick, loosely interwoven. Photobiont layer 15~70 μm thick with green algae. Lower surface pale

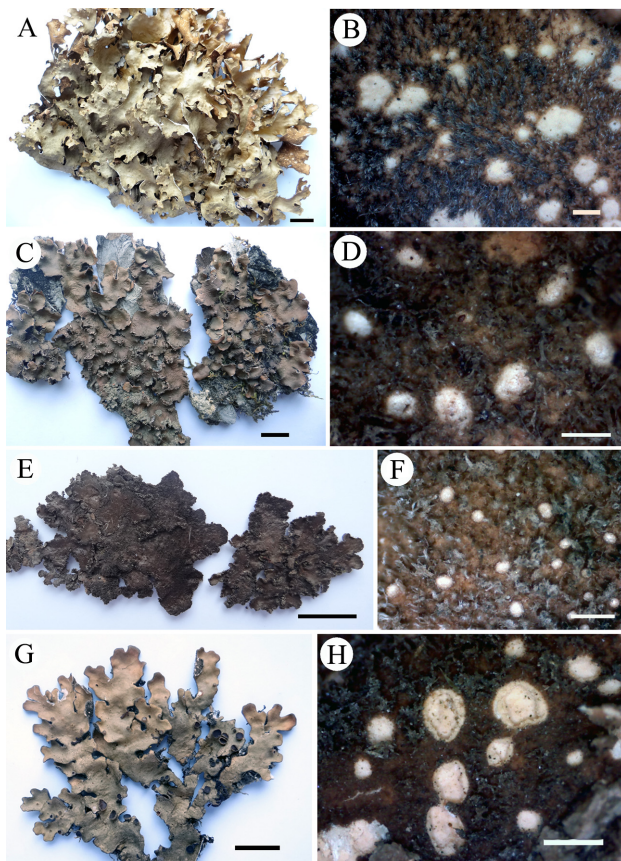


Fig. 2. *Sticta* species. A, *S. nylanderiana* (J. S. Hur, 030664); B, *S. nylanderiana*—lower surface with cyphellae; C, *S. sublimbata* (J. S. Hur, 070686); D, *S. sublimbata*—lower surface with cyphellae; E, *S. weigeli* (J. S. Hur, 030646); F, *S. weigeli*—lower surface with cyphellae; G, *S. yatabeana* (J. S. Hur, 040504); H, *S. yatabeana*—lower surface with cyphellae (scale bars: A, C, E, G = 1 cm, B, D, H = 1 mm, F = 0.5 mm).

to dark brown, densely tomentose in the middle, glabrous at the margins. Cyphellae round, shallowly excavate with non-prominent margins. Apothecia somewhat rare, 0.6–3 mm in diameter; disc brown, matt, epruinose; margins entire; proper exciple well-developed, smooth to dentate, without photobiont cells. Ascospores fusiform-ellipsoidal to linear, $40\text{--}60 \times 4\text{--}6 \mu\text{m}$, 5–7 septate, colorless. Pycnidia laminal, crowded on thallus margin. Conidia $3.0\text{--}5.0 \times 1\text{--}1.5 \mu\text{m}$, bacilliform (Fig. 2A).

Chemistry: Cortex K–, C–, KC–, P–; medulla K–, C–, KC+ (rose to red), P–. TLC: gyrophoric acid, congyrophoric acid, pseudocyphellarin A, unknown 1 to 5 (Fig. 3).

Remarks: *S. nylanderiana* is characterized by the presence of glabrous, shiny upper surface without soredia, isidia, or lobules, rounded cyphellae without non-prominent margins, and gyrophoric acid in the medulla. This species is similar to *S. yatabeana* in external appearance, but the latter species differs in having small cortical hairs on the upper surface and not having gyrophoric acid in the medulla.

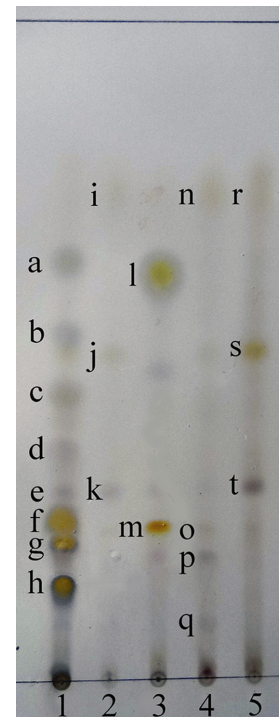


Fig. 3. Thin layer chromatography profile of *Sticta* species in solvent system A. 1, *S. nylanderiana* with pseudocyphellarin A (a), congyrophoric acid (g), gyrophoric acid (h), unknown 1 (b), unknown 2 (c), unknown 3 (d), unknown 4 (e), and unknown 5 (f); 2, *S. sublimbata* with unknown 1 (i), unknown 2 (j), and unknown 3 (k); 3, Control [*Lethariella cladonioides* (Nyl.) Krog] with atranorin (l) and norstictic acid (m); 4, *S. weigeli* with unknown 1 (n), unknown 2 (o), unknown 3 (p), and unknown 4 (q); 5, *S. yatabeana* with unknown 1 (r), unknown 2 (s), and unknown 3 (t).

Selected specimens examined: Mt. Duta, on bark, $37^{\circ}25'49.8''$ N, $128^{\circ}58'40.5''$ E, elev. 1,377 m, 11 May 2008, J. S. Hur, 080175; Mt. Taebaek, on bark, $37^{\circ}06'08.2''$ N, $128^{\circ}55'53.1''$ E, elev. 1,308 m, 20 Aug 2003, J. S. Hur, 030664; on bark, $37^{\circ}06'11.4''$ N, $128^{\circ}55'55.8''$ E, elev. 1,254 m, 20 Aug 2003, J. S. Hur, 030658.

Ecology and distribution: *S. nylanderiana* is a common species on barks at higher elevations around 900 m to 1,700 m in South Korea (Fig. 4). This species was reported for the first time in South Korea by Kim [24]. According to Takahashi [14], this species is more widely distributed in Japan. This species has also been found in other Asian and Southeast Asian countries including Bhutan, China, Nepal, Thailand, and Tibet [10, 14, 25].

Sticta sublimbata (J. Steiner) Swinscow & Krog, in Galloway, N. Z. J. Bot. 21: 198 (1983).

Thallus lobate, green to bluish-gray, non-stipitate, polyphyllous frond, loosely attached, 5–8 cm wide. Lobes irregularly branching, 0.5 to 1.5 cm wide, rounded apex, with sorediate isidia at the margins. Isidia minute, granular-coralloid, dark gray-brown, soon eroding and becoming sorediate.

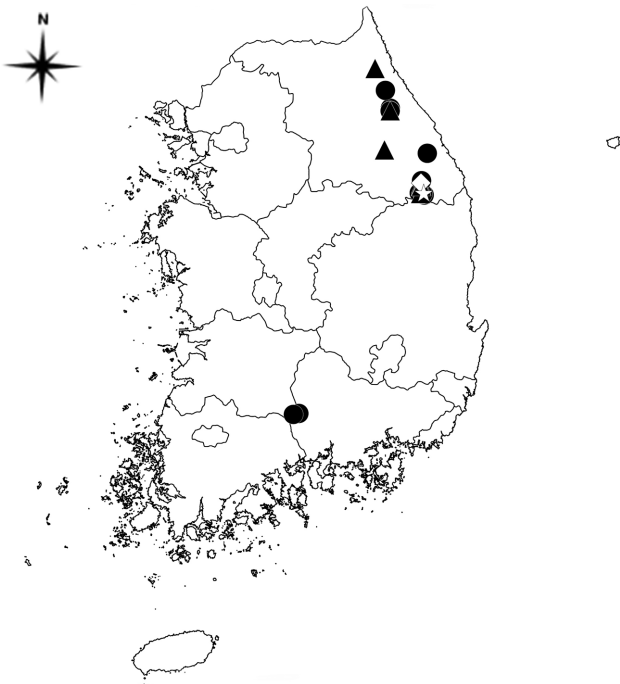


Fig. 4. Distribution of *Sticta* species in South Korea: *S. nylanderiana* (●), *S. sublimbata* (◇), *S. weigelii* (☆), and *S. yatabeana* (▲).

Upper surface smooth, shallowly wrinkled, maculate. Medulla white, 20–80 μm thick, loosely interwoven. Photobiont layer 25–60 μm thick with cyanobacteria *Nostoc*. Lower surface dark brown to black, densely tomentose. Cyphellae round, shallowly excavate without prominent margins. Apothecia and pycnidia not seen (Fig. 2C).

Chemistry: Cortex K–, C–, KC–, P–; medulla K–, C–, KC–, P–. TLC: unknown 1 to 3 (Fig. 3).

Remarks: This species is newly found in South Korea during this study. *S. sublimbata* is characterized by the presence of a non-stipitate thallus and sorediate isidia along the margin. According to Takahashi [14], the appearance of isidia varies with the development stage of the thallus. This species closely resembles *S. weigelii*, but the latter species has marginal coralloid isidia. This species is closely related to *S. limbata* (Sm.) Ach. by external appearance, but the latter species has a sorediate instead of a sorediate-isidiate margin [14].

Selected specimens examined: Mt. Hambaek, on bark, 37°11'27.3" N, 128°54'52.9" E, elev. 1,445 m, 19 Jun 2007, J. S. Hur, 070686.

Ecology and distribution: This species is very rare in South Korea. Only a single specimen was found on a bark at higher elevation (Fig. 4). This species has also been found in Japan [14], East Africa [26], South America [7], New Zealand [11], and Australia [5].

Sticta weigelii (Ach.) Vain., Acta Soc. Fauna Flora Fenn. 7: 189 (1890).

Thallus lobate, bluish-gray to grayish brown, non-stipitate,

polyphyllous frond, deeply dissected, loosely attached, 5–7 cm wide. Lobes irregularly branching, 0.5 to 1.5 cm wide, rounded apex, with isidia at the sinuous margins, esorediate, etomentose. Isidia minute, cylindrical to coralloid, dark gray-brown, densely crowded at the margins. Upper surface glabrous, flat to concave, maculate. Medulla white, 80–120 μm thick, densely interwoven. Photobiont layer 60–100 μm thick with cyanobacteria *Nostoc*. Lower surface dark brown to black, densely tomentose. Cyphellae round, deeply excavate with prominent margins. Apothecia and pycnidia not seen (Fig. 2E).

Chemistry: Cortex K–, C–, KC–, P–; medulla K+ (yellow), C–, KC–, P–. TLC: unknown 1 to 4 (Fig. 3).

Remarks: *S. weigelii* is characterized by the presence of a non-stipitate thallus and marginal to submarginal coralloid isidia. Further, the color reaction with KOH is specific in this species. According to Takahashi [14], the well-developed thallus of this species contains coralloid isidia not only at the margins but also on the lamina. This species very closely resembles *S. sublimbata* owing to the presence of marginal coralloid isidia, but the latter species is distinguished by the presence of sorediate isidia and absence of chemicals in the medulla.

Selected specimens examined: Mt. Taebaek, on rock, 37°06'16.4" N, 128°56'31.8" E, elev. 1,011 m, 20 Aug 2003, J. S. Hur, 030646.

Ecology and distribution: This species is somewhat rare in South Korea. Only a single specimen was found on a rock at higher elevation (Fig. 4). This species widely distributed in tropical and southern-temperate regions [5]. In East Asia, this species has been found in Japan [14] and China [10, 25].

Sticta yatabeana Müll. Arg., Flora 74: 111 (1891).

Thallus lobate, green to grayish-green, non-stipitate, polyphyllous frond, loosely attached, 5–10 cm wide. Lobes irregularly branching, 0.5 to 1.5 cm wide, rounded to truncate apex, without isidia, soredia, or phyllidia. Upper surface smooth, somewhat glossy, maculate, tomentose. Medulla white, 150–200 μm thick, loosely interwoven. Photobiont layer 15–30 μm thick with green algae. Lower surface pale to dark brown, densely tomentose at the center, glabrous at the margins. Cyphellae irregular, scarcely collapsed with non-prominent margins. Apothecia stalked, 0.8–5 mm in diameter; disc brown, matt, epruinose; margins entire; proper exciple well-developed, smooth to dentate, with photobiont cells. Ascospores fusiform-ellipsoidal to linear, 40–100 \times 3–4 μm , 3–7 septate, colorless. Pycnidia laminal, crowded on thallus margin. Conidia 2.0–6.0 \times 1–1.5 μm , bacilliform (Fig. 2G).

Chemistry: Cortex K–, C–, KC–, P–; medulla K–, C–, KC–, P–; TLC: unknown 1 to 3 (Fig. 3).

Remarks: *Sticta yatabeana* is characterized by a hirsute upper surface without soredia, isidia, or lobules; irregular cyphellae without non-prominent margins; stalked apothecia with thalloid exciple; and the absence of gyrophoric acid in

the medulla. This species is similar to *S. nylanderiana* in external appearance, but the latter species differs in having a glabrous upper surface and the presence of gyrophoric acid in the medulla.

Selected specimens examined: Mt. Odae, on bark, 37°46'21.6" N, 128°36'04.9" E, elev. 1,460 m, 8 May 2004, J. S. Hur, 040504; Mt. Taebaek, on bark, 37°06'23.1" N, 128°54'37.5" E, elev. 1,290 m, 9 Apr 2004, J. S. Hur, 040182; Mt. Gariwang, on bark, 37°27'26.1"N, 128°32'44.6"E, elev. 1,297 m, 10 May 2008, J. S. Hur, 080096.

Ecology and distribution: *S. yatabeana* is a common species on barks at higher elevations of about more than 1,200 m in South Korea (Fig. 4). This species was reported for the first time in South Korea by Park [15]. This species has only been found in Japan [14].

Species excluded in this study. According to the literature, lichen species *S. fuliginosa* (Dicks.) Ach., *S. gracilis* (Müll. Arg.) Zahlbr., *S. limbata* (Sm.) Ach., and *Sticta wrightii* Tuck. have previously been reported in South Korea. However, the specimens of those species are untraceable, and therefore, the descriptions are based on the previous literatures.

Sticta fuliginosa (Dicks.) Ach., Method. Lich.: 280 (1803). This species is characterized by the presence of umbilicate monophyllous thallus, granular to coralloid isidia on upper surface, and round cyphellae with prominent margins [14]. According to Galloway [27], *S. limbata* (Sm.) Ach. has similar morphology with *S. fuliginosa*, but it differs by the presence of somewhat eroded soralia on margins and upper surface. The presence of this species in South Korea was reported for the first time by Kim [24]. This is a cosmopolitan species, especially common in temperate areas [5, 10, 14, 27].

Sticta gracilis (Müll. Arg.) Zahlbr., Zahlbruckner's Cat. Lich. Univ. 3: 386 (1925).

This species is characterized by the presence of a stipitate thallus without isidia and soredia, hirsute near the stalk and central stalk. This species closely resembles *Sticta duplolimkata* (Hue) Vain. by the presence of stalked thallus, but is distinguished by the absence of marginal cilia and isidia [14]. The presence of this species in South Korea was first reported by Park [28]. This species has also been found in New Caledonia, China, Japan [10, 14], and Tibet [25].

Sticta limbata (Sm.) Ach., Method. Lich.: 280 (1803).

This species is characterized by the presence of a monophyllous thallus with umbilicus on the central part of the lower cortex, marginal to submarginal soredia, branched tomentum, and deeply excavate cyphellae with prominent margins. This species is closely related to *Sticta sublimkata* in having marginal soredia, but the latter is distinguished by the presence of a polyphyllous thallus and sorediate

isidia [5, 14]. The presence of this species in South Korea was first reported by Park [29]. This species is widely distributed in the Northern Hemisphere, East Africa, South America, Australia, and New Zealand. In the East Asian region, this species has been found in China [10, 25] and Japan [14].

Sticta wrightii Tuck., Am. J. Sci. Arts, Ser. 2 28: 204 (1859).

According to Takahashi [14], two morphotypes of *S. wrightii* exist. The chloromorph is characterized by the presence of glabrous foliose thallus with green algal photobiont, rounded to irregular cyphellae with non-prominent margins, and the absence of gyrophoric acid in the medulla. The cyanomorph of this species is characterized by the presence of a dendriscocauloid thallus with articulately ridged main branches and terete secondary branches, which are densely covered with tomentum. The presence of this species in South Korea was first reported by Kim [24]. The morphotype concept was not developed during that time, but the reported species would have belonged to the chloromorph type. This species has been found in other Asian countries including China [25], Japan [14], and Mongolia [30].

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