Diversity of the genus *Lyonia* Nuttall, 1818 (Ericaceae: Vaccinioideae) in the Eastern Himalayas, India

Subhasis Panda

Summary

The genus *Lyonia* Nuttall, a superior-ovaried member of the family Ericaceae Juss., consists of 35 species globally, with only 3 species reported from India. Members of the genus are distributed in tropical Asia and America. The genus is well-known to plant systematists due to its recent evolutionary significance, uniqueness in flowering-phenology, woody habit and increasing ethnomedicinal potentialities for herbal drug discovery. As a result of extensive fieldwork and herbarium consultation in Indian Herbaria (CAL, BSIS, ASSAM, BSHC, DD, APFH, ARUN, Lloyd Botanical Garden Herbarium) and abroad (K, BM, E, P: cibachrome images!), a total of three species are reported from the Indian Eastern Himalayas, viz., *Lyonia ovalifolia* (Wall.) Drude, *L. villosa* (Wall. ex C. B. Clarke) Hand.-Mazz. and *L. macrocalyx* (J. Anthony) Airy Shaw. The present work summarizes detailed field and herbarium-based exomorphological diversity, laboratory-based microscopical works viz., leaf stomata, leaf vein islets and trichomes, ethnobotanical-ITKs, IUCN Status, regional as well as global distribution pattern and unusual flowering in *L. ovalifolia*. The purpose of the present study is to evaluate the contribution of leaf stomata, leaf vein islets and trichomes along with field-based exomorphological unique data to stabilize species circumscription in the genus *Lyonia* Nutt.

Key words: Ericaceae, *Lyonia*, diversity, Eastern Himalaya, India

Introduction

The genus *Lyonia* Nuttall, a superior-ovaried member of the family Ericaceae Juss., was first described and named by Nuttall in 1818 (Quattrocchi 2000) who described the genus based on the species *L. ferruginea* (Walter) Nutt. He named the genus "*Lyonia*" in memory of the Scottish botanist John Lyon (1765 – 1814) who was a gardener in Philadelphia and W Pennsylvania and an indefatigable collector of North American plants (Quattrocchi 2000).
The genus is well-known to plant systematists due to its recent evolutionary significance, uniqueness in flowering-phenology, woody habit and increasing ethnomedicinal potentialities for herbal drug discovery. The genus consists of *c*. 35 species (Judd 1981, Stevens et al. in Kubitzki 2004), but Mabberley (2008) reported 36 species. Panda (2006, 2008), Panda & Sanjappa (2014) recognized 3 species, viz. *L. ovalifolia*, *L. villosa* and *L. macrocalyx* from India. Members of the genus are distributed in tropical Asia including India, as well as North America.
Detailed investigations of the genus *Lyonia* Nutt. were made by several workers like Judd (1981), Hara (1966), Hara in Hara et al. (1982), Kron et al. (2002), Stevens et al. in Kubitzki (2004), Ruizheng & Stevens in Wu et al. (2005), Panda (2006, 2008), and Panda & Sanjappa (2014). In India, the genus was studied in detail by Panda who revised the genus and described 3 species.

The present work embodies detailed field-based exomorphological diversity, ethnobotanical-ITKs, IUCN status, regional as well as global distribution and unusual flower in *L. ovalifolia*. This work also includes diversity in light microscopic studies of leaf stomata, leaf vein islets and trichomes in these three species of *Lyonia* Nutt. Scientists like Niedenzu (1890), Cox (1948), Hagerup (1953), Watson (1964, 1965), Lems (1964), Stevens (1970, 1971), Judd (1981) contributed a little or no works on leaf-stomata, leaf vein islets and leaf trichomes. Judd (1981) investigated a detailed account on *Lyonia* Nutt. excluding most of the Indian taxa.

The purpose of the present study is to evaluate the contribution of leaf stomata, leaf vein islets and trichomes along with field-based exomorphological unique data to stabilize species delimitation in the genus *Lyonia* Nutt.

Materials and Methods

The present work is the result of a critical study of literature in the library of the Central National Herbarium, Howrah (India), relevant references available in websites, consultations of herbaria (CAL, BSIS, ASSAM, BSHC, DD, Lloyd Botanical Garden Herbarium, Arunachal Pradesh Regional Centre, Botanical Survey of India, Itanagar, ARUN, Arunachal Pradesh Forest Herbarium, APFH), consultation of Cibachrome images of type and authentic specimens (K, BM, P, E) and extensive field survey (2002-2021) in different parts of Eastern Himalaya. This work also includes laboratory-based leaf-anatomical investigations among the said three species of *Lyonia* Nutt.

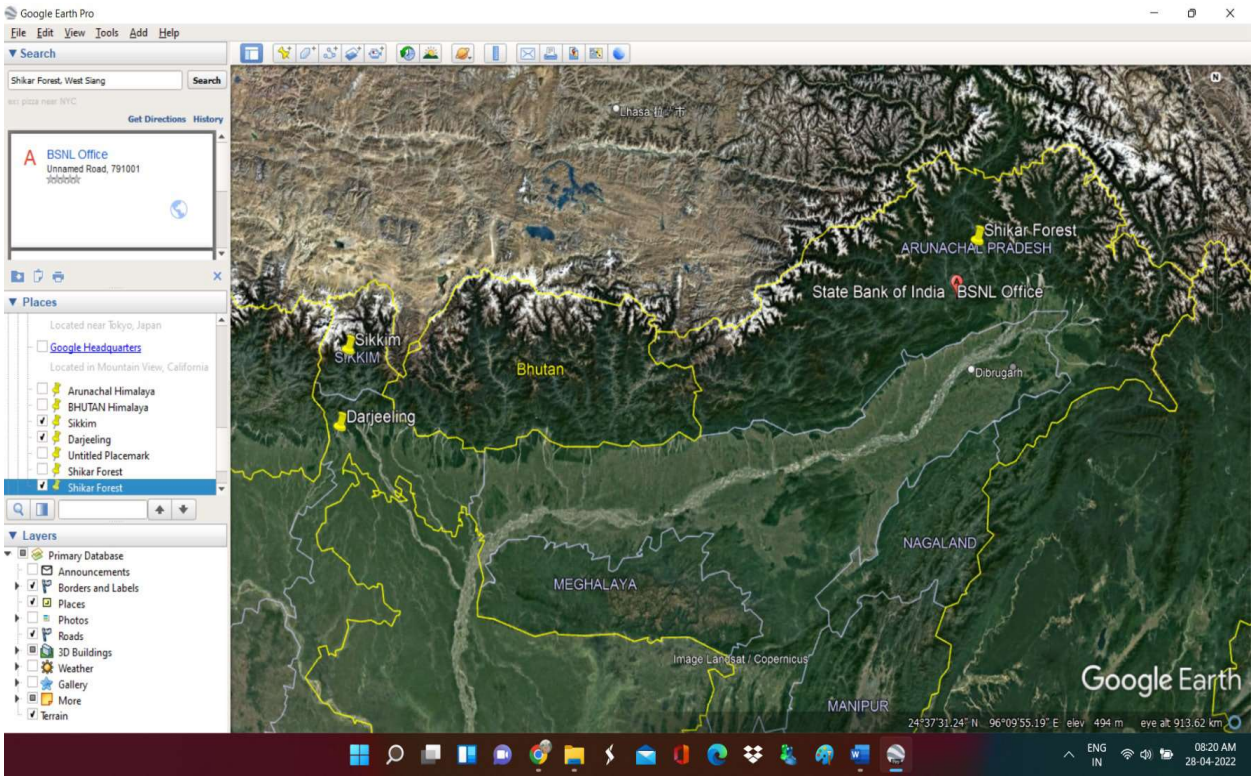
Morpho-taxonomical descriptions were made based on data from live plants except one species, *L. macrocalyx* which is based on herbarium material at APFH (to record the colour of flowers, fragrance and indumentum, the range of variations, their abundancy and rarity, presence of nectaries, associated plants, altitude, habitat and to assess the conservation status), as well as relevant taxonomic literature and herbarium data. For ethnomedicinal investigation, the first hand Indigenous Traditional Knowledge (ITK) or information were recorded during field visits at different localities in Eastern Himalaya through an oral interview of experienced and elderly tribal people, local medicinemen and field guides. Detailed information regarding local names(s), part(s) used, mode of administration or preparation and dosimetry are recorded in the field note book. Botanical identity is confirmed with herbarium consultations in Central National Herbarium (CAL). Important voucher specimens are deposited in Central National Herbarium (CAL), in the laboratory of Angiosperm Taxonomy & Biosystematics, Post Graduate Dept. of Botany, Barasat Govt College, in the laboratory of Angiosperm Taxonomy & Ecology, Botany Dept., Darjeeling Govt. College, Darjeeling and Maulana Azad College Herbarium (MAC), Kolkata. The work was carried out in Central National Herbarium (1999 – 2004), Botanical Survey of India and partly in the laboratory of Angiosperm Taxonomy & Biosystematics, Barasat Govt College and Darjeeling Govt College. For detailed study, floral parts of these plants were dissected and examined. The terminology for different external morphological characters were followed

mainly Lawrence (1951), Featherly (1954), Stearn (1983), Radford (1986) and Veldkamp in Vogel (1987). All measurements were given in metric system. Botanical identity is confirmed with herbarium consultations in Central National Herbarium (CAL). Authors of scientific names are abbreviated according to Brummitt & Powell (1992), Authors of Plant Names. Herbarium acronyms are followed according to Holmgren et al. (1990), Index Herbariorum, part 1 (ed.8). Anatomical materials used in this investigation were taken from the live materials collected from Darjeeling, Sikkim and Arunachal Himalayas which are deposited at Central National Herbarium (CAL), Barasat Govt College (BGC) and Darjeeling Govt. College (DGC) Herbaria. For *Lyonia macrocalyx*, one leaf sample was taken from duplicate herbarium material with permission from APFH.

Stomatal slide preparation: Small cubical pieces (c. 1 sq.cm) were excised from the base, middle and apical regions of the leaf blade. Several existing methods viz., 10% HNO_3-boiling for 10 minutes, 5% KOH overnight (12 – 24 hours) treatment without boiling and with boiling were done. Pieces were ringed in sterilized water until clear. After clearing, pieces were dehydrated in an ethanol series followed by staining with 10% safranin and mounted onto microscope slide in DPX (pieces of basal, middle and apical regions in one slide). The slide was examined under an Olympus (Tokyo, Japan) light microscope using 10X, 45X and 100X objectives and Camera Lucida drawings were made with the help of drawing prism. The descriptive terminology followed Metcalfe & Chalk (1950), Dilcher (1974), Stace (1965, 1989), Fahn (1997) and Carpenter (2005).

Methodology of leaf clearing for venation study: Entire mature leaves were immersed in 2.5% NaOH solution until clear (closed condition). In the present study, most of the leave were cleared after 6 days of NaOH treatment. Leaf samples were then washed in distilled water. After clearing, pieces were dehydrated in an ethanol series followed by staining with 1% safranin and mounted onto microscope slide in DPX. The descriptive terminology follows Hickey (1973) and Dilcher (1974).

Preparation of trichomes: trichomes were removed using a sharp razor blade and forceps from different parts viz., stem, branches, leaves, peduncles, rachis, pedicels, bract, bracteoles, corolla, calyx, stamens, ovary and fruits and their morphological nature were observed under Olympus light microscope (10X, 45X). The descriptive terminology follows Metcalfe & Chalk (1950); Stace (1965) and Fahn (1997).



Overview of the Study Area: The Indian Eastern Himalayas consist of three Himalayan States viz. West Bengal (Darjeeling and Kalimpong hills), Sikkim and Arunachal Pradesh. Geographically, they are located 27°06′N to 29°19′N and 88°01′E to 97°23′E. They harbour diverse elements of tropical, subtropical, temperate and alpine vegetation. Populations of three species of *Lyonia* Nutt. were mostly observed in temperate forests altitudes ranging from 1800 m to 4000 m. Although *L. ovalifolia* is occasionally observed in subtropical forest at an altitude of c. 900 m in Arunachal Himalaya. *L. ovalifolia* is commonly observed at altitudes ranging from 1500 m to 3500 m at different places in all three Himalayan states. Populations of *L. villosa* are mostly (Map 1) Indian Eastern Himalayan distribution of the genus *Lyonia* Nutt. (satellite imagery) observed in temperate forests to occasionally in subalpine forests at altitudes ranging from 2500 m to 4000 m. (Tonglu in Darjeeling, Lachen, Lachung, Lachung-Kupup, Lachung-Yumesangdong, Tsoka-Dzongri, Kyongnosla in Sikkim Himalaya and Mandala, Tawang, Naga jeje, Talle WLS, Malinja-hotspring in Arunachal Himalaya), while populations of *L. macrocalyx* are restricted only to Arunachal Himalaya and observed in broad-leaved temperate forest of West Siang (Shikar forest) at an altitude of c. 1800 m.

Sequence of presentation of data:

- Correct or accepted name of the species.
- Original citation and other references including Type information.
- Synonyms, if any.
- A reference to illustration or photo.
- Vernacular names along with languages in parenthesis where available.
- Brief field description.
- Natural Distribution (globally).
- Habitat.
- Flowering & Fruiting
- Specimens examination
- Ethnic Use.
- IUCN Status
- Leaf stomata (LM)
- Leaf vein islets (LM)
- trichomes (LM)

Source of Anatomical materials

Name of species	Collector and Number	Voucher information (Herbarium acronym)
Lyonia ovalifolia (Wall.) Drude	S. Panda 29947; S. Panda 71; S. Panda 717	India: Sikkim, on the bank of Karthak Lake, Yuksum, 1880 m, 18.05.2002, S. Panda 29947 (CAL); West Bengal, Darjeeling, Alubari Jungle busty, 2040 m, 02.06.2014, S. Panda 71 (DGC); Arunachal Pradesh, West Kameng district, Salari forest, 1900 m, 10.05.2010, S. Panda 717 (BGC).
2. *L. villosa* (Wall. ex C. B. Clarke) Hand.-Mazz.	S. Panda 30801; S. Panda 129	India: Sikkim, on the bank of Zema River, Lachen, 8000 ft, 31.05.2002, S. Panda 30801 (CAL); West Bengal, Darjeeling, Tonglu, 10000 ft, 21.05.2022, S. Panda 004 (MAC) & 12.05.2012, S. Panda 129 (DGC).
3. *L. macrocalyx* (J.Anthony) Airy Shaw	Haridasan 7283	India: Arunachal Pradesh, West Siang district, Shikar forest, 18.11.1994, Haridasan 7283 (APFH).

Results

Taxonomic Treatment

Lyonia Nuttall, gen. N. Amer. pl. 1: 266. 1818, nomen cons., not *Lyonia* Ell., Sketch Bot. S. Carolina and Georgia 1: 316. 1817 (= *Cynanchum* L., Asclepiadaceae), not *Lyonia* Raf., Med. Repos. New York 5 (ser. 2): 353. 1803 (= *Polygonella* Michaux, Polygonaceae); D. Don, Edinburgh N. Philos. J. 17: 158. 1834; G. Don, Gen. Syst. 3: 830. 1834; Endl., Gen. Pl.: 755. 1839; DC., Prodr. 7 (2): 599. 1839; Hook. f. in Benth. & Hook. f., Gen. Pl. 2: 587. 1876; Drude in Engl. & Prantl, Nat. Pflanzenfam. 4 (1): 44. 1889; Judd, J. Arnold Arbor. 62 (2): 129. 1981. Panda & Sanjappa in Sanjappa & Sastri, Fasc. Fl. India no. 25 (Ericaceae): 229. 2014. *Xolisma* Raf., Amer. Mon. Mag. Crit. Rev. 4: 193. 1819. Lectotype: *Lyonia ferruginea* (Walter) Nutt. (= *Andromeda ferruginea* Walter).

Distribution: INDIA: Himalayas (Jammu & Kashmir, Himachal Pradesh, Uttarakhand, Sikkim, West Bengal and Arunachal Pradesh) and NE India (Meghalaya, Nagaland, Manipur and Mizoram); PAKISTAN; NEPAL; BHUTAN; BANGLADESH; CHINA; TAIWAN; N MYANMAR; THAILAND; VIETNAM; CAMBODIA; MALESIA; JAPAN and N AMERICA (including Mexico, West Indies and Greater Antilles). The genus consists of *c*. 35 species (Judd 1981, Stevens et al. in Kubitzki 2004, Panda & Sanjappa in Sanjappa & Sastri 2014), but Mabberley (2008) reported 36 species. Panda (2006, 2008) and Panda & Sanjappa (2014) recognized 3 species, viz., *L. ovalifolia*, *L. villosa* and *L. macrocalyx* from India. Members of the genus are distributed in tropical Asia including India and North America.

Key to the Indian taxa

(Based on exomorphology, leaf stomata, leaf vein islets and trichomes)

Anther-filament junction spurred; Stomatal index (SI) less than 6.0; average stomatal dimensions more than 25.0µm×22.0µm (37.3µm×24.0µm); larger areoles ca. 312µm long; number of average areoles per 1 mm^2=10; average vein endings=30; stem and branches possess many-celled non-glandular trichomes of ca. 176 µm long ..... 1. *Lyonia ovalifolia*

Anther-filament junction spurless; Stomatal Index (SI) always more than 9.0; average stomatal dimensions always less than 25.0µm×22.0 µm; larger areoles 182-216 µm long; number of average areoles per 1 mm^2=5; average vein endings=13; stem and branches possess many-celled nonglandular trichomes varying from 280-322 µm long 2

Leaf apices strongly mucronate while leaf bases rounded to slightly cordate; epidermal cells usually irregular to occasionally polygonal-elongated; maximum length of stomata 21.8 µm; vein endings bulbous to rounded; larger areoles 216 µm long; number of average areoles per 1 mm^2 = 4; abaxial leaf surface shows many-celled non-glandular trichomes varying from 231-323 µm long.....2. *L. villosa*

Leaf apices acuminate, long acuminate to rarely mucronate, while leaf bases always cuneate; epidermal cells isodiametric-polygonal; maximum length of stomata 24 µm; vein endings elongated-bulbous, but never rounded; larger areoles 182 µm long; number of average areoles per 1 mm^2 = 6; abaxial leaf surface shows many-celled non-glandular trichomes of about 78.6 µm long.....3. *L. macrocalyx*

1. *Lyonia ovalifolia* (Wall.) Drude in Engl. & Prantl, Nat. Pflanzenfam. 4 (1): 44. 1889; Judd, J. Arnold Arbor. **62** (2): 149. 1981; Hara in Hara et al., Enum. Fl. Pl. Nepal **3**: 55. 1982; Rae in A. J. C. Grierson & D. G. Long, Fl. Bhutan **2** (1): 395. 1991; F. Ruizheng & P. F. Stevens in Wu et al., Fl. China **14**: 461. 2005; Panda & Sanjappa in Sanjappa & Sastry, Fasc. Fl. India **25**: 231. 2014. *Andromeda ovalifolia* Wall., Asiat. Res. 13: 391. 1820 and Numer. List: no. 763. 1829. *Pieris ovalifolia* (Wall.) D. Don, Edinburgh N. Philos. J. 17: 159. 1834. *Xolisma ovalifolia* (Wall.) Rehder, J. Arnold Arbor. **5**: 52. 1924. **Type**: Nepal, May, 1818, Wallich s.n. (K, photo!). (Plates 1 -4)
Vernacular names*:* Sikkim: Piazay (Bhutias), Kangchior, Taksol (Lepchas), Angeri (Nepalese); West Bengal (Darjeeling): Aigiri, Anjir (Tamang Nepalese); Arunachal Pradesh: Aule Angeri (Monpas of Bomdi-La).

Stout, erect shrub, treelet to medium-sized tree, 0.5 – 12 (–18) m high. Stem terete, **greyish-brown with longitudinally furrowed reddish bark**, profusely branched, glabrous or sparsely hairy; branches greyish-brown to pale-green, glabrous to rarely sparsely glandular hairy. Leaves chartaceo-coriaceous to papery, lamina shape and size variable from broadly ovate, ovate, elliptic, ovate-elliptic, oblong-elliptic, lanceolate, oblanceolate, oblong-lanceolate to rarely obovate, 2 – 15 (– 22) × 2 – 8 (–12) cm, **obscurely serrulate-ciliate at margin**, cilia deciduous and up to 5 mm long, rounded, **slightly cordate to broadly cuneate at base**, acuminate or mucronate to mucronulate at apex, dark green to rarely reddish-brown, villous along midvein above, light green to rarely yellowish-brown, villous beneath; venation conspicuous brochidodromous with 10 – 14 pairs lateral veins; petioles stout, 2 – 15 (– 20) mm long, villous. Racemes axillary, **with 1 – 7 basal foliaceous bracts of 2 – 7 cm long**; rachis light green to light greenish-pink, 5 – 15 (– 20) cm long, **10 – 35-flowered**, densely white puberulous. **Flowers 11 – 16 mm long**; pedicels greyish-white to light green, 3 – 6 mm long, densely white puberulous; bract 1, basal, greyish-white to orange brown or green, lanceolate to ovate-triangular, 3 – 8 × 1 – 4 mm, entire to ciliate at margin, short acuminate, acute to mucronulate at apex, glabrous or pilose; bracteoles 2, opposite, basal to subbasal, caducous, greyish-white to orange brown, elliptic to oblong-elliptic, *c*. 1 × 0.5 mm, ciliolate at margin, short acuminate at apex, pilose outside, glabrous inside. Calyx lobes equal, united basally, **pale yellow to light green**, coriaceous, ovate-triangular to ovate-oblong, c. 2 × 1.5 mm, entire at margin, acute at apex, sparsely to densely puberulous. Corolla **tubular, white, snow white, yellowish-white to greenish-white, 7 – 12 × 3 – 6 mm, villous outside, glabrous or villous inside**, lobes equal, ovate-deltoid, c. 1 × 1 mm. Stamens 10, 5 – 8.5 mm long, **loosely epipetalous; filaments greenish-white**, 4.5 – 7 mm long, slender, geniculate, pilose, dilated at base with c. 1 mm long or minute, **2 equal spurs at or below the anther-filament junction**; anthers orange brown, oblong-ovoid to oblong, 1 – 1.5 mm long, glabrous, each lobe with single apical minute appendage. Pistil 4 – 13 mm long; ovary ovoid to oblong, dark green, 1.5 – 2.5 × 1.5 mm, glabrous, ovules numerous on axile placenta in each locule; **disc minutely 10-dentate or obscure**; **style** impressed, pale yellow, 1.5 – 8 mm long, slender, columnar, straight, **slightly swollen near middle**, pilose or glabrous, with or without 4 – 8 fluted central canals; stigma usually truncate, rarely capitate. Capsule loculicidally 5-valved with a dry persistent calyx, globose, subglobose to rarely ovoid, greenish-white (immature) to cinnamon brown (mature), 3 – 4 × 3 – 5 mm with 4 – 9 mm long puberulous pedicel, glabrous. Seeds numerous, blackish-brown, cylindrical, **acicular to sickle-shaped with pointed ends**, c. 1 mm long, scariose.
Unusual Flowers Observation (Plate 4): During field studies at Alubari Jungle Busty vicinity Rock valley forest in Darjeeling Himalaya in June 2018, unusual flowering noticed in two individual plants at an altitude of about 7000 ft. Flowering peduncles 8-10 cm long, consisting of 24-42-flowering buds, but only single unusual flower seen in two peduncles. Flower ca. 10 mm long excluding pedicels, bracts and bracteoles not seen; pedicels ca. 8 mm long. Calyx lobes 5, grey white, succulent. Corolla grey white, ca. 8 mm long, not tubular like usual corolla, but campanulate to wide deltoid-campanulate, 5 lobes, succulent. Stamens 10, all steriles, without anther lobes. Pistils deltoid with 10 prominent discs alternating with stamens, greenish white, style not seen, stigma capitate.
Distribution: INDIA: Himalayas (Jammu & Kashmir, Himachal Pradesh, Uttarakhand, Sikkim, West Bengal and Arunachal Pradesh) and NE India (Meghalaya, Nagaland, Manipur and Mizoram); PAKISTAN; NEPAL; BHUTAN; BANGLADESH; W CHINA (including Hainan); TAIWAN; N MYANMAR; THAILAND; VIETNAM; CAMBODIA; MALESIA; INDONESIA and JAPAN.
Habitat: This variety grows gregariously in dry as well as moist rocky slopes, sometimes in loose humus-covered rocky soils, rarely hanging down from rock crevices in association with *Quercus incana*, *Q. griffithii*, *Pinus kesiya*, *Lithocarpus dealbatus*, *Cedrus deodara*, *Rhododendron arboreum*, *Vaccinium vacciniaceum*, *Gaultheria fragrantissima*, *G. codonantha* and *Pieris formosa* at altitudes ranging from (500 –) 900 – 3100 (– 3500) m.
Flowering: Late March – June. **Fruiting:** July – December.
Specimens examined: **Arunachal Pradesh**: Tawang district: Mukto, 08.12.1999, G. D. Pal 8341 (ARUN); West Kameng district: 7 km from Rupa toward Chilipam, 5000 ft, 26.12.2002, S. Panda 30836; Salari forest, 1900 m, 10.05.2010, *S. Panda* 717 (BGC). Lower Subansiri district: Ziro to Tamin, 14.06.1961, G. V. Subba Rao 25022 (ASSAM); West Siang district: summit between Lalik & Serpo, 5100 ft, 28.01.1912, I. H. Burkill 36362; Lohit district: Walong Hill top, 1400 m, 28.04.2003, S. Panda 30888. **Sikkim**: East district: Pangthang, 16.12.1980, B. Krishna 1208; North district: Lachung to Chungthang, 6000 ft, 06.05.2001, S. Panda 30095; South district: Way to Namchi, 09.06.1987, B. Krishna 7475 (BSHC); West district: Hilley to Versay, 2700 m, 27.04.2001, S. Panda 30021. **West Bengal** (Darjeeling): Senchal forest, 7000 ft, 28.06.1862, T. Anderson 780; West Bengal, Darjeeling, Alubari Jungle busty, 2040 m, 02.06.2014, S. Panda 71 (DGC) & Alubari jungle busty, 2080 m, 07.06.2015, S. Panda 117 (DGC: irregular flower).
Uses: Sikkim & Darjeeling: i. juice of tender leaves used in severe skin diseases by the Nepalese of Darjeeling and Sikkim (S. Panda 71 & 30702). ii. Lepchas of Lachung and Chungthang in North Sikkim (S. Panda 29956; ref. Gamble, 1896; Chopra et al., 1956; Gurung, 2002): Vernacular names-Kangshior, Taksol, Kechung. Mode of use: the infusion of tender leaves is used in cutaneous diseases. iii. Also powder of tender leaves are often used as insecticides by the Lepchas of Lachung. iv. Bhutias of Lachung (S. Panda 30067). Vernacular

names-Piazay, Senchum. Mode of use: powder of tender leaves are often used as domestic insecticides. **Arunachal Pradesh**: i. older leaves are used for making 'Biri' and smokes from 'Biri' is administered to the asthma patients to relieve pain by the Nepalese of Walong, Arunachal Pradesh; (S. Panda 30861). ii. Leaves are used as insecticides to kill harmful soil-borne insects in rice cultivation at Kahao village in Anjaw district by the Miju Mishmese.

IUCN Status: LC (Least Concern: assessed in 2018).

Leaf-Stomata (Plate 3A-B): The study of Light Microscopic stomatal architecture (45X, 100X) includes number, form, stomatal index and arrangement of specialized epidermal cells associated with the stomatal guard cells. These stomatographic studies have shown that stomata can be used as valuable taxonomic marker to delimit taxa. **Type:** The investigated species shows only one type, paracytic (Dilcher 1974). **Dimensions of stomata:** The average dimension is 37.3 µm × 24 µm. **Size of guard cells:** The average dimension is 37.3 µm × 6.3 µm. **Size of epidermal cells:** Epidermal cells are irregular: polygonal. Epidermal walls in surface view are slightly arched. The epidermal walls in the adaxial surface are also slightly arched, cells are irregular-polygonal. **Dimensions of epidermal cells:** The average dimension is 25.7 µm × 12.6 µm. The maximum length is 31.7 µm and breadth is 16.5 µm. The minimum length is 19.8 µm and breadth is 8.8 µm. **Stomatal Index (SI):** 5.7. The average number of stomata is 14 per 228 epidermal cells per microscopic field. SI = S/S+E × 100 = 14/14 + 228 × 100 = 5.7.

Leaf areoles (vein islets) (Plate 3C-F): Mostly quadrangular, pentagonal to occasionally triangular and irregular in shape. Larger areole: 312×172 µm. Smaller areole: 91×72 µm. Areoles: 10 (average) per 1 mm^2. Vein endings: 30 (average) per 1 mm^2; veinlets simple unbranched to branched (once). Branched and unbranched veinlets occur in the same areole. Vein ends: bulbous to rounded, never taperated.

Trichomes (Plate 1E-F): abaxial leaves shows simple, unbranched, multicellular (many-celled), non-glandular trichomes of *c*. 218 µm long. Stem and branches possess multicellular (many-celled) non-glandular (*c*. 176 µm long) trichomes. Floral parts viz., corolla, pedicels, calyx lobes possess multicellular (2 to many-celled) trichomes of 140.8 µm – 529.6 µm long.

2. *L. villosa* (Wall. ex C. B. Clarke) Hand.-Mazz., Symb. Sin. **7**: 789. 1936; Judd, J. Arnold Arbor. **62** (2): 177. 1981; Hara in Hara et al., Enum. Fl. Pl. Nepal **3**: 56. 1982; Rae in A. J. C. Grierson & D. G. Long, Fl. Bhutan **2** (1): 395. 1991; F. Ruizheng & P. F. Stevens in Wu & al., Fl. China **14**: 463. 2005; Panda & Sanjappa in Sanjappa & Sastry, Fasc. Fl. India **25**: 240. 2014. *Andromeda villosa* Wall., Numer. List: no. 762. 1829. *Pieris villosa* Wall. ex C. B. Clarke in Hook. f., Fl. Brit. India **3**: 461. 1882; *Xolisma villosa* (Wall. ex C. B. Clarke) Rehder, J. Arnold Arbor. **5**: 53. 1924. Type: Nepal, Gossainthan, 1821, Wallich [in Wall. list no. 762] *s.n*. (K, photo!). (Plates 5-7)

Vernacular name: Sikkim & Darjeeling: Lekh Angeri (Nepalese).

Stout, erect shrub to treelet, 1–4 m high. **Stem greyish-white**, terete, profusely branched, glabrous; **branches blackish-brown**, terete, glabrous. Leaves chartaceo-coriaceous to papery, lamina elliptic, oblong-elliptic to rarely ovate-elliptic, 4–8×2–4 cm, **obscurely serrulate-ciliate at margin, cilia deciduous and up to 3 mm long**, **rounded to slightly cordate at base**, **mucronate at apex**, villous, dark green above, light green beneath; venation brochidodromous with 6–9 pairs lateral veins; petioles stout, 4–7 mm long, villous. Racemes axillary; rachis light green, 3–5 cm long, **6–8-flowered, densely white puberulous with 2–5 basal foliose bracts** of 2–4×1–2 cm. **Flowers 12–18 mm long, scented**; pedicels light green, 5–7 mm long, sparsely white puberulous; pedicellar bract 1, basal, dark green to light green with pinkish stripes, oblanceolate to obovate-elliptic, 7–30×3–10 mm, ciliolate at margin, mucronulate at apex, sparsely villous outside, glabrous inside; bracteoles not seen. Calyx lobes equal, basally united, **light green with pinkish stripes**, oblong to oblong-obovate, c. 3.5×1 mm, ciliolate at margin, acute at apex, sparsely villous outside, glabrous inside. Corolla **tubular to rarely tubulo-urceolate, pinkish-white**, 7–11 × 4–7 mm, villous outside, glabrous inside, lobes ovate, c. 1×1 mm. Stamens 10, c. 6 mm long, **loosely epipetalous; filaments greyish-white**, c. 4.5 mm long, slender, geniculate, pilose, slightly dilated near base, **anther-filament junction spurless**; anthers orange brown, oblong, c. 1.5 mm long, glabrous. Pistil c. 7 mm long; ovary globose to subglobose, light green, c. 2×2.5 mm, glabrous, ovules numerous on axile placenta in each locule; disc minutely 10-dentate; style greyish-white, c. 5 mm long, slender, columnar, straight, slightly swollen near middle, glabrous; stigma truncate. Capsule loculicidally 5-valved with a dry persistent calyx, globose, dark brown, c. 4×4 mm including c. 7 mm long puberulous pedicel, glabrous. Seeds numerous, **blackish-brown, cylindrical, acicular to sickle-shaped with pointed ends**, c. 1 mm long, scariose.

Distribution: INDIA: Himalayas (Uttarakhand, Sikkim, West Bengal and Arunachal Pradesh); NEPAL; BHUTAN; W CHINA and N MYANMAR.

Habitat: This species is rare and grown in moist rocky slopes, sometimes in loose humus-covered rocky soils in association with *Enkianthus deflexus*, *Gaultheria hookeri*, *Quercus incana* and *Vaccinium glauco-album* at altitudes ranging from 2500–4000 (–4400) m.

Flowering: Late May–June. **Fruiting*:*** July–Sept.

Specimens examined: Arunachal Pradesh: Se La, Tawang district, 28.10.1955, R. S. Rao 1583. Sikkim: East district: Yak La, 12000 ft, 17.10.1869, C. B. Clarke 10068; North district: on the bank of Zema River, Lachen, 8000 ft, 31.05.2002, S. Panda 30801; Yakche to Phuney, 10000 ft, 01.06.2002, S. Panda 30808. West Bengal (Darjeeling): Sandakphu, 10000 ft, July, 1881, J. S. Gamble 9524; Tonglu, 10000 ft, 21.05.2022, S. Panda 004 (MAC); Tonglu, 12.05.2012, S. Panda 129 (DGC).

Uses: juice of tender leaves applied in scabies, severe skin diseases, boils and pimples by the Nepalese of Chungthang in North Sikkim (S. Panda 30801, CAL) and Tumling in Darjeeling (S. Panda 129, DGC).

IUCN Status: LC (Least Concern: assessed in 2018). But populations are very smaller and fragmented compare to *L. ovalifolia*.

Leaf-Stomata (Plate 7A-B): The study of Light Microscopic stomatal architecture (45X, 100X) includes number, form, stomatal index and arrangement of specialized epidermal cells associated with the stomatal guard cells. **Type:** The investigated species shows three types, **paracytic, pericytic**

and **desmocytic** (Dilcher, 1974). **Dimensions of stomata:** The average dimension is 21.8 µm × 16.7 µm. **Size of guard cells:** The average dimension is 21.8 µm × 5.7 µm. **Size of epidermal cells:** Epidermal cells are irregular: polygonal to elongated. Epidermal walls in surface view are slightly arched. The epidermal walls in the adaxial surface are also slightly arched, cells are irregular-polygonal. **Dimensions of epidermal cells:** The average dimension is 22.1 µm × 12.6 µm. The maximum length is 31.7 µm and breadth is 16.5 µm. The minimum length is 12.6 µm and breadth is 8.8 µm. **Stomatal Index (SI):** 14.1. The average number of stomata is 45 per 272 epidermal cells per microscopic field. SI = S/S+E × 100=45/45+272×100 = 14.1.

Leaf areoles (vein islets) (Plate 7C-E): Mostly quadrangular, pentagonal to hexagonal in shape. Larger areole: 216×142µm. Smaller areole: 112×92µm. Areoles: 4 (average) per 1 mm^2. Vein endings: 14 (average) per 1 mm^2; veinlets simple unbranched to branched (once). Branched and unbranched veinlets occur in the same areole. Vein ends: bulbous to rounded, never taperated.

Trichomes (Plate 7F): abaxial leaf shows simple, unbranched, unicellular and multicellular (many-celled), non-glandular trichomes of 231 µm – 323.6 µm long. Stem and branches possess multicellular (many-celled) non-glandular (*c*. 322 µm long) trichomes.

3. *Lyonia macrocalyx* (J. Anthony) Airy Shaw in Curtis's Bot. Mag. **160**: t. 9490. 1937; Judd, J. Arnold Arbor. **62** (2): 185. 1981; Panda, Bull. bot. Surv. India **48** (1 – 4): 207 – 210. 2006; F. Ruizheng & P. F. Stevens in Wu & al., Fl. China **14**: 463. 2005; Panda & Sanjappa in Sanjappa & Sastry, Fasc. Fl. India **25**: 238. 2014. *Pieris macrocalyx* J. Anthony, Notes Roy. Bot. Gard. Edinburgh **15**: 241. 1927. Type: China, Yunnan, Hills north of Lung-fan, 25º 54′ N, 98º 33′E, 9000 ft, June, 1925, G. Forrest 26800 (holotype: E, n.v.; iso: A, n.v.). (Plates 8-9)

Stout, erect shrub to treelet, 1–2 m high. Stem **greyish-white to pale brown**, terete, profusely branched, glabrous; **branchlets light green**, **densely villous or covered with long headed glandular hairs**. Leaves chartaceo-coriaceous to papery, lamina elliptic, oblong-elliptic to rarely ovate, 5–11×2–4 cm, **obscurely serrulate-ciliate at margin**, **broadly cuneate at base**, acuminate, **long acuminate to mucronate at apex**, dark green, glaucous, densely villous along midvein above, light green, villous along lateral veins beneath; venation brochidodromous with 10–12 pairs lateral veins; petioles 4–7 mm long, villous. Infructescence **blackish-brown, 3–7 cm long**, rachis consisting of 8–12 fruits, glabrous or densely puberulous. Calyx lobes of capsule 5-lobed, equal, basally united, ovate, ovate-elliptic to lanceolate, *c*. (7–)12 × (1.5–)4 cm, entire at margin, acuminate at apex, glabrous. Capsule loculicidally 5-valved with a dry persistent calyx, subglobose to globose-ovoid, 5–6×4–6 mm with (6–)11–17 mm long glabrous or puberulous pedicel, glabrous. Seeds numerous, linear, elliptic, **acicular, sickle-shaped**, 1–2.5 mm long, scariose.

Distribution: INDIA: E Himalayas (Arunachal Pradesh) and NE India (Nagaland); W CHINA and N MAYANMAR.

Habitat: This species grows gregariously in discontinuous patches in dry as well as moist rocky slopes in association with *Lyonia ovalifolia*, *Rhododendron arboreum* and *Rubus* sp. at an altitude of c. 1800 m.

Flowering: not known in India (May to July in China). **Fruiting**: November – December; March – April.

Specimens examined: Arunachal Pradesh: Shikar forest, West Siang district, 28.11.1984, Haridasan 1825 (APFH); 18.11.1994, Haridasan 7283 (APFH).

IUCN Status: Not assessed yet. But populations are very smaller and fragmented compare to both *L. ovalifolia* and *L. villosa*.

Leaf-Stomata (Plate 9A-B): The study of Light Microscopic stomatal architecture (45X, 100X) includes number, form, stomatal index and arrangement of specialized epidermal cells associated with the stomatal guard cells. **Type:** The investigated species shows only one type, paracytic (Dilcher, 1974). **Dimensions of stomata:** The average dimension is 24 µm × 21.5 µm. **Size of guard cells:** The average dimension is 24 µm × 6.3 µm. **Size of epidermal cells:** Epidermal cells are more or less isodiametric-polygonal. Epidermal walls in surface view are slightly arched. The epidermal walls in the adaxial surface are also slightly arched, cells are isodiametric-polygonal. **Dimensions of epidermal cells:** The average dimension is 22.1 µm × 12.6 µm. The maximum length is 25.3 µm and breadth is 9.2 µm. The minimum length is 9.1 µm and breadth is 5.7 µm. **Stomatal Index (SI):** 9.1. The average number of stomata is 29 per 288 epidermal cells per microscopic field. SI = S/S+E × 100 = 29/29 + 288 × 100 = 9.1.

Leaf areoles (vein islets) (Plate 9C-E): Quadrangular, pentagonal, hexagonal to occasionally irregular in shape. Larger areole: 182×141µm. Smaller areole: 78×62µm. Areoles: 6 (average) per 1 mm^2. Vein endings: 12 (average) per 1 mm^2; veinlets simple unbranched to branched (once). Branched and unbranched veinlets occur in the same areole. Vein ends: elongated-bulbous, but not rounded and taperated.

Trichomes (Plate 9F): abaxial leaf shows simple, unbranched, multicellular (several-celled), non-glandular trichomes of *c*. 78.6 µm long. Stem and branches possess multicellular (many-celled) non-glandular (*c*. 281.7 µm long) trichomes. Floral parts viz., sepals, corolla possess both unicellular and multicellular trichomes of 423 µm – 835 µm long.

Discussion

The diversity of the genus *Lyonia* Nutt. in the Indian Eastern Himalayas based on extensive field surveys to remote areas of Darjeeling, Sikkim and Arunachal Himalayas, herbarium consultations in Indian herbaria as well as abroad (Cibachrome images), field-based morpho-taxonomical amplified description (unique field-based characters are highlighted as bold-italics), detailed global and regional distribution, locality-based reports under Specimen examinations, IUCN status, ITKs as well as light microscopic leaf anatomical investigations, has not been studied in detail earlier. This work includes several first hand new and noteworthy ITKs based on oral interviews with elderly ethnic people of the Eastern Himalayas, mostly dealing with ethnomedicinal uses, which may be subjects for herbal medicines in the global market following proper laboratory-based research. This work also provides identification keys to the species (based on vegetative, floral and leaf anatomical unique data) for the purpose of identification in the field as well as in herbaria. The purpose of the present study is to evaluate the contribution of leaf stomata, leaf vein

islets and trichomes along with field-based exomorphological unique data to stabilize species delimitation in the genus *Lyonia* Nuttall.

Acknowledgements

I am thankful to the Herbarium-In-Charges of CAL, DD, ASSAM, ARUN, APFH, BSHC & Lloyd Botanical Garden Herbaria for their permission to consult herbarium specimens, to Dr. P. Lama, Officer-In-Charge of Darjeeling Govt College for his all types of cooperation and permission to carry out the work. I am also grateful to Dr. Partha Roy, Head Department of Botany and Dr. Subhasis Dutta, Principal of Maulana Azad College, Kolkata for their kind permission and providing necessary facilities during the course of the work.

References

Brummitt, R. K. & C. E. Powell (1992): Authors of Plant Names. - Royal Botanic Gardens, Kew, United Kingdom. p.1-732.

Carpenter, K. J. (2005): Stomatal architecture and evolution in basal angiosperms. - American Journal of Botany **92** (10): 1595-1615.

Cox, H. T. (1948): Studies in the comparative anatomy of the Ericales II: Ericaceae - subfamily Arbutoideae. - The American Midland Naturalist **40** (2): 493-498.

Dilcher, D. L. (1974): Approaches to the identification of angiosperm leaf remains. - Botanical Review **40** (1): 1-53.

Fahn, A. (1997): Plant Anatomy. - 4th ed., Aditya Books (P) Ltd., New Delhi (India), p. 168.

Featherly, H. I. (1954): Taxonomic Terminology of the Higher Plants. - Ames, Iowa (USA), 1-166.

Hagerup, O. (1953): Leaf morphological and systematics in Bicornes. - Phytomorphology **3**: 459- 464.

Hara, H. (1966). The Flora of Eastern Himalaya – Results of the Botanical Expedition to Eastern Himalaya organized by the University of Tokyo, 1960 – 1963. - University of Tokyo Press, Japan, 234, 647.

Hara, H.; A. O. Charter & L. H. J. Williams (1982): An Enumeration of the Flowering Plants of Nepal **3. -** Trustees of British Museum (Natural History), London, 55-59.

Hickey, L. J. (1973). Classification of the architecture of dicotyledonous leaves. - American Journal of Botany **60** (1): 17-33.

Holmgren, P. K.; N. H. Holmgren & L. C. Barnett (1990): Index Herbariorum, part 1: The Herbaria of the World. - ed. 8, New York Botanical Garden, Bronx, New York (USA).

Judd, W. S. (1981): A monograph of *Lyonia* (Ericaceae). - Journal of the Arnold Arboratum **62**: 63-209, 315-436.

Kron, K. A.; W. S. Judd, P. F. Stevens, D. M. Crayn, A. A. Anderberg, P. A. Gadek, C. J. Quinn & J. L. Luteyn (2002): Phylogenetic classification of *Ericaceae*: Molecular and morphological evidence. - Botanical Review **68** (3): 335-423.

Lawrence, G. H. M. (1951): Taxonomy of Vascular Plants. - Macmillan Co., New York (USA).

Lems, K. (1964): Evolutionary studies in the Ericaceae II. Leaf anatomy as a phylogenetic index in the Andromedeae. - Botanical Gazzette **125** (3): 178-186.

Mabberley, D. J. (2008): *Lyonia* Nutt. Mabberley's Plant-Book - A portable dictionary of plants, their classification and uses. - 3rd ed., Cambridge University Press, Cambridge, UK, pp. 507.

Metcalfe, C. R. & L. Chalk (1950). Anatomy of Dicotyledons, Vol. **1**. - Clarendon Press, Oxford.

Niedenzu, F. (1890): Über den anatomischen Bau der Laubblätter der Arbutoideae und Vaccinoideae in Beziehung zu ihrer systematische Gruppierung und geographischen Verbreitung. - Botanische Jahrbücher **11**: 134-263.

Panda, S. (2006): *Lyonia macrocalyx* (J. Anthony) Airy Shaw (Ericaceae) – A new record for India from Arunachal Pradesh and Nagaland. - Bulletin of the Botanical Survey of India **48**: 157- 164.

- (2008): *Lyonia* Nutt. - In: Taxonomic revision of some selected genera of Ericaceae in India, Published Ph. D. thesis, Vidyasagar University, Midnapore (India), pp. 204-222.

Panda, S. & M. Sanjappa (2014): *Lyonia* Nutt. - In: Sanjappa, S. & A. R. K. Sastry (eds.): Fascicles of Flora of India: Fascicle 25 Ericaceae, Botanical Survey of India. Kolkata, pp. 229-242.

Quattrocchi, U. (2000). CRC World Dictionary of Plant Names (vols. 1-4). - CRC Press, LLC, Corporate Blvd., NW Boca Raton, Florida (USA).

Radford, A. E. (1986): Ericaceae. Fundamentals of Plant Systematics. - Harpers & Row, New York (USA): 365-367.

Ruizheng, F. & P. F. Stevens (2005): *Lyonia* Nutt. - In: Wu, Z. Y.; P. H. Raven & D. Y. Hong (eds.): Flora of China – Apiaceae through Ericaceae, vol. 14, Science Press (China) and Missouri Botanic Garden Press (USA). pp. 461-463.

Stace, C. A. (1965): Cuticular studies as an aid to plant taxonomy. - Bulletin of the British Museum, Botany **4**: 3-78.

- (1989): Plant Taxonomy and Biosystematics. - 2nd ed., Edward Arnold. 78-80.

Stearn, W. T. (1983): Botanical Latin. - 3rd revised ed., David & Charles Inc., USA.

Stevens, P. F. (1970): *Calluna*, *Cassiope* and *Harrimanella*: a taxonomic and evolutionary problem. - The New Phytologist **69**: 1131-1148.

- (1971): A classification of the Ericaceae: Subfamilies and tribes. - Botanical Journal of the Linnean Society **64**: 1-53.

Stevens, P. F.; J. Luteyn, E. G. H. Oliver, T. L. Bell, E. A. Brwon, R. K. Crowden, A. S. George, G. J. Jordan, P. Ladd, K. L.; C. B. McLean, Y. Menadue, J. S. Pate, H. M. Stace & C. M. Weiller (2004): Ericaceae. - In: Kubitzki, K. (ed.): The Families and Genera of Vascular Plants **6**: Springer, Berlin, 145-194.

Veldkamp, J. F. (1987): Manual for the description of Flowering Plants. - In: Vogel, E. F.: Manual of Herbarium Taxonomy: Theory and Practice, UNESCO Regional Office, Jakarta (Indonesia): 20-64.

Watson, L. (1964): The taxonomic significance of certain anatomical observations on Ericaceae. The Ericoideae, *Calluna* and *Cassiope*. - The New Phytologist **63** (1): 274-280.

- (1965): The taxonomic significance of certain anatomical variations among Ericaceae. - Journal of the Linnean Society (Botany) **59**: 111-126.

IUCN Standards and Petitions Committee (2019): Guidelines for Using the IUCN Red List Categories and Criteria, version 14. Prepared by the Standards and Petitions Committee of IUCN Species Survival Commission. p. 1-110. http://www.iucnredlist.org/documents/RedListGuidelines.pdf

Author's address:

Subhasis Panda
Biodiversity Conservation Laboratory
Government General Degree College
Chapra, University of Kalyani
Shikra, PO. Padmamala, PS. Chapra
Nadia-741123
West Bengal, India
Email: bgc.panda@gmail.com

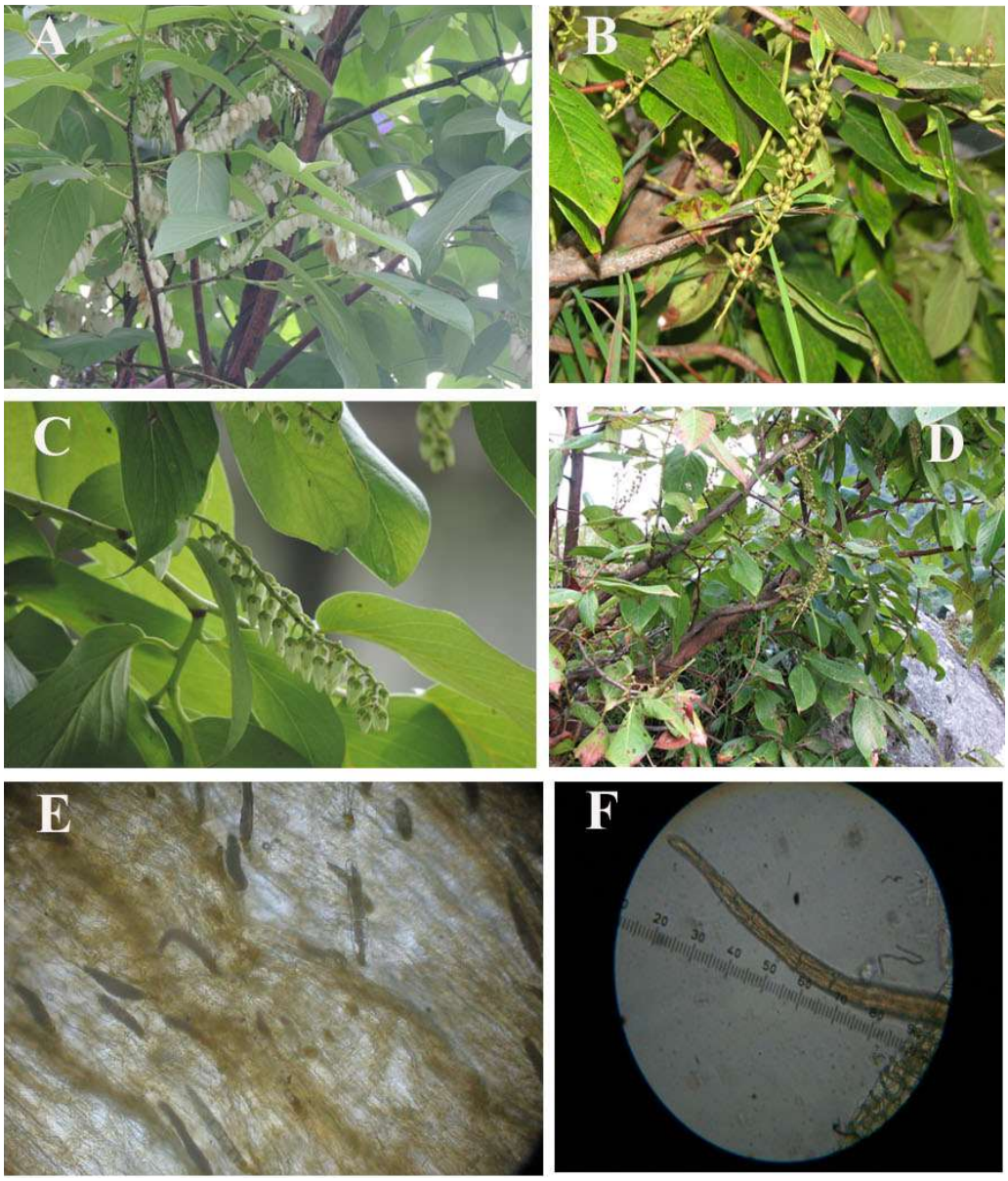


Plate 1. *Lyonia ovalifolia* (Wall.) Drude: A, C: Habit and Flowering twig observed at Jungle Busty Darjeeling in June, 2016; B, D: Fruiting plants observed at Chungtam in North Sikkim in September 2010; E, F: Leaf abaxial surfaces Trichomes (10X & 40X, S. Panda 71 & 717).

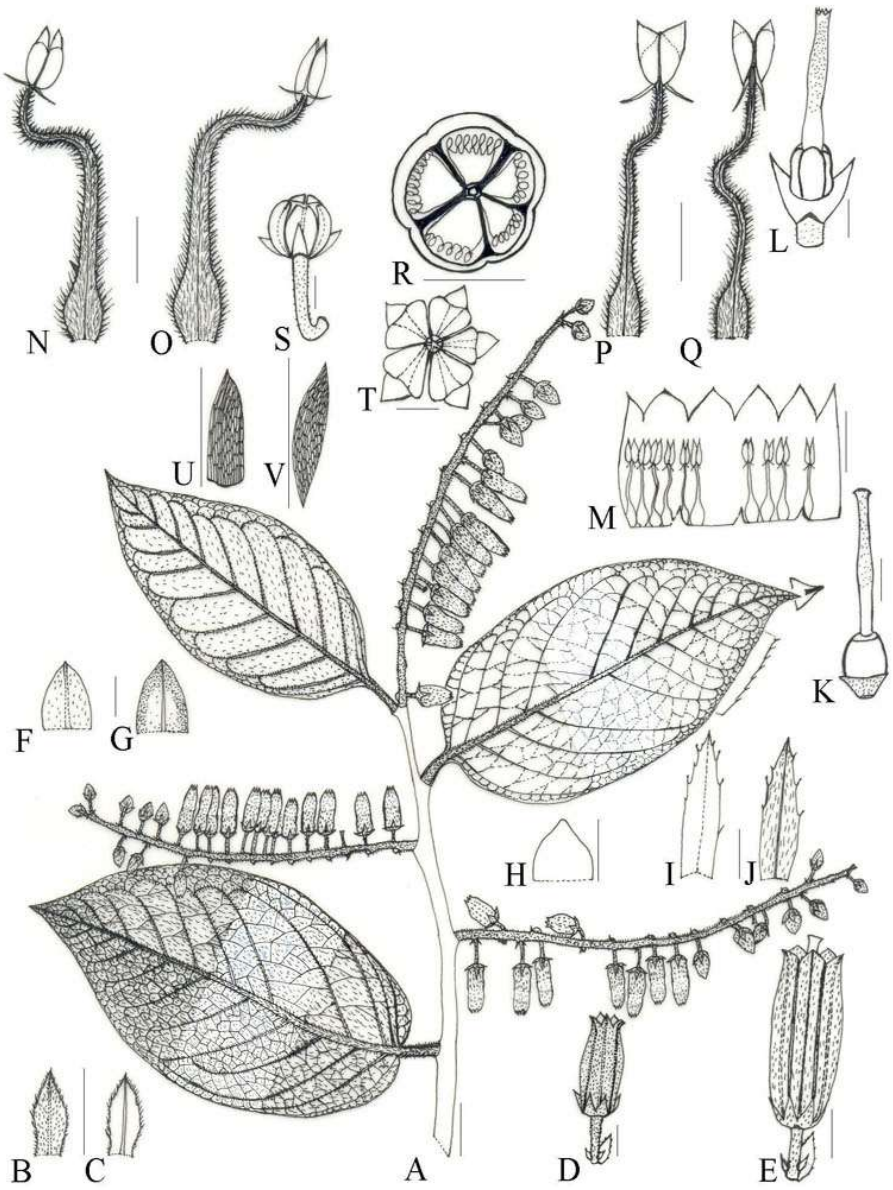


Plate 2. *Lyonia ovalifolia* (Wall.) Drude: A: habit; B, C: bracteoles; D, E: flowers; F, G: calyx lobes; H: corolla lobe; I, J: bracts; K, L: pistils; M: corolla split open; N – Q: stamens; R: ovary (t.s.); S, T: fruits; U, V: seeds. Scale bars: A = 1 cm; B, C, F, G-J, R, U, V = 1 mm; D, E = 3 mm; K, L, N-Q, S, T = 2 mm; M = 5 mm (A-D, F-K, M, N, O, R-V: drawn from S. Panda 29947, CAL; E, L, P, Q: S. Panda 30095, CAL).

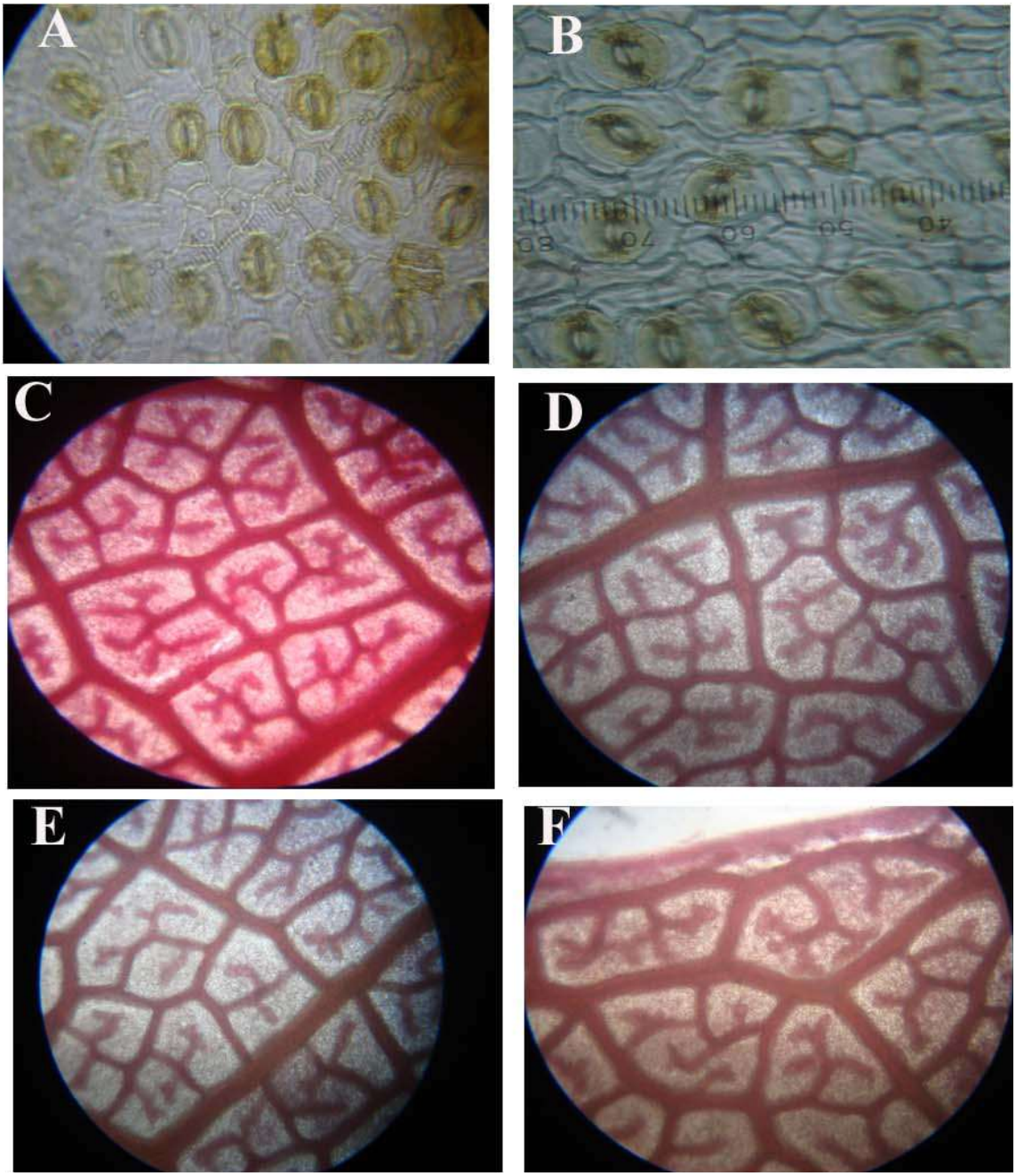


Plate 3. *Lyonia ovalifolia* (Wall.) Drude (S. Panda 71): A, B: leaf stomata (10X, 40X); C - F: leaf vein islets (C: 5X, apex; D: 5X, middle; E - F: 5X & 10X, basal parts).

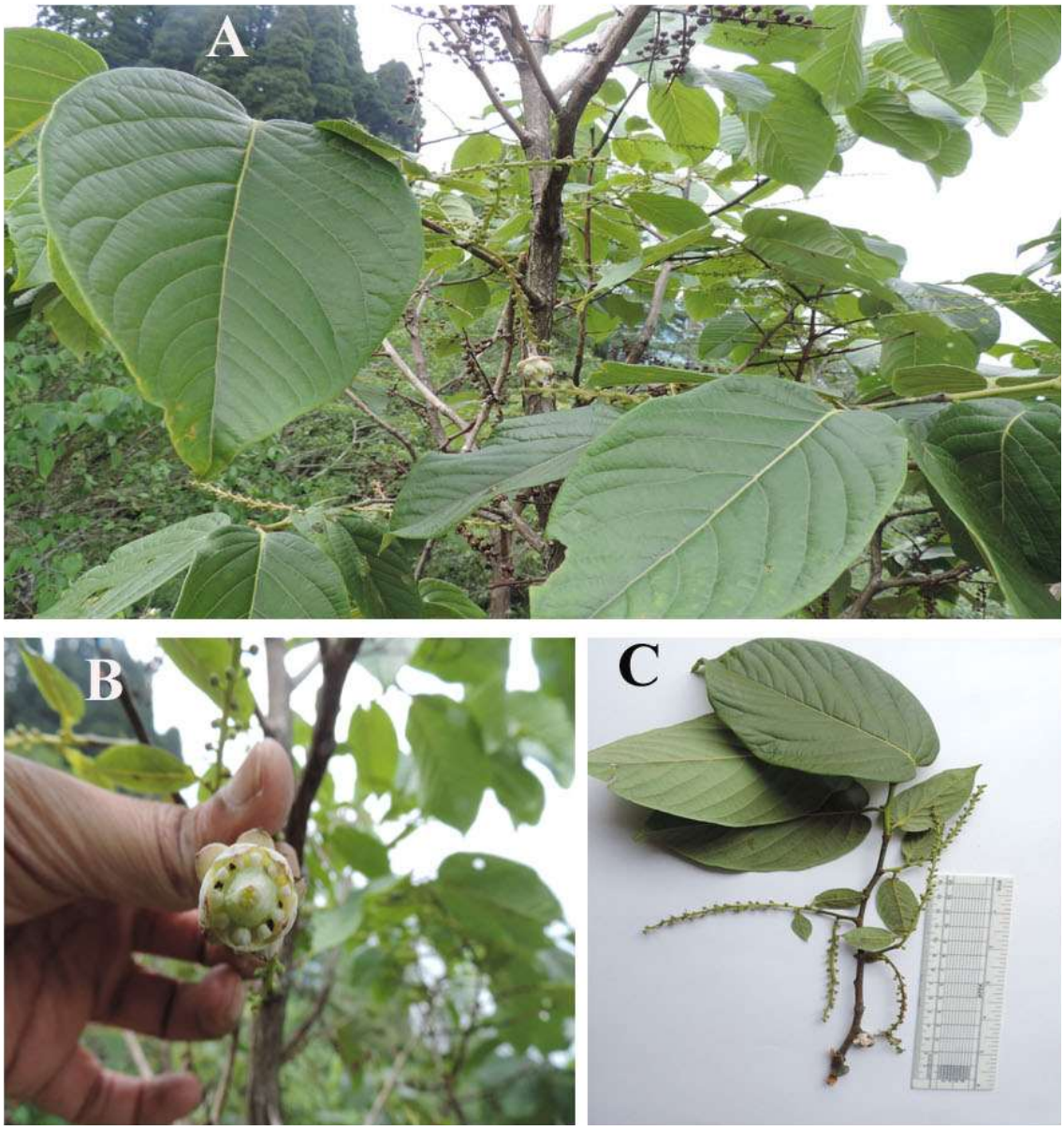


Plate 4. Unusual flower formation of *Lyonia ovalifolia* (Wall.) Drude: observed at Alubari Jungle Busty in Darjeeling Himalaya in June 2015. C: habit with unusual flowering branch with measuring scale (S. Panda 712, DGC).

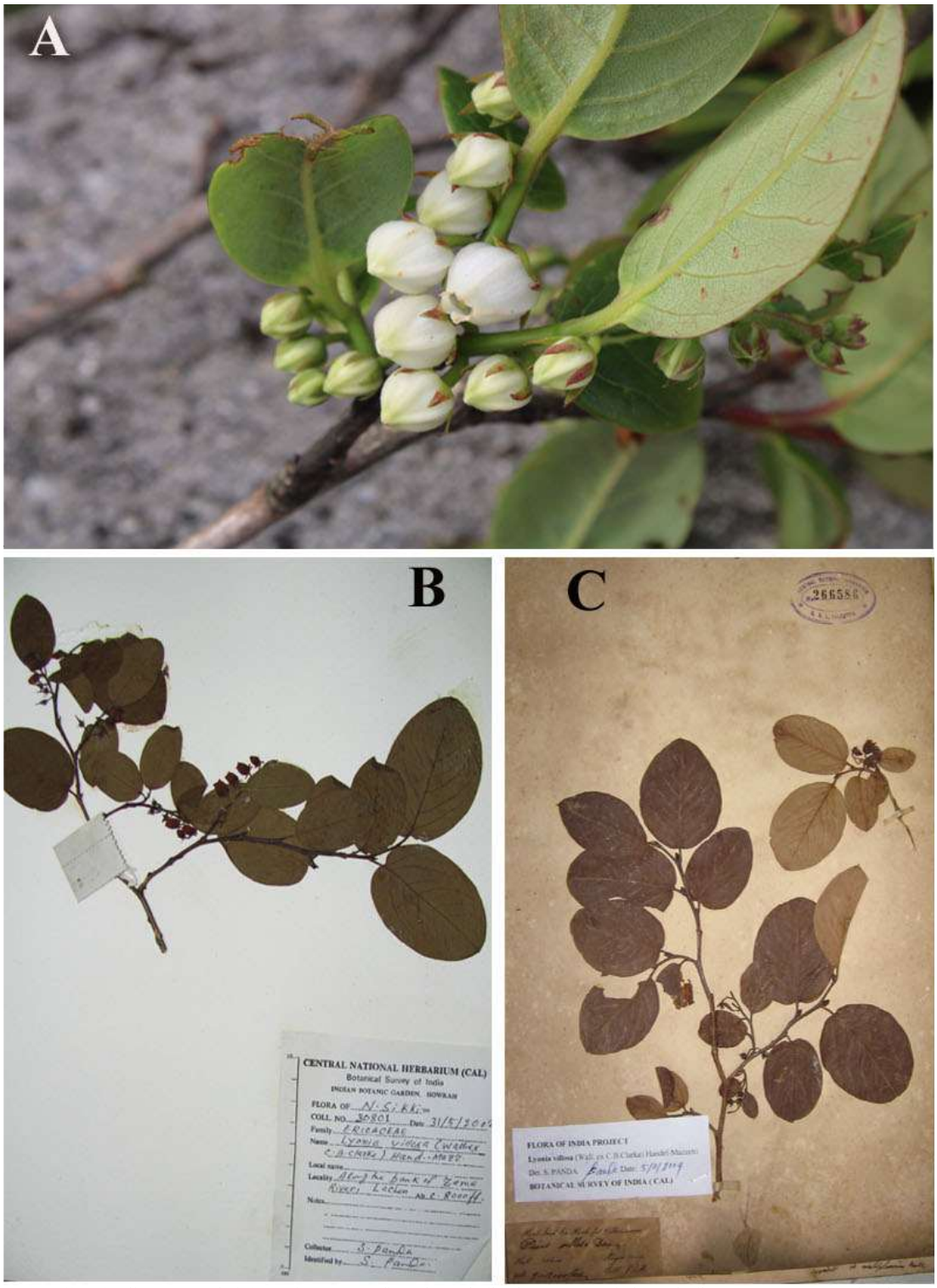


Plate 5. *Lyonia villosa* (Wall. Ex C.B. Clarke) Hand.-Mazz.; A: flowering twig live imgae at Tonglu in Darjeeling, May 2022; B: Lachen in North Sikkim in May, 2002, S. Panda 30801, CAL; C. from North Sikkim by J.D. Hooker, CAL.

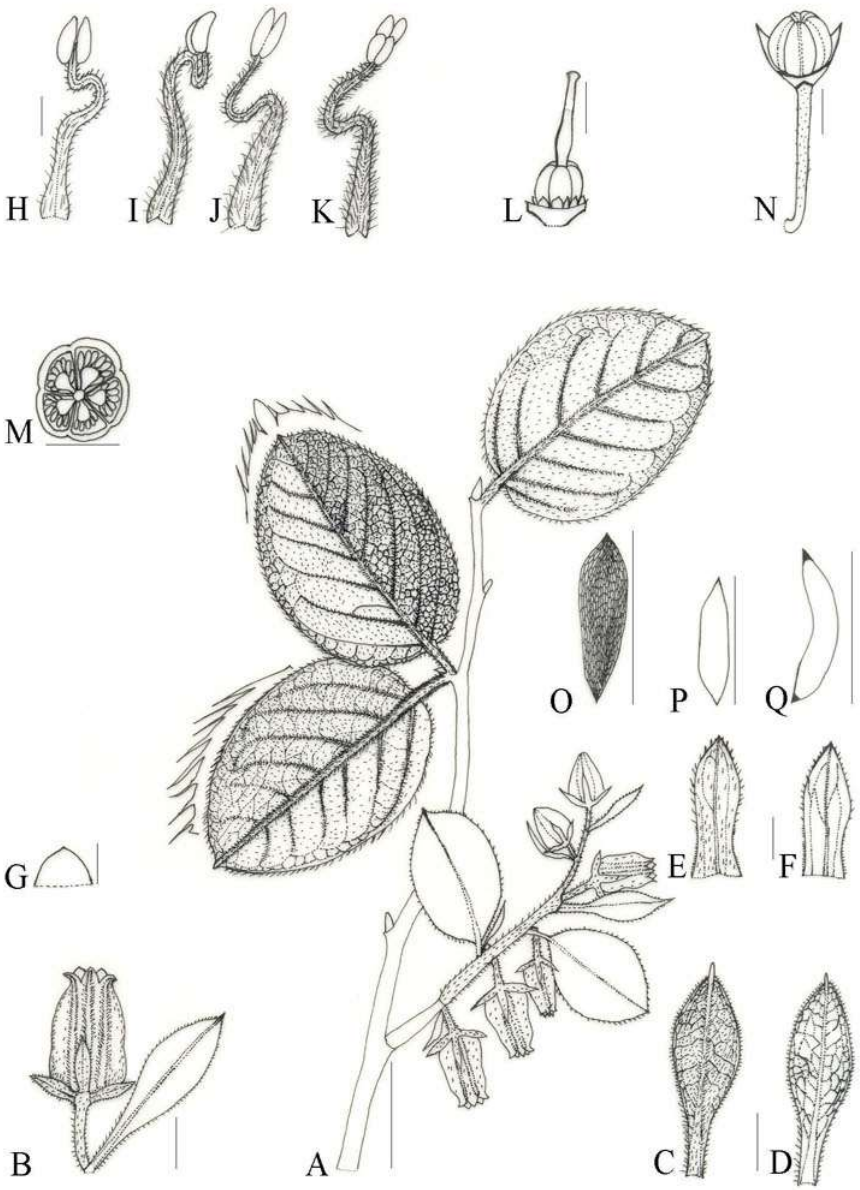


Plate 6. *Lyonia villosa* (Wall. Ex C.B. Clarke) Hand.-Mazz.: A: habit; B: flower; C, D: bracts; E, F: bracteoles; G: corolla lobe; H – K: stamens; L: pistil; M: ovary (t.s.); N: fruit; O – Q: seeds. Scale bars: A = 2 mm; B – D = 4 mm; E – K, O -Q = 1 mm; L – N = 2 mm (A – M: drwan from S. Panda 30801; N – Q: s.l., acc. no. 266594).

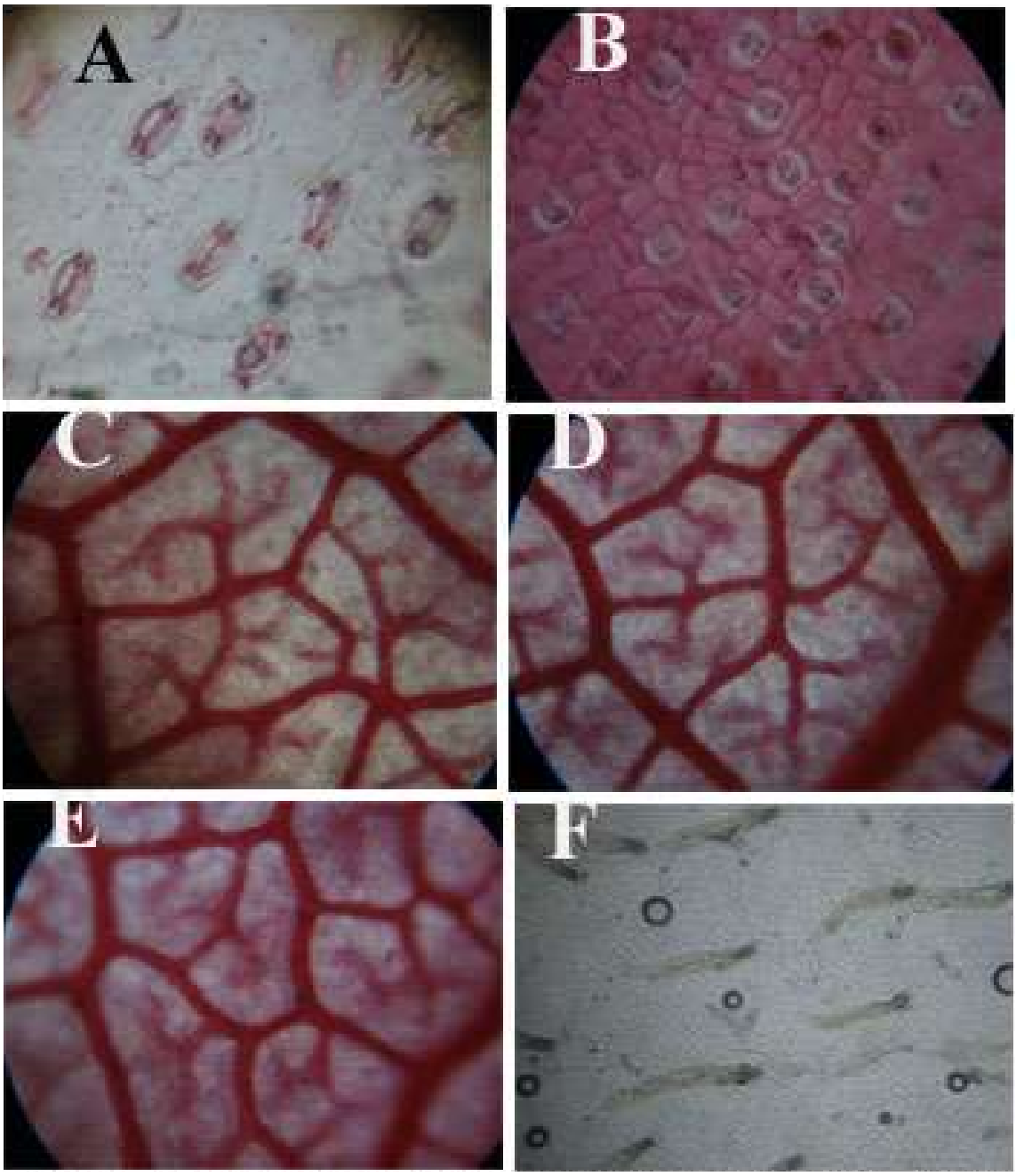


Plate 7. *Lyonia villosa* (Wall. Ex C.B. Clarke) Hand.-Mazz.: A, B: leaf stomata (45X); C – E: leaf vein islets - apex, base & middle (10X); F: leaf trichome (5X), S. Panda 129, DGC.

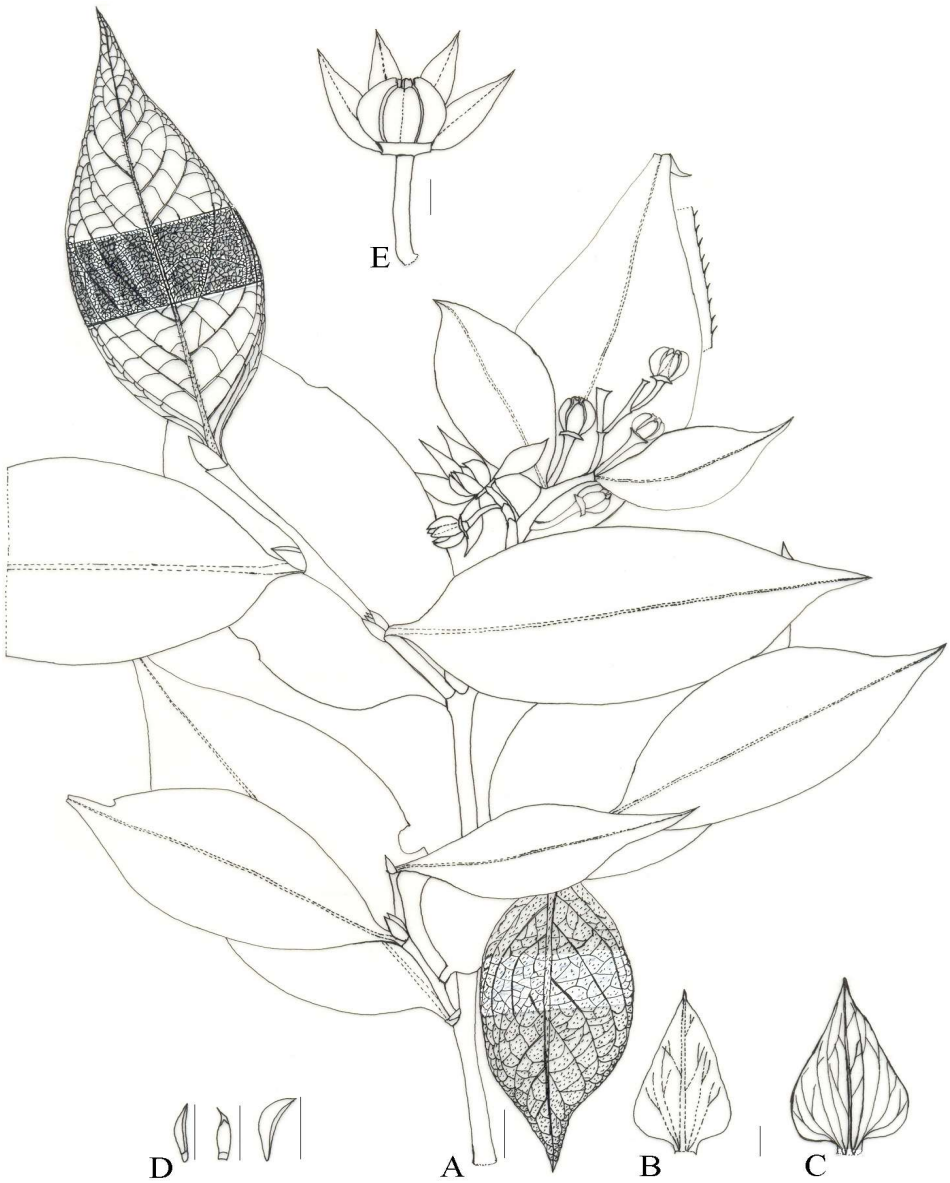


Plate 8. *Lyonia macrocalyx* **(** J.Anthony) Airy Shaw: A: habit; B, C: calyx lobes; D: seeds; E: fruit. Scale bars: A = 1 cm; B, C = 2 mm; D = 2.5 mm; E = 3 mm (A – E: drawn from Haridasan 7283).

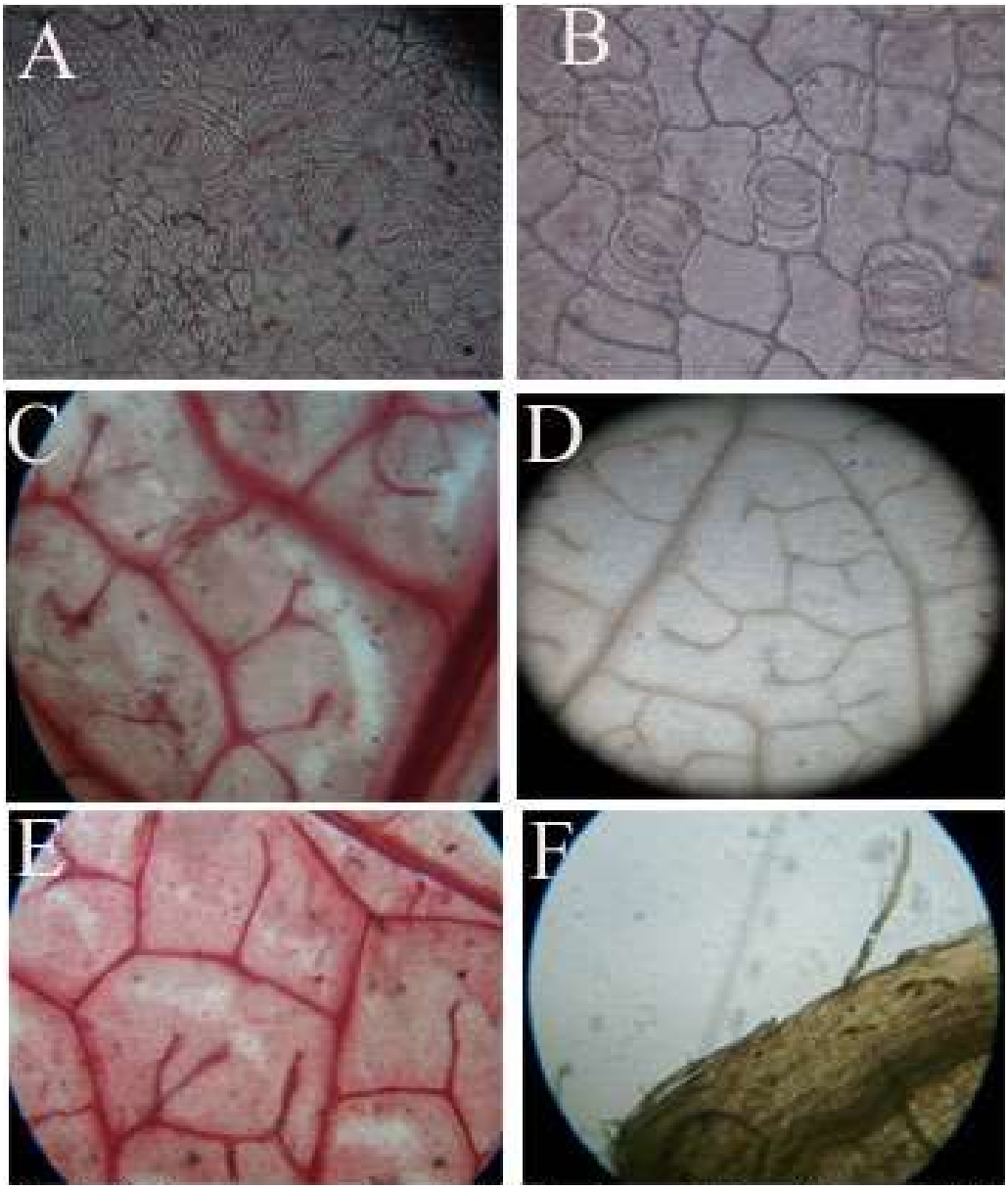


Plate 9. *Lyonia macrocalyx* **(** J.Anthony) Airy Shaw: A, B,: leaf stomata (10X, 40X); C **-** E: leaf vein islets apex, middle & base (10X, 5X, 10X), Haridasan 7283, APFH.