A revision of Middletonia (Gesneriaceae) in Thailand

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ABSTRACT

The genus *Middletonia* in Thailand is revised. We recognise four species, including the newly described *Middletonia glebosa* C.Puglisi and the resurrected *M. reticulata* (Barnett) C.Puglisi. A key to the species, full descriptions and proposed conservation assessments are provided.

KEYWORDS: taxonomy, new species, Flora of Thailand.

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INTRODUCTION

Middletonia C.Puglisi belongs to subtribe Loxocarpinae, tribe Trichosporeae (Weber et al., 2013) of the family Gesneriaceae. Subtribe Loxocarpinae includes all species of Southeast Asian Gesneriaceae with twisted fruits (plus many species with straight fruits). The genus was segregated from Paraboea (C.B.Clarke) Ridl. by Puglisi et al. (2016) as a result of a study of the Loxocarpinae which showed that the species now in Middletonia did not form a monophyletic group with the rest of Paraboea. The segregation from Paraboea is supported by both molecular data and the morphological characters discussed below. This group of species had already been noted by Xu et al. (2008) as being distinctive although no formal infrageneric classification was proposed.

Like *Paraboea*, *Middletonia* presents a matted, interwoven indumentum of long and fine hairs on the abaxial side of the leaf, a flat-faced corolla and a capsular fruit. The characters that separate *Middletonia* from *Paraboea* are the erect anthers (borne at a right angle in *Paraboea*) and the minutely glandular indumentum on the anthers and ovary. Other characters that can aid in the distinction of *Middletonia* from *Paraboea* are the reticulate tertiary venation, which is visible at least by the leaf margin, and the considerably smaller corolla and shorter fruit compared to most species of *Paraboea*.

MATERIALS AND METHODS

The investigation is based on an examination of specimens from the herbaria A, AAU, ABD, BK, BKF, BM, E, K, L, MO, P, SING (herbarium codes from Thiers, continuously updated), and on the living and spirit collections of the Royal Botanic Garden Edinburgh and Singapore Botanic Gardens. All the specimens cited have been seen unless otherwise stated. Measurements of the fruit and the vegetative parts were taken from dry specimens, while flowers were measured from fresh, rehydrated or preserved samples. Accuracy of the fine measurements should be estimated at 0.05 mm.

MIDDLETONIA

C.Puglisi, Taxon 65: 286. 2016. Type species: *Middletonia multiflora* (R.Br.) C.Puglisi.— *Boea* sect. *Caulescentes* Fritsch in Engl. & Prantl, Nat. Pflanzenfam. 4(3B): 150. 1894. Type species: *Boea multiflora* R.Br., lectotype designated by Burtt (1954: 194).

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Lithophytic herbs with short stems. *Leaves* petiolate, opposite, rarely alternate or subopposite at the base of the stem and then becoming opposite apically. *Lamina* elliptic to obovate, often with unequal base, abaxial surface with a matted eglandular indumentum and visible tertiary venation. *Inflorescence* axillary and cymose; bracts inconspicuous; flowers 5-merous. *Calyx* with sepals divided to base. *Corolla* with short tube and spreading limb, slightly bilabiate, with 2 lobes on the upper lip and

three on the lower. *Stamens* 2; filaments straight; anthers erect, dorsifixed, with a minute glandular indumentum; staminodes 2, often reduced. *Ovary* ovoid, minutely glandular; ovules many; stigma capitate. *Fruit* a straight or twisted capsule, bivalved, orthocarpic and longitudinally dehiscing. *Seeds* minute and elliptic.

Five species, found in India, Bangladesh, Bhutan, Myanmar, China, Thailand, Laos, Cambodia, Vietnam and Malaysia. Four in Thailand.

KEY TO THE THAI SPECIES

1. Inflorescence longer than the subtending leaf; fruit strongly twisted	
2. Leaves pubescent above (hispid and glandular)	1. M. evrardii
2. Leaves glabrous or glabrescent above	4. M. reticulata
1. Inflorescence shorter than or as long as the subtending leaf; fruit straight or slightly twisted	
3. Fruit slightly twisted, valves smooth; habit distinctly caulescent	3. M. regularis
3. Fruit straight, valves lumpy; habit shortly caulescent	2. M. glebosa

1. Middletonia evrardii (Pellegr.) C.Puglisi, Taxon 65: 286. 2016.—*Boea evrardii* Pellegr. in Lecomte, Fl. Indo-Chine 4: 550. 1930.—*Paraboea evrardii* (Pellegr.) B.L.Burtt, Notes Roy. Bot. Gard. Edinburgh 41: 428. 1984. Type: Vietnam, Lam Dong, Pongour pres Dijring [Di Linh], 24 Aug. 1924, *Evrard 1177* (lectotype **P** [P00622885], designated by Puglisi *et al.* (2016: 287); isolectotype **P** [P00556499]).

Shortly caulescent herb. Stem 3-7 cm long, woody, densely covered in a thick layer of matted long, brown hairs. Leaves congested or with internodes to 0.5 cm long, opposite or subopposite; petioles 0.5-4 cm long, tomentose as on stem, the indumentum slightly less appressed; lamina $2.5-8 \times 1.2-4$ cm, 2–2.6 times as long as wide, elliptic; apex acute; base shortly attenuate; margin serrulate to crenulate; adaxial surface hispid with scattered uniseriate multicellular hairs, abaxial surface tomentose with brown hairs, appressed along the venation, looser over the rest of the lamina; 8-9 pairs of secondary veins, tertiary venation seldom visible on the adaxial surface, evident on the abaxial. Inflorescence an axillary, compound cyme, tomentose and glandular, longer than the subtending leaf, many-flowered; peduncles 1–1.3 cm long; bracts $4.5-5 \times 0.7-0.8$ mm, ligulate, tomentose outside, glandular inside; pedicels 3-7 mm long, delicate. Calyx divided to base; lobes triangular, ca 1.4×0.4 mm, apex broadly acute, densely glandular and sparsely tomentose outside, sparsely glandular inside. Corolla almost flat-faced, white and densely glandular inside; otherwise poorly known (see note below). *Stamens* exserted, inserted ca 1 mm from corolla base; filaments ca 0.8 mm long, straight; anthers free, ca 1.5×0.8 mm, thecae parallel, glabrous and dehiscing longitudinally; staminodes not seen. *Ovary* ca 2 mm long, ca 0.7 mm diameter, markedly distinct from style and covered in glands; style 3.5-4 mm long, ca 0.1 mm diameter, glabrous, straight; stigma capitate, ca 0.2 mm long. *Capsule* to 16 mm long, densely covered in glands, twisted. *Seeds* not seen.

Thailand.— EASTERN: Nakhon Ratchasima [Khao Yai National Park, along River Huyta Kon, 600-800 m, 19 July 1973, *Murata et al. T-16373* (**BKF**)]; CENTRAL: Nakhon Nayok [Nang Rong Waterfall, ca 100 m, 29 July 1959, *Smitinand & Floto 6106* (**BKF**, **E**)].

Distribution.- Lao PDR, Vietnam.

Ecology.— Evergreen forest.

Proposed IUCN conservation assessment.— Endangered (EN B2ab(iii)). In Thailand this species is only known from two collections in the Khao Yai area, of which the most recent was made in 1973 despite Khao Yai being frequently explored. It is also known from a few collections in Lao PDR and Vietnam over a wide distribution. Although its EOO is large the few and localised collections give it an AOO in the Endangered range. It also has a fragmented distribution, not all populations are in protected areas and those that are may be subject to pressure from tourists.

Note.— The material from Thailand differs slightly from the Vietnamese material, especially in the leaf shape which is more elongated and with an acute apex in the Thai specimens. As there are rather few specimens from throughout the distribution range, with no floral material from Thailand available for dissection, further collections are necessary to assess whether the Thai and Vietnamese plants belong to one or more species.

2. Middletonia glebosa C.Puglisi, sp. nov.

Similar to *Middletonia regularis* in having the inflorescences shorter than the leaves and to *Middletonia reticulata* in the shortly caulescent habit. Differs from both in having a dense indumentum on the adaxial side of the leaf, with glands and eglandular hispid hairs (upper leaf surface glabrescent or minutely glandular in *Middletonia regularis*, glabrescent in *M. reticulata*), and in the short, non-twisted, lumpy and irregular fruit (longer, weakly to strongly twisted, smooth and regular in *M. regularis* and *M. reticulata*). Type: Thailand, Sukhothai, Khiri Mat, Ram Kham Haeng National Park, Khao Luang, alt. 520 m, 11 Oct. 2012, fr., *Middleton, Karaket, Suddee & Triboun 5559* (holotype E [E00547434]; isotypes **BK**, **BKF** [SN201922]). Fig. 1A–B.

Stoloniferous, very shortly caulescent herb. Stem to 2 cm long, densely covered in a thick layer of matted long, brownish hairs. Leaves congested, opposite; petioles 1-4.5 cm long, tomentose as on stem; lamina $5-11 \times 2-4.5$ cm, 2-3 times as long as wide, elliptic, with a rugose surface; apex broadly acute to obtuse; base seemingly attenuate in herbarium specimens, rounded to subcordate in living plants; margin more or less crenate and undulate, sometimes appearing irregularly denticulate; adaxial surface mid-green and hispid with scattered uniseriate multicellular hairs and glands; abaxial surface pale green, with glands throughout and long, thin hairs which entirely cover the surface but become restricted to the veins in mature leaves; 6-7 pairs of secondary veins; tertiary venation seldom visible on the adaxial surface, more evident on the abaxial. Inflorescence an axillary, compound cyme, tomentose, shorter than the leaf subtending it, many-flowered; peduncles 3-6 cm long; bracts to ca 3 mm long, narrow, hirsute on the abaxial surface, glabrous or glandular on the adaxial; pedicels white, 1-7 mm long, delicate and slightly pendulous, glandular and less tomentose than the lower axes. Calyx divided to base; lobes white, becoming green towards the apex, narrowly ovate, 1-2 mm long, apex obtuse, glabrous or with a scattered glandular and eglandular indumentum. Corolla almost flat-faced, slightly bilabiate, white and densely glandular outside; tube ca 1 mm long; lip lengths not measured; upper lobes elliptic, ca 4 $\times 2.5$ mm; lower lobes broader, ca 4 $\times 3$ mm. *Stamens* inserted ca 0.5 mm from corolla base; filaments ca 2 mm long, geniculate in the middle and with a small hump below the attachment point of the anther; anthers pale yellow and free, ca 1×2 mm, thecae subparallel and reniform, glandular towards the middle line and dehiscing longitudinally; staminodes absent. Ovary ca 1 mm long, markedly distinct from style and covered in minute glands conferring it a pale yellow-green colour; style ca 3 mm long, glabrous, more or less straight; stigma inconspicuous, slightly bilabiate, with lower lip slightly bifid and a little longer than the upper. Capsule green, to 6 mm long, densely covered in glands and with a lumpy, uneven surface, not twisted, sometimes curved. Seeds elliptic, ca 0.2×0.1 mm.

Thailand.— NORTHERN: Sukhothai [Khiri Mat, Ram Kham Haeng National Park, Khao Luang, alt. 520 m, 11 Oct. 2012, fr., *Middleton et al. 5559* (**BK**, **BKF**, **E**); ibid. 450 m, 24 Oct. 2014, *Middleton et al. 5848* (**SING**)].

Distribution.— Cambodia [Stung Treng, 1866–1868, *Thorel 2268* (**P**×2)].

Ecology.— Mixed deciduous forest with bamboo on granite bedrock.

Etymology.— The specific epithet is the Latin adjective glebosus, -a, -um, which means "lumpy" and refers to the peculiar surface of the capsule.

Proposed IUCN conservation assessment.— Endangered (EN B2ab(iii)). This species is only known from two localities, Ram Kham Haeng National Park in Thailand and Stung Treng in Cambodia. There have been no collections from the Cambodian site since the nineteenth century and the area is poorly collected overall. Although the potential EOO is large, given the wide separation of the two known localities, the AOO is less than 500 km², even considering the possibility of a much more widespread distribution in the National Park than has been collected. Although the status of the Cambodian population(s) needs to be verified, in Ram Kham Haeng National Park this plant has been observed growing in areas subject to considerable tourist pressure.

Notes.— In Ram Kham Haeng National Park this species grows in a mixed population with *Middletonia reticulata*. The two are readily told apart in the field by *Middletonia glebosa* having softer, paler leaves, a shorter inflorescence, generally slightly smaller flowers, and the curious short, straight, lumpy fruit. The species is known only from Ram Kham Haeng National Park and a single collection made around 150 years ago in Cambodia. The Cambodian collection, placed in *Paraboea regularis* by Xu *et al.* (2008), is only in fruit so the identification will need to be verified with flowering material. However, the fruit of this species is rather distinctive.

3. Middletonia regularis (Ridl.) C.Puglisi, Taxon 65: 287. 2016.— *Didymocarpus regularis* Ridl., J. Linn. Soc., Bot. 32: 515. 1896.— *Paraboea regularis* (Ridl.) Ridl., J. Straits Branch Roy. Asiat. Soc. 44: 68. 1905. Type: Malaysia, Kedah, Langkawi, 1893, cult. Hort. Bot. Sing., *Curtis s.n.* (lectotype **SING** [0042998], designated by Burtt (1984: 435); isolectotype **E** [E00451499]).

— Paraboea monticola Triboun & D.J.Middleton, Gard. Bull. Singapore 64: 346. 2012.— Middletonia monticola (Triboun & D.J.Middleton) C.Puglisi, Taxon 65: 287. 2016. Type: Thailand, Phangnga, Tai Toy, Triboun 3662 (holotype **BK**; isotype **E** [E00564788]).

Paraboea multiflora var. caulescens Z.R.Xu & B.L.Burtt, Edinburgh J. Bot. 48: 7. 1991.
Middletonia multiflora var. caulescens (Z.R.Xu & B.L.Burtt) C.Puglisi, Taxon 65: 287. 2016. Type: Thailand, [Kanchanaburi], Kwae Noi Basin, 150 m, 13 June 1946, G. Den Hoed Exp. No. 946 (holotype L [L0003189]).

Rhizomatous, caulescent herb. *Stem* 10–40 cm tall. *Leaves* alternate along the stem, becoming opposite and sometimes tightly congested apically; petioles 0.5-7 cm long, densely tomentose; lamina $2.9-10.5 \times 1.5-5.5$ cm, 1.6-3.2 times as long as wide, ovate, obovate or elliptic; apex acute; base

often unequal, acute or shortly so; margin irregularly serrate to crenulate; adaxial surface dark green and minutely glandular or glabrescent; abaxial surface brownish green, densely tomentose with fine and long hairs which are rusty brown, woolly and caducous along the veins and pale brown and sparse over the rest of the lamina; 5-10 pairs of secondary veins; tertiary venation prominent on both sides, on dry specimens seldom visible on the adaxial surface, reticulate and clearly visible on the abaxial. Inflorescence an axillary, compound cyme, with the same loose indumentum of the stems and lower leaf surface (sometimes reduced in mature plants or missing in old specimens), as long as or shorter than the subtending leaf, 4 to many-flowered; peduncles 1–4.5 cm long; bracts reduced, $1-3.5 \times 0.2-1.5$ mm, ligulate to lanceolate, tomentose outside, sparsely so or glabrescent inside; pedicels 10-15 mm long, tomentose. Calyx greenish brown; tube 0.2-0.5 mm long; lobes ligulate to lanceolate, 1-2 mm long, apex broadly acute to obtuse, tomentose outside, sparsely glandular inside. Corolla almost flat-faced, slightly bilabiate, white and glabrous outside and inside; tube 1.5-1.8 mm; upper lip 2.2-2.7 mm long, lower lip 3.2–3.3 mm long; lobes spreading, upper lobes elliptic, ca 3×3.1 mm, lateral lobes ca 2.8×2.4 mm, lower lobe ca 2.7 × 3.3 mm. Stamens inserted ca 0.7 mm from corolla base; filaments white, 2-2.5 mm long, straight or geniculate, with minute glands towards the apex; anthers pale orange and free, ca $2 \times 1.6-2$ mm, glandular dorsally and dehiscing longitudinally; staminodes 2, 1.5-6 mm long, arising ca 0.4 mm from corolla base. Ovary pale yellow to light green, 1.3-2.1 long, 0.6-0.7 mm diameter, distinct from the style and covered in minuscule glands; style 2.5-3.8 mm long, white, glabrous, straight; stigma green at tip, capitate. Capsule pale green, 5–10 mm long, densely covered in glands, straight or slightly twisted. Seeds elliptic, ca $0.2 \times$ 0.1 mm.

Thailand.— PENINSULA: Surat Thani [Phanom, Khlong Phanom National Park, trail from Park headquarters, 200 m, 7 Sept. 2008, *Middleton et al.* 4363 (**BKF**, **E**); Phanom, Khao Sok National Park, 100–200 m altitude, 12 Dec. 1979, *Shimizu et al. T-27069* (**BKF**, **L**); Khiri Rat Nikhom, Khao Phra Rahoo, 50–100 m, 27 Aug. 1982, *Shimizu et al. T-28871* (**BKF**); Phanom, Khlong Sok, 23 Sept. 2010, *Triboun 4577* (**E**)]. Phangnga [Mueang Phangnga, 25 Sept. 2006, *Triboun 3662* (**BK**, **E**); Pulau Panji (Koh Panyee), 2 Dec. 1928, *Haniff & Nur 4013* (**K**, **SING**); Thap Put, Khao Tao, 10 m, 21 June 2006, *Williams et al. 2036* (**A**, **BKF**, **E**)]; SOUTH-WESTERN: Kanchanaburi [Kwae Noi Basin, near Wangka, 150 m, 13 June 1946, *Den Hoed 946* (**L**)].

Distribution.— Malaysia.

Ecology.— Lowland evergreen forest on limestone soils.

Proposed IUCN conservation assessment.— Least Concern (LC). This species is relatively widespread with an EOO of around 40,000 km², well beyond the threshold for a threat category. Its status should, however, be monitored as not all of the limestone localities where it is found are in protected areas.

Note.— The protologue of *Paraboea monticola* suggests the flowers to be much larger: "lobes ... $6-9 \times c$. 6.5 mm". However, none of the material available for this study had lobes longer than 3.3 mm.

4. Middletonia reticulata (Barnett) C.Puglisi, **comb. nov.**— *Boea reticulata* Barnett, Nat. Hist. Bull. Siam Soc. 20: 20. 1961. Type: Thailand, Chiang Mai, Mae Wang, *Kerr 6356* (lectotype **K**, designated by Barnett (1961: 256); isolectotypes **ABD**, **BM**). Fig. 1C–F.

Rhizomatous, very shortly caulescent herb. Stem to 1.5 cm. Leaves tightly congested, seemingly opposite; petioles 0.3–9 cm long, densely tomentose; lamina 2–9.8 × 1.9–5 cm, 1.2–2.3 times as long as wide, ovate, obovate or elliptic, often irregular; apex obtuse to rounded; base unequal, shortly attenuate and forming small auricles at the insertion on the upper surface; margin irregularly crenate to crenulate; adaxial surface dark green and glabrescent, abaxial surface brown, densely tomentose with hairs that are somewhat woolly, dark brown along the veins, pale brown over the rest of the lamina and which can be removed very easily (the base of the hair is very fine); 5–9 pairs of secondary veins, secondary venation deeply sunk on the upper surface of the leaf, slightly raised beneath; tertiary venation sometimes visible on the adaxial surface on dry specimens, clearly visible and reticulate on the abaxial. Inflorescence an axillary, compound cyme, with a loose reddish brown tomentum that becomes patchy on dry specimens, longer than the subtending leaf, dense and many-flowered; peduncles 4.5-17 cm long; bracts $1.6-3 \times 0.2-0.5$ mm, ligulate, tomentose; pedicels 1-12 mm long, tomentose. Calyx brown, tube 0.2-0.7 mm long; lobes triangular, 1-2.5 mm long, apex broadly acute, tomentose outside, sparsely glandular inside. Corolla almost flat-faced, slightly bilabiate, white and glabrous outside and inside; tube 1-1.8 mm; upper lip 0.1-4.5 mm, lower lip 3.2–6.9 mm; upper lobes elliptic, $3-5 \times 2.5-4.7$ mm, lateral lobes $2.8-4.7 \times 2.4-5.2$ mm, lower lobe $2.7-4.5 \times 3-4.7$ mm. Stamens inserted at 0.3-0.7mm from corolla base; filaments white, 1.5-3 mm long, straight, with minute glands towards the apex; anthers pale orange, free, $1.3-2 \times 1.6-1.8$ mm wide, apically confluent, glandular dorsally and dehiscing longitudinally; staminodes absent or ca 2.6 mm long, arising ca 0.4 mm from corolla base. Ovary pale yellow, 2-2.5 mm long, 0.6-1 mm diameter, markedly distinct from style and covered in minuscule glands; style 2.5-3.8 mm long, glabrous, straight; stigma capitate and densely papillose. Capsule pale green, 7-14 mm long, densely covered in glands, twisted. Seeds elliptic, $0.2-0.3 \times 0.1-0.2$ mm.

Thailand.— NORTHERN: Mae Hong Son [Payap, West of Muang Hot, 350 m, 23 Sept. 1958, Sørensen et al. 5201 (BKF)]; Chiang Mai [Mae Wang, ca 400 m, 19 July 1922, Kerr 6356 (ABD, **BM**, **K**); Chom Thong, Doi Inthanon National Park, Mae Ya Falls, 550 m, 11 Sept. 1994, Palee 257 (A, BKF); Doi Inthanon, Mae Klang waterfall, 310-400 m, 18 July 1988, Tamura T-60094 (BKF); ibid., Takahashi T-62973 (BKF); ibid., 400 m, 19 Sept. 2008, Middleton et al. 4516 (BKF, E); ibid., 10 Nov. 1965, Sangkhachand 25 (BK); ibid., 25 Nov. 1965, Phusomsaeng 5 (BKF); ibid., 430 m, 3 Nov. 1967, Burtt 5612 (E); Doi Inthanon National Park, 7 km below Wachiratan Waterfall, 400 m, 19 Aug. 2004, Nielsen et al. 1783 (BKF); Doi Inthanon, along Mae Klang river, 450 m, 31 July 1988, Fukuoka T-62387 (BKF, L); Hot, Op Luang Nature Park, Doi Op Luang, Mae Jam river, 550 m, 23 Oct. 1987, Maxwell 87-1281 (BKF); Hot, Op Luang Gorge, 325 m, 24 Sept. 1989, Maxwell 89-1134 (A, MO); Hot, Ob Luang National Park, 9 Aug. 2014, Prommanut et al. P528 (BK)]; Lamphun [Li, Mae Ping, Ko Luang waterfall, 300-400 m, 27 Sept. 2006, Dongkumfu 2 (E); Pa Sang, Erawan Cave, 575 m, 5 Dec. 2004,



Figure 1. *Middletonia gebosa* C.Puglisi: A. Habit, showing young fruit; B. Flowers; *M. reticulata* (Barnett) C.Puglisi: C. Habitat with fruiting plants; D. Habit; E. Flowers; F. Fruit. Photographs by Preecha Karaket (A, C–E) and David Middleton (B, F).

Maxwell 04-772 (**BKF**)]; Lampang [Mae Ngow, 420 m, 25 Aug. 1922, *Winit 740* (**BM**)]; Phrae [Song, Mae Yom National Park, Mae Den, Sahiab, Yom river, 225 m, 12 Nov. 1991, *Maxwell 91-1028* (**AAU**, **E**)]; Sukhothai [Khiri Mat, Ram Kham Haeng National Park, Khao Luang, 445 m, 11 Aug. 2012, *Middleton et al. 5557* (**BKF**, **E**, **SING**); ibid., 518 m, 24 Oct. 2014, *Middleton et al. 5844* (**BKF**, **SING**); ibid., 420 m, 24 Oct. 2014, *Middleton et al. 5849* (**BKF**, **E**, **SING**); ibid., 1 Dec. 1987, *Paisooksantivatana & Sangkhachand y2186-87* (**BK**].

Ecology.— Mixed deciduous forest on granite bedrock.

Distribution.— Currently endemic to Thailand but possibly also occurring in Lao PDR and Myanmar.

Proposed IUCN conservation assessment.— Near Threatened (NT). This species has an EOO of more than 20,000 km² but occurs in many areas subject to tourist pressure and microclimate changes due to surrounding agricultural practices.

Note.— This species is resurrected from synonymy of *Middletonia multiflora* (R.Br.) C.Puglisi from which it differs in the distinctively reticulate venation pattern covered in loose brown indumentum on the lower surface of the leaves, and in the auriculate leaf base. *Middletonia multiflora* has a denser indumentum on the leaf and the tertiary venation is less densely reticulate and less visible altogether. *Middletonia multiflora* is widespread along the northern distribution of the genus (from India to Vietnam), but none of the material from Thailand studied can be attributed to *M. multiflora*.

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