

CHRISTA BEURTON

Angostura ossana* (Rutaceae), a component of the Cuban flora*Abstract**

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The enigmatic *Galipea ossana*, described in 1822 and known from the type specimen said to originate from Cuba and only one other collection made before 1841 in a locality called San Diego, is reconsidered. Light microscopic and scanning electron microscopic examination of flower structure, fruits and pollen grains of the scarce material available show that the species actually is the only representative of the genus *Angostura* on the Antilles and a W Cuban endemic, closely related to *A. trifoliata* from Venezuela. The combination *A. ossana* is validated and the diagnostic features of this species and of *A. trifoliata* are illustrated.

Introduction and taxonomic history

Galipea ossana DC. is listed in the Flora de Cuba as a very rare W Cuban endemic (León & Alain 1951: 378). It was originally described in a detailed analysis by Augustin Pyramus de Candolle in 1822 on the basis of a specimen received from Cuba, and he also gave an illustration of this specimen (Candolle 1822: t. 10, see Fig. 1). The specimen, preserved in the Candolle Herbarium (G-DC), includes a twig with leaves, an inflorescence with flower buds and an infructescence with the fruits dehiscent, and is still in good condition. Candolle received this specimen from La Ossa, at that time director of (the old) Botanical Garden La Habana, Cuba. It is supposed to have been collected in the vicinity of La Habana. Fruiting material of the same species was collected at another time in a locality cited as “San Diego” (Richard 1841: 323) and presumably referable to San Diego de los Baños, in the province Pinar del Río, W Cuba. A corresponding specimen collected in Cuba from the herbarium of Richard is preserved in the Muséum National d’Histoire Naturelle Paris (P) (Fig. 2).

Candolle included the new species in the genus *Galipea* Aubl. This genus belongs to the neotropical subtribe *Galipeinae*, one of the two subtribes of the *Galipeae* Kallunki (previously

known as *Cusparieae* DC., an illegitimate name; see Kallunki & Pirani 1998) of the *Rutaceae* subfamily *Rutoideae*. The *Galipeinae* comprise 27 genera, which are mostly unispecific or with less than 10 species, of trees or shrubs with greenish, whitish or pink flowers. They differ from all other *Rutaceae* by a tendency towards noticeably zygomorphic flowers (mainly due to the position of elements of corolla and androecium) in combination with a tubular corolla (petals connate or coherent), a filament tube (adherent or adnate to the corolla tube), a reduction in number of fertile stamens from five to two combined with a transformation of stamens into staminodes (the latter with free apical lobes), and frequently with appendages at the base of the fertile anthers (Kallunki 1994, 1998, Kallunki & Pirani 1998, Morton & Kallunki 1993).

Engler (1874, 1896, 1931) transferred *Galipea ossana*, without having seen any specimen, because of its reported choricarpous gynoecium into the genus *Raputia* Aubl. Kallunki (1994) again excluded it from that genus, but it does not belong to *Galipea* either because that genus is characterised by syncarpous gynoecium and fruit, fertile anthers that are sterile at base above the point of attachment to the filament, and connate appendages at the base of these anthers (Table 1). With the exception of the alternate leaves, all traits characteristic of *Galipea* differ from Candolle's description of *Galipea ossana*. This was the cause of the doubts about this material belonging to *Galipea* Aubl.

Like other authors (Alain 1958: 37), I also had doubt about the Cuban origin of La Ossa's plant. However, that it was collected in Cuba is supported by a second finding of the species at a different location in Cuba. Unfortunately both locations were documented incompletely, and the species has not been collected again.

Material and methods

Specimens (cited below and see also Fig. 2 & 8) from the herbaria B-W, G-DC and P (abbreviations according to Holmgren & al. 1990) were analysed.

For studies of flower morphology including measurements the material was rehydrated. For the analysis of pollen grains the material was prepared according to the acetolysis method of Erdtman (1960).

Angostura ossana (DC.) Beurton, comb. nova

≡ *Galipea ossana* DC. in Mem. Mus. Hist. Nat. 9: 149. 1822 ≡ *Raputia ossana* (DC.) Engl. in Mart., Fl. Bras. 12(2): 104. 1874. – Holotype: Cuba, “ad Habana”, [c. 1820], *La Ossa* (G-DC) – Fig. 1-2, 3A-B, 4, 6).

Description (abbreviations in brackets: LOs: specimen collected in the vicinity of La Habana; LSa: specimen collected in the vicinity of San Diego; DC: according to the description of De Candolle; !: contrary to the description of Candolle). – *Branches* terete [LOs] or semiterete [LSa], glabrous [LOs] to white-tomentose of short, stellate hairs [LSa]. *Leaves* trifoliolate, petiole 5.3-9.1 cm long, semiterete; petiole, petiolules, rachis and leaflets (especially abaxially) from nearly glabrous [LOs] to white-tomentose of short, stellate hairs [LSa]. *Leaflets* elliptic, apex acuminate, margins entire, base attenuate; lateral leaflets subsessile to shortly petiolulate, up to 13.3 × 6 cm, slightly asymmetric; terminal leaflet nearly equal, shortly petiolulate, up to 14.5 × 6 cm, frequently somewhat larger than the lateral ones; chartaceous, gland-dotted. *Thyrse* long-stalked, multiflorous, partly corymboid ([LSa]; Fig. 1b-c, 2); peduncle about 14-24 cm long, glabrous [LOs] to densely stellate hairy [LSa], flowering zone about 5-11 cm long; peduncles of the partial inflorescences (cymes) about 1-3.5 cm long, lower partial inflorescences longer than the upper ones. *Flower buds* 8-12 mm long; pedicel 0.8-4 cm long, densely stellate hairy. *Calyx* c. 3.3 mm long, abaxially densely stellate hairy and glandular, the lobes 2-2.4 mm long, elliptic, the apex acute. *Corolla (buds)* 6-10 mm long, petals either connate toward base [DC] or coherent [not ascertainable by the author], older buds slightly recurved, indument on abaxial surface



Fig. 1. *Angostura ossana* (taken from Candolle 1822: t. 10) – a: twig with leaves; b: inflorescence; c: infructescence; d-f: flower (d-e: male phase; f: female phase), d: early anthesis just beginning, e: androecium, f: calyx and gynoecium; g-m (g-k, m: adaxial view): dehiscent follicle and its components, g: follicle with exocarp, endocarp and placental endocarp, h: exocarp, i: endocarp with remaining placental endocarp and seed, k: endocarp, l: seed, m: placental endocarp; n: base of a fruit cluster with persistent sepals and disc (with 5 bidentate lobes; follicles fallen off).



Fig. 2. *Angostura ossana* – Cuba without date, *Ramon de la Sagra* (P).

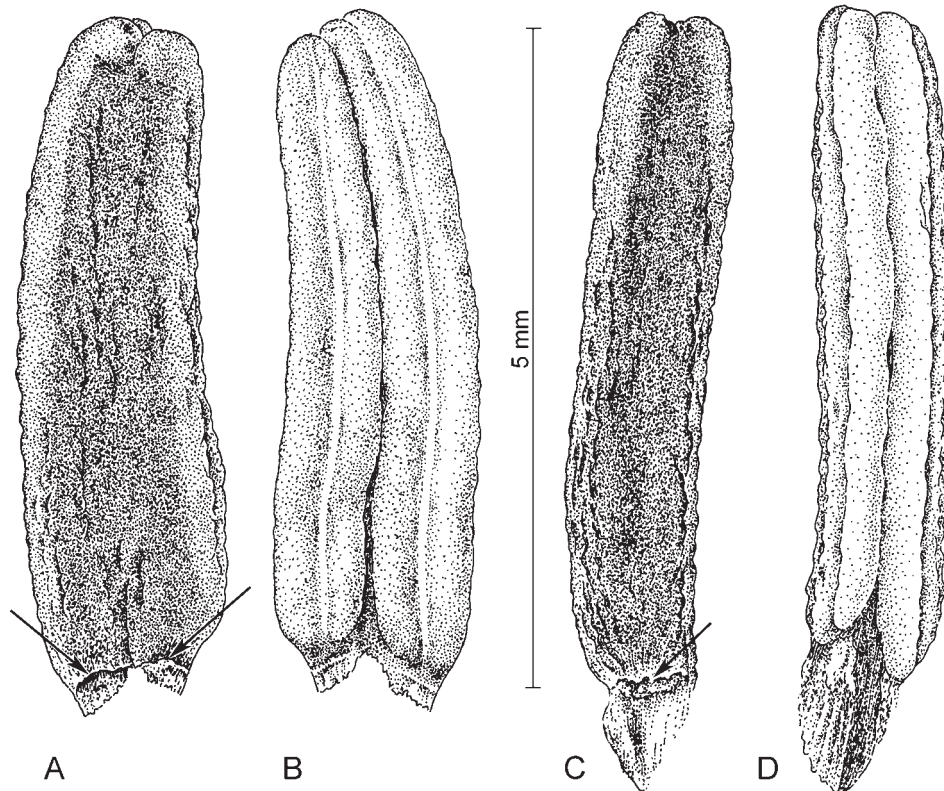


Fig. 3. Fertile anthers with appendages – A-B: *Angostura ossana* (holotype of *Galipea ossana*, G-DC); C-D: *Angostura trifoliata* (holotype of *Bonplandia trifoliata*, B-W 4812). – A, C: abaxial view; B, D: adaxial view; arrows mark the junction between anther and filament.

densely brightly silky, hairs stellate with long branches directed toward base; indument on adaxial surface less dense, hairs stellate with longer or shorter branches radiating in all directions (Fig. 4A-B). *Androecium* of 2 fertile stamens and 4 [!]-5 [DC] staminodes; filaments (except the bases of the fertile stamens) flattened, free from each other but coherent by curled hairs on adjacent margins (Fig. 4D) and adherent by abaxial hairs to the corolla tube as a short pseudo-tube; free apices of the staminodes subulate, 1.2-2.2 mm (in bud) and glandular; fertile anthers (in bud) about 5×1.4 mm, free from each other, either bearing tiny [!] appendages at the base or not [DC], broadly attached to filament (Fig. 3A-B, arrow). *Pollen grains* (Fig. 6A-D) with 5 short colpae, suboblate spheroidal, large, polar axis $49.5\text{-}56.1$ ($\bar{n} = 52.2$) μm , equatorial axis $59.4\text{-}62.7$ ($\bar{n} = 61.4$) μm , P/E-index 0.85 ($n = 5$), exine baculate and with small rod-like structures between the bacula. *Disc* cupular, deeply 5-lobate, each lobe bidentate (Fig. 1n). *Gynoecium* of 5 choricarpous carpels (Fig. 4C); carpels anacrostylous, in bud about 0.3 mm long, glabrous when young, later sericeous; styles 5, united at base into a common tube (about 0.5 mm diameter); stigma in bud nearly 5-partite-capitate, style tube and stigma enclosed between the fertile anthers. *Ovules* 2, superposed. *Fruit* of 5 modified follicles free or (sub)choricarpous at base; follicles ovoid-angular, $10\text{-}12 \times 6$ mm, partly densely blackish glandular [LSa] and short stellate hairy, apex beaked; endocarp elastic, horny, orange, placental endocarp small, triangular, rounded at base and pointed at apex of follicle, membranaceous (Fig. 1c, g-k, m, and 2). *Seeds* globose (Fig. 1l), smooth; testa thin, fragile, black [DC, n.v.].

Table 1. Character states in *Raputia*, *Galipea* and *Angostura* s.str. (compiled partly from data provided by Kallunki 1994, Kallunki & Pirani 1998, Morton & Kallunki 1993).

Character		<i>Raputia</i>	<i>Galipea</i>	<i>Angostura</i> (S & Central America)	<i>Angostura ossana</i>
Leaves	opposite	●			
	alternate		●	●	●
Hairs (calyx, corolla)	simple	●*	●		
	stellate			●	●
Gynoecium	syncarpous		●		
	choricarpous	●		●	●
Fertile anthers	free			●	●
	connate	●	●**		
	completely fertile	●		●	●
	sterile at the base		●		
Appendages	present	●	●	●	●
	absent				●
Pollen	reticulate	●	●		
	baculate			●	●

* glabrous to densely strigulose (Kallunki 1994); ** by their sterile bases and appendages

Distribution. – W Cuba (probably vicinity of La Habana and vicinity of San Diego de los Baños). Extinct?

Further specimen seen. – Cuba [“circa San Diego, fructos maturos ferebat abeunte martio” (Richard 1841: 323)], *Ramon de la Sagra* [or *José Maria Valenzuela*, according to Urban 1898: 143] (P [ex herb. Richard]).

Relationship

The plants collected in the vicinity of San Diego are much more hairy than those collected in the vicinity of La Habana. Their infructescences are partly nearly corymboid, but clearly belong to the same species (see also Richard 1841: 323). The flowers, present only in the type collection, show a specific combination of characters found in the *Galipeinae* only in *Angostura* Roem. & Schult. s.str. (Table 1; Kallunki & Pirani 1998). This is confirmed by a comparative study of the holotype [Venezuela, Golfe Sta., Fé, près Cumaná, 1800, *Humboldt & Bonpland* (B-W 4812), see Fig. 8] of the Venezuelan *Angostura trifoliata* (Willd.) T. S. Elias. The ascertained character states are the following: choricarpous carpels (Fig. 4C), (sub)choricarpous fruitlets, the presence of two fertile stamens per flower with completely fertile anthers, the presence of small appendages at the base of these anthers (Fig. 3), the presence of stellate (not simple) hairs abaxially on calyx and corolla (Fig. 4A-B, 5A), and the baculate pollen type (Fig. 6-7, Table 1).

Among the seven hitherto known *Angostura* species only the Venezuelan *A. trifoliata* (Fig. 8, Kallunki & Pirani 1998) shows some similarity with the Cuban material. Both species possess trifoliolate leaves, unwinged petioles, relatively long secondary inflorescence axis and relatively long anthers. In both species the filaments are adherent rather than adnate to the corolla tube (Fig. 5C-D). However, in view of the differences in the morphology of the inflorescence and the androecium as well as in pollen size, the Cuban taxon is specifically distinct from *A. trifoliata*: in *A. ossana* the lower cymes are clearly longer than the upper ones, not only slightly longer (Fig. 1, 2, 8); the appendages at the base of the fertile anthers are rudimentary or missing (Fig. 3A-B and 3C-D); the staminodes are shorter and not gland-tipped (Fig. 4D, 5B); and the pollen grains are smaller (Fig. 6A, 7A; see also Morton & Kallunki 1993).

In contrast to the present finding, Candolle observed no appendages at the base of the fertile anthers of *Angostura* (*Galipea*) *ossana*. The appendages are indeed rudimentary, and because Candolle's investigation is generally very precise, it may be assumed that appendages are not always formed.

Given that *A. ossana* and *A. trifoliata* are close relatives, the observed W Cuban-S American pattern of distribution is highly interesting, since it is also known within the *Rutaceae* from the species pair *Zanthoxylum ekmanii* and *Z. sobreviolae* (Beurton, in prep.).

Is *Angostura ossana* still extant in Cuba?

The bark of *Angostura* species (and related genera) was widely used by the natives of South America and the West Indies as a stimulant tonic (as a substitute for quinine, containing different bitter alkaloids and the bitter substance angosturin). Today the bark is also a source of antipyretics (Cortex *Angosturae*) and the base for *Angostura* bitters. The Cuban vernacular name 'Quina del país' for *A. ossana* (Gómez de la Maza 1889: 93) suggest usage of the bark as a substitute of quinine. *A. ossana* was probably also rare in the 19th, since it was collected only twice between 1820 and 1841. Perhaps populations of the species were strongly reduced by collecting of the bark. It is unknown whether the species occurs in Cuba today. In view of the case of *Zanthoxylum duplicipunctatum*, which was not collected for 136 years before its rediscovery in 2000 (Berazaín & al. 2000: 15), a possible occurrence of *A. ossana* in the hardly accessible mountains of the Sierra de los Organos in W Cuba cannot be excluded, even though natural vegetation has been much destroyed by agriculture nearly to the summits of the mountains.

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Address of the author:

Christa Beurton, Botanischer Garten und Botanisches Museum Berlin-Dahlem, Freie Universität Berlin, Königin-Luise-Str. 6-8, D-14195 Berlin; e-mail: c.beurton@bgbm.org

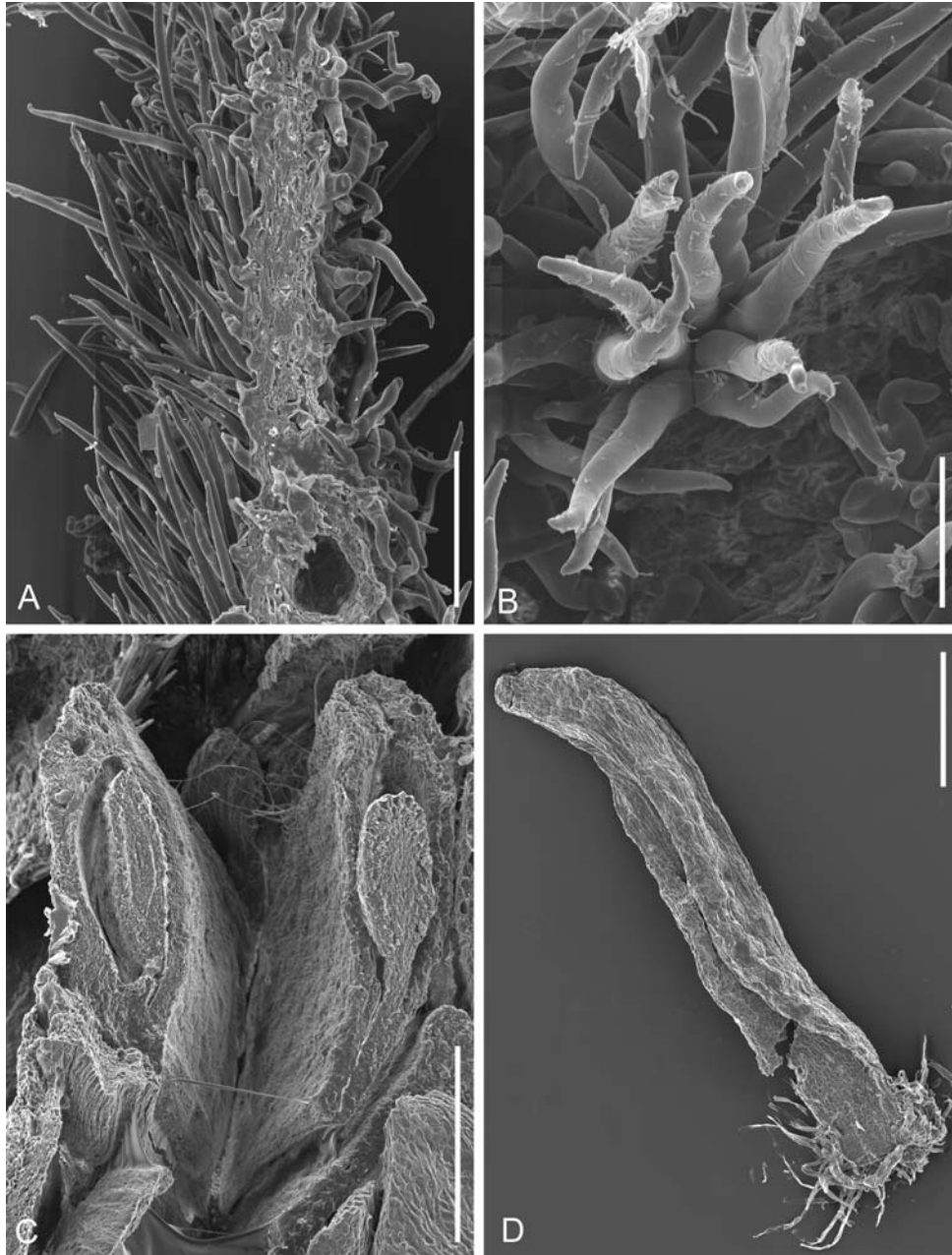


Fig. 4. *Angostura ossana*, scanning electron micrographs of corolla, gynoecium and staminode (holotype of *Galipea ossana*, G-DC) – A: corolla (lower part) in cross section (long stellate hairs on the abaxial surface, shorter spreading hairs on the adaxial surface); B: long stellate hairs (detail of A); C: part of a gynoecium (cross section) with two (of the five) carpels; D: staminode. – Scale bars A = 90 μm , B = 40 μm , C = 200 μm , D = 300 μm .

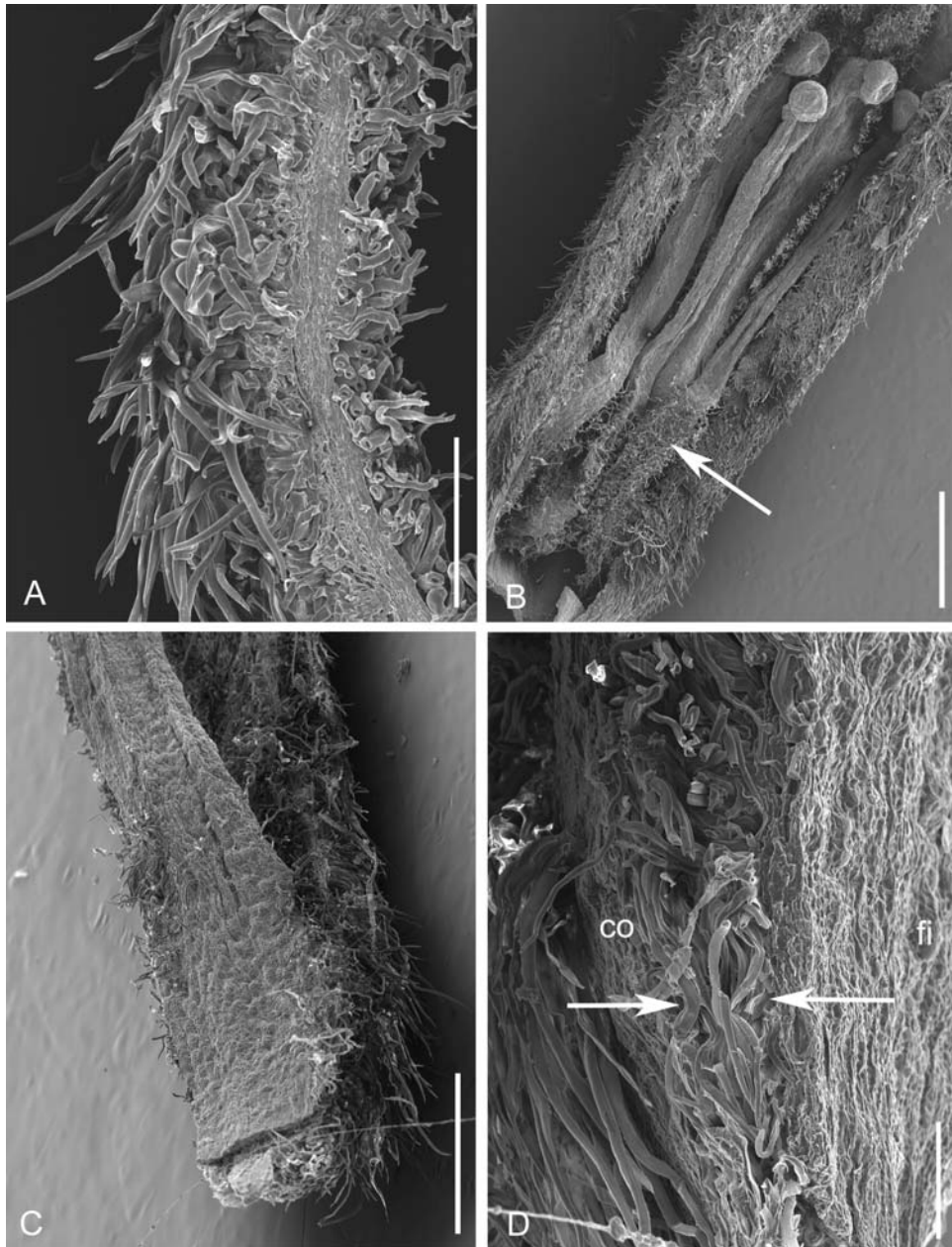


Fig. 5. *Angostura trifoliata*, scanning electron micrographs of flower bud, corolla, and filaments (holotype of *Bonplandia trifoliata*, B-W 4812) – A: corolla (lower part) in cross section (long stellate hairs on the abaxial surface, shorter, spreading hairs on the adaxial surface); B: four staminodes in a flower bud (longitudinal section) with gland-like anthers and with curled hairs abaxially and marginally in the area of the filament pseudo-tube (arrow); C: base of corolla tube and filament pseudo-tube, note the glabrous adaxial surface of the filaments; D: detail of C, note the intertwining of the adaxial hairs of the corolla with the abaxial hairs of the filaments. – Abbreviations: fi = filament; co = corolla; scale bars A = 200 μ m, B = 1 mm, C = 600 μ m, D = 100 μ m.

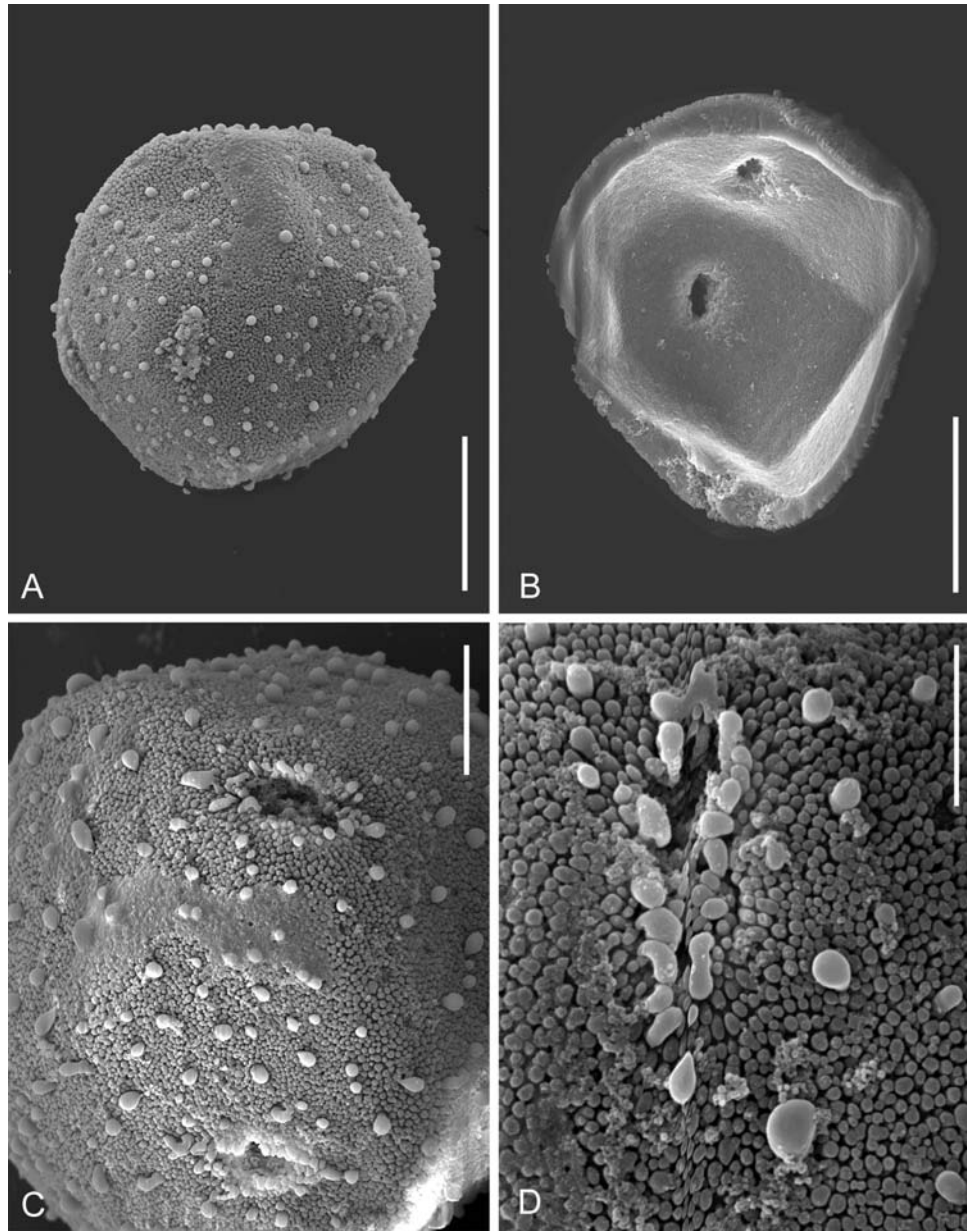


Fig. 6. *Angostura ossana*, scanning electron micrographs of pollen grains (holotype of *Galipea ossana*, G-DC) – A: oblique equatorial view; B: view into a broken pollen grain showing the sporoderm with two short-colporate apertures; C: details of apertures and sculpturing, D: baculae surrounding a colpus, surface view. – Scale bars A-B = 20 μ m, C = 10 μ m, D = 5 μ m.

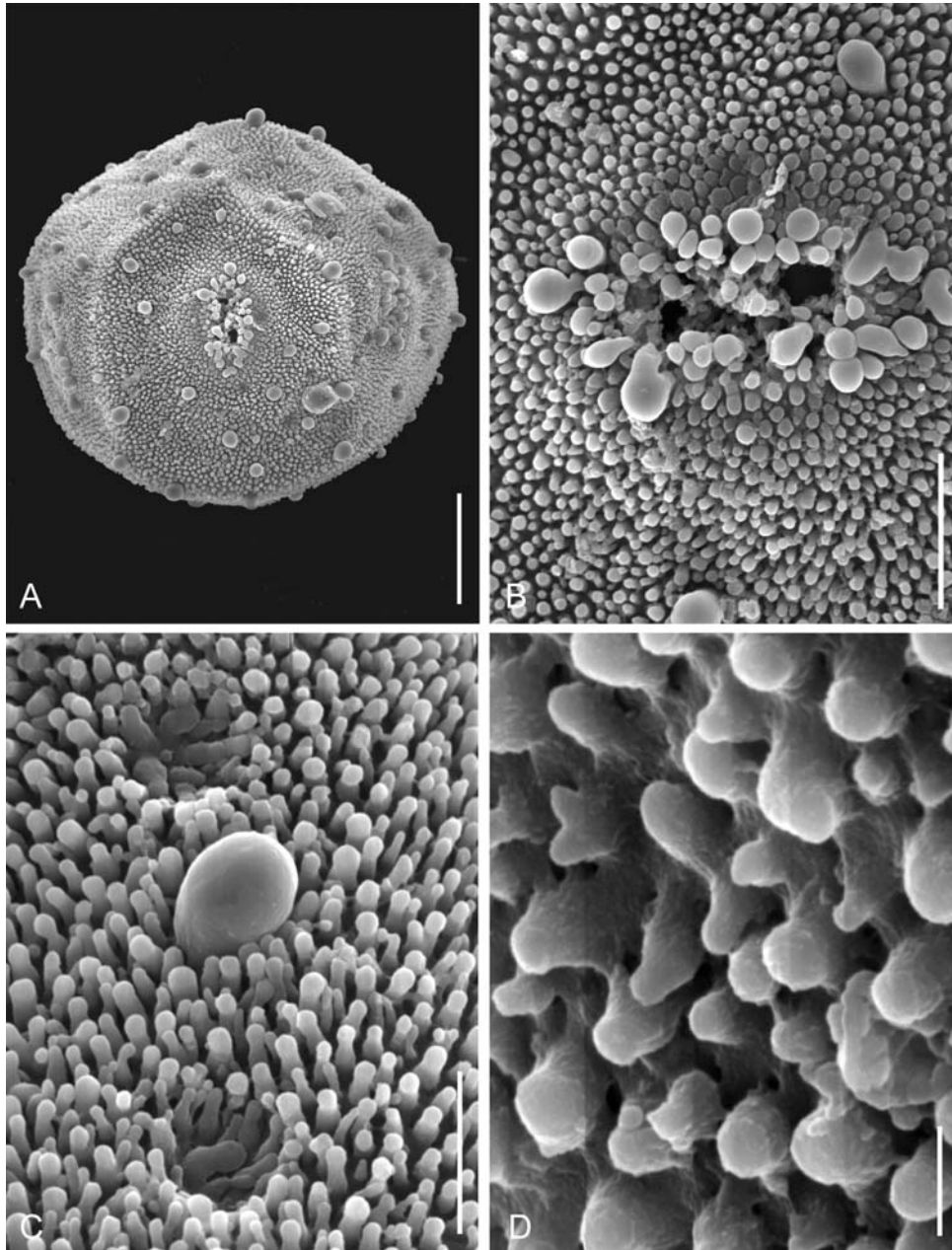


Fig. 7. *Angostura trifoliata*, scanning electron micrographs of pollen grains (holotype of *Bonplandia trifoliata*, B-W 4812) – A: equatorial view; B: detail of A, baculae surrounding a colpus; C: baculate sculpturing with rod-like structures, surface view; D: footlayer with pori, surface view. – Scale bars A = 20 μm , B = 7 μm , C = 4 μm , D = 1 μm .

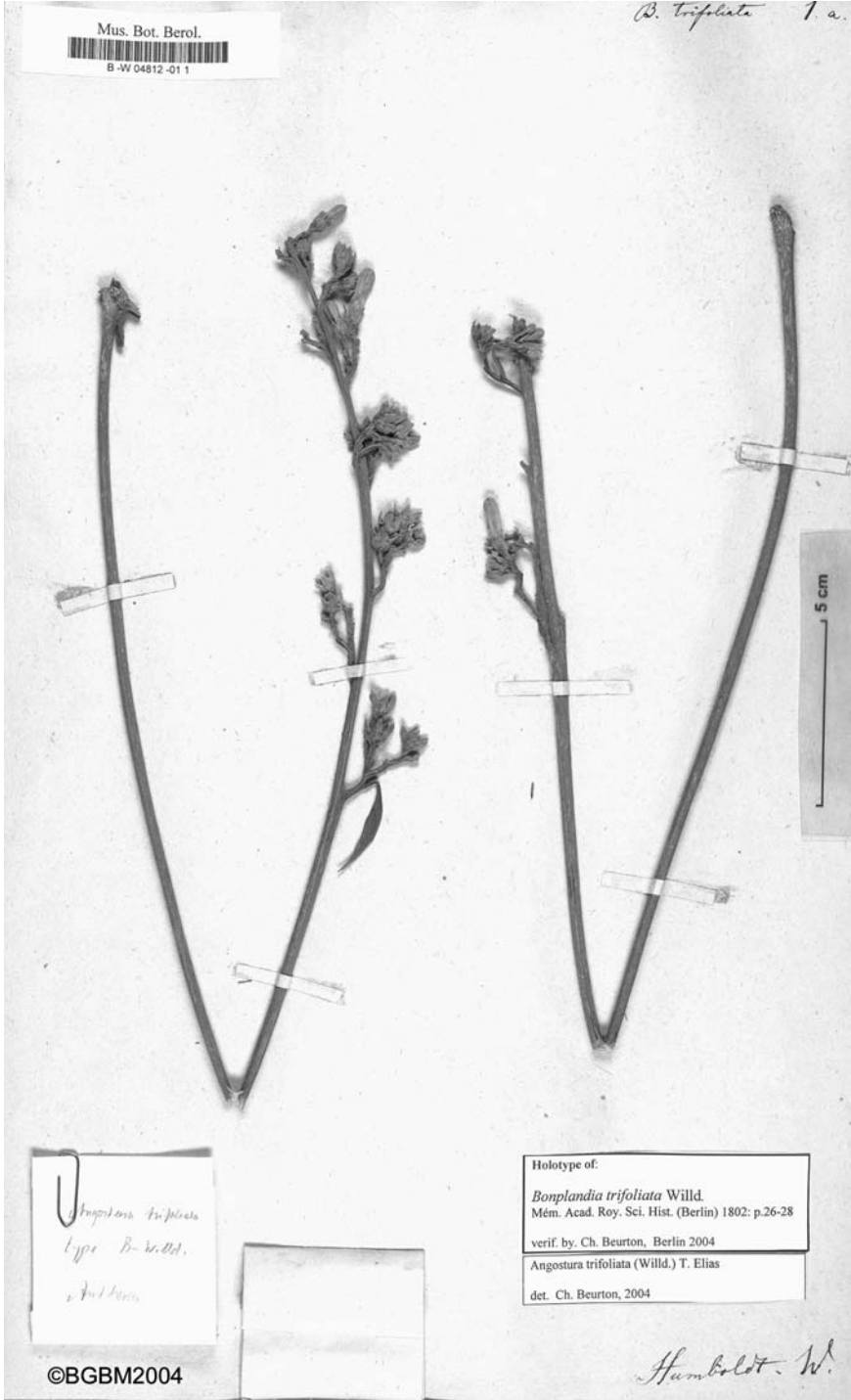


Fig. 8. *Angostura trifoliata* – holotype of *Bonplandia trifoliata* (B-W 4812).