

Croton laceratoglandulosus (Euphorbiaceae s.s.), a new glandular-stipulate species from Brazil and Bolivia, and its systematic position based on molecular analysis

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Croton laceratoglandulosus, a new species from the dry forests of the Brazilian states of Piauí, Ceará, Bahia and Minas Gerais, and the Bolivian department of Santa Cruz, is described and illustrated here. Molecular sequence data demonstrate that it is most closely related to the taxa of *Croton* section *Cascarilla*, and not to sections *Medea* or *Barhamia*, which also have glandular calyces and lacinate stipules. © 2008 The Linnean Society of London, *Botanical Journal of the Linnean Society*, 2008, 158, 493–498.

ADDITIONAL KEYWORDS: *Croton* section *Cascarilla* – dry forest – molecular phylogenetics – systematics.

INTRODUCTION

Croton L. is the second largest genus in the family Euphorbiaceae, with an estimated 1223 species (Govaerts, Frodin & Radcliffe-Smith, 2000), mainly from warmer regions of the world, but also from the subtropics and northern temperate areas. In the Neotropics, its most important centres of diversity are in Brazil, the West Indies and Mexico (Burger & Huft, 1995). In Brazil, *Croton* is represented by around 300 species, including herbs, shrubs and trees in all types of vegetation, but with the largest number of species concentrated in the eastern part of the country, mainly in areas of open vegetation, in semi-arid climates with irregular rains known as 'caatinga', or in tropical climates characterized by two well-marked seasons, dry and rainy, in 'cerrado' and 'campos rupestres'.

The new species described below, *Croton laceratoglandulosus*, is known from the Brazilian states of Piauí, Ceará, Bahia, and Minas Gerais and the Bolivian department of Santa Cruz, where it grows in semi-deciduous forests and caatingas.

RESULTS

CROTON LACERATOGLANDULOSUS CARUZO & CORDEIRO, SP. NOV.

Type: Brazil, Bahia, Malhada, rodovia BR-030 Malhada/Palmas de Monte Alto, 19.2 km da Sede Municipal, 14°17'11"S, 43°36'39"W, caatinga arbórea, altitude 500 m, 1.iv.2001, fl. fr., J.G. Jardim, F. Juchum, S. Sant'Ana, B. Santos & R. Querino 3372 (holotype, CEPEC; isotypes, HUEFS, SP) (Fig. 1).

Diagnosis: Similis *Croton ciliatoglandulifer* Ortega sed foliis petiolisque maioribus; stipulis profundi lacerato-glandulosis; inflorescentiis maioribus,

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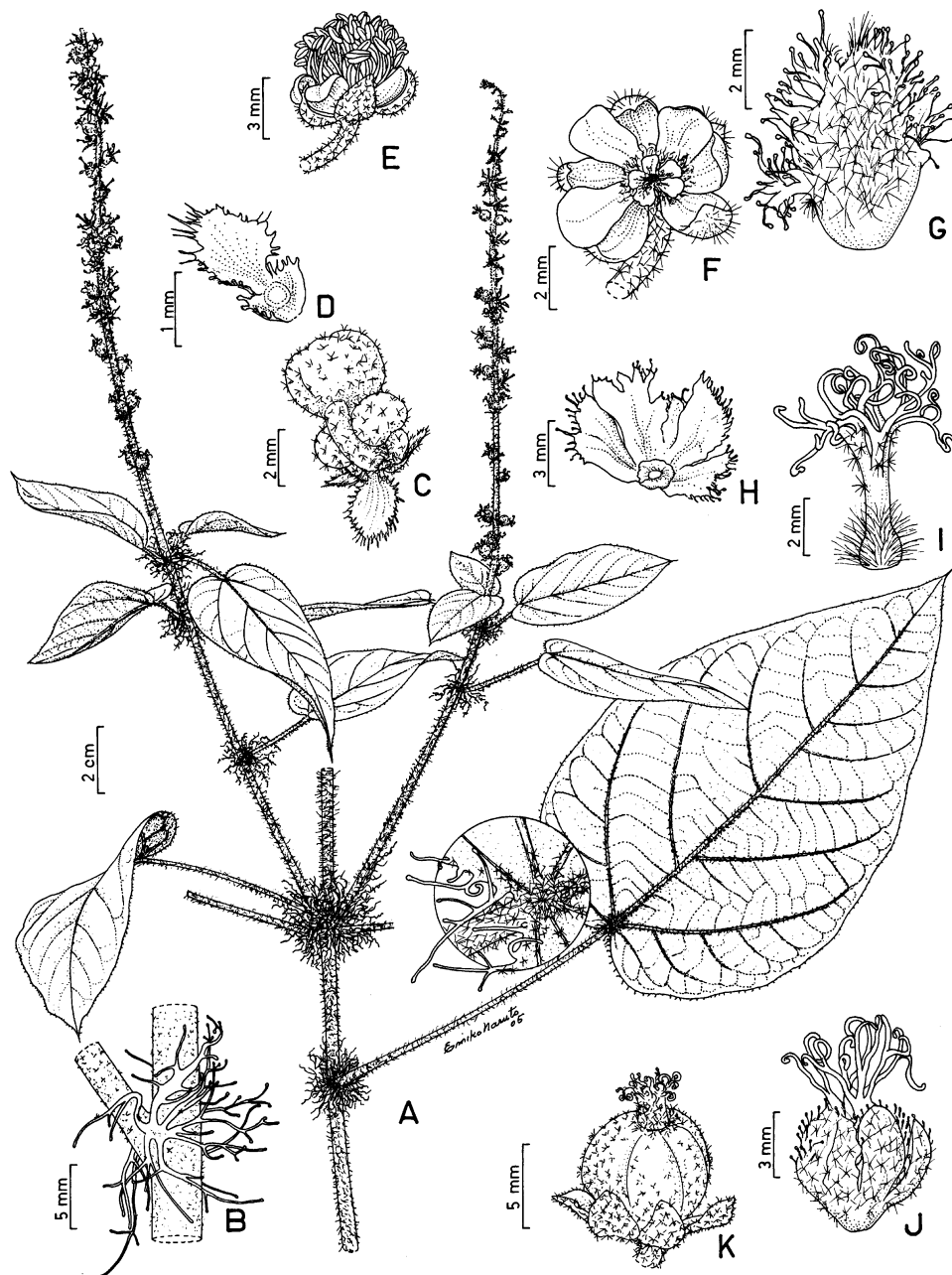


Figure 1. *Croton laceratoglandulosus* Caruzo & Cordeiro: A, habit; B, stipules deeply lacerate with glands on margins; C, cymule with male buds; D, bracts lacerate with glands on margins; E, male flower; F, detail of male flower disc; G, detail of female sepal; H, detail of female flower disc; I, gynoecium; J, female flower; K, fruit.

bracteis spatulatis, lacerato-glandulosis, fructibus globosis, glabrescentibus differt.

Description: Shrub 1–2 m tall, monoecious, indument of stellate trichomes; branchlets cylindrical, slightly striate. Leaves alternate; lamina 3.5–14.5 × 3–9 cm, ovate, discolorous, the underside whitish, apex acute to acuminate or rarely apiculate, base obtuse to

cordate, margins serrate to ciliate, with sessile or stalked glands, upper surface pubescent to glabrescent, lower surface velutinous, venation pinnate, brochidodromous, secondary veins joining the supradjacent secondary on an acute angle, tertiary veins percurrent, oblique, convex; petiole 1.5–9 cm long, without glands; stipules filiform to deeply lacerate, c. 10 × 2–10 mm, with marginal glands. Inflorescence

10–30 cm, terminal, without interruption between the female and male cymules; rachis cylindrical, striate, pubescent; cymules unisexual; bracts *c.* 2 mm, spatulate, lacerate with marginal glands. Male flowers in triads, on distal cymules, *c.* 6 mm long, subcampanulate, pedicels 2.5–3 mm long; calyx lobes 5, 2–2.5 × 1–1.5 mm, entire, equal, valvate, ovate, pilose externally, glabrescent internally, apex acute; petals 2–2.5 mm long, 1–1.5 mm wide, obovate to spatulate, apex obtuse to rounded, glabrescent; disc deeply five-segmented, segments opposite the sepals, glabrescent, *c.* 0.5 × 0.3 mm; stamens 50–55, filaments filiform; receptacle villose with simple trichomes. Female flowers solitary on proximal nodes of the inflorescence, 5–6 mm long, campanulate, sessile or rarely subsessile; calyx lobes 5, 3–4 mm long, 1.5–2 mm wide, slightly unequal, valvate, lanceolate to ovate-lanceolate, lacerate, glandular on margins, pubescent externally, glabrescent internally; petals reduced to inconspicuous glands or lacking; disc entire, shallowly five-lobed; ovary globose, pilose; styles multifid, slightly joined at the base. Fruits 5–6 mm in diameter, globose, glabrescent, smooth; seeds 3–3.5 × 2–2.5 mm, subglobose, slightly verrucose.

Paratypes: BRAZIL. Piauí: Serra da Capivara, 1979, *L. Emperaire 623* (IPA). Ceará: Aiuaba, Estação Ecológica de Aiuaba, *E. Nunes* (EAC 12615); Estação Ecológica de Aiuaba, caatinga, entre 400–600 m, 06°36'01"–06°44'35"S, 40°07'15"–40°19'19"W, 3.iii.2003, *J.R. Lemos & P. Matias 158* (HUVA); Quixadá, Serra do Estevão, *A. Fernandes & Matos* (RB 177116). Bahia: Caetité, 28.xi.1992, *M.L. Guedes 2686* (HRB); remanescente de floresta estacional semidecidual com elementos de caatinga arbórea, 14°04'03"S, 42°38'12"W, 820 m, 9.ii.1997, *B. Stannard et al. PCD 5309* (ALCB, CEPEC, K, SPF); Correntina, 22.i.2005, *W. Ribeiro 63* (SP); Iraquara, Fazenda Pratinha, margin of Rio San Antonio, 16 km SSE of Iraquara, 600 m, caatinga, 14.vi.1981, *S.A. Mori et al. 14420* (CEPEC, NY); Itanajé, 13°42'00"S, 42°01'00"W, 27.ii.1993, *M.L. Guedes et al. 2842* (ALCB, SP); Macaúbas, Subida Serra Poções, caatinga, 18.i.1997, *G. Hatschbach et al. 65961* (MBM, SP); 11 km W de Marcionílio Souza na estrada para Itaeté, caatinga arbórea, solo arenoso de cor vermelha, margem de estrada, 305 m, 13°03'S, 40°35'W, 22.iii.1998, *S. Ginzburg et al. 809* (SP, TEX-LL); cerca de 5 km da cidade de Pindai, 12.v.1978, *J.S. Silva 473* (SP); mistura de mata decidual e caatinga, 750 m, 16.iii.1981, *G.C.P. Pinto 140/81* (CEPEC, HRB, RB); Morpará, Quixaba, caminho para Ipupiara, a 3 km, caatinga arbórea antropizada, 11°48'S, 43°10'W, *M.L. Guedes & D. de Paulo Filho 7906* (ALCB, CEPEC, HUEFS); Rio de Contas, 12.5 km da cidade, na estrada para Jussiape,

beira de estrada, 13°35'55"S, 41°45'12"W, caatinga, altitude 707 m, 26.xii.2004, *R.M. Harley et al. 54970* (HUEFS, SP). Minas Gerais: Francisco Sá, vegetação secundária (caatinga), 16.xii.1989, *S. D'Angelo Neto 10* (ESAL); entre Sitio Barbacena, 1873, *A. Glaziou 5933a* (P); 1907, *A. Glaziou 15393* (BR, P); Monte Azul, subida via Montevídiu, 14.i.1997, *G. Hatschbach et al. 65759* (MBM, SP), 14.i.1997, *G. Hatschbach et al. 65762* (MBM, SP). BOLÍVIA. Santa Cruz. Chiquitos Province (probably San José de Chiquitos), Serrania Sansas, Campamento Bocamina, 12 km camino a Quebrada La Vera, 17°50'S, 59°45'W, 5.xi.1997, *F. Mamani & A. Jardim 1060* (USZ); 2 km W of Taperas on road to San José de Chiquitos, 400 m, dry forest merging into well-developed cerrado, on dry sandy soil, locally frequent, in moister areas of dry forest, 1.xi.1998, *J.R.L. Wood & F. Mamani 14095* (USZ, K).

DISTRIBUTION AND ECOLOGY

Croton laceratoglandulosus is a shrubby species from semi-deciduous forests and caatingas at altitudes of 300–820 m. It grows from Piauí and Ceará in north-east Brazil southwards into Bahia and Minas Gerais, and then reappears in Santa Cruz in east Bolivia (Fig. 2).

This disjunct distribution between north-east Brazil and south-east Bolivia is found in many species of dry forests and caatingas, and it can be understood as a vestige of a once extensive seasonal woodland formation, which may have reached its maximum extent during the dry, cool climatic period of the Pleistocene, as pointed out by Prado & Gibbs (1993).

MORPHOLOGICAL AFFINITIES

Following the synopsis of *Croton* sections by Webster (1993), this species keys out as intermediate in character between *Croton* section *Adenophyllum* Griseb., *Croton* section *Barhamia* (Klotzsch) Baill., and *Croton* section *Medea* (Klotzsch) Baill., as a result of its leaf margins with sessile or stalked glands, dissected stipules with glandular margins, inflorescences with unisexual cymules, female flowers with dissected calyx lobes and glandular margins, reduced petals and multifid styles. *Croton* section *Adenophyllum*, however, includes only Mexican, Mesoamerican and Caribbean species (Webster, 1993). Sections *Barhamia* and *Medea* are mostly South American, but they differ in the smaller number of stamens, usually with a maximum of 12, although Webster (1993) included *Croton luetzelburgii* Pax & K.Hoffm. from Bahia, Brazil, with 25–50 stamens, as described by Cordeiro (1995), in *Croton* section *Medea*. *Croton ciliatoglandulifer*, a mesoamerican species, is similar in

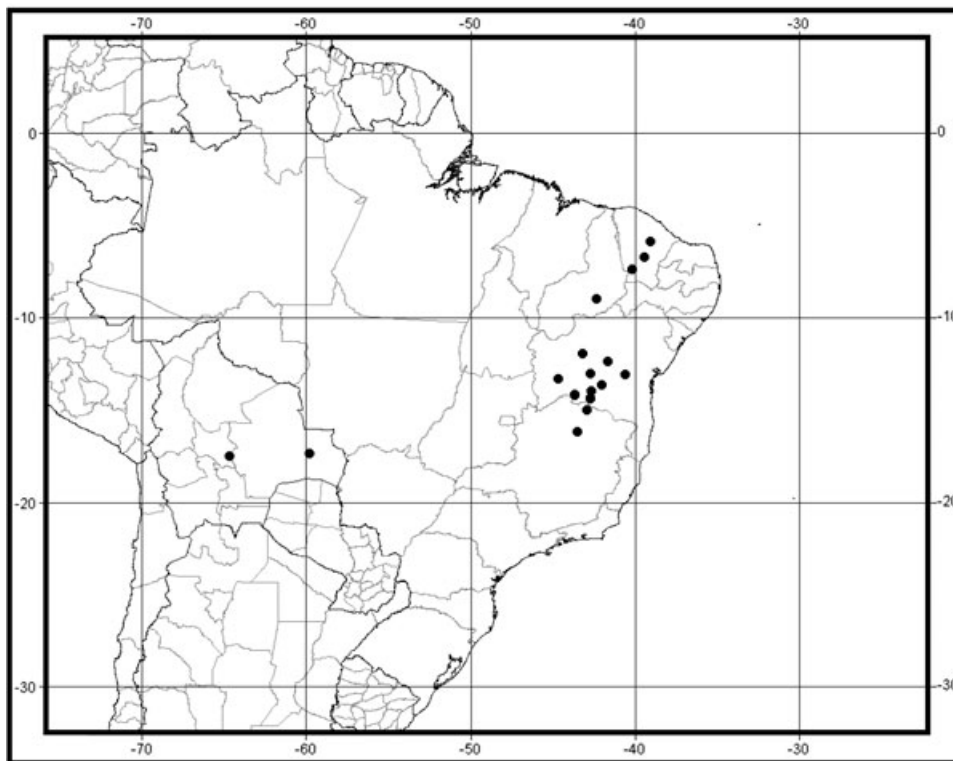


Figure 2. Geographical distribution of *Croton laceratoglandulosus* Caruzo & Cordeiro.

habit and general morphology to this new species, and it also occurs in deciduous forests from sea level to 1600 m (Martínez-Gordillo, 1995).

MOLECULAR ANALYSIS

DNA was extracted from a collection of *C. laceratoglandulosus* from Santa Cruz, Bolivia (Wood & Mamani 14095, USZ) and sequenced for the same two regions [nuclear internal transcribed spacer (ITS) and plastid *trnL-trnF*] as those used to produce the molecular phylogeny in Berry *et al.* (2005), employing the same laboratory methods. The sequences are deposited in GenBank (DQ836744; DQ836745). These sequences were integrated into the data matrix of Berry *et al.* (2005) available from TreeBASE (<http://www.treebase.org/treebase>). The combined data, with the new accession, were reanalysed using maximum parsimony and maximum likelihood, employing the same search strategies and likelihood model as in Berry *et al.* (2005). Once the position of *C. laceratoglandulosus* in the full data set of Berry *et al.* (2005) was established, a reduced data set consisting of 20 accessions from clades C-2 to C-11, the new accession of *C. laceratoglandulosus* and two outgroup taxa was analysed using both parsimony and likelihood. The outgroups, *Brasiliocroton mamoninha* and

Croton alabamensis, were selected based on their outlying positions in the phylogenetic analysis of Berry *et al.* (2005). In the reduced data set, congruence between the nuclear and plastid regions was evaluated using the incongruence length difference (ILD) test (Farris *et al.*, 1994), and a likelihood model was selected in Modeltest 3.5 (Posada & Crandall, 1998).

In the reduced 23-taxon data set, the ILD test revealed incongruence ($P = 0.01$) between the two regions. A visual examination of the phylograms recovered by analysing the ITS and *trnL-trnF* regions separately revealed that the two accessions from section *Cyclostigma* (*C. coriaceus* and *C. speciosus*) were the source of the incongruence. In the ITS-only analysis, they were recovered in the same position as depicted in Figure 3, as well as in figure 5 of Berry *et al.* (2005), whereas, in the *trnL-trnF*-only analysis, they were recovered embedded in the ingroup clade comprising taxa other than those of clade C-3, a position we consider to be erroneous. Repeating the ILD test after removing these two taxa indicated strong congruence ($P = 0.97$) between the data partitions. When the null hypothesis evaluated is that there is no incongruence between the data partitions, the ILD test has been found to have a high rate of type I error (Hipp, Hall & Sytsma, 2004). In their

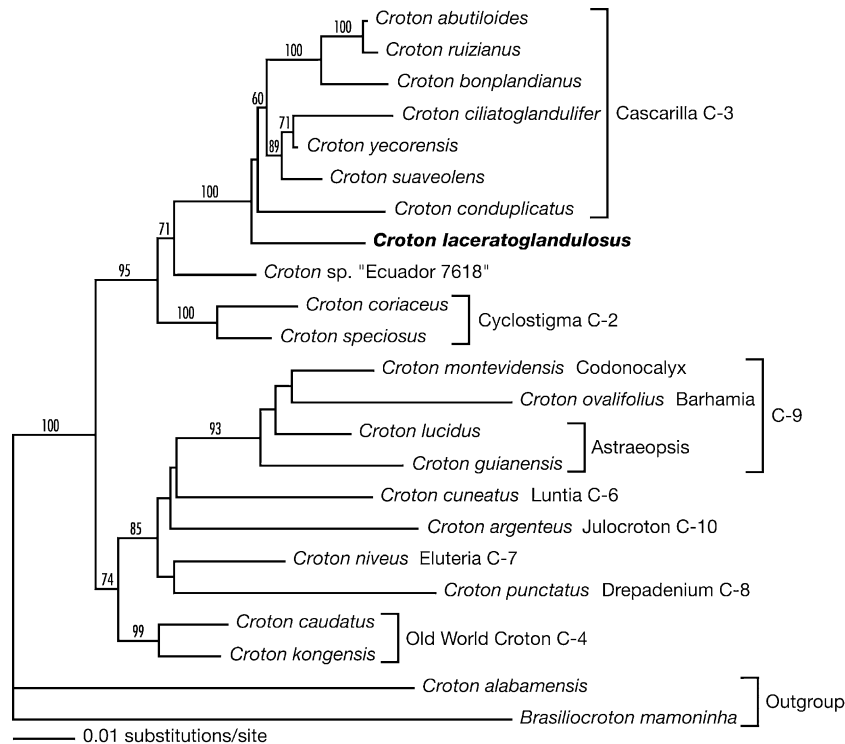


Figure 3. Phylogram of the maximum likelihood analysis of combined internal transcribed spacer (ITS) and *trnL-trnF* data. The numbers above the branches are parsimony bootstrap percentages. The labels on the right refer to the sections of *Croton* to which the taxa were assigned by Webster (1993), with the exception of those in clade C-3, which is shown as a more broadly circumscribed section *Cascarilla*. The clades beginning with 'C' refer to the groups defined by Berry *et al.* (2005).

91-taxon (excluding *C. setiger* and *C. insularis*) combined ITS and *trnL-trnF* data set, including all of the accessions from this work except for *C. laceratoglandulosus*, Berry *et al.* (2005) found congruence ($P = 0.214$) between the two regions. Our conclusion is that the indication of incongruence between the gene regions is a case of type I error, which, in this case, appears to be an artefact of the reduced taxon sampling. We therefore analysed the two gene regions together (analysing them separately does not yield a different phylogenetic placement of *C. laceratoglandulosus*). Using hLRT in Modeltest (Posada & Crandall, 1998), GTR + G + I was selected as the best-fitting likelihood model. In the single maximum likelihood tree obtained (Fig. 3), *C. laceratoglandulosus* occupies a position at the base of a clade consisting of members of clade C-3 from Berry *et al.* (2005), either in a basal trichotomy, or in a weakly supported position sister to all included members of clade C-3, including *C. conduplicatus* but excluding *Croton* sp. 'Ecuador 7618' (Fig. 3). Although the sister relationship of *C. laceratoglandulosus* is unresolved in our analyses, its inclusion in clade C-3 is strongly supported (100% bootstrap, Fig. 3). Although *C. lacer-*

atoglandulosus is not recovered as sister to *C. ciliatoglandulifer*, as would be suggested by the morphological data, its position indicates that it is as closely related to *C. ciliatoglandulifer* as to any other member of clade C-3, excluding *Croton* sp. 'Ecuador 7618'. The position of *C. laceratoglandulosus* in the molecular phylogeny is consistent with our interpretation of its morphological characters as being intermediate between several different sections of *Croton*, but ultimately closest to clade C-3. The grade of three South American taxa (*Croton* sp. 'Ecuador 7618', *C. laceratoglandulosus*, and *C. conduplicatus*) at the base of section *Cascarilla* suggests a South American origin for this section, the largest within *Croton*.

In the first molecular survey of *Croton* (Berry *et al.*, 2005), the species sampled with glandular stipules and calyces were grouped into two different and distant clades, named clade C-3 and clade C-9. These groups differ morphologically, with some members of clade C-3, such as *C. ciliatoglandulifer* and *C. yecorensis*, possessing stipitate-glandular leaf margins, whereas members of clade C-9 do not. With its ciliate-glandular leaf margins, *C. laceratoglandulosus* fits morphologically much better within clade C-3. This

clade is composed of taxa that were previously assigned to *Croton* sections *Adenophyllum*, *Medea*, *Cascarilla*, *Velamea* and *Cyclostigma* (Berry *et al.*, 2005). However, most, if not all, of these taxa should probably be assigned to a more broadly circumscribed section *Cascarilla*, with the other sections either being reduced to synonymy (as for section *Velamea* in Berry *et al.*, 2005), or recircumscribed as monophyletic groups. Within clade C-3, *C. laceratoglandulosus* is morphologically most similar to members of Webster's section *Adenophyllum*, although that section includes no South American species.

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