

A new species of *Picrasma*, *P. nanophylla* (Simaroubaceae), from the Dominican Republic

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Abstract: Recent collections in the Sierra Martín García represent a new species of the genus *Picrasma*, which because of its small leaves in comparison with closely related species, we describe as *P. nanophylla*. We document its phylogenetic position within the clade, compare it with close relatives and phenetically similar species, provide an illustration of the species and provide an identification key for the Greater Antillean species.

Keywords: Hispaniola, Sapindales, Seasonally Dry Tropical Forest, Sierra Martín García.

Resumen: Recientes colecciones en la Sierra Martín García representan a una especie nueva del género *Picrasma*, la que por sus hojas diminutas en comparación con otras especies la nombramos *P. nanophylla*. Mostramos su posición filogenética dentro del clado *Picrasma*, la comparamos con parientes cercanos y especies fenéticamente similares, proveemos una ilustración de la especie y una clave para las especies de las Antillas Mayores.

The circumscription of the Simaroubaceae has undergone several significant changes over the past 20 years with the realization that some genera, such as *Alvaradoa* Liebm. and *Picramnia* Sw., do not form part of that familial clade and should be recognized separately as Picramniaceae (Picramniales; Fernando et al., 1995; Logacheva & Shipunov, 2017), and *Suriana* L. belongs to the Surianaceae (Fabales; Crayn et al., 1995; Persson, 2001). Clayton et al. (2007) published the most comprehensive phylogeny of Simaroubaceae to date and showed that the mostly tropical genera *Castela* Turpin and *Picrasma* Blume were sister taxa, based on a combination of plastid and nuclear DNA sequence data.

The genus *Picrasma* is widespread in the Old and New World tropics (Cronquist, 1944; Thomas, 1990) consisting of ten known species (Palacios, 2015; Noa-Monzón & González-

Gutiérrez, 2019) and forms a small radiation in the Greater Antilles, with five species known, *P. cubensis* Radlk. & Urb., *P. excelsa* (Sw.) Planch., *P. pauciflora* A. Noa & P.A. González, *P. selleana* Urb., and *P. tetramera* (Urb.) W.W. Thomas, J.D. Mitchell & A. Noa (Acevedo-Rodríguez & Strong, 2012; Noa-Monzón & González-Gutiérrez, 2019). Three species are endemic to Cuba (*P. cubensis*, *P. pauciflora*, *P. tetramera*; Thomas et al., 2011; Noa-Monzón & González-Gutiérrez, 2019) and one to Hispaniola (*P. selleana*; Liogier, 1985). *Picrasma excelsa* Planch. occurs on all four islands of the Greater Antilles (Acevedo-Rodríguez & Strong, 2012) and is found elsewhere in the Neotropics. Liogier (1985) treated the family Simaroubaceae for Hispaniola, where he included the widespread *P. excelsa* and the endemic *P. selleana* from Haiti, as well as two

species of the genus *Simarouba*. He also included one species of *Castela*, *C. depressa* Turpin, endemic to the Dominican Republic.

Recent fieldwork in 2014 and 2016 (Franklin et al., 2019), in the Dominican Republic led to the discovery of the existence of a new species of *Picrasma* from the Sierra Martín García (southwestern Dominican Republic; Fig. 1) from seasonally dry tropical forest (SDTF). We describe the new species here as *P. nanophylla*, place the species in a phylogenetic context, and provide a

key to the genera of Simaroubaceae from Hispaniola and species of *Picrasma* from the Greater Antilles.

Materials and methods

Herbarium material of *Picrasma selleana* and *P. excelsa*, the other two species of *Picrasma* currently known from Hispaniola, was consulted from FLAS, FTG, JBSD, NY and S for

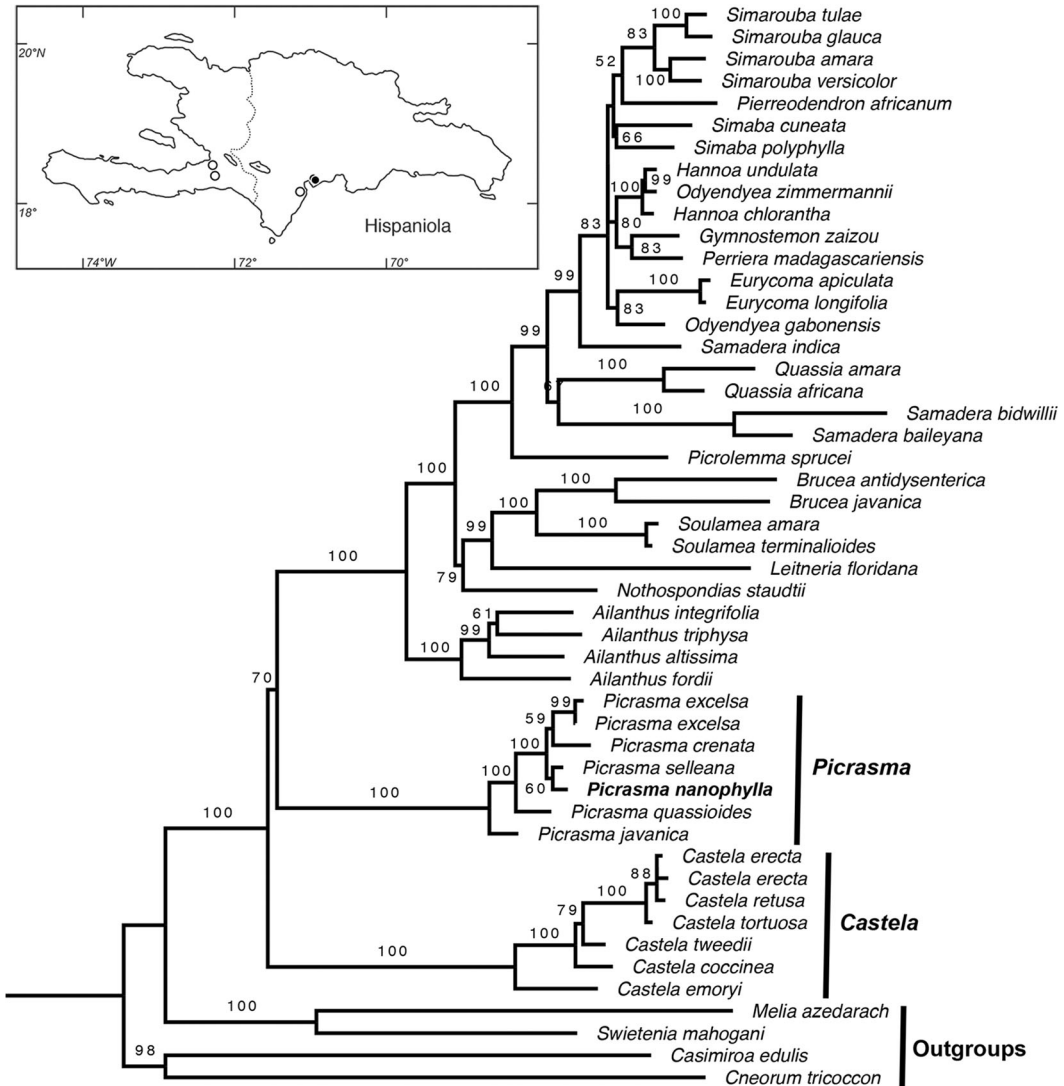


FIG. 1. Maximum likelihood phylogeny of *Picrasma* showing the placement of *P. nanophylla* (in bold) as sister to the Hispaniolan endemic *P. selleana* (bootstrap values are given above branches), and map of the distribution of *P. nanophylla* (black circles) in the Sierra Martín García, southwestern Dominican Republic and *P. selleana* (white circles) in Massif de la Selle, Haiti and Sierra de Bahoruco, Dominican Republic.

comparisons with the material collected from Sierra Martín García (*Majure 6473, Clase 8980*; see specimens cited below). We also compared the new material with previous descriptions and accounts of several Cuban endemic species (*P. cubensis*, *P. pauciflora*, and *P. tetramera*), which were treated by Thomas et al. (2011) and Noa-Monzón and González-Gutiérrez (2019).

We downloaded plastid sequence data published by Clayton et al. (2007) from GenBank and attempted to generate sequences through PCR for the loci used in that study (*atpB*, *matK*, *rbcL*). Whole genomic DNA was extracted from *P. selleana* and the new species using a modified CTAB protocol where the supernatant was directly column-cleaned to remove secondary compounds and other contaminants (Neubig et al., 2014; Majure et al., 2019). DNA degradation resulted in minimal amplification in our specimen of *P. selleana* (Judd 4400; FLAS) from Massif de la Selle, Haiti, and thus, we decided to undertake a genome skimming approach (Straub et al., 2012) using short-read sequencing via Illumina, which is amenable to fragmented DNA. Whole genomic DNA extracted from *P. nanophylla* and *P. selleana* was sent to Rapid Genomics for library preparation and sequencing on the Illumina HiSeq X platform with paired-end reads yielding 150 bp reads. Raw reads were imported into Geneious (Biomatters, LLC.) and reference mapping was performed using the plastid loci for *P. excelsa* (*atpB*, *matK*, *rbcL*) from the Clayton et al. (2007) dataset. We then generated majority consensus sequences for each locus. Our newly generated sequence data from *P. nanophylla* and *P. selleana* was incorporated into a reduced dataset (reduced from 64 to 47 terminals for our analysis) from Clayton et al. (2007), aligned using MAFFT (Katoh & Standley, 2016), and then manually corrected by eye in Geneious. We carried out a maximum likelihood (ML) analysis on the concatenated, unpartitioned dataset of all three plastid loci using RAxML (Stamatakis, 2014) under the default model of molecular evolution for smaller datasets, GTR + G, and a search for the mostly likely tree. We then undertook 1000 rapid bootstrap pseudoreplicates. Results were

visualized in Geneious initially and then in Figtree v1.4.4 (<http://tree.bio.ed.ac.uk/software/figtree/>).

Results

Picrasma nanophylla is most morphologically similar to *P. pauciflora* from Cuba (not sampled in our phylogeny), because both taxa have 3–5 leaflets, much smaller leaves than the rest of the species in the genus, reduced inflorescences (Figs. 2–3) and occur in similar SDTF habitats. *Picrasma nanophylla* differs from *P. pauciflora* in its growth form (i.e., shrub vs. tree in *P. pauciflora*), the size of its leaves (0.75–2.9 cm long vs. 3–6 cm long in *P. pauciflora*), number of flowers (1–2 vs. 2–4 in *P. pauciflora*), as well as floral and fruit indumentum (unicellular-pubescent vs. glabrous in *P. pauciflora*) (Noa-Monzón & González-Gutiérrez, 2019). *Picrasma nanophylla* is also similar to its closest relative inferred in our phylogeny (Fig. 1), *P. selleana*, in its growth habit (a shrub), hairy stamens, and hairy petals (see specimens examined). However, the number of flowers per inflorescence (1–2 vs. 5–10 in *P. selleana*), as well as leaf size (0.75–2.9 cm vs. 4.5–10 cm in *P. selleana*) and number of leaflets (3–5 vs. 5–9 in *P. selleana*) easily differentiate the two species. Likewise, the two species occur in very different habitat types, with *P. nanophylla* occurring in SDTF and *P. selleana* occurring in moist, relatively high elevation cloud forest dominated by *Brunellia comocladifolia* Bonpl. and *Pinus occidentalis* Sw. in the Massif de la Selle or lower elevation broadleaf mesic forest in the eastern Sierra de Bahoruco (see specimens examined).

Based on our analyses of the three plastid loci from Clayton et al. (2007), *Castela* is resolved as sister to *Picrasma* plus the rest of Simaroubaceae (bs = 70) and does not form a clade with *Picrasma*. *Picrasma nanophylla* is resolved as sister to *P. selleana*, although this relationship was not strongly supported (bs=60). There was strong support (bs=100) for a Neotropical clade including *P. crenata*, *P. excelsa* and the two Caribbean species, *P. nanophylla* and *P. selleana* (Fig. 1).

Key to the genera of Simaroubaceae on Hispaniola and the species of *Picrasma* in the Greater Antilles

1. Plants with thorns, leaves unifoliolate, not compound *Castela*
1. Plants without thorns, leaves compound 2
 2. Petals 5, stamens double the number of petals, each with an appendage at the base, trees *Simarouba*
 2. Petals 4–5, stamens the same number as the petals, without basal appendages, trees or shrubs *Picrasma* (3)
 3. Leaves 3–5 foliate, flowers 1–4 per inflorescence 4
 4. Trees 4–5 m tall, leaves 3–6 cm long, abaxial leaf surface dark green in dried specimens, flowers 2–4 per inflorescence, petals, staminal filaments, and fruit glabrous, Holguín, Cuba *P. pauciflora*
 4. Shrubs to 2.5 m tall, leaves 0.75–2.9 cm long, abaxial leaf surface light green in dried specimens, flowers 1–2 per inflorescence, petals, staminal filaments, and fruit unicellular-pubescent, Sierra Martín García, Dominican Republic *P. nanophylla*
3. Leaves 5–13 foliate, flowers 5–numerous per inflorescence 5
 5. Trees 6–25 m tall, leaves 5–13 foliate, leaflet margins entire, widespread in the Neotropics *P. excelsa*
 5. Shrubs or small trees 1–6 m tall, leaves 5–7 foliate, leaflet margins toothed, Cuba and Hispaniola 6
 6. Leaf rachis not winged, leaflet margins revolute, adaxial leaflet veins sunken, inflorescences 10 + flowered, Central Cuba *P. tetramera*
 6. Leaf rachis winged or not, leaflet margins not revolute, adaxial leaflet veins not sunken, inflorescences 5–10 flowered 7
 7. Leaflets 7, leaflet margins dentate throughout, leaflets pubescent abaxially only at the base of the primary vein, sepals pubescent abaxially along midrib, W Cuba *P. cubensis*
 7. Leaflets 5–9, leaflet margins dentate throughout or restricted to distal 1/2 to 1/3 of leaflet, leaflets unicellular-pubescent abaxially throughout the primary and secondary veins, sepals pubescent abaxially throughout, Massif de la Selle-Sierra de Bahoruco, Hispaniola *P. selleana*

Taxonomic treatment

***Picrasma nanophylla* Majure & Clase, sp. nov.**

Type: La Española. República Dominicana. Prov. Barahona: Sierra Martín García, ca. 0.6 km al noreste del Cruce de Vicente Noble, al noreste ca. 12.3 km de la Carretera 44, 18.396441°N, 71.042634°E, 840 m, 13 Nov 2016 (fl staminate), *L.C. Majure* et al. 6473 (holotype: JBSD; isotypes: FLAS (272844), K, MO, NY, S, ULV, US).

Diagnosis: Differing from its putative sister species *P. selleana* by the smaller leaves (0.75–2.9 × 0.5–2.0 cm vs. 6–10 × 6–9.6 cm in *P. selleana*) with fewer leaflets (3(–5) in *P. nanophylla* vs. 5–9 in *P. selleana*), and inflorescence of 1–2 flowers vs. 5–6 flowers in *P. selleana*. Differing from the phenetically similar species *P. pauciflora* in growth form (shrub vs. small tree), leaf size (0.75–2.9 cm long in *P. nanophylla* vs. 3–6 cm long in *P. pauciflora*), number of flowers per inflorescence (1–2 in *P. nanophylla* vs. 2–4 in *P. pauciflora*), petal, stamen, and fruit indumentum (pubescent in *P. nanophylla* and glabrous in *P. pauciflora*).

Androdioecious shrubs to 2.5 m tall, leaves alternate, 3 (–5) foliate, 0.75–2.9 × 0.5–2.0 cm, mostly glabrous or with occasional, unicellular hairs at the base of the leaflets along the primary veins adaxially, apical leaflets 6.7–22 × 3.7–8.1 mm, obovate to occasionally elliptic, base acute, apex acute to uncommonly rounded, lateral leaflets 3.3–14 × 2–6.5 mm, elliptic, opposite, base acute, apex acute to uncommonly rounded, leaflet margins serrate, with upwardly arching teeth (these oftentimes slightly overlapping the upper leaf surface, leaflet venation craspedodromous), terminating in a black gland (when young), these quickly deciduous, secondary veins slightly impressed adaxially and upraised abaxially, petiole of compound leaf 1.4–5.5 mm long. Staminate plants: inflorescences highly reduced, with 1(–2) flowers, peduncles to 3 mm long (when present), pedicels 5–10 mm long, pubescent with unicellular hairs, flowers 4-merous, sepals green, 1.4–1.5 × 0.2–0.4 mm, narrowly ovate, pubescent, petals yellow-green, 2.5–2.8 × 1.2–1.5 mm, the base clawed, obovate to elliptic, pubescent along the midrib on both adaxial and abaxial surfaces,

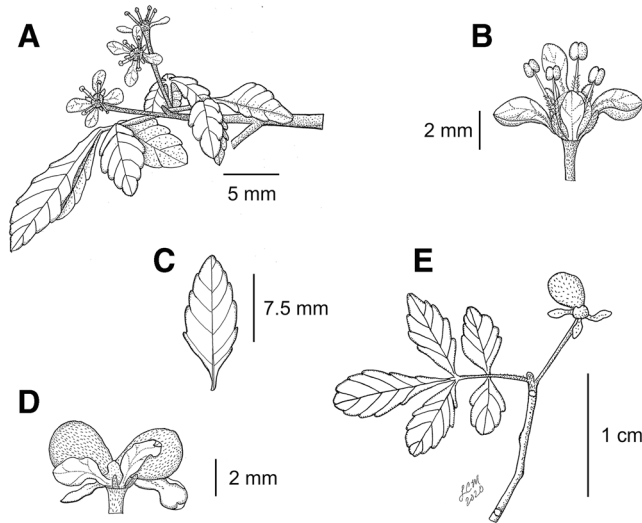


FIG. 2. Illustration of *Picrasma nanophylla*. A. Habit, showing tri- and quinquefoliolate leaves and position of the inflorescence (Majure et al. 6473). B. Staminate flower showing clawed petals and pubescent stamens (Majure et al. 6473). C. Terminal leaflet showing craspedodromous venation (Clase 8980). D. Immature fruit (pubescent) with persistent petals (Clase et al. 8980). E. Portion of stem with quinquefoliolate leaf and immature fruit with persistent petals (Clase et al. 8980).

stamens 4, arising from 4-lobed intrastaminal disk, filaments pubescent, 1.8–2 mm long. Hermaphroditic plants: inflorescences highly reduced, with 1(–2) flowers, peduncle to 8.7 mm long, pedicels 4–8 mm long, pubescent, flowers 4 (5)-merous, sepals green, 1.2–1.6 × 0.2–0.3 mm, narrowly ovate, pubescent, petals light yellow, 2.5–2.7 × 1.1–1.6 mm, the base clawed, obovate to elliptic, pubescent along the midrib on both adaxial and abaxial surfaces, stamens 4, arising from 4-lobed intrastaminal disk, filaments pubescent, 0.8–1.1 mm long, carpels 2, ovaries separate, pubescent (hairs unicellular), the styles fused and stigmas separate, sepals and petals often persistent in immature fruit, fruit a cluster of drupes (drupelets), 3–4.5 × 2.5–4 mm, green (immature), mature fruit pinkish-red, poorly known.

Distribution and habitat.—*Picrasma nanophylla* is restricted to the Sierra Martín García in southwestern Dominican Republic, where it is found in SDTF over limestone at 700–840 m elevation. The species occurs with *Akrosida floribunda* Fryxell & Clase, *Amyris elemifera* L., *Croton corylifolius* Lam., *Damburneya coriacea* (Sw.) Trofimov & Rohwer, *Erythroxyllum rotundifolium* Lunan, *Eugenia foetida* Pers., *Guarea guidonia* (L.) Sleumer, *Krugiodendron ferreum* Urb., *Leersia monandra* Sw., *Leptocereus weingartianus* Britton & Rose, *Mammillaria prolifera* (Mill.) Haw., *Morisonia americana* L., *Opuntia taylori*

Britton & Rose, *Selenicereus triangularis* (L.) D.R.Hunt, *Sideroxylon obovatum* Lam., and *Trichostigma octandrum* (L.) H.Walter. It is likely that more occurrences of this species will be found in the poorly explored SDTF in both the Dominican Republic and Haiti.

Phenology.—The type (Majure 6473) was collected in flower in November (Fig. 3) and the species was collected in flower and with immature/mature fruits in June (Clase 8980; Fig. 2), so it seems likely that *P. nanophylla* flowers sporadically throughout the year possibly corresponding with seasonal precipitation.

Etymology.—The specific epithet “*nanophylla*” refers to the very small leaves of the species, which are smaller than those of any other currently recognized species of *Picrasma*.

Conservation status.—The new species is known only from an approximately 0.25 km² area in the Sierra Martín García. The known population consists of very few individuals (<20), although, extensive work has not been carried out to determine the exact number of individuals in the area. This species should be considered data deficient based on the paucity of detailed information regarding population size, the numbers of hermaphroditic versus staminate plants within the population and based on the lack of comprehensive exploration among other SDTF sites where the species could occur in both the

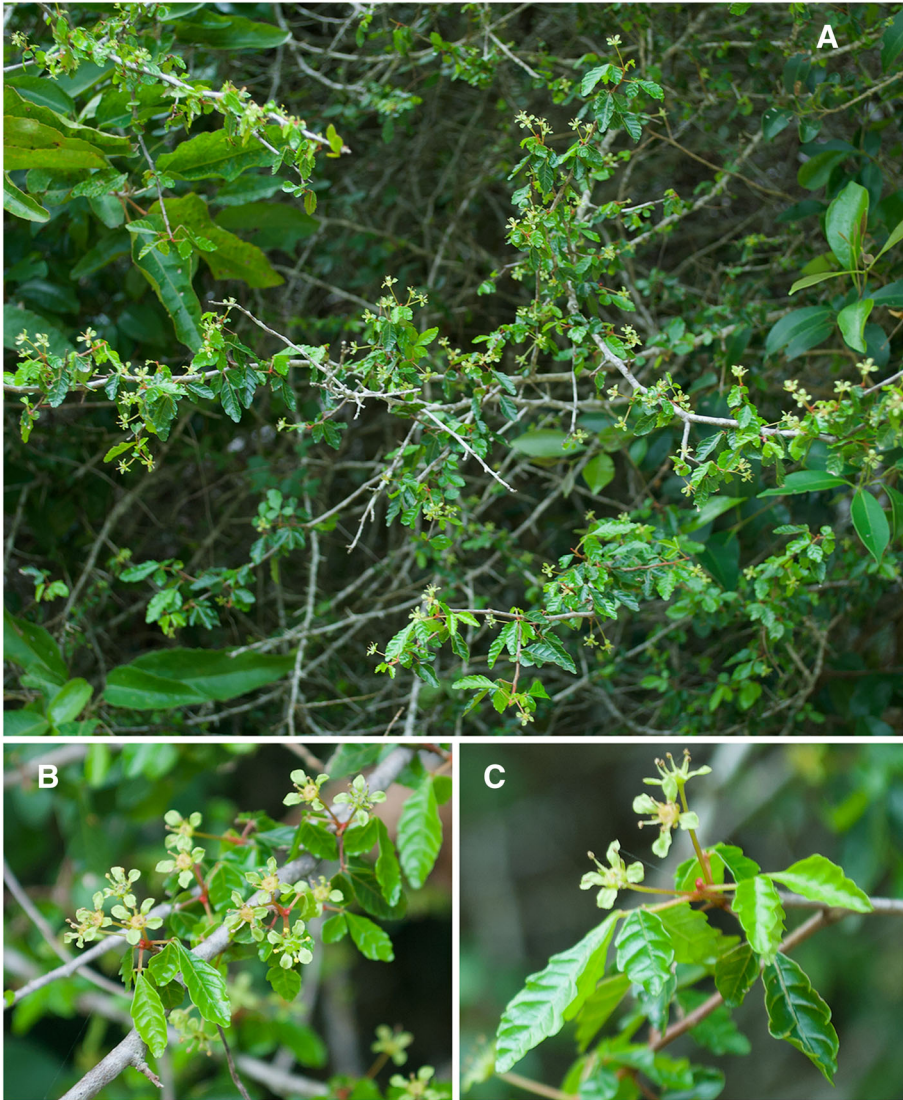


FIG. 3. Photos of *Picrasma nanophylla* in flower. A. Habit. B-C. Staminate flowers and structure of the inflorescence (Majure et al. 6473) along with mostly trifoliolate leaves. (Photos taken by L.C. Majure).

Dominican Republic and Haiti. That the species occurs in a protected area, the Parque Nacional Sierra Martín García of the Dominican Republic, greatly enhances the potential for its protection.

Species concept.— As is discussed more fully in the results section above, *P. nanophylla* is morphologically distinguishable from close relatives, such as *P. selleana*, and the phenetically similar species from Cuba, *P. pauciflora*, based on numerous morphological characters.

Therefore, our delimitation of the new species adheres to the morphological-phenetic species concept of Judd (2007) and diagnostic species concept of Wheeler & Platnick (2000). Likewise, because the species is restricted to the Sierra Martín García, as far as is currently known, it must certainly be reproductively isolated from other close relatives, which are all allopatric (see Fig. 1), therefore, making the biological species concept (Mayr, 2000) applicable.

Additional specimens examined. — *Picrasma nanophylla*: HISPANIOLA. DOMINICAN REPUBLIC. [Prov. Barahona]: Sierra Martín García, Municipio Vicente Noble, en una cañada seca subiendo hacia Alto de la Bandera, 18°23'56"N, -71°04'02.6"O, 700 m, 26 Jun 2014, (fl hermaphroditic, and with mostly immature fruit), *Clase et al. 8980* (F, FLAS-272843, JBSD, NY, US).

Additional specimens examined. — *Picrasma selleana*: HISPANIOLA. DOMINICAN REPUBLIC. Prov. Barahona: Las Filipinas, Finca Sucesión. 850 m, 4 Jan 1977, *Liogier 26,258* (FTG). HAITI. Dept. de l'Ouest: Massif de la Selle, Morne Cabaio, in virgin forest of good soil Laubwald, 25 Aug 1924, *Ekman H1636* (Type-S, online); Parc National Morne La Visite, lower cascade of Riviere Blanche, ca. 2 km SW of park headquarters, S of Morne La Visite, 1650–1800 m, 10 May 1984, *Judd 4400* (FLAS, JBSD). Massif de la Selle, 8 km “Norte” de Seguin en la carretera a Furcy y Petionville; “Cajacque”, 18°20'N, -72°16'O, elev. 1900 m, 21 Nov 1982, *Zanoni 24,528* (JBSD).

Selected specimens examined. — *Picrasma excelsa*: COSTA RICA. Prov. Puntarenas: Cantón de Puntarenas Monteverde, upper community on farms of Wolf Guindon & John Campbell, 14 Jan 1992, *Haber 10,987* (NY). EL SALVADOR. Dept. de Sonsonate: A la orilla del bosque nebuloso del Cerro El Pilón; 24 km al SO de la ciudad de Santa Ana, municipio de Juayúa, April 1995, *Linares 2489* (NY). FRANCE. GUADELOUPE. La Désirade: ravine de la Rivière, 19 June 1982, *Barrier 3582* (NY). GRENADA. Mt. Gilbert, NE of Mt. Maitland, 4–10 March 1979, *Howard 18,797* (NY). HISPANIOLA. HAITI. Dept. du Sud: Massif de la Hotte. en la entrada del este del poblado Port-Salut, 25 Jan 1985, *Zanoni 33,187* (NY). JAMAICA. St. Elizabeth Parish: Fields near retirement hotel at Malvern, 11 Sept 1954, *Howard 13,666* (NY). PANAMA. Prov. De Cocle: region north of El Valle de Anton, 27 Sept 1946, *Allen 3725* (US). UNITED STATES. PUERTO RICO. Municipio de Quebradillas, Barrio Terranova, along old railway right-of-way 0.3 km due E of Hwy. 2 bridge, 9 Sept 1989, *Proctor 45,915* (NY).

Discussion

Picrasma nanophylla is morphologically distinct from other species of Greater Antillean *Picrasma* based on the size of its leaves, reduced

numbers of leaflets, and small, shrubby, growth form (Figs. 2–3). Likewise, the inflorescences are axillary, reduced cymes (or more frequently one-flowered; Figs. 2–3) and smaller than those of all other species.

The putative close relationship with *P. selleana* from the Massif de la Selle and Sierra de Bahoruco is a common pattern found in a number of other taxa from Sierra Martín García. Floristically Sierra Martín García is in some ways similar to the adjacent Sierra de Bahoruco (García et al., 2007), which is an eastward extension of the Massif de la Selle. *Miconia paralongicollis* (Judd, Ionta, Clase & Skean) Judd & Bécquer, for example, is known from populations in both Sierra de Bahoruco and Sierra Martín García (Judd et al., 2008).

With the addition of *Picrasma nanophylla* to the flora of Hispaniola, it is clear that the Greater Antilles is a major hotspot for endemism and diversification of the genus considering the two, endemic species from Hispaniola, *P. nanophylla* and *P. selleana*, and the three, endemic species from Cuba, *P. cubensis*, *P. tetramera* and *P. pauciflora* (Noa-Monzón & González Gutiérrez, 2019).

Picrasma nanophylla is phenetically most similar to *P. pauciflora*, and both of those taxa are extremely restricted in their distribution (Sierra Martín García, Dominican Republic vs. Holguín, Cuba, respectively), occurring only in sclerophyllous vegetation in SDTF. The same pattern can be seen in *P. cubensis* and *P. selleana*, two species that are morphologically very similar (see key), and which are greatly restricted on Cuba and Hispaniola (to Pinar del Río, Cuba, and the Massif de la Selle-Sierra de Bahoruco, Hispaniola, respectively). Although we have not included the Cuban species in our phylogenetic analyses, it seems likely based on morphological characters that *P. nanophylla* and *P. pauciflora* are close relatives or even sister species, as may be the case for *P. cubensis* and *P. selleana*. All four of these species are known from very few collections, and *P. cubensis* is still only known from the type (Thomas et al., 2011). Fieldwork is essential for discovering more populations of these taxa and for filling in the potential gaps in their distribution. Based on recent discoveries (Noa-Monzón & González-Gutiérrez, 2019), the species described here, and others in preparation (Majure et al., in revision), it is clear that we still have much to understand about the diversity of the often poorly studied SDTF of the Greater Antilles.

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APPENDIX 1

Accessions used in our phylogenetic analysis with associated GenBank numbers (*rbcL*, *matK*, *atpB*). *Picrasma nanophylla* (Majure 6473-FLAS; MN517916, MN517917, MN517918). *Picrasma selleana* (Judd 4400-FLAS; MN517913, MN517914, MN517915).