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## Taxonomic Revision and Phylogenetic Position of the Brazilian Endemic Genus *Sphaerorrhiza* (Sphaerorrhizinae, Gesneriaceae) Including Two New Species

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**Abstract**—*Sphaerorrhiza*, a genus of herbaceous plants endemic to the Cerrado domain in Brazil, was recently segregated from the genus *Gloxinia* and placed in its own monotypic subtribe Sphaerorrhizinae. Most information on this little-known taxon derived from the observation of a single species and a limited number of collections. Using new material representative of all recognized species we reassess the monophyly of *Sphaerorrhiza* and its phylogenetic placement within Gesnerioideae based on the analysis of nuclear (ITS) and plastid (atpB-rbcL, matK, rpl16, rps16, trnL-trnF) DNA sequences. Our results support the monophyly of the genus and its segregated position from the other clades of Gesneriaceae. A taxonomic revision of *Sphaerorrhiza* is presented, including new anatomical and morphological data and the first reported chromosome number for the genus. Four species are recognized and two species are described as new, *S. rosulata* and *S. serrata*. Descriptions, illustrations, and a distribution map, as well as a key to the species of *Sphaerorrhiza*, are provided.

**Keywords**—Cerrado, *Gloxinia*, molecular phylogeny, rhizomes, tuber, underground storage organs.

*Sphaerorrhiza* is a genus of herbaceous plants endemic to the Cerrado domain in Brazil. Species of this genus are found growing on rocky outcrops close to rivers within gallery forest in central Brazil, or in rocky savanna-like habitats along the Espinhaço mountain range (Araujo et al. 2015). The genus *Sphaerorrhiza* Roalson & Boggan and the tribe Sphaerorrhizeae were established by Roalson et al. (2005a) to accommodate two species previously classified in *Gloxinia* L’Hérit: *G. sarmentiana* Gardner ex Hook. and *G. burchellii* (S. M. Phillips) Wiehler. This decision was mainly supported by phylogenetic results showing that *G. sarmentiana* was clearly misplaced in *Gloxinia* and lacked affinity with any of the recognized taxa in the subfamily Gesnerioideae (Zimmer et al. 2002; Roalson et al. 2005b). In a recent new formal classification of the family Gesneriaceae, the tribe Sphaerorrhizeae received a new status as subtribe Sphaerorrhizinae, within tribe Gesnerieae together with Columneinae, Gesneriinae, Gloxiniinae, and Ligeriinae (Weber et al. 2013).

Morphologically, *Sphaerorrhiza* has been defined primarily on the basis of its underground storage organs, described as “lumpy rhizomes” (Zimmer et al. 2002) or small tuber-like swellings (Roalson et al. 2005a). This structure appears distinct from what is found in other clades of Gesnerieae, like the scaly rhizomes in the Gloxiniinae and the tuber in most species of Sinningia Nees (Kvist and Skog 1992, Weber 2004). Besides the underground system, *Sphaerorrhiza* closely resembles Gloxiniinae genera occurring in the same saxicolous habitat such as *Mandirola* Decne. and *Goyazia* Taub., with which it shares a similar vegetative habit and campanulate flowers (Araujo et al. 2005; Araujo 2007). These similarities explain why this taxon has not been foreseen as a separate entity until the use of molecular data (Roalson et al. 2005a).

Phylogenetic and morphological characterization of *Sphaerorrhiza* has been so far based on the analysis of a restricted number of specimens of *S. sarmentiana*. This narrow sampling has prevented an accurate assessment of its

monophyly and diversity. In addition, the relationship between *Sphaerorrhiza* and the other lineages within Gesnerieae is not clear. Previous results have variously suggested that *Sphaerorrhiza* could be sister to a clade composed of Ligeriinae and Columneinae (Zimmer et al. 2002), to the Ligeriinae only (Roalson et al. 2008), or to all other Gesnerieae subtribes (i.e. Gloxiniinae, Gesneriinae, Columneinae, and Ligeriinae; Perret et al. 2013). However, none of these alternative hypotheses is well supported, indicating that additional molecular data are still needed to resolve the phylogenetic placement of *Sphaerorrhiza* within the Gesnerieae.

Here we present a revision of the genus *Sphaerorrhiza* and describe two new species based on an extensive survey of specimens and field observations collected during several trips to different localities within the Cerrado domain. In addition, the phylogenetic placement of this taxon within the Gesnerioideae and its monophyly was tested using all recognized species of *Sphaerorrhiza* and a larger number of loci than previously considered. Finally, to further evaluate the taxonomic utility of the underground system in *Sphaerorrhiza* and whether it is a synapomorphy for this genus, we improved its morphological and anatomical characterization in the four recognized species and compared it with other types of underground storage organs found in the related clades of Gesnerieae.

### Materials and Methods

**Taxonomic Study**—This study is based on field observation and examination of herbarium specimens of *Sphaerorrhiza* deposited in 39 herbaria (B, BHCB, BM, BR, CEN, CGE, E, ESA, ESAL, F, G, GZU, HB, HPL, HTO, HUEFS, HUFABC, IBGE, IAN, IPA, K, L, LE, MBM, MG, MO, NY, P, R, RB, S, SP, SPF, U, UB, UFG, US, W, and WAG; acronyms following Thiers 2015). We have studied the species in their natural habitats during field excursions in 2002, 2004, 2005, and 2013 in Brazil. This resulted in the collection of new specimens that were deposited in the herbaria ESA, G, HPL, and HUFABC. Details of the herbarium material

consulted are listed in the taxonomic treatment. Morphological observations and measurements were done on living plants cultivated in the private collection of Mauro Peixoto at Sítio Primavera near Mogi das Cruzes (SP, Brazil), in Instituto Plantarum in Nova Odessa (SP, Brazil), and at the Conservatoire et Jardin botaniques de la Ville de Genève in Switzerland. Geographic occurrences were obtained from labels of herbarium specimens and geolocalized specimens in the field. Species distributions were generated and mapped using ArcView GIS 3.2 (ESRI 2000).

**Phylogenetic Analysis**—Representatives of all four recognized species of *Sphaerorrhiza* were sampled. To investigate the tribal position of *Sphaerorrhiza*, 42 other genera of Gesnerioideae were sampled, representing all tribes and subtribes of the subfamily and 72% of the genera. The monotypic genus Sanago Bunting & Duke [*S. racemosum* (Ruiz & Pav.) Barringer], which has been shown to be sister to all other Gesneriaceae (Perret et al. 2013), was used as the outgroup.

Total DNA was isolated from silica-gel dried leaf tissue using the NucleoSpin Plant II (Macherey-Nagel, Düren, Germany) following the manufacturer's protocol. Sequences of *atpB-rbcL*, *matK*, *rpl16* and *rps16* introns, *trnL-F* intron-spacer, and ITS were newly acquired for this study or gathered from published data (Perret et al. 2003; Araujo et al. 2010; Perret et al. 2013; Serrano-Serrano et al. 2015). The primers used to amplify and sequence the *atpB-rbcL*, *rpl16* intron, *trnL-F* intron-spacer and ITS are indicated in Perret et al. (2003) and Araujo et al. (2010). The *matK* region was amplified and sequenced using the two primers pairs *matK1F/matK943R* and *matK762F/matK1R* as described in Perret et al. (2013). Species names, voucher information and GenBank accession numbers for sequences used in this study are provided in the Appendix 1.

All DNA regions were initially aligned with Muscle (Edgar 2004) and then imported into Mesquite 2.75 (Maddison and Maddison 2011) for verification and manual adjustment. Regions of uncertain homology in ITS, totaling nine characters (positions 519–527), were excluded from the dataset. For parsimony analyses, all characters were equally weighted and gaps were treated as missing characters. Heuristic searches were conducted in PAUP\* 4.0b10 (Swofford 2002), using 1,000 random taxon addition sequence replicates with tree-bisection-reconnection (TBR) branch swapping. To evaluate internal support, heuristic searches were completed for 1,000 bootstrap replicates (BS; Felsenstein 1985), with TBR swapping. The number of trees saved at each replicate was limited to 1,000. Bayesian inferences (BI) were performed with MrBayes version 3.1 (Ronquist and Huelsenbeck 2003). The combined data set was divided into two partitions: ITS and plastid DNA sequences (*atpB-rbcL*, *matK*, *rpl16*, *rps16*, and *trnL-trnF*). For each partition, the optimal model of DNA substitution was selected by the Akaike information criterion (AIC) using Modeltest 3.7 (Posada and Crandall 1998). In each case the optimal model was the general time reversible model, with rate heterogeneity modeled by assuming that some proportion of sites are invariable and that rate of evolution at other sites is modeled using a discrete approximation to a gamma distribution (GTR+I+ $\Gamma$ ). All parameter values were unlinked across partitions and estimated during the Markov Chain Monte Carlo (MCMC) runs. Two independent analyses were run from different random trees ( $N_{runs} = 2$ ) with temperature of 0.15. The chains were run for 10,000,000 generations, with trees sampled every 100th generation. We discarded 25% of the samples obtained during the first 1,000,000 generations. Posterior probability (PP) and branch lengths were averaged across remaining sampled trees. To examine potential incongruences between datasets, we inspected whether the topology resulting from the combined analyses of all the regions conflicted with the topologies obtained through separate analyses of the plastid and nuclear sequences. Only conflicting nodes with 70% BS or higher were considered.

**Morphology and Underground System Anatomy**—Morphology of the underground system was observed in all *Sphaerorrhiza* species based on samples collected from native populations and from cultivated plants of wild origin. Anatomical studies of underground storage organs were performed in *Sphaerorrhiza sarmentiana* (voucher = Araujo 1099). Samples were obtained from material stored in 70% ethanol, dehydrated, and embedded using standard methods for Leica historesin (Heraeus Kulzer, Hanau, Germany). Cross or longitudinal sections (10  $\mu$ m thickness) were stained with toluidine blue O (Sakai 1973) and mounted in water. Observations and digital images were captured with an Olympus BX53 compound microscope equipped with an Olympus Q-Color 5 digital camera and Image Pro Express 6.3 software. Evolution of the underground system in Gesnerioideae was reconstructed on the Bayesian 50% majority rule consensus tree using maximum likelihood method and the Mk1 model as implemented in Mesquite 2.75 (Maddison and Maddison 2011). Underground storage organs were coded as perennial tuber, moniliform tuber, scaly rhizome, or absent, based on examination of live material as well as reports from the literature (e.g. Weber 2004).

**Chromosome Number**—Mitotic chromosome number of *Sphaerorrhiza sarmentiana* was investigated using material collected in the region of Ipu, Ceará (Brazil) and propagated in the living collection of the Instituto Plantarum (voucher = Lorenzi 2684). Growing root tips of the underground structure were prepared by removing the epidermis. This material was pre-treated with hydroxyquinoline (300 ppm) and cycloheximide (3.12 ppm) for 3 hr at 28°C, and then fixed in Carnoy fixative (= Farmer's

Fluid; see Jong and Möller 2000) (3 ethyl alcohol: 1 acetic acid glacial P. A.) for 24 hr at room temperature. After a washing step in distilled water for 5 min, the material was hydrolyzed in 1N HCl for 8 min at 60°C, washed again, and transferred to Schiff reagent for 45 min in the dark. Finally, the tips were washed for 10 min, put in 45% acetic acid for 3 min and mounted on slides with a drop of 45% acetic acid, then squashed and stained with 1% acetic carmine.

## Results

**Phylogenetic Analysis**—The combined matrix of all DNA regions comprised a total of 6,530 characters with 1,881 variable sites and 901 parsimony-informative characters. Of these, 5,809 (1,447 variable, 582 parsimony-informative) were derived from plastid sequences and 721 (434 variable, 319 parsimony-informative) from ITS. The result of the BI analyses is shown in Fig. 1. The comparison of this tree with results derived from separate analyses of the nuclear and plastid datasets did not indicate supported topological incongruences (BS > 70%). Our results support the monophyly of *Sphaerorrhiza* (1.0 PP, 100% BS) and its placement in the tribe Gesnerieae (1.0 PP, 100% BS). Subtribes currently recognized by Weber et al. (2013) are all found to be monophyletic (1.0 PP, 100% BS) but their relationships are not resolved. Indeed, *Sphaerorrhiza* forms a polytomy with *Gloxiniinae* and *Columnneinae*, and the position of *Ligeriinae* as sister to this clade is not supported (0.55 PP, BS < 50%).

Relationships among the four species of *Sphaerorrhiza* are fully resolved but our sampling, limited to one accession per species, does not allow testing of their monophyly.

**Morphology and Underground System Anatomy**—*Sphaerorrhiza sarmentiana* presents an underground structure that can be described as a moniliform tuber, as it resembles a string of beads composed of swellings of a few mm connected by a narrow thread-like stalk (Fig. 2A–C). The whole structure is a tuber derived from stem, in which the protuberances are composed of a single epidermis, many layers of starch parenchyma and medullar vascular bundles (Fig. 2D). Developing vegetative buds can be visualized at the cortical region (Fig. 2E). The internal structure is maintained along the narrow thread-like stalk (Fig. 2E), differing only in the number of layers of starch parenchyma. Thus, at longitudinal view, it is possible to visualize the continuous vascularization along the narrow region of the tuber (Fig. 2E). Ancestral state reconstruction indicates that this underground structure is a synapomorphy for *Sphaerorrhiza*, which differs from the perennial tuber found in the *Ligeriinae* or the scaly rhizomes that characterize the *Gloxiniinae* (Fig. 1).

Besides the underground system, we observed that sepals in all species of *Sphaerorrhiza* are valvate and sealed in buds, and that the corolla is always glabrous. These features are useful additional characters for distinguishing *Sphaerorrhiza* from other *Gloxiniinae* genera with similar flower morphology, including *Mandirola* and *Goyazia* which sometimes co-occur with *Sphaerorrhiza* in central Brazil. The four recognized species in *Sphaerorrhiza* differ by traits such as plant size, leaf blade, type of florescence, and corolla shape and color (Figs. 3–5; Table 1).

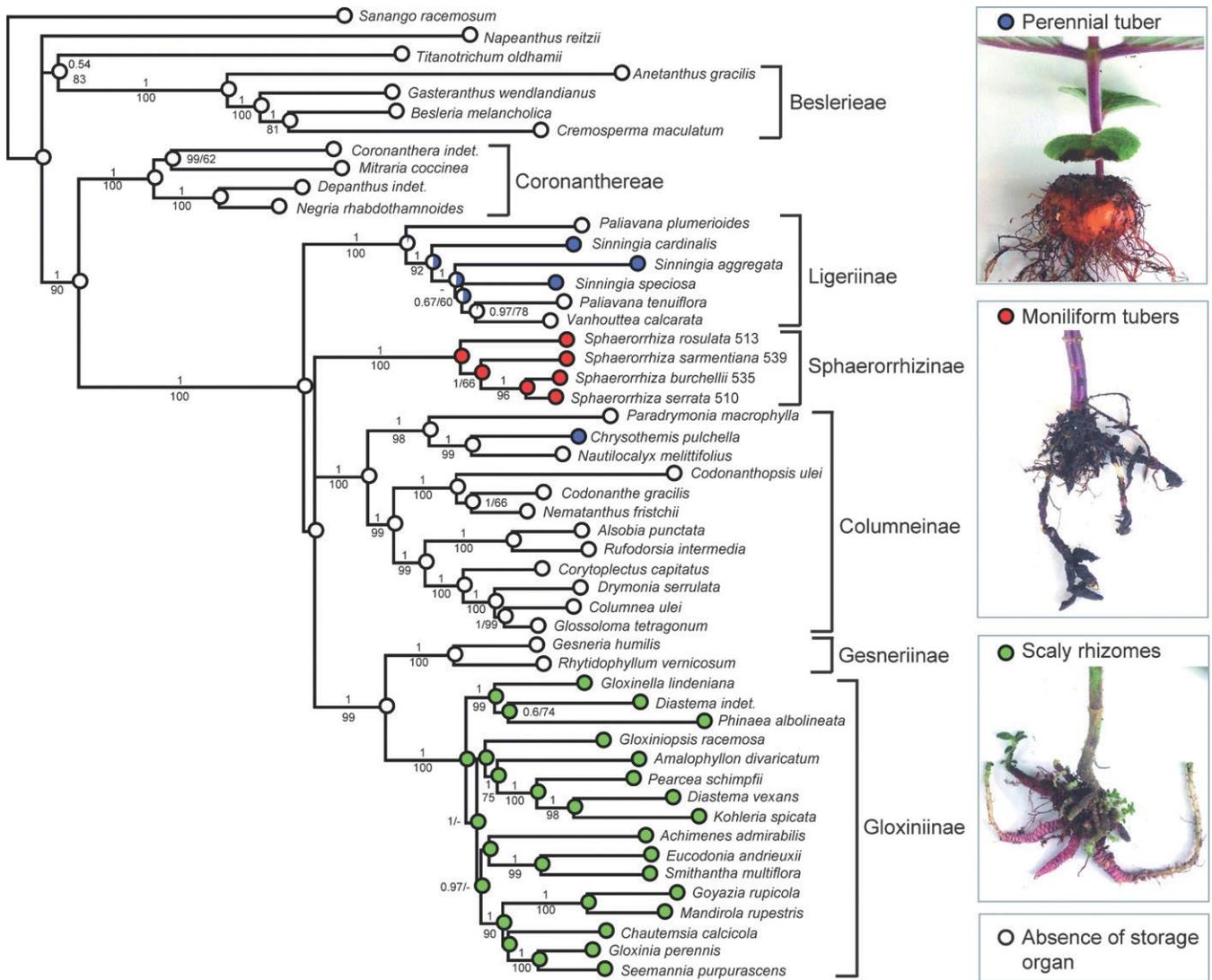


Fig. 1. Bayesian 50% majority rule consensus tree resulting from the combined analysis of the *atpB-rbcL*, *matK*, *rp116* intron, *rps16* intron, *trnL-F* region, and ITS datasets including 51 accessions representing all tribes of Gesnerioideae. Numbers above branches are Bayesian posterior probabilities and those below are parsimony bootstrap values when  $\geq 50\%$ . Types of underground storage organs are coded as perennial tuber (blue), moniliform-like tuber (red), scaly rhizomes (green) and absent (white). Pie charts represent the ancestral state reconstructed at each node using maximum likelihood. On the right-hand side, images of a tuber of *Sinningia canescens* (Mart.) Wiehler (top), moniliform tuber of *Sphaerorrhiza sarmentiana* (middle), and scaly rhizomes of *Kohleria warszewiczii* (Regel) Hanst. (bottom) are shown to represent each type of underground storage organ. Classification follows Weber et al. (2013).

**Chromosome Number**—A chromosome number  $2n = 26$  was counted for *Sphaerorrhiza sarmentiana*. This is the first report for the genus.

**Discussion**

**Phylogenetic Position of *Sphaerorrhiza***—Our phylogenetic analysis clearly supports that the species currently recognized in *Sphaerorrhiza* form a monophyletic group. In agreement with previous results, we found that *Sphaerorrhiza* is not embedded in any other subtribes of Gesnerieae, justifying the treatment of this genus in its own subtribe Sphaerorrhizinae (Roalson et al. 2005a; Weber et al. 2013). Relationships among *Sphaerorrhiza* and the other monophyletic subtribes Columneinae, Gesneriinae, Gloxiniinae and Ligeriinae are not resolved. Previous results based on a more limited number of DNA loci have suggested that *Sphaerorrhiza* could be

sister to a clade composed of Ligeriinae and Columneinae (Zimmer et al. 2002), or to all other Gesnerieae subtribes (i.e. Gloxiniinae, Gesneriinae, Columneinae), although none of these relationships were supported (Perret et al. 2013). Unfortunately, our analysis, based on a larger amount of molecular characters, did not provide additional resolution and support. This lack of resolution could indicate that the main clades of Gesnerieae diverged within a short period of time, probably during the Oligocene/Miocene boundary (Perret et al. 2013).

**Morphology and Underground System Anatomy**—Although the underground structure of *Sphaerorrhiza* had been mentioned in the literature (Zimmer et al. 2002; Roalson et al. 2005a, 2005b), all the observations were based on a single accession of *S. sarmentiana*, originally collected in Goiás (Chautems et al. 407). The added accessions of *S. sarmentiana* and the three other recognized species allowed us to characterize this structure more accurately. This underground structure

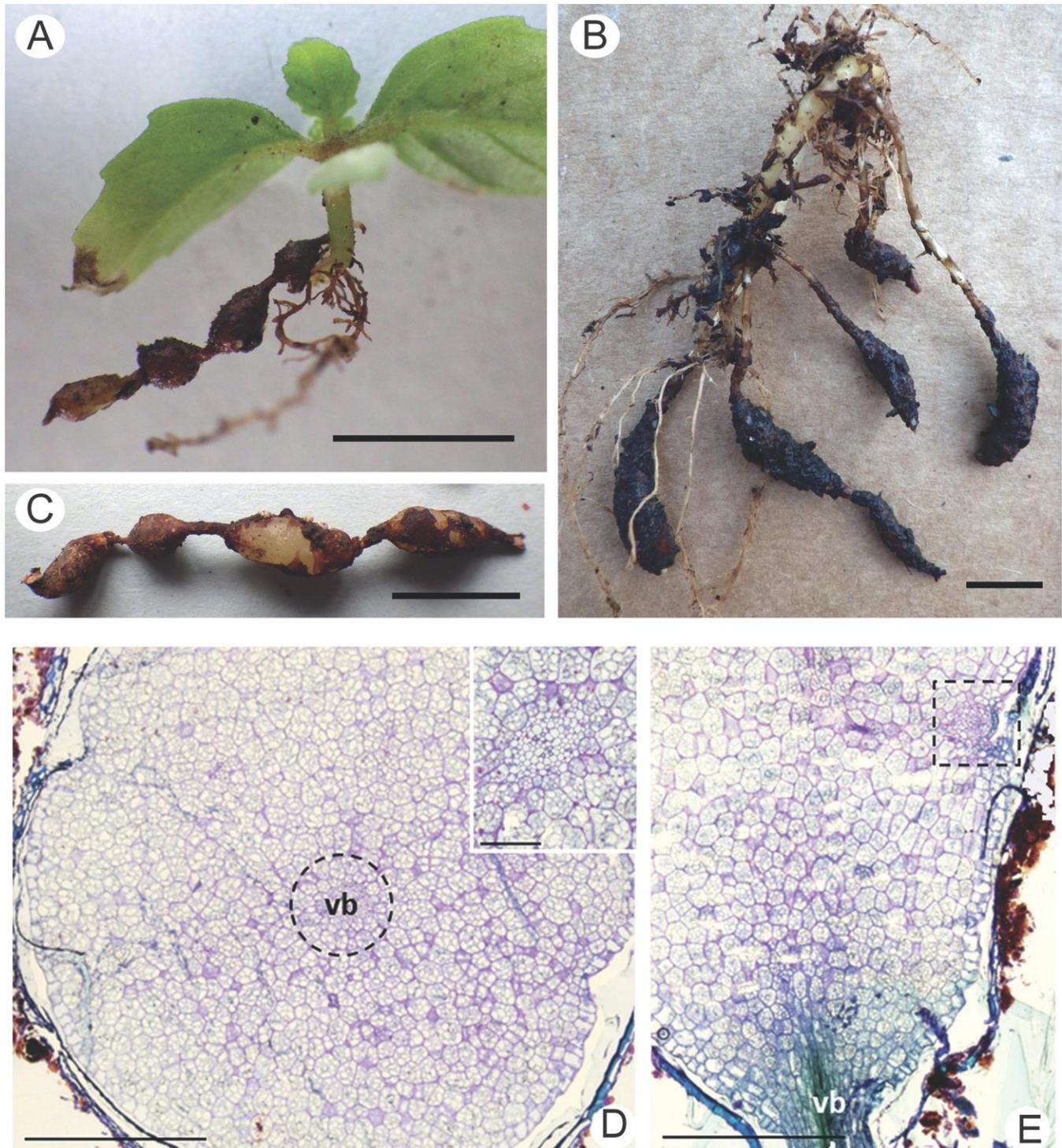


Fig. 2. Morphology and anatomy of the underground storage organs in *Sphaerorrhiza*. A. Young plant of *S. serrata* with moniliform tubers. B. Moniliform tubers of *S. sarmentiana*. C. Moniliform tubers of *S. burchellii*. D–E. *Sphaerorrhiza sarmentiana*. D. Transverse section of the swelling; it presents many layers of starch parenchyma and vascular bundles (vb) in the medullar region (inset). E. Longitudinal section of the region connecting two swellings; note the longitudinal view of vascular bundles at the narrow portion, while a vegetative bud (dashed square) can be observed in the large portion of the swelling. Scale bars: 5mm (A–C); 300  $\mu$ m (D, E); 75  $\mu$ m (inset in D). A: (cult. in Geneva under AC-3804). B: (cult. at Instituto Plantarum, Lorenzi 2684). C: (cult. in Geneva under AC-3806). D–E: (Araujo 1099).

is easier to observe in plants in cultivation because the tuberous swellings are only a few mm long and wide and in the field they may be unnoticed or confused with small stones in the ground. The structure is fragile and the swellings break

apart easily. The presence of amyloplasts indicates that the structure has a storage role that allows the plant to survive until the next season after the aerial stems dry out at the end of the growing period. New storage organs are produced each

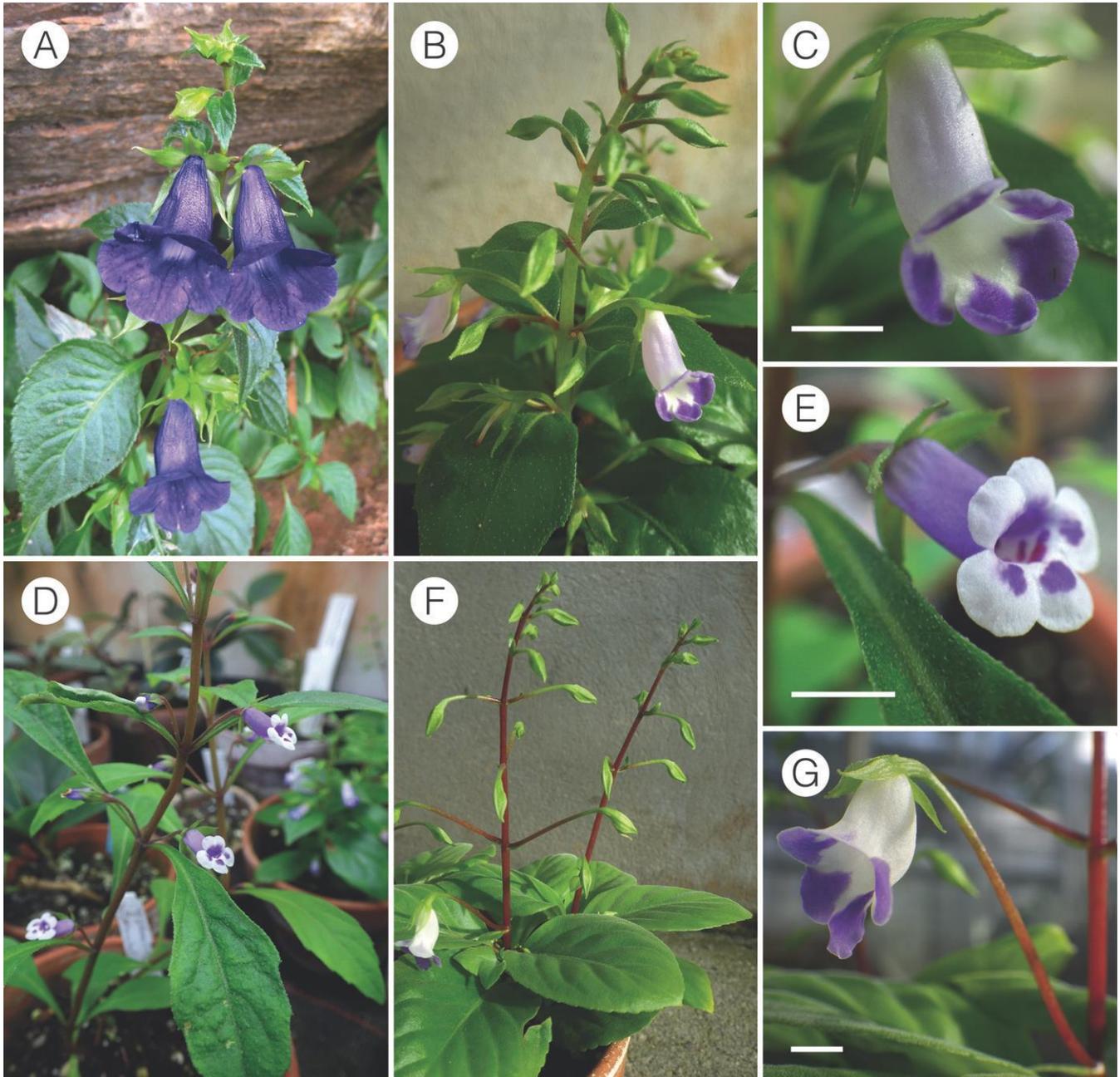


Fig. 3. Pictures of the four *Sphaerorrhiza* species. A. *Sphaerorrhiza sarmentiana*. B–C. *S. serrata*. D–E. *S. burchellii*. F–G. *S. rosulata*. Scale bars: 5 mm. A: Photo by V. Dittrich. B–C: (cult. in Geneva under AC-3804). D–E: (cult. in Geneva under AC-3806). F–G: (cult. in Geneva under AC-3805).

year and give rise to new leafy stems that eventually develop flowers, enabling sexual reproduction. These aerial stems then dry out at the end of the growing season and the plant survives until the next season only through the underground storage organs. These tubers play an effective role for vegetative propagation, each swelling acting as a propagule, when it breaks apart from the moniliform tuber structure. The ability of producing additional buds is likely the first step to the production of new plantlets.

Underground storage structures and vegetative propagation exist in other clades of neotropical Gesneriaceae, but they involve different parts of the plants (Fig. 1). In Gloxininiinae, the scaly rhizomes represent appendages of the

shoot axis that are formed by a thin central stalk closely packed with pairs or whorls of small, fleshy leaf scales that are the starch-storage organs (Kvist and Skog 1992; Weber 2004). Scales constitute the propagation units, able to sprout and produce new annual shoots when favorable conditions return. By contrast, in Ligeriinae, the perennial tubers are hypocotyledonous structures from which new shoots are produced each growing season. Therefore, the tubers of the Sphaerorrhizinae are not homologous to the underground storage organs found in the other clades of Gesnerioideae.

**Chromosome Numbers**—Chromosome numbers are used as a diagnostic feature in Gesnerioideae, as they are fairly stable within subtribes. The values range from  $n = 8$  to  $n = 9$

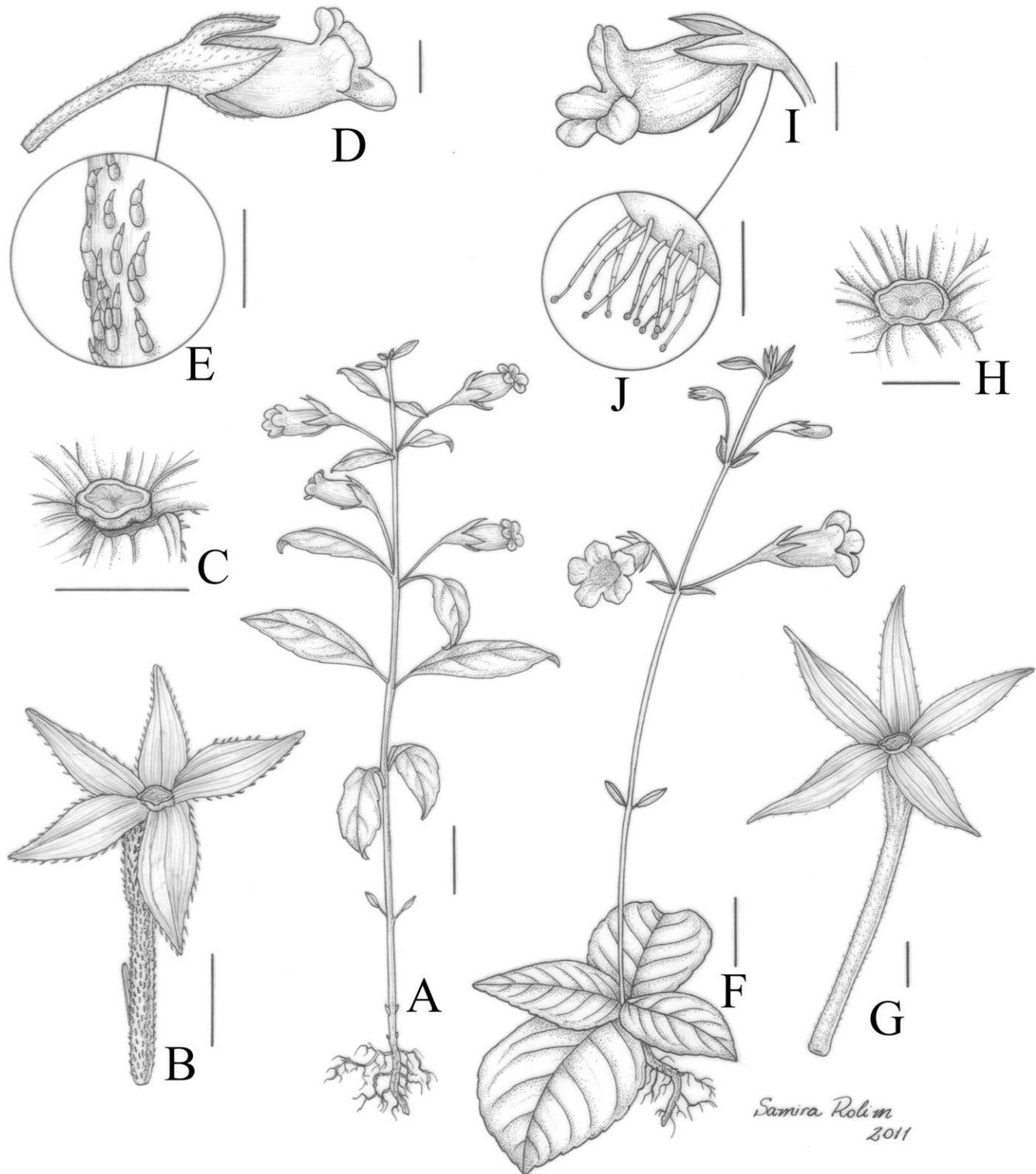


Fig. 4. A–E. *Sphaerorrhiza burchellii*. A. Habit. B. Pedicel, calyx, and nectary (gynoecium removed). C. Nectary in detail. D. Flower. E. Detail of the calyx trichomes. F–J. *Sphaerorrhiza rosulata*. F. Habit. G. Pedicel, calyx, and nectary (gynoecium removed). H. Nectary in detail. I. Flower. J. Detail of the calyx trichomes. Scale bars: 1 cm (A–F); 3 mm (B, G); 2 mm (C, H); 2 mm (D, I); 0.5 mm (E, J). A: (Calago 94). B–E: (Peixoto 28). F, I, J: (Araujo 535). G–H: (Peixoto 27).

in Columneinae,  $n = 13$  in Ligeriinae,  $n = 14$  in Gesneriinae, and  $n = 16$  in Besleriinae (Wiehler 1983; Möller and Pullan 2016). In Gloxiniinae the chromosome numbers are more variable: from  $n = 10$  to  $n = 13$  (Wiehler 1983; Skog 1984). The chromosome number found in *Sphaerorrhiza sarmentiana* is

$n = 13$ . This number is identical to the number found in Ligeriinae and in some genera included in Gloxiniinae (e.g. *Diastema*, *Seemannia*, *Gloxinia*) (Möller and Pullan 2016). Therefore it cannot be used as a discriminating feature for defining *Sphaerorrhiza*. Further studies should be done with

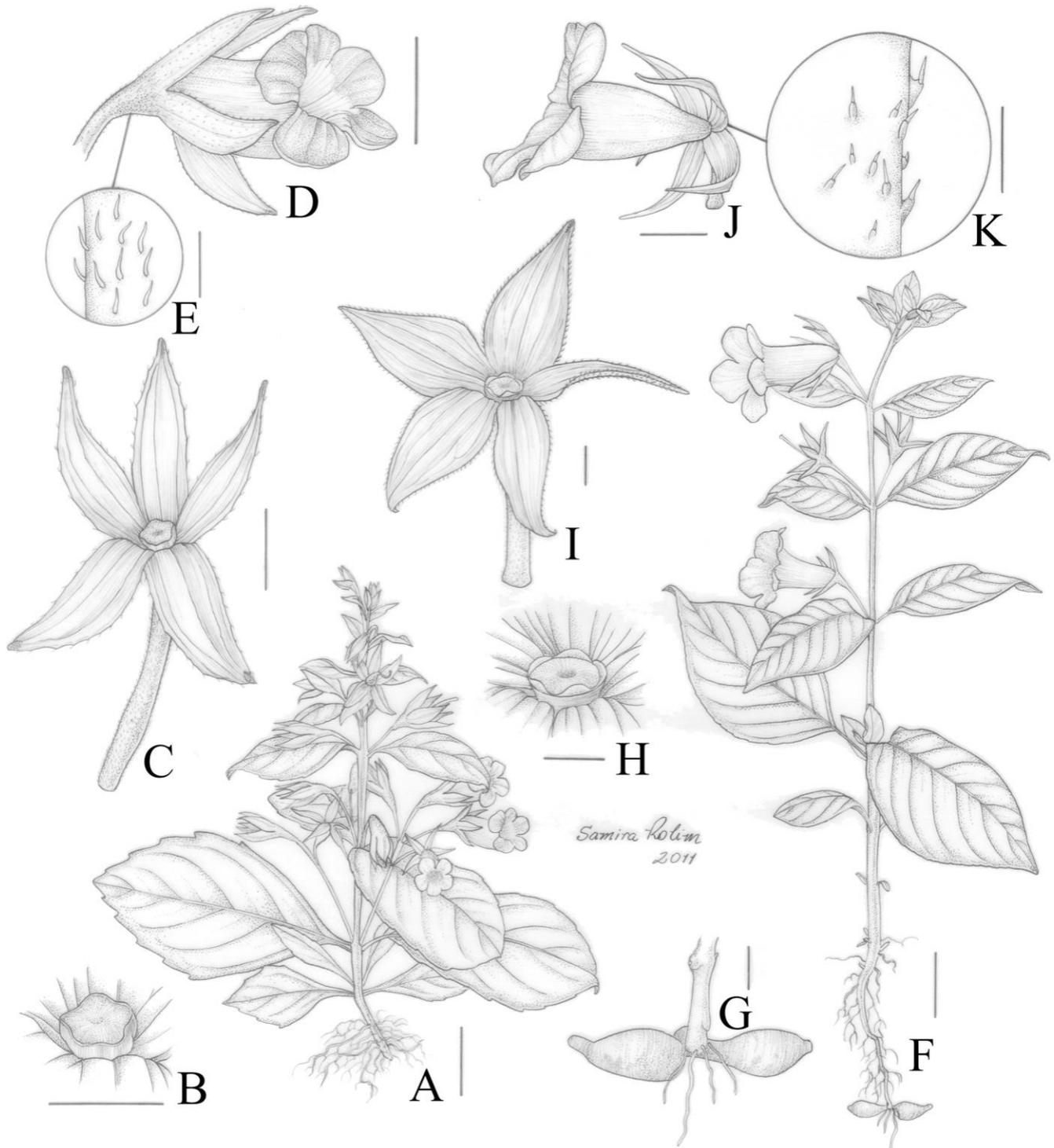


Fig. 5. A–E. *Sphaerorrhiza serrata*. A. Habit. B. Nectary in detail. C. Pedicel, calyx, and nectary (gynoecium removed). D. Flower. E. Detail of the calyx trichomes. F–K. *Sphaerorrhiza sarmentiana*. F. Habit. G. Underground system. H. Nectary in detail. I. Pedicel, calyx, and nectary (gynoecium removed). J. Flower. K. Detail of the calyx trichomes. Scale bars: 1 cm (A, J); 2 mm (B, H); 3 mm (C, I); 5 mm (D); 0.5 mm (E, K); 2 cm (F, G). A: (Peixoto 26). B–E: (Araujo 510). F (Souza 28143). G–K: (Lorenzi 2684).

the three other species in order to check if this character is shared throughout the whole genus.

#### Taxonomic Treatment

*Sphaerorrhiza* Roalson & Boggan, *Selbyana* 25(2): 236 (2005).—TYPE: *S. sarmentiana* (Gardner ex Hook.)

Roalson & Boggan (= *Gloxinia sarmentiana* Gardner ex Hook.).

Perennial herbs, terrestrial. Underground structure with small moniliform tuber. Stems usually unbranched. Leaves opposite, subequal or equal, rarely whorled, petiolate, blade margin serrate, serrulate or crenate, trichomes evenly spread on the adaxial surface, 3–7(–8) pairs of secondary veins.

Table 1 Comparisons of morphological characters among species of *Sphaerorrhiza*.

	<i>S. burchellii</i>	<i>S. rosulata</i>	<i>S. sarmentiana</i>	<i>S. serrata</i>
Plant size (cm)	8–25	3–10	(3.5–)5–50	4–8
Leaf distribution	Along the stem	Clustered at the base of the plant	Along the stem	Along the stem
Leaf blade size (cm)	0.8–4.5 × 0.4–1.8	1.2–5.3 × 0.6–4.5	2.4–16.0 × 1.3–9.1	0.7–5.0 × 0.5–1.9
Leaf blade margin	Serrulate, entire at base	Crenate, sometimes only sinuate at apex	Serrate or serrulate, sometimes irregularly serrate, rarely weakly crenate	Serrate, entire at base
Florescence	Frondose	Bracteose	Frondose	Frondo-bracteose
Corolla tube length (cm)	0.5–0.9	0.7–0.8	1.2–3.0	0.5–0.7
Corolla color:				
- Tube (outside)	Violet or lilac with darker spots on the ventral face	Always white	Purple or dark violet with pink to white base	Always white
- Lobes	White with lilac or violet spots	Lilac	Concolor or with some marks slightly lighter than the rest of the corolla	Lilac
Nectary	Unlobed annular disk, inconspicuous	Unlobed annular disk, inconspicuous	5 separate glands or unlobed annular disk, conspicuous	Unlobed annular disk, inconspicuous

Florescence frondose, sometimes bracteose or frondo-bracteose, with solitary flowers, rarely a fascicle of 2 flowers with a peduncle. Flowers zygomorphic, without bracteoles; calyx aestivation valvate, sepals free, equal in size; corolla narrowly campanulate, dorsiventrally depressed or not, or campanulate and with a small ventral gibbosity in the middle of the tube, aestivation imbricate, generally totally glabrous, tube white, violet, purple, or lilac outside or white at the base and purple at the apex, or dark-violet with pinkish or white base; throat not constricted or barely constricted; lobes patent, of the same color as corolla tube, or lilac, pink, or white when the tube is white, or white when the tube is violet or lilac, depressed-obovate or depressed-orbicular, entire, sinuate or slightly emarginate at tip; stamens 4 included in the corolla, staminode present, anthers opening by longitudinal slits; nectary an unlobed and annular disk or composed of 5 free glands; ovary inferior to semi-inferior; style glabrous; stigma stomatomorphic or bilobed. Capsule dry, dehiscing only apically, with prominent costae at base. Seeds numerous, minute, 0.1–0.5 mm.

**Distribution and Ecology**—The range of distribution of *Sphaerorrhiza* encompasses an area of about 1.2 million km<sup>2</sup> that includes the basin of the Paraná, São Francisco, Tocantins-Araguaia, and Amazonas rivers and also a small part of the South Atlantic Basin (North and Northeast region). According to the phytogeographic domains proposed by Fiaschi and

Pirani (2009), this genus is endemic to the Cerrado. Machado et al. (2004) estimated that 70% of the herbaceous and shrubby plants that grow in the Cerrado are endemic to this domain, *Sphaerorrhiza* being a good example. All the species of *Sphaerorrhiza* are terrestrial growing on rocky outcrops located in semi-shady to shady gallery forest or in open, sun-exposed locations in savannah. These outcrops can be limestone, sandstone, or clay. *Sphaerorrhiza* presents a wide altitudinal distribution range, at elevations between 200 and 1,100 m. *S. burchellii* occurs at the lowest altitudes in the town of Porto Nacional (TO), *S. serrata* grows around 950 m, whereas *S. rosulata* thrives at the highest altitudes near Chapada dos Veadeiros (GO). The most widespread species of the genus, *S. sarmentiana*, covers most of the Cerrado area, including at least a part of the states of Bahia, Ceará, Goiás, Mato Grosso, Minas Gerais, Pará, Piauí, Tocantins, and Distrito Federal (Fig. 6). The three other species present a much more restricted distribution. *S. rosulata* was found so far only in a preserved area inside a conservation unit (RPPN Raizama), near the Chapada dos Veadeiros National Park in Goiás state. *S. burchellii* is found in the Tocantins river valley (center of Tocantins state) and *S. serrata* occurs in one locality near the city of Brasília in the Distrito Federal. This last species is the only one that occurs at rather short distance (a few km) from the widespread *S. sarmentiana*.

#### Key to the Species of SPHAERORRHIZA

1. Plants with all leaves clustered at the base, appearing like a rosette, leaf blade margin crenate, sometimes sinuate only at the apex; florescence bracteose . . . . . *S. rosulata*
1. Plants with leaves distributed along the stem, leaf blade margin serrate or serrulate, base entire or not; florescence frondose or frondo-bracteose . . . . . 2
  2. Leaf blades 2.4–16.0 × 1.3–9.1 cm; calyx lobes 1.0–1.9 × 0.3–0.7 cm; corolla tube 1.2–3.0 cm long; throat 1.0–2.3 cm diam . . . . . *S. sarmentiana*
  2. Leaf blades 0.7–5.0 × 0.4–1.9 cm; calyx lobes 0.3–0.6 × 0.1–0.3 cm; corolla tube 0.5–0.9 cm long; throat 0.4–0.5 cm diam . . . . . 3
    3. Herbs 4–8 cm tall; leaf blade margin serrate; florescence frondo-bracteose; corolla tube white outside and lobes lilac; plants from Distrito Federal . . . . . *S. serrata*
    3. Herbs 8–25 cm tall; leaf blade margin serrulate; florescence frondose; corolla tube violet outside or lilac with darker spots on the ventral face, lobes white with lilac or violet spots; plants from Tocantins . . . . . *S. burchellii*

1. *Sphaerorrhiza burchellii* (S. M. Phillips) Roalson & Boggan, Selbyana 25(2): 225–238 (2005). *Achimenes burchellii* S. M. Phillips, Kew Bull. 24(1): 225. 1970. *Gloxinia burchellii* (S. M. Phillips) Wiehler, Selbyana 1(4): 387. 1976.—TYPE: BRAZIL. Tocantins: Porto Real [nowadays

Porto Nacional], 08 Feb 1829, W. J. Burchell 8615 [holotype: K! (K000479984); isotypes: WAG!, L!].

Herbs 8–25 cm tall. Internodes 1–3.5 cm long. Leaves distributed all over the plant, opposite, equal to subequal in

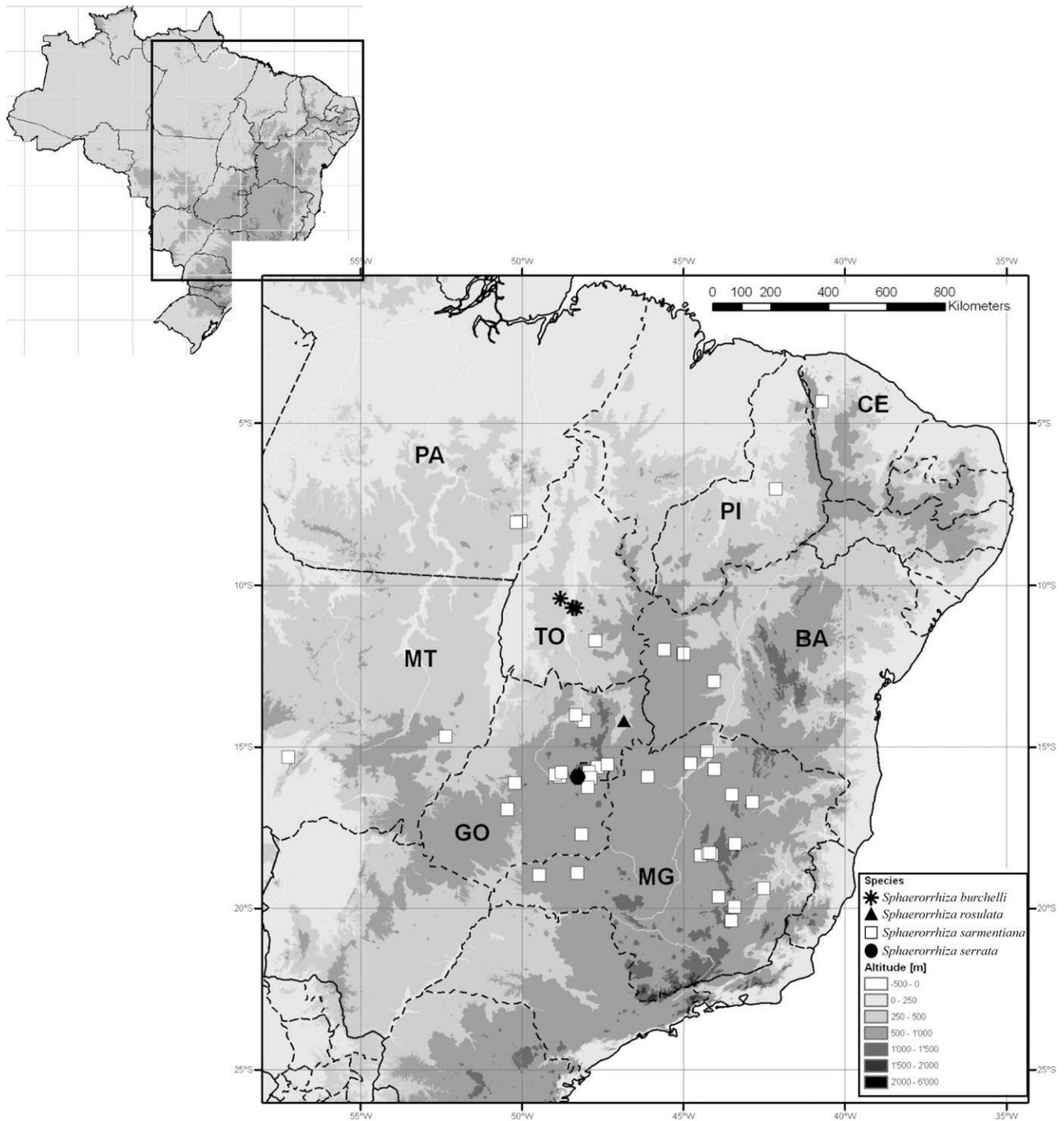


Fig. 6. Distribution of *Sphaerorrhiza burchellii*, *S. rosulata*, *S. sarmentiana* and *S. serrata* in Brazil.

pairs; petioles 0.1–1.5 cm long; blades 1–5 × 0.4–1.8 cm, elliptic or ovate, rarely obovate, apex acute to acuminate, base cuneate and decurrent into the petiole, margin serrulate, entire at base, above sparsely pubescent, below glabrous or sparsely pubescent with trichomes only on the veins, secondary veins 3–4 pairs. Florescence frondose, with solitary flowers, opposite or alternate in the stem, rarely 2–4 flowers in the same axis; subtending leaves 1.0–2.0 × 0.3–0.8 cm, flowering internodes 0.5–2.0 cm long. Flowers with pedicel 0.5–1.8 cm long, reddish; calyx green, lobes 0.3–0.5 × 0.15–0.3 cm, outside sparsely pubescent, inside glabrous, lanceo-

late or elliptic, margin entire, frequently ciliate; corolla axis slightly oblique in relation to pedicel; corolla narrowly campanulate, totally glabrous, with a small ventral gibbosity in the middle of the tube, tube outside violet or lilac with darker spots on the ventral face, tube 0.5–0.9 cm long; throat without differentiated coloring or throat with lilac dots and the tube with red dots or tube and throat with violet dots, not constricted or barely constricted, 0.4–0.5 cm diam; lobes white with lilac or violet spots, about 0.2 cm long, entire or slightly emarginate at tip; stamens 0.3–0.4 cm long, included, filament glabrous; nectary an unlobed and annular disk, inconspicuous;

ovary inferior; stigma stomatomorphic, sometimes bilobed. Capsule about  $0.5 \times 0.2\text{--}0.3$  cm, pubescent, fusiform, apex pointed upwards.

**Distribution and Ecology**—This species occurs in restricted areas of the northern part of the Cerrado, in Tocantins state (Fig. 6), where it is found in sandy-clay outcrops within gallery forests.

**Phenology**—Flowering and fruiting from February to April. During the dry season, from May to September, plants are dormant and reduced to their underground system hidden in rock crevices. This species is in cultivation since 2005 and seems to have kept the same rhythm of flowering, fruiting or dormancy periods as in its natural environment.

**Conservation status**—The species is critically endangered, based on criteria such as extent of occurrence ( $<100$  km),<sup>2</sup> number of mature individuals ( $<250$ ), and a continuing decline, and less than 50 mature individuals in the largest subpopulation (IUCN 2001). Following the Brazilian list of threatened plant species, this species is endangered (MMA 2014). However, in that list three subpopulations were included in the analysis, totaling 5,000 km<sup>2</sup> of extent of occurrence for the species.

**Notes**—The first documented collection of this species dates back to 1829 in Porto Nacional (TO), but the species was only recognized and described in 1970 in the genus *Achimenes* Pers. After some 140 yr, it was collected in Paraíso do Tocantins (Irwin et al. 21737) in 1968, and in the vicinity of the type locality (Boracão, in Porto Nacional — TO) in 2005, where it has survived despite the urbanization of the area. Burchell's "Catalogus Geographicus Plantarum Brasiliae Tropicae" refers to the locality in the following terms: "in a walk to the Boracão, bearing south of my house about a mile; the Boracão is a dark woody glen in which rises a small rivulet, overshadowed by large forest trees." Although the species is in cultivation since 2005, additional environmental restoration and in situ conservation measures should urgently be taken to guarantee its survival in nature, because degradation of the site near Porto Nacional is likely to occur, as it sits within an urban area.

**Additional Specimens Examined**—BRAZIL. Tocantins: Porto Nacional, margem direita do Rio Tocantins, mata de galeria a poucos metros do riacho, 26 Apr 1994, Alves & Santo 1233 (HTO); Porto Nacional, Buracão, próximo à igreja São Judas Tadeu, local bastante alterado, rochas formando uma pequena baía na margem direita do Rio Tocantins, 250 m alt.,  $10^{\circ}42'57.3''\text{S } 48^{\circ}25'05.2''\text{W}$ , 27 Jan 2005, Araujo et al. 535 (ESA); ca. 24 km S of Paraíso, soil-filled crevices, in outcrops, and rocky slopes, ca. 600 m alt., 24 Mar 1968, Irwin et al. 21737 (F, K, NY, MO, RB, SEL, UB, WAG). In cultivation: São Paulo, Mogi das Cruzes. Introduzido em cultivo no sítio Primavera, Biritiba Ussu, perto de Mogi das Cruzes (a partir de material originalmente coletado em botão, Araujo et al. 535) e quando floresceu a presente exsicata foi preparada, 10 Feb 2011, Peixoto 27 (HPL).

2. *Sphaerorrhiza rosulata* A. O. Araujo & Chautems, sp. nov.—**TYPE**: BRAZIL. Goiás: Alto Paraíso de Goiás, próximo a São Jorge; beira de barranco, local sombreado, 894 m alt.,  $14^{\circ}11'39''\text{S } 47^{\circ}50'27''\text{W}$ , 14 Mar 2007, J. F. B. Pastore, R. M. Harley & A. A. Santos 1849 (holotype: HUEFS!).

Ab omnibus speciebus generis foliis omnis congestis basi plantae laminarum margine crenato et inflorescentia racemosa differt.

Herbs 3–10 cm tall. Internodes about 0.1 cm long. Leaves clustered at the base of the plant, at most 3 pairs of leaves each plant, opposite, apparently rosulate because of the short internodes, subequal in pairs; petioles 0.1–0.3 cm long; blades  $1.2\text{--}5.3 \times 0.6\text{--}4.5$  cm, orbicular or elliptic, apex rounded or

acute, base symmetrical, cuneate, margin crenate, sometimes sinuate only at the apex, above pubescent or sericeous, below glabrous to sparsely pubescent only on the veins or sericeous, secondary veins 4–5 pairs. Florescence bracteose in the stem apex, with internodes 1.0–1.7 cm long, flowers solitary opposite in the stem; bracts  $0.2\text{--}0.8 \times 0.1\text{--}0.5$  cm. Flowers with pedicel 0.6–3.0 cm long; calyx green, reddish when dry, lobes  $0.3\text{--}0.6 \times 0.1\text{--}0.3$  cm, sparsely pubescent in both faces, lanceolate, margin entire and ciliate; corolla axis oblique to pedicel; corolla campanulate, with a ventral gibbosity in the middle of the tube, tube always white, 0.7–0.8 cm long, corolla totally glabrous or tube sparsely pubescent outside with glandular trichomes; throat white, not constricted, about 0.6 cm diam; lobes lilac, 0.3–0.4 cm long, margin entire or sinuate or slightly emarginate at tip; stamens 0.3–0.4 cm long, filament glabrous to puberulent; nectary an unlobed and annular disk, inconspicuous; ovary inferior to semi-inferior; stigma stomatomorphic. Capsule about  $0.7\text{--}0.9 \times 0.3$  cm, pubescent, fusiform, apex pointed upwards.

**Distribution and Ecology**—The species has been found so far only in Chapada dos Veadeiros (area of Cerrado) (Fig. 6), in rocky outcrops (clayey) in gallery forest, on river banks.

**Phenology**—In the field, the species was only found with buds and old fruits in January. During the dry season, from May to September, plants are dormant and reduced to small tubers hidden in rock crevices.

**Etymology**—From Latin *rosulata* = rosulate. The specific epithet makes reference to the leaves clustered at the base of the plant, apparently rosulate because of the short internodes. This trait helps distinguish this species from the other species of *Sphaerorrhiza*, which present leaves distributed along the stem.

**Conservation Status**—The species is critically endangered, based on criteria such as extent of occurrence ( $<100$  km),<sup>2</sup> known to exist at only a single location and continuing to decline in area, extent and/or quality of habitat (IUCN 2001).

**Notes**—*Sphaerorrhiza rosulata* can be distinguished from the other species in the genus by crenate leaf margin and flowers always arranged in the bract axils (Table 1). Because of the leaves clustered at the base of the stem and their size larger than the bracts, the bracteose florescence looks like a terminal raceme. The other *Sphaerorrhiza* species present leaves distributed along the stem that are larger than those that subtend the flowers. In these species, the flowers are usually uniformly distributed along the stem, or sometimes concentrated at the apex, but never in bract axils.

**Additional Specimens Examined**—BRAZIL. Goiás: Alto Paraíso, Reserva Raizama/São Jorge, barranco próximo a cachoeira, relevo ondulado, solo argiloso-úmido, 1060 m alt.,  $14^{\circ}42'52''\text{S } 47^{\circ}55'34''\text{W}$ , 27 Feb 1999, Calago 94 (CEN); Alto Paraíso, Vila de São Jorge, Reserva Raizama, trilha próximo à hidromassagem, ca. 10 m do final da passarela de madeira, nas rochas, na borda do rio, 780 m alt.,  $14^{\circ}12'09''\text{S } 47^{\circ}50'11.7''\text{W}$ , 23 Jan 2005, Araujo et al. 513 (ESA); idem, 849 m alt.  $14^{\circ}12'09.2''\text{S}, 47^{\circ}50'09.5''\text{W}$ , 02 Mar 2013, Araujo et al. 1121 (G, HUFABC, RB); idem, living plant for cultivation, bloomed at sítio Primavera, Biritiba Ussu, near Mogi das Cruzes under the number MP-1329 (from Araujo et al. 513 in bud), 10 Feb 2011, Peixoto 28 (HPL).

3. *Sphaerorrhiza sarmentiana* (Gardner ex Hook.) Roalson & Boggan, Selbyana 25(2): 225–238. 2005. *Gloxinia sarmentiana* Gardner ex Hook., Icon. Pl. 4: pl. 378. 1841.—**TYPE**: BRAZIL. Piauí: Oeiras, on shady rocks in the Buraco do Inferno, about a league from Oeiras in a ravine, Apr 1839, G. Gardner 2226 [lectotype designated here: K! (K000479972); isolectotypes: BM!, CGE!, G!, K! (K000479973), L!, P!, W!].

*Gloxinia attenuata* Hanst., *Linnaea* 27: 716. 1856.—TYPE: BRAZIL. Tocantins: Natividade, Near Villa de Natividade, in shady cliff of calcareous rocks, Dec 1839, G. Gardner 3303 [lectotype designated here: K! (K000479976); isolectotypes: BM!, CGE!, E!, F!, G!, K! (K000479975, K000479977, K000479974), NY!, P!, SP!, W!].

*Gloxinia stolonifera* Fritsch, *Bot. Jahrb. Syst.* 37: 493. 1906.—TYPE: BRAZIL. Minas Gerais: Ouro Preto, Tripuhy, 08 Jan 1883, A. F. M. Glaziou 15168 [holotype: B, destroyed; lectotype designated here: P! (P00587416); isolectotypes: B!, BR!, F!, G!, IAN!, K!, LE!, MO!, NY!, P! (P00587415), R!, RB!, US!].

Herbs (3.5–)5–50 cm tall, rarely producing filiform aerial stolons, reaching up to 20 cm long and bearing spaced and minute scale-like leaves. Stems usually unbranched, sappy when fresh. Internodes 0.7–12.0 cm long. Leaves distributed all over the plant, opposite, rarely whorled, generally equal in pairs, rarely subequal in pairs; petioles 0.3–4.0 cm long; blades 2.4–16.0 × 1.3–9.1 cm, narrowly ovate to ovate or elliptic to broadly elliptic, apex acuminate, sometimes acute (mainly in the basal leaves) or rarely retuse, base asymmetric or not, attenuate or narrowly cuneate, margin serrate or serrulate, sometimes irregularly serrate, rarely weakly crenate, above sparsely pubescent to pubescent, below totally glabrous, sometimes sparsely pubescent only on the veins, rarely sparsely pubescent all over the blade, secondary veins (3–)5–7(–8) pairs. Florescence frondose, with solitary flowers, and generally opposite in the stem, rarely alternate and with peduncle (0.1–0.8 cm long) supporting 2 flowers; subtending leaves 1.8–11.5 × 0.8–5.4 cm, flowering internodes 1.0–7.0 cm long. Flowers with pedicel (0.4–)0.7–2.0 cm long; calyx green, lobes 1.0–1.9 × 0.3–0.7 cm, outside sparsely pubescent to pubescent, rarely glabrous, inside glabrous, lanceolate, ovate or elliptic, margin entire, frequently ciliate; corolla axis slightly oblique in relation to pedicel; corolla narrowly campanulate, dorsiventrally compressed with a small ventral gibbosity in the middle of the tube, totally glabrous, tube outside purple or dark violet with pink to white base, inside with nectar guide with purple dots, 1.2–3.0 cm long; throat uniformly colored or with some marks slightly lighter than the rest of the corolla, not constricted, 1.0–2.3 cm diam; lobes concolor or with some marks slightly lighter than the rest of the corolla, 0.4–1.1 cm long, sometimes somewhat emarginate at tip; stamens 1.2–1.5 cm long, included, filament glabrous; nectary 5 separate glands or unlobed and annular disk, conspicuous; ovary inferior; stigma bilobed, sometimes stomatomorphic. Capsule 0.7–1.2 × 0.3–0.5 cm, pubescent, fusiform, apex pointed upwards.

**Distribution and Ecology**—The species is the most common of the genus and it is widely distributed in Brazil, from the Espinhaço Range and surroundings in Minas Gerais to the Cerrado of the central region of Piauí state and to the limits of caatinga in northwest of Ceará state; it also occurs in Cerrado and “campos rupestres” of the Central Brazilian Plateau reaching the Amazon biome in the southeast of Pará state (Fig. 6; Araujo et al. 2015). It thrives at elevations varying from 300–1,000 m, generally on rocky outcrops (sandy, sandy-clay or lime) inside deciduous forests, gallery forests or margin of rivers or in “campos rupestres” such as in the Espinhaço Range, but it occurs rarely in seasonal semi-deciduous forests or in Cerrado with gravel substrate.

**Phenology**—Flowering from December to June; fruiting in April.

**Conservation Status**—The species is least concern, based on criteria such as extent of occurrence (about 1.2 million km<sup>2</sup>), i.e. it is widespread (IUCN 2001).

**Notes**—*Sphaerorrhiza sarmentiana* can be differentiated from the other three species in the genus by the larger size of the plant and much larger flowers.

This species was described by Hooker and, according to this author, the epithet (*sarmentiana*) was given in homage to Casimiro Sarmento (resident of Oeiras who showed that species to Gardner). It was described under *Gloxinia* and had never been removed from the genus until the molecular evidence produced by Zimmer et al. (2002), Roalson et al. (2005a), and the present study.

There are some morphological variations in this widespread species, like the overall size of the plant or the length of the corolla. Flowers are single in the axils of the upper leaves, but in some duplicates of the collections Irwin 19138 (U) and Gardner 3303 (E, G, K), two flowers supported by a small peduncle were observed.

Fritsch (1906), in the protologue of *Gloxinia stolonifera*, said that he also examined material of *Gloxinia attenuata* and *Gloxinia sarmentiana* and concluded that the absence of aerial stolon, the presence of coarsely serrate leaves and the wide and shorter calyx lobes differentiate these taxa from the first species. Hoehne (1964) adopted the same position as Fritsch (1906) and recognized the three species as different taxa. When analyzing all the types involved, we found that the differences pointed out by Fritsch are superficial and that the characters overlap, except for the presence of aerial stolon. The few differences in the types of *Gloxinia attenuata*, *G. sarmentiana* and *G. stolonifera* are not consistent enough to keep them as distinct taxa. Characters like size of the plants (25–50 cm tall in the first one, 6–25 cm tall in the second, and 20–30 cm in the third), presence of stolon only in *G. stolonifera* (although absent in the type collection in the B and US herbaria), or flowers in the leaf axils in pairs sustained by a short peduncle only in a few duplicates of *G. attenuata*, but with solitary flowers in the others, do not allow a clear separation of the three species. A single taxon is therefore recognized under *Sphaerorrhiza sarmentiana* with the other names, *Gloxinia stolonifera* and *Gloxinia attenuata*, included in synonymy.

It may be worth noting here that the occasional presence of aerial and filiform stolons must be an additional way of vegetative propagation, but it is a rare phenomenon probably linked to particular growing conditions. It was never observed during our field excursions nor in the three accessions in cultivation.

Fritsch (1906) commented that the collections used for the description of new species of his work were in Berlin (B); the holotype of *Gloxinia stolonifera* should therefore be in this herbarium. Nevertheless, following the bombing suffered by the herbarium, this material was likely destroyed, as we did not locate it in B (Sleumer 1949; Pilger 1953; Hiepko 1987). No Gesneriaceae negatives were produced based on the list provided by the Chicago Field Museum ([http://emuweb.fieldmuseum.org/botany/search\\_berlin.php](http://emuweb.fieldmuseum.org/botany/search_berlin.php)). Another sheet of this taxon was encountered in B that bears a note “acc 2 OKT 64”. This number “64” likely indicates that the material was received at B in 1964. This date is later than the description of *Gloxinia stolonifera*: 1906. This material

is identified as *Gloxinia attenuata* without signature or date and as *Gloxinia sarmentiana* by Hans Wiehler in 1981. As there is no evidence that this material was identified by Fritsch, it cannot be the holotype of *Gloxinia stolonifera*, and therefore a lectotype is designated here based on the Glaziou 15168 specimen held in the Paris herbarium (P). Locality information varies among the different duplicates' labels of Glaziou 15168, but we assume that the correct one is "Ouro Preto, Tripuhy" in Minas Gerais state.

Additional Specimens Examined—BRAZIL. Bahia: São Desidério, Sítio Grande, paredão na borda do Rio Grande, em baixo da ponte, 520 m alt., 12°25'57.2"S, 45°05'04.0"W, 22 Feb 2013, Araujo et al. 1099 (HUFABC); Barreiras, Rio de Janeiro, Cachoeira do Acaba Vidas, mata ciliar, Apr 2000, Harley 53806 (HUEFS, G); Próximo Rio Preto (Formosa do Rio Preto), 11°12'S 45°57'W, 03 Apr 1978, Miranda 214 (RB); Próximo do Aeroporto de Barreiras, mata com afloramento rochoso, 675 m alt., 12°06'0"S 44°59'0"W, 14 May 1997, Proença et al. 1751 (UB); Santana, Serra a N de Santana na estrada para a torre de TV, 14 Feb 2000, Queiroz 6004A (HUEFS). Ceará: Ipu, Curva do Boqueirão, 6 Apr 2001, Lorenzi 2684 (HPL); Bica de Ipu, 29 Mar 2002, Lorenzi 3258 (HPL); subida da Serra de Ibiapaba, 07 Jan 2004, Lorenzi 4308 (HPL). Distrito Federal: Brasília, Reserva Ecológica do IBGE, 01 Mar 1992, Azevedo & Figueiras 971 (IBGE); Brasília, Fazenda Maranhão via DF-2-Serra, 27 Feb 1979, Heringer 1084 (R, IBGE, K, NY, US); Brasília, Cultivada na Estação Biológica da Universidade de Brasília, ripado das orquídeas, 21 Feb 1979, Heringer 1102 (R, IBGE, NY, MO); Brasília, UNB, cultivada no ripado de mudas, trazida de Luziânia, mata de galeria do Rio Corumbá – GO, 16 Apr 1975, Heringer 14561 (HB, NY); Planaltina, Cerrado aberto, cascalhoso, sujeito ao fogo anualmente, 05 Mar 1976, Heringer 15419 (IBGE); Brasília, Estação Experimental de Biologia, UNB, cultivada de semente colhida em matas de galeria no Rio Corumbá, 30 km de Luziânia, local fresco, encosta, 15 Mar 1976, Heringer 15492 (HB, R, IBGE, NY, UB); Brasília, Forest rocky slopes, Córrego Landim, ca. 25 km N of Brasília, 950 m alt., 16 Mar 1966, Irwin et al. 14002 (F, K, MO, NY, RB, UB); Brasília, Reserva Ecológica do IBGE, ripado no viveiro, 15 Feb 1985, Mendonça & Ribeiro 454 (IBGE); Reserva Ecológica do IBGE, ripado no Viveiro, Mar 1979, Mendonça & Ribeiro 1102 (IBGE); Brasília, Reserva Ecológica do IBGE, viveiro, local alterado, 17 Jan 1990, Silva & Figueiras 927 (IBGE). Goiás: Alto Paraíso de Goiás, Vila de São Jorge, Reserva Raizama, nas rochas, na borda da cachoeira, próximo ao canion, 849 m alt. 14°12'09.2"S, 47°50'09.5"W, 02 Mar 2013, Araujo et al. 1123 (HUFABC); Formosa, Parque Municipal Salto do Itiquira, afloramento na borda do rio, em dois locais: atrás do restaurante e do lado direito da trilha para a cachoeira. 721m alt. 15°22'02.3"S, 47°27'12.6"W, 21 Feb 2013, Araujo et al. 1095 (G, HUFABC); Paraúna, Estrada GO-411, ca. 90 km após a cidade de Paraúna, Ponte de Pedra, margens do Ribeirão Corrente, 25 Jan 1991, Barros 2192 (SP); Colinas do Sul, estrada Serra da Mesa-Colinas, ca. 17 km do portão do canteiro de obras, mata de galeria, substrato arenoso com folhedo, relevo montanhoso, 640 m alt., 14°12'S 48°04'W, 12 Mar 1992, Cavalcanti et al. 1199 (CEN, RB, G); Estrada Corumbá de Goiás a Niquelândia, ca. 16 km ao N do trevo com a estrada de Brasília, área de mineração abandonada, paredão úmido com capoeira, 30 Jan 1991, Chautems 407 (G, UFG); Descida da Serra Dourada, beira d'água, 27 Jan 1968, Eunice et al. s. n. (NY, R, UB); Formosa, JK, 09 Jan 1977, Hatschbach 39357 (MBM, WAG); Luziânia, Vale do Rio Corumbá, cultivada na Est. Exp. de Biologia, 05 Apr 1969, Heringer 11792 (UB); Luziânia, Rio Corumbá, Mar 1973, Heringer 14561 (NY); Corumbá de Goiás, 26 Mar 1978, Heringer 17013 (IBGE, NY); Pirenópolis, Serra dos Pirineus, 50 km N of Corumbá de Goiás on road to Niquelândia, Goiás in valley of Rio Maranhão, forested limestone outcrop, ca. 800 m alt., 24 Jan 1968, Irwin et al. 19138 (F, K, NY, UB, RB, W, WAG, U); Mossâmedes, Serra Dourada, da Reserva Biol. até o córrego Cafundó e Picarrão, 2° transecto, s.d., Rizzo et al. 18294 (UFG); Ipameri, margem esquerda do Rio Corumbá, aproximadamente 200 m da G.O. 28, mata de galeria antropizada, solo arenoso-argiloso com deposição de matéria orgânica, relevo acidentado, 17°43'S 48°09'W, 28 Apr 1994, Santos et al. 301 (CEN, G); Cocalzinho, Fazenda Funil, propriedade do Grupo Itaú, faz divisa com a fazenda do Sr. Osmar Carneiro, nas proximidades do Rio Preto, 755 m alt., 15°38'25" S 48°42'56"W, 23 Mar 2002, Silva et al. 5166 (IBGE, CEPEC); Niquelândia, Lago em processo de enchimento no AHE Serra da Mesa - segmento Rio Bagagem, mata de galeria de pequeno afluente do Rio Bagagem - Serra Negra; relevo acidentado, substrato: solo areno-argiloso entre cascalho, muitas e grandes rochas nuas, 13°59'57"S 48°19', 30 Jan 1997, Walter et al. 3706 (CEN, G). Mato Grosso: Chapada dos Guimarães, Parque Nacional da Chapada dos Guimarães, Trilha para o poço do Vêú da Noiva, 03 Mar 2011, Giacomini et al. 1300 (BHCb); Missão Salesiana Santa Terezinha,

no rio da Mortes, 21 Apr 1958, Lima 58–3073 (K); Porto Estrela, Serra das Araras, Exped. Regnellian, 14 Feb 1894, Lindman 3001 (GZU, S); s.d., Smith 142 (R). Minas Gerais: Francisco Sá, Mata de encosta, s.d., Angelo Neto 339 (ESAL); Lagoa Santa, APA de L.S., Oct. 1995-Feb 1996, Brina & Costa s. n. (BHCb 32715); Cristália, barra do Rib. Soberbo, 19 Mar 1992, Carvalho 942 (BHCb); Triângulo Mineiro, Serra São Vicente, 28 Mar 1959, Castellanos 22428 (R); Monjolos, Rodeador - distrito de Monjolos, Mata calcárea, 14 Jan 1990, Filgueiras 1976 (IBGE, R); Lagoa Preta, Margem do Rio Paraopeba, margem da lagoa, 31 Mar 1958, Heringer 6372 (UB); Serra do Espinhaço, ca. 5 km NE of Francisco Sá, road to Salinas, 950 m alt., 13 Feb 1969, Irwin et al. 23198 (NY, UB); São Gonçalo do Rio Preto, Parque Est. do Rio Preto, 08 Apr 2000, Lombardi et al. 3882 (BHCb, ESA, SPF, MBM); Campina Verde, Morro Limpo, Fazenda do Barro, Campina Verde, 25 Jan 1945, Macêdo 644 (SP, E, MO); Ituiutaba, "S.Vicente", 25 Feb 1951, Macêdo 3210 (NY, SP, US); Ituiutaba, S. Vicente, 10 Mar 1956, Macêdo 4493 (K); Arinos, Assentamento Chico Mendes, drenagem entre calcário e granito, 600 m alt., 15°51'40.8"S 46°15'54.2"W, 26 Feb 2006, Melo 1439 (ESAL); Arinos, Assentamento Chico Mendes, na base de afloramento de calcário, 600 m alt., 15°51'52.7"S 46°15'05.9"W, 27 Feb 2006, Melo 1474 (ESAL); Corinto, Fazenda do Diamante, Olhos de Água, 01 Apr 1931, Mexia 5503 (BM); Pandeiros, Refúgio da Natureza, afloramento calcário nas margens do Rio Pandeiros, 22 Feb 2007, Mota et al. 691 (BHCb); Estrada do Centro de Maquiné para Cordisburgo, 31 Mar 1957, Pereira & Pabst 2740 / 3376 (HB, R); Santo Hipólito, estrada Corinto-Cons.Mata, 6 km de Santo Hipólito, 550 m alt., 12 Jan 1998, Pirani et al. 3913 (SPF); Januária, Vale do Rio Peruaçu, Boqueirão da Onça, 15°07'8.5"S 44°15'17"W, 24 Apr 1997, Salino 3076 (BHCb, G); Varzelândia, Campo Redondo, 06 Feb 1985, Silva & Menezes 1128 (R); Santo Hipólito, estr. Sto. Hipólito-Cons. Mata, ca. 5 km de Sto. Hipólito, 21 Feb 2002, Souza et al. 28143 (ESA); Uberlândia, UHE Capim Branco, 04 Mar 1996, Temeirão Neto 1964 (BHCb); Lagoa Santa, s.d., Warming s. n. (P). Pará: Redenção, Rodovia BR-287, ca. 22 km oeste de Redenção em direção a Cumaru do Norte, Serra na Chácara São Bernardo, mata, 340 m alt., 08°03'58.5"S 50°11'23.5"W, 29 Jan 2005, Araujo et al. 539 (ESA); Conceição do Araguaia, range of low hills ca. 20 km W of Redenção, near Córrego São João and Troncamento Santa Tereza, 350–620 m alt., 8°03'S 50°10'W, 08 Feb 1980, Plowman et al. 8464 (K, MG, US). Piauí: Serra da Capivara, 1979, Empeaire 681 (IPA); An Felsen der Serra Branca, Jan 1907. Goyaz, 1837–1841, Gardner s. n. (P). Ule 7567 (K, L). Tocantins: Natividade, Near Villa de Natividade, in shady cliff of calcareous rocks, Dec 1839, Gardner 3303 (BM, CGE, E, G, K, NY, P, SP, W). Sem Procedência, s.d., Gardner s. n. (K); mato, Mar, Glaziou s. n. (P); s.d., Loeffgren s. n. (R 20923).

#### 4. *Sphaerorrhiza serrata* A. O. Araujo & Chautems, sp. nov.—

TYPE: BRAZIL. Distrito Federal: Brasília, Margem esquerda do Rio Belchior, Cerrado, borda de mata de galeria, relevo suave ondulado, solo areno-argiloso, área de influência indireta, 950 m alt., 15°53'56"S 48°13'04"W, 10 Mar 2003, J. P. Silva, G. A. Moreira & J. M. Rezende 7302 (holotype: CEN!).

*S. burchellii* affinis, sed differt inflorescentia frondo-bracteosa, margine foliorum serrata, corolla cum tubo albo et lobis lilacinis.

Herbs 4–8 cm tall. Internodes 0.2–1.0 cm long. Leaves distributed all over the plant, those subtending the flowers decrease in size toward the apex, opposite, equal to subequal in pairs; petioles 0.2–0.3 cm long; blades 0.7–5.0 × 0.5–1.9 cm, elliptical, sometimes ovate, apex acute to acuminate, base symmetrical, attenuate, margin serrate toward apex, entire at base, above sparsely pubescent, below glabrous or sparsely pubescent with trichomes only on the veins, secondary veins 3–

4 pairs. Florescence frondo-bracteose, 1–2 flowers per axil, opposite in the stem, sometimes a florescence sprout of the basal leaf axils; subtending leaves 0.3–2.7 × 0.2–0.7 cm or flowers without leaves at the base, flowering internodes 0.2–1.0 cm long. Flowers with pedicel 0.3–2.3 cm long; calyx green, lobes 0.4–0.5 × 0.1–0.3 cm, outside sparsely pubescent, inside glabrous, lanceolate or elliptical, with entire margin, frequently ciliate; corolla axis slightly oblique in relation to pedicel; corolla narrowly campanulate, with a small gibbosity in the middle of the tube, glabrous, except by the lobes margin, tube outside white, 0.5–0.7 cm long; throat white, not constricted or

barely constricted, 0.4–0.5 cm diam; lobes lilac, 0.2–0.3 cm long, entire or slightly emarginate at tip; stamens 0.3–0.4 cm long, included, filament glabrous; nectary an unlobed and annular disk, inconspicuous; ovary inferior; stigma stomatomorphic, sometimes bilobed. Capsule about  $0.5 \times 0.2\text{--}0.3$  cm, pubescent, fusiform, apex pointed upwards.

**Distribution and Ecology**—This species occurs in a restricted area of the Distrito Federal (Fig. 6), in sandy-clay outcrops within gallery forests near a river, in small but dense populations.

**Phenology**—Flowering and fruiting from February to March. This species was observed in vegetative stage in the natural environment in the District of Samambaia (Brasília - DF). During the dry season, from May to September, plants are dormant and reduced to small tubers hidden in rock crevices.

**Etymology**—From Latin serratus = serrate. The specific epithet makes reference to the leaf margin, which is serrate.

**Conservation Status**—The species is critically endangered, based on criteria such as extent of occurrence ( $< 100$  km)<sup>2</sup>, a single location of occurrence, and continuing decline in area, extent, and/or quality of habitat (IUCN 2001). Signs of degraded environment were observed at this unique location.

**Notes**—The corolla color is similar to *S. rosulata* (tube white with lilac lobes without spots) and the leaves are distributed all over the plant, like in *S. burchellii* (Table 1). Leaves with serrate margin are also found in *S. sarmentiana*, but the small size of the plant and the size and color of corolla distinguish this species from the latter.

**Additional Specimens Examined**—BRAZIL. Distrito Federal: Brasília, Distrito Samambaia, Sítio de propriedade do Sr. Nena localizado em uma estrada de terra saindo à direita da rodovia para Goiânia, rochas na margem esquerda do Rio Belchior, 909 m alt., 15°53'51.4"S 48°13'0.8"W, 21 Jan 2005, Araujo et al. 510 (ESA). In cultivation. São Paulo: Mogi das Cruzes. Introduzido em cultivo no sítio Primavera, Biritiba Ussu (a partir de material originalmente coletado em botão ou frutos passados, Araujo et al. 510) e quando floresceu a presente exsiccata foi preparada, 10 Feb 2011, Peixoto 26 (HPL).

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Appendix 1. Taxon, collector, collection number (herbarium acronym), and GenBank accession numbers (ITS, atpB-rbcL, matK, rpl16, rps16, and, trnL-trnF. N.A.: no data) for material used in the molecular phylogenetic analyses. Samples taken from cultivated material with indicated accession numbers were collected in CJBG (Conservatoire et Jardin botaniques de la Ville de Genève greenhouse), GRF (Gesneriad Research Foundation, Sarasota, USA), or MP (Mauro Peixoto private collection, Brazil).

*Achimenes admirabilis* Wiehler, Chautems & Perret 01-033 (G), GQ344504, AJ439982, JX195949, AJ487784, JX195806.1, AJ439827.1; *Alsobia punctata* (Lindl.) Hanst., Perret & Chautems 80 (G), KX011572, KX011546, JX195950, KX011564, JX195807, JX195723.1; *Amalophyllon divaricatum* (Poepp.) Boggan, L. E. Skog & Roalson, Perret & Chautems 89 (G), KU991233, KU991206, JX196056, KU991246, JX195882.1, JX195796; *Anethanthus gracilis* Hiern, Peixoto & Chautems 28 (G), KU991234, KU991207, JX195951, N.A., JX195808.1, JX195724.1; *Besleria melancholica* (Vell.) C. V. Morton, Forzza 2419 (G), GQ344505, AJ490923, JX195952, GQ383630, GQ383580, GQ383542; *Codonanthe gracilis* (Mart.) Hanst., Chautems & Perret 99-009 (G), KT958324, KT958440, JX195967, KT958381, JX195823.1, JX195739.1; *Chautemsia calcicola* A. O. Araujo & V. C. Souza, Araujo et al. 500 (ESA), GQ344506, KU991208, JX195975, GQ383631, GQ383581, GQ383543; *Chrysothemis pulchella* (Donn ex Sims) Decne., Araujo et al. 602 (G), GQ344507, KT958447, JX195977, GQ383632, GQ383582, GQ383544; *Columnea ulei* Mansf., Chautems & Perret 10-109 (G), KT958333, KT958449, JX195981, KT958390, JX195832.1, JX195750.1; *Coronanthera* indet., Callmänder 830 (G), KU991235, KU991209, JX195982, KU991247, JX195833, JX195751; *Corytoplectus capitatus* (Hook.) Wiehler, cult. CJBG 936189, KT958334, KT958450, JX195983, KT958391, JX195834.1, JX195752.1; *Codonanthopsis ulei* Mansf., Chautems & M. Perret 07-002 (G), KT958336, KT958452, JX195985, KT958392, JQ954145.1, JX195754.1; *Creosperma maculatum* L. E. Skog, Perret & Chautems 23 (G), KU991236, KU991210, JX195986, KU991248, JX195837.1, JX195755.1; *Depanthus* indet., Savolainen 19211 (K), GQ344509, KU991211, JX195987, GQ383633, GQ383584, GQ383545; *Diastema* indet., cult. MP 0872, GQ344510, KU991212, JX195988, GQ383634, GQ383585, GQ383546; *D. vexans* H. E. Moore, cult. GRF 840 306, GQ344511, KU991213, JX195989, GQ383635, GQ383586, GQ383547; *Drymonia serrulata* (Jacq.) Mart., Araujo et al. 601 (G), GQ344512, KT958453, JX195992, GQ383636, GQ383587, GQ383548; *Eucodonia andrieuxii* (DC.) Wiehler, Perret & Chautems 81 (G), KU991237, KU991214, JX195996, KU991249, JX195843.1, JX195760.1; *Gasteranthus wendlandianus* (Hanst.) Wiehler, Perret & Chautems 9 (G), KU991238, KU991215, JX195997, KU991250, JX195844.1, JX195761.1; *Gesneria humilis* L., Chautems & Perret 97-020 (G), GQ344513, AJ439976, JX195998, AJ487778, GQ383588, AJ439821; *Glossoloma tetragonum* Hanst., Perret & Chautems 11 (G), KX011576, KX011550, JX196002, KX011568, JX195845.1, JX195762.1; *Gloxinia lindeniana* (Regel) Roalson & Boggan,

Araujo 604 (G), GQ344515, KX011555, JX196000, GQ383638, GQ383590, GQ383550; *Gloxinia perennis* (L.) Fritsch, Araujo et al. 536 (ESA), GQ344516, KX011556, JX196001, GQ383639, GQ383591, GQ383551; *Gloxiniopsis racemosa* (Benth.) Roalson & Boggan, Araujo 548 (ESA), GQ344514, KX011554, JX195999, GQ383637, GQ383589, GQ383549; *Goyazia rupicola* Taub., Araujo et al. 545 (ESA), GQ344518, KU991216, JX196004, GQ383641, GQ383592, GQ383553; *Kohleria spicata* (Kunth) Oerst., Chautems & Perret 97-021 (G), GQ344520, KT958456, JX196006, AJ487777, GQ383594, AJ439820; *Mandirola rupestris* (Gardner) Roalson & Boggan, Araujo 530 (ESA), GQ344529, KU991217, JX196013, GQ383650, GQ383603, GQ383561; *Mitraria coccinea* Cav., Charpin 25967 (G), GQ344530, KU991218, JX196014, GQ383651, GQ383604, GQ383562; *Napeanthus reitzii* (L. B. Sm.) B. L. Burt ex Leeuwenb., Cervi et al. AC479 (G), GQ344531, KU991219, JX196041, GQ383652, GQ383605, GQ383563; *Nautilocalyx mellitifolius* (L.) Wiehler, Chautems & Perret 01-025 (G), GQ344532, KT958481, JX196043, AJ487786, GQ383606, AJ439829; *Negria rhabdanthoides* F. Muell., Savolainen 19043 (K), GQ344533, KU991220, JX196044, GQ383653, GQ383607, GQ383564; *Nematanthus fritschii* Hoehne, Chautems & Perret 07-404 (G), KT958350, KT958464, JX196022, KT958404, JX195855.1, JX195772.1; *Paliavana plumerioides* Chautems, Chautems 460 (G), GQ344537, AJ439964, JX196048, AJ487766, GQ383611, AJ439809; *P. tenuiflora* Mansf., Salviani 1541 (HPL), GQ344536, AJ439962, JX196051, AJ487764, GQ383610, AJ439807; *Pearcea schimpffii* Mansf., Chautems & Perret 10-020 (G), KU991239, KU991221, JX196054, KU991251, JX195881.1, JX195795.1; *Phinaea albolineata* (Hook.) Benth. ex Hemsl., Araujo et al. 538 (ESA), GQ344538, KU991222, JX196055.1, GQ383655, GQ383612, GQ383566; *Rhytidophyllum vernicosum* Urb. & Ekman, Araujo et al. 600 (G), GQ344539, KU991223, JX196057, GQ383656, GQ383613, GQ383567; *Rufodorsia intermedia* Wiehler, Perret & Chautems 4 (G), KU991240, KU991224, JX196058.1, KU991252, JX195883, JX195797.1; *Sanango racemosum* (Ruiz & Pav.) Barringer, Neill 9458 (US), KU991241, KU991225, JX196060, KU991245, JX195885, JX195799; *Seemannia purpurascens* Rusby, cult. GRF 9670, GQ344545, KU991226, GQ383617, GQ383660, GQ383617, GQ383572; *Sinningia aggregata* (Ker Gawl.) Wiehler, Chautems & Perret 97-001 (G), KU991242, AJ439913, JX196067, AJ487715, JX195886.1, AJ439757.1; *S. cardinalis* (Lehm.) H. E. Moore, Chautems & Perret 97-015 (G), KU991243, AJ490931, JX196076, AJ487733, JX195896.1, AJ492318.1; *S. speciosa* (Lodd.) Hiern, Chautems & Perret 98-003 (G), GQ344551, AJ439935, JX196113, AJ487737, GQ383622, AJ439779; *Smithiantha multiflora* (Martens & Galeotti) Fritsch, Perret & Chautems 95 (G), KU991244, KU991227, JX196122, KU991253, JX195942.1, JX195805.1; *Sphaerorrhiza burchellii* (S. M. Phillips) Roalson & Boggan, Araujo et al. 535 (ESA), GQ344553, KU991228, JX196123, GQ383662, GQ383624, GQ383575; *S. sarmentiana* (Gardner ex Hook.) Roalson & Boggan, Araujo et al. 539 (ESA), GQ344555, KU991230, JX196125, GQ383664, GQ383626, GQ383577; *S. serrata* A. O. Araujo & Chautems, Araujo et al. 510 (ESA), GQ344554, KU991229, JX196124, GQ383663, GQ383625, GQ383576; *S. rosulata* A. O. Araujo & Chautems, Araujo et al. 513 (ESA), GQ344556, KU991231, JX196126, GQ383665, GQ383627, GQ383578; *Titanotrichum oldhamii* (Hemsl.) Solereder, Chautems & Perret 01-021 (G), GQ344557, KU991232, JX196127, GQ383666, GQ383628, GQ383579; *Trichodrymonia macrophylla* (Wiehler) M. M. Mora & J. L. Clark, Perret & Chautems 25 (G), KX011578, KX011552, JX196052, KX011570, JX195879.1, JX195793.1; *Vanhouttea calcarata* Lem., Carvalho et al. 526 (CEPEC), GQ344558, AJ439972, JX196129, AJ487774, GQ383629, AJ439817.