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Saxifraga luoxiaoensis (Saxifragaceae), a new species from Hunan and Jiangxi, China

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Abstract

Saxifraga luoxiaoensis, a new species of the genus *Saxifraga* sect. *Irregulares* (Saxifragaceae) from Hunan and Jiangxi, China, is described and illustrated. This new species is most similar to *S. daqiaoensis*, which can be easily distinguished from the later by its leaf margin 7- or 9-lobed and winged capsule. The systematic position of this species within *Saxifraga* sect. *Irregulares* is assessed based on molecular phylogenetic analysis of the chloroplast regions sequences together with morphological comparisons.

Keywords: China, Hunan and Jiangxi, molecular phylogeny, new species, *Saxifraga*

Introduction

Saxifraga Linnaeus (1753: 398), the largest genus of Saxifragaceae, with more than 440 species widely distributed throughout the Northern Hemisphere (Tkach *et al.* 2015). The genus displays remarkable morphological variation. Based on Engler & Irmscher's (1913) system, Gornall (1987) presented a revised classification with 15 sections, 19 subsections and 34 series. Recent molecular phylogenetic research covered at least 13 sections and 9 subsections within the genus (Tkach *et al.* 2015).

In China, *Saxifraga* comprises about 222 species, including the recently reported new species, *S. banmaensis* Pan (2006: 443) and *S. dingqingensis* Pan (2006: 445), *S. xiaozhongdianensis* Pan (2007: 512) and *S. ludingensis* Pan (2007: 513), *S. daqiaoensis* F.G. Wang & F.W. Xing (2008: 237), and *S. kegangii* D.G. Zhang, Y. Meng & M.H. Zhang (Zhang *et al.* 2017: 159). Sect. *Irregulares* Haworth (1803: 155) is the ancestral clade of *Saxifraga,* which is usually characterized by zygomorphic flowers (Tkach *et al.* 2015). This section includes 2 series, i.e., ser. *Rufescentes* Pan (1991: 1) and ser. *Stoloniferae* Pan (1991: 1) and 9 species occurring in China.

In 2016, during investigations in the Luoxiao mountain range, China, an undescribed specimen was collected from the Mount Nanfengmian between Hunan and Jiangxi province. This species is morphologically similar to *S. epiphylla* Gornall & H. Ohba (Gornall *et al.* 2000: 375), *S. mengtzeana* Engl. & Irmsch. (1913: 36) and *S. daqiaoensis*. But the new species was characterized by leaf blade reniform without foliar embryos, winged capsule, leaf margin 7- or 9-lobed. Subsequent morphological comparisons and phylogenetic analyses support the status of the taxon as a new species which is described herein.

Materials and Methods

Morphological observation:—Morphological data for description of this new species were collected from observation of the specimens (10 specimens from 3 populations) and compared to *S. daqiaoensis* (6 specimens from 1 population), *S. epiphylla* (3 specimens from 1 population) and *S. mengtzeana* (3 specimens from 1 population), both in the wild

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and herbarium. The voucher specimens are deposited in the Herbarium of Sun Yat-sen University (SYS!), Guangzhou, China.

Molecular analyses: We sampled 6 populations representing 5 species of Saxifraga sect. *Irregulares* (six individuals each population), including the presumed new species. Genomic DNA was extracted from silica-gel-dried leaves using the modified CTAB method (Doyle *et al.* 1987). Sequences for other taxa were obtained from GenBank (see Table 1). Voucher information and GenBank accession numbers were provided in the Table 1.

Phylogenetic reconstruction was performed using Bayesian inference (BI) and maximum likelihood (ML). The chloroplast rbcL region was amplified using the primers provided by Prieto *et al.* (2013). The phylogenetic tree based on Bayesian inference was generated using MrBayes version 3.2.6 (Huelsenbeck *et al.* 2001). The phylogenetic analysis based on maximum likelihood was conducted with PhyML version 3.0 (Guindon *et al.* 2010). Detected by the jModeltest 2.1.7, the GTRGAMMA+I model was used in BI and ML analyses (Posada 2008).

TABLE 1. Voucher information and GenBank accessions for phylogenetic analysis.

Species	Collected locations	Accession number
S. stolonifera (six individuals)	Taoyuandong Natural Nature Reserve, Yanling county, Hunan, China; 26°20′N, 113°59′E, 1450 m; Voucher No. LXP-13-24775	MG589455
S. mengtzeana (two individuals)	Mount. Yuanbao, Rongshui, Guangxi, China; 25°24′N, 109°08′E; 1200 m; Voucher No. FHZ-1608	MG589457
***S. luoxiaoensis*1** (six individuals)	Ceyuan Town, Yanling County, Hunan, China; 26°36′N, 114°31′E;1673 m; Voucher No. LXP-13-24953	MG589462
***S. luoxiaoensis*2** (six individuals)	Daijiapu Town, Suichuan County, Jiangxi, China; 26°18′N, 114°03′E; 1574 m; Voucher No. LXP-13-24717	MG589460
S. epiphylla (two individuals)	Mount Qingchen, Dujiangyan City, Sichuan, China; 30°54′N, 103°33′E; 1089 m; Voucher No. Q. Fan 15680	MG589456
S. daqiaoensis (six individuals)	Daqiao Town, Ruyuan County, Guangdong, China; 24°59′N, 113°08′E; 394 m; Voucher No. RY-2017-031	MG589458
	Sequences for other species downloaded from NCBI	
S. mertensiana		U06216.1
S. hirsuta		KC749993.1
S. hirsuta		KF997488.1
S. cuneifolia		KC749992.1
S. spathularis		KC749994.1
S. clusii		KC749989.1

Results

The morphological characters of our presumed new species, *S. daqiaoensis*, *S. mengtzeana* and *S. epiphylla* are presented in Table 2. These four species shared features such as stolons absent, and leaves abaxially with spots which distinguish them from other species in sect. *Irregulares*. The new species differs by its winged capsule, leaf margin 7- or 9-lobed, and lobe margin 5−7 -dentate, leaf blade reniform without foliar embryos, adaxially hispid and abaxially with red or brown spotted. *S. epiphylla* differs from the new species chiefly in that it produces a foliar embryo in the sinus of the basal leaf blades. The leaf blades of *S. mengtzeana* has no foliar embryo, but it has blades glabrous adaxially. *S. daqiaoensis* differs from the new species in its peltate leaves and leaf margin remotely shallowly dentate or subentire. Crucially, the mature carpels of the new species is winged, which makes it distinctly different from the above three species.

Three chloroplast rbcL region (584bp, 331bp, 450bp) of *Saxifraga luoxiaoensis* was concatenated by Sequence Matrix (Vaidya *et al.* 2011), yielding a total alignment length of 1365 bp for phylogenetic analyses.

The 50% majority-rule consensus tree based on Bayesian posterior probability (PP) and maximum likelihood bootstraps (LP) of the rbcL sequences both showed that most species of the *Saxifraga* sect. *Irregulares* were divided

into two well-supported subclades, corresponding to ser. *Stoloniferae* and ser. *Rufescentes* (PP=1.0, LP=91). Two accessions of the new species were grouped together (PP=1.0, LP=98), sister to *S. daqiaoensis* with strong supports (PP=1.0, LP=91).

Discussion

The new species *Saxifraga luoxiaoensis* has zygomorphic flowers and stolons absent, which indicate a position in Sect. *Irregulares* ser. *Rufescentes*. *Saxifraga luoxiaoensis* is similar to *S. daqiaoensis*, *S. epiphylla* and *S. mengtzeana* for the leaf base cordate and the leaf page abaxially glabrous spotted with red or brown, but it differs from the other three species for the leaf blade reniform without foliar embryos, the leaf page abaxially hispid, the mature capsule winged, and the leaf margin 7- or 9-lobed and lobe margin 5−7 -dentated.

According to our phylogenetic study, *S. daqiaoensis*, *S. epiphylla, S. mengtzeana* and the new species formed a clade (PP=1.00, LP=74). Two accessions of the new species were grouped together (PP=1.00, LP=98), sister to S. *daqiaoensis* with strong supports (PP=1.00, LP=91). The result of the phylogenetic reconstruction shows that *S. daqiaoensis* is the closest species relative to *Saxifraga luoxiaoensis*. The close relationship of the two taxa is also supported by similar morphological features. However, as discussed above, *S. daqiaoensis* is characterized by leaf base peltate and leaf margin remotely shallowly dentate or subentire. Furthermore, the above three species occur in Jiangxi Province and Hunan Province at 400−900 m a.s.l., whereas the new species occurs at higher elevation (1400−1800 m a.s.l.), supporting the distinction of our finding as a new species. Here we considered that the elevation gradient plays an important role in the differentiation of the species in Sect. *Irregulares*. The vertical distribution and the long-time geographical isolation may be a driver to speciation (Sobel 2014).

TABLE 2. Diagnostic characters of *Saxifraga luoxiaoensis* and comparison with other related species of sect. *Irregulares*

Characters	*S. luoxiaoensis*	*S. daqiaoensis*	*S. mengtzeana*	*S. epiphylla*
Foliar embryo	absent	absent	absent	present
leaf shape	reniform	nephroid to orbicular	ovate to broadly so	ovate
leaf base	cordate	peltate, cordate	cordate	cordate
leaf margin	margin 7- or 9-lobed	Remotely shallowly dentate or subentire	margin lobed	margin undulate, thickly dentate
trichomes on leaf	adaxial surface sparsely hispid ca. 2.5 mm long, abaxially glabrous with red or brown spotted	adaxial surface sparsely hispid ca. 4 mm long, abaxial surface glabrous, with brown or yellow-green spotted	adaxial surface glabrous, abaxial sparsely hispid and brown spotted	both surfaces glandular hispid and spotted
leaf size	1.5−8.5 × 1.2−7.3 cm	3−6.4 × 3.8−8 cm	1.5−7.6 × 1.2−6cm	1.2−10 × 1−8.4 cm
petals	shortest 3 petals ovate, yellow spotted, 3-veined; longer petal lanceolate-oblong, ca. 0.8–2.0 cm × 1.3–3 mm, 3−5-veined; longest petal linear-lanceolate, ca. 1.6−2.5 cm × 1.3−3 mm, 3−5-veined	shortest 3 petals triangular-ovate, ca. 3.5−4×2 mm, 3-veined, base with a claw; longer 2 petals linear, ca. 18–22 × 3 mm, 3-veined, apex obtuse, base gradually contracted into a claw.	shortest 3 petals triangular-ovate, 3-veined; longer petal narrowly ovate, ca. 0.9 cm × 2.2 mm, 3-veined; longest petal sublanceolate, ca. 1.9 cm × 3.4 mm, 8-veined	shortest 3 petals ovate, (1−)3−5-veined, base with a claw; longer petal linear-lanceolate, 0.3−1.8 cm × 1.2−2 mm, 3−5(−12)-veined; longest petal ca. 1.3−2.9 cm × 2.2−7.5 mm, 5−9(−15)-veined
flowering time	April to June	March to May	May	May to July
inflorescence	ca. 12−50 cm. 10–65-flowered	ca. 8−17.5 cm, 17−27-flowered	ca. 8.7 cm, 16−18-flowered	ca. 13−22 cm, 12−30-flowered
capsule shape	Mature carpels winged	Carpels without wings	Carpels without wings	Carpels without wings

Taxonomic Treatment

Saxifraga luoxiaoensis W. B. Liao, L. Wang & X. J. Zhang, *sp. nov.* (Fig. 2)

Type:—CHINA. Jiangxi Province, Suichuan County, Daijiapu Town, in wet limestone under of gully, 26°16′N, 114°02′E, Elev. 1466 m, 18 May 2016, W. Y. Zhao, Q. L. Ding, X. J. Zhang *et al.*, *LXP-13-16785* (SYS!).

Diagnosis:—*Saxifraga luoxiaoensis* is similar to *S. daqiaoensis*, *S. epiphylla* and *S. mengtzeana*. *S. epiphylla* differs from the new species chiefly in that it produces a foliar embryo in the sinus of the basal leaf blades. The leaf blades of *S. mengtzeana* has no foliar embryo, but it has blades glabrous adaxially. *S. daqiaoensis* differs from the new species in its peltate leaves and leaf margin remotely shallowly dentate or subentire. The most distinctive characters of *S. luoxiaoensis* is the winged capsule.

Description:—Perennial herbs, 12−50 cm tall. Stolons absent. Rhizomes rather short. Leaves all basal; petiole 5–18 cm, sparsely short glandular piliferous or glabrous; leaf blade reniform, papery, 1.5−8.5 × 1.2−7.3 cm, base cordate, margin 7- or 9-lobed, apex acute, lobes irregularly 5−7-dentate at margin, adaxially sparsely hispid, abaxially glabrous with red or brown spots. Inflorescence paniculate, ca. 12−50 cm. 10−65-flowered, branches 2−20 cm, sparsely glandular pubescent, 2−10-flowered, flowers zygomorphic; pedicels slender, 0.6−2.1 cm, glandular pubescent. Sepals spreading to reflexed, triangular lanceolate, 1.5−3 × 0.5−2 mm, adaxially glabrous, abaxially and marginally with sparse glandular hairs, 3-veined; veins confluent at apex. Petals 5, white or pink, apex acute; the two largest lanceolate-oblong, 3−5-veined, penninerved, the two longest ca. 0.8−2.0 (2.5) cm × 1.3−3 mm, the three smallest three ovate, yellow spotted, 2.3−3.5 mm × 1.5−2 mm, 3-veined. Stamens 4.3−5.6 mm. Ovary ovoid, with a semiannular nectary disc; styles divergent ca. 0.8−3 mm. Capsule winged when mature, carpels 5−7 mm. Seeds elliptic, the two sides slightly bent, ca. 0.8 mm, surface ribbed. Fl. and fr. Apr–July.

Distribution and ecology:—The new species *Saxifraga luoxiaoensis* occurs in the centre of Luoxiao mountain range between Hunan and Jiangxi province, China, and grows on moist rocks nearby valleys, alt. 1200–1900 m.

Etymology:—The specific epithet is derived from Luoxiao mountain range.

Paratypes:—CHINA. Jiangxi Province, Suichuan County, Daijiapu Town, 26°18′N, 114°03′E, Elev. 1574 m, 24 Oct. 2017, W. B. Liao, W. Y. Zhao, Z. C. Liu, F. Ye, X. J. Zhang *et al*., LXP-13-24717 (SYS!). CHINA. Hunan Province, Yanling County, Ceyuan Town, 26°36′N, 114°31′E, Elev. 1673 m, 28 Oct. 2017, W. Y. Zhao, Z. C. Liu, F. Ye, X. J. Zhang *et al.*, LXP-13-24953 (SYS!); same locality, 28 Oct. 2017, W. Y. Zhao, Z. C. Liu, F. Ye, X. J. Zhang *et al*., LXP-13-24990 (SYS!).

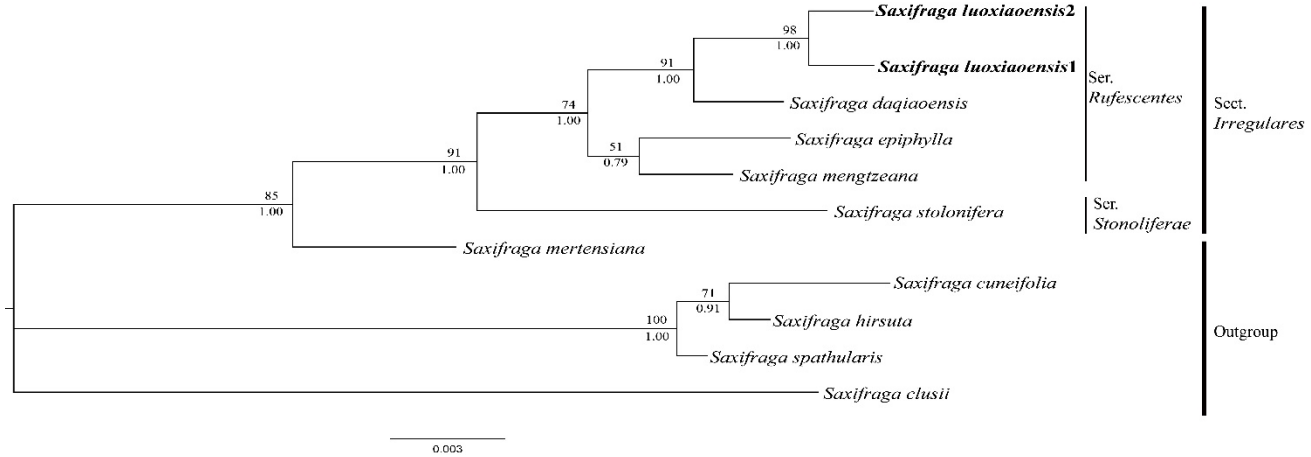

FIGURE 1. Bayesian consensus tree of the new species and related species. Numbers above branches are ML bootstraps, numbers below branches indicate Bayesian posterior probability; the new species is shown in bold.

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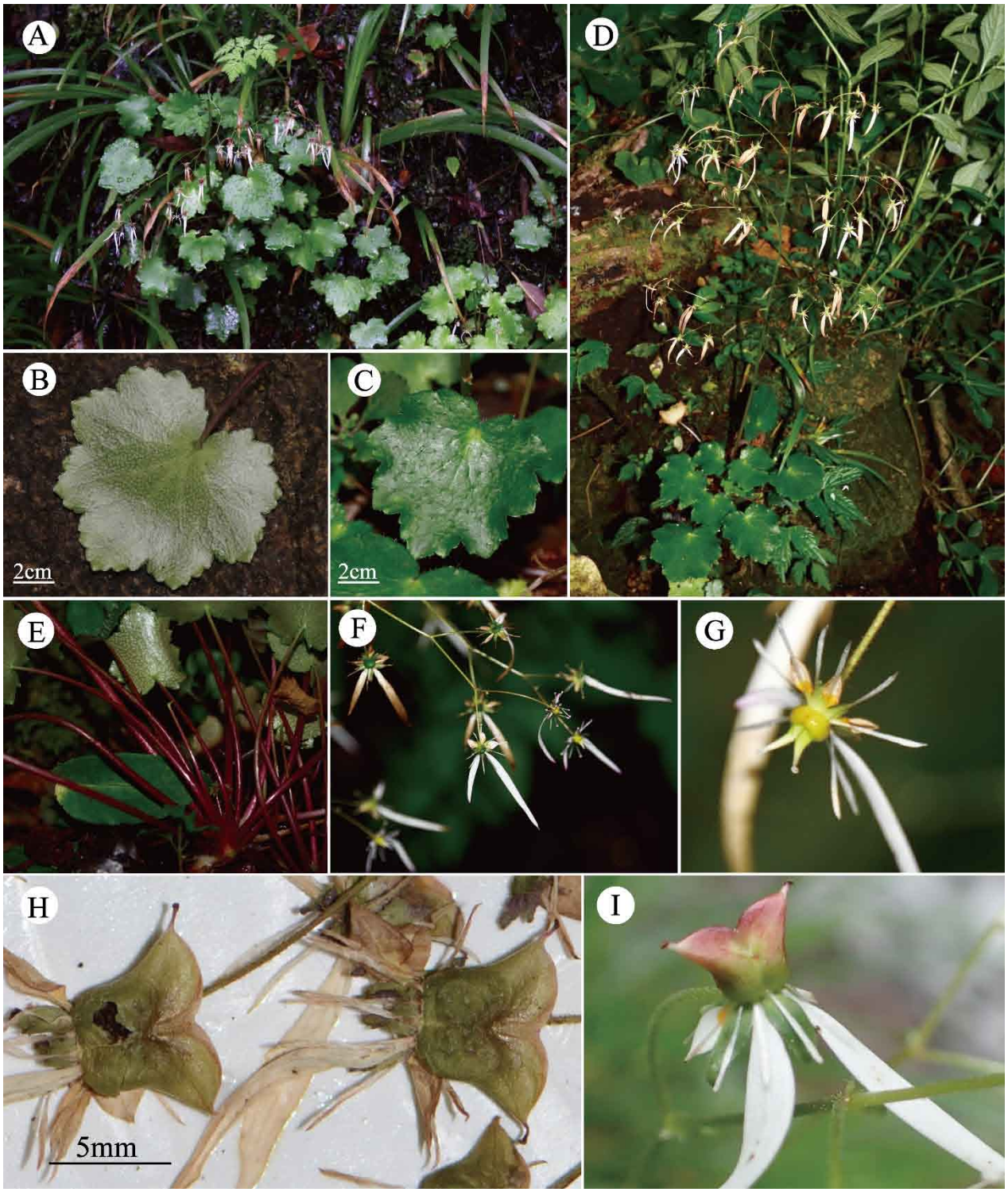

FIGURE 2. *Saxifraga luoxiaoensis* W. B. Liao, L. Wang & X. J. Zhang. **A.** Habitat; **B.** adaxial surface of leaves; **C.** abaxial surface of leaves; **D.** plants and inflorescence; **E.** rhizomes and petiole; **F.** flowers; **G.** semiannular disc; **H.** fruits on dry specimen; **I.** young fruit.

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