



Sedum lipingense (Crassulaceae) identifying a new stonecrop species in SE Guizhou, China, based on morphological and molecular evidence

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Abstract

We describe and illustrate *Sedum lipingense* (Crassulaceae), a new species of stonecrop found in the limestone areas of SE Guizhou, China. Based on the presence of adaxially gibbous carpels and follicles, this taxon belongs to sect. *Sedum* S.H. Fu. The new species superficially resembles *S. subtile* Miquel and *S. bulbiferum* Makino but differs from these two taxa in its development of a basal leaf rosette during florescence. The nrDNA internal transcribed spacer (ITS) sequences also support the claim that this plant is a new species in the *Sedum* genus.

Keywords

flora of Guizhou, karst, limestone flora, new taxon, Sedum lipingense

Introduction

Sedum Linnaeus is the largest genus in the Crassulaceae family, containing about 430 species, with the greatest diversity centering in eastern Asia (Thiede and Eggli 2007, Ito et al. 2017a). Approximately 121 Sedum species (91 endemics) occur in China, and 49 of these species (34 endemics) belong to sect. Sedum, a subclass which possess adaxially gibbous carpels and follicles (Wu et al. 2013). There are 23 species within five genera of Crassulaceae found in the Guizhou Province (Li et al. 1985). From 2005, a number of new species of Sedum were reported across mainland China, in areas includ-

ing Zhejiang (Wang et al. 2005, Jin et al. 2010, 2013), Anhui (Xie et al. 2014, Chen et al. 2017) and the Guizhou Province (Yang et al. 2012). In China, only a few species in this genus retain rosette leaves during florescence, such as *S. balfourii* Hamet and *S. drymarioides* var. *saxifragiforme* X. F. Jin & H. W. Zhang. *Sedum balfourii* was formerly placed in sect. *Aizoon*, within the genus *Sedum* (Fu and Fu 1984), but was then moved to the genus *Ohbaea* (Raymond-Hamet) V. V. Byalt & I. V. Sokolova (Fu and Ohba 2001) based on its conspicuous lateral flowering stems that derive from rosettes during florescence.

During our fieldwork, a new species of *Sedum* was discovered in Liping County, Qiandongnan Prefecture, Guizhou Province, China. This particular species has conspicuous rosettes during florescence, an attribute similar to *O. balfourii*. However, the new species differs from *O. balfourii* as it possesses central flowering stems rather than lateral ones (Fig. 2D). It also differs from *S. drymarioides* var. *saxifragiforme*, a species which is glandular-pubescent throughout, despite its rosette leaves. Based on its adaxially gibbous carpels, we place the new species in Sect. *Sedum*. Macro-morphological character studies indicated that this species is also somewhat similar to *S. subtile* Miquel and *S. bulbiferum* Makino, sharing a number of traits with these species, including opposite leaves on proximal stems and alternate leaves mainly on distal stems. We conducted morphological comparisons and molecular phylogenetic analysis to elucidate the presumed new *Sedum* species.

Materials and methods

All morphological characters were measured using dissecting microscopes. Specimen checking was done at PE, IBK, ZY, with the additional use of some web database, including the Plant Photo Bank of China (http://ppbc.iplant.cn/) and Global Plants (http://plants.jstor.org/).

Leaf material from the presumed new species was collected in the field, and immediately dried in silica gel for DNA extraction. The nuclear ribosomal internal transcribed spacer (ITS) regions were used as molecular markers. ITS-F (TGAACCT-GCGGAAGGATCAT) and ITS-R (GGTAGTCCCGCCTGACCTG) primers (Wu et al. 2013) were selected to amplify the ITS sequences. DNA extraction and PCR amplification of the new species followed the procedure of Wu et al. (2013). Primer synthesis and PCR product sequencing were carried out at the Shanghai Sangon Biotech Institute, China.

The ITS sequence of the new species, as well as the ITS sequences of the congeners downloaded from GeneBank (Table 1), were aligned using MEGA7 and then manually adjusted. Bayesian inference was implemented using MrBayes v3.2.6. Prior to the Bayesian analysis, the Akaike information criterion (AIC) implemented in mrModel-Test v1.0 was used to select the best-fit model (GTR+I+G) of molecular evolution. For the BI analyses, four Markov Chain Monte Carlo (MCMC) chains were run, sampling one tree every 100 generations for 2,000,000 generations starting with a random tree (Xie et al. 2014). When the log-likelihood scores were found to have stabilized, a

Table 1. Accession information relating to internal transcribed spacer (ITS) sequences downloaded from GeneBank.

Species	Voucher	Accession no.
Aeonium lancerottense	MEM 1518	AY082143
Aeonium viscatum	MEM 1432	AY082154
Greenovia aizoon	MEM 1425	AY082112
Sedum alfredii	WUK415208	FJ919953
Sedum baileyi	LBG0064555	FJ919935
Sedum bergeri	Ni et al.	AY352897
Sedum bulbiferum_416	Ito416	LC229234
Sedum bulbiferum_hs41	130514hs41	KM111166
Sedum bulbiferum_qz09	130524qz09	KM111165
Sedum emarginatum	130512hs27	KM111145
Sedum erici-magnusii	Ito 2077	LC229235
Sedum erythrospermum	Tsutsumi 504	AB906473
Sedum formosanum	Ito 1260	LC229279
Sedum hakonense	S. Mayuzumi C00005	AB088625
Sedum hangzhouense	Ito2604 (TNS)	LC260130
Sedum japonicum	Kokubugata 16749	AB906475
Sedum jiulungshanense	CMQ20150076	LC229243
Sedum kiangnanense	Ito 1030	LC229244
Sedum lineare	Mayuzumi C00120	AB088623
Sedum lipingense	ZRB1479	MN150061
Sedum lungtsuanense	Ito3563	LC260131
Sedum makinoi	Kokubugata 16730	AB906476
Sedum mexicanum	Ito 647	LC229247
Sedum morrisonense	Ito2765	LC229290
Sedum multicaule	Miyamoto et al. TI9596136	AB088631
Sedum nagasakianum	Ito2064	LC229249
Sedum nokoense	Kokubugata 10426	AB906478
Sedum oligospermum	CMQ 74	LC229250
Sedum oreades	G. Y. Rao 090803-03	KF113733
Sedum polytrichoides	CMQ1057	LC229251
Sedum rupifragum	Ito 2070	LC229254
Sedum sarmentosum	Ito 978	LC229255
Sedum satumense	Ito2295	LC229256
Sedum trullipetalum	9420132	AB088630
Sedum subtile_1999	A. Shimizu 1999	AB088622
Sedum subtile_2259	Ito2259	LC229257
Sedum subtile 624	Ito 624	AB930277
Sedum taiwanianum	Ito2770	LC229297
Sedum tetractinum	Ito3623	LC260135
Sedum tianmushanense	LP 67	LC229261
Sedum tosaense	Kokubugata 16726	AB906483
Sedum triactina	9596091	AB088629
Sedum tricarpum	Ito 2269	LC229259
Sedum trullipetalum	Miyamoto et al.9420132	AB088630
Sedum truncastigmum	Ito3254	LC229306
Sedum yabeanum	S. Mayuzumi C00029	AB088626
Sedum zentaro-tashiroi	H. Ohba 1998	AB088619

consensus tree was calculated after omitting 5,000 sampled trees as burn-in. *Aeonium lancerottense*, *A. viscatum* and *Greenovia aizoon* were selected as the outgroups referring to Ito et al. (2017b).

Results

Molecular analyses

In this study, the sequences of 40 species (44 samples) were treated as ingroups. Sequence length was 584 bp for the ITS region, of which 234 characters were constant, 45 characters were parsimony-uninformative and 305 characters were parsimony-informative.

The sequence of the ITS region taken from *S. lipingense* aligned with the genus *Sedum*, confirming its generic identity (Fig. 1). The new species was resolved as sister to *S. bulbiferum* (Bayesian posterior probabilities (PP) was 97) but turned out to be

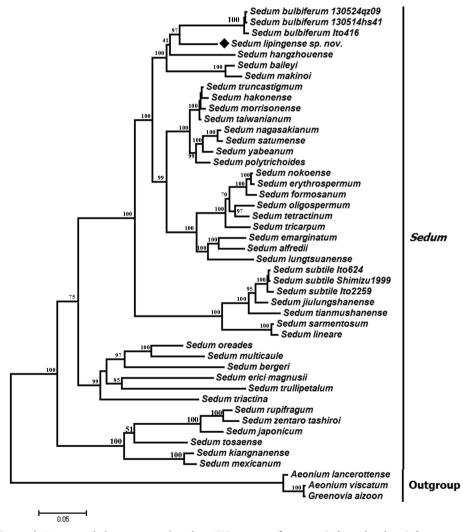


Figure 1. Bayesian phylogenetic tree based on ITS sequence for genus *Sedum* related to *S. lipingense* and three outgroups. Bayesian posterior probabilities are shown.

genetically distant from *S. subtile*. There were 50 nucleotides differ between *S. lipingense* and *S. bulbiferum*, suggesting the high variation compared to the closest relatives was remarkable.

S. lipingense and S. bulbiferum were found to be nested with S. hangzhouense (PP = 41, suggesting a weak support), and then to be nested with S. baileyi and S. makinoi (PP = 100), all species with alternate or opposite stem leaves. Except for S. lipingense, the above four (or perhaps two-three) species were also clustered as a distinct clade (Wu et al. 2013, Xie et al. 2014, Ito et al. 2017a), suggesting that the four species are closely related. Sedum lipingense is a close member to this clade, but these species form a polytomy and it is hard to say for sure, which one is the closest relative of S. lipingense. Sedum subtile is not within the same clade as S. bulbiferum, S. hangzhouense, S. baileyi, and S. makinoi (Wu et al. 2013) or with S. hangzhouense, S. baileyi, and S. makinoi (Ito et al. 2017a), suggesting that the relationship between S. subtile and S. lipingense is relatively distant.

Taxonomy

Sedum lipingense R.B. Zhang, D. Tan & R.X. Wei, sp. nov. urn:lsid:ipni.org:names:77202732-1
Figs 2–4

Diagnosis. *S. lipingense* can be distinguished from the closely related *S. subtile* and *S. bulbiferum* by the presence of rosettes, absent sterile shoots and bulbils, subequal lanceolate-oblong sepals, and other traits (Table 2).

Type. CHINA. Guizhou Province, Kaili City, Liping County, Mengyan Township, on moist rocks, 26°07′N, 108°42′E, 800 m alt., 13 April 2019, *ZRB1479* (fl., holotype ZY!, isotype IBK!), 16 June 2019, *ZRB1495* (fr., paratype ZY!)

Description. Biennial (or perennial?) herb. *Sterile stems* absent. *Rosette* present during florescence; rosette leaves alternate, broadly obovate, base attenuated and shortly spurred, $0.5-1.5 \times 0.4-0.7$ cm. *Flowering stems* 1 to 3 (-4), erect, slender, 3-7 cm; single stems shoot from rosette centers, others shoot from the rosette leaf axils; lateral proximal leaves sometimes opposite, akin to rosette leaves but smaller, $0.6-0.8 \times 0.3-0.5$ cm, base shortly spurred; distal leaves alternate, spatulate-obovate to spatulate-oblanceolate, $0.7-1.2 \times 0.3-0.4$ cm, apex obtuse, base shortly spurred. *Cymes* scorpioid, 2 to 3 branched; branches 1 to 2 flowered; bracts obliquely oblanceolate, apex obtuse, $4-9 \times 2-4$ mm. *Sepals* 5, lanceolate-oblong, subequal, ca. 2 mm, base shortly spurred, apex obtuse. *Petals* 5, yellow, broadly lanceolate, ca. 4 mm, apex mucronate. *Stamens* 10; antesepalous one ca. 3 mm; antepetalous one inserted ca. 1 mm above petal base, slightly shorter than the antesepalous stamens. *Nectar scales* broadly cuneate, ca. 0.6×0.4 mm, apex truncate. *Carpels* erect, lanceolate, ca. 3.5 mm, base connate for ca. 1 mm. *Styles* slender, ca. 1 mm. *Follicles* stellately divergent at maturity. *Seeds* oblong, ca. 0.6×0.4 mm, papillate.

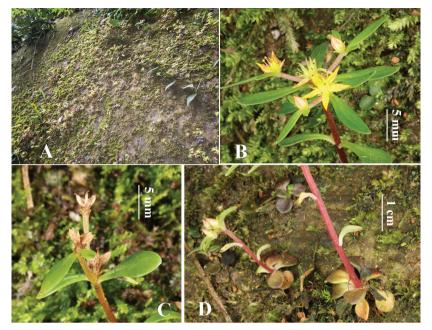


Figure 2. *Sedum lipingense* **A** natural habitat **B** 3-branched scorpioid cyme **C** follicles and bracts **D** single flowering stems derived from rosette centers. Charted by Ren-Bo Zhang.

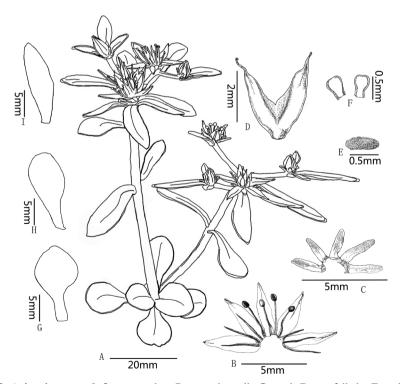


Figure 3. *Sedum lipingense* **A** flowering plant **B** opened corolla **C** sepals **D** two follicles **E** seed **F** nectar scales **G** rosette leaf **H** distal leaf **I** bract of flower. Drawn by Tan Deng.

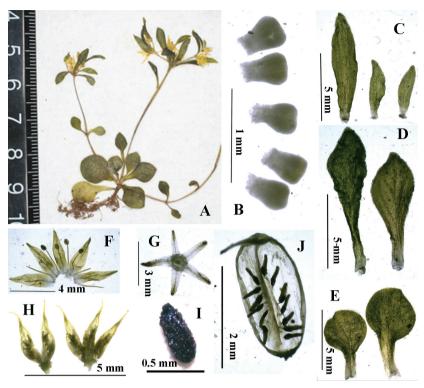


Figure 4. Sedum lipingense **A** rosette, central and lateral flowering stems **B** nectar scales **C** bracts of flowers **D** distal leaves **E** rosette leaves **F** opened corolla **G** sepals **H** split carpels **I** seed. Charted by Ren-Bo Zhang.

Table 2. Comparing the diagnostics of *Sedum lipingense* sp. nov., *S. subtile* and *S. bulbiferum*.

T	raits	S. lipingense	S. subtile	S. bulbiferum
Rosette leaves during florescence		present	absent	absent
Sterile shoots		absent	present	absent
Flowering stem		3–7 cm	5–10 cm	7–22 cm
Proximal stem	Phyllotaxy	alternate, sometimes opposite	opposite or	opposite
leaves		on lateral flowering stem	3–6-verticillate	
	Leaf blade	broadly obovate	obovate	ovate-spatulate
Distal stem	Phyllotaxy	alternate (sometimes	alternate	alternate
leaves		subopposite)		
	Leaf blade	spatulate-oblanceolate	oblanceolate-linear	spatulate-oblanceolate
	Bulbils in axils	absent	absent	present
Cymes	Branches	(2-) 3	2- or 3-branched	3-branched, branches
				2-forked
	Branch flowers	1- to two	3- to several	many
Sepals		lanceolate-oblong, subequal	broadly linear to narrowly	lanceolate to
			lanceolate, unequal	oblanceolate, unequal
Nectar scales		broadly cuneate, ca. 0.6×0.4	broadly cuneate, ca. 0.4 ×	obovate, ca. 0.6 mm
		mm, apex truncate	0.5 mm, apex truncate	
Carpels		ca. 3.5 mm base connate for	ca. 5 mm base connate for	ca. 4 mm base connate
		ca. 1 mm	ca. 2 mm	for ca. 1 mm
Styles		ca. 1 mm	ca. 2 mm	ca. 1 mm
Fl.		Apr–May	Apr–Jun	Apr–May
Fr.		May–Jun	Jul-Aug	Jun-Jul

Distribution and habitat. At this time, based on our field observations, *Sedum lipingense* is only known to occur in Longxi village, Mengyan town, Liping County, Guizhou Province. It grows on moist limestone rocks, at ca. 800 m altitude, in groups of several hundred individuals.

Conservation status. This species is currently known to occur in a single valley and we suggest its placement in the Data Deficient category of IUCN (2017).

Phenology. This new species was observed flowering from April to May and fruiting from May to June.

Etymology. The specific epithet '*lipingense*' is derived from the plant's locality, Liping County, Guizhou Province, China.

Acknowledgments

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References

- Chen ML, Han X, Zhang LF, Cao XH (2017) *Sedum peltatum* (Crasulaceae): A new species from Anhui, China. Bangladesh Journal of Botany 46(3): 847–852.
- Fu SX, Fu KJ (1984) Crassulaceae. Flora of China (vol 34-1). Science Press, Beijing, 31–220.
- Fu KJ, Ohba H (2001) Crassulaceae. In: Wu ZY, Raven PH (Eds) Flora of China, vol 8., Brassicaceae through Saxifragaceae. Science Press, Beijing and Missouri Botanical Garden Press, St Louis, 202–268.
- Ito T, Nakanishi H, Chichibu Y, Minoda K, Kokubugata G (2017a) *Sedum danjoense* (Crassulaceae), a new species of succulent plants from the Danjo Islands in Japan. Phytotaxa 309(1): 23–34. https://doi.org/10.11646/phytotaxa.309.1.2
- Ito T, Yu CC, Nakamura K, Chung KF, Yang QE, Fu CX, Qi ZC, Kokubugata G (2017b) Unique parallel radiations of high-mountainous species of the genus *Sedum* (Crassulaceae) on the continental island of Taiwan. Molecular Phylogenetics and Evolution 113: 9–22. https://doi.org/10.1016/j.ympev.2017.03.028
- IUCN (2017) Guidelines for using the IUCN red list categories and criteria. Version 13. Prepared by the Standards and Petitions Subcommittee. https://www.iucnredlist.org/resources/redlistguidelines
- Jin XF, Zhang HW, Xie JB, Zhao YJ (2010) Taxonomic Notes on *Sedum* s. str. (Crasullaceae) from Zhejiang Province, China. Journal of Hangzhou Normal University 9(3): 165–171. [Natural Science Edition]
- Jin SH, Zhou YY, Ding BY, Wang RW, Jin XF (2013) *Sedum kuntsunianum* (Crassulaceae: Sedoideae), a new species from southern Zhejiang, China. Phytotaxa 105(2): 33–38. https://doi.org/10.11646/phytotaxa.105.2.1

- Li YK, Huang WL, Wang XG, Zhang XS, Wu JR (1985) Crassulaceae. In: Flora of Guizhou, vol 2. Guizhou People's Press, Guiyang, 173–197.
- Thiede J, Eggli U (2007) Crassulaceae. In: Kubitzki K (Ed.) The families and Genera of Vascular Plants (Vol. 9). Springer, Hamburg, 83–118. https://doi.org/10.1007/978-3-540-32219-1_12
- Wang H, Song XJ, Liu QW (2005) *Sedum hoi*, a New Species of the Crassulaceae from Zhejiang, China. Yunnan Zhi Wu Yan Jiu 27(3): 381–382.
- Wu LH, Liu YJ, Zhou SB, Guo FG, Bi D, Guo XH, Baker AJM, Smith JAC, Luo YM (2013) Sedum plumbizincicola X.H. Guo et S.B. Zhou ex L.H. Wu (Crassulaceae): A new species from Zhejiang Province, China. Plant Systematics and Evolution 299(3): 487–498. https://doi.org/10.1007/s00606-012-0738-x
- Xie DM, Peng DY, Fang CW, Qin MJ, Wang DQ, Huang LQ (2014) *Sedum spiralifolium* (Crassulaceae): A new species from Anhui Province, China. Phytotaxa 183(3): 171–182. https://doi.org/10.11646/phytotaxa.183.3.4
- Yang CD, Wang XY, Gou GQ (2012) *Sedum fanjingshanensis* C. D. Yang et X. Y. Wang, a new species of *Sedum* L. Bulletin of Botanical Research 32(4): 389–391.