Gentianella macrosperma, a new species of Gentianella (Gentianaceae) from Xinjiang, China

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Abstract

Gentianella macrosperma Ma ex H.F. Cao, J.D. Ya & Q.R. Zhang, a new species of Gentianaceae from Xinjiang, Northwest China is described and illustrated. This new species is unique in having equal length of corolla lobe and corolla tube, nectaries located at the throat of the corolla tube and large seeds up to 1.6 mm in diameter. In addition, an updated identification key to the Chinese species of Gentianella is provided.

Keywords
Gentianella, ITS, matK, Morphology, Swertiinae, Taxonomy, Xinjiang

Introduction

Gentianella Moench (Gentianaceae) consists of approximately 300 species distributed from the temperate, arctic and alpine regions of the Northern Hemisphere, to South America, Australia and New Zealand (Pringle 2017). About 70% of species (ca. 200 spe-
cies) occur in South America, where new species continue to be discovered (Pfanzelt et al. 2015; Pringle 2015, 2017; Pringle and Grant 2017). Molecular phylogenetic studies indicated that Gentianella was polyphyletic, and the new circumscription of Gentianella s. str. contains species with one nectary per petal lobe (von Hagen and Kadereit 2001, 2002). However, the taxonomic placement of the Asiatic species with two nectaries per corolla lobe has yet to be determined. Before the phylogenetically-based concept of Asiatic gentianellas proposed, the description of this genus published in Flora of China (Ho and Pringle 1995) remains applicable in the present context. There are 10 species of Gentianella reported from China and mainly distributed in northern China and alpine areas of southwest China mountains (Ho and Pringle 1995, Chen et al. 2011).

During the field expedition to west of Xinjiang, China, an unusual species of Gentianaceae was collected. Its corolla campanulate without plicae and fringed scale, lobed to middle of corolla, two nectaries per corolla lobe located at the corolla tube fit the main characters of Gentianella. Subsequent morphological investigation and molecular study supported this species as new to science and described here.

**Materials and methods**

Specimen collections of Gentianella were carefully examined, especially the relevant species, including G. holostoides Schott & Kotschy ex N.M. Pritch., G. longicarpa (Gilli) Holub, G. sibirica (Kunst.) Holub, G. stoliczkae (Kurz ex C.B. Clarke) Holub and G. umbellata (M. Bieb.) Holub. Collections at the following herbaria (BM, FR, GH, GLM, HIMC, HNWP, JE, K, E, KFTA, KUN, MA, MPU, MW, P, PE, PEY, W, WAG) were checked on-site and via Chinese Virtual Herbarium (CVH, http://www.cvh.ac.cn/), Global Biodiversity Information Facility (GBIF, https://www.gbif.org/) and Global Plants on JSTOR (https://plants.jstor.org/). The high-resolution images of type specimen of G. sibirica (LE01043410, LE01043411, LE00050650) were obtained from curators of LE. Relevant literatures were investigated (Gillett 1957; Shishkin and Bobrov 1967; Omer et al. 1988; Ho and Pringle 1995; Omer 1995; Struwe et al. 2002; Aitken 2007; Chen et al. 2011; Mohd et al. 2018). Line drawings, description and most of photographs were based on the latest collections (J.D. Ya et al. 17CS16327), except that the images of seeds were from the type specimen (Shun-Li Chen Tianyi281, PE00029466). The conservation status of the new species was evaluated according to the guidelines of the IUCN Red List Categories and Criteria (IUCN 2017).

Fresh leaves of this new species were dried immediately by using silica gel for DNA extraction. Genomic DNA extraction, amplification and DNA sequencing of ITS and the plastid matK followed the protocol described by Xi et al. (2014) and sequences of relevant species were downloaded from GenBank (Appendix 1).

The molecular phylogenetic tree of 88 species representing 13 genera of Gentianaceae was reconstructed using Bayesian Inference (BI) and Maximum Likelihood (ML). Chelonanthus alatus (Aubl.) Pulle (Gentianaceae: Helieae) was chosen as outgroup (Figure 1). ITS and matK datasets were combined for analysis. BI analysis was
Figure 1. The major-rule consensus tree of ML analysis based on the total dataset, including ITS and matK. ML bootstrap values and BI posterior probabilities are shown on branches.
performed using MrBayes 3.26 (Ronquist and Huelsenbeck 2003). Markov Chain Monte Carlo (MCMC) analysis was performed using MrBayes for 10,000,000 generations for the combined dataset, with two simultaneous runs, with each run comprising four incrementally heated chains. BI analysis was started with a random tree and sampled every 1000 generations. The combined dataset was partitioned and the best-fit DNA substitution model for two DNA regions using Bayesian Information Criterion (BIC) was estimated using jModeltest 2 (Darriba et al. 2012). ML analysis was conducted with RAxML 8.2.10 (Stamatakis et al. 2008) using the GTR substitution model with gamma-distributed rate heterogeneity amongst sites and the proportion of invariable sites estimated from the data. Support values for nodes/clades were estimated from 1000 bootstrap replicates.

Results

The ITS matrix was 689 bp in length including 376 variable sites and 266 parsimony-informative sites and the matK matrix was 821 bp in length including 286 variable sites and 198 parsimony-informative sites. The best-fit BIC model of ITS and matK datasets was SYM+G and TVM+G, respectively. The major-rule consensus tree of both BI and ML analyses with support values is shown in Figure 1.

Phylogenetic analyses using ML and BI methods identified that Gentianella, Swertia L. and other genera in subtribe Swertiinae are not monophyletic, which shows a similar conclusion as previous studies (von Hagen and Kadereit 2001, 2002; Xi et al. 2014). Current new species and 44 other Gentianella species were strongly supported as monophyletic (BI PP = 1.00, ML BS = 93; Figure 1). G. arenaria (Maxim.) T.N. Ho, G. angustiflora H. Smith, G. azurea (Bunge) Holub, G. gentianoides (Franch.) H. Smith and G. moorcroftiana (Wall. ex G. Don) A. Shaw formed different clades with Comastoma Toyok., Lomatogonium A. Braun, Swertia and other genera in Swertiinae.

Phylogenetic analyses showed that this new species and G. holosteoides formed a clade (BI PP = 0.93), then sister to the clade including G. aurea (L.) H. Smith, G. umbellata and G. longicarpa (Figure 1). Three samples of G. stoliczkae were located at most basal of the new species clade (BI PP = 1.00, ML BS = 79).

Taxonomic treatment

Gentianella macrosperma Ma ex H.F. Cao, J.D. Ya & Q.R. Zhang, sp. nov. urn:lsid:ipni.org:names:60479356-2 Figures 2, 3

Diagnosis. Resembles G. holosteoides, G. longicarpa, G. sibirica, G. stoliczkae and G. umbellata, but differs from them by having even flower size, corolla white, corolla lobe as long as corolla tube, nectaries located close to the throat of the corolla tube and larger seeds.
Figure 2. Gentianella macrosperma, sp. nov. A plant B flower, top views C–D show opened corollas, 4- and 5-merous, respectively E flower, showing the length of calyx and corolla subequal F calyx, showing 4-merous G calyx, showing 5-merous H capsule I seeds. Drawn by R.M. Zhang. H and I from the isotype S.L. Chen Tianyi281 (PE00029471), others from the paratype J.D. Ya, Q.R. Zhang & X.J. Hu 17CS16327 (KUN1443565). Scale bars: 2 cm (A); 5 mm (B); 2 mm (C–H); 0.5 mm (I).
Figure 3. *Gentianella macrosperma*, sp. nov. A plant in nature habitat B flowers and inflorescence C flowers, showing pedicels and upper leaves D–E front view and side view of corolla, showing nectaries located close to the throat of the corolla tube F middle cauline leaf, abaxial view, showing veins G plants specimen (from KUN1443554) H opened corolla (5-merous) showing ovary I calyx J seed, front view (left and middle) and side view (right) (from S.L. Chen Tianyi281 (PE00029471)). I, H from the paratype J.D. Yu, Q.R. Zhang & X.J. Hu 17CS16327 (KUN1443565). Scale bars: 5 cm (A, G); 2 cm (B); 2 mm (C–E, I, H); 1 mm (J).
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**Type.** CHINA. Xinjiang: Ili Kazak Autonomous Prefecture, Gongliu County, Ji’ergelang Township, Qiaxi Village, on the mountain ridge in the forest, 1780 m elev., 6 September 1956, Shun-Li Chen Tianyi281 (holotype: PE00029466; isotype: PE00029453; PE00029471).

**Description.** Herbs, annual. Roots slender, yellow. Stems 30–40 cm, erect, subquadrangular, glabrous, yellowish-green, 2.0–2.5 mm in diameter; branched from the base in axils of each node, more slender, suberect or slightly ascending. Leaves opposite, basal leaves not rosette and withered at anthesis, petiole conspicuous, 7–10 mm long, leaves oblong-spatulate, 14–17 × 2–6 mm, base tapering into petiole, margin entire, apex rounded, veins 3–5, raised abaxially and slightly sunken adaxially; lower cauline leaves obovate-spatulate or rounded-spatulate, petiole 10–18 mm long, leaf blades with petiole 18–31 × 10–11 mm, both surfaces glabrous, base tapering into conspicuous petiole, margin entire, apex rounded, veins 5–7 raised abaxially and slightly sunken adaxially; middle leaves on primary stem elliptic, ovate-elliptic, 25–38 × 10–15 mm, base rounded or truncate, inconspicuously short or subsessile, both surfaces glabrous, margin entire, apex rounded, veins 5–9, raised abaxially and slightly sunken adaxially; upper stem leaves ovate-elliptic to ovate, 15–25 × 7–12 mm, with terminal two pairs of leaves nearly in whorls, both surfaces glabrous, base rounded, sessile, margin entire, apex acute, veins 3–5, raised abaxially and sunken adaxially; lateral branches leaves smaller, 10–15 × 4–7 mm. Cymes terminal and axillary, 3–4 flowers per leaf axil, terminal inflorescence 8–10 flowers, dense, inflorescence flowering at different times, pedicel variable in length and up to 36 mm. Flowers 4-merous (rarely 5-merous), all flowers almost the same size (terminal corolla as long as or slightly longer than others), rotating arrangement. Calyx 3.5–4.5 mm long, slightly shorter than corolla or as long as corolla, divided almost to the base, calyx tube 0.7–0.8 mm long, membranous, lobes green, distinctly unequal, 2 slightly larger, ob lanceolate to linear-ob lanceolate, 3.0–3.5 × 0.7–1.0 mm, 2 (–3) slightly smaller, linear, 2.3–3.0 × 0.4–0.5 mm, apex acute or acuminate, margin scabrous, midvein raised abaxially, sinus obtuse. Corolla white, campanulate, 4.0–4.5(5.0) mm long; corolla tube 2.1–2.4 mm long; lobes ovate, with light brown fine longitudinal veins, 2.2–2.5 × 1.5–1.8 mm, apex obtuse and mucronate, margin entire. Nectaries 8(–10), green, oblong, naked and indistinct, two nectaries per corolla lobe located very close to the throat of the corolla tube, ca. 0.2 mm from the top of corolla tube. Stamens inserted at middle of corolla tube, filaments white, linear, 1.1–1.4 mm long, anthers blue, rectangular, 0.2–0.3 mm long; ovary ellipsoidal, ca. 2.0 mm long. Style short, linear, 0.4–0.5 mm long, stigma small, 2-lobed. Gynophore short, 0.2–0.3 mm long. Capsule elliptic, a concavity sometimes present in the centre, 2.5–4.0 mm long, usually with 2–8 seeds each capsule. Seeds brown, glossy, flat-ellipsoid, 1.2–1.6 × 0.5–0.9 mm, seed coat wrinkled-reticulate (smooth when immature).

**Phenology.** Flowering and fruiting from June to September.

**Distribution and habitat.** *G. macrosperma* is distributed in Gongliu county and Xinyuan county, west of Xinjiang, China. It grows in thickets on the slope or on the mountain ridge in the forest of *Picea schrenkiana* Fisch. & Mey. at an elevation of 1729–1780 m.
Etymology. The specific epithet “macrosperma” refers to the larger seeds of this new species.

Vernacular name. Chinese mandarin: da zi jia long dan (大籽假龙胆)

Conservation status. Currently only known from three localities in west of Xinjiang, therefore considered to be Vulnerable (VU D2) (IUCN 2017).

Additional specimens examined (paratypes). CHINA. Xinjiang: Ili Kazak Autonomous Prefecture, Xinyuan County, on the road from Xinyuan County to the gold mine, 43°16'06.45"N, 83°17'42.90"E, 1729 m elev., 1 July 2017, J.D. Ya, Q.R. Zhang & X.J. Hu 17CS16327 (KUN1443565!, KUN1443566!, KUN1443554!); Ili Kazak Autonomous Prefecture, Gongliu County, Mohuer Township, Damohe Village, 8 August 1976, Shu-Run Liu s.n. (HIMC0026063!, HIMC0026064!). The sheet 0026064 presents a mixture of Swertia dichotoma Linn. which was labelled as “A” and G. macrosperma labelled as “B”)

Discussion

It was Prof. Yu-Quan Ma (also as Yu Chuan Ma), a specialist of Gentianaceae, who first recognised this plant as a distinct new species and inscribed the name “Gentianella macrosperma Ma” on the specimen kept at PE. Later the same year, he proposed another name “Gentianella procumbens Ma” to the same collections, corresponding to its procumbent stems. However, both names were never published. Based on field observation and specimen examination, procumbent stems occurred occasionally in some individuals, the character of larger seeds being easily distinguished from other Gentianella species.

In all the known Chinese species of Gentianella, the length of corolla lobes is shorter than that of the corolla tube and nectaries which are located at the base or middle of the corolla tube. The same length of corolla lobes and corolla tube and nectaries positioned at the throat of the corolla tube make G. macrosperma a distinctive species amongst them. Its large seeds up to 1.6 mm in diameter are perhaps unique amongst the Asiatic species of Gentianella.

G. macrosperma is similar in size and shape of the corolla lobe to G. sibirica and G. longicarpa, but further differs from them both in the lack of rosette basal leaves, predominant 4-merous flowers and smaller corolla, no more than 5 mm long, except the corolla lobed to the middle, nectaries position and seeds size. Gentianella longicarpa, which is endemic to Afghanistan, is also distinct from G. macrosperma in its light-pink, pale blue or lilac-violet flower and larger corolla up to 8 mm long and all calyx lobes are shorter than the corolla tube. G. macrosperma is similar in habit and inflorescences to G. umbellata and G. stoliczkae. The flower of G. umbellata is larger than those of G. macrosperma and, although the size of the corolla lobe in the two species overlaps, the corolla lobe is much shorter than the corolla tube in G. umbellata. In G. stoliczkae, flowers are in densely clustered cymes, the corolla are generally much larger up to 20 mm long with various colours from purple, pink, pale blue to yellow and the capsule has a short gynophore ca. 1–2 mm long.
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The molecular evidence shows that *G. macrosperma* has the closest relationship with *G. holosteoides* which is native to Turkey and Pakistan and they also share similar floral whorls and basal leaves shape, but plants of *G. holosteoides* are smaller in stature, no more than 5 (7) cm height; it further differs from *G. macrosperma* in its smaller basal leaves, larger flowers with corolla lobes shorter than corolla tube, nectaries position at corolla base and smaller, numerous seeds. A detailed morphological comparison is given in Table 1.

Von Hagen and Kadereit (2001) proposed *Gentianella* s. str. to only include species with one nectary per petal lobe, however, *G. umbellata* and *G. stoliczkae* represented in their study are both binectariate species. Current molecular analyses also shows the binectariate *G. macrosperma* clustered into von Hagen and Kadereit’s *Gentianella* s. str. A careful selection of species across wider geographic regions of this genus and data from more nuclear and chloroplast sequences may clarify the generic circumscription in *Gentianella*.

### Key to species of *Gentianella* in China

The following key is based on Flora of China (Ho and Pringle 1995), Flora of the U.S.S.R. (Shishkin and Bobrov 1967) and other literature (Omer et al. 1988; Aitken 2007; Chen et al. 2011). It includes 11 species of *Gentianella* in China.

1. Corolla lobes fimbriate at base ................................................................. *G. acuta*
   - Corolla lobes glabrous at base .......................................................... 2
2. Nectaries above the middle of corolla tube ........................................... 3
   - Nectaries at the base of corolla tube .................................................. 4
3. Plant 12–40 cm tall, nectaries close to the throat of corolla tube, seeds 1.2–1.6 mm in diameter ................................................................. *G. macrosperma*
   - Plant 1–4 cm tall, nectaries just above the middle of corolla tube, seeds 0.7–0.8 mm in diameter ......................................................... *G. pygmaea*
4. Margin and midvein of calyx lobe blackish ............................................ *G. azurea*
   - Calyx not as above ............................................................................. 5
5. Stem densely purple pilose .................................................................... *G. gentianoides*
   - Stem glabrous (sometimes sparsely pilose in *G. moorcroftiana*) .......... 6
6. Flowers often angled, corolla tube 3–4 times longer than lobe ... *G. angustiflora*
   - Flowers not angled, corolla tube 1–3 time(s) longer than lobe .............. 7
7. Corolla lobes apically obtuse or round .................................................. 8
   - Corolla lobes apically mucronate ....................................................... 9
8. Flowers 5-merous, stem leaf blades linear .......................................... *G. moorcroftiana*
   - Flowers 4-merous, stem leaf blades spatulate to oblong-spatulate .... *G. arenaria*
   - Corolla lobes densely papillate outside ........................................... *G. anomala*
9. Corolla 7–20 mm long, terminal ones ca. 20 mm, lobes 3–7 mm .... *G. stoliczkae*
   - Corolla 4–10 mm long, terminal ones up to 10 mm, lobes ca. 2 mm .... *G. sibirica*
Table 1. Morphological comparison between *Gentianella macrosperma* and related species.

|                      | *G. macrosperma* | *G. holosteoides* | *G. longicarpa* | *G. sibirica* | *G. stoliczkae* | *G. umbellata* |
|----------------------|-----------------|------------------|----------------|---------------|----------------|----------------|
| Plant height (cm)    | 12–40           | up to 5          | 9–22           | (1–)10–20     | 10–45          | (4–)10–35      |
| Basal leaves (mm)    | not roslate, obovate-spathulate 14–17 × 2–6 | roslate, spathulate-ovate or lanceolate, 3–5 × 1–3 | roslate, spathulate, oblong-obovate, 7–16 × 3–8 | roslate, oblong-obovate, 6–20 × 2–6 | roslate, ovate-lanceolate to ovate, 10–35 × 6–20 | roslate, spathulate, obovate-lanceolate, 8–25 × 5–12 |
| Cauline leaves (mm)  | oval to ovaloid, apex rounded, the uppermost sometimes acute, 15–38 × (7–)10–15 | lanceolate-oblanceolate or elliptic, apex acute, 5–15 × 2–6 | ovate-oblong, ovate or ovate-lanceolate, apex acute, 3–6–9 | ovate-oblong, ovate-lanceolate, apex acute, 6–20 × 3–9 | oblong-lanceolate, lanceolate to ovate-lanceolate, apex acute, (20–)25–40 × (2–)10–15 | oblong-ovate, oblong-lanceolate, apex acute, 8–25 × 4–18 |
| Calyx length (mm)    | 3.5–4.5         | 4–8              | 4–5            | 3–6           | 8–11           | 4–10           |
| Floral whorls        | 4(5)–merous     | 4(5)–merous      | 5-merous       | 5(4)–merous   | 5-merous       | 5-merous       |
| Flower size          | almost all of the same size | variable in size, terminal ones 1–2 × larger than others | variable in size, terminal ones 1–1.5 × larger than others | variable in size, terminal ones 1–2 × larger than others | variable in size, terminal ones 2–3 × larger than others | variable in size, terminal ones 2–3 × larger than others |
| Corolla colour       | white           | pale blue to blue | pale blue, light-pink, or lilac-violet | predominantly pink, yellowish or white, rarely pale blue | purple, pink, pale blue or yellowish | pale azure, purple, pink, yellowish or mixture of these, rarely white |
| Corolla shape        | campanulate     | tubular to campanulate-tubular | tubular to campanulate-tubular | tubular or tubular-infundibular | tubular to campanulate-tubular | tubular to campanulate-tubular |
| Corolla length (mm)  | 4.0–4.5(–5.0)   | 6–12             | (5–) 6–8       | (5–) 6–7(–10) | 7–20           | (5–) 8–11(–15) |
| Corolla lobes        | 2.0 mm long, the same length as corolla tube | 1.5–3.0 mm long, much shorter than corolla tube | 2–3 mm long, shorter than corolla tube | ca. 2 mm long, much shorter than corolla tube | 3–7 mm long, much shorter than corolla tube | 2–3(4) mm long, much shorter than corolla tube |
| Nectaries            | 8(10), at top of corolla tube | 8(10), at basal part of corolla tube | 10, at basal part of corolla tube | 8–10, at basal part of corolla tube | 10, at basal part of corolla tube | 10, at basal part of corolla tube |
| Stamens              | 1.1–1.4 mm      | –                | –              | 2–4 mm        | ca. 7 mm       | 1–5 mm         |
| Anthers              | blue, 0.2–0.3 mm | –                | –              | blue, 0.5–0.7 mm | yellow, 1.0–1.2 mm | – |
| Gynophore            | 0.2–0.3 mm      | subsecille       | sessile        | subsecille    | 1.5–2.2 mm     | sessile        |
| Seeds                | 2–8 per capsule, 1.2–1.6 mm in diameter | numerous per capsule, ca. 0.8–1.0 mm in diameter | numerous per capsule, 0.2–0.3 mm in diameter | numerous per capsule, 0.1–0.2 mm in diameter | numerous per capsule, ca. 0.8 mm in diameter | numerous per capsule, 0.2–0.3 mm in diameter |
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Appendix

Appendix 1. Samples for phylogenetic analysis using matK and ITS sequences with voucher information, GenBank accession number.

| Species                          | Voucher specimen (Herbarium/No.) | Locality                  | matK          | ITS          |
|----------------------------------|----------------------------------|---------------------------|---------------|--------------|
| OUTGROUP                         |                                  |                           |               |              |
| Chelonanthis alatus (Aubl.) Pulle | Maas 9316 (U)                    | French Guiana             | KX904551      | KX904610     |
| **GENTIANEAE GROUP**             |                                  |                           |               |              |
| Crawfurdia speciosa Wall.         | KEKE 1244 (K)                    | N/A                       | AJ010512/     | AJ294586/    |
|                                  |                                  |                           | AJ011441      | AJ294646     |
| Gentiana crassicaulis Duthie ex Burkill | xuechy090107 (KUN) | China                   | KC861277      | KC861348     |
| Gentiana dahurica Fisch.          | xuechy0076 (KUN)                 | China                     | KC861279      | KC861350     |
| Gentiana frigida Haenke          | N/A                              | Germany (Schachen Bot. Garden), cultivated | AJ388166/     | AJ294588/ |
|                                  |                                  |                           | AJ388236      | AJ294648     |
| Species                          | Voucher specimen (Herbarium/No.) | Locality | matK       | ITS          |
|---------------------------------|----------------------------------|----------|------------|--------------|
| *Gentianella macrosperma*       | xuechy0065 (KUN)                 | China    | KC861282   | KC861353     |
| **SWERTIIINAe GROUP**           |                                  |          |            |              |
| *Comastoma cyananthiflorum*     | XHC120021 (KUN)                  | China    | KC861250   | KC861320     |
| (Franch.) Holub                 | CEE-88 (E 00025334)              | China    | AJ406324/  | AJ294585/    |
| *Comastoma jigsawiae* T.N. Ho  | Chen0423 (KUN)                   | China    | AJ406353/  | AJ294645     |
| & J.Q. Liu                      | xuechy090036 (KUN)               | China    | AJ406351/  | AJ294647     |
| *Comastoma pulmonarium*         | GLM-081307 (KUN)                 | China    | KC861238   | KC861306     |
| (Turcz.) Toyok.                 | K. Gutsche 20 (MJG)              | N/A      | AJ406325/  | AJ294587/    |
| *Praesia alciculata* Griseb.    |                                  |          | AJ406354/  | AJ294657     |
| *Gentianella amarella* (L.)     | W.J. Schrenk (FR)                | N/A      | AJ406326/  | AJ294591/    |
| Börner                          |                                  |          | AJ406355/  | AJ294651     |
| *Gentianella angustiflorum*     | Edinburgh Makalu Expedition 430 | Nepal    | AJ406327/  | AJ294592/    |
| (E 00025322)                    |                                  |          | AJ406356/  | AJ294652     |
| *Gentianella antipoda* (Kirk)   | CHR 510015                       | New Zealand | –      | AJ136500     |
| T.N. Ho & S.W. Liu              |                                  |          |            |              |
| *Gentianella arenaria* (Maxim.) | T.N. Ho et al. 435 (E 00025341)  | N/A      | AJ406328/  | AJ294593/    |
| T.N. Ho                        | K. Gutsche 45 (MJG)              | N/A      | AJ101517/  | AJ294594/    |
| & Skalicky, Chricht & Gill      |                                  |          | AJ101146/  | AJ294654     |
| *Gentianella azuensis* (Petric) | CHR 509942                       | New Zealand | –      | AJ136504     |
| T.N. Ho & S.W. Liu              | H. Smith 4131 (E 00025348)       | N/A      | AJ406329/  | AJ294595/    |
| *Gentianella aurea* (L.) H.     |                                  |          | AJ406357/  | AJ294655     |
| Smith                          |                                  |          | AJ406358/  | AJ294656     |
| *Gentianella auriculata* (Pall.)|                                  |          | AJ406330/  | AJ294596/    |
| J.M. Gillett                   |                                  |          | AJ406350/  | AJ294657     |
| *Gentianella austrica* (A. Kern. & Jos.Kern.) Holub | N/A (MJG) | Germany (Schachen Bot. Garden), cultivated | – | AJ294597/ |
| *Gentianella azurea* (Bunge)    | xuechy090033 (KUN)               | China    | KC861284   | KC861355     |
| Holub                          |                                  |          |            |              |
| *Gentianella azurea* (Bunge)    | T.N. Ho, B. Bartholomew, M.      | China    | AJ406331/  | AJ294598/    |
| Gilbert 1312 (E 00025339)       |                                  |          | AJ406359/  | AJ294658     |
| *Gentianella azurea* (Bunge)    | Yangyp-Q-0255 (KUN)              | China    | MN067526*  | MK416127*    |
| Holub                          |                                  |          |            |              |
| *Gentianella bellidifolia*      | 19923274                         | Scotland | AJ388162/  | AJ294599/    |
| (Hook.f.) Holub                |                                  | Edinburgh Bot. Garden, cultivated | AJ388232 | AJ294659     |
| *Gentianella bohemica* Skalicky | 015                               | Czech Republic | –      | AJ580570     |
| *Gentianella carnosii* G.L.     | S. Gonzales, S. Acevedo 2033 (TEX) | N/A      | AJ406332/  | AJ294600/    |
| Nesom & B.L. Turner             |                                  |          | AJ406360/  | AJ294660     |
| *Gentianella caseosa* (Lodd. ex | J. C. Archibald 8208 (E 00025347) | N/A      | AJ294601/  | AJ294661     |
| Sims) Holub                    |                                  |          | AJ294602/  | AJ294662     |
| *Gentianella cerastoides*       | R. Greissl (MJG)                 | N/A      | AJ101518/  | AJ294603/    |
| (Kunth) Fabris                 |                                  |          | AJ101147/  | AJ294663     |
| *Gentianella cernua* (Kunth)    | C.Viteri 4410 (MO)               | N/A      | –          | AJ294603/    |
| Fabris                         |                                  |          | –          | AJ294663     |
| *Gentianella cosmantha* (Griseb.) J.S. Pringle | J.G. Haukes, J.P.Hjirting, K. Rahn 3659 (L. 424359) | N/A      | AJ406333/  | AJ294604/    |
| *Gentianella diemensis* (Griseb.) J.H. Willis | H. Hurka (MJG) | N/A      | AJ406336/  | AJ294664     |
| *Gentianella engadensis*        | Ge002                            | Switzerland | –      | AJ580559     |
| (Wettst.) Holub                |                                  |          |            |              |
| *Gentianella fastigiata*        | K. Gutsche (MJG)                 | N/A      | –          | AJ294606/    |
| (Benth.) Fabris                |                                  |          | –          | AJ294666     |
| *Gentianella florida* (Griseb.) | R. Ehrich 444 (MJG)              | N/A      | AJ406334/  | AJ294607/    |
| Holub                          |                                  |          | AJ406362/  | AJ294667     |
| *Gentianella foliosa* (Kunth)   | 1994-508                         | England (Kew Bot. Garden), cultivated | – | AJ294608/ |
| Fabris                         | Ge002                            | Switzerland | –      | AJ294668     |
| *Gentianella gentianoides*      | xuechy090065 (KUN)               | China    | KC861285   | KC861356     |
| (Franch.) H. Smith             |                                  |          |            |              |
| *Gentianella germanica* (Wild.)| xuechy090094 (KUN)               | China    | KC861286   | KC861357     |
| E.F. Warburg                   | 20                               | Germany  | –          | AJ580562     |
| Species                                | Voucher specimen (Herbarium/No.) | Locality       | matK            | ITS                  |
|----------------------------------------|----------------------------------|----------------|------------------|----------------------|
| Gentianella germanica (Willd.) E.F. Warburg | J.W. Kadereit (MJG) | N/A            | AJ406335/ AJ406363 | AJ294609/ AJ294669 |
| Gentianella hirculus (Griseb.) Fabris    | J.L. Clarke 1787 (QCNE) | N/A            | –                | AJ294610/ AJ294670   |
| Gentianella holosteoides Schott & Korsky ex N.M. Pritch. | Southampoton University 179 (K) | N/A            | –                | AJ294611/ AJ294671   |
| Gentianella macroperma Ma ex H.F. Cao, J.D. Ya & Q.R. Zhang | 1/CS16527 (KUN) | China          | MN067/523*       | MK416132*            |
| Gentianella lineata (Kirk) T.N. Ho & S.W. Liu | CHR 509866 | New Zealand | –                | AJ136503             |
| Gentianella longicarpa (Gilli) Holub     | D. Podliech 12436 (M) | N/A            | –                | AJ294612/ AJ294672   |
| Gentianella magellanica (Gaudich.) Fabris | K. Kubitzki, T. Feurer 99-10 (MJG) | N/A            | AJ406336/ AJ406364 | AJ294613/ AJ294673   |
| Gentianella microcalyx (Lemmon) J. M. Gillett | E. Joyal, J. Enrique 1853 (TEX) | N/A            | AJ406337/ AJ406365 | AJ294614/ AJ294674   |
| Gentianella montana (G. Forst.) Holub    | CHR 509944 | New Zealand | –                | AJ136491             |
| Gentianella moorcroftiana (Wall. ex G. Don) A. Shaw | R. McBeath 2093 (E 00025318) | N/A            | AJ406338/ AJ406366 | AJ294615/ AJ294675   |
| Gentianella narcissoides (Gilg) T.N. Ho & S.W. Liu | L. Naessany 14 (MJG) | N/A            | –                | AJ294616/ AJ294676   |
| Gentianella patula (Kirk) Holub           | 19932978 | Scotland (Edinburgh Bot. Garden), cultivated | AJ406339/ AJ406367 | AJ294617/ AJ294677   |
| Gentianella peruviana (Griseb.) Fabris    | 19950534 | Scotland (Edinburgh Bot. Garden), cultivated | AJ388163/ AJ388233 | AJ294618/ AJ294678   |
| Gentianella propinqua (Richardson) J.M. Gillett | G. Halliday A 333/75 (E 00025300) | North America | AJ406340/ AJ406368 | AJ294619/ AJ294679   |
| Gentianella quinquefolia (L.) Small       | Bozeman, Ramseur, Radford 45200 (E 00025241) | North America | AJ406341/ AJ406365 | AJ294620/ AJ294680   |
| Gentianella quinquefolia (L.) Small       | D. Pitillo 12106 (WCUH) | America       | –                | EU812469             |
| Gentianella rapunculoides (Willld. ex Schult.) J.S. Pringle | R. Greissl 616 (MJG) | N/A            | –                | AJ294621/ AJ294681   |
| Gentianella ruizii (Griseb.) Holub        | Weigend, Weigend 2000/386 (NY) | N/A            | AJ406342/ AJ406370 | AJ294622/ AJ294682   |
| Gentianella ruticola (Kunth) Holub        | 199930516 | Scotland (Edinburgh Bot. Garden), cultivated | –                | AJ294623/ AJ294683   |
| Gentianella saxosa (G. Forst.) Holub      | Gutsche (MJG) | N/A            | –                | AJ406343/ AJ406371   |
| Gentianella splendens (Gilg) Fabris       | J.L. Clarke 1855 (QCNE) | N/A            | AJ295336/ AJ295337 | AJ294624/ AJ294684   |
| Gentianella stoliczkae (Kurz ex C.B. Clarke) Holub | LiuJQ0028 (KUN) | China          | MN067/524*       | MK416130*            |
| Gentianella stoliczkae (Kurz ex C.B. Clarke) Holub | LiuJQ0071 (KUN) | China          | MN067/525*       | MK416131*            |
| Gentianella stoliczkae (Kurz ex C.B. Clarke) Holub | O. Anders 18178 (M 50043) | N/A            | AJ406344/ AJ406372 | AJ294625/ AJ294685   |
| Gentianella sulphurea (Gilg) Fabris       | J.L. Clarke 1833 (QCNE) | N/A            | –                | AJ294626/ AJ294686   |
| Gentianella thyroides (Hook. f.) Fabris   | D.N. Smith, F. Escalona 10134 (MO) | N/A            | –                | AJ294627/ AJ294687   |
| Gentianella tristicha (Gilg) Fabris ex T.N. Ho & S.W. Liu | D.N. Smith, F. Escalona 10125 (MO) | N/A            | –                | AJ294628/ AJ294688   |
| Gentianella umbellata (M. Bieb.) Holub    | K91-G3 | Georgia       | –                | Z48102Z48132       |
| Gentianella willizemii (Engelm.) J.M. Gillett | M. Lavin 4947 (TEX) | N/A            | –                | AJ294630/ AJ294690   |
| Gentianopsis barbata (Froel.) Ma           | xueschy090085 (KUN) | China          | –                | KC861287/ KC861358   |
Gentianella macrosperma, a new species of Gentianella from Xinjiang, China

| Species | Voucher specimen | Locality | matK | ITS |
|---------|------------------|----------|------|-----|
| Gentianella macrosperma (C.B. Clarke) C.B. Clarke | XHC120060 (KUN) | China | AJ406348/ AJ406376 | AJ294638/ AJ294698 |
| Gentianella crinita (Froel.) Ma | GLM-081543 (KUN) | Germany (Mainz Bot. Garden), cultivated | AJ406345/ AJ406373 | AJ294631/ AJ294691 |
| Halenia elliptica D. Don | GLM-081957 (KUN) | China | AJ406347/ AJ406375 | AJ294640/ AJ294700 |
| Halenia palmeri A. Gray | K.B.v. Hagen 98/41 (MJG) | N/A | AJ406349/ AJ406377 | AJ294639/ AJ294699 |
| Jaeschkea oligosperma (Griseb.) Knobl. | R. McBeath 2300 (E 00025275) | N/A | AJ388171/ AJ388241 | AJ294633/ AJ294693 |
| Lomatogonium bellum (Hemsl.) H. Smith | GLM-06075 (KUN) | China | AJ406346/ AJ406374 | AJ294634/ AJ294694 |
| Lomatogonium carinthiacum (Wulfen) Rchb. | V. Zuev 6649 (BR) | N/A | AJ406346/ AJ406374 | AJ294634/ AJ294694 |
| Lomatogonium forrestii (I.B. Baliour) Fernald | XHC120061 (KUN) | China | AJ388177/ AJ388247 | AJ294635/ AJ294695 |
| Lomatogonium gamosepalum (Burkill) H. Smith | GLM-081372 (KUN 1272996) | China | AJ406346/ AJ406374 | AJ294634/ AJ294694 |
| Lomatogonium oreocharis (Diels) C. Marquand | CLD-90 1106 (K) | N/A | AJ388174/ AJ388247 | AJ294635/ AJ294695 |
| Megacodon stylophorus (C.B. Clarke) H. Smith | GLM-081957 (KUN) | China | AJ388177/ AJ388247 | AJ294636/ AJ294696 |
| Megacodon stylophorus (C.B. Clarke) H. Smith | Kuming, Edinburgh, Gothenburgh Exp. 1378 (E 00025279) | China | AJ388174/ AJ388247 | AJ294635/ AJ294695 |
| Swertia bifolia Batalin | Chem02388 (KUN) | China | AJ388177/ AJ388247 | AJ294636/ AJ294696 |
| Swertia bimaculata (Sieb. & Zucc.) Hook. f. & Thomson ex C.B. Clarke | XHC120026 (KUN) | China | AJ388177/ AJ388247 | AJ294636/ AJ294696 |
| Swertia cincta Burkhill | XCY090050 (KUN) | China | JF956557 | JF978820 |
| Swertia cimosa Gilg. | XCY090098 (KUN) | China | JF956561 | JF978825 |
| Swertia deoria Franch. | XCY090077 (KUN) | China | JF956561 | JF978825 |
| Swertia erythrosticta Maxim. | xuechy090044 (KUN) | China | AJ406346/ AJ406374 | AJ294639/ AJ294699 |
| Swertia franchetiana H. Smith | XHC120048 (KUN) | China | AJ406346/ AJ406374 | AJ294639/ AJ294699 |
| Swertia japonica (Schult.) Makino | N/A (KYO) | Japan (Kyoto Bot. Garden), cultivated | AJ406348/ AJ406376 | AJ294638/ AJ294698 |
| Swertia macrosperma (C.B. Clarke) C.B. Clarke | XHC120060 (KUN) | China | AJ406348/ AJ406376 | AJ294638/ AJ294698 |
| Swertia macrosperma (C.B. Clarke) C.B. Clarke | J.H. de Haas 2765 (U 500099) | N/A | AJ406349/ AJ406377 | AJ294639/ AJ294699 |
| Swertia nervosa (G. Don) Wall. ex C.B. Clarke | XHC120053 (KUN) | China | AJ406349/ AJ406377 | AJ294639/ AJ294699 |
| Swertia patens Burkhill | 09CS1123 (KIB) | China | AJ406349/ AJ406377 | AJ294639/ AJ294699 |
| Swertia perennis L. | K.B. Hungerer (MJG) | N/A | – | AJ294640/ AJ294700 |
| Swertia panicul Hemsl. | 19943574 | Scotland (Edinburgh Bot. Garden), cultivated | AJ406350/ AJ406378 | AJ294641/ AJ294701 |
| Swertia racemosa (Wall. ex Griseb.) C.B. Clarke | J.H. de Haas 2725 (U 500131) | N/A | AJ406351/ AJ406379 | AJ294642/ AJ294702 |
| Swertia volvensii Gilg | U. Hecker 1093 (MJG) | N/A | AJ406352/ AJ406380 | AJ294643/ AJ294703 |
| Swertia yunnanensis Burkhill | XCY090089 (KUN) | China | JF956585 | JF978836 |
| Veratrilla baillonii Franch. | Kuming, Edinburgh, Gothenburgh Exp. 1326 (E 00025273) | China | AJ388196/ AJ388266 | AJ294644/ AJ294704 |

* indicates the taxon was newly sequenced in the present study.