The complete plastome sequence of *Lilium primulinum* var. *burmanicum*, an ornamental and medicinal plant endemic to China

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**ABSTRACT**

*Lilium primulinum* var. *burmanicum* (W. W. Smith) Stearn 1948 is an ornamental and medicinal plant that has an extremely limited distribution in Yunnan, China. Here, we obtained the complete plastome of *L. primulinum* var. *burmanicum* via next-generation sequencing, and conducted phylogenomic analyses with existing species from *Lilium*. The total length of *L. primulinum* var. *burmanicum* was 152,206 bp with a typical quadripartite structure. The whole plastome contained a pair of inverted repeats (IRa/IRb; 26,399 bp) which divided a large single-copy (LSC; 81,854 bp) and a small single-copy (SSC; 17,563 bp). The average GC content among the whole plastome sequence and the LSC, SSC, and IR regions were 37%, 34.8%, 30.6%, and 42.5%, respectively. There were 134 genes detected from the whole plastome sequence, including 87 protein-coding genes, 39 tRNAs, and 8 rRNAs. Phylogenetic analyses using maximum likelihood showed congruent results that *L. primulinum* var. *burmanicum* together with *L. primulinum* var. *ochraceum* formed a single branch. These results demonstrate a close relationship between these variation species. The newly characterized chloroplast genome presented here will provide essential data for further phylogenomic analyses of the intraspecific relationship among *Lilium* species and for conservation genetics research of *L. primulinum* var. *burmanicum*.

*Lilium* (Liliaceae) species are cultivated as potted plants or cut flowers for their bright-colored flowers and extraordinary fragrance (Shahin et al. 2011). More than this, Bulbus Lilii is an important traditional Chinese drug due to its medicinal properties, such as nourishing the lung and alleviating mental stress (Chinese Pharmacopoeia Commission 2020). Therefore, many species of *Lilium* are at risk of extinction in the field due to overutilization.

*Lilium primulinum* var. *burmanicum* (W. W. Smith) Stearn, 1948, is a perennial herb that inhabits the edge of mountainous forests at elevations ranging from ~ 1200 to 2700 m in China (Chen et al. 2000). According to herbarium, *L. primulinum* var. *burmanicum* had been widely distributed, with at least 15 specimen occurrence records in Yunnan. However, based on our field work during 2011–2020, only 4 populations with approximately 20 individuals per population were found. We inferred that with illegal removal from natural habitats for horticultural and medicinal use, *L. primulinum* var. *burmanicum* has likely become vulnerable and decreased in the number of populations, which may lead to extinction in the wild and should draw attention to this nearly endangered species. Here, we first reported the complete chloroplast genome of *L. primulinum* var. *burmanicum* and determined the phylogenetic analyses within *Lilium* species.

Samples of *L. primulinum* var. *burmanicum* were collected from Malipo County in Yunnan, China (23°09′49″N, 104°49′35.4″E), and the specimens were deposited in the Herbarium of Kunming University (YGS1107023; Genshen Yin, yingenshen@126.com). The total genomic DNA of *L. primulinum* var. *burmanicum* was extracted from dry leaves stored in silica using a Plant Genomic DNA Kit (DP305; Tiangen, Beijing) according to the manufacturer’s instructions. The quality and concentration of total DNA were determined by Nanodrop 2000 Spectrophotometers (Thermo Fisher Scientific Inc., Waltham, MA). The total genome was sequenced with paired-end reads of 150 bp on the Illumina HiSeq platform (Illumina Inc., San Deigo, CA). All raw reads were assembled using GetOrganelle software (Jin et al. 2020) with 47 published plastid genome sequences of the genus *Lilium*, referred to as “embplant_pt.” Genome annotation was performed by PGA (Qu et al. 2019) and GeSeq (Tillich et al. 2017) with the plastid genome of *L. pardanthinum* (GenBank accession: MG704135) as the reference. Transfer RNA (tRNA) genes were identified with the tRNAscan-SE program with default parameters (Schattner et al. 2005). OrganellarGenomeDRAW (OGdraw) was used to draw a circular chloroplast genome map (Marc et al. 2018). Finally, the whole chloroplast genome of *L. primulinum* var. *burmanicum* was deposited into GenBank (Accession no. MZ188968).

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The complete chloroplast genome of *L. primulinum* var. *burmanicum* showed a typical quadripartite cycle with a length of 152,309 bp, containing two inverted repeat (IR) regions of 26,394 bp each, which were separated by a large single-copy (LSC, 81,956 bp) region and a small single-copy (SSC, 17,565 bp) region, similar to *L. primulinum* var. *ochraceum* (accession no.: KY748298). The GC content of the complete plastid genome was 37%, whereas the corresponding values of the LSC, SSC, and IR regions were 34.8%, 30.6%, and 42.5%, respectively. We detected 134 genes from the cp genome sequence, consisting of 87 protein-coding genes, 39 tRNA genes, and 8 rRNA genes. Among these, seven protein-coding genes, eight tRNA genes, and four rRNA genes were duplicated. However, the protein-coding genes *cemA* and *ndhD* were annotated in duplicate in a previous study carried out by Du et al. (2017). In both *L. primulinum* species, 25 introns contained genes.

Maximum-likelihood (ML) phylogeny was inferred by IQ-tree software (Minh et al. 2020) from the total chloroplast genome sequence of 19 species, including a representative of *Fritillaria* that was used as outgroup (Figure 1). The best-fit models, GTR + F + I + G4, were used in the analysis with 1000 bootstrap replicates. The phylogenetic results showed a close relationship of *L. primulinum* var. *burmanicum* with *L. primulinum* var. *ochraceum*. There was only one singleton variable site (G/C) between these two species, located in the intergenic regions of *trnT-UGU* and *trnL-UAA*. In addition to the variable site, five InDel events were also detected among the single-nucleotide microsatelliteregions. Information on the complete chloroplast genome of *L. primulinum* var. *burmanicum* could provide valuable insight into conservation and exploitation efforts for this endangered species.

**Ethics of experimentation statement**

The authors ensure that the plant material (*Lilium primulinum* var. *burmanicum*) reported in submitted papers was handled in an ethical and responsible manner and the study was conducted in full compliance with all relevant codes of experimentation and legislation. The total genomic DNA extraction from dry leaves saved in silica does not harm the growth of plants. In addition, the plant samples used in this research were not collected from private land or a nature reserve, so ethics approval was not required for this research.

**Disclosure statement**

No potential conflict of interest was reported by the authors.

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**Data availability statement**

The genome sequence data that support the findings of this study are openly available in GenBank under accession no. MZ188968 (https://www.ncbi.nlm.nih.gov/nuccore/MZ188968.1/). The associated BioProject,
SRA, and Bio-Sample numbers are PRJNA729634, SRR14520563, and SAMN19136515, respectively.

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