

The complete chloroplast genome of *Epimedium brevicornu* Maxim (Berberidaceae), a traditional Chinese medicine herb

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ABSTRACT

Epimedium brevicornu Maxim. is a traditional Chinese medicine herb with good effects on many diseases. In the present paper, the complete chloroplast genome of *E. brevicornu* was sequenced. The complete chloroplast genome of *E. brevicornu* was 159572 bp in length with 38.83% GC content, including four distinct regions: large single-copy region (86535 bp), small single-copy region (17641 bp), and a pair of inverted repeat regions (27698 bp). The chloroplast genome encoded 112 unique genes, which included 78 protein-coding genes, 30 tRNA genes, and 4 rRNA genes. Phylogenetic analysis with the previously reported chloroplast genomes of *Epimedium* showed that *E. brevicornu* with small flowers at first clustered with large-flowered *E. acuminatum* into a strongly supported branch, but not with *Epimedium* species having similar floral characters.

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Epimedium L. is the largest perennial herbaceous genus of Berberidaceae (Ranales). The genus comprises about 62 species, of which about 52 species are distributed in China (Stearn 2002; Ying et al. 2011; Wei et al. 2017). Since *Epimedium* plants have been identified as having good curative effects on many diseases, the development and utilization of *Epimedium* resources have attracted unprecedented attention by research institutions and enterprises (Jiang and Song 2015; Indran et al. 2016; Tan et al. 2016; Xi et al. 2018). However, *Epimedium* has abundant morphological variations inter- and intra-species, especially on the morphology of leaf, which greatly increases the difficulty of identification of the medicinal materials (Zhang et al. 2014; Chen et al. 2015). In previous studies, 15 *Epimedium* chloroplast genome sequences from 10 species had been reported, which contributes a lot to the clarification of phylogenetic relationships and identification of the plants of the genus (Lee et al. 2016; Zhang et al. 2016; Sun et al. 2018; Guo et al. 2019). Here, we at first reported the complete chloroplast genome sequence of *E. brevicornu*, which is one of the four botanical origins of Herba Epimedii embodied in Chinese pharmacopeia (The State Pharmacopeia Committee of China 2015).

Fresh leaves of *E. brevicornu* were collected from Lixian, Gansu, China (N34°11'25.84", E105°10'29.53") with Voucher Yanjun Zhang 556 (HIB). Chloroplast DNA was then extracted according to Vieira et al. (2014) and sheared with a Covaris

LE220 (Covaris Inc., Woburn, USA). The sequencing libraries were constructed with the Mate Pair Library Preparation Kit (Illumina Inc, San Diego, CA, USA) and then were paired-end sequenced on an Illumina HiSeq 4000 platform. The raw reads each with 150 bp in length were assembled using CLC Genomics Workbench 11.0 software with 146.44× coverage and no gaps (CLC Bio, Aarhus, Denmark). Using Geneious 9.1.2 (Biomatters, Auckland, New Zealand), the contigs were furthermore assembled into complete single circular cp genome and the gene annotation of each cp genome was performed followed by manual correction, through comparison with the previously published cp genome sequences of *Epimedium* (Lee et al., 2016; Zhang et al. 2016; Sun et al. 2018; Guo et al. 2019).

The complete chloroplast genome of *E. brevicornu* (Genbank accession MN714008) was 159,572 bp in length with 38.83% GC content, including four distinct regions: Large single copy (LSC) region of 86535 bp, small single copy (SSC) region of 17641 bp, and a pair of inverted repeat (IR) regions of 27698 bp. In accordance with the previously reported cp genome sequences of *Epimedium* (Lee et al. 2016; Zhang et al. 2016; Sun et al. 2018; Guo et al. 2019), the chloroplast genome encoded 112 unique genes, which included 78 protein-coding genes, 30 tRNA genes, and 4 rRNA genes. However, *Epimedium* cp genomes exhibited obviously different at the IR/SC boundary regions. For the cp

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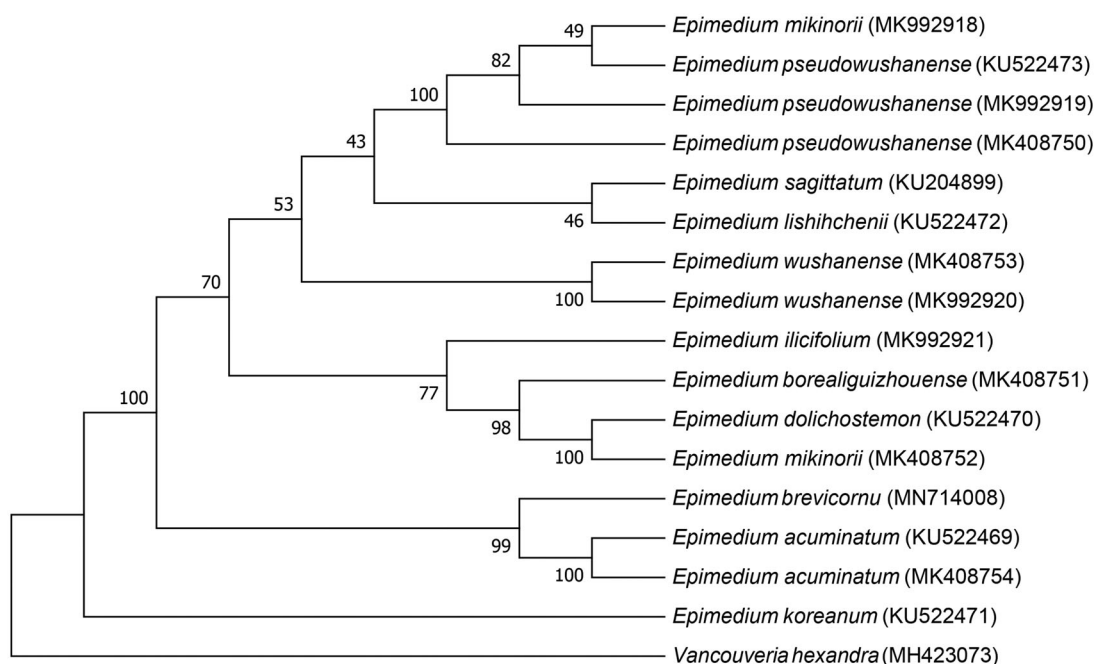


Figure 1. A phylogenetic ML tree constructed using MEGA7.0 based on the complete chloroplast sequence of 11 *Epimedium* species including *E. brevicornu* and *Vancouveria hexandra* as an outgroup.

genome of *E. brevicornu*, the gene *ycf1* crossed the SSC/IRB region, and the pseudogene fragment $\psi ycf1$ was located at the IRA region with 2237 bp, while the gene *rpl22* crossed the LSC/IRA region, and $\psi rpl22$ with 296 bp was located at IRB region.

The phylogenetic analysis was carried out using the chloroplast complete genome sequences of *E. brevicornu*, ten reported *Epimedium* species and *Vancouveria hexandra* as outgroup (Lee et al. 2016; Zhang et al. 2016; Sun et al. 2018; Guo et al. 2019) (Figure 1). The results showed that all the ten species of the sect. *Diphyllum* of *Epimedium*, endemic to China, clustered into a strongly supported branch and were separated from *E. koreanum* of sect. *Macroceras* distributed in Korea. Small-flowered *E. brevicornu* firstly clustered with large-flowered *E. acuminatum* into a strongly supported branch, not with *Epimedium* species having similar floral characters. Inconsistent with Stearn's (2002) system about *Epimedium*, the phylogenetic relationships of 11 species of sect. *Diphyllum*, based on the chloroplast genome data, were not closely related to floral characters. It needs further research to achieve natural phylogenetic relationships of *Epimedium*.

Disclosure statement

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

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