

## Complete sequence of *Cynanchum rostellatum* (Apocynaceae: Asclepiadoideae) chloroplast genome and its phylogenetic analysis

Lixin Pei<sup>a</sup>, Shengnan Shu<sup>a,c</sup>, Baoyu Ji<sup>a</sup> and Ning Cui<sup>b</sup>

<sup>a</sup>College of Pharmacy, Henan University of Chinese Medicine, Zhengzhou, China; <sup>b</sup>Central Laboratory, Shandong Academy of Chinese Medicine, Ji'nan, China; <sup>c</sup>School of Pharmacy and Chemical Engineering, Zhengzhou University of Industry Technology, Zhengzhou, China

### ABSTRACT

*Cynanchum rostellatum* (Turcz.) Liede and Khanum 2016 is a perennial herbaceous twining vine that is widely distributed in Japan, South Korea, the United States of America, and China. In this study, the complete chloroplast (cp) genome of *C. rostellatum* was sequenced using the Illumina platform and assembled for the first time. This plastome has a circular structure with a length of 160,641 bp. The GC content of the plastome was 37.82%. The cp genome contained 113 unique genes, including 79 protein-coding, 30 transfer RNA, and four ribosomal RNA genes. Phylogenetic analysis based on the complete cp genome sequences of the Asclepiadoideae subfamily showed that *C. rostellatum* was closely related to *C. bungei* in the genus *Cynanchum*. These results provide useful information for both phylogenetic research and the utilization of *C. rostellatum*.

### ARTICLE HISTORY

Received 10 May 2022  
Accepted 12 July 2022

### KEYWORDS

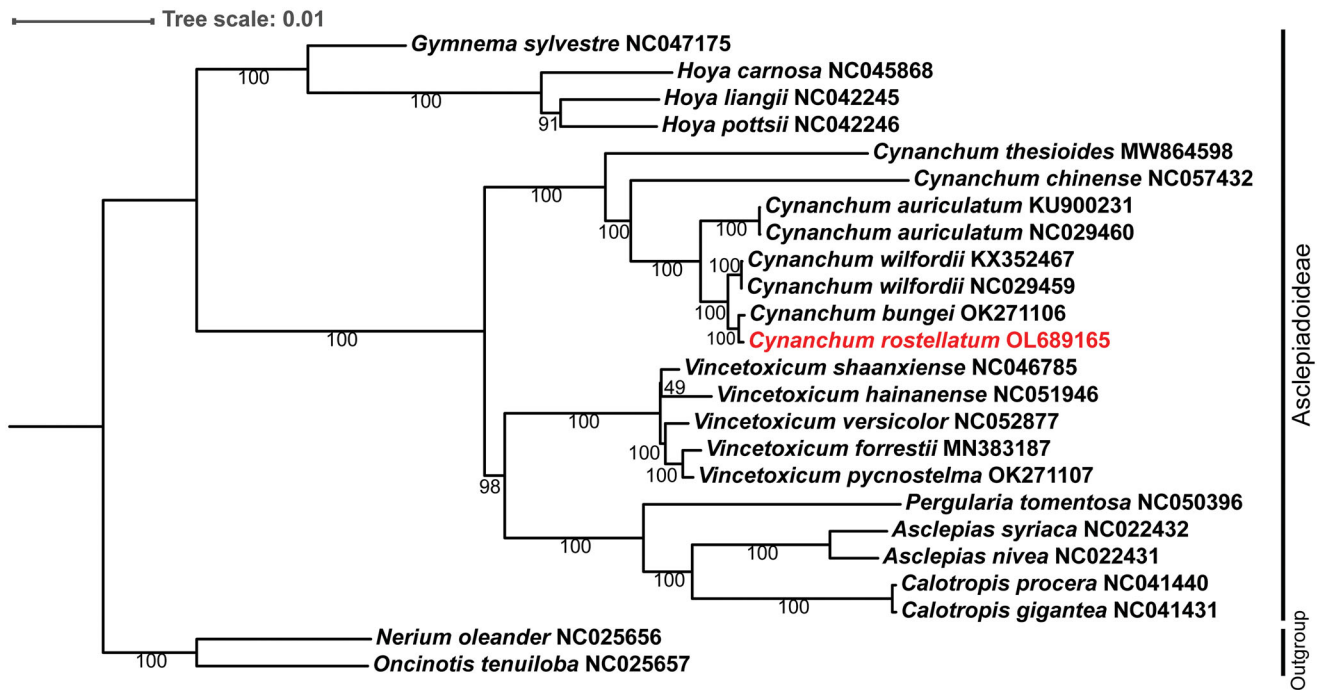
*Cynanchum rostellatum*;  
chloroplast genome;  
phylogeny

*Cynanchum rostellatum* (Turcz.) Liede and Khanum 2016 is a perennial herbaceous twining vine of the Asclepiadoideae subfamily of Apocynaceae, with the heterotypic synonym *Metaplexis japonica* in the *Flora Reipublicae Popularis Sinicae* database (<http://www.iplant.cn/>; Ma and Clemants 2006). The herb contains milky latex and is widely distributed in Japan, South Korea, the United States of America, and China (Welsh and Anderson 1962). The fiber of *C. rostellatum* has considerable industrial application prospects, and the fruit, root, stem, leaf, seed hair, and milky juice of *C. rostellatum* have a long history in traditional Chinese medicine (Yang et al. 2006; Wang et al. 2020). The chemical components of *C. rostellatum* have been shown to have a variety of pharmacological activities, such as antitumor, antioxidant, antibacterial, immunosuppressive, and neuroprotective activities (Zhang et al. 2014; Wei et al. 2019). Although *C. rostellatum* has significant economic and medicinal value, genetic and evolutionary research is extremely rare. In this study, we report the complete chloroplast (cp) genome of *C. rostellatum* and examine its phylogenetic position within the subfamily Asclepiadoideae. It is expected to lay the foundation for further molecular studies and the utilization of this species.

Fresh leaves of *C. rostellatum* were collected from Xixia County, Nanyang City, Henan Province (33°38'N, 111°43'E). The specimen and DNA were deposited at the Herbarium of Henan University of Traditional Chinese Medicine, Henan, China (contact person: Lixin Pei, [xlpxlp@aliyun.com](mailto:xlpxlp@aliyun.com)), under

voucher number HNPS2020-12-032. Total genomic DNA was extracted using a Dneasy Plant Mini Kit (Qiagen, CA, USA) according to the manufacturer's instructions (Cui et al. 2020). The genomic library for 150 bp paired-end sequencing was produced using the Illumina HiSeq 1500 platform (Illumina Inc., USA). The cp genome of *C. auriculatum* (NC029460) was used as a reference for assembly and annotation. The complete chloroplast genome was assembled using GetOrganelle (v. 1.1.1; Jin et al. 2020), and annotated by Plann (Huang and Cronk 2015). The novel cp genome was submitted to the NCBI database ([www.ncbi.nlm.nih.gov](http://www.ncbi.nlm.nih.gov)) under the GenBank accession number OL689165.

The cp genome of *C. rostellatum* contained circular double-stranded DNA and displayed a typical quadripartite structure, including two inverted repeat (IR) regions with lengths of 23,841 bp and 23,836 bp, separated by a large single-copy (LSC) region of 92,051 bp and a small single copy (SSC) region of 20,913 bp. The GC content of the plastome was 37.82%. The genome contained a set of 132 genes, of which 113 were unique and 19 were duplicated. A total of 87 protein-coding genes (79 unique genes) were annotated and were mainly involved in processes related to photosynthesis and gene expression. Eleven protein-coding genes (*rps16*, *atpF*, *rpoC1*, *petB*, *petD*, *rpl16*, two *rpl2* genes, two *ndhB* genes, and *ndhA*) and eight tRNA genes (*trnK-UUU*, *trnG-UCC*, *trnL-UAA*, *trnV-UAC*, *trnI-GAU*, *trnA-UGC*, *trnA-UGC*, and *trnI-*



**Figure 1.** Phylogenetic tree constructed by using the whole chloroplast genome sequences of Asclepiadoideae subfamily with the maximum likelihood method. Numbers near each branch are the bootstrap values.

*GAU*) contained one intron, and two genes (*clpP* and *ycf3*) contained two introns.

To elucidate the evolutionary relationship of *C. rostellatum*, the cp genomes of 21 Asclepiadoideae species were downloaded from the NCBI GenBank database. We aligned the plastomes using MAFFT and constructed a maximum likelihood (ML) tree (Figure 1) using RAxML (v.8.2.9), using the GTRGAMMA model with 1000 rapid bootstrap replicates (Cui et al. 2020). Our plastome phylogeny showed that *C. rostellatum* is most closely related to *C. bungei* in the genus *Cynanchum*. *Cynanchum rostellatum* and seven *Cynanchum* species were clustered into one branch indicating that *C. rostellatum* was likely placed in the *Cynanchum* genus, and not in *Metaplexis*, during taxonomic classification. In conclusion, the cp genome of *C. rostellatum* provides a theoretical basis for a better understanding of the evolutionary patterns and for improving its taxonomic classification.

## Ethical approval

No permission was necessary in this study for the sample collection. *Smilax moranensis* is widely distributed in North China and is not listed as a national key protected plant.

## Authors' contributions

N. Cui designed and conceived this work; L.X. Pei collected the samples and carried out the experiment; B.Y. Ji and S.N. Shu analyzed the data and wrote the first version of the manuscript; All authors read, revised, and approved the final manuscript.

## Disclosure statement

The authors declare that there is no potential conflict of interest exists.

## Funding

This work was supported by the Shandong Province Traditional Chinese Medicine Science and Technology development Program [2019-0650], Research Incubation Fund Project of Shandong Academy of Chinese Medicine [2021SACM-3], Medical and Health Science and Technology Development Project of Shandong Province [202102041135], "National Survey of Traditional Chinese Medicine Resources", a special subsidy program for United States Public Health Service of traditional Chinese medicine in 2017 [caishe[2017] No. 66] and "National Survey of Traditional Chinese Medicine Resources", a special subsidy program for United States Public Health Service of traditional Chinese medicine in 2018 [caishe[2017] No.183].

## Data availability statement

The complete chloroplast genome of *Cynanchum rostellatum* assembled here is available in the GenBank of NCBI (<https://www.ncbi.nlm.nih.gov/genbank>, accession no. OL689165). The associated BioProject, BioSample and SRA numbers are PRJNA785091, SAMN23527168 and SRR17082022, respectively.

## References

- Cui N, Liao BS, Liang CL, Li SF, Zhang H, Xu J, Li XW, Chen SL. 2020. Complete chloroplast genome of *Salvia plebeia*: organization, specific barcode and phylogenetic analysis. *Chin J Nat Med.* 18(8): 563–572.
- Huang DI, Cronk QC. 2015. Plann: a command-line application for annotating plastome sequences. *Appl Plant Sci.* 3(8):1500026. apps.1500026.

- Jin JJ, Yu WB, Yang JB, Song Y, dePamphilis CW, Yi TS, Li DZ. 2020. GetOrganelle: a fast and versatile toolkit for accurate de novo assembly of organelle genomes. *Genome Biol.* 21(1):241.
- Ma J, Clemants S. 2006. A history and overview of the Flora Reipublicae Popularis Sinicae (FRPS, Flora of China, Chinese edition, 1959-2004). *Taxon.* 55(2):451–460.
- Wang Z, Wang D, Li Z, Wang Y. 2020. *Metaplexis japonica* seed hair fiber: a hydrophobic natural fiber with robust oil-water separation properties. *Cellulose.* 27(5):2427–2435.
- Wei L, Yang M, Huang L, Lin Li J. 2019. Antibacterial and antioxidant flavonoid derivatives from the fruits of *Metaplexis japonica*. *Food Chem.* 289:308–312.
- Welsh SL, Anderson DE. 1962. *Metaplexis japonica*: an oriental milkweed from an Iowa cornfield. *Brittonia.* 14(2):186–188.
- Yang M, Wu J, Xu X, Jin Y, Guo Y, Chen J. 2006. A new lignan from the Jian-er syrup and its content determination by RP-HPLC. *J Pharm Biomed Anal.* 41(2):662–666.
- Zhang J, Chen J, Liang Z, Zhao C. 2014. New lignans and their biological activities. *Chem Biodivers.* 11(1):1–54.