

The complete chloroplast genome sequence of traditional Chinese medicine *Uncaria macrophylla* (Rubiaceae)

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ABSTRACT

Uncaria macrophylla (Rubiaceae) is a medicinal vine plant of the Rubiaceae family that was distributed in East Asia and Southeast Asia. The first complete chloroplast genome of *Uncaria macrophylla* was sequenced and assembled in this study. The genome is 155,138 bp in length and contained 129 encoded genes in total, including 79 protein-coding genes, eight ribosomal RNA genes, and 37 transfer RNA genes. The phylogenomic analysis showed that *U. macrophylla* was closely related to *Uncaria rhynchophylla* according to the current sampling extent.

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Uncaria macrophylla Wall. 1824 (Rubiaceae) is a medicinal herb with double or single hooks in leaf axils. It is mainly distributed in tropical areas, concentrated in Malaysia to Solomon Islands in Southeast Asia, other parts of Asia and Africa, South America also have distribution (Ndagijimana et al. 2013). As antihypertensive, antioxidation, antipyretic, and anticonvulsant, it is used in the treatment of headache, vertigo, and epilepsy, and also has a certain effect on Alzheimer's disease (Fujiwara et al. 2006). Previous studies have shown that alkaloids in *Uncaria*, such as rhynchophylline and isorhynchophylline, can be developed as chemical drugs for the treatment of human hypertension (Wang et al. 2019). In this study, the first complete chloroplast (cp) genome of *U. macrophylla* was reported. To provide genomic resources for investigating the evolution of *U. macrophylla*, the complete cp genome of this species was analyzed from high-throughput Illumina sequencing reads. It will provide a better understanding on the evolution and genetics of Rubiaceae.

The leaves of *U. macrophylla* were collected from Nanning, Guangxi, China (GPS: E108°30'3.22", N22°47'53.21"). The specimen was deposited at Zhejiang Province Key Laboratory of Plant Secondary Metabolism and Regulation (<http://sky.zstu.edu.cn/>), in Zhejiang Sci-tech University, under the voucher number ZSTU09554 (Miao Zhang: Zhwater1063@163.com). Permission was granted by Guangxi University of Chinese Medicine to carry out research on the species and there was no endangered or protected species was involved in this study. The plant was identified as *U. macrophylla* by Professor Zhe-chen Qi of Zhejiang Sci-tech University.

The total genomic DNA was extracted from its silica dried leaves using DNA Plantzol Reagent (Invitrogen, Carlsbad, CA) in accordance with the manufacturer's instructions. The plasmid sequences were generated using the Illumina HiSeq 2500 platform (Illumina Inc., San Diego, CA). In total, ca. 18.3 million high-quality clean reads (150 bp PE read length) were generated with adaptors trimmed. Aligning, assembly, and annotation were conducted by GetOrganelle v1.7.0c, BLAST, GeSeq (Michael et al. 2017) and GENEIOUS v11.0.5 (Biomatters Ltd., Auckland, New Zealand).

The full length of *U. macrophylla* cp sequence (GenBank accession no. OK534535) is 155,138 bp, consisting of a large single-copy (LSC with 85,750 bp) region, a small single-copy (SSC with 18,157 bp) region, and two inverted repeat regions (IR with 25,615 bp). The GC content of *U. macrophylla* cp genome was 37.5%. A total of 129 genes were contained in the genome (79 protein-coding genes, eight rRNA genes, and 37 tRNA genes). Seventeen genes had two copies, which were comprised of protein-coding genes (*ndhB*, *rpl2*, *rpl23*, *rps7*, and *ycf2*), seven tRNA genes (*trnA-UGC*, *trnI-CAU*, *trnI-GAU*, *trnL-CAA*, *trnN-GUU*, *trnR-ACG*, and *trnV-GAC*), and all four rRNA species (*rrn16*, *rrn23*, *rrn4.5*, and *rrn5*). In the genome, seven protein-coding genes (*ndhB*, *rpl2*, *rps16*, *atpF*, *rpoC1*, *rpl2*, and *ndhA*) had one intron, and two genes (*clpP*, *ycf3*) contained two introns.

To confirm the phylogenetic position of *Uncaria macrophylla*, we obtained 10 published cp genomes of Rubiaceae from NCBI, including *Galium aparine* (KY562587), *Paederia scandens* (MN567112), *Coffea arabica* (MK875244), *Coffea canephora* (NC030053), *Gardenia jasminoides* (NC057593),

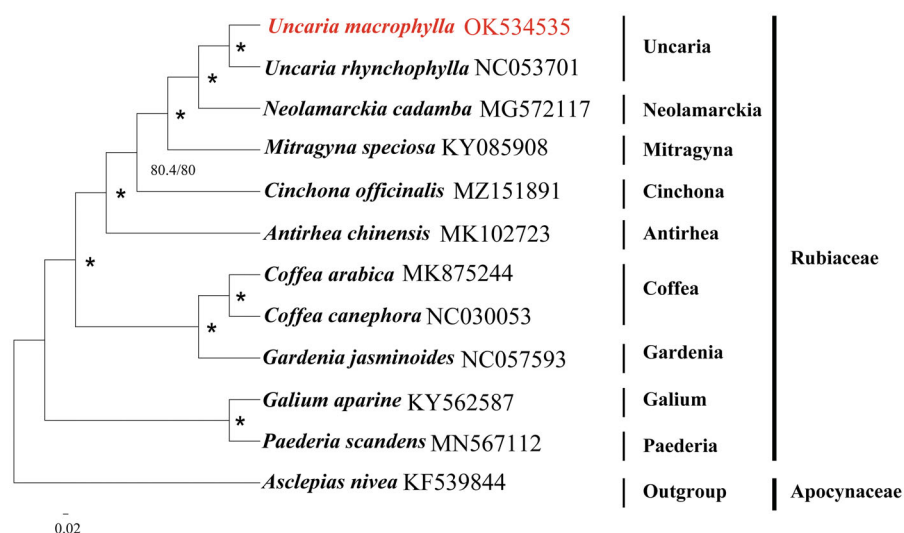


Figure 1. Phylogeny of *U. macrophylla* and other Rubiaceae based on complete chloroplast genomes. Accession numbers were listed behind each taxon. Statistical support values are shown on nodes. The ‘*’ approval rating for delegates is 100%.

Antirhea chinensis (MK102723), *Cinchona officinalis* (MZ151891), *Mitragyna speciosa* (KY085908), *Neolamarckia cadamba* (MG572117), and *Uncaria rhynchophylla* (NC053701). *Asclepias nivea* from the family of Gentianales was used as outgroups. The sequence alignment was conducted using MAFFT v7.3 (Kato and Standley 2013). The maximum-likelihood (ML) phylogenetic analyses were constructed using IQTREE v1.6.7 (Nguyen et al. 2015), with the best selected TVM + F + R3 model and 5000 bootstrap replicates. The phylogenetic tree revealed that *U. macrophylla* was closely related to *Uncaria rhynchophylla* (Miq.) Jacks (Rubiaceae) according to the current sampling extent (Figure 1).

Acknowledgements

Ethical approval: The collection of plant material was conducted according to the guidelines provided by Guangxi University of Chinese Medicine and Zhejiang Sci-Tech University. Studies on the species comply with Guangxi province legislation and Zhejiang province legislation.

Authors contributions

Miao Zhang and Zong-Suo Liang conceived and designed this study. Min-Min Chen and Miao Zhang conducted analysis. Xue-Min Zhang and Shao-Ning Chen contributed the analytical methods. Miao Zhang wrote the manuscript. Shao-Ning Chen revised the manuscript. All authors have read and agreed to the published version of the manuscript.

Disclosure statement

No potential conflict of interest was reported by the author(s).

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Data availability statement

The genome sequence data that support the findings of this study are openly available in GenBank of NCBI (<https://www.ncbi.nlm.nih.gov>) under the accession no. OK534535. The associated BioProject, SRA, and Bio-Sample numbers are PRJNA769592, SRR16267456, and SAMN22147662, respectively.

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