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PLASTOME ANNOUNCEMENT

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The complete chloroplast genome sequence of *Ardisia crispa* Thunb.

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ABSTRACT

Ardisia crispa (Thunb.) A. DC. belongs to the genus *Ardisia* (Myrsinaceae). It is a traditional medicinal plant widely used to treat inflammatory-related diseases in southern China. Here, we provide the complete chloroplast genome of *A. crispa* from Laibin, Guangxi, PR China using Illumina high-throughput sequencing approach. The total length of the chloroplast genome is 156,709 bp, including a large single-copy (LSC) region, a small single-copy (SSC) region, and a pair of inverted repeats IRa and IRb regions which are separated by the LSC and SSC, with lengths of 86,301 bp, 18,411 bp, and 25,999 bp, respectively. In general, 132 genes were identified, including 93 protein-coding genes, 31 transfer RNA (tRNA) genes, and eight ribosomal RNA (rRNA) genes. The overall GC content is 47.82%. Phylogenetic analysis revealed that *A. crispa* is close to congeneric species *A. mamillata*.

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Ardisia crispa (Thunberg) A. de Candolle, Trans. Linn. Soc. London, Bot. 17:124.1834 belongs to the genus *Ardisia* (Myrsinaceae). The main bioactive component of the genus *Ardisia* is triterpenoid saponins, and it is rich in quinines (Zheng et al. 2008; Liu et al. 2016; Blin et al. 2021). The root of *A. crispa* has primarily been used as traditional medicines for a long time by Asians (Yin et al. 2022). For instance, it is widely used in Malaysia to treat diseases such as ever, pain, swelling, rheumatism, and blood circulation (Blin et al. 2021). As a kind of Chinese Traditional Medicine as well, the root of *A. crispa* is known as 'Bai-Liang-Jin' in China. Additionally, the modern pharmacological studies revealed that *A. crispa* have notable pharmacological activities in antipyretic, antiangiogenic, antitumor, and anti-inflammation (Jansakul et al. 1987; Sulaiman et al. 2012; Nordin et al. 2018; Wen Jun et al. 2019; Blin et al. 2021). The triterpenoid saponins inside of the *A. crispa* root may be the main components that exert cytotoxicity in the biological activities of anti-tumor (Yin et al. 2022). In Guangxi province of China, *A. crispa* is also called 'Zhu Ye Feng', which is a kind of classical Yao Medicine (Hou et al. 2014). Although it has been used as an herb for many years, genetic information of *A. crispa* is still lacking. In this study, the whole chloroplast genome of *A. crispa* was reported for the first time.

Five pieces of the mature fresh *A. crispa* leaves were collected from Jinxiu Yao Autonomous County, Guangxi Province, China (N: 24°09′4.32″, E: 110°12′53.4312″). A specimen was stored at the School of Food and Biochemical Engineering of Guangxi Science & Technology Normal University (https://www.gxstnu.edu.cn/, the contact person is Song Guo and the email is guosong0804@163.com) under the voucher number ZYF202006. Total genomic DNA was extracted from approximately 30 g of leaves of *A. crispa* by using a DNeasy Plant Mini Kit (QIAGEN, Hilden, Germany) following the manufacturer's instructions. Micro spectrophotometer was used to assess the quantity and quality of the extracted DNA and agarose gel electrophoresis was used to assess the integrity of the DNA. High quality DNA was sent to Biomarker Technologies (Beijing, China) for genomic library construction and sequencing using the Illumina HiSeq platform (Illumina, San Diego, CA). About 4.5 Gb high quality, 2×150 bp pair-end raw reads were obtained and were used to assemble the complete chloroplast genome of *A. crispa* (Chen et al. 2022). The chloroplast genome was assembled using the program NOVOPlasty v4.3.1 (Dierckxsens et al. 2017), with the complete chloroplast genome of *C. formosana* chloroplast genome (GenBank accession number: MW252167.1) as the reference.

Comparing with the chloroplast genomes of species with close phylogenetic relationships, Geneious v 11.1.5 (Biomatters Ltd., Auckland, New Zealand) was mainly used to annotate the chloroplast genomes. Then the annotation result was confirmed and modified by CPGAVAS online tool (Zuo et al. 2017; Wang et al. 2018). An accession number (MW829277) was gotten from GenBank after the annotated genomic sequence was registered. The chloroplast genome of *A. crispa*, which is a circular DNA molecule, is 156,709 bp in length. Its whole chloroplast genome consists of a large single-copy (LSC, 86,301) region, a small single-copy (SSC, 18,411 bp) region, and two inverted repeat regions (IRa and IRb, 25,999 bp). Overall GC content of *A. crispa* chloroplast genome is 47.82%, with corresponding GC values of LSC,

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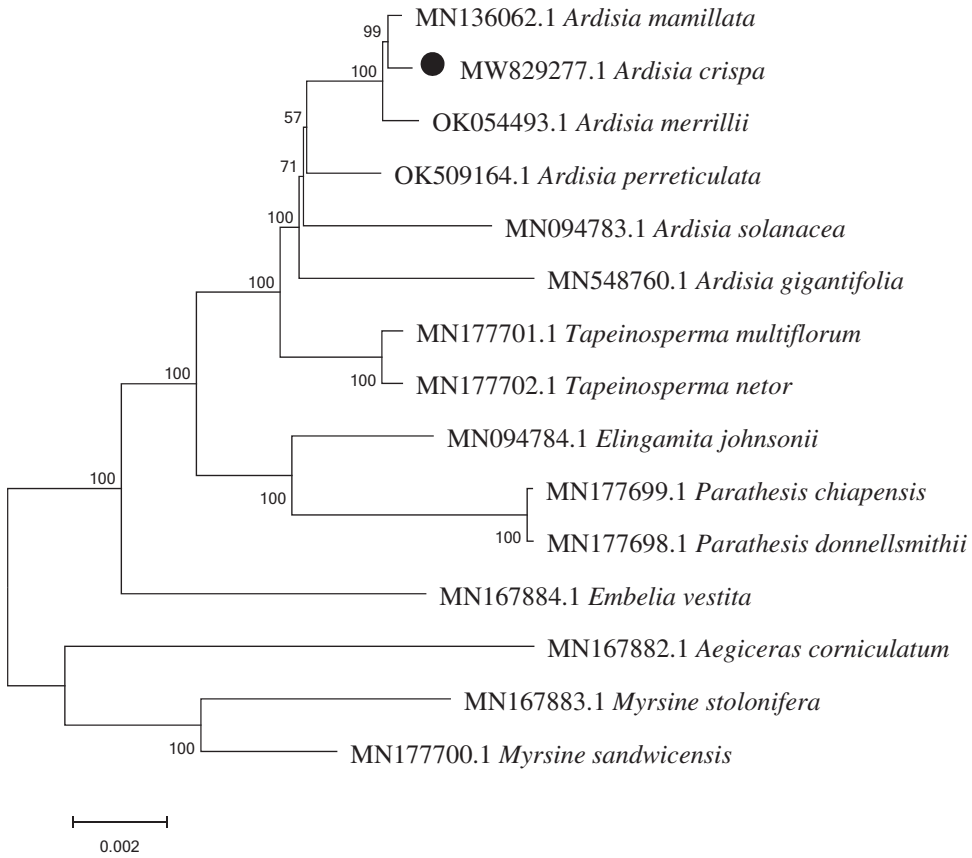

Figure 1. Phylogenetic position of *Ardisia crispa* resolved by maximum-likelihood method (bootstrap values were calculated out of 1000 replicates) based on the complete chloroplast genome. The bootstrap values are listed on nodes.

SSC, and IR regions as 34.97%, 30.22%, and 42.98%, respectively. In total, 196 genes were identified, while 132 genes were unique genes. Among the unique genes, there are 93 protein-coding genes, eight ribosomal RNA (rRNA) genes, and 31 transfer RNA (tRNA) genes.

Phylogenetic analysis was carried out using chloroplast sequences of *A. crispa* and other 15 species within family (Figure 1). The sequences were aligned using the default settings in MAFFT v7 (Katoh et al. 2019). The phylogenetic analyses were conducted using MEGA6 software (Koichiro et al. 2013), and were analyzed with using maximum-likelihood (ML) method (bootstrap values were calculated out of 1000 replicates) (Hu et al. 2019). The sisterhood of *Ardisia crispa* with genus *Ardisia* has been confirmed in this study and *A. crispa* has a relatively close phylogenetic relationship with congeneric species *A. mamillata*. Our data may be useful for further research in *Ardisia* phylogeny and the molecular identification of this promising medicinal plant.

Author contributions

Song Guo and Wuwei Wu involved in the conception, design, and financial support; Zeyang Li and Song Guo collected the sample; Yu Liu and Song Guo analyzed the data. Wuwei Wu was involved in the drafting of the paper, and Song Guo revised the manuscript and final approval of the version to be published. All authors agreed to be accountable for all aspects of the work.

Ethics statement

Ethical approval for the study was obtained from the Ethical Committee of Guangxi Science and Technology Normal University. The collection of specimens conformed to the requirement of International Ethics, which did not cause damage to the local environment. The process and purpose of this experimental research were in line with the rules and regulations of our institute. There are no ethical issues and other conflicts of interest in this study.

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 at https://www.ncbi.nlm.nih.gov under the accession number MW829277. The associated BioProject, SRA, and Bio-Sample numbers are PRJNA715070, SRR13985456, and SAMN18325375, respectively.

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