

Chloroplast Genome Analysis of The Famous Chinese Herbal Medicine *Yizhiren* (Zingiberaceae)

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Research

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Abstract

Background: *Yizhiren* is the fruit of *Alpinia oxyphylla* (Zingiberaceae), a well-known Chinese herbal medicine from China. The complete chloroplast genome of *Alpinia oxyphylla* was studied in this paper, which laid the foundation of the further study of genetic information and data of *Alpinia oxyphylla*.

Methods: The complete chloroplast sequences of 19 the family Zingiberaceae species were aligned using MEGAX software. The phylogenetic tree was constructed by the Maximum-Likelihood method and edited by the Evolview online.

Results: The chloroplast gene group is a typical tetragonal structure, which is formed by 161,351 base pairs. Each genome has a large single-copy region (LSC) of 87,248 bp, a small single-copy region (SSC) of 16,175 bp and a pair of inverted-repeat regions (IRs) of 28,964 bp in each. The complete nucleotide composition of chloroplast genome is: 31.5% A, 32.4% T, 18.2% C, 17.9% G, and the total GC content is 36.2%. Among them 28 exons and 15 introns. A total of 137 genes were annotated, which included 92 protein coding genes (PCGs), 37 metastatic RNA (tRNAs) and 8 ribosomal RNA (rRNAs).

Conclusions: The phylogenetic ML tree shown the conclusion that *Alpinia oxyphylla* is closely related to *Alpinia chinensis* on genetic position relationship. This result is of great value to the study of biological inheritance, species identification and medicinal value. Meanwhile, it provides references for the study of biological inheritance, species identification and medicinal value.

Background

The fruit of *Alpinia oxyphylla* is the mature fruitage of *Alpinia oxyphylla* (Zingiberaceae) that is distributed in Guangdong, Hainan, Fujian, Guangxi, Yunnan and other provinces of China. As a Chinese medicinal material of "One Root of Medicine and Food", it has been widely used for the clinical of TCM. Over the years, *Alpinia oxyphylla* is collected for the production of Chinese patent medicine and food, which causes the reduction of wild resources. The fruit of *Alpinia oxyphylla* contains flavonoid, sesquiterpenes, monoterpenes, diterpenes, diarylheptanoids and so forth (Wei et al. 2018) and 1%-2% of volatile oil that contains eucalyptol, zingiberene and zinginerol (Feng et al. 2018). Its peel is rich in B vitamins and vitamin C. In addition, it has micronutrients such as manganese, zinc, potassium, sodium, calcium, magnesium, phosphorus, iron, copper and so on. These ingredients provide *Alpinia oxyphylla* the pharmacological effects such as anti-microbial (Wang et al. 2018), anti-diabetic (Xie et al. 2018), anti-angiogenic (He et al. 2018), nerve production (Duan et al. 2018) and so on. Over the years, the wild resources of *Alpinia oxyphylla* have been gradually reduced on account of the widely use in the production of Chinese patent drug and food. In this paper, the chloroplast genome of *A. oxyphylla* was studied to provide references for the research of the superior cultivation and medical development of *Alpinia oxyphylla* and other family Zingiberaceae species in the future.

Methods

Alpinia oxyphylla was collected from the herb market located at Qiaocheng, Bozhou, Anhui, China (33.50N, 115.47E). Total genomic DNA was extracted from fresh stems of *Alpinia oxyphylla* with the application of the Plant Tissues Genomic DNA Extraction Kit (TIANGEN, BJ and CN). The chloroplast genome DNA was kept in storage in Zhejiang Chinese Medical University (No. SCMC-ZJU-TCM-08) .

After the chloroplast genome of *Alpinia oxyphylla* was purified and sequenced by the sequencer, the collected raw sequences were quality controlled and removed by the FastQC (Andrews 2015). The chloroplast genome of *Alpinia oxyphylla* was assembled and annotated by MitoZ (Meng et al. 2019). Then the chloroplast genome map was generated by the OrganellarGenomeDRAW (Lohse et al. 2013).

Results

The chloroplast genome of *Alpinia oxyphylla* (KY985237) has a length of 161,351 base pairs (bp) and has a typical quadripartite structure. It is consisted of a large single-copy region (LSC of 87,248 bp), a small single-copy region (SSC of 16,175 bp) and a pair of inverted repeat regions (IRs of 28,964 bp in each). The overall nucleotide composition of chloroplast genome has 31.5% of A, 32.4% of T, 18.2% of C, 17.9% of G and the total content of GC is 36.2%, and there are 28 exons and 15 introns among these. The chloroplast genome of *Alpinia oxyphylla* contains 137 genes, which includes 92 protein-coding genes (PCGs), 37 transfer RNA genes (tRNAs) and 8 ribosomal RNA genes (rRNAs). Twenty-one duplicated genes were found in each IR region, which includes 9 PCGs species (*ndhB*, *orf79*, *orf1006*, *rpl2*, *rpl23*, *rps7*, *rps19*, *ycf2*, *ycf68*), 8 tRNAs species (*trnH-GTG*, *trnM-CAT*, *trnL-CAA*, *trnV-GAC*, *trnI-GAT*, *trnA-TGC*, *trnA-ACG* and *trnA-GTT*) and 4 rRNAs species (*rRNA16*, *rRNA23*, *rRNA4.5* and *rRNA5*). The large single-copy region contains 84 genes, 64 protein-coding genes, 13 exons and 8 introns. The small single-copy region contains 14 genes, 13 protein-coding genes, 2 exons and 1 intron.

Discussion

In order to confirm the phylogenetic position of *Alpinia oxyphylla*, the complete chloroplast sequences of 19 species of the family Zingiberaceae were aligned with the application of MEGA X software (Kumar et al. 2018) and the phylogenetic tree was constructed by the Maximum-Likelihood (ML) method. The phylogenetic tree was analyzed by ML method with TVM + F + R2 model, and strong support from all of the nodes were received by duplicating and inferring the 2,000 bootstrap values of each node. The phylogenetic tree was generated by Mega X software and edited by the Evolview online (www.evolgenius.info/evolview) (Subramanian et al. 2019). The phylogenetic ML tree shown the conclusion that *Alpinia oxyphylla* is closely related to *Alpinia chinensis* (NC_050165.1) on genetic position relationship (Fig. 1).

Conclusions

This result not only is of great value to the research on metabolism, biosynthesis and regulation of *Alpinia oxyphylla*, but also provides references for the study of biological inheritance, species identification and

medicinal value.

Declarations

Ethics approval and consent to participate

Not applicable

Consent for publication

Not applicable

Availability of data and materials

Not applicable

Competing interests

The authors declare no potential conflict of interest.

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Authors' contributions

Pan J.Y. and Sun S.Y. performed the experiments, analyzed the data, and wrote the manuscript. He F.L. conceived and designed this study, initiated and supervised the study, Xia Q. and Lv X.H. wrote and revised the manuscript, Yang X.X. prepared figures 1. All authors reviewed the manuscript and given final approval of the version to be published. All authors agreed to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

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References

1. Wei N, Wang Y, Li HF, Zhang JQ, Li YH. Chemical Constituents of the Rhizomes of *Alpinia oxyphylla*. *Chem Nat Compd*. 2013;49(5):934–5.
2. Feng HH, Luo JY, Kong WJ, Dou XW, Wang YT, Zhao XS, Zhang WP, Li Q, Yang MH. Enhancement effect of essential oils from the fruits and leaves of *Alpinia oxyphylla* on skin permeation and deposition of indomethacin. *RSC Advances*. 2015;5(49):38910–7.

3. Dan W, Wen XC, Yue YH, Mei FP. Optimization, Modeling and Antibacterial Activity of Extraction from *Fructus Alpinia oxyphylla*. *Adv Mater Res*. 2013;2606:827–31.
4. Xie YQ, Xiao M, Li D, Liu HQ, Yun FL, Wei Y, Sang SG, Du G, K. Anti-diabetic effect of *Alpinia oxyphylla* extract on 57BL/KsJ db-/db- mice. *Experimental Therapeutic Medicine*. 2017;13(4):1321–8.
5. He ZH, Ge W, Yue GG, Lau CB, He MF, But PP. Anti-angiogenic effects of the fruit of *Alpinia oxyphylla*. *Journal of Ethnopharmacology*. 2010;132(2):443–9.
6. Duan LH, Li M, Wang CB, Wang QM, Liu QQ, Shang WF, Shen YJ, Lin ZH, Sun TY, Wu ZZ, Li YH, Wang YL, Luo X. Protective effects of organic extracts of *Alpinia oxyphylla* against hydrogen peroxide-induced cytotoxicity in PC12 cells. *Neural Regeneration Research*. 2020;15(4):682–9.
7. Andrews S. (2015). FastQC: a quality control tool for high throughput sequence data. Available online at: <http://www.bioinformatics.babraham.ac.uk/projects/fastqc/>.
8. Meng GL, Li YY, Yang CT, Liu SL. MitoZ: a toolkit for animal mitochondrial genome assembly, annotation and visualization. *Nucleic Acids Res*. 2019;47(11):e63.
9. Lohse M, Drechsel O, Kahlau S, Bock R. OrganellarGenomeDRAW—a suite of tools for generating physical maps of plastid and mitochondrial genomes and visualizing expression data sets. *Nucleic Acids Res*. 2013;41(W1):W575–81.
10. Kumar S, Stecher G, Li M, Knyaz C, Tamura K. MEGA X: molecular evolutionary genetics analysis across computing platforms. *Molecular Biology Evolution*. 2018;35(6):1547–9.
11. Subramanian B, Gao S, Lercher MJ, Hu S, Chen W-H. Evolview v3: a webserver for visualization, annotation, and management of phylogenetic trees. *Nucleic Acids Res*. 2019;47(W1):W270–5.

Figures

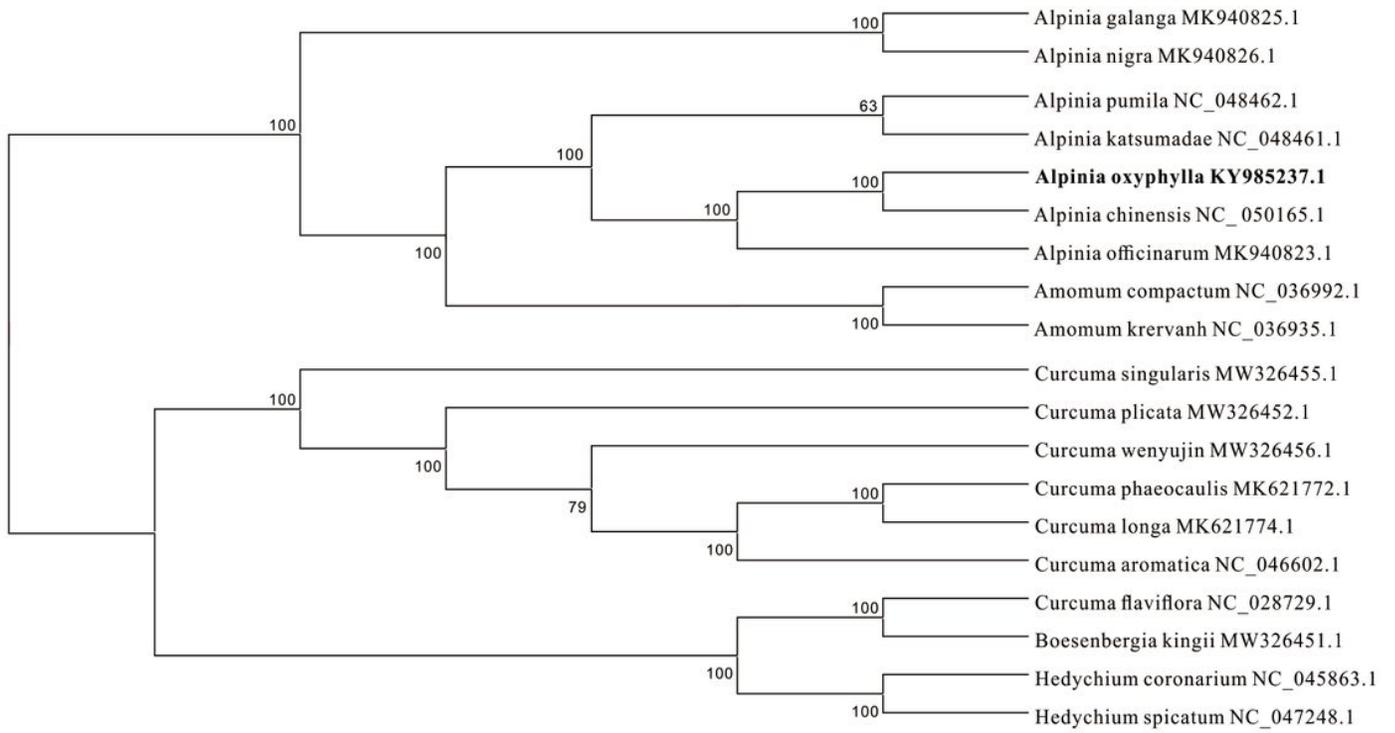


Figure 1

the phylogenetic ML tree of *Alpinia oxyphylla* based on complete chloroplast genomes from 19 species of Zingiberaceae and each node is the bootstrap value from 2000 duplicates.