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Protocols for "The genome analysis of Tripterygium wilfordii reveals TwCYP712K1 and TwCYP712K2 responsible for oxidation of friedelin in celastrol biosynthesis pathway"



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Tianlin Pei[1,2], Mengxiao Yan[1], Yu Kong[1], Hang Fan[1,2], Jie Liu[1], Mengying Cui[1], Yumin Fang[1], Binjie Ge[1], Jun Yang[1,2], Qing Zhao[1,2]

[1]Shanghai Key Laboratory of Plant Functional Genomics and Resources, Shanghai Chenshan Botanical Garden, Shanghai Chenshan Plant Science Research Center, Chinese Academy of Sciences, Shanghai, China;
[2]State Key Laboratory of Plant Molecular Genetics, CAS Center for Excellence in Molecular Plant Sciences, Shanghai Institute of Plant Physiology and Ecology, Chinese Academy of Sciences, Shanghai, China

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Tianlin Pei
Shanghai Chenshan Botanical Garden

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We use this protocol and it's working

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Abstract

Tripterygium wilfordii is a vine from the Celastraceae family that is used in traditional Chinese medicine (TCM). The active ingredient celastrol is a friedelane-type pentacyclic triterpenoid with putative roles as antitumor, immunosuppressive, and antiobesity agent. Here, we report a reference genome assembly of *T. wilfordii* with high-quality annotation by using a hybrid sequencing strategy, obtaining a 340.12 Mb total genome size, a contig N50 reaching 3.09 Mb. We successfully anchored 91.02% of the sequences into 23 pseudochromosomes using Hi-C technology and the superscaffold N50 reached 13.03 Mb. In addition, we annotated 31,593 structural genes, and a repeat percentage of 44.31%. Comparative evolutionary analyses showed that *T. wilfordii* diverged from species of Malpighiales approximately 102.4 million years ago. Based on integration of the genome, transcriptome and metabolite analyses, as well as *in vivo* and *in vitro* enzyme assays of the two CYP450 genes *TwCYP712K1* and *TwCYP712K2* the second biosynthesis step of celastrol was investigated. Syntenic analysis revealed that *TwCYP712K*1 and *TwCYP712K2* were derived from a common ancestor. These results provide insights for further investigating pathways related to celastrol andvaluable information to aid the conservation of resources. In addition, they helped us understand the evolution of Celastrales.

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