

# Multi-omics Techniques Analysis of Ganfule Capules in Improving Nonalcoholic Fatty Liver Disease

Yu Pan<sup>1</sup>, Yu Liu<sup>1</sup>, Danna Huang<sup>1</sup>, Fei Xu<sup>2</sup>, Dan Huang<sup>3</sup>, Lu Chen<sup>1,\*</sup>

<sup>1</sup>National Engineering Research Center of Southwest Endangered Medicinal Resources Development, Guangxi Botanical Garden of Medicinal Plants, Nanning, China

<sup>2</sup>Hunan Engineering Research Center of Bioactive Substance Discovery of Chinese Medicine, Hunan University of Chinese Medicine, Changsha, China

<sup>3</sup>State Key Laboratory of Chinese Medicine Powder and Medicine Innovation in Hunan (Incubation), Science and Technology Innovation Center, Hunan University of Chinese Medicine, Changsha, China

## Email address:

panyu1226@126.com (Yu Pan), 52888147@qq.com (Yu Liu), dhuang202@126.com (Danna Huang), 393077215@qq.com (Fei Xu), huangdan110@hnuocm.edu.cn (Dan Huang), chenlulu982@hotmail.com (Lu Chen)

\*Corresponding author

## Abstract

*(Background)* Compound Chinese medicines exhibit holistic regulatory advantages with multiple targets and pathways in treating nonalcoholic fatty liver disease (NAFLD). This study aims to clarify the key components and mechanisms of Ganfule (GFL) in treating NAFLD using multi-omics techniques. *(Methods)* Firstly, untargeted metabolomics (LC-MS) was used to identify differential metabolites in the feces of NAFLD rats treated with GFL and the metabolic pathways involved. Subsequently, high-throughput sequencing (HTS) combined with bioinformatics techniques analyzed differentially expressed genes (DEGs) in the livers of NAFLD rats. Immunobiochemical techniques such as HE, IHC, RealTime-qPCR, and Western Blot were employed to validate morphological changes in the liver and variations in key DEGs. Finally, network pharmacology was applied to dissect the key herb combinations, active ingredients, and core targets of GFL in treating NALFD. *(Results)* The GFL treatment group significantly improved lipid levels and pathological morphology in the livers of NAFLD rats. Seven herbs, including rhubarb, bupleurum, capillaris, poria, codonopsis, astragalus, and eaglewood, may constitute the core effective formula. We identified 87 differential metabolites, 9 core DEGs, 12 chemical components, and 16 core targets. GFL can also regulate the expression of key proteins such as PPARA, PPARD, PRKACA, and PIK3CG, involving seven pathways including lipid metabolism, immune regulation, inflammation inhibition, JAK-STAT, leptin-insulin signaling, and apoptosis. *(Conclusions)* The key targets and pharmacological components of GFL in improving liver steatosis in NALFD rats hold promise as potential biomarkers for targeted drug design in clinical NAFLD treatment.

## Keywords

Multi-omics Techniques, Multiple Validations, Ganfule (GFL), Nonalcoholic Fatty Liver Disease (NAFLD), Pharmacological Mechanism

## **Funding**

This study was supported by the National Natural Science Foundation of China (Grant No. 81960797) and the Natural Science Foundation of Guangxi (Grant No. 2020GXNSFAA297156).