MULTI-OMICS REVEALED THAT THE POSTBIOTIC OF HAWTHORN-PROBIOTIC ALLEVIATED CONSTIPATION CAUSED BY LOPERAMIDE IN ELDERLY MICE

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Constipation is a common gastrointestinal disorder with high prevalence and recurrence, making people suffer. However, the treatment for constipation remains ineffectual. Our previous research found the postbiotic of hawthorn-probiotic alleviates constipation. We aimed to study the mechanisms of it on loperamide-modeled old KM mice. Constipated mice were grouped and treated with 10% lactulose (Y), hawthorn group (S), probiotic group (F), and postbiotic of hawthorn-probiotic (FS). Constituents of F, S, and FS were detected by UPLC-MS metabolomics. The components with higher FS than S were screened for network pharmacological analysis. The target of FS action on constipation was found. The contents of these targets in the colon of mice were detected by RT-qPCR, and the contents of IL-6 and IL-17A were also detected. The elevated components of FS were molecular-docked with these targets using AutoDock Tools1.5.6, and with previously discovered TNF-a, AQP3, and Enac-r. At the same time, these components were added to the intestinal organoid model for observation, and the changes in the target were detected by RT-qPCR. The small molecular components whose docking results were consistent with organoid results were selected and molecular dynamics analysis was performed with corresponding targets. The content of short-chain fatty acids in the stool of mice was detected by targeted metabolomics. The metabolomics of UPLC-MS showed that F, S, and FS were different. Compared with S, FS showed a decrease in toxic substances and an increase in beneficial substances. There were 19 cross-targets of FS and constipation found by network pharmacology. RT-qPCR detected the colons of mice and found that NR1I2 and SULT1A1 decreased and GLP-2r increased in FS compared model group. At the same time, the inflammatory cytokines IL-6 and IL-17A were reduced. Organoid dosification and molecular dockings revealed TNF-Baicalin, AQP3-Quinacridone, Enacγ-Quinacridone, IL17A-Baicalin, SULT1A1- N-Phenyl-2 naphthylamine, GLP2R- L-Isoleucine is well combined. After binding, small molecules adjust the content of the target back to the content of the normal group. However, IL-6 had a poor bonding effect and was not docked. The results of RMSD, RMSF, and RG of the above small molecules and targets were all good. Targeted metabolomics showed that the contents of various short-chain fatty acids in the feces of the F, S, and FS groups were different. 5/6 Postbiotic of hawthorn-probiotic relieved constipation Except through the combined effects of regulating intestinal water and sodium metabolism, in addition to maintaining intestinal barrier and gut microflora, can also promote epithelial cell proliferation, reduce inflammatory response, and improve short-chain fatty acid metabolism.

KEYWORDS: postbiotic, hawthorn, probiotic, constipation, inflammation, intestinal microenvironment