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Soyabean glycinin depresses intestinal growth and function in juvenile Jian carp (*Cyprinus carpio* var Jian): protective effects of glutamine

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Abstract

This study investigated the effects of glycinin on the growth, intestinal oxidative status, tight junction components, cytokines and apoptosis signalling factors of fish. The results showed that an 80 g/kg diet of glycinin exposure for 42 d caused poor growth performance and depressed intestinal growth and function of juvenile Jian carp (*Cyprinus carpio* var. Jian). Meanwhile, dietary glycinin exposure induced increases in lipid peroxidation and protein oxidation; it caused reductions in superoxide dismutase (SOD), catalase and glutathione peroxidase (GPx) activities; and it increased *MnSOD, CuZnSOD, GPx1b and GPx4a* mRNA levels, suggesting an adaptive mechanism against stress in the intestines of fish. However, dietary glycinin exposure decreased both the activity and mRNA levels of nine isoforms of glutathione-*S*-transferase (*GST*) (α, μ, π, ρ, θ, κ, *mGST1*, *mGST2* and *mGST3*), indicating toxicity to this enzyme activity and corresponding isoform gene expressions. In addition, glycinin exposure caused partial disruption of intestinal cell–cell tight junction components, disturbances of cytokines and induced apoptosis signalling in the distal intestines > mid intestines > proximal intestines of fish. Glycinin exposure also disturbed the mRNA levels of intestinal-related signalling factors *Nrf2*, *Keap1a*, *Keap1b*, eleven isoforms of protein kinase C and target of rapamycin/*4E-BP*. Interestingly, glutamine was observed to partially block those negative influences. In conclusion, this study indicates that dietary glycinin exposure causes intestinal oxidative damage and disruption of intestinal physical barriers and functions and reduces fish growth, but glutamine can reverse those negative effects in fish. This study provides some information on the mechanism of glycinin-induced negative effects.

Key words: **Glycinin: Intestine: Antioxidant: Tight junction: Cytokine: Apoptosis**

The growing demand to substitute fish meal in aqua feed has resulted in the need to search for alternative less-expensive and protein-rich sources(1). Soyabean meal (SBM) is one of the most promising plant protein sources as a fish meal substitute in fish feeding(2). However, its use is also limited because of the occurrence of the antinutritional factors(3). Glycinin, the main storage protein in soyabean, has been identified as a major antinutritional factor in soyabean(4). Studies have shown that glycinin could reduce weight gain in piglets(5). Our previous studies demonstrated that poor growth performance is always attributed to a reduction in the digestive and absorptive ability, which is partially related to the impaired intestinal integrity of fish(6,7). Limited studies have observed that glycinin could cause intestinal epithelium damage in piglets(5). However, until now, the underlying mechanisms by which glycinin caused damage to the intestine epithelium in animals have been largely unknown. Nevertheless, this topic is very important for understanding and resolving the problems of using high doses of SBM.

The intestinal structural integrity relies on the integrity of the intestinal physical barrier, which comprises intestinal epithelial cells(8). Our previous studies observed that the intestinal epithelial cell structure of fish is very sensitive to oxidative damage(6,9).

Abbreviations: AKP, alkaline phosphatase; CAT, catalase; DI, distal intestine; Gln, glutamine; γ-GT, γ-glutamyl transpeptidase; GPx, glutathione peroxidase; GSH, glutathione; GST, glutathione-*S*-transferase; IPC, intestinal protein content; MDA, malondialdehyde; MI, mid intestine; PC, protein carbonyl; PI, proximal intestine; ROS, reactive oxygen species; SBM, soyabean meal; SOD, superoxide dismutase; TGF-β2, transformed growth factor-β2; TJ, tight junction; TOR, target of rapamycin.

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