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Studies on the effect of taurine supplementation to low or non fishmeal diet on growth, nutrient digestibility, intestinal morphology and cytokines gene expression in juvenile red seabream, *Pagrus major*

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## The Dissertation summary

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論文題目 Title	Studies on the effect of taurine supplementation to low or non fishmeal diet on growth, nutrient digestibility, intestinal morphology and cytokines gene expression in juvenile red seabream, <i>Pagrus major</i> (マダイ稚魚の成長、消化率、消化管形態、サイトカイン遺伝子の発現に対する低・無魚粉飼料へのタウリンの添加効果に関する研究)		

**The purpose**

Plant protein (PP) sources have been receiving considerable attention over the past few decades as a partial or total fishmeal alternatives in aquafeed industry. And among the ingredients investigated, the products, soybean meal (SBM) and corn gluten meal (CGM) are the most promising. However, the presence of anti-nutritional factors (ANF's), the imbalances in amino acids, especially the low content of taurine in SBM and CGM have adverse impact on fish growth performance, feed utilization and health. Intestinal health is vital for body health. Once normal intestinal function is damaged, it will cause many diseases, such as loss of appetite, low food intake, growth retardation and the decline of digestive and absorptive capacity of nutrients. In fish, feed composition, especially the plant protein can always cause oxidative damage and structural damage of intestinal tract of fish, and then induce enteritis. In many inflammatory diseases, the increase of the cytokine levels can be detected. PPs have very low inclusion of taurine and FM is rich in taurine. Consequently, the removal of fishmeal may create several taurine deficiency symptoms. However, these physiological defects can be overcome by dietary taurine supplementation. Meanwhile, several studies have suggested that enzyme complex and some other additives supplementation help recovery from negative impact caused by PP.

**Methodology**

Three studies were designed to, i) evaluate the supplement effect of taurine and enzyme complex in plant protein based low fishmeal diet on growth performance and body health of red sea bream with, ii) evaluate the effect of complete replacement of fishmeal with PP and taurine supplement on growth performance, feed utilization, morphology and cytokine gene expression in intestine of juvenile red seabream and iii) evaluate the effect of graded levels of taurine supplementation to non fishmeal diet on growth performance, feed utilization and nutrient digestibility of juvenile red seabream.

**Experiments/analysis**

In the first experiment, a 12-week feeding trial of juvenile red seabream with initial weight of 7.6g was conducted. In this trial, four iso-nitrogenous (47% protein) diets were employed: fishmeal-based control diet (FM); low fishmeal diet (60% of fish meal was replaced by PP), designated LFM; the diet supplemented with 0.5% taurine to LFM, designated as TLFM and the diet supplemented with 0.5% taurine and 0.05% enzyme complex to LFM, designated as TELFM.

Second experiment was conducted to evaluate the effect of complete replacement of fishmeal with PP and taurine supplement on taurine on growth performance, feed utilization, morphology and cytokine gene expression in intestine of juvenile red seabream. Four iso-nitrogenous diets with 47% protein were prepared. Red sea bream (5.4g) were fed one of the diets formulated with 50% fishmeal diet (FM, control) or soy and corn protein based non fishmeal diets supplemented with increasing level of 1.0-2.0% taurine (NFM+T1.0, NFM+T1.5 and NFM+T2.0)

for 10 weeks.

The third experiment (5.8g initial weight) was conducted to evaluate the effect of graded levels of taurine supplementation to non fishmeal diet on growth performance, feed utilization and nutrient digestibility of juvenile red seabream for 12 weeks. Diet formulations in this study were almost same with the experiment 2, except two non fishmeal diets with 0 and 0.5% taurine were added. Six diets were designated as FM (control), NFM, NFM+T0.5, NFM+T1.0, NFM+T1.5 and NFM+T2.0. Fish fed NFM diets performed equally well final weight compared with the control group.

### **Results and discussion**

In the first experiment, fish fed LFM showed significantly lower weight gain (%WG) and specific growth ratio (%SGR) than control group while TLFM and TELFM groups showed similar growth performance with control. Feed efficiency in terms of feed efficiency ratio (FCR) and protein efficiency ratio (PER) in LFM group was significantly lower than control and TELFM groups. Daily feed intake (DFI) showed no difference among diet groups. Typical pathological change (Neutrophil infiltration and blood congestion) mainly appeared in intestine submucosa of fish fed LFM. The expression of inflammation-relative genes (IL-1 $\beta$ , IL-8, TNF- $\alpha$  and TGF- $\beta$ 1) were up-regulated in intestine of fish fed low fishmeal diets. And in low fishmeal groups, fish have significantly higher expression level than the control. However, in liver, the cytokine genes expression levels showed no difference among diet groups. Results in this experiment suggested that fish fed LFM indicated poorer growth, feed utilization and severer inflammatory response while taurine and/or enzyme complex supplementation ameliorated soybean-induced adverse impacts.

In the second experiment, fish fed NFM diets performed equally well final weight and DFI compared with the control group. Growth parameters in terms of %WG and SGR in FM group were significantly higher than those in NFM groups. Among non-fishmeal groups, WG and SGR showed no significant difference with increasing levels of taurine supplementation. Fish fed NFM+1.0T performed significantly lower FCR than control. And PER in fish fed NFM+1.0T was significantly lower than control. Among non fishmeal groups, PER gradually increased with increasing level of taurine and PER of NFM+2.0 was significantly higher than NFM+1.0. Typical pathological change for soybean enteropathy such as neutrophil infiltration was observed in intestinal submucosa of fish fed NFM. Inflammatory cytokine genes were significantly up-regulated in intestine of NFM groups than FM group. Although there was no significant difference in gene expression level, gene expression levels decreased with increasing taurine supplementation levels. These results demonstrated that complete replacement of fishmeal in red seabream diet by PP caused pathological changes and high expression of cytokine genes in intestine, but these changes can be improved by taurine supplementation.

In the third experiment, % WG and SGR in non fishmeal groups with less inclusion of taurine were significantly lower than the other groups. For DFI, there was no difference among diet groups. Fish fed NFM and NFM+0.5T performed significantly lower feed utilization efficiencies in terms of feed efficiency and PER than the other groups. Among non fishmeal groups, growth parameters and feed utilization efficiency were improved with graded levels of taurine supplementation to diets. Apparent digestibility coefficient (ADC) of dry matter, protein and lipid were significantly higher in FM group than NFM, NFM+T0.5, NFM+T1.0 group. Comparing the ADCs among non fishmeal groups, values were gradually increased as elevation of taurine supplement. ADCs of dry matter and lipid in NFM+T2.0 were significantly higher than NFM groups. Among the non fishmeal groups, the ADC values were tended to elevate with increasing taurine supplementation, even though there was no significant difference.

In conclusion, dietary taurine improved fish growth performance and feed utilization and ameliorates pathological changes of distal intestine such as intestinal inflammation. FM in red seabream diet can be completely replaced without negative effect on growth with sufficient level of taurine supplementation. However, from the viewpoint of intestinal health, FM complete replacement along with PP was not feasible.