Dietary Lactosucrose Decreases Hepatic and Plasma Lipid Contents in a Marine Teleost, Red Sea Bream Pagrus major

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Abstract: The aims of this study were to confirm the effects of dietary lactosucrose (LS), a fermentable oligosaccharide, on plasma and liver lipid levels in red sea bream. Fish were assigned to two groups and fed either a commercial diet or a commercial diet supplemented with 0.24% LS for two months. Fish fed LS had lower liver lipid contents than did control diet-fed fish (P < 0.001). Levels of plasma cholesterol and triglyceride but not HDL-cholesterol in fish fed LS were significantly lower than in control fish (P < 0.001). The above results indicate that the dietary fermentable saccharide LS affected lipid metabolism in this fish.

Key words: Red sea bream Pagrus major; Lactosucrose; Lipid metabolism; Oligosaccharide

Body fat contributes to the supply of high metabolic energy in fish. However, excessive fat accumulation in viscera reduces commercial value, especially in cultured fish. Red sea bream is one of the most popular cultured fish in Japan, but consumers are not satisfied with relative high lipid contents in this fish (Forster and Ogata 1996). One study indicates that lipid content of the cultured red sea bream was 2.5 to 4.8 times higher than in the wild type (Aoki et al. 1991). Some fermentable dietary fiber lowers plasma cholesterol concentrations (Anderson 1995, Hara et al. 1998). Hara et al. (1998) suggests that the suppressive effect of sugar-beet fiber, a highly fermentable dietary fiber, on plasma cholesterol levels is at least partly caused by cecal fermentation products, which are short-chain fatty acids (SCFA) such as acetic, propionic and n-butyric acids. Previous study demonstrated that hind-gut microbes of red sea bream Pagrus major could ferment lactosucrose and produce SCFA in vivo and in vitro (Kihara et al. 1995, Kihara 2008). Therefore, if dietary fermentable saccharides affect lipid metabolism in the fish, plasma cholesterol levels might be modified by dietary lactosucrose, as in mammals (Anderson 1995, Hara et al. 1998). Accordingly, this study measured levels of plasma lipids and lipid contents in the liver of red sea bream.

Approximately 70 g body weight red sea bream Pagrus major were divided into 2 groups of 50, namely control and lactosucrose groups. The fish were reared in 1,000 l transparent polycarbonate tanks and supplied with filtered and UV-sterilized seawater maintained at 23.0°C and circulated at a rate of 28,000 l/day to each tank. Tanks were placed under natural light conditions. Fish in the control group were fed a conventional commercial pelleted diet for red sea bream, while fish in the lactosucrose group were fed the above commercial diet containing 0.24% lactosucrose (Kihara 2008) for two months. Diets were fed at 1.5 g/100g body weight at 08:00, 11:00, 14:00 and 17:00 every day. On the sampling day, seven fish from each tank were randomly selected and anesthetized with 0.2 ml/l of 2-phenoxyethanol, then weighed individually. Fish were sacrificed by blood collection from the caudal vein with a heparinized syringe (SS-01T, Terumo, Tokyo) through a 27G needle (NN-2719S, Terumo) and blood was centrifuged at 2,000 g for 12 min in a micro test-tube (Eppendorf, Hamburg, Germany). After opening the abdominal cavity, the liver was collected. Then the liver was rinsed with ice-cold phosphate buffered saline (pH 7.0) and blotted on dry filter paper (No. 2, Toyo Roshi Kaisha, Ltd., Tokyo) before weighing. Liver and plasma were frozen at −30°C until analysis. The thawed liver was minced with scissors, then homogenized with an ultra-disperser (LK-22, Yamato Scientific Co., Ltd., Tokyo) equipped with a G-10 generator (Yamato Scientific Co., Ltd.) at 20,500 r.p.m. for 3 min. One gram of homogenized liver was further homogenized with 60 ml of a mixture of chloroform and methanol (2:1, v/v) in a homogenizer (AM-9S, NISSEI, Tokyo) at 14,000 r.p.m. for 5 min. The extracts were filtered through No. 2 filter paper under reduced pressure. The filtrate and 25 ml of 0.03M MgCl2 solution were added to a separation funnel. The gas phase in the funnel was substituted with N2 and left for a night after vigorous shaking. The lower layer was collected in a flask and evaporated with a rotary evaporator at 30°C. This residue was weighed and arbitrarily termed ‘crude lipid’. Total cholesterol, triglyceride and HDL-cholesterol in the plasma were measured using an automated biochemical analyzer (DryLab 80M, Konica Corporation, Tokyo). Values were given as means with SD. Differences in
mean body weight, lipid content of the wet liver, plasma triglyceride, cholesterol and HDL-cholesterol content were tested by one-way ANOVA. When the variances varied significantly between groups ($P < 0.05$, by Bartlett test), Kruskal-Wallis analysis of variance was conducted. The differences were considered significant at $P < 0.05$. All statistical analyses were conducted using a computer program (STATISTICA, StatSoft, Inc., Tulsa, OK).

Final body weight did not differ between groups (control, $112.4 \pm 11.8$ g; lactosucrose, $90.7 \pm 20.0$ g; mean $\pm$ SD). Feed conversion efficiency calculated from each tank was $53.9$ in fish given control diet and $43.9$ in fish given lactosucrose diet, respectively. Relative wet weight of the liver did not vary between groups. Crude lipid content per wet liver varied between groups ($P < 0.001$, Fig. 1). Liver crude lipid level was approximately $30\%$ lower in fish fed lactosucrose than in fish fed the control diet. Plasma triglyceride content in fish fed lactosucrose diet was lower than in fish fed the control diet ($P < 0.001$, Fig. 2). Fish fed the lactosucrose diet had lower plasma cholesterol level than did fish fed the control diet ($P < 0.001$, Fig. 2). Plasma HDL-cholesterol level did not vary between groups (Fig. 2).

The dietary fermentable saccharide lactosucrose, that is fermented by hindgut microbes of red sea bream (Kihara et al. 1995, Kihara 2008), affected lipid metabolism in this fish (Fig. 1 and 2). This agrees with results in mammals that some fermentable dietary fiber lowers plasma cholesterol concentrations in mammals (Anderson 1995, Hara et al. 1998). Hara et al. (1998) suggests that the suppressive effect of fermentable fiber on plasma cholesterol levels is at least partly caused by cecal fermentation products, SCFA. Thus the effect of dietary lactosucrose on the host fish, probably produced by SCFA from bacterial metabolism, might be concerned with energy metabolism. So far, there is no explanation for this suppressive effect of fermentable oligosaccharide on the lipid levels in fish. If the gut fermentation products SCFA affect lipid metabolism in fish, dietary fermentable materials might be expected to improve lipid over-accumulation in cultured fish. We should confirm if the above lowering effect on plasma and liver lipid levels in fish is also caused by SCFA, in order to define the physiological effect of intestinal fermentation on the host fish.

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**References**


