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Aquaculture 257 (2006) 437–445
Aquaculture
www.elsevier.com/locate/aqua-online

Growth and phosphorus loading by partially replacing fishmeal with tuna muscle by-product powder in the diet of juvenile Japanese flounder, *Paralichthys olivaceus*

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Received 6 December 2005; received in revised form 27 February 2006; accepted 27 February 2006

Abstract

Tuna muscle by-product powder (TMP), obtained from residues of tuna muscle, was replaced with fishmeal to evaluate its quality, substitution level and phosphorus loading for juvenile Japanese flounder. TMP was substituted for fishmeal on a crude protein basis with levels of 25%, 50%, 75% and 100%, respectively. The feeding trial continued for 40 days. Weight gain, specific growth rate and feed intake of fish were not significantly different among the fish fed the control (0% TMP inclusion), 25% and 50% TMP containing diets. However, these parameters significantly ($P<0.05$) decreased in fish fed diets containing TMP at 75%, 100% with or without phosphorus supplements compared to those fed diets containing TMP at 0%, 25% and 50%. The dietary free histidine level increased with increasing TMP levels while other free amino acids decreased. Negative correlation ($R^2=0.978$) was observed between dietary free histidine level and feed intake. Phosphorus (P) loading markedly decreased with increasing TMP level in the diets. The present study demonstrated that TMP is a promising feed ingredient to reduce P discharge into the environment, and could replace 50% of fishmeal protein without reduction of growth performances, resulting in about 50% lower P loading than the control group.

Keywords: Alternative protein source; Fishmeal replacement; Tuna muscle by-product powder; Phosphorus loading; *Paralichthys olivaceus*

1. Introduction

Worldwide aquaculture production continues to increase, thereby increasing the demand for fish feeds containing fishmeal (Watanabe, 2002). Due to limited sources, supplies and high cost of fishmeals, many studies have been made to use alternative protein sources such as plant proteins (Riche and Brown, 1999; Kissil et

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doi:10.1016/j.aquaculture.2006.02.060

al., 2000; Kaushik et al., 2004), terrestrial animal-based proteins (Millamena and Golez, 2001; Williams et al., 2003) and fish by-product meals (Sugiura et al., 2000), for reducing fishmeal in aquafeeds.

Japan is the world's major tuna catching country, and is also known as the major world market of tuna for direct consumption such as sashimi and sushi. Edible parts of tuna muscle (mainly blue-fin, big-eye, yellow-fin and skipjack) are cut and shaped into meat blocks before releasing to the Japanese market. During this process, unused parts are available as a by-product and are collected, drum-dried to make a powdered product. Although tuna muscle by-product powder (TMP) has been used in pet foods, none is used in aquafeeds.

Phosphorus (P) is an essential mineral required for growth, bone mineralization, reproduction, synthesis of nucleic acids and structure of phospholipids as well as for energy metabolism in fish (Tacon, 1990; NRC, 1993; Roy and Lall, 2003). On the other hand, P is also a major nutrient for growth of microorganisms in the aquatic environment, and can contribute to excessive algae and macrophyte growth in receiving waters (Pillay, 1992; Bureau and Cho, 1999; Storebakken et al., 2000). Fishmeal, containing 16.7–42.1 mg/g P, is the main P source in aquafeeds, and accounts for 30–50% by weight in most carnivorous fish feeds (NRC, 1993). Furthermore, P from fishmeal is not well utilized by aquatic animals so one strategy to reduce P output from aquaculture operations is to decrease fishmeal levels in aquafeeds by using low P containing ingredients (Lall, 1991; Talbot and Hole, 1994). Recently, the inclusion of low P de-boned fishmeal has become popular among feed manufacturers to reduce P content of fish diets (Sugiura et al., 2004).

Due to its high protein (~80% in dry matter) and low P content (~15 mg/g in dry matter), TMP would be a promising candidate to reduce P level in aquafeeds minimizing environmental impact. Since dietary TMP has not been tested in aquatic animals, this study was designed to evaluate the effects of dietary TMP level on growth and P loading of juvenile Japanese flounders by partially or completely replacing fishmeal.

2. Materials and methods

2.1. Test diets

The formulation of the experimental diets is shown in Table 1. The protein source was brown fishmeal (73.9% protein, 10.7% lipid, 27.6 mg/g P in dry matter). TMP (79.9% protein, 14.1% lipid, and 15.5 mg/g P in dry matter) was provided by a private company, and incorporated to replace fishmeal protein at 0%, 25%, 50%, 75% and 100% (diet 1, 2, 3, 4, and 5, respectively). All diets were isonitrogenous and isolipidic, and diets 6 and 7 were formulated by adding 1% inorganic P, $Ca(H_2PO_4)_2H_2O$ (Kanto Chemicals, Tokyo, Japan), to diets 4 and 5, respectively, in order to clarify the possible lack of adequate dietary P level in diets 4 and 5. Pollack liver oil served as the lipid source, starch and dextrin were the carbohydrate sources in the diets.

TMP was ground to the desired particle size prior to preparing the diet. To prepare diets, all dry ingredients were well mixed for 30 min in a food mixer. Then pollack liver oil was added, and mixed for 15 min. Finally, water (35% of the dry weight of ingredients) was put, and again mixed for 15 min. The pH of the diets was adjusted to 7.0–7.5 with 4 N sodium hydroxide. The pellets were (1.2 to 2.2 mm diameter) made with a meal grinder, and dried in a dry-air mechanical convection oven (DK 400, Yamato Scientific, Japan) at 70 °C for 2 to 4 h to obtain approximately 10% moisture level. The experimental diets were stored at −30 °C until used.

Test diets (Table 1) had similar levels of crude protein (45–46%) and crude lipid content (13–15%). Ash and total P content decreased with inclusion level of TMP in the diets.

2.2. Fish and experimental design

Japanese flounder, *Paralichthys olivaceus*, juveniles (average weight, means ± S.E., 3.60 ± 0.04 g) were obtained from a commercial hatchery (Matsumoto Suisan Co., Miyazaki, Japan) and randomly allocated to 21 polycarbonate circular tanks (100 l/tank) with triplicate groups consisting of 20 fish each. The fish were maintained on a commercial formulated flounder diet ("Feed for marine juvenile fish", Nippon Suisan, Tokyo, Japan) for 10 days prior to the experiment. The juveniles were fed the respective test diets at a rate of 5% of body weight per day. Daily ration size was divided into two equal feedings at 8:30 and 16:30 h. Every 10 days, all fish were weighed in bulk to adjust the ration size. The feeding trial was conducted for 40 days. All rearing tanks were provided with continuous aeration and maintained under natural light/dark regime (12 : 12 h). Filtered seawater was continuously provided at a flow rate of 1.5 l/min. Water temperature, pH and salinity (mean ± S.E.) were 21.0 ± 1.6 °C, 8.0 ± 0.2, and 33.3 ± 0.4‰, respectively, during the trial.

2.3. Evaluation of the data from feeding trial

Feed intake was recorded by subtracting the amount of uneaten diet from total amount of diet fed on a dry