Thesis for the Degree of Master of Science

Dietary substitution of fishmeal with tuna byproduct meal on growth, feed utilization, body composition, serum chemistry and amino acid profiles of juvenile Korean rockfish (*Sebastes schlegeli*)

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by

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요 약

본 연구에서는 국내 주요 양식어종 중 하나인 조피볼락(*Sebastes schlegeli*)를 대상으로 하여 참치 가공부산물을 이용한 사료 내 어분대체에 따른 조피볼락의 성장, 사료 이용성, 체조성, 혈액성상학적 변화 및 체구성 아미노산 변화에 미치는 영향을 조사하였다.

810마리의 조피볼락을 임의로 선별하여 27개의 50L 유수식 수조에 수용하였다. 모든 실험어는 손으로 만복시까지 1주일에 7일간 1일 2회 (08:00, 17:00) 매일 사료를 공급하여 주었으며, 사육실험 기간은 총 8주간이었다.

실험에 이용된 실험사료는 총 9종류의 사료를 준비하였다. 어분을 55%을 첨가하여 제조한 대조구 사료(Con), 어분을 참치 가공부산물로 대체한 어분대체 10%(TBM10), 어분대체 20%(TBM20), 어분대체 30%(TBM30), 어

분대체 40%(TBM40), 어분대체 60%(TBM60), 어분대체 80%(TBM80)와 어분대체 100%(TBM100) 사료를 준비하였다. 또한 어분과 대두박을 모두 참치 가공부산물로 대체한 어분 및 대두박 대체 100%(TBM100-S) 사료를 준비하였다. 모든 사료는 동일한 함량의 단백질(isonitronic)과 지질(isolipidic)을 가지게끔 제조하였으며, 각 실험구는 3반복구를 두었다.

배합사료 내 어분이 참치 가공부산물로 대체됨에 따라서 사료 내 lysine 함량이 증가하는 경향을 나타내었다. 그리고 배합사료 내 methionine의 경우 어분을 40% 이상 대체함에 따라서 급격히 감소하였다.

조피볼락의 어체중 증가량(weight gain)은 TBM10, TBM20과 TBM30 사료를 공급한 실험구에서 TBM40, TBM60, TBM80, TBM100과 TBM 100-S 사료를 공급한 실험구보다 높게 나타났지만 대조구(Con) 사료를 공급하는 실험구와는 유의적인 차이가 없었다. 일일성장률(Specific growth rate, SGR)은 TBM20과 TBM30 사료를 공급한 실험구에서 TBM40, TBM60, TBM80, TBM100과 TBM 100-S 사료를 공급한 실험구보다 높게 나타났지만 TBM10과 대조구 사료를 공급한 실험구와는 유의적인 차이를 보이지 않았다.

사료전환효율(Feed efficiency ratio, FER)은 대조구와 TBM10, TBM20과 TBM30 사료를 공급한 실험구에서 다른 모든 실험구보다 높게 나타났다. 단백질전환효율(Protein efficiency ratio, PER)의 경우 TBM10, TBM20과 TBM30 사료를 공급한 실험구에서 TBM40, TBM60, TBM80, TBM100과 TBM 100-S 사료를 공급한 실험구보다 높게 나타났지만 대조구 사료를 공급한 실험구와는 유의적인 차이가 없었다.

8주간의 사육 실험 종료시 생존한 어체를 대상으로 하여 수행한 조피볼락의 혈액성상학적 분석 결과, total protein, glucose, GOT(glutamate oxaloacetate transaminase), GPT(glutamate pyruvate transaminase)와

triglyceride의 함량은 실험구간에 유의적인 차이가 나타나지 않았다. 또한 실험어의 체구성 아미노산 분석 결과도 실험구간에 유의적인 차이가 나타나지 않았다.

이상의 결과를 고려할 때 조피볼락용 배합사료 내 어분을 참치 가공부산물로 대체시 성장률(SGR)의 경우 어분의 40%까지, 사료 효율의 경우 어분의 30%까지 대체가 가능한 것으로 판단되며, 어분의 가격이 급격히 상승하고 있는 현실을 고려할 때 양어사료 시장에서의 어분 절감 효과를 기대할 수 있다.

Keywords 조피볼락; 참치 가공부산물; 어분; 성장률; 일일성장률; 사료전환효율; 단백질전환효율

I. Experiment

Dietary substitution of fishmeal with tuna byproduct meal on growth, feed utilization, body composition, serum chemistry and amino acid profiles of juvenile Korean rockfish (*Sebastes schlegeli*)

Abstract

Effect of dietary substitution of fishmeal with tuna byproduct meal (TBM) in the diet on growth, body composition, serum chemistry and amino acid profiles of juvenile rockfish was determined. Eight hundred ten juvenile fish were randomly distributed into 27 of 50-l flow-through tanks. Nine experimental diets were prepared in triplicate. A 55% fishmeal was included into the control (Con) diet. The 10, 20, 30, 40, 60, 80 and 100% fishmeal were substituted with TBM, referred to as the TBM10, TBM20, TBM30, TBM40, TBM60, TBM80 and TBM100 diets, respectively. Finally, the 100% fishmeal and soybean meal were replaced with TBM to determine the combined substitution effect of fishmeal and soybean meal with TBM, referred to as TBM100-S diet. All experimental diets were prepared at isonitronic and isolipidic. Lysine content tended to decrease with an increased TBM in the experimental diets. Methionine content sharply dropped above 40% TBM substitution. Weight gain of fish fed the TBM10, TBM20 and TBM30 diets were higher than that of fish fed the TBM40, TBM60, TBM80, TBM100 and TBM100-S diets, but not different from that of fish fed the Con diet. Specific growth rate (SGR) of fish fed the TBM20 and TBM30 diets were higher than that of fish fed the TBM40, TBM60, TBM80, TBM100 and TBM100-S diets, but not different from that of fish fed the Con and TBM10 diets. Feed efficiency ratio (FER) of fish fed the Con, TBM10, TBM20 and TBM30 diets was higher than that of fish fed the all other diets. Protein efficiency ratio (PER) of fish fed the TBM10, TBM20 and TBM30 diets was higher than that of fish fed the TBM40, TBM60, TBM80, TBM100 and TBM100-S diets, but not different from that of fish fed the Con diet. It can be concluded that substitution of fishmeal with TBM up to 40 and 30% in the diets could be made without detrimental effect on growth (SGR) and feed utilization (FER and PER) of juvenile rockfish, respectively.

Keywords Rockfish (*Sebastes schlegeli*); Tuna byproduct meal; Fishmeal Weightgain; Specific growth rate; Feed efficiency ratio; Protein efficiency ratio

1. Introduction

Rockfish (*Sebastes schlegeli*) is a commercially important marine fish species, whose annual aquaculture production in 2012 reached 17,338 metric tones in Korea (MFAFF 2013). Therefore, many feeding trials to determine dietary nutrient requirements (Kim et al. 2001; Wang et al. 2003; Yan et al. 2007), digestibility of feed ingredients (Bai et al. 2001; Lee 2002; Lee et al. 2002), alternative animal and/or plant protein sources for fishmeal in the diets (Lee et al. 1996; Lim et al. 2004), optimum feeding frequency (Lee et al. 2000; Seo and Lee 2008), feeding strategy (Oh et al. 2008), comparison of the extruded pellet with the raw fish-based moist pellet for performance (Kim et al. 2003) and dietary additive to improve immune response (Kim et al. 1999; Bai et al. 2001) of rockfish have been performed.

Fishmeal has been used as one of the most popular protein sources in the diets for fish culture, but its international market price has kept skyrocketing. Therefore, the several candidate plant products for aquafeeds were suggested (Gatlin et al. 2007). Lim et al. (2004) reported that dietary soybean meal could be replaced with fishmeal up to 20% without amino acid and 30% with supplementation for fingerling and growing rockfish, respectively. In addition, dietary substitution of fishmeal with corn gluten meal up to 15%, meat meal up to 10%, meat and bone meal up to 10% and blood meal up to 10% could be made without retardation of weight gain and feed utilization of rockfish (Lee et al. 1996).

However, animal protein sources such as meat meal, meat and bone meal, blood meal and feather meal can not be safely used as an alternative protein source for fishmeal in the diet for fish culture anymore because of high risk of disease transfer or infection of land-animals originated such as foot-and-mouth disease, mad cow disease and bird-flu to human being when used for feed ingredient. In addition, although Hardy (2010) had proposed that the alternative ingredients from plant protein sources whose global production should be sufficient to supply the needs of aquafeeds for future, plant protein sources such as soybean and corn gluten meals can not be economically used as the alternative protein source for fishmeal anymore because of their high price resulted from an expansion of biofueling industry to develop soybean and corn as a seed stock for ethanol production over the world.

Therefore, a new feed ingredient that is safe from disease transfer or infection from land-animal originated and economically cheap to replace for fishmeal in the diet for fish culture is highly needed. Tuna byproduct is primary consisted of tuna head, bone, fin, blood and skin after the process of tuna canning. About 40 ton tuna byproduct is daily produced from the process of tuna canning by Ottogi SF and HIF Co., LTD in Korea. This has the high potential as the alternative protein source for fishmeal. Tuna byproduct is mixed with soybean meal at the ratio of 8:2 and then fermented by *Bacillus* sp. for 48 hours and then dried, referred to as tuna byproduct meal (TBM). TBM has been recently developed as the new feed ingredient to replace for fishmeal in the diet for fish culture because of its high nutrient content with crude protein of above 55% and crude lipid of above 10% and stable supply. Recently, Kader and Koshio (2012) reported that 80% fishmeal could be replaced with the combined seafood byproduct and soybean proteins in a typical commercial diet without retardation of weight gain of red sea bream (*Pagrus major*).

In this study, therefore, effect of dietary substitution of fishmeal with TBM in the diet on growth, body composition, serum chemistry and amino acid profiles of juvenile rockfish was determined.

2. Materials and Methods

2.1. Fish and the Experimental Conditions

Juvenile rockfish were purchased from a private hatchery and acclimated to the experimental conditions for 2 weeks before an initiation of the feeding trial. Eight hundred ten juvenile (an initial body weight of 3.2 g) fish were randomly chosen and distributed into 27 of 50-l flow-through tanks (water volume: 40-l) (thirty fish per tank). The flow rate of water into each tank was 600-ml/min/tank. The water source was sand-filtered natural seawater and aeration was supplied into each tank. Water temperature monitored daily from 16.0 to 24.9°C (mean ± SD: 20.8 ± 2.61°C) and photoperiod followed natural conditions.

2.2. Design of the Feeding Trial and Preparation of the Experimental Diets

Nine experimental diets were prepared in triplicate (Table 1).

Fishmeal, TBM, dehulled soybean meal and casein were used as the protein source in the experimental diets. Wheat flour, and squid liver and soybean oils were used as the carbohydrate and lipid sources, respectively. A 55% fishmeal was included into the control (Con) diet to satisfy dietary nutrients requirements for rockfish (Kim et al. 2001; Lee et al. 2002). The 10, 20, 30, 40, 60, 80 and 100% fishmeal were substituted with TBM, referred to as the TBM10, TBM20, TBM30, TBM40, TBM60, TBM80 and

Table 1. Ingredient and nutrient composition of the experimental diets

	Experimental diets								
	Con	TBM10	TBM20	TBM30	TBM40	TBM60	TBM80	TBM100	TBM100-S
Ingredients (%, DM)									
Fishmeal[1]	55.0	49.5	44.0	38.5	33.0	22.0	11.0	0	0
Tuna byproduct meal[2]	0	5.5	11.0	16.5	22.0	33.0	44.0	55.0	53.7
Dehulled soybean meal[3]	5.0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	0
Casein	0	0.8	1.6	2.4	3.2	4.9	6.5	8.1	10.9
Wheat flour	33.5	32.7	31.9	31.1	30.3	28.6	27.0	25.4	28.9
Squid liver oil	2	2	2	2	2	2	2	2	2
Soybean oil	2	2	2	2	2	2	2	2	2
Vitamin premix[4]	1	1	1	1	1	1	1	1	1
Mineral premix[5]	1	1	1	1	1	1	1	1	1
Choline	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Nutrients (%, DM)									
Dry matter	91.2	91.6	91.1	91.2	91.4	91.3	91.1	91.5	91.6
Crude protein	50.1	49.4	49.7	50.2	50.1	49.8	50.0	50.2	49.8
Crude lipid	11.3	10.9	11.1	11.9	11.8	11.3	11.1	11.3	11.2
Ash	8.0	8.6	8.5	8.6	8.6	9.2	9.6	9.6	9.6

[1]Fishmeal (CP: 75.0%, CL: 8.2%, ash: 16.1%) and [3]Dehulled soybeanmeal (CP: 55.1%, CL: 6.6%, ash: 6.4%)were provided by Jeilfeed Co Ltd., Haman-gun, Gyeongsangnam-do, Korea.

[2]Tuna byproduct meal (CP: 57.3%, CL: 8.0%, ash: 11.7%) was purchased from HIF Co., LTD, Changwon-si,

Gyeongsangnam-do, Korea

[4]Vitamin premix contained the following amount which were diluted in cellulose (g kg^{-1}mix): L-ascorbic acid, 121.2; DL-α-tocopheryl acetate, 18.8; thiamin hydrochloride, 2.7; riboflavin, 9.1; pyridoxine hydrochloride, 1.8; niacin, 36.4; Ca-D-pantothenate, 12.7; myo-inositol, 181.8; D-biotin, 0.27; folicacid, 0.68; p-aminobenzoicacid, 18.2; menadione, 1.8; retinyl acetate, 0.73; cholecalciferol, 0.003; cyanocobalamin, 0.003.

[5]Mineral premix contained the following ingredients (g kg^{-1}mix): $MgSO_4 \cdot 7H_2O$, 80.0; $NaH_2PO_4 \cdot 2H_2O$, 370.0; KCl, 130.0; Ferric citrate, 40.0; $ZnSO_4 \cdot 7H_2O$, 20.0; Ca-lactate, 356.5; CuCl, 0.2; $AlCl_3 \cdot 6H_2O$, 0.15; KI, 0.15; $Na_2Se_2O_3$, 0.01; $MnSO_4 \cdot H_2O$, 2.0; $CoCl_2 \cdot 6H_2O$, 1.0.

TBM100 diets, respectively. Finally, the 100% fishmeal and soybean meal were replaced with TBM to determine the combined substitution effect of fishmeal and soybean meal with TBM, referred to as TBM100-S diet. All experimental diets were prepared at isonitronic and isolipidic.

The ingredients of the experimental diets were well mixed with water at the ratio of 3:1 and pelletized by pellet-extruder. The experiment diets dried at room temperature overnight and stored into -20°C until use. All fish were hand-fed to apparent satiation twice a day (08:00 and 17:00 h) for 7 days a week throughout the 8-week feeding trial.

2.3. Analytical Procedures of the Experimental Diets and Fish

Fifteen fish from each tank at the termination of the feeding trial were sampled and sacrificed for proximate analysis. Crude protein was determined by the Kjeldahl method (Kjeltec 2100 Distillation Unit, Foss Tecator, Hoganas, Sweden), crude lipid was determined using an ether-extraction method (Soxtec TM 2043 Fat Extraction System, Foss Tecator, Sweden), moisture was determined by oven drying at 105°C for 24 h, fiber was determined using an automatic analyzer (Fibertec, Tecator, Sweden) and ash was determined using a muffle furnace at 550°C for 4 h, all methods were according to standard AOAC (1990). Amino acid composition of the experimental diets was determined by using an automatic amino acid analyzer (Sykam S4330, Eresing, Germany) after which the samples were hydrolyzed in 6 N HCl for 22 h at 110°C.

Blood samples were obtained from the caudal vein of randomly chosen seven fish from each tank by using syringes after they were starved for 24h at the end of the 8-week feeding trial. Serum was collected after centrifugation (3,000 rpm for 10 min), stored freezer at -70°C as separate aliquots for analysis of total protein, glucose, glutamate oxaloacetate transaminase (GOT), glutamate pyruvate transaminase (GPT) and triglyceride, and analyzed by using automatic chemistry system (Vitros DT60 II, Vitros DTE II, DTSC II Chemistry System, Johnson and Johnson Clinical Diagnostics Inc., New York, USA).

2.4. Statistical analysis

One-way ANOVA and Duncan`s multiple range test (Duncan 1955) were used to analyze the significance of the difference among the means of treatments through SAS version 9.3 (SAS Institute, Cary, NC, USA).

3. Results and Discussion

Amino acid profiles of the experimental diets were given in Table 2. An essential amino acid, lysine content tended to decrease with an increased TBM in the experimental diets. Another essential amino acid, methionine content slightly changed from 0.89 to 1.00% in the diets substituting fishmeal up to 40% with TBM, but sharply dropped above 40% TBM substitution. Dietary methionine requirement for maximum growth of juvenile rockfish was estimated to be 1.37% in the presence of 0.12% cystine, being equivalent to 2.8% of dietary protein (crude protein level of 48.7%) when fishmeal was used as intact protein source and crystal amino acid was used as a part of dietary protein (Yan et al. 2007). The optimal dietary methionine requirements were 0.91, 0.97, 1.0, 1.11, 1.19, 1.31, 1.44, 1.49 and 1.71% of the diets in the presence of 1.29, 0.03 0.04, 0.31, 0.67, 0.26, 0.29, 0.06 and 0.31% cystine for sea bass *Dicentrarchus labrax*, yellow perch *Perca flavescens*, rohu *Labeo rohita*, yellowtail *Seriola quinqueradiata*, cobia *Rachycentron canadum*, grouper *Epinephelus coioides*, yellow croaker *Pseudosciaena corcea*, black sea bream *Sparus macrocephalus*, flounder *Paralichthys olivaceus*, respectively (Hidalgo et al.,

Table 2 Amino acid profiles of the experimental diets (% in the diet)

	Experimental diets								
	Con	TBM10	TBM20	TBM30	TBM40	TBM60	TBM80	TBM100	TBM100-S
Alanine	2.86	2.70	2.68	2.64	2.67	3.22	2.58	2.71	2.75
Arginine	2.64	2.51	2.49	2.42	2.42	2.42	2.26	2.33	2.29
Aspartic	4.16	4.00	4.00	3.92	4.01	3.62	3.76	3.99	3.96
Cystine	0.72	0.80	0.82	0.82	0.92	0.93	1.02	1.11	1.14
Glutamic	7.32	7.04	7.03	7.00	7.12	5.78	7.01	7.65	7.83
Glycine	2.94	2.75	2.73	2.68	2.67	2.54	2.57	2.67	2.68
Histidine	1.73	1.59	1.53	1.45	1.59	1.51	1.62	1.89	1.91
Isoleucine	2.04	1.96	1.95	1.93	1.95	1.97	1.88	2.00	2.02
Leucine	3.43	3.34	3.40	3.43	3.55	3.70	3.71	4.04	4.21
Lysine	3.41	3.19	3.10	2.99	2.99	2.82	2.68	2.75	2.83
Methionine	0.97	1.00	0.96	0.89	0.93	0.82	0.77	0.76	0.82
Phenylalanine	1.87	1.77	1.84	1.86	1.93	1.91	1.98	2.19	2.22
Proline	2.19	2.20	2.31	2.40	2.49	2.86	2.85	3.21	3.47
Serine	0.97	1.17	1.13	1.18	1.40	0.76	1.38	1.49	1.51
Threonine	1.56	1.64	1.58	1.55	1.70	1.30	1.57	1.66	1.70
Tyrosine	0.45	0.61	0.70	0.84	0.73	1.01	0.82	0.75	0.79
Valine	2.51	2.43	2.46	2.48	2.53	2.63	2.61	2.88	2.93

1987; Ruchimat et al. 1997; Alam et al. 2000; Luo et al. 2005; Zhou et al. 2006; Twibell et al. 2000; Mai et al. 2006; Abidi and Khan 2011; Zhou et al. 2011).

Total sulfur amino acid ranged from 1.69% in the Con diet, which was lowest, being equivalent to 3.37% of dietary protein to 1.96%, which was highest, being equivalent to 3.94% of dietary protein in the TBM100-S diet. High (0.72-1.14%) cystein content in the experimental diets in this study compared to that (0.12%) in Yan et al. (2007)' study lowered methionine requirement to the some extent. Dietary cystine has a methionine-sparing effect in several fish species (Wilson and Halver 1986) and thus results in lower dietary methionine requirement of fish: cystine was able to spare approximately 39, 50, 51 and 60% of the dietary methionine requirement for rohu (Abidi and Khan 2011), red drum *Sciaenops ocellatus* (Goff and Gatlin 2004), yellow perch (Twibell et al. 2000) and channel catfish *Ictalurus punctatus* (Harding et al. 1977), respectively. This is why rockfish fed the Con, TBM10, TBM20, TBM30 and TBM40 diets that containing lower methionine content than dietary requirement (1.37%) estimated by Yan et al. (2007)'s study achieved good growth in this study.

Survival (%), weight gain (g/fish) and specific growth rate (SGR) of rockfish fed the experimental diets substituting fishmeal with TBM were presented in Table 3.

Survival of fish ranging from 95.6 to 100% was not significantly ($P > 0.05$) different among the experimental diets. However, weight gain of fish fed the TBM10, TBM20 and TBM30 diets were significantly ($P < 0.05$)

Table 3 Survival, weight gain (g/fish) and specific growth rate (SGR) of juvenile rockfish fed the experimental diets substituting fishmeal with tuna byproduct meal (TBM) for 8weeks (means of triplicate ± SE)[1]

Experimental diets	Initial weight (g/fish)	Final weight (g/fish)	Survival (%)	Weight gain (g/fish)	SGR[2]
Con	3.2 ± 0.01	11.6 ± 0.13[ab]	97.8 ± 1.11	8.4 ± 0.12[ab]	2.29 ± 0.020[ab]
TBM10	3.2 ± 0.01	11.8 ± 0.13[ab]	100.0 ± 0.00	8.6 ± 0.12[a]	2.32 ± 0.013[ab]
TBM20	3.2 ± 0.02	12.3 ± 0.54[a]	96.7 ± 3.33	9.1 ± 0.54[a]	2.40 ± 0.081[a]
TBM30	3.2 ± 0.00	11.9 ± 0.23[a]	96.7 ± 1.92	8.7 ± 0.23[a]	2.33 ± 0.035[a]
TBM40	3.2 ± 0.01	11.0 ± 0.23[bc]	95.6 ± 1.11	7.8 ± 0.22[bc]	2.19 ± 0.034[bc]
TBM60	3.2 ± 0.00	10.3 ± 0.35[cd]	96.7 ± 1.92	7.1 ± 0.35[cd]	2.08 ± 0.061[cd]
TBM80	3.2 ± 0.01	9.9 ± 0.20[de]	95.6 ± 2.22	6.7 ± 0.19[de]	2.01 ± 0.031[de]
TBM100	3.2 ± 0.01	9.4 ± 0.11[e]	95.6 ± 2.22	6.2 ± 0.11[e]	1.91 ± 0.022[e]
TBM100-S	3.2 ± 0.01	9.2 ± 0.16[e]	95.6 ± 2.94	6.0 ± 0.16[e]	1.89 ± 0.029[e]

[1]Values in the same column sharing a common superscript are not significantly different ($P > 0.05$).

[2]SGR = (Ln final weight of fish - Ln initial weight of fish) × 100/days of feeding trial.

higher than that of fish fed the TBM40, TBM60, TBM80, TBM100 and TBM100-S diets, but not significantly ($P > 0.05$) different from that of fish fed the Con diet. SGR of fish fed the TBM20 and TBM30 diets were significantly ($P < 0.05$) higher than that of fish fed the TBM40, TBM60, TBM80, TBM100 and TBM100-S diets, but not significantly ($P > 0.05$) different from that of fish fed the Con and TBM10 diets. No significant difference in weight gain and SGR of fish fed the TBM40 and Con diets in this study indicated that fishmeal could be replaced with TBM up to 40% in the diet without retardation of growth and SGR of juvenile rockfish.

Replacement of fishmeal with the combined animal and plant protein sources in the diets was effective to improve growth of carp (*Cyprinus carpio*), flounder and drum (*Nibea miichthioides*) rather than the single protein source (Hossain and Jauncey 1989; Kikuchi 1999; Guo et al. 2007). Dietary inclusion of soybean meal and cottonseed meal fermented by *Aspergillus oryzae* improved antioxidant activities in the diets and Nile tilapia (*Oreochromis niloticus*), and reduced the toxicity of gossypol in cottonseed meal (Lim and Lee 2011). In addition, supplementation of duckweed (*Lemna polyrhiza*) leaf meal fermented by *Bacillus* sp. into the diets effectively improved weight gain and feed utilization of rohu (Bairagi et al. 2002). A slightly improvement in weight gain of rockfish fed the TBM10, TBM20 and TBM30 diets compared to fish fed the Con diet in this study could be explained by the promising combined substitution effect of fishmeal with animal and plant protein sources and/or fermentation effect of plant (soybean meal) protein source in the diets. Dietary substitution of

60% and 80% fishmeal with the combined seafood (squid byproduct mixture, scallop byproduct, fish soluble, krill meal and squid meal) byproduct and fermented soybean meals in a typical commercial diet did not deteriorate weight gain and feed efficiency of red sea bream, but did for 100% substitution of fishmeal (Kader and Koshio 2012). Zhou et al. (2011) also reported that dietary substitution of fishmeal up to 20% with soybean meal fermented by *Candida utilis* could be made for juvenile black sea bream (*Acanthopagrus schlegeli*) when the 60% fishmeal-based diet was supplied, and proposed that fermented soybean meal contained higher crude protein and lipid content, and lower antinutritional components such as phytic acid, lectin and urease as compared with non-fermented soybean meal.

Feed consumption (g/fish), feed efficiency ratio (FER), protein efficiency ratio (PER) and protein retention (PR) of juvenile rockfish fed the experimental diets substituting fishmeal with TBM were given in Table 4.

Feed consumption of fish was not significantly ($P > 0.05$) different among the experimental diets. However, FER of fish fed the Con, TBM10, TBM20 and TBM30 diets was significantly ($P < 0.05$) higher than that of fish fed the all other diets. PER of fish fed the TBM10, TBM20 and TBM30 diets was significantly ($P < 0.05$) higher than that of fish fed the TBM40, TBM60, TBM80, TBM100 and TBM100-S diets, but not significantly ($P > 0.05$) different from that of fish fed the Con diet. PR of fish fed the TBM10 and TBM20 diets was significantly ($P < 0.05$) higher than that of fish fed the all other diets. And PR of fish fed the Con, TBM30 and

Table 4 Feed consumption (g/fish), feed efficiency ratio (FER), protein efficiency ratio (PER) and protein retention (PR) of juvenile rockfish fed the experimental diets substituting fishmeal with tuna byproduct meal (TBM) for 8 weeks (means of triplicate ± SE)[1]

Experimental diets	Feed consumption	FER[2]	PER[3]	PR[4]
Con	8.8 ± 0.16	0.96 ± 0.002[a]	1.90 ± 0.009[ab]	30.2 ± 0.73[b]
TBM10	8.8 ± 0.10	0.98 ± 0.002[a]	1.98 ± 0.005[a]	37.6 ± 1.81[a]
TBM20	9.2 ± 0.54	0.98 ± 0.003[a]	1.99 ± 0.003[a]	37.7 ± 2.12[a]
TBM30	9.0 ± 0.20	0.97 ± 0.004[a]	1.93 ± 0.010[a]	33.8 ± 0.71[b]
TBM40	8.9 ± 0.63	0.92 ± 0.022[b]	1.77 ± 0.084[c]	30.8 ± 0.75[b]
TBM60	8.0 ± 0.65	0.91 ± 0.016[bc]	1.79 ± 0.058[bc]	26.0 ± 1.19[c]
TBM80	7.6 ± 0.26	0.88 ± 0.003[c]	1.77 ± 0.016[c]	24.8 ± 0.29[cd]
TBM100	7.5 ± 0.27	0.83 ± 0.012[d]	1.64 ± 0.035[d]	22.4 ± 1.18[cd]
TBM100-S	8.2 ± 0.37	0.75 ± 0.009[e]	1.48 ± 0.033[e]	21.2 ± 1.27[d]

[1]Values in the same column sharing a common superscript are not significantly different ($P > 0.05$).

[2]Feed efficiency ratio (FER) = Weight gain of fish/feed consumed.

[3]Protein efficiency ratio (PER) = Weight gain of fish/protein consumed.

[4]Protein retention (PR) = Protein gain of fish/protein consume.

TBM40 diets was significantly (P < 0.05) higher than that of fish fed the TBM60, TBM80, TBM100 and TBM100-S diets.

No difference in FER and PER of fish fed the diets substituting fishmeal up to 30% with TBM compared to those of fish fed the Con diet in this study indicated that fishmeal up to 30% could be replaced with TBM without deterioration of feed utilization by rockfish, resulted from an effective improvement in weight gain of fish. Similarly, feed utilization of fish commonly improved when fishmeal was successfully replaced with the alternative animal and/or plant protein sources in the diets (Kikuchi 1999; Yang et al. 2004; Zhang et al. 2006; Kader and Koshio 2012). No improvement in weight gain, but poorer FER and PER of fish fed the TBM100-S compared to those of fish fed TBM100 in this study indicated that no promising effect of additional substitution of soybean meal with TBM at the whole fishmeal substitution in the diet.

Proximate composition of the whole body of fish at the end of the 8-week feeding trial was presented in Table 5.

Moisture content of fish fed the TBM80, TBM100 and TBM100-S diets was significantly (P < 0.05) higher than that of fish fed the Con, TBM10, TBM20, TBM30 and TBM40 diets, but not significantly (P > 0.05) different from that of fish fed the TBM60 diet. Crude protein content of fish fed the TBM10 and TBM20 diets significantly (P < 0.05) higher than that in fish fed the Con, TBM60, TBM80, TBM100 and TBM100-S diets, but not significantly (P > 0.05) different from that in fish fed the TBM30 and TBM40 diets. Crude lipid content of fish fed the TBM10 and TBM30 diets

Table 5 Proximate composition (% of wet weight) of the whole body of juvenile rockfish at the end of the 8-week feeding trial (means of triplicate ± SE)[1]

Experimental diets	Moisture	Crude protein	Crude lipid	Ash
Con	71.6 ± 1.36^{bc}	16.0 ± 0.22^{bc}	5.1 ± 0.32^{bcd}	5.0 ± 0.20
TBM10	69.4 ± 0.51^{c}	18.3 ± 0.69^{a}	5.7 ± 0.15^{a}	5.0 ± 0.05
TBM20	69.6 ± 0.42^{c}	18.3 ± 0.81^{a}	5.6 ± 0.07^{ab}	5.0 ± 0.12
TBM30	70.2 ± 0.52^{c}	17.2 ± 0.26^{ab}	5.7 ± 0.14^{a}	4.9 ± 0.13
TBM40	70.5 ± 0.57^{c}	17.1 ± 0.46^{ab}	5.3 ± 0.15^{abc}	5.1 ± 0.08
TBM60	72.9 ± 0.43^{ab}	15.1 ± 0.32^{c}	4.9 ± 0.23^{cd}	5.2 ± 0.09
TBM80	74.8 ± 0.80^{a}	14.8 ± 0.18^{c}	4.9 ± 0.15^{cd}	4.8 ± 0.13
TBM100	73.9 ± 0.85^{a}	14.6 ± 0.33^{c}	4.8 ± 0.14^{cd}	5.1 ± 0.38
TBM100-S	74.6 ± 0.38^{a}	15.0 ± 0.40^{c}	4.6 ± 0.15^{d}	5.0 ± 0.05

[1]Values in the same column sharing a common superscript are not significantly different ($P > 0.05$).

was significantly (P < 0.05) higher than that of fish fed the Con, TBM60, TBM80, TBM100 and TBM100-S diets, but not significantly (P > 0.05) different from that of fish fed the TBM20 and TBM40 diets. The lowest crude lipid was observed in fish fed the TBM100-S diet. However, no significant (P > 0.05) difference in ash content was observed among fish fed the experimental diets. An increased TBM substation in the diets tended to elevate moisture content, but lowered crude protein content of fish in this study. Similarly, the whole body of fish was affected by the various alternative animal and/or plant protein sources for fishmeal in the diets (Deng et al. 2006; Lee et al. 2010; Zhou et al. 2011; Kader and Koshio 2012). Zeitler et al. (1984) reported that the whole body composition of fish correlated with fish species, water temperature, weight gain, feeding and diet formulation.

Amino acid profiles of the whole body of rockfish at the end of 8-week feeding trial were not significantly (P > 0.05) different among the experimental diets (Table 6).

Although the differences in the essential amino acids, such as lysine and methionine content in the experimental diets were observed, it would not lead to a significant change in amino acid profiles of the whole body of fish. However, amino acid profiles of the whole body flounder were affected by dietary substitution of fishmeal with soy protein concentrate (Deng et al. 2006).

Table 6 Amino acid profiles of juvenile rockfish at the end of the 8-week feeding trial (% in the whole body)

	Experimental diets								
	Con	TBM10	TBM20	TBM30	TBM40	TBM60	TBM80	TBM100	TBM100-S
Alanine	1.16 ± 0.14	1.18 ± 0.13	1.17 ± 0.09	1.07 ± 0.10	1.05 ± 0.06	1.10 ± 0.09	0.93 ± 0.14	1.10 ± 0.10	1.22 ± 0.08
Arginine	1.05 ± 0.12	1.10 ± 0.10	1.12 ± 0.09	1.25 ± 0.17	1.18 ± 0.15	1.10 ± 0.03	0.99 ± 0.01	1.10 ± 0.01	1.15 ± 0.07
Aspartic acid	1.68 ± 0.20	1.72 ± 0.16	1.67 ± 0.15	1.65 ± 0.05	1.60 ± 0.03	1.74 ± 0.04	1.66 ± 0.10	1.72 ± 0.03	1.73 ± 0.10
Glutamic acid	2.47 ± 0.30	2.65 ± 0.38	2.47 ± 0.20	2.48 ± 0.09	2.41 ± 0.06	2.61 ± 0.09	2.58 ± 0.26	2.60 ± 0.04	2.60 ± 0.12
Glycine	1.47 ± 0.19	1.49 ± 0.21	1.55 ± 0.11	1.56 ± 0.06	1.58 ± 0.16	1.41 ± 0.16	1.15 ± 0.22	1.46 ± 0.13	1.68 ± 0.14
Histidine	0.47 ± 0.06	0.60 ± 0.05	0.60 ± 0.12	0.43 ± 0.04	0.42 ± 0.05	0.49 ± 0.01	0.53 ± 0.09	0.50 ± 0.02	0.58 ± 0.09
Isoleucine	0.76 ± 0.09	0.79 ± 0.06	0.66 ± 0.06	0.67 ± 0.08	0.64 ± 0.07	0.77 ± 0.02	0.70 ± 0.01	0.76 ± 0.02	0.75 ± 0.04
Leucine	1.22 ± 0.15	1.29 ± 0.09	1.25 ± 0.08	1.13 ± 0.08	1.08 ± 0.08	1.23 ± 0.02	1.16 ± 0.02	1.23 ± 0.03	1.25 ± 0.06
Lysine	1.31 ± 0.17	1.44 ± 0.09	1.40 ± 0.11	1.17 ± 0.12	1.12 ± 0.14	1.28 ± 0.05	1.09 ± 0.14	1.25 ± 0.09	1.32 ± 0.08
Phenylalanine	0.68 ± 0.08	0.72 ± 0.06	0.70 ± 0.06	0.57 ± 0.12	0.55 ± 0.11	0.71 ± 0.02	0.71 ± 0.06	0.72 ± 0.01	0.72 ± 0.03
Proline	0.85 ± 0.12	0.82 ± 0.09	0.86 ± 0.05	0.84 ± 0.05	0.87 ± 0.06	0.82 ± 0.04	0.80 ± 0.08	0.86 ± 0.01	0.95 ± 0.08
Serine	0.46 ± 0.02	0.52 ± 0.06	0.53 ± 0.07	0.54 ± 0.01	0.56 ± 0.03	0.46 ± 0.06	0.63 ± 0.10	0.45 ± 0.04	0.50 ± 0.02
Threonine	0.59 ± 0.05	0.64 ± 0.05	0.65 ± 0.06	0.61 ± 0.02	0.61 ± 0.03	0.61 ± 0.04	0.60 ± 0.02	0.59 ± 0.03	0.65 ± 0.03
Tyrosine	0.31 ± 0.02	0.33 ± 0.04	0.37 ± 0.06	0.28 ± 0.05	0.27 ± 0.03	0.32 ± 0.02	0.36 ± 0.03	0.31 ± 0.02	0.33 ± 0.02
Valine	0.85 ± 0.11	0.90 ± 0.07	0.87 ± 0.06	0.75 ± 0.12	0.72 ± 0.10	0.88 ± 0.02	0.77 ± 0.03	0.86 ± 0.01	0.87 ± 0.05

Serum total protein level ranged from 2.1 to 2.6 g/dl, glucose level ranged from 91.7 to 139.7 mg/dl, GOT level ranged from 32.0 to 48.0 IU/l, GPT level ranged from 4.3 to 8.3 IU/l and triglyceride level ranged from 121.7 to 291.0 mg/dl of fish at the end of the 8-week feeding trial was not significantly ($P > 0.05$) different among the experimental diets due to wide variation within the same diets (Table 7).

Similarly, dietary substitution of fishmeal with the various protein sources did not affect serum chemistry of flounder (Lee et al. 2012). Unlike this study, however, dietary alternative protein sources affected plasma constituents of flounder (Kikuchi et al. 1994; Kikuchi 1999). In addition, serum total protein, triglyceride and cholesterol concentration of flounder would be affected by dietary substitution of fishmeal with soybean meal although the whole body composition of fish was not affected (Ye et al. 2011).

Table 7 Serum chemistry of juvenile rockfish at the end of the 8-week feeding trial (means of triplicate ± SE)[1]

Experimental diets	Total protein (g/dl)	Glucose (mg/dl)	GOT (IU/l)	GPT (IU/l)	Triglyceride (mg/dl)
Con	2.3 ± 0.17	95.3 ± 15.34	43.0 ± 8.72	7.0 ± 3.61	144.3 ± 46.88
TBM10	2.5 ± 0.10	112.7 ± 1.45	36.0 ± 4.73	5.3 ± 1.20	291.0 ± 47.71
TBM20	2.3 ± 0.11	91.7 ± 17.61	32.0 ± 7.23	8.0 ± 2.00	190.0 ± 52.52
TBM30	2.5 ± 0.16	139.7 ± 16.48	45.0 ± 7.37	4.3 ± 1.86	202.3 ± 27.34
TBM40	2.6 ± 0.17	134.0 ± 24.25	39.0 ± 6.93	6.0 ± 2.31	224.3 ± 14.72
TBM60	2.3 ± 0.23	103.3 ± 16.76	32.3 ± 9.26	5.3 ± 2.60	128.7 ± 27.72
TBM80	2.3 ± 0.04	139.3 ± 44.43	47.3 ± 11.29	8.3 ± 1.20	176.7 ± 21.67
TBM100	2.1 ± 0.05	92.7 ± 12.67	48.0 ± 11.93	4.3 ± 2.33	138.7 ± 28.82
TBM100-S	2.3 ± 0.34	132.0 ± 11.27	47.7 ± 16.23	6.0 ± 3.06	121.7 ± 27.17

[1]Values in the same column sharing a common superscript are not significantly different ($P > 0.05$).

II. Conclusion

Effect of dietary substitution of fishmeal with tuna byproduct meal (TBM) in the diet on growth, body composition, serum chemistry and amino acid profiles of juvenile Korean rockfish (*Sebastes schlegeli*) was determined.

Weight gain of fish fed the TBM10, TBM20 and TBM30 diets were higher than that of fish fed the TBM40, TBM60, TBM80, TBM100 and TBM100-S diets, but not different from that of fish fed the Con diet. Specific growth rate (SGR) of fish fed the TBM20 and TBM30 diets were higher than that of fish fed the TBM40, TBM60, TBM80, TBM100 and TBM100-S diets, but not different from that of fish fed the Con and TBM10 diets. Feed efficiency ratio (FER) of fish fed the Con, TBM10, TBM20 and TBM30 diets was higher than that of fish fed the all other diets. Protein efficiency ratio (PER) of fish fed the TBM10, TBM20 and TBM30 diets was higher than that of fish fed the TBM40, TBM60, TBM80, TBM100 and TBM100-S diets, but not different from that of fish fed the Con diet.

Based on these results, it can be concluded that substitution of fishmeal with TBM up to 40 and 30% in the diets could be made without detrimental effect on growth (SGR) and feed utilization (FER and PER) of juvenile Korean rockfish, respectively.

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