

## EFFECTS OF REPLACEMENT OF FISH MEAL WITH POULTRY BY-PRODUCT MEAL ON GROWTH PERFORMANCE IN PRACTICAL DIETS FOR RAINBOW TROUT, *Onchorynchus mykiss*\*

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### Abstract

A feeding trial was conducted to evaluate the potential of replacing fish meal (FM) with poultry by-product meal (PBM) in practical diets for rainbow trout (*Onchorynchus mykiss*, Walbaum, 1972). Five isocaloric (5600 kcal GE/ kg) and isonitrogenous (45% CP) diets including PBM at the levels of 0 (Control), 10 (PBM10), 20 (PBM20), 30 (PBM30) and 40% (PBM40), respectively were formulated. and had a lipid content of 21% approximately. Seven hundred and fifty rainbow trout (initial body weight, 34.50±0.43 g) were randomly assigned to fifteen 500-l fiber-glass tanks. Each dietary treatment was tested in triplicate groups of 50 fish per tank arranged in a completely randomized design. Fish were fed three times (08:30, 12:00 and 15:30 h) daily for 12 weeks. Final weight (W<sub>f</sub>) and body weight gain (BWG) of groups fed PBM up to 20% were similar to the control, while significantly lower values were obtained from the groups receiving higher levels of PBM (p<0.05). Hepatosomatic index, viscerasomatic index and dressing percentage data indicated no significant differences among the treatments; however, condition factor and specific growth ratio of PBM30 and PBM40 groups were lower than those of the control diet (p < 0.05). Fish fed PBM40 showed lower feed intake and the groups containing PBM 20% and higher levels had lower feed conversion than the control group (p < 0.05). Results of the study indicated that dietary PBM could be included up to 20% in rainbow trout diets without any significant impairment in the performance criteria evaluated.

**Key Words:** Rainbow trout, poultry by-product meal, growth performance

### Gökkuşığı Alabalığı Rasyonlarında Tavuk Mezbaha Artıkları Ununun Balık Unu Yerine Kullanılmasının Büyüme Performansı Üzerine Etkileri

#### Özet

Bu çalışma Gökkuşığı alabalığı (*Onchorynchus mykiss*, Walbaum, 1972) rasyonlarında balık unu (BU) yerine tavuk mezbaha artıkları unu (TMAU)'nun kullanılma olanaklarını belirlemek amacıyla yapılmıştır. Denemede % 21 yağ içeren, 5600 kcal/kg brüt enerji ve %45 ham proteine sahip, isokalorik ve isonitrojenik karma yemler kullanılmıştır. Karma yemlerde sırasıyla, %0 (Kontrol), 10 (TMAU10), 20(TMAU20), 30 (TMAU30) ve 40 (TMAU40) oranlarında TMAU kullanılmıştır. Deneme başı canlı ağırlıkları 34.50±0.43 g olan 750 gökkuşığı alabalığı, tesadüfen, 500 l kapasiteli 15 fiber-glas tanka, her birine 50 adet balık ve her mumeleye de 3 tank düşecek şekilde, 5 muamele grubuna dağıtılmışlardır. 12 haftalık deneme süresince balıklar günde 3 kez (saat 08:30, 12:00 ve 15:30'da) yemlenmişlerdir. %20 TMAU ile beslenen grupta deneme sonu canlı ağırlıkları ile canlı ağırlık artışları bakımından kontrol grubu ile benzer sonuçlar elde edilirken, daha yüksek seviyelerde TMAU kullanılan gruplarda bu özellikler bakımından önemli derecede daha düşük değerler elde edilmiştir (p < 0.05). Hepatosomatik indeks, viskerosomatik indeks ve karkas randımanları dikkate alındığında gruplar arasında önemli farklılıklar çıkmazken, TMAU30 ve TMAU40 gruplarında, kontrol grubuna göre daha düşük kondüsyon faktörü ve spesifik büyüme oranı elde edilmiştir (p < 0.05). Yem tüketimi bakımından TMAU40 grubu ile, yemden yararlanma bakımından %20 ve üzerinde TMAU içeren gruplar, kontrol grubundan göre daha düşük ortalamalara sahip olmuşlardır (p < 0.05). Bu çalışmadan elde edilen sonuçlara dayanarak, TMAU'nun alabalık rasyonlarına %20 oranında katılabileceğini söylemek olasıdır.

**Anahtar Kelimeler:** Gökkuşığı Alabalığı, Tavuk Mezbaha Artıkları Unu, Büyüme Performansı

### 1. Introduction

Most high-value species of fish raised by aquaculture are carnivores requiring feeds containing 400 g/kg or more protein, generally supplied by fish meal (FM)

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(Hardy, 1996). The price of FM will substantially increase due to the increasing demand and decreasing production. One approach to reduce feed cost is partially or totally replace expensive FM protein with less expensive other animal protein sources.

Poultry by-product meal (PBM) is a by-product of the poultry industry consisting of ground, dry-rendered parts of slaughtered poultry such as heads, feet, undeveloped eggs, intestines and feathers. Steffens (1994) showed that PBM is suitable as a partial or complete replacement in diet for rainbow trout; however, complete substitution required amino acid supplementation, principally lysine and methionine. Similarly, Alexis et al. (1985) using a diet containing 25 % PBM obtained satisfactory performance data in trout. Pokorny (1982) reported that diets including up to 40 % PBM and 10 % poultry fat could successfully be used as a substitute for FM in trout. However, Lee et al. (2002) found juvenile rainbow trout fed animal protein mixture, consisting equal amounts of meat and bone meal, blood meal, PBM and feather meal, showed significantly lower growth performance and feed conversion than the fish fed a FM based diet.

PBM is high in ether extract (EE), ash and indigestible components (feathers, etc.), resulting in reduced digestibility (Robinson and Li, 1996). Thus, PBM varies in nutritional value and composition depending on the processing and the materials included in the meal (Nengas et al., 1999). However, if suitable raw ingredients are chosen and then properly processed, a high-quality protein feed can be achieved. Kureshy et al. (2000) reported that flash-dried PBM and enzyme-digested diets for juvenile red drum did not show any significant difference in percent weight gain and feed conversion ratio than a FM and soybean meal based diet. Davis and Arnold (2000) observed that replacement of FM with co-extruded soybean and PBM resulted in equivalent values for final weight, percent weight gain and feed conversion ratio for Pacific White shrimp. Although, Webster et al. (2000) determined that feed conversion ratio of sunshine bass fed PBM (%30) and soybean meal (%30) based diet; was significantly

higher when compared to fish fed a FM (%30) and soybean meal (%30) based diet, percentage weight gain, specific growth rate, feed intake, hepatosomatic index were not significantly different among treatments of the same study. Abdel-Warith et al. (2001) proved that PBM may successfully replace up to 40% of the protein component in practical feeds for African catfish. However, these researchers reported growth performance, feed intake, feed conversion ratio and specific growth rate were all depressed for catfish fed the highest levels of PBM diets compared to the FM group.

The main purpose of this experiment was to evaluate the dietary inclusion level of PBM as a replacement for FM in rainbow trout (*Oncorhynchus mykiss*, Walbaum, 1972) diets in terms of growth performance and feed conversion ratio.

## 2. Materials and Methods

The nutrient composition of the feed ingredients used in the study are given in Table 1. Five isocaloric (5600 kcal GE/kg) and isonitrogenous (45% CP) diets including PBM at the levels of 0 (Control), 10 (PBM10), 20 (PBM20), 30 (PBM30) and 40% (PBM40), respectively were formulated (Table 2) to meet the requirements (NRC, 1993) (Table 3). Amino acid contents of FM, PBM and the experimental diets are given in Table 4. PBM used in the study was produced by heating at 150-200 °C and 2.5 atmospheric pressure for about 10 h (Abalıoğlu Feed Plant, Denizli, Turkey). The powdered ingredients except fish oil were mixed into the ground feed mixture in an horizontal mixer for 5 min and pelleted (3 mm in diameter). Fish oil supplemented to cover the pellets in order to prevent them from dispersion in the water. The diets were stored at regular ambient temperature in airtight bags.

The feeding experiment was performed at the Kepez Aquaculture Center (Antalya-Turkey) with rainbow trouts (*Oncorhynchus mykiss*, Walbaum, 1972) averaging 34.5±0.43 g initial weight. Prior to the trial fish were fed with the control diet for two weeks for adaptation to the

Table 1. Nutrient composition of feed ingredients used in the study (as fed basis, %)

Ingredients	DM <sup>1</sup>	CP <sup>1</sup>	EE <sup>1</sup>	CA <sup>1</sup>	CF <sup>1</sup>
Fish meal	94.33	68.50	10.19	10.94	0.96
Poultry by-product	94.84	57.75	28.93	11.54	1.26
Corn gluten meal	93.94	71.75	5.33	1.74	3.86
Soybean meal	92.55	47.62	3.69	7.48	5.85
Wheat middlings	91.95	15.87	6.15	3.49	11.12

<sup>1</sup> DM=Dry matter; CP=Crude protein; EE=Ether extracts; CA=Crude ash; CF=Crude fiber

Table 2. Ingredient composition of experimental diets (%)

	Diets				
	Control	PBM10	PBM20	PBM30	PBM40
Fish meal	46.83	38.22	29.61	21.01	12.40
Poultry by-product	0	10.00	20.00	30.00	40.00
Corn gluten meal	10.00	10.00	10.00	10.00	10.00
Soybean meal	5.00	5.00	5.00	5.00	5.00
Wheat middling	21.21	21.97	22.73	23.50	24.26
Fish oil	15.31	13.16	11.01	8.84	6.69
Vitamin mix. <sup>1</sup>	0.50	0.50	0.50	0.50	0.50
Mineral mix. <sup>2</sup>	0.20	0.20	0.20	0.20	0.20
Choline chloride	0.15	0.15	0.15	0.15	0.15
Pellet binder <sup>3</sup>	0.30	0.30	0.30	0.30	0.30
Cr <sub>2</sub> O <sub>3</sub> <sup>4</sup>	0.50	0.50	0.50	0.50	0.50

<sup>1</sup> Per kg mix: 4 000 000 IU vitamin A, 480 000 IU vitamin D<sub>3</sub>, 40 000 mg vitamin E, 2400 mg vitamin K<sub>3</sub>, 4 000 mg vitamin B<sub>1</sub>, 6 000 mg vitamin B<sub>2</sub>, 40 000 mg niacin, 10 000 mg Ca-D-pantothenate, 4 000 mg vitamin B<sub>6</sub>, 10 mg vitamin B<sub>12</sub>, 100 mg D-biotin, 1 200 mg folic acid, 40 000 mg vitamin C ve 60 000 mg inositol.

<sup>2</sup> Per kg mix: 23 750 mg Mn, 75 000 mg Zn, 5 000 mg Zn, 2 000 mg Co, 2 750 mg I, 100 mg Se, 200 000 mg.

<sup>3</sup> Calcium lignosulfonate

<sup>4</sup> Cr<sub>2</sub>O<sub>3</sub> was used as a inert marker for determination of the apparent digestibility in the other part of this project, SIGMA Product No: 34,296-3

Table 3. Chemical composition of experimental diets (as dry matter basis)

Prox. composition (g kg <sup>-1</sup> DM)	Diets				
	Control	PBM10	PBM20	PBM30	PBM40
DM	93.66	94.06	94.14	93.99	93.89
CP	45.11	45.02	43.78	43.24	43.09
EE	21.64	21.36	21.98	20.53	20.07
CA <sup>1</sup>	7.46	7.60	7.64	7.60	7.98
Carbohydrate (NFE) <sup>2</sup>	25.32	25.60	26.16	28.15	28.43
Gross energy (kcal kg <sup>-1</sup> )	5658	5637	5649	5566	5524
Cr <sub>2</sub> O <sub>3</sub> <sup>3</sup>	0.459	0.425	0.446	0.479	0.447

<sup>1</sup> Except Cr<sub>2</sub>O<sub>3</sub>

<sup>2</sup> Determined by [ NFE = 100-(% water + CP + % EE + % CA+% CF)].

experimental conditions. The trial was conducted in 500-l flow-trough circular fiberglass tanks, supplied with well water at a flow rate of 15-l min<sup>-1</sup>. Fish were randomly distributed into the groups replicated 3 times and each replicate had so fish of 50 (3 groups per treatment). Average water quality parameters measured such as temperature, dissolved oxygen (daily) and pH (once in each three days), were 14.2±0.9°C, 8.8±0.3 mg l<sup>-1</sup> and 7.5±0.2, respectively. Solid particles were removed

from the water through a sand filter. The fish were subjected to natural photoperiod over the trial. Each diet was fed to the groups to apparent satiation three times a day (08:30, 12:00 and 15:30 h) except the weighing days in which only the 08:30 meal was offered. The tanks were siphoned daily after the last meal. All the fish in each group were weighed together to calculated growth rates, biweekly.

Dry matter (DM), crude ash (CA), crude protein (CP), ether extracts (EE),

Table 4. Some essential and nonessential amino acid compositions of FM, PBM and test diets (%)

Amino acids <sup>1</sup>	FM	PBM	Control	PBM10	PBM20	PBM30	PBM40
Arginine	5.09	5.08	5.21	5.31	5.15	5.04	5.10
Histidine	2.62	0.96	2.54	2.32	2.09	1.86	1.76
Isoleucine	4.06	3.66	4.37	4.18	4.09	4.00	4.03
Leucine	6.26	5.79	7.69	7.52	7.33	7.44	7.42
Lysine	7.24	3.03	6.87	5.86	5.32	4.73	4.36
Methionine	2.60	0.87	2.59	2.37	2.05	1.80	1.90
Cystine <sup>2</sup>	0.60	0.84	0.49	0.52	0.55	0.59	0.62
Tryptophan <sup>2</sup>	0.75	0.46	0.47	0.45	0.44	0.42	0.40
Phenylalanine+tyrosine	5.82	4.90	4.16	3.72	3.45	3.54	4.22
Threonine	4.06	3.72	3.79	3.99	3.79	3.48	3.61
Valine	4.58	5.06	4.83	4.81	4.74	4.97	4.79
Aspartic acid	8.91	6.38	8.12	8.40	8.22	7.16	7.15
Glutamic acid	7.77	6.75	8.96	9.30	9.19	8.34	8.80
Glycine	4.29	7.45	4.22	4.60	4.97	5.11	5.23
Serine	5.07	8.98	5.59	6.44	6.07	6.70	7.33
Proline	3.73	7.31	2.40	3.68	4.30	3.17	5.32

<sup>1</sup> Determined by chemical analysis except tryptophan and cystine value (AOAC, 1990).

<sup>2</sup> NRC (1993).

crude fiber (CF) and carbohydrate in terms of N free extracts (NFE) of all ingredients and diets were determined according to the standard methods of AOAC (1984). Gross energy was calculated using the multiplier factors of 5.65, 9.45 and 4.2 kcal g<sup>-1</sup> for CP, EE and NFE, respectively (Henken et al., 1986). Amino acid contents of FM, PBM and the diets were determined by using Eppendorf LC 3000 Amino Acid Analyzer (AOAC, 1990).

At the end of the experiment all fish were weighed to calculate body weight gain [BWG = final body weight – initial body weight], feed conversion ratio [FCR = dry feed consumed (g) / wet weight gain (g)] and specific growth rate [SGR = (ln final body weight – ln initial body weight) x 100 / time (day)]. Five fish randomly selected from each tank were anaesthetized with fenoxo ethanol to evaluate condition factor [CoF = (wet body weight / total body length, cm) x 100], dressing percentage (DP = eviscerated fish weight, g / body weight, g), hepatosomatic index [HSI = (wet liver weight/wet body weight) x 100] and viscerosomatic index [VSI = (visceral weight, g / body weight, g) x 100].

Each diet was offered to three groups in a completely randomized design. Differences among the dietary treatments were tested by ANOVA (Düzgüneş et al.

1987) and means were compared using Duncan's multiple range tests (Duncan 1955).

### 3. Results and Discussions

At the end of the 12-week trial, growth of the rainbow trout receiving the control diet was similar to the fish offered a high quality practical diet. Table 5 summarizes the mean initial ( $W_i$ ) and final ( $W_f$ ) weights (g fish<sup>-1</sup>), BWG, feed intake (g fish<sup>-1</sup>) (FI), FCR, SGR, CoF, DP, HSI, VSI, and cost analysis data obtained from the groups fed experimental diets.

Fishes fed PBM10 and PBM20 diets had similar final body weight and weight gain to the control group (Table 5). However, increasing the level of PBM up to 30 and 40 % significantly reduced same parameters ( $P < 0.05$ ). Similarly, Alexis et al. (1985) using a diet containing 25 % PBM obtained satisfactory performance in trout. Pokorny (1982) found that diets including up to 40 % PBM and 10 % poultry fat were useful as in substituting a FM diet in trout. Gouveia (1992) reported that the optimum inclusion level of PBM replacement was about 80 % of the protein for rainbow trout. The author used a FM lower in protein and certain essential amino acids (EAA) than PBM. Lee et al (2001) concluded that a

Table 5. Growth performance, FCR, DP, HIS, VSI of fish fed the test diets and costs analyses of the diets<sup>1</sup>

	Diets				
	Control	PBM10	PBM20	PBM30	PBM40
W <sub>i</sub>	34.24±0.37	34.17±0.20	34.88±0.14	34.35±0.24	34.59±0.12
W <sub>f</sub>	174.52±6.20 <sup>a</sup>	174.30±2.65 <sup>a</sup>	171.22±4.40 <sup>a</sup>	152.81±1.64 <sup>b</sup>	130.50±4.08 <sup>c</sup>
BWG	140.28±6.53 <sup>a</sup>	140.14±2.82 <sup>a</sup>	136.34±4.42 <sup>a</sup>	118.46±1.51 <sup>b</sup>	95.90±4.00 <sup>c</sup>
FI	122.83±5.88 <sup>a</sup>	123.48±4.69 <sup>a</sup>	125.90±5.83 <sup>a</sup>	117.90±0.57 <sup>ab</sup>	106.52±3.56 <sup>b</sup>
SGR	1.94±0.06 <sup>a</sup>	1.94±0.03 <sup>a</sup>	1.89±0.03 <sup>a</sup>	1.78±0.01 <sup>b</sup>	1.58±0.04 <sup>c</sup>
FCR	0.87±0.01 <sup>a</sup>	0.88±0.02 <sup>a</sup>	0.92±0.02 <sup>b</sup>	1.00±0.07 <sup>c</sup>	1.11±0.01 <sup>d</sup>
CoF	1.39±0.02 <sup>a</sup>	1.38±0.02 <sup>ab</sup>	1.33±0.02 <sup>abc</sup>	1.32±0.02 <sup>bc</sup>	1.28±0.03 <sup>c</sup>
DP, %	85.20±0.39	85.20±0.88	84.98±0.20	84.38±0.36	84.29±0.52
HSI, %	1.67±0.08	1.51±0.10	1.58±0.10	1.62±0.08	1.75±0.04
VSI, %	13.84±0.43	13.92±0.85	13.74±0.16	14.74±0.26	14.45±0.30
USD/kg feed <sup>2</sup>	0.641	0.573	0.506	0.438	0.370
USD/kg BWG <sup>2</sup>	0.557	0.504	0.465	0.438	0.411

<sup>1</sup> Mean within a row having different letters are significantly different (p<0.05)

<sup>2</sup> Cost analyses were calculated on the prices at the date of 28<sup>th</sup> Sept. 2001.

mixture of animal by-products (25% meat and bone meal, 24.5% leather meal, 20% squid liver powder, 15% feather meal, 7.5% blood meal, 7.5% PBM, and 0.25% each of methionine and lysine) could be replaced up to 28% of FM protein in diets without any adverse effect on growth of juvenile rainbow trouts.

SGR and CoF followed trends similar to W<sub>f</sub> and BWG, being significantly lower in PBM30 and PBM40 groups compared to the others (P<0.05); although, Gouveia (1992) reported that high level of PBM in rainbow trout diets caused no significant difference in SGR and CoF.

HSI, VSI and DP data in this study also indicated no significant differences among the treatments. This result is in accordance with the findings of Alexis et al. (1985) and Webster et al. (2000) who reported high PBM levels in rainbow trout diets did not affect HSI, while conflicts with the findings of Zoccarato et al. (1996) who reported that high PBM levels could cause increased HSI and VSI. Furthermore, high diet carbohydrate level could cause enlarged liver. However, data of HSI of the groups were not different since NFE contents of the diets were close to each other.

Traits relating to the growth performance have been depressed in fishes fed PBM30 and PBM40, compared to the others. The lower growth rates in these two groups could be attributed to insufficient amino acid, and also high feather,

connective tissue and skin contents which are considered to be difficult to digest for the fish. When the results compared to the apparent digestibility data driven (Erturk and Sevgili, 2003), this suggestion can be explained by the lower digestibility coefficients of the same diets.

The essential and nonessential amino acid profiles of FM, PBM and experimental diets were compared in Table 4. When they were compared with NRC (1993) requirements, methionine appeared as the first limiting EAA in PBM and followed by histidine, lysine and phenylalanine+tyrosine. In PBM including diets the first limiting EAA methionine followed by same amino acids in a similar order (except phenylalanine is exceeding the requirement). In previous studies conducted with rainbow trout with various EAAs, leucine (Gouveia, 1992), lysine and isoleucine (Zoccarato et al., 1996), methionine and phenylalanine (Dong et al., 1993) have been marked as first limiting amino acids. Luzzana et al. (1998) claimed that arginine requirement of Pacific salmon is the highest among the other salmonid species. It was found, however, in a study aiming to determine the order of limiting EAAs in a blend of 4 PBMs produced in USA for chicks that cystine was the first limiting and the least digestible (46%) amino acid (Wang and Persons, 1998). According to the Sadiku and Jauncey (1998) the least available amino acid in soybean flour-PBM blended diets for

tilapia and sharp-toothed catfish was cystine. Even though cystine contents of PBM and test diets were not determined, sulphur amino acid content of PBM might have a more important role than the others in the current study.

Feed intake was found to be the lowest in PBM40 group ( $P < 0.05$ ). This result supports Fowler (1991) and Quartararo et al. (1998) who reported that high PBM levels could cause reduced palatability for Chinook salmon and Australian snapper; however, contradicted with the findings of Davis and Arnold (2000) who reported that PBM didn't indicate any apparent palatability problem. Similarly, Shepherd (1998) was not able to observe any serious problem related to palatability of rendered products in fish diets. Yamamoto et al., (2000), claimed that rainbow trout preferred a balanced amino acid diet to an imbalanced or a protein free diet regardless to the protein level. The fact that deficiency of any EAA usually reduces palatability of the diet (Moon and Gatlin, 1994; Abdel-Warith et al., 2001). Thus, increasing the level of PBM in the diet up to 40%, possibly reduced feed intake in the present study.

FCR data suggested that PBM can be used as the highest level of only 10% in rainbow trout diets (Table 5). In fact, FCRs of fish fed PBM20, PBM30, PBM40 diets were significantly higher (0.92, 1.00, 1.11,) compared to the control and PBM10 diets (0.87, 0.88). These results confirm the findings of Webster et al. (2000) and Lee et al. (2002) who reported replacement of FM with PBM resulted in significantly higher FCR ( $P < 0.05$ ). However, Davis and Arnold (2000) found that replacement of FM with co-extruded soybean-PBM mixture resulted in equivalent FCR value. Similarly, Steffens (1994) showed that

Reasons for the differences between the results of some previous reports and this experiment could be attributed to variations in raw materials, and different rendering processes. PBM is suitable as a partial or complete replacement in rainbow trout diets; but, a complete substitution required amino acid supplementation, principally lysine and methionine. However, in this study, increasing PBM caused some decrease in

energy and protein contents of the 4 and 5<sup>th</sup> groups. Similarly, the rich fat content of PBM changed fish oil levels of diets causing decrease in omega-3 fatty acids such as eicosapentaenoic (EPA) and docosahexanoic (DHA). Thus, the reduced energy, protein and omega-3 fatty acids content of diets should be taken into consideration in evaluating the significantly lowered performance data obtained particularly from these two groups.

In brief, this study indicated that PBM might be included in rainbow trout diets up to 20% without any significant impairment on growth. It was also determined that when 20% PBM used, feed cost per kg of BWG was lower 16.5 percent than the control.

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