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Effects of the Feeds Containing Different Plant Protein Sources on Growth Performance and Body Composition of Rainbow Trout (*Oncorhynchus mykiss*, W.)

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Abstract

In this study, a total of 720 rainbow trout (*Oncorhyncus mykiss*) having 50.08±0.19 g of average initial weight were used to compare of usage of the soybean meal, sunflower meal, canola meal and cottonseed meal as the best plant protein source. Four different feeds which contain soybean meal (SBM), canola meal (CM), sunflower meal (SFM) and cotton seed meal were used, respectively. Four trial groups were tested in triplicate during 12 weeks. Experimental diets were prepared as isonitrogenous (42% CP) and isocaloric (13.9 MJ DE/kg feed). At the end of the study, it was observed that there was no significantly difference among the groups for average feed consumption, condition factor and survival rate, however, average live weight, live weight gain, feed conversion ratio (FCR), protein efficiency ratio (PER) and specific growth rate (SGR) were found statistically significant ($P<0.05$). Moreover, the difference in the composition of carcass and total body between the groups were found statistically significant ($P<0.05$). Consequently, it is concluded that soybean meal, canola meal and sunflower meal can be used as a part of fish meal, however cottonseed meal can be used as only smolt or older trout feeds if it is cheap and has good quality.

Keywords: Rainbow trout, soybean meal, canola meal, sunflower meal, cotton seed meal, growth performance, body composition.

Gökkuşağı Alabalıklarının (*Oncorhynchus mykiss*) Besi Performansı ve Vücut Bileşimi Üzerinde Farklı Protein Kaynakları İçeren Yemlerin Etkisi

Özet

Araştırmada; soya, ayçiçeği tohumu, küspesi, kanola ve pamuk tohumu küspelerinden hangisinin en iyi bitkisel protein kaynağı olduğunun karşılaştırılması için başlangıç ortalama canlı ağırlığı 50,08±0,19 g olan 720 adet gökkuşağı alabalığı (*Oncorhynchus mykiss*, W.) kullanılmıştır. Sırasıyla soya küspesi (SFK), ayçiçeği tohumu küspesi (ATK), kanola küspesi (KK) ve pamuk tohumu küspesi (PTK) içeren 4 yem kullanılmıştır. Deneme, her biri 3 paralelli olmak üzere 4 deneme grubu ile 12 hafta boyunca sürmüştür. Tüm deneme yemleri izonitrojenik (%42 HP) ve izokalorik (13.9 MJ SE/kg yem) olarak hazırlanmıştır. Deneme sonunda gruplar arasında yem tüketimi, kondisyon faktörü ve yaşama gücü bakımından farklılık gözlenmezken ($P>0,05$), ortalama canlı ağırlık, ortalama canlı ağırlık kazancı, yemden yararlanma oranı (YYO), proteinden yararlanma oranı (PYO) ve özel büyüme oranı (ÖBO) bakımından saptanan farklılık önemli bulunmuştur ($P<0,05$). Ayrıca gerek karkas gerekse de toplam vücut bileşimi bakımından gruplar arasında gözlemlenen farklılık da önemli olmuştur ($P<0,05$). Sonuç olarak, soya, ayçiçeği tohumu ve kanola küspesinin balık ununun bir kısmı yerine kullanılabileceğine, ancak pamuk tohumu küspesinin ucuz ve kaliteli olması halinde daha büyük balıkların yemlerinde kullanılabileceğine karar verilmiştir.

Anahtar Kelimeler: Gökkuşağı alabalığı, soya küspesi, ayçiçeği tohumu küspesi, kanola küspesi, pamuk tohumu küspesi, besi performansı, vücut bileşimi.

Introduction

The production of fish meal used in traditional fish feeds has decreased gradually, whereas its share in aquaculture has increased. Total amount of fish meal used in aquaculture in 2002 was 46%, it is expected that using fish meal demand in 2010 will be 56% (Miles and Chapman, 2006). Recent studies focus on alternative protein sources for fish meal because of its usage problems (high price, variable quality, availability etc.). Therefore, the most important feedstuffs as alternative protein sources are

oil industry by-products.

Soybean (SBM) and its several by-products are used in aquaculture feeds largely and frequently. Cho *et al.* (1974), found that decreased fish meal in young rainbow trout (*Oncorhynchus mykiss*) feeds did not affect the growth performance and feed conversion ratio negatively where fish meal ratio are decreased from 35% to 16% and soybean meal ratio are increased from 10 to 39%. Refstie *et al.* (1997) conducted a study on the adaptation of rainbow trout on the soybean meal in their feeds. They found that the growth was 1.4 times slower in 28 days for the trout fed with the diet containing 60% soybean, after which adaptation has occurred and the difference in growth was not significant after this period. These results indicate that given an adaptation period, trout previously adapted to fish meal diets consume and grow well on diets containing high levels of soybean meal.

Canola meal (CM) has been used as a feedstuff in salmon and trout feeds over 20 years. It can be used up to 20% in salmon feeds without any negative effects of glikozinolate on the growth performance and thyroid glands (Satoh *et al.,* 1998). However, Hardy and Sullivan (1983) had observed that even though there was no significant difference on the average live weight and feed conversion ratio (FCR), there were an increase in secretion of thyroid gland and hyperplasia in rainbow trout fed with a diet containing 0, 10, 15, 20% canola meal through 108 days experiment. The poor dry matter digestibility of canola meal is attributed to content of high level cellulose and complex carbohydrates.

Crude protein content of dehulled sunflower meal is about 40% and if shortage of lysine can be fulfilled, it can be used 60-70% in fish meal (Tacon *et al.,* 1984). Navneet *et al*. (2006) reported that 27% sunflower meal can be used in smolt Atlantic salmon feeds without any negative effect. Martinez (1986) found that a reduction in the dietary brown fish meal concentration and an increase in sunflower meal concentration have resulted in no loss in growth performance and diet utilization efficiency.

In addition to using low gossypol containing cotton seed in agriculture, cotton seed meal (CSM) is used more and more in fish feeds in recent years. Cheng and Hardy (2002) replaced to fish meal with cotton seed meal at 5, 10, 15 and 20% in trout feed contained 25% fish meal and 0% cottonseed meal and reported that cotton seed meal can be replaced to fish meal at 10% easily.

Consequently, it was investigated to compare of the effects of using soybean, canola, sunflower and cotton seed meal as a part of fish meal on growth performance, feed consumption and body composition of rainbow trout in this study, since these meals were abundant in Turkey. Although there are large numbers of studies about the topic there are not any studies comparing more than two different feedstuffs.

Materials and Methods

Fish and Diets

This study have performed in a commercial trout farm with a total of 720 rainbow trout (*Oncorhyncus mykiss,* W.) (initial mean body weight, 50.08±0.19 g) was stocked into twelve cages with 60 fish per cage. Young fish were obtained from another fish farm. Fish were assigned randomly to these four diets. The cages made by plastic profile in 1×1×1 m and encircled with knotless net with 2 cm mesh size were settled into same channel type pond with 80 cm deep. Water temperature was 15.3-16.7°C and pH was 7.42-7.54 (WTW, PH/OXI 340i/SET, Germany) and water flow rate was 10 L/s during the trial.

All feeds used in the experiment contained about 35% fish meal and four different feedstuffs, soybean meal (SBM), canola meal (CM), sunflower meal (SFM) and cotton seed meal (CSM), are used as plant protein sources. Experimental diets were prepared as isonitrogenous (42% CP) and isocaloric (13.9 MJ DE/kg feed, Table 1). Fish oil was added to all the diets to increase the palatability. The whole feedstuffs are grinded to medium fine size (0.3 mm) before pelleting. Pellet size was 3 mm diameter and 6 mm in length. Table 2 shows nutritional composition of these four trial feeds.

Feeding Trial

Each feed was given to four trial groups tested in triplicate. The trial continued for 84 days at biweekly periods and ended when all fish reached over 200 g weight. Every two weeks, the whole fish starved fish before 24 h, was taken from each cage then weighed as a group. Fish was fed twice a day, at 8 o'clock in the morning and at 6 o'clock in evening according to free feeding (*Ad libitum*) method. It is supposed that all given feeds was consumed by fish. The amount of consumed feed was calculated by determining weight of lacking total feed.

Live weight gain was determined by fish final weight-initial weight; feed conversion ratio was calculated as feed consumed/weight gain. Number of dead fish was taken into account before feed conversion ratio (FCR) was calculated. Fish weight gain, gain (%), feed conversion rate (FCR), survival rate, protein efficiency rate (PER) specific growth rate (SGR), condition factor (CF), fat deposition rate (FDR) and protein growth rate (PGR) were estimated.

PER=(Average Live Weight Gain In A Definited Period, g)/(Consumed Crude Protein With The Diet In A Definited Period, g) (Hepher, 1989)

$SGR= [(\ln W_t-\ln W_0)/(t-t_0)]"\times 100$ " (Hepher, 1989)

$CF = W/L^3$ (Brown, 1957)

FDR= [(ln (Final body lipid)-ln (Initial body

Table 1. Composition of experimental fish diets

	Test Diets, %			
Feedstuffs	SBM	CM	SFM	CSM
Fish Meal	34.4	34.4	34.4	34.4
Bone-Meat Meal	5.0	5.0	5.0	5.0
Corn Gluten	9.0	9.0	9.0	9.0
Soybean Meal	11.3	-	-	-
Canola Meal	-	16.4	-	-
Sunflower Meal	-	-	23.0	-
Cotton Seed Meal	-	-	-	25.0
Wheat Bran	30.3	25.2	18.6	15.6
Fish Oil	7.6	7.6	7.6	7.6
Cotton Seed Oil	-	-	-	1.0
Vitamin Complex [a]	0.5	0.5	0.5	0.5
Mineral Complex [b]	0.1	0.1	0.1	0.1
Dicalcium Phosphate	0.3	0.3	0.3	0.3
DL-Methionine[c]	0.6	0.6	0.6	0.6
L-Lysine[c]	0.3	0.3	0.3	0.3
Lignobond [d]	0.5	0.5	0.5	0.5
Butil Hydroxi Toluen [e]	0.1	0.1	0.1	0.1
TOTAL, g	100.0	100.0	100.0	100.0

[a] Vitamin Premix (mg/kg or IU/kg of dry matter): thiamine 40 mg, riboflavin 50 mg, pyridoxine 40 mg, calcium pantothenate 117 mg, nicotinic acid 200 mg, biotin 1 mg, folic acid 10 mg, cyanocobalamin 0.5 mg, choline chloride 2700 mg, inositol 600 mg, ascorbic acid 5000 mg, alpha tocopherol 300 mg, menadione 20 mg, cholecalciferol 2000 IU, retinol acetate 5000 IU and α-cellulose was used as a carrier.
.[b] Mineral Premix (g/kg of dry matter): calcium orthophosphate 1.80 g, calcium carbonate 5 g, ferrous sulphate 1.7 g, magnesium sulphate 1.8 g, potassium phosphate 3.0 g, sodium phosphate 1 g, aluminium sulphate 0.02 g, zinc sulphate 0.24 g, copper sulphate 0.20 g, manganese sulphate 0.08 g, potassium iodate 0.02 g. α-cellulose was used as carrier.
[c] These additives were obtained by Sigma.
[d] This commercial product is used as pellet binder.
[e] Antioxidant powder.

Table 2. Nutritional composition of experimental diets

Ingredient	Test Diets (% as fed basis)			
	SBM	CM	SFM	CSM
Dry Matter	92.37	92.22	92.90	92.99
Crude Protein	42.34	42.17	42.11	42.01
Crude Oil	12.30	12.61	12.53	12.61
Crude Fibre	1.66	3.21	3.87	4.05
Nitrogen Free Extract	27.79	25.70	25.92	25.70
Ash	8.28	8.53	8.47	8.62
Digestible Energy, MJ/kg	13.92	13.89	13.99	13.93

lipid))/(Number of feeding days)]×100 (Luzzana *et al*., 2005)

PGR=[(ln (Final body nitrogen)-ln (Initial body nitrogen))/(Number of feeding days)]×100 (Luzzana *et al*., 2005)

At the end of the experiment, nine fish from each treatment were sacrificed and pooled for total body and carcass composition analyses. The chemical compositions of the total body–carcass, complete feeds and feedstuffs were measured following AOAC methods (Anonymous, 1995).

Statistical Analyses

In this trial, Randomized Block Design Model was performed to observe the differences. All statistical analyses were performed using the statistical package, SPSS (v17) for Windows (2008). The significance of treatment effects on the different parameters measured were determined by one-way ANOVA followed by Tukey's multiple comparison test where appropriate. Differences were reported as significant if $P<0.05$. Results presented in Table 3 are reported as means ± SD (n=3) unless otherwise stated and total body and carcass quality parameters results as means ± SD (n=9).

Results

The results obtained in this experiment are summarized in Table 4. There are no important differences among the groups for initial live weight, but important differences among final weights ($P<0.05$). While differences between SBM and CM; SBM and CSM; SFM and CSM are significant

Table 3. Average initial weight, final weight, weight gain, FCR, SGR, PER and survival rate for rainbow trout fed different diets for 84 days*

Item	Test Diets			
	SBM $X\pm S_{\bar{x}}$	CM $X\pm S_{\bar{x}}$	SFM $X\pm S_{\bar{x}}$	CSM $X\pm S_{\bar{x}}$
Trial Period, day	84	84	84	84
Total Fish Number	180	180	180	180
Initial Weight, g	49.86±1.19	49.97±0.71	50.17±0.47	50.30±0.99
Final Weight, g	231.6±4.96^{a}	213.1±7.40^{bc}	221.0±6.68^{ab}	197.1±8.54^{c}
Average Live Weight Gain, g	181.8±3.87^{a}	163.1±6.86^{bc}	170.8±6.97^{ab}	147.1±8.68^{c}
Average Feed Consumption, g	250.0±10.0	246.8±17.56	241.7±20.21	230.0±18.03
Feed Conversion Rate, g	1.37±0.05^{a}	1.51±0.06^{ab}	1.41±0.07^{a}	1.56±0.03^{b}
Protein Efficiency Rate (PER)	1.86±0.01^{a}	1.82±0.01^{ab}	1.84±0.02^{a}	1.78±0.03^{b}
Specific Growth Rate (SGR)	1.83±0.01^{a}	1.73±0.03^{bc}	1.76±0.04^{ab}	1.67±0.02^{c}
Gain [a], %	364.5	326.5	340.5	291.8
Fat Deposition Rate (FDR)	0.75	0.75	0.78	0.71
Protein Growth Rate (PGR)	0.14	0.11	0.11	0.09
Survival Rate, %	98.3	97.2	96.6	95.5
Condition Factor	1.27±0.04	1.20±0.02	1.15±0.03	1.14±0.02

*Results are means±SD (n=3). Means in the same row that do not share a common superscript letter differ significantly (P<0.05).
[a] Percentage of initial weight.

Table 4. Total body and carcass composition in the beginning and end of the trial*

Item		Test Diets (% wet weight)			
		SBM $X\pm S_{\bar{x}}$	CM $X\pm S_{\bar{x}}$	SFM $X\pm S_{\bar{x}}$	CSM $X\pm S_{\bar{x}}$
Initial Total Body	Moisture	74.0±0.61			
	Crude protein	14.72±0.34			
	Fat	4.88±0.17			
	Ash	2.58±0.09			
Initial Carcass	Moisture	75.51±0.58			
	Crude protein	13.87±0.22			
	Fat	4.04±0.14			
	Ash	2.66±0. 09			
Final Total Body	Moisture	70.9±1.02^{a}	72.3±0.49^{ab}	71.2±0.47^{ab}	73.2±0.35^{b}
	Crude protein	16.6±0.42	16.1±0.38	16.2±0.27	15.9.3±0.45
	Fat	9.18±0.61	9.16±0.53	9.40±0.57	8.85±0.44
	Ash	2.98±0.12	2.16±0.10	2.28±0.12	2.01±0.08
Final Carcass	Moisture	72.36±0.78	72.69±0.49	72.82±0.44	73.19±0.66
	Crude Protein	18.8±0.48^{a}	18.1±0.10^{ab}	17.86±0.61^{ab}	17.72±0.24^{b}
	Fat	6.26±0.19	6.25±0.28	6.08±0.29	5.92±0.12
	Ash	2.08±0.27	1.99±0.27	2.22±0.17	2.14±0.12

* Results are means±SD (n=9). Means in the same row that do not share a common superscript letter differ significantly (P<0.05).

statistically (P<0.05), the differences between SBM and SFM; CM and CSM are not significant in terms of final weight or live weight gain.

Protein efficiency ratio (PER) can be defined as a ratio of live weight gain to the consumed crude protein in a sample period. The differences between SBM and SFM, and CSM were found to be statistically important (P<0.05). There are statistically significant difference between SBM and, CM and CSM; SFM and CSM in terms of specific growth rate (SGR), shows ratio of daily live weight gain during the trial (P<0.05).

Fish deaths were found to be random and different plant protein sources did not affect survival rate. Moreover, the differences among the groups for condition factors, total body composition (except moisture) and carcass composition (except crude protein content) are not significant (P>0.05). Differences on crude protein in carcass composition and moisture content in total body composition between SBM and CSM were found to be statistically important (P<0.05).

Discussion

Trout need good quality protein sources in their feeds for better growing. There is a considerable difference among the results obtained by the feeding

experiment of the present study. Although the difference between soybean meal, canola meal and sunflower meal in terms of protein efficiency ratio is the same, it can be said that the best result for the live weight gain is obtained with soybean meal. At the same time, the most protein accumulation was obtained by the group fed the ration containing SBM. Findings of Carter and Hauler (2000) supported to ours. They determined that soybean meal protein concentrate had the best potential for replacing at least 33% of the fish meal protein in extruded salmon feeds on the weight gain and feed efficiency ratio. Our findings were also supported by Martinez (1986), Cheng and Hardy (2002) and Barros *et al.* (2002) findings. They have reported that the acceptable level of cotton seed meal inclusion in the ration is not higher than the maximum of 10% and more than 55% substitution of soybean meal by cotton seed meal could result in reduced live weight gain. Dorsa *et al.* (1982) indicated that more than 17.4% of cotton seed meal in the ration inhibited the growth. Similarly, Hepher (1989) has reported that cotton seed meal has a lower nutritional value than soybean meal, which is also supported by the current study. Observed difference between soybean and sunflower meal, and canola and cotton seed meal can be attributed to high level complex carbohydrate and high level crude fibre content of canola and cotton seed meal and their unbalanced essential amino acid profile. In addition, they have another important disadvantage such as higher FCR. In contrast, Dadgar *et al.* (2009) has shown that cotton seed meal did not negatively affect the live weight gain, even when 100% replaced the soybean meal. This finding suggests that cotton seed meal can be included in the rainbow trout diet up to 31%.

The present research was performed to compare the different plant protein sources on the rainbow trout. Therefore, it does not include any suggestion on usage level of them. However soybean is the most used feed ingredient with fish meal in traditional fish feeds, if deficiency of some essential amino acids can be achieved by supplemental amino acids like methionine and lysine (Bloom *et al.*, 2001; Robinson, 1991), it is thought that soybean, sunflower, canola and cotton seed meal can be used as a protein sources in fish feeds easily. However, canola and cotton seed meals should be included into fish feed only after fingerling period, as reported by Hilton and Slinger (1986).

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