

Effects of canola meal on growth and digestion of rainbow trout (*Oncorhynchus mykiss*) fry

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Abstract: A 12-week feeding trial was conducted with rainbow trout fry (initial weight of 1.57 ± 0.01 g) to examine the effects of partial substitution of canola meal in prepared diets on growth, feed conversion ratio (FCR), nutrient digestibility, somatic indices, and survival rate. Five isonitrogenous (44% crude protein) and isocaloric (4000 kcal/kg digestible energy) diets were formulated to contain 8%, 16%, 24%, and 32% canola meal against no canola meal (control group). A total of 375 rainbow trout were distributed into 5 experimental groups with 3 replicates. At the end of the experiment, the fish that were fed diets containing 8% and 16% canola meal exhibited similar growth performance, FCR, and protein digestibility as those receiving the control diet ($P > 0.05$). Feed intake was reduced in the groups fed a diet containing a level of canola meal higher than 8%. A decrease in growth performance and FCR was found in the fish fed diets with 24% and 32% canola meal. Fish fed the diet containing 32% canola meal had the worst growth and FCR. No significant differences in lipid digestibility or somatic indices were found ($P > 0.05$). This study showed that canola meal could replace standard diets by up to 8% in rainbow trout fry without adversely affecting performance.

Key words: Canola meal, feed conversion ratio, growth, nutrient digestibility, rainbow trout fry

Introduction

Fish meal usage in aquaculture is estimated to reach well over 4 million metric tons by 2015 (1). Given that total production of fishmeal is approximately 6 million tons per year and that this level of production is expected to remain constant or decrease slightly in the future, there are concerns regarding the long-term sustainability of this resource (2). A number of studies have examined the effect of the replacement of fish meal with plant proteins in fish diets, including soybean meal (3,4), maize gluten meal (5), lupins (6), rapeseed (7), cottonseed meal (8), and canola meal (9). Canola meal is a suitable protein replacement

for fish meal as it has relatively high protein content (38%) and the protein is highly digestibility for fish (10). Canola is the name given to selected varieties of rapeseed that are low in glucosinolate and erucic acid (11). The amino acid profile of canola meal protein is similar to that of herring meal protein and superior to soybean meal protein (12-14). However, the cost of canola meal is generally lower than that of fish meal and soybean meals (15).

Information pertaining to the use of canola meal in fish diets is presented in the literature (9-13); nevertheless, the literature for trout fry diets in specific is scarce (14). The aim of this study

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was to evaluate the use of canola meal as a partial replacement for fish meal in the diets of rainbow trout fry by examining growth, feed conversion ratio (FCR), nutrient digestibility, survival rate, and somatic indices.

Materials and methods

Fish and experimental conditions

Rainbow trout (*Oncorhynchus mykiss*) were obtained from the production unit of Eğirdir Fisheries Faculty, Süleyman Demirel University, Isparta, Turkey. The feeding trial was conducted in 15 tanks (50 × 50 × 70 cm). Twenty-five fish (average weight: 1.57 ± 0.01 g) were randomly stocked into each tank with 3 replications per treatment. All of the fish were fed ad libitum for 12 weeks. During the experimental period, the dissolved oxygen level, temperature, and pH were maintained at 6.5 ± 0.06 mg/L, 11 ± 1 °C, and 7.03 ± 0.27, respectively.

Experimental diets

Five isonitrogenous (44% protein) and isocaloric (4000 kcal/kg digestible energy) diets were formulated to contain 0%, 8%, 16%, 24%, and 32% canola meal (16). Ingredients and chemical compositions of the diets are given in Table 1. The solvent-extracted canola meal used in the experiment was obtained from Ukraine. Proximate analysis of the canola meal is presented in Table 2.

In preparing the diets, the ingredients were first ground to a small particle size in a mill. The ingredients were thoroughly mixed, and then oil and water were added. The diets were cold-pelleted into 2-mm-diameter size using a mincing machine, and then they were dried for about 12 h in a ventilated oven at 45 °C. After drying, the diets were broken up and sieved into appropriate pellet sizes, and they were stored at -20 °C.

Analysis and measurement

At the end of the feeding trial, 5 fish per tank were sacrificed by a lethal dose of anesthesia (150 mg/L MS-222), homogenized in a blender, and stored at -20 °C for analysis. The moisture, crude protein, crude fiber, and ash contents of the feed ingredients, experimental diets, and feces samples were determined according to standard AOAC methods (17). The total lipids of

all of the samples were determined by the chloroform-methanol extraction method (18).

Digestibility study

Apparent digestibility coefficients (ADCs) of the different diets were measured by the indicator method (19), using 0.5% chromic oxide as a marker. The collection of fish feces was carried out during the feeding trial. The collection and preparation of fish feces for chemical analysis were previously described by Lim et al. (20). ADCs were calculated using the following equation (21):

$$\text{ADC} = 100 - [100 \times (\text{Cr}_2\text{O}_3 \text{ in diet } [\%] / \text{Cr}_2\text{O}_3 \text{ in feces } [\%]) \times (\text{nutrient in feces } [\%] / \text{nutrient in diet } [\%])].$$

Statistical analysis

One-way ANOVA was used to compare growth, FCR, nutrient digestibility, and somatic indices among the treatments. All of the data were analyzed using SPSS for Windows (22). Duncan's multiple range test was used to determine the mean differences among the treatments ($P = 0.05$)

Results

The growth performance of rainbow trout fry that were fed the different canola meal diets is presented in Table 3. Final weight, weight gain, specific growth ratio, and FCR of the fish fed diets containing 8% and 16% canola meal were similar to those of the fish fed the control diet ($P > 0.05$). Feed intake was reduced in the groups fed a diet containing a level of canola meal higher than 8%. Growth performance and FCR of the fish fed the diets with 24% and 32% canola meal were lower. Fish fed the diet containing 32% canola meal showed the lowest final weight, weight gain, and specific growth rate. Moreover, the group fed the diet containing 32% canola meal appeared to have the lowest feed intake.

The highest protein digestibility was found in the control group. However, protein digestibility of the groups fed 8% and 16% canola meal did not differ significantly from that of the control group. The addition of 24% and 32% canola meal to the diets resulted in lower protein digestibility.

There were no significant differences in the digestibility of crude lipids or somatic indices values among the dietary groups ($P > 0.05$).

Table 1. Ingredients and nutrient composition of diets used in the experiment.

Ingredients (%)	Diets				
	Control	CM8	CM16	CM24	CM32
Fish meal	45.00	41.00	37.00	33.00	29.00
Solvent-extracted canola meal	0.00	8.00	16.00	24.00	32.00
Soybean meal	13.80	13.80	13.80	13.80	13.80
Corn gluten meal	12.70	12.70	12.70	12.70	12.70
Corn flour	17.10	12.80	8.55	4.30	0.01
Fish oil	9.90	10.20	10.45	10.70	10.99
Vitamin*	0.50	0.50	0.50	0.50	0.50
Cr ₂ O ₃	0.50	0.50	0.50	0.50	0.50
Mineral**	0.10	0.10	0.10	0.10	0.10
Choline chloride	0.20	0.20	0.20	0.20	0.20
Binder	0.20	0.20	0.20	0.20	0.20
Chemical analysis					
Dry matter (%)	87.34	88.48	88.50	89.39	89.75
Crude protein (%)	44.85	44.66	44.47	44.28	44.09
Crude fat (%)	15.51	15.40	15.24	15.08	14.96
Crude fiber (%)	1.60	2.52	3.44	4.36	5.29
Digestible energy (kcal/kg)***	4000	4000	4000	4000	4000

CM8: diet contained 8% canola meal, CM16: diet contained 16% canola meal, CM24: diet contained 24% canola meal, and CM32: diet contained 32% canola meal. *Vitamin premix contained the following per kilogram: 4,000,000 IU vitamin A, 480,000 IU vitamin D3, 2400 mg vitamin E, 2400 mg vitamin K3, 4000 mg vitamin B1, 6000 mg vitamin B2, 4000 mg niacin, 10,000 mg calcium D-pantothenate, 4000 mg vitamin B6, 10 mg vitamin B₁₂, 100 mg D-biotin, 1200 mg folic acid, 40,000 mg vitamin C, and 60,000 mg inositol. ^bMineral premix contained the following per kilogram: 23,750 mg manganese, 75,000 mg zinc, 5000 mg copper, 2000 mg cobalt, 2750 mg iodine, 100 mg selenium, and 200,000 mg magnesium. ***Digestible energy value was calculated from published values for the diet ingredients (16).

Table 2. Nutrient composition of canola meal used in the experiment.

Nutrient composition	%
Crude protein	34.25
Total lipid	1.52
Crude fiber	12.99
Crude ash	7.12
Dry matter	89.89
Glucosinolates (µmol/g)	9.04

The survival rate was not affected by canola meal levels in the diets ($P > 0.05$) and ranged between 96.66%-98.33% in all of the treatments.

Discussion

The results of this study suggested that commercial canola meal could comprise up to 8% of the diet of rainbow trout fry without compromising their growth, survival, feed intake, FCR, and protein and lipid digestibility.

Table 3. Growth, feed efficiency, digestibility, survival rate, and somatic indices of rainbow trout fed diets containing various levels of canola meal (mean \pm SE) for 12 weeks.

	Control	CM8	CM16	CM24	CM32
Growth					
Initial weight (g)	1.55 \pm 0.03	1.57 \pm 0.03	1.56 \pm 0.03	1.57 \pm 0.02	1.58 \pm 0.03
Final weight (g)	14.21 \pm 0.61 ^a	13.06 \pm 0.47 ^{ab}	12.82 \pm 0.57 ^{ab}	11.79 \pm 0.47 ^{bc}	10.48 \pm 0.39 ^c
WG (g)	12.65 \pm 0.62 ^a	11.51 \pm 0.83 ^{ab}	11.24 \pm 0.89 ^{ab}	10.20 \pm 0.41 ^{bc}	8.88 \pm 0.42 ^c
SGR (% daily)	2.45 \pm 0.05 ^a	2.36 \pm 0.06 ^{ab}	2.3 \pm 0.06 ^{ab}	2.24 \pm 0.05 ^{bc}	2.10 \pm 0.03 ^c
Feed intake (g)	12.80 \pm 0.17 ^a	12.77 \pm 0.44 ^a	12.55 \pm 0.32 ^{ab}	12.35 \pm 0.55 ^{ab}	11.49 \pm 0.23 ^b
FCR	1.04 \pm 0.04 ^a	1.10 \pm 0.04 ^{ab}	1.09 \pm 0.03 ^{ab}	1.19 \pm 0.03 ^{bc}	1.30 \pm 0.05 ^c
Survival rate (%)	98.33	98.33	98.33	98.33	96.66
Apparent digestibility coefficients					
Crude protein	92.76 \pm 0.17 ^a	90.44 \pm 0.45 ^{ab}	90.32 \pm 0.04 ^{ab}	89.98 \pm 0.98 ^b	89.48 \pm 1.11 ^b
Crude lipid	92.98 \pm 0.13	93.32 \pm 0.25	93.54 \pm 0.51	93.49 \pm 0.33	93.33 \pm 1.28
Somatic indices					
Hepatosomatic index	1.08 \pm 0.05	1.27 \pm 0.07	1.16 \pm 0.06	1.11 \pm 0.10	1.23 \pm 0.12
Renosomatic index	0.99 \pm 0.07	0.98 \pm 0.06	0.93 \pm 0.06	0.97 \pm 0.07	1.02 \pm 0.05

^{a-c}: Values in the line having the same superscript are not significantly different ($P > 0.05$).

WG (g) = (final body weight, g - initial body weight, g).

FCR = (total feed intake, g) / (final body weight, g - initial body weight, g).

SGR (% daily) = [(ln final body weight - ln initial body weight) / days] \times 100.

Hepatosomatic index (HIS) = 100 \times liver weight, g / body weight, g.

Renosomatic index (RSI) = 100 \times kidney weight, g / body weight, g.

Shafaeipour et al. (14) indicated that a diet with 30% canola meal may be acceptable for use in rainbow trout diets. The difference of their study from the present study could be due to fish age, but also to the processing and source of the canola. The effects of the substitution of canola meal are different in different fish species. Thus, Higgs et al. (23) found that rapeseed could comprise 13% to 16% of the dietary protein of Chinook salmon, while using a higher level of rapeseed meal depressed growth. Cheng et al. (24) indicated that canola meal could substitute for less than 20% of the fish meal protein without influencing the growth of Japanese sea bass. Furthermore, higher substitution levels of canola meal induced negative influences on the growth and survival of Japanese sea bass. Li and Robinson (25) reported that feeds for channel catfish can contain at least 25% canola meal by replacement of soybean

meal without detrimental effects. Davies et al. (7) suggested an inclusion limit of 15% rapeseed meal in the diet of tilapia (*Oreochromis mossambicus*) fry. The differences of these studies from the present study could be due to different fish species and the glucosinolate content of the canola meal.

The present study showed that there was an inverse relationship between the growth of the rainbow trout fry and the dietary canola meal levels. In this study, a decrease of growth of the fish with an increase of canola meal level in the diet may be due to high dietary fiber and the glucosinolate content of the canola meal. This is similar to the suggestion of Higgs et al. (23), who reported that a decrease of growth in Chinook salmon that were fed rapeseed protein concentrate may be due to high dietary fiber. Shiao and Kwok (26) indicated that increased dietary fiber can reduce growth in tilapia. Liang (27) reported that

fiber can induce a faster passage rate, reducing the opportunity for digestion and increasing endogenous nitrogen loss through abrasive action or binding endogenous protein. In addition, studies with fish also provoked deleterious effects of glucosinolate on growth inhibition (28-30). Burel et al. (28) reported that 300 g/kg rapeseed meals containing a low level of glucosinolate (5 $\mu\text{mol/g}$) could not be used in practical rainbow trout diets without deleterious effects. Furthermore, feed efficiency was adversely affected by rapeseed meal and glucosinolate intake. Burel et al. (29) suggested that heat treatment is effective in reducing the glucosinolate content of feed materials in rapeseed meal from 40 to 26 $\mu\text{mol/g}$ after wet pressure cooking; thus, heat-treated rapeseed meal at the level of 30% did not lead to any significant decrease in growth performance of turbot. Burel et al. (30) indicated that dietary incorporation of rapeseed meal with a low (26 $\mu\text{mol/g}$) content of glucosinolate at a level of 30% led to a decrease in the growth performance of rainbow trout after only 80 days of feeding.

In the present study, feed intake was reduced in fish fed a high level (8%) of dietary canola meal. This result was similar to the studies of Shafaeipour et al. (14), which showed that feed intake decreased with increasing dietary canola meal level in trout. Hilton and Slinger (31) indicated that suppression of feed intake could be the main reason for reduced growth performance of rainbow trout as dietary canola meal level increased. These results are in contrast with the studies of Cheng et al. (24), who observed an increasing trend in feed intake with increasing dietary canola meal level in Japanese sea bass. Differences between these studies and the present study may be due to the use of different fish species.

In the present study, the addition of 24% and 32% canola meal to diets resulted in lower protein

digestibility. The lower protein digestibility was most likely attributable to the large amount of poorly digestible fiber in canola meal. It is well known that fish do not secrete cellulase (16), and higher levels of fiber can decrease the absorption of other nutrients. Mwachireya et al. (13) concluded that high levels of fiber have the greatest adverse effects on the digestibility of canola protein products for rainbow trout.

Yigit and Olmez (32) found that the addition of canola meal to tilapia diets resulted in lower protein digestibility. Glencross et al. (33) reported that the influence of heat had a considerable negative impact on the nutritional value of canola meal. Burel et al. (34) reported that in trout, no significant difference was found between solvent-extracted and heat-treated rapeseed meal for the digestibility of dry matter. However, in turbot, the digestibility of rapeseed meal was improved by a thermal treatment. Mwachireya et al. (13) indicated that in general, solvent washing and enzymatic treatments significantly depressed dry matter digestibility coefficients of commercial canola meal. Burel et al. (28) reported that a decrease in the digestibility of dry matter and protein was observed with a high level of rapeseed meal (300 and 500 g/kg).

In conclusion, there are obvious limitations to the use of canola meal as an ingredient in trout fry diets. Constraints arise from either the presence of various antinutritional factors (fiber, glucosinolate, etc.) or feed intake. In this study, although fish fed diets containing 8% and 16% canola meal exhibited similar growth performance as those fed the control diet, feed intake was reduced in the groups fed a diet containing levels of canola meal higher than 8%. Therefore, canola meal can replace up to about 8% of the diet of rainbow trout fry without adversely affecting their growth, FCR, digestibility, and somatic index.

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