

Replacement of Soybean Meal with Guar Meal in the Diet of Rainbow Trout, *Oncorhynchus mykiss* (Walbaum 1792): Biological Parameters and Flesh Quality

VAHID YADOLLAHI¹, MEHDI SHAMSAIE², MEHRAN JAVAHERI BABOLI³, FROUD YADOLLAHI⁴, ESFANDYAR NAJAFI HAJIVAR¹ AND FATEH MOËZZI^{5*}

¹Research and Development Center, Faradaneh Feed Mill Co., Shahr-e Kord, Islamic Republic of Iran

²Department of Fisheries, Faculty of Agriculture and Natural Resources, Science and Researches Branch, Islamic Azad University, Tehran, Islamic Republic of Iran

³Department of Fisheries, Ahvaz Branch, Islamic Azad University, Ahvaz, Islamic Republic of Iran

⁴Department of Food Science and Technology, Shahr-e kord Islamic Azad University Branch, Shahr-e kord, Islamic Republic of Iran

⁵Department of Fisheries, University of Tehran, Karaj, Islamic Republic of Iran

Abstract

The present study investigated the effects of a diet containing guar meal, as a substitute for soybean, in rainbow trout *Oncorhynchus mykiss* (Walbaum 1792). The juvenile fish were fed with a soybean meal diet as basal diet and guar meal diets as substitute diets with replacement levels of 25, 50, 75 and 100 % (represented by 25PR, 50PR, 75PR and 100PR, respectively). Growth performance, feed utilisation, and body composition were investigated. Condition factor of fish fed 100PR was significantly lower than other treatments. Maximum and minimum survival of fish were observed in 50PR and 75PR treatments, respectively. Eviscerated fat of fish in 75PR and 100PR groups were significantly ($P < 0.05$) higher than other treatments. There was no significant difference in the other parameters between experimental treatments. The results indicate that there was no adverse effect in rainbow trout juveniles fed guar meal in the diet and thus could be an appropriate substitute for soybean meal in fish diet.

Keywords: guar meal, soybean meal, growth performance, feed utilisation, body composition

*Corresponding author. Email: fmoezi.fateh@gmail.com

Introduction

The major amount of the current cost of aquaculture production is feed. Primary sources of protein in fish feed include fish meal and soybean meal (Trushenki et al. 2006; Gatlin et al. 2007). These are expensive ingredients that are high in demand, exceeding supply. The increase in soybean meal in the diet can lead to poor digestibility (Oliva-Teles and Goncalves 2001; Soltan 2005), decrease in feed conversion ratio (FCR) and protein efficiency ratio (PER) (Robaina et al. 1998), decrease in growth (Lim and Dominy 1991; Sintayehu et al. 1996; Tacon 1997; Zead et al. 2008) and in some cases fish mortality (Hepher 1998) due to anti-nutritive factors of soybean meal (Smith 1977; Oliva-Teles and Goncalves 2001; Gatlin et al. 2007). Thus, in recent years, there has been more emphasis on soybean replacement with other plant protein that is easily available at lower cost with higher protein content and meets the nutritional requirements of fish (Hassan et al. 2015).

Guar (*Cyamopsis tetagonoloba*) is a yearling plant of the family Leguminacea (Storebakken 1985). The main derivative of guar seeds is guar gum, which is used at low levels as a binder in animal feeds and is the main anti-nutritional factor in guar seeds (Francis et al. 2001). Guar meal is a secondary component of the guar gum extraction process with a high protein level of 35–45 %, that is used as a supplement in livestock feed (Storebakken 1985). Using guar meal at high levels in fish feeds can lead to decrease in nitrogen retention and consumption, and fat absorption in the gastrointestinal tract that can cause weight loss and even death of fish (Enes et al. 2013).

Guar meal has some properties that make it an appropriate substitute for soybean meal in fish feed. Guar meal has a higher protein content, better amino acid profile and lower cost per unit of produced protein compared to soybean meal (Hardy 1999). Methionine and cysteine are the determinative amino acids in soybean meal (0.57 % and 0.70 %). These amino acids are much higher in guar meal (1.57 % and 2.53 %) (Hardy 1999). In addition, other benefits of guar meal include higher digestibility and higher pellet quality due to the low content of guar gum as a binder (Brinker and Friedrich 2012). Accordingly, guar meal can be a potential alternative to soybean meal in fish diet. Thus, replacing soybean meal with guar meal and determining its effects on the growth performance, feed utilisation and proximate composition of the fish fillets is required.

The aim of the present study was to investigate the possibility of soybean meal replacement with guar meal in the fish diet at different replacement levels and their effect on growth performance, feed utilisation and body composition of rainbow trout (*Oncorhynchus mykiss*) juveniles.

Materials and Methods

Fish conditioning and rearing

A total of 345 rainbow trout *Oncorhynchus mykiss* (Walbaum 1792) juveniles (weight: 82 ± 0.6 g) were obtained from a private farm in Bazoft region, Chaharmahal-o-Bakhtiari province, Iran and were stocked in 15 fiberglass tanks (125 L; 23 fish per tank; ≈ 5 L per fish). Water used in the experimental tanks was obtained from a groundwater well. Water flow was maintained at $3 \text{ L}\cdot\text{min}^{-1}$ and the photoperiod of experimental tanks was fixed at 12L:12D. The fish were acclimatised for 1 week and fed a commercial diet (Fara-Daneh Co. extruded diet, Iran) three times daily (8:00 a.m., 12:00 p.m., 4:00 p.m.) until apparent satiation. After the acclimatisation period, feeding of fish began with a basal diet and 4 experimental diets for 60 days. There were three replicates for each diet treatment. Fish were fed at a rate of 6 % of the body weight (Ramos et al. 2015), which was divided between three feeding times. The excess food in each tank was measured at the end of each day to record daily food intake.

Water parameters

Water parameters (pH, dissolved oxygen (DO) and temperature (T)) were measured daily using a multi-parameter field meter (WTW MultiLine® 3410 IDS, USA). The values (mean \pm SD) of water temperature, pH and dissolved oxygen were 15.1 ± 2.1 ($^{\circ}\text{C}$), 8.2 ± 0.6 and 8.1 ± 0.8 ($\text{mg}\cdot\text{L}^{-1}$), respectively.

Diet formulation

Five diets were used including soybean meal as one basal diet (C) and guar meal (Aria-Shirin-Noush Co., Iran) as substitute diets at 25 %, 50 %, 75 % and 100 % represented by 25PR, 50PR, 75PR and 100PR, respectively (Table 1).

Experimental diets were regulated according to the digestible energy level and the guar gum content (Drew 2004), and soybean meal replacement with guar meal was performed based on iso-energetic equivalents. Dry ingredients included: fish meal (68 % protein), wheat flour, choline chloride, sodium chloride, calcium carbonate, gelatin, canola oil (Fara-Daneh Co., Shahr-e-Kord, Chahar Mahall-o-Bakhtyari, Iran); mineral premix (Amineh-Gostar Animal Health Co., Iran); vitamin premix (Science Laboratories Co., Iran); high protein guar meal (68 % protein) (Arya-Shirin-Noush Co., Iran); soybean meal (Gol-bahar Co., Iran). Preparation of experimental diets was done using Brinker and Reiter (2011).

Table 1. Feed ingredients and proximate composition of experimental diets. (C: basal diet; 25, 50, 75 and 100PR: experimental diets with different levels of guar meal replacement).

	Experimental diet				
	C	25PR	50PR	75PR	100PR
<i>Feed ingredients (%)</i>					
Fishmeal	42.65	42.4	42.2	41.9	41.7
Soybean meal	20	15	10	5	0
Guar meal	0	3.88	7.8	11.63	15.5
Wheat flour	20	21.41	22.8	24.38	25.8
Oil	13.52	13.41	13.3	13.19	13.1
Gelatin	2	2	2	2	2
Choline chloride (60 %)	0.1	0.1	0.1	0.1	0.1
Sodium chloride	0.3	0.3	0.3	0.3	0.3
Calcium carbonate	1.2	1.2	1.2	1.2	1.2
Mineral supplement	0.15	0.15	0.15	0.15	0.15
Vitamin supplement	0.15	0.15	0.15	0.15	0.15
<i>Calculated ingredients</i>					
Digestible energy (kcal.kg ⁻¹)	4391	4374	4359	4380	4339
Protein (%)	42.00	42.00	42.06	42.01	42.04
Fat (%)	20.00	20.00	20.01	20.00	20.03
Energy/protein ratio	104.54	104.14	103.64	104.26	103.21

Growth trial and chemical analysis

The weight of the fish in the experimental tanks was taken at the beginning of the experiment. Juvenile fish were reared for 60 days and fed with basal and experimental diets. At the end of the study period, weight and length of the fish in each tank were recorded and biometric indexes were calculated as follows:

Weight gain (g) = final weight - initial weight.

Feed intake (g.day⁻¹) = consumed feed (g) / rearing period (day)

Feed conversion ratio [FCR] = dry feed consumed (g) / wet weight gain (g)

Protein efficiency rate [PER] = weight gain (g) / protein intake (g)

Specific growth rate [SGR] (% day⁻¹) = [(ln final weight - ln initial weight) / time (days)] × 100

Condition factor [CF] (g.cm⁻³) = 100 × body weight (g) / body length (cm³)

Survival (%) = (final fish number / initial fish number) × 100

Fishes were starved for 48 h at the end of the experimental period, and eight fish were randomly selected for chemical analysis of their carcass. Diets and fish fillets were analysed for ash, crude protein, and crude fat using standard methods of Association of Official Analytical Chemists (AOAC 1990). Diet and fish were dried at 100 °C for 48 h in an oven to constant weight. For calculating ash content of the samples, they were maintained in a furnace at 550 °C for 12 h, and thereafter, ash content (%) was calculated as ash content (%) = [(ash weight/sample weight) × 100]. Crude fat was determined using a Soxhlet apparatus and n-hexane as the solvent, and crude fat was calculated as: [(fat weight/sample weight) × 100]. Total nitrogen (N) levels were determined using the Kjeldahl method, and crude protein content was calculated as N × 6.25.

Statistical analysis

Statistical analyses were performed using SPSS version 19.0. All results are presented as mean ± SD. Normality of obtained data was analysed using Kolmogorov-Smirnov (K-S) test. One-way analysis of variance (ANOVA) was used for comparing parameters between different fish diets. Level of significance was determined at $P < 0.05$.

Results

The growth performance results are given in Table 2. There were no significant differences ($P > 0.05$) in initial and final weights, as well as weight gain of the fish fed both the basal and experimental diets. The CF value of the fish fed diet with 100 % replacement ($3.76 \pm 0.84 \text{ g.cm}^{-3}$) was significantly ($P < 0.05$) lower than the fish fed basal diet and other experimental diets with lower replacement levels. The calculated SGR values showed no significant difference between experimental dietary groups. The survival of fish at 50PR ($89.85 \pm 6.6 \%$) was significantly higher than 75PR ($79.71 \pm 5.02 \%$), however not significantly different ($P > 0.05$) from the fish fed basal diet.

Table 2. Growth performance indices (mean ± SD) of the fish fed with different diets. (C: basal diet; 25, 50, 75 and 100PR: experimental diets with different levels of guar meal replacement).¹

	C	Experimental diet			
		25PR	50PR	75PR	100PR
Initial weight (g.fish ⁻¹)	82.88 ± 2.11	79.52 ± 1.86	86.45 ± 3.6	81.82 ± 2.8	78.40 ± 3.3
Final body weight (g.fish ⁻¹)	144.25 ± 7.16	153.84 ± 10.98	147.73 ± 5.63	142.38 ± 5.54	139.53 ± 11.26
Weight gain (g.fish ⁻¹)	61.37 ± 6.47	74.31 ± 12.27	61.29 ± 12.39	60.56 ± 14.84	61.12 ± 8.39
Condition factor	4.70 ± 0.25 ^a	4.91 ± 0.14 ^a	4.76 ± 0.07 ^a	4.82 ± 0.15 ^a	3.76 ± 0.84 ^b
SGR	0.92 ± 0.074	1.09 ± 0.147	0.90 ± 0.233	0.93 ± 0.258	0.96 ± 0.096
Survival (%)	84.06 ± 5.02 ^{ab}	88.41 ± 2.50 ^{ab}	89.85 ± 6.6 ^a	79.71 ± 5.02 ^b	82.61 ± 0.0 ^{ab}

¹Means within a row having different superscripts were significantly different ($P < 0.05$).

Feed utilisation data are shown in Table 3. There were significant differences ($P < 0.05$) in FI between experimental treatments. The lowest FI belonged to the fish fed with basal diet (2.23 ± 0.31 g.day⁻¹), and highest values belonged to 25PR (3.07 ± 0.41 g.day⁻¹) and 100PR (3.37 ± 0.43 g.day⁻¹). The FCR and PER values showed no significant difference between fed fishes with different diets. The results of the body composition analysis are presented in Table 4. There was no significant difference between them, except for the eviscerate fat, where its values in 75PR and 100PR treatments were significantly ($P < 0.05$) higher than the basal diet and other treatments.

Table 3. Feed utilisation parameters (mean \pm SD) of the fish fed with different diets. (C: basal diet; 25, 50, 75 and 100PR: experimental diets with different levels of guar meal replacement).¹

	C	Experimental diet			
		25PR	50PR	75PR	100PR
Feed intake (g.day ⁻¹)	2.23 \pm 0.31 ^b	3.07 \pm 0.41 ^a	2.66 \pm 0.38 ^{ab}	2.94 \pm 0.39 ^{ab}	3.37 \pm 0.43 ^a
FCR	1.19 \pm 0.285	1.24 \pm 0.188	1.37 \pm 0.362	1.51 \pm 0.299	1.62 \pm 0.345
PER	2.08 \pm 0.48	1.95 \pm 0.32	1.82 \pm 0.53	1.64 \pm 0.39	1.51 \pm 0.31

¹Means within a row having different superscripts were significantly different ($P < 0.05$).

Table 4. Body composition (mean \pm SD) of the fish fed with different diets. (C: basal diet; 25, 50, 75 and 100PR: experimental diets with different levels of guar meal replacement).¹

	C	Experimental diet			
		25PR	50PR	75PR	100PR
Eviscerate weight (g)	132.36 \pm 7.09	141.57 \pm 10.3	135.31 \pm 6.32	130.92 \pm 2.32	1127.57 \pm 9.6
Visceral mass (g)	11.89 \pm 1.38	12.27 \pm 1.40	12.42 \pm 0.58	11.46 \pm 0.95	11.96 \pm 1.39
Liver weight (g)	1.75 \pm 0.26	1.69 \pm 0.31	1.48 \pm 0.19	1.70 \pm 0.27	1.52 \pm 0.21
Hepatosomatic index	0.011 \pm 0.001	0.014 \pm 0.006	0.011 \pm 0.001	0.015 \pm 0.004	0.013 \pm 0.002
Dry matter (g)	27.40 \pm 0.67	27.77 \pm 0.41	27.56 \pm 0.21	27.83 \pm 0.28	27.36 \pm 0.22
Ash content (%)	2.32 \pm 0.34	2.11 \pm 0.08	2.22 \pm 0.19	2.30 \pm 0.17	2.15 \pm 0.19
Eviscerate fat (%)	0.94 \pm 0.11 ^b	0.80 \pm 0.11 ^b	0.84 \pm 0.12 ^b	1.58 \pm 0.38 ^a	1.67 \pm 0.49 ^a
Liver fat (%)	2.03 \pm 0.21	1.96 \pm 0.20	2.42 \pm 0.29	2.43 \pm 0.37	2.27 \pm 0.24
Eviscerate protein (%)	16.94 \pm 0.51	16.77 \pm 0.66	16.84 \pm 0.22	17.18 \pm 0.66	16.78 \pm 0.34

¹Means within a row having different superscripts were significantly different ($P < 0.05$).

Discussion

The substitution of animal protein in the fish diet with plant protein that is cheap and is readily available is a crucial factor in aquafeed production. Protein from plant sources that have suitable essential amino acids profile for fish nutrition could be incorporated in the fish diet (Cho and Bureau 2001).

Since feed intake (FI) is among the main factors that affect fish growth, the acceptability of diet by fish is very important (Brinker and Reiter 2011). The level of anti-nutritional factors and other components of plant protein that disrupt nutrient digestibility and utilisation in the gastrointestinal tract and consequently the health of the fish must be minimised in the diet (Francis et al. 2001). Guar meal, which is a rich plant protein, is cheap and easily available compared to the soybean meal and hence may be an appropriate alternative to soybean and fish meal in fish feed. Most of the studies conducted using guar seeds derivatives in fish diet were focused on using guar gum as a binder in the diets (Brinker and Friedrich 2012; Enes et al. 2013; Ramos et al. 2015) and there were few studies that investigated using guar meal as a protein source in fish diets (Al-Hafedh and Siddiqui 1998).

The results of the present study showed that an increase in replacement level of soybean meal with guar meal showed no significant effect on weight alterations of fish compared to basal diet. Al-Hafedh and Siddiqui (1998) reported that an increase in replacement level of guar meal resulted in decrease growth performance due to the increase of anti-nutrient components as well as non-starch polysaccharides (NSPs) in the diets with higher levels of substitution, which are indigestible by fish. Moreover, the increase in replacement level of guar meal had no significant effect on SGR. In previous studies, growth reduction was reported with an increase in replacement level of guar meal due to an increase of anti-nutrients and digestive system inflammation, as well as damage of mucosa in the digestive tract (Rumsey et al. 1994; Baeverfjord and Kroghal 1996). The lack of difference in weight gain between fish fed basal, and treatment diets might be due to the appropriate processing of guar meal and reduction of anti-nutrients in the experimental diet. The significant decrease of condition factor in 100PR treatment (complete replacement) demonstrates that complete replacement of soybean meal with guar meal results in fish with lower weight than 25, 50 and 75PR. The lowest survival was seen in 75PR treatment, and a similar trend was also reported by Al-Hafedh and Siddiqui (1998).

There was an increase in feed intake with the increase in guar meal level as substituted diet, except for 25PR treatment. A similar trend was reported by Al-Hafedh and Siddiqui (1998) in which FI of fish fed on diets with 75 % guar meal replacement was lower than other substitution levels. The highest FCR value in their study was obtained at 25 % replacement level of guar meal. In the present study, no difference was observed for experimental diets. Body composition analysis of fish fed different experimental diets showed that using guar meal instead of soybean meal, except for eviscerate fat, had no effect on other body composition constituents (Table 4). The fat content of the body significantly increased in 75PR and 100PR treatments compared to basal and other two substitute diets. The increase in body fat content could be due to the inappropriate metabolism of proteins and imbalance of amino acids in guar meal. According to de Francesco et al. (2004) replacing fish meal with plant proteins in the diet of rainbow trout showed a decrease in the body fat content and Webster et al. (1997) working on the same species reported that replacing fish meal with mixed plant proteins can lead to changed moisture content of the body.

Moisture and fat content of the body varies greatly in different fish, but protein content is almost constant (Belal and Assem 1995). The size of fish is associated with the protein content in fish fed on diets containing mainly plant proteins (Brinker and Reiter 2011), where high correlation had been reported between wet weight and body protein content in fish (Shearer 1994; Dumas et al. 2007). In the present study, liver weight and HSI index values of fish in different treatments showed no difference in the control group. High levels of anti-nutrients and plant NSPs in fish diet can stimulate the immune system of fish and consequently increase liver weight and HSI (Nordmo et al. 1995; Skjermo et al. 2006). It is possible that the lack of alteration in liver weight and HSI in the present study was due to the effective processing of guar meal, where the level of anti-nutrients in guar meal replaced diets are nearly equal to that of basal diet.

Conclusion

The results of the present study indicate that guar meal could be a possible substitute for soybean meal in fish diets. Replacement of soybean meal with guar meal showed no adverse effects on growth performance and body composition of fish when fed as substitute diet with replacement levels at 25, 50, 75 and 100 %. The optimum level of substitution of guar meal needs further investigation.

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