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## Optimal Dietary Protein Levels in Juvenile Electric Blue Cichlid (*Sciaenochromis fryeri*)

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### Abstract

A feeding trial was conducted to determine the dietary protein requirement of juvenile electric blue cichlids (*Sciaenochromis fryeri*). Groups of fish ( $0.50 \pm 0.01$  g) were fed one of four isocaloric diets containing protein levels ranging 35-50% for 12 weeks. According to the broken-line model, the dietary protein requirement was estimated as 38.8%. The feed conversion ratio ranged from 1.73 in the 50%-protein group to 2.16 in the 35% group. There were statistically significant differences in feed intake and protein efficiency ratio. In general, the protein efficiency ratio decreased as the dietary protein level increased. For optimum growth, feed conversion, and protein utilization, a diet containing 39-40% protein, 11% crude lipid, and 20.7 MJ gross energy/kg diet is recommended for juvenile *S. fryeri*.

### Introduction

The live aquarium animal trade is a global multi-million dollar industry. Cichlid species represent approximately 95% of the world's ornamental fish. The electric blue cichlid, *Sciaenochromis fryeri*, is an African cichlid cultured throughout the world (Smith, 2000). It is a bright blue freshwater cichlid and one of the most commercially valuable ornamental fish species.

Ornamental fish obtain most of their energy from dietary lipid. However, to avoid hepatic lipidosis, lipids should not exceed 15% of the daily intake. Therefore, determination of the protein requirement of ornamental fish is important. Dietary protein plays a major role in determining the rate of fish growth (NRC, 1993). Since protein is the most expensive component of formulated fish feeds, the pre-

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cise protein requirement of cultured fish must be known for economic and environmentally-sound aquaculture production.

Like other fish species, the reported protein requirement of ornamental fish is relatively high compared to terrestrial animals (NRC, 1993). Optimum dietary protein levels for ornamental fishes depend on species, age, feeding level, quality of diet ingredients, protein-energy ratio, diet composition, etc. (NRC, 1993; Sales and Janssens, 2003). For example, dietary protein requirements vary from around 50% for growing carnivorous discus, *Symphysodon aequifasciata* (Chong et al., 2000) to 30% for the omnivorous goldfish, *Carassius auratus* (Lochmann and Phillips, 1994).

The energy requirement of electric blue cichlids has recently been investigated (Royes et al., 2006). The purpose of this study was to determine the optimum protein requirement of juvenile *S. fryeri* in a semi-recycled water system.

#### Materials and Methods

**Diets.** Four isoenergetic (21 MJ/kg) experimental diets were formulated to supply crude protein levels of 35%, 40%, 45%, and 50% using fishmeal and soybean meal as sources of protein, wheat as the carbohydrate source, and fish oil as the lipid source (Table 1). Diets were prepared by thoroughly mixing the dry ingredients with oil and water in a kitchen mixer and extruding the mixture through a 1-mm die. The moist pellets were fan-dried, ground in a food blender, and stored frozen at -20°C until needed.

**Rearing systems, fish, experimental design.** Juvenile electric blue cichlids (*Sciaenochromis fryeri*) were obtained from a commercial aquarium in Istanbul and transported to the Aquarium Unit of the Fisheries Faculty in Canakkale Onsekiz Mart University. Prior to the start of the feeding trial, the fish were kept in a 100-l aquarium and fed a commercial diet (38% protein; 6% fat) for 2 weeks to adjust to experimental conditions.

At the start of the trial, the fish were randomly distributed amongst twelve 30-l glass aquaria (30 x 30 x 40 cm) inside a 750-l semi-

recirculating system annexed to 210-l sand filter system at a rate of 8 fish per tank, three tanks for each dietary treatment. Water flow was 1 l/min. Temperature (26.5±0.8°C), pH (7±0.5), and dissolved oxygen (7.2±0.4 mg/l) were measured daily. The system was installed in a climate-controlled laboratory with an artificial photoperiod of 12 h light:12 h dark. Experimental groups were fed three times a day (8:00, 12:00, 16:00) by hand to visual satiation for 12 weeks.

**Chemical analyses.** Proximate analyses of feedstuffs and diets were performed using standard methods (AOAC, 2000). Dry matter was measured by drying at 105°C until a constant weight was reached, crude protein by the Kjeldahl method after acid digestion using the Gerhardt system, crude fat by ether extraction, and crude ash by incineration at 525°C for 12 h in a muffle furnace. Crude fiber was determined by acid alkali hydrolysis and ignition of the dried sample for 3 h. Nitrogen-free extracts (NFE) were calculated as:  $NFE = 100 - (\%protein + \%lipids + \%ash + \%fiber)$ . Gross energy was estimated using the following values: 23.7 kJ/g for protein, 39.5 kJ/g for lipids, and 17.2 kJ/g for carbohydrates (Brett and Groves, 1979). To analyze essential amino acids (excluding tryptophan), about 50 mg of diet was hydrolyzed in 10 ml 6 N HCl at 110°C for 24 h. After removal of the HCl by evaporation under vacuum, the amino acids were separated by ion-exchange chromatography on a Shimadzu RF-10AXL sodium column and detected following postcolumn derivatization with ninhydrin, by measuring absorbance at 350-450 nm.

**Evaluation of fish performance and production costs.** Growth was determined biweekly by collectively weighing the fish from each aquarium. Feed intake was recorded daily to calculate the feed conversion ratio (FCR). The specific growth rate was calculated as  $SGR = 100 \times [(\ln \text{ final fish wt}) - (\ln \text{ initial fish wt})] / \text{no. experimental days}$ , the feed conversion ratio as  $FCR = \text{feed intake (g)} / \text{wt gain (g)}$ , the protein efficiency ratio as  $PER = \text{wt gain (g)} / \text{dietary protein intake (g)} \times 100$ , and the economic conversion ratio as  $ECR = \text{feed cost} \times FCR$ .

Table 1. Formulation, chemical composition, and amino acid contents of experimental diets.

	<i>Dietary protein level (%)</i>			
	35	40	45	50
<i>Ingredient (%)</i>				
Fishmeal	38.0	47.0	56.0	64.0
Soybean meal	10.0	10.0	10.0	10.0
Wheat meal	42.0	34.0	26.0	19.0
Fish oil	7.0	6.0	5.0	4.0
Vitamin	1.0	1.0	1.0	1.0
Mineral	0.8	0.8	0.8	0.8
Binder (guar gum)	1.2	1.2	1.2	1.2
<i>Chemical composition</i>				
Crude protein (%)	35.1	40.8	45.2	50.1
Crude lipid (%)	11.1	11.0	10.9	11.2
Crude ash (%)	7.4	8.7	9.9	11.1
Crude fiber (%)	1.3	1.1	1.0	0.9
Nitrogen free extract (%)	45.1	38.4	33	26.7
Gross energy (MJ/kg)	20.5	20.6	20.7	20.9
<i>Amino acid content (% of protein)</i>				
Arginine	1.9	2.2	2.5	2.7
Cystine	0.4	0.4	0.4	0.5
Histidine	0.8	0.9	1.0	1.1
Isoleucine	1.8	2.1	2.4	2.6
Leucine	2.3	2.7	3.1	3.4
Lysine	2.3	2.8	3.2	3.6
Methionine	0.8	0.9	1.1	1.2
Phenylalanine	1.4	1.6	1.8	2.0
Threonine	1.2	1.4	1.6	1.8
Valine	1.6	1.9	2.2	2.4

*Statistical analysis.* Data were subjected to one-way analysis of variance (ANOVA). When a significant difference was found among treatments, Duncan's multiple range test was performed to rank the groups using Statgraphics 4.0 statistical software (Manugistics Inc., Rockville MD) as per Zar (2001). Estimation of the dietary protein

requirement was made from nonlinear regression of the SGR against the dietary protein level using broken-line model analysis (SAS statistical software) as per Robbins et al. (2006) and second order polynomial analysis using SigmaPlot (Lovell, 1989). All values were considered significant at a 5% level of confidence.

### Results

All the experimental diets were well accepted by the fish. No pathological signs were observed during the trial and no losses occurred. Final weight, SGR, FCR, feed intake (%), PER, feed cost, and ECR are presented in Table 2. The optimal dietary protein requirement for growth was estimated at 38.8% (Fig. 1). Second order polynomial regression analysis showed that the maximum SGR response point ( $Y_{max}$ ) occurred at 46.4% (Fig. 2).

### Discussion

This study indicates that 40.0% dietary protein is the optimal level for weight gain in juvenile electric blue cichlids under the culture conditions used in this study. The broken-line model has been used to determine protein requirements of several fish species (Ng et al., 2001; Kim et al., 2001). Broken-line analysis of the SGR in our study shows that 38.8% dietary protein was optimum when fishmeal was the main protein source and the dietary energy value was 20.7 MJ/kg diet.

Comparison of protein requirements among fish species is complicated by differences in fish size, diet formulation, and culture conditions among studies. The optimum dietary protein level for juvenile electric blue cichlids determined by this study was lower

than for other carnivorous ornamental fish such as 45% for swordtails (*Xiphorus helleri*; Kruger et al., 2001) and discus (*Symphysodon* spp.; Chong et al., 2000), but higher than for herbivorous or omnivorous fish such as 29% for goldfish (*Carassius auratus*; Lochmann and Phillips, 1994) or 25% for dwarf gourami (*Colisa lalia*; Shim et al., 1989). In addition to the difference in species, the high protein requirement of the electric blue may be due to its small size, as protein requirements of fish decrease with increasing size and age (NRC, 1993). The optimum dietary protein level for fish is influenced by the dietary protein:energy ratio, the essential amino acid composition, digestibility of the dietary protein, and the amount of non-protein energy sources in the diet (Akiyama et al., 1997; Wilson, 2002). In this study, dietary essential amino acids were influenced by the protein level as they increased with the dietary protein level.

Our growth performance results compared well with African cichlids in similar experimental conditions (Royes et al., 2005, 2006). For instance, Royes et al. (2005) reported that increasing the protein level for juveniles of the omnivorous *Pseudotropheus socolofi* from 42.5% to 58.9% did not enhance growth performance. Similarly, Royes et al. (2006) did not obtain significant increases in weight gain

Table 2. Growth and feed utilization of electric blue fed experimental diets.

	Dietary protein level (%)			
	35	40	45	50
Initial mean wt (g)	0.57	0.56	0.56	0.57
Final mean wt (g)	2.97±0.03 <sup>a</sup>	3.56±0.09 <sup>b</sup>	3.39±0.08 <sup>b</sup>	3.57±0.22 <sup>b</sup>
Specific growth rate	2.01±0.01 <sup>a</sup>	2.25±0.04 <sup>b</sup>	2.19±0.01 <sup>b</sup>	2.24±0.08 <sup>b</sup>
Feed intake (%)	6.07±0.45 <sup>b</sup>	5.01±0.01 <sup>a</sup>	5.67±0.72 <sup>a</sup>	4.92±0.26 <sup>a</sup>
Feed conversion ratio	2.16±0.15 <sup>b</sup>	1.82±0.06 <sup>a</sup>	1.85±0.02 <sup>a</sup>	1.73±0.09 <sup>a</sup>
Protein efficiency ratio	1.16±0.09 <sup>b</sup>	1.23±0.04 <sup>b</sup>	1.07±0.01 <sup>a</sup>	1.07±0.06 <sup>a</sup>
Feed cost (€/kg)	0.70	0.79	0.87	0.94
Economic conversion ratio (€/kg)	1.52	1.43	1.61	1.63

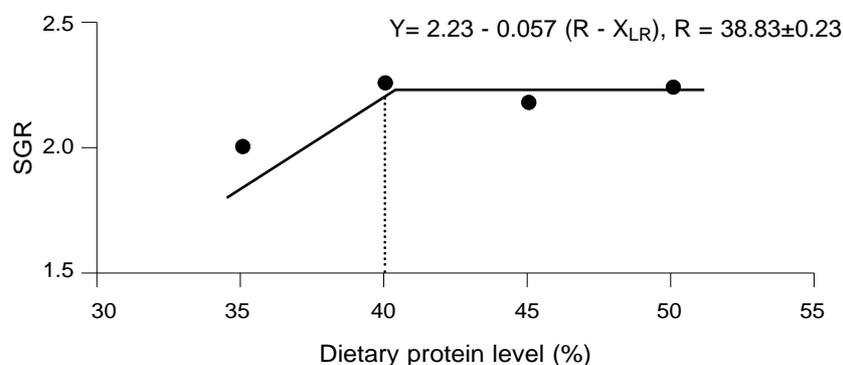


Fig. 1. Broken line analysis of specific growth rate (SGR) in juvenile electric blue cichlids (*Sciaenochromis fryeri*) fed diets with different protein levels.

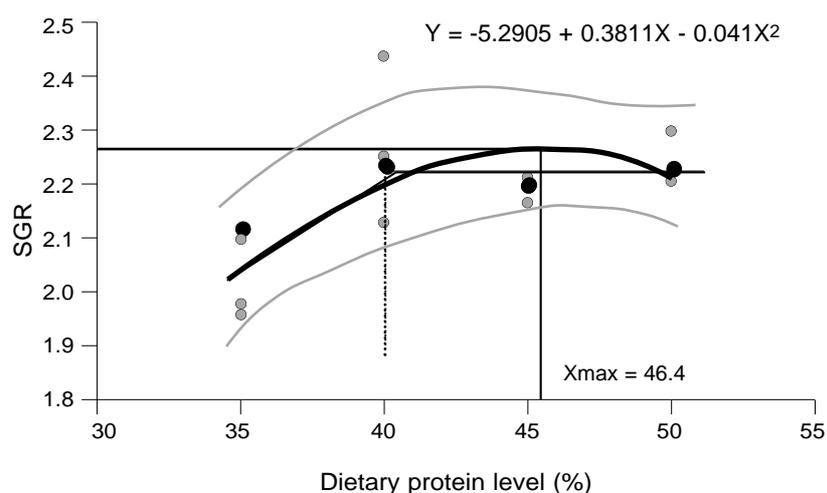


Fig. 2. Second order polynomial relationship between specific growth rate (SGR) and dietary protein levels for juvenile electric blue cichlids (*Sciaenochromis fryeri*) fed diets with different protein levels.

in carnivorous *S. fryeri* fed increasing levels of dietary protein and lipid levels (36-55% protein and 10-20% lipid). They suggest protein and lipid levels of 36% and 10%, respectively. In our experimental conditions, the optimum dietary protein level was determined to be 40% without significant pathological changes to the liver.

Our SGR was lower than in Royes et al. (2006). The slower growth rate seems to be due to a difference in body weight. In Royes et al. (2006), fish with an average weight of about 2.0 g were fed experimental diets for 8 weeks and gained more than a twofold in final weight in all treatment groups. In the present study, fish of 0.5 g were fed experimental

diets for 12 weeks and attained 5.2 to 6.4-fold increases in final weight.

Fish regulate feed intake to satisfy their energy requirements (Wilson, 2002). In this feeding trial, fish were fed to visual satiation and achieved maximum growth by controlling feed intake. In general, the feed intake dropped as the dietary protein level increased. This is in agreement with results for discus (Chong et al., 2000). Likewise, the FCR improved as the protein level increased, well-known in ornamental fish (Kruger et al., 2001; Royes et al., 2005). The FCR in this study was excellent (1.73-2.16) and quite lower than reported for similar small size African cichlids (Royes et al., 2006). The FCR for omnivorous African cichlids *P. socolofi* was 2.75-3.35 (Royes et al., 2005).

The PER tended to decrease as the dietary protein level increased, as reported for other ornamental fish species (Elengovan and Shim, 1997; Royes et al., 2005). The low PER in the groups fed the 45% and 50% protein diets, together with the lack of significant improvement in growth, indicates that excessive protein in those diets was used for metabolic purposes rather than growth. PER in this study increased until the dietary protein requirement was met.

Based on growth performance, PER, and ECR, 39-40% protein in diets with 11% crude lipid and 20.7 MJ gross energy/kg is recommended for juvenile electric blue cichlids under the experimental conditions used in this study.

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