Dietary Protein Requirements of Juvenile Black Sea Bass, *Centropristis striata*

MD. SHAH ALAM¹, WADE O. WATANABE, AND PATRICK M. CARROLL

Center for Marine Science, Aquaculture Program, University of North Carolina Wilmington, 601 S. College Road, Wilmington, North Carolina 28403-5927 USA

Abstract

A feeding trial was conducted in a recirculating system to determine the dietary protein requirement for juvenile black sea bass. Six isocaloric diets were formulated to contain varying levels of crude protein (CP) ranging from 36 to 56% (36, 40, 44, 48, 52, and 56%) by substituting a mixture of carbohydrates and lipid for fish meal. The feeding experiment was carried out in 18-75 L aquaria stocked at a density of 15 juveniles (initial average weight 6.7 g) per tank. Fish were fed test diets in triplicate tanks to apparent satiation twice a day for 8 wk. Whole-body proximate composition was analyzed after the feeding trial. After the feeding trial, weight gain and specific growth rate of fish fed the 44% CP diet were not significantly different from those fed the 48, 52, and 56% CP diets, but were significantly higher (*p* < 0.05) than those fed the 36 and 40% CP diets. Feed conversion efficiency and protein efficiency ratio were significantly affected by dietary protein level. The dietary requirement of protein for maximum growth of black sea bass juveniles, estimated using broken-line regression analysis on weight gain, was 45.3% and maximum weight gain occurred at 52.6% based on polynomial regression analysis.

Black sea bass, *Centropristis striata*, are found in waters along the Atlantic coast from the Gulf of Maine to Northern Florida, and a subspecies inhabits the eastern Gulf of Mexico. Black sea bass are an important commercial and recreational fishery throughout its range (Musick and Mercer 1977; Shepherd and Terceiro 1994) but are currently overfished in many areas and regulated under a variety of management strategies (NCDENR DMF 2006). Landings, however, are not expected to meet increasing consumer demand.

Investigations into commercial production of the black sea bass were initiated in response to the need for diversification of the marine finfish aquaculture industry in the Northeastern USA. Like many other species in the family Serranidae, black sea bass have great potential for mariculture as they are hardy, have high market value, and have limited seasonal supply (Tucker 1984; Kim 1987; Costa and Provenzano 1993; Berlinsky et al. 2000; Walker and Moroney 2000). In response to this, several studies have focused on captive spawning (Watanabe et al. 2003), larviculture (Berlinsky et al. 2000), grow-out of juveniles and subadult captive wild black sea bass (Copeland et al. 2002; Cotton et al. 2003), and economic evaluation of ongrowing of captive wild black sea bass (Copeland et al. 2005). The methodology of spawning and larval rearing is well documented (Watanabe et al. 2003; Copeland and Watanabe 2006), but research is lacking on the growth of hatchery-reared fingerlings to market size. One of the most significant constraints to commercial production of black sea bass is the lack of a reliable diet.

Knowledge of dietary protein requirement of a fish species is of fundamental importance in aquaculture because protein provides the essential amino acids and the nitrogen sources for nonessential amino acid synthesis and also provides energy for maintenance and growth. Protein influences the economics of a farming industry by determining the feed cost, which is typically the largest operational cost in aquaculture (NRC 1993). Basic information on the nutrient requirements of black sea bass is needed to develop a cost-effective commercial feed. A series of nutritional studies were conducted at the University of North Carolina Wilmington

¹ Corresponding author.

© Copyright by the World Aquaculture Society 2008

656
(UNCW), Center for Marine Science (CMS), to develop a cost-effective nutritionally balanced diet for juvenile black sea bass. The present study was designed to quantify the protein requirements of the juvenile black sea bass fed isocaloric diets containing graded levels of protein from 36 to 56%, with 4% increments.

**Materials and Methods**

**Experimental Diets**

Six experimental diets in which herring meal was used as the main protein source were prepared to contain different crude protein (CP) levels of 36, 40, 44, 50, 52, and 56% at the expense of wheat starch, dextrin, and menhaden oil (Table 1). The analyzed values for CP were 37.2, 41.7, 44.9, 49.8, 52.8, and 57% in the diets (Table 1). Krill meal was included (10%) in all diets. All other ingredients were formulated according to the studies on nutrient requirements of other marine fish (Alam et al. 2000, 2003).

<table>
<thead>
<tr>
<th>Ingredients</th>
<th>36%</th>
<th>40%</th>
<th>44%</th>
<th>48%</th>
<th>52%</th>
<th>56%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Herring meal</td>
<td>44</td>
<td>50</td>
<td>56</td>
<td>62</td>
<td>68</td>
<td>74</td>
</tr>
<tr>
<td>Krill meal</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>Menhaden fish oil</td>
<td>9.2</td>
<td>7.8</td>
<td>6.3</td>
<td>4.9</td>
<td>3.4</td>
<td>2.0</td>
</tr>
<tr>
<td>Soybean lecithin</td>
<td>2.5</td>
<td>2.5</td>
<td>2.5</td>
<td>2.5</td>
<td>2.5</td>
<td>2.5</td>
</tr>
<tr>
<td>Carboxymethyl cellulose (binder)</td>
<td>4.5</td>
<td>4.5</td>
<td>4.5</td>
<td>4.5</td>
<td>4.5</td>
<td>4.5</td>
</tr>
<tr>
<td>Wheat starch</td>
<td>10</td>
<td>8</td>
<td>6</td>
<td>4</td>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>Dextrin</td>
<td>5</td>
<td>4</td>
<td>3</td>
<td>2</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Vitamin mix</td>
<td>2.5</td>
<td>2.5</td>
<td>2.5</td>
<td>2.5</td>
<td>2.5</td>
<td>2.5</td>
</tr>
<tr>
<td>Mineral mix</td>
<td>2.5</td>
<td>2.5</td>
<td>2.5</td>
<td>2.5</td>
<td>2.5</td>
<td>2.5</td>
</tr>
<tr>
<td>Attractants</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Cellulose</td>
<td>8.8</td>
<td>7.2</td>
<td>5.7</td>
<td>4.1</td>
<td>2.6</td>
<td>0</td>
</tr>
<tr>
<td>Total</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Gross energy (GE) (calculated, kJ/g diet)</td>
<td>14.4</td>
<td>14.4</td>
<td>13.9</td>
<td>14.0</td>
<td>13.7</td>
<td>13.6</td>
</tr>
<tr>
<td>P/E ratio (mg protein/kJ GE)</td>
<td>25.8</td>
<td>28.9</td>
<td>32.3</td>
<td>35.5</td>
<td>38.5</td>
<td>41.9</td>
</tr>
<tr>
<td>Analyzed proximate composition (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Moisture</td>
<td>11.6</td>
<td>11.1</td>
<td>9.8</td>
<td>9.5</td>
<td>11.9</td>
<td>11.7</td>
</tr>
<tr>
<td>Protein</td>
<td>37.2</td>
<td>41.7</td>
<td>44.9</td>
<td>49.8</td>
<td>52.8</td>
<td>57.0</td>
</tr>
<tr>
<td>Lipid level</td>
<td>15.4</td>
<td>14.5</td>
<td>13.0</td>
<td>12.3</td>
<td>11.6</td>
<td>10.3</td>
</tr>
<tr>
<td>Ash</td>
<td>11.9</td>
<td>12.8</td>
<td>13.9</td>
<td>14.7</td>
<td>15.8</td>
<td>16.7</td>
</tr>
</tbody>
</table>

1 Integral Fish Foods, Inc., Grand Junction, CO, USA.
2 ADM, Co., Decatur, IL, USA.
3 Sigma–Aldrich, St. Louis, MO, USA.
4 Alam et al. (2000).
5 Attractants: alanine, betaine, glycine, and taurine (each 0.25%).
6 VWR International, Co., Suwanee, GA, USA.
temperature oven (DKM 600; Yamato Scientific, Co., Ltd., Tokyo, Japan). The proximate composition of the diets was analyzed (Table 1). The experimental diets were stored at −20°C until used.

**Fish and Experimental Conditions**

The feeding trial was conducted at the UNCW-CMS Aquaculture Facility, Wrightsville Beach, Wilmington, North Carolina. Adult broodstock held in photothermally controlled tanks were induced to spawn using luteinizing hormone-releasing hormone analogue (LHRHa) (Watanabe et al. 2003). Eggs were hatched and reared through juvenile stage in 150-L tanks. These fish were hatched and reared at the same facility where the feeding trial was conducted.

The experimental units consisted of 18 rectangular glass aquaria supported by a recirculating system supplied by filtered seawater. Water level in each tank was maintained at 72 L (holding capacity 75 L). Six triplicate groups of fish (initial body weight 6.7 ± 0.03 g) were randomly distributed into each of 18 tanks (15 fish per tank) and acclimated to the experimental conditions for 1 wk prior to the starting the experiment. Photoperiod was 13:11 (light:dark). Water temperature (23.0 ± 0.5°C, mean ± SD), salinity (33.5 ± 0.61 p.p.t.), pH (7.8 ± 0.5), and dissolved oxygen (6.5 ± 0.5 p.p.m.) were maintained throughout the experimental period. Each tank was covered with a plastic lid to minimize disturbance and to prevent fish from jumping out. Fish were fed to apparent satiation twice per day 0900 and 1600 h as much as they could consume during a 30-min period, and the amount of diet consumed was recorded daily. Fish were bulk weighed every 2 wk, with feed being withdrawn for 24 h prior to weight measurements. The experiment was conducted for 8 wk and at the end of the experiment; five fish from each tank were sacrificed for whole-body proximate analysis.

**Biochemical and Statistical Analyses**

CP (nitrogen combustion; AOAC 1990) and crude fat content (ether extraction; AOAC 1990) of the diets and fish whole body were determined at New Jersey Feed Laboratory, Inc., Trenton, New Jersey. Ash and moisture contents were analyzed by standard methods of AOAC (1990) at UNCW-CMS. All data were subjected to statistical verification using one-way analysis of variance (JMP, version 6.0; SAS Institute, Inc., Cary, NC, USA). Significant differences between means were evaluated by Tukey–Kramer test (Kramer 1956). Ps < 0.05 were considered significant. The optimum dietary protein level was determined according to the broken-line regression method (Robbins et al. 1979). The 95% confidence interval of the break point was estimated as described by Jones and Molitoris (1984). Second-order polynomial regression analysis (Zeitoun et al. 1976) was also used to determine the break point for optimum dietary protein requirement. Regression analysis was performed using software package JMP, version 6.0.

**Results**

Data on mean body weight gain (BWG), specific growth rate (SGR), daily feed intake (FI), feed conversion efficiency (FCE), protein efficiency ratio (PER), and survival of the black sea bass juveniles fed graded levels of protein after 8 wk are shown in Table 2. After the feeding trial, BWG and SGR of fish fed the 44% CP diet were significantly higher than those of fish fed the 36 and 40% CP diets (P < 0.05), but there were no significant differences in these parameters among fish fed the 44, 48, 52, and 56% CP diets (Table 2).

The dietary protein requirement of juvenile black sea bass based on BWG was found to be 45.3% (95% confidence interval 43–48) by broken-line regression analysis (Fig. 1) on the basis of the analyzed protein level in the diets. Second-order polynomial regression analysis showed that the maximum weight gain occurred at 52.6% (Fig. 2). The relationship being $Y = -1557.9650 + 73.4308X - 0.6981X^2$ ($R^2 = 0.88$).

FCE was significantly higher at 44–56% CP (1.44–1.52) than at 36% CP (1.0). But FCE values did not differ between fish fed the 40 and 44% CP. The FCE ($Y$) to dietary levels of protein ($X$) relationship was estimated by the following second-order polynomial regression equation: $Y = -4.9220 + 0.2545X - 0.0025X^2$. 
(\(R^2 = 0.94\)). The best FCE occurred at 50.7% dietary protein.

The highest PER was observed for fish fed the 44% CP. There were no statistical differences in PER among the fish fed 36 and 40% CP diets, whereas the lowest PER was found in fish fed the 56% CP diets. The PER (\(Y\)) to dietary protein levels (\(X\)) relationship was described by the following second-order polynomial regression equation: \(Y = -574.04 + 19.79X ≤ 45.3\), \(Y = 322.74\) when \(X > 45.3\) \((R^2 = 0.81)\). The best PER occurred at 46.4% dietary protein.

The FI data showed a little higher feed consumption for the fish fed below the 44% CP level but did not show any statistical difference among the treatments. No gross deficiency signs were observed in any groups. Survival of the fish among the treatments was high (>93%), with no significant differences. Whole-body moisture and ash contents in the fish fed 36% CP were significantly higher than those in the fish fed the other diets, but lipid content was lowest for the juveniles fed 36% CP (Table 3). However, whole-body protein did not show any significant differences among the dietary treatments.

### Figure 1
Relationship between weight gain of black sea bass juvenile and analyzed dietary protein level as described by the broken-line regression model (Robbins et al. 1979). The requirement of protein for black sea bass juvenile was estimated to be 45.3% of diet.

### Table 2
BWG, SGR, FI, FCE, PER, and survival (SR) of juvenile black sea bass fed diets with graded levels of protein for 8 wk.1

<table>
<thead>
<tr>
<th>CP level in the diets</th>
<th>36%</th>
<th>40%</th>
<th>44%</th>
<th>48%</th>
<th>52%</th>
<th>56%</th>
</tr>
</thead>
<tbody>
<tr>
<td>BWG</td>
<td>175 ± 4.5(^a)</td>
<td>220 ± 6.0(^b)</td>
<td>332 ± 14.9(^c)</td>
<td>319 ± 7.6(^c)</td>
<td>326 ± 12.1(^c)</td>
<td>326 ± 7.4(^c)</td>
</tr>
<tr>
<td>SGR</td>
<td>1.81 ± 0.05(^a)</td>
<td>2.07 ± 0.03(^b)</td>
<td>2.56 ± 0.07(^c)</td>
<td>2.55 ± 0.03(^c)</td>
<td>2.59 ± 0.05(^c)</td>
<td>2.60 ± 0.03(^c)</td>
</tr>
<tr>
<td>FI</td>
<td>1.55 ± 0.02(^a)</td>
<td>1.61 ± 0.10(^a)</td>
<td>1.43 ± 0.07(^a)</td>
<td>1.41 ± 0.08(^a)</td>
<td>1.52 ± 0.08(^a)</td>
<td>1.53 ± 0.03(^a)</td>
</tr>
<tr>
<td>FCE</td>
<td>1.07 ± 0.03(^a)</td>
<td>1.28 ± 0.06(^ab)</td>
<td>1.50 ± 0.05(^b)</td>
<td>1.52 ± 0.04(^b)</td>
<td>1.46 ± 0.08(^b)</td>
<td>1.44 ± 0.03(^b)</td>
</tr>
<tr>
<td>PER</td>
<td>2.70 ± 0.09(^ab)</td>
<td>2.88 ± 0.14(^abc)</td>
<td>3.34 ± 0.12(^c)</td>
<td>3.13 ± 0.15(^bc)</td>
<td>2.77 ± 0.16(^abc)</td>
<td>2.52 ± 0.06(^a)</td>
</tr>
<tr>
<td>SR</td>
<td>91 ± 2(^a)</td>
<td>96 ± 2(^a)</td>
<td>93 ± 3(^a)</td>
<td>95 ± 4(^a)</td>
<td>93 ± 3(^a)</td>
<td>98 ± 2(^a)</td>
</tr>
</tbody>
</table>

1 Values are means ± SEM of triplicate groups. Means with different superscript letters in the same column differ significantly (\(P < 0.05\)).
In the present study, BWG and SGR increased with increasing protein in the diet up to 44% CP. These parameters significantly improved as the dietary protein level increased up to an optimum level, beyond which they plateaued or slightly decreased, consistent with results reported for other species including Arctic char, Salvelinus alpinus (Tabacheck 1986); red drum, Sciaenops ocellatus (Jirsa et al. 1997); and Florida pompano, Trachinotus carolinus (Lazo et al. 1998).

In some species, when the protein requirement is exceeded, the growth rate remains constant or decreases (Jauncey 1982). However, no decrease in growth beyond the optimum requirement level was observed in the present study. A number of studies have compared the growth rates of black sea bass on commercially prepared diets with different protein levels. Cotton and Walker (2004) previously reported that juvenile black sea bass (initial weight 0.82 g) grew well on a high-protein (>50%) commercial feed. Walker and Moroney (2000) found that adult black sea bass fed a salmon diet (45% protein and 26% lipid) grew significantly larger than fish fed a trout diet (38% protein and 8% lipid). Similarly, Berlinsky et al. (2000) found that subadult black sea bass fed a diet containing 52% protein and 18% lipid were significantly larger than fish fed a diet containing 38% protein and 12% lipid. Copeland et al. (2002) also reported higher growth rates in subadult black sea bass fed commercial diets containing between 50 and 56% proteins when compared with diets containing 41–45% protein. All these studies were conducted using different commercial diets where protein sources were not provided, and these studies were not designed to determine optimum requirements.

The quality as well as the quantity of protein in aquafeeds is well recognized (Jauncey 1982; Shiau and Huang 1989). In this study, we used fish with 6.7 g initial weight, and protein source in the diets was mainly herring meal. The nutritive value of dietary protein for fish is influenced by their amino acid composition (Wilson and Poe 1985; Alam et al. 2002). Many factors may affect protein requirements, including species, age, dietary protein sources, amino acid profile, environmental conditions, and experimental design (Tacon and Cowey 1985; Moon and Gatlin 1991; Teshima et al. 2006).

In the present study, fish fed the diet containing 48% CP had the highest FCE (1.52) among all the dietary treatments; however, there was no significant difference among fish fed the

![Second-order polynomial relationship between dietary protein level and body weight gain of juvenile black sea bass. Maximum body weight gain occurred at 52.6% dietary protein.](image)

**FIGURE 2.** Second-order polynomial relationship between dietary protein level and body weight gain of juvenile black sea bass. Maximum body weight gain occurred at 52.6% dietary protein.

**TABLE 3.** Effects of dietary protein level on body composition (% wet basis) of black sea bass.1

<table>
<thead>
<tr>
<th>CP level % diet</th>
<th>Moisture</th>
<th>Crude protein</th>
<th>Crude lipid</th>
<th>Crude ash</th>
</tr>
</thead>
<tbody>
<tr>
<td>36</td>
<td>68.1 ± 0.23b</td>
<td>16.4 ± 0.05a</td>
<td>9.5 ± 0.08a</td>
<td>5.4 ± 0.04b</td>
</tr>
<tr>
<td>40</td>
<td>67.4 ± 0.14ab</td>
<td>16.5 ± 0.19a</td>
<td>10.3 ± 0.10ab</td>
<td>4.5 ± 0.08a</td>
</tr>
<tr>
<td>44</td>
<td>66.5 ± 0.07a</td>
<td>16.6 ± 0.09a</td>
<td>10.9 ± 0.11b</td>
<td>4.4 ± 0.10a</td>
</tr>
<tr>
<td>48</td>
<td>66.3 ± 0.05a</td>
<td>16.8 ± 0.14a</td>
<td>10.7 ± 0.09b</td>
<td>4.5 ± 0.03a</td>
</tr>
<tr>
<td>52</td>
<td>66.7 ± 0.38a</td>
<td>16.4 ± 0.21a</td>
<td>11.1 ± 0.4b</td>
<td>4.6 ± 0.10a</td>
</tr>
<tr>
<td>56</td>
<td>66.1 ± 0.49a</td>
<td>16.8 ± 0.19a</td>
<td>10.6 ± 0.12b</td>
<td>4.4 ± 0.20a</td>
</tr>
</tbody>
</table>

1 Values are means ± SEM of triplicate groups. Means with different superscript letters in the same column differ significantly (P < 0.05).
40–56% CP (Table 2). The FCE values obtained in our study for black sea bass (1.07–1.52) are similar to those of the others species, such as European sea bass (Perez et al. 1997) and Asian sea bass (Catacutan and Coloso 1995). The PER values (3.34) for a dietary protein level of 44% is higher than those obtained in a previous experiment (1.21) where black sea bass subadults were fed commercial diets (Copeland et al. 2002) containing 45% CP. These differences could be related to a more balanced amino acid composition in the practical diets used in this study and to different growth stage of the fish used in these studies. Our PER results indicate that the diets used here were adequate to meet protein requirements of the juveniles. In the present study, FCE and PER were still useful indicators of fish response to dietary protein because FCE showed a similar trend to growth responses, and protein was used less efficiently (PER decreased) with increasing dietary protein above the optimum level (44%).

The CP of whole body did not show any differences related to protein level in the diets. Whole-body moisture concentration decreased with increasing protein level, whereas whole-body lipid increased. These changes in whole-body composition were similar to those observed in yellow puffer (Bai et al. 1999), plaice (Cowey et al. 1972), and Japanese flounder (Kim et al. 2001).

In conclusion, in terms of BWG based on broken-line and polynomial regression analysis of BWG, FCE, and PER, it is recommended that the dietary protein requirement for juvenile black sea bass for maximum growth is greater than 45% but less than 52.6% when herring meal was used as a main protein sources and the diet contains 14.0 kJ/g diet energy.

Acknowledgments
This research was supported by the MARBIONC (Marine Biotechnology in North Carolina) Center for Marine Science, University of North Carolina Wilmington, and United States Department of Agriculture-Cooperative State Research Education and Extension Service (USDA-CSREES). The authors also wish to acknowledge the Lab of Aquatic Animal Nutrition, Kagoshima University, Japan, for providing the vitamin and mineral premix for their study. We thank to Dr. Barry Wray, associate professor of quantitative science (Information System & Operations Management, Cameron School of Business), and Christopher Bentley, UNCW, for their assistance in statistical analysis. We also thank Troy Rezek and Walker D. Wright-Moore for their technical assistance.

Literature Cited


