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The effect of complete substitution of fish-meal with crab-meal at different dietary protein levels on growth and feed utilization of *Heterobranchus longifilis* fingerlings

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Abstract

A feeding experiment was conducted in plastic aquaria ($46 \cdot 38 \cdot 28 \text{ cm}^3$) to investigate the effect of replacing fish-meal (FM) with crab-meal (CM) on growth, feed utilization and body composition of *Heterobranchus longifilis* fingerlings with body weight, $3.11 \pm 0.56 \cdot 8.48 \pm 1.70 \text{ g}$ and total length, $73.07 \pm 4.99 \cdot 98.69 \pm 7.64$ mm. Fish and crab-meal were separately used to produce feeds containing 30, 35 and 40% crude protein (CP) and fed to fish for 10 weeks at 5% body weight. The results show that fish fed with 40% CP fish-meal diet had the best growth as indicated in mean weight gain, $6.99 \pm 1.70 \text{ g}$; specific growth rate, $4.05 \pm 0.19\% \text{ day}^{-1}$; feed conversion ratio, 1.12 ± 0.06 and protein efficiency ratio of 2.28 ± 0.12 . Diets with FM performed better than CM diets. Percentage survival rate for FM and CM containing diets varied between 91.0 ± 1.63 and 100% and the condition of fish was not significantly different (P > 0.05). Apart from whole body levels of CP in FM fed fish, moisture and fat contents were unaffected by dietary treatments. Deficiency in some essential amino acids and higher dietary ash levels in CM probably impaired effective utilization of these diets compared to FM. Crab-meal diets show appreciable performance and could have potential use as a partial replacer of FM at a particular level.

Keywords: Heterobranchus longifilis, fish-meal, replacement crab-meal, growth, food utilization.

INTRODUCTION

Feed is generally acknowledged by fish farmers to be the most expensive input in fish culture operations and accounts for about 70% of the production cost (Omitoyin, 2006). The high cost of fish feeds arise from the inclusion of fish-meal as the main protein source (Lim et al., 1977). Fish-meal is incorporated in fish feed because of its high biological value and excellent protein quality (Adikwu and Haruna, 1999) and palatability (Webster et al., 2000). In order to reduce the high cost of fish-meal without adversely affecting the quality of feed, several studies have been conducted to substitute fishmeal either wholly

or partially with other low-priced alternate and good protein sources (Madu and Akilo, 2001). Notable animal protein source which could replace fish-meal in fish diets among others is the brackish water crab, *Uca tangeri* (Fiddler crab), family Ocypodida. This crab is found in the inter-tidal zone of the Nigerian coast. It lives in burrows and is abundant. This species contains 31.21% CP, 2.4% lipid, 4.98% crude fibre, 43.5% ash and organic content of 56.5% (Deekae and Idoniboye-Obu, 1995). Sesarmahuzardii (Hairy mangrove crab) is also abundant and could be harvested for incorporation as valuable animal

protein source in African giant catfish (Heterobranchuslongifilis) feeds.

Ozogul (2000) reported that crustacean waste products supplemented with amino acids could be used to partially replace fish-meal in rainbow trout diets. Meyers (1987) also stated that crustaceans and molluscs have attractants that contain nitrogenous compounds such as amino acid, peptides, nucleotides and chitin which could promote feed consumption, hence resulting in increased growth. Shrimp waste meal which is an animal protein source of considerable potential (Fanimo et al., 2000; Rutanaporn et al., 2005)) is rich in lysine and chitin (Fanimo et al., 1996). The use of shrimp waste meal may be limited due to its high fibre, chitin and ash contents (Cavalheiro et al., 2007). These substances have been reported to reduce crustacean meal digestibility in tilapia (Koprucu and Ozdemir, 2005), decrease lipid absorption and increase water content in the faeces of the Atlantic salmon, Salmo salar (Olsen et al., 2006).

African giant catfish is a popular freshwater catfish with farmers and consumers in Nigeria and attracts high market value (Alatise et al., 2005). There has not been any documented report on the effect of complete substitution of fish-meal with crab-meal at different dietary protein levels on growth responses and feed utilization of *H. longifilis* fingerlings. This study was therefore carried out to investigate the effect of complete substitution of fish-meal with crab-meal at different dietary protein levels on growth and feed utilization of *H.longifilis* fingerlings.

MATERIALS AND METHODS

Experimental procedure

This study was carried out in the Fisheries Laboratory of Rivers State University of Science and Technology, Port Harcourt, Nigeria between November 2001 and January 2002. Fingerlings of H.longifilis with body weight, 3.11 \pm 0.56 to 8.48 \pm 1.70 g and totallength 73.07 \pm 4.99 to 98.69 \pm 7.64 mm were purchased from the African Regional Aquaculture Centre (ARAC), Aluu-Port Harcourt and used for this research. The fish were transported to the laboratory in plastic bags with well-oxygenated water. The fish were acclimated for 7 days and fed with a commercial feed containing 30% crude protein at 5% body weight.

Ten fish as stocking rate were randomly distributed into each of 18 experimental plastic aquaria of 46 x 38 x 28 cm³ dimension. A constant volume of 40 L of water was maintained throughout the experimental period. Fish in each aquarium were bulk weighed using a triple beam balance (Model MB-2610) and the total length also measured with a metal metre rule at the beginning of the experiment and thereafter on a weekly basis. Six diets containing protein levels 30, 35 and 40% each for FM (*Tilapia*) and CM mixture of *U. tangeri* and *S. huzardii* protein sources (Table 1) were administered simultaneously at 5% fish body weight according to treatment. Each diet was divided into two halves and fed daily at 0800 and 1600 h, respectively. The amount of diet was also adjusted from weekly recorded weights. The feeding trial lasted for 10 weeks.

The six experimental diets were analyzed for moisture content, crude protein, fat, fibre and ash using standard methods (AOAC,

1990) (Table 1). During the experimental period, water quality parameters were determined. The water temperature was measured daily using a laboratory mercury thermometer (0 to 100°C). The dissolved oxygen (DO) was determined by Winkler's method, unionized ammonia by Nesslerization, water hardness was monitored titrimetrically and nitrite, steam distillation method (Boyd, 1979). The pH of the water was measured using a portable pH meter (Model Jenway 3150).

Fish performance parameters

Mean weight gain (MWG), percent weight gain (PWG), mean length increase (MLI), specific growth rate (SGR), condition factor (CF), percentage survival and feed conversion ratio (FCR) were computed as described by Brown (1957), Bagenal and Tesch (1978), Okoye et al. (2001), Adikwu (2003), Alatise and Otubusin (2006) and Orisamuko (2006). The protein efficiency ratio (PER) was according to Zeitoun et al. (1974). At the beginning and end of feeding period, a random sample of 2 fish from each tank was taken, sacrificed and analyzed for proximate compositions using standard methods (AOAC, 1990).

Data analysis

All data were subjected to analysis of variance (ANOVA) and Duncan's Multiple Range Test (Duncan, 1955) to determine their significant differences (P > 0.05). The Statistical Analysis System (SAS) User's Guide, SAS/STAT Version (2003) was used in all the analyses.

RESULTS

The proximate composition of the analyzed experimental diets showed that the expected protein levels of 30, 35 and 40% in the formulations were fairly met (Table 1). Diet CM-30 had a slightly lower protein level of 29.5% CP while Diets FM-40 and CM-40 had crude protein levels of 40.50 and 41.0%. Diets FM-30 and FM-40 had highest lipid levels of 11.49 and 12.94%, respectively. Diet CM-30 contained higher amount of crude fibre (6.45%) and ash (18.15%) than other diets. Crab-meal containing diets had highest ash levels (14.14 to 18.15%) than FM diets (13.07 to 14.25%). Except for diets FM-40 and CM-40, all other diets had a digestible energy ranging 3075.0 to 3404.1 kcal/kg.

The water quality parameters monitored during the experimental period showed that the water temperature was highest for Diet FM-30 with a mean value of 28.0° C. All other diets had a mean temperature of 27.5° C. The mean pH value fluctuated between 7.3 (Diet FM-35) and 7.8 (Diet CM-35). Dissolved oxygen was highest for Diet FM-35 with a mean value of 5.5 mg/l. The mean unionized ammonia (NH₃–N) ranged from 0.02 mg/l (Diet CM-35) to 0.10 mg/l (Diet CM-30); nitrite, 0.36 mg/l (Diet FM-30) to 2.75 mg/l (Diet CM-30) and water hardness ranged from 20.05 mg/l (Diet CM-40) to 55.3 mg/l (Diet CM-35).

Table 2 represents the growth and nutrient utilization parameters. Mean weight gain of fish fed diet containing

Table 1. Ingredients in and proximate (g) of experimental diets fed to Heterobranchus longifilis fingerlings.

Lance Paris	Diets						
Ingredient	FM-30	FM-35	FM-40	CM-30	CM-35	CM-40	
Tilapia fishmeal- (64.25% CP)	35.22	44.53	53.84				
Crab meal- (37.20% CP)				71.00	60.00	11.00	
Soybean meal- (45.24% CP)					25.00	77.15	
Wheat bran- (18.54% CP)	15.00	15.00	15.00	15.00	5.60	5.25	
Corn- (10.55% CP)	43.18	33.87	24.56	7.40	2.80		
Vitamin and mineral premix ^a	0.50	0.50	0.50	0.50	0.50	0.50	
Ascorbic acid ^b	0.10	0.10	0.10	0.10	0.10	0.10	
Table salt	0.20	0.20	0.20	0.20	0.20	0.20	
Red palm oil	2.80	2.80	2.80	2.80	2.80	2.80	
Binder (starch)	3.00	3.00	3.00	3.00	3.00	3.00	
Total	100.0	100.0	100.0	100.0	100.0	100.0	
Analyzed composition (%dry matter)							
Moisture	5.57	5.73	5.53	5.75	5.84	5.67	
Crude protein	30.38	35.25	40.50	29.50	35.50	41.00	
Crude lipid	11.49	9.42	12.94	8.80	10.84	9.28	
Crude fibre	6.25	5.86	5.21	6.45	5.35	4.25	
Ash	13.34	14.25	13.07	18.15	17.78	14.14	
Nitrogen free extract	32.97	29.49	22.75	31.35	24.69	25.66	
Digestible energy (Calculated) ^c , kcal/kg	3388.1	3327.1	3640.3	3075.0	3404.1	3559.0	

Optimix vitamin – mineral premix: Vit. A, D₃, K, B₁, B₂, B₆, B₁₂. Niacin, Pantothenic acid, Folic acid, Biotin, Choline chloride, Antioxidant, Manganese, Zinc, Iron Copper, Iodine, Selenium and Cobalt. Produced by Animal Care for Animal Services Konsult (Nig.) Ltd., Agege, Lagos. Supplied by L-ascorbyl-2 phosphate (25% activity). Digestible energy was calculated as 3.0, 4.25, 3.8 and 8.0 kcal/g of carbohydrate (non-legume), proteins (animal), proteins (plant) and fats (New, 1987). FM: Fish-meal; CM: Crab-meal; CP: Crude protein; 30, 35 and 40: protein levels in %.

Table 2. Effect of protein sources at different levels on growth and feed utilization of *H. longifilis*fingerlings.

Parameter	Fishmeal diets			Crab meal diets			
raiailletei -	30%	35%	40%	30%	35%	40%	
Initial weight of fish (g)	3.11±0.56 ^c	3.98±0.71 ^c	8.48±1.70 ^a	3.57±0.63 ^c	4.13±0.68 ^b	4.40±0.60 ^b	
Final weight (g)	7.30 ^b	5.36 ^c	7.26 ^b	13.71 ^a	5.77 ^c	6.64 ^b	
Weight gain (g)	2.05±0.56 ^b	2.88±0.71 ^D	6.99±1.70 ^a	2.20±0.61 ^b	2.54±0.69 ^b	2.67±0.60 ^b	
Total Length of fish (mm)	73.07±4.99 ^d	79.33±5.98 ^b	98.69±7.64 ^a	76.47±4.17 ^c	80.90±5.05 ^b	82.24±4.38 ^b	
Length gain (mm)	24.64±4.99 ^c	30.20±5.98 ^b	43.58±7.64 ^a	16.03±3.82 ^e	20.00±5.04 ^d	18.24±4.38 ^d	
Weight Gain (%)	65.92 ^c	72.36 ^b	82.43 ^a	61.62 ^c	61.50 ^c	60.68 ^d	
Specific growth rate (%day ⁻¹)	2.14±0.20 ^b	2.92±0.06 ^b	4.05±0.19 ^a	2.01±0.13 ^b	2.07±0.09 ^b	2.28±0.06 ^b	
Condition factor (K)	0.72±0.01	0.74±0.02	0.77±0.02	0.72±0.01	0.73±0.01	0.74±0.02	
Survival (%)	97.0±0.81 ^b	96.0±0.66 ^b	100.0±0 ^a	94.50±1.16 ^c	97.36±0.44 ^b	91.0±1.63 ^d	
Feed conversion ratio	2.64±0.74 ^a	1.56±0.04 ^b	1.12±0.06 ^b	2.42±0.32 ^a	2.17±0.17 ^a	2.07±0.07 ^a	
Protein efficiency ratio	1.61±0.15 ⁰	1.84±0.05 ^D	2.28±0.12 ^a	1.51±0.11 ⁰	1.30±007 ⁰	1.21±0.04 ^D	

Means with same letter for a given parameter in same horizontal row are not significantly different (P>0.5). 30, 35, 40: Protein levels in %.

FM with 40% protein was significantly higher (P < 0.05) than fish fed 30 or 35% dietary protein levels in all diets. Fishmeal diets at 30, 35 and 40% CP had the best weight gains than fish fed CM diets. Diets FM–40 had highest PWG (82.43%) and SGR (4.05 \pm 0.197% day ⁻¹). Crabmeal diets resulted in least PWG with values ranging

from 60.68 to 61.62% while SGR values ranged 2.01 \pm 0.13 – 2.28 \pm 0.06% day $^{-1}$. Feed conversion ratios were significantly better in diets FM-40 (1.12 \pm 0.6) and FM-35 (1.56 \pm 0.04) than CM diets with FCR range of 2.07 \pm 0.07 - 2.42 \pm 0.32. Fish-meal diet containing 30%CP had the poorest FCR, 2.64 \pm 0.74. The results of protein

Table 3. Proximate body composition (% wet weight) of *H. longifilis* fingerlings before and after feeding graded protein levelsfrom different sources for 70 days.

Parameter -	Nutrient parameters						
	Moisture (%)	Crude protein (%)	Crude lipid (%)	Ash (%)			
Fish at start of experiment	15.14 ^c	78.15 ^a	1.15 ^c	3.26 ^a			
FM-30	17.71 ^b	71.25 ^c	2.54 ^b	3.45 ^a			
FM-35	18.83 ^b	72.58 ^b	2.72 ^a	3.06 ^a			
FM-40	22.92 ^a	70.59 ^c	2.18 ^b	2.95 ^a			
CM-30	17.62 ^b	73.05 ^b	1.85 ^c	3.35 ^a			
CM-35	17.90 ^b	73.85 ^b	3.38 ^a	3.12 ^a			
CM-40	17.64 ⁰	73.92 ^D	2.93 ^a	2.82 ^a			

Values within the same vertical row with same superscripts are not significantly different (P>0.05). FM = Fish meal; CM = Crab meal.

utilization obtained in all the diets showed FM-40 diet to be significantly higher (P < 0.05), 2.28 \pm 0.12 than other diets. Similarly, crab-meal diet of 30% CP (1.51 \pm 0.11) had better PER as FM-30 diet (1.61 \pm 0.15). Percentage survival ranged from 91.0 \pm 1.63% to 100% in the treatments.

The body composition values are given in Table 3. Moisture ranged from 70.59% (Diet FM–40) -73.92% (Diet CM–40). Carcass protein ranged from 15.14% in fish at the beginning of trial to 22.92% in FM containing diet of 40%CP which was significantly higher (P < 0.05) than other diets. Carcass protein in fish fed crab-meal diets ranged 17.62 to 17.90% and similar to that of FM–30 fed fish (17.71%). Carcass lipid content was significantly higher (P < 0.05) in fish fed crab-meal diets with 35 and 40% CP levels. Ash content showed no significant differences (P > 0.05) in all diets.

DISCUSSION

Water quality parameters were not significantly different between treatments and were within the recommended range for the culture of *Clarias gariepinus* (Viveen at al., 1986). The good water quality observed during the experimental period probably favoured the increase in size and higher percentage survival of *H. longifilis* fed FM and CM diets.

Protein requirement for optimal growth and feed efficiency of fingerling fish usually ranged from 40 to 45% depending upon species (Harpaz et al., 2001). Cowey et al. (1972) stated that Plaice (*Pleuronectess platessa*), a carnivorous fish required 40 to 45% dietary protein. The percent range of dietary crude protein (30 to 40%) used in this study was equally found to maximize good growth of carnivorous fish such as *H. longifilis*. The increase in weight gain obtained indicates that the diets were capable of supporting growth. However, the 40% FM protein containing diet resulted in highest growth. The CM diets even at 40% CP performed less. Similar results using FM gave best growth at 40% CP for pure *H*.

longifilis fingerlings (Eyo, 1995), and *C. gariepinus* (Maduand Olurebi, 1987).

The trend in fish growth across the diets with CM as total replacement for FM shows decrease. This could be due to lower feed consumption, stress resulting from excessive amounts of ash and slightly lower survival of fish in CM containing diets. Growth response is depended on dietary protein level and quality. Fish meal appears superior in essential amino acids balance and high digestibility (Dabrowski and Kozak, 1979) compared to CM as a protein source.

The percent weight gain (PWG) and SGR increased almost linearly with increasing dietary protein with FM containing diet at 40% CP, producing maximum growth rates than CM diets as in common carp (Ogino and Saito, 1970). A probable reason for the difference in growth rate was that the diets were not equal in amino acids, digestible energy and environmental (Olufeagba et al., 2002). Fish fed FM diets showed slight signs of superiority in condition and survival. Feed conversion ratio (FCR) decreased with dietary protein levels from 30 to 40% in both FM and CM diets. The small variation in the FCR values obtained probably reflects similarity in the composition of the diets from a protein source (Hepher, 1988). The mean FCR values of 2.07 ± 0.07 to 2.42 ± 0.32 for *H. longifilis* in this study compared favourably with those of gariepinusfingerlings (Alegbeleve et al., 2001), Heterobranchus bidorsalis fry (Dada et al., 2001) and C. gariepinus fedfermented shrimp head waste meal (Nwanna, 2003).

The PER values increased with increasing dietary protein in FM diets and were also higher than those of CM fed fish. This result indicated that fish fed FM diets utilized the protein more than those fed CM protein source. The CM fed fish might have found it increasingly necessary to metabolize part of the protein consumed when FM was completely replaced in the diets. Hence, utilization of protein for incorporation into the body structure was consequently reduced. The observation on reduced growth pattern with CM diets as indicative of lower PER values could be adduced to quality and profile

of some essential amino acids (Carlos et al., 1988) and higher dietary ash levels coupled with lower digestibility (Keremah, 2008). Due to this low digestibility chitin found in crustaceans and molluscs could physically block the access of digestible enzymes to lipids and proteins, thus affecting the utilization of these nutrients (Karasov, 1990 cited by Jean et al., 2012). Apart from some variations in the body protein content, *H. longifilis* fingerlings in this study have not shown any noticeable variations in body of moisture, fat and ash.

Conclusion

This study showed that fish fed fish-meal diet had the best growth and nutrient utilization than crab meal as a complete fish protein replacer. The higher ash level in crab meal wholly could not give the best growth performance in this instance but may have the potential use as a partial replacer at a particular level of inclusion in *H. longifilis* diets. This conclusion is based on the fact that crab-meal has the potential and compares favourably in biological performance with FM diets of 30% CP and had 50% performance of 35% CP and 40% CP fish meal diets. Further research is required to evaluate the influence of CM at various levels of FM substitution in fish diets.

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