DIETARY CRUDE PROTEIN AND DIGESTIBLE ENERGY REQUIREMENTS OF *Heterobranchus bidorsalis* (Geoffrey St. Hilaire, 1809) FINGERLINGS

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ABSTRACT

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This study determined the optimum dietary crude protein and digestible energy requirement for clariid catfish (Heterobranchus bidorsalis) fingerlings which is commonly farmed in tropical world. Sixteen (16) practical diets were formulated and used in the feeding trials. The diets were made up of four (4) digestible energy levels (2400, 2600, 2800 and 3000 kcal kg^{-1}), each at four (4) crude protein levels, (25, 30, 35 and 40%). Fingerlings were sourced from the hatchery unit of the Experimental Fish Farm in the Department of Fisheries, University of Benin, Benin City, Nigeria. Fingerlings were fed for 70days in three (3) replicates for each treatment. The general aim of this study is to determine quantitatively, the optimum protein and energy levels for catfish (Heterobranchus bidorsalis) fingerlings for optimum performance using locally sourced feed stuffs. Weekly data were collected based on weight gain and feed intake. Feed and fish carcasses were analyzed for proximate composition. All data collected were subjected to two-way analysis of variance. The total feed intake increased with increase in dietary protein level up to 35% crude protein which corresponded with the optimal level especially for absolute growth rate, specific growth rate (SGR), feed conversion ratio (FCR) and utilization. In terms of caloric intake, optimum level stood at 2600-2800 kcalkg^{-1}for H. bidorsalis fingerlings. interaction between probability significantly influenced feed intake (FI), specific growth rate (SGR), feed conversion ratio (FCR), crude protein (CP), net protein utilization (NPU) and protein efficiency ratio (PER) by H. bidorsalis fingerlings were optimized at 40% crude protein and 2800 kcalkg^{-1} digestible Energy (DE). In general, a lower limit of 35% CP and an upper limit of 40% were found optimal and may be recommended for catfish fingerlings in Nigeria. Similarly, lower limit 2600 KcalKg^{-1} and an upper limit of 2800 kcalkg^{-1} digestible energy are recommended.

Keywords*: Heterobranchus bidorsalis, fingerlings, optimum energy, protein levels, fish diet.*

INTRODUCTION

Aquaculture requires optimization of nutrition to efficiently raise fish for the purpose of food production. Nutrition plays a critical role in intensive aquaculture as it influences not only the production cost but also fish growth, health and waste production and waste management. Fish nutrition is the study of nutrients and energy sources essential for fish health, growth and reproduction (Hixson, 2014). Fish requires high quality nutritionally balanced diet for growth and attainment of market size within the shortest possible time (Gabriel *et al.,* 2007). Catfish farming has continued to attract private sector initiative compared to earlier public or government-sponsored programmes (Adewumi and Olaleye, 2011). *Clarias gariepinus* is one of the two main genera of the African mud catfish (*Clarias* and *Heterobranchus*) widely cultured in Africa, Asia and Europe (Adewolu and Adoti, 2010). This is due to their outstanding culture characteristics such as ability to withstand unfavorable environmental conditions, efficient in utilizing various types of locally formulated fish feed, resistance to diseases, high economic potential and simple techniques in the propagation of their fingerlings (Owodeinde and Ndimele, 2011).The purpose of this study is to ascertain the optimum growth and utilization responses to crude protein and digestible energy variation of *Heterobranchus bidorsalis* fingerlings

MATERIALS AND METHODS

Feeding trials were conducted to determine the optimum protein and digestible energy levels for the clariid catfish (*H. Bidorsalis*) fingerings in the Experimental Fish Farm of Department of Fisheries, University of Benin, Benin City, Nigeria.

Experimental diets

Sixteen (16) diets were used for the feeding trials. The diets were formulated to contain four digestible energy (DE) levels of 2400, 2600, 2800 and 3000 KcalKg^{-1}, each at four (4) crude protein levels of 25, 30, 35 and 40% respectively. The layout of the dietary treatment is shown in Table 1. Each diet constituted a treatment. Detail of nutrient composition of feedstuffs of experimental diets and proximate analysis is shown in Table 2. The levels of feed ingredients used to formulate the diets were adjusted to obtain the desired levels of DE and CP. Calculation of the DE levels of the diets were based on the cumulative of DE of the ingredients as recommended for channel catfish (*Heterobranchus bidorsalis*) by Lovell (1984). The recommended levels of crude protein, lysine and methionine by New (1987) were used. These values are shown in Table 2. In preparing the diets, ingredients were milled, mixed and prepared as described by Martinez-Palacios *et al*. (1996). The milled ingredients were sieved through standard sieve Nos. 16 and 20 (maximum of 1.19mm). The homogenous feed combinations were

processed into pellets or granules (2 mm) with gelatinized corn starch component as the binder. After the preparation, pelleted diets were oven-dried at 70 °c for 24 hours. Feed samples were stored in polythene bags in cupboard at laboratory temperature. Dried granules of feed samples were taken for proximate analysis. All ingredients were locally sourced for the study.

Table 1: Dietary treatment combinations used in the experiment

Digestible Energy (DE kcal kg^{-1})	Diets (% Crude protein)			
	25%	30%	35%	40%
2400	2400(1)	2400(5)	2400(9)	2400(13)
2600	2600(2)	2600(6)	2600(10)	2600(14)
2800	2800(3)	2800(7)	2800(11)	2800(15)
3000	3000(4)	3000(8)	3000(12)	3000(16)

Numbers in parenthesis represent the various treatment codes.

There were four trials, one trial for each type of feed. Glass tank was used for the trials. Each tank was connected to a central aerator. Water supplied by the University of Benin Campus Domestic Water Services was maintained at 35 litre mark/level throughout the experiment. Fingerings were fed test-diets twice daily during daylight (9:30 am and 4:00pm). At each time of feeding, animals were fed to satiation [i.e. hand fed access to food, during which diet was provided in small amount at a time, so that the fish will eat nearly all the diet offered]. Water temperature was measured twice daily during feeding. Dissolved oxygen (DO) was measured once a week using Winkler's method. Daily observations were made to detect any abnormality and fish mortality. Unconsumed diets and faecal wastes were removed by siphoning daily. Each trial lasted 70days. Weight of fish per treatment and per replicate was recorded weekly. Weight of food consumed by fish was also recorded weekly for each replicate. In order to obtain the weights of the fish, fish were batch weighted in a dish containing pre-weighed water.

Carcass Analysis

All the diets and carcasses were subjected to proximate analysis at the end of the trials. Crude protein (N X 6.25) was determined by the micro-kjeldahl method and crude fibre (CF) was by the system based on acid-alkaline digestion. Lipids, ash and moisture were determined using standard methods in triplicate (AOAC, 1990).

Growth and Nutrient Utilization indices

Weight of fish and quantity of feed consumed were obtained at weekly intervals from each replicates of each treatment. From the values recorded, the following parameters were determined:

Weight Gain (WG) = $W_2 - W_{1\,(g)}$

Where; W_1 = initial weight

W_2 = final weight

Feed Intake = initial weight of feed – final weight of feed

Specific Growth Rate (SGR) % = $\frac{\text{Loge W2} - \text{loge W1}}{\text{T2} - \text{T1}}$ X 100

Where: T_1 and T_2 are time of experiment in days.

W_2 = final weight at T_2

W_1 = initial weight at T_1

Loge = natural logarithm.

Relative Weight Gain (RWG) % = $\frac{\text{Weight Gain}}{\text{Initial Weight}}$ X 100

Food Conversion Ratio (FCR) = $\frac{\text{Feed Intake(g)}}{\text{Wet Weight Gain(g)}}$

Protein Efficiency Ratio (PER) = $\frac{\text{Weight Gain (g)}}{\text{Protein Intake}}$

Net Protein Utilization (NPU) = $\frac{\text{Final body protein–Initial body protein}}{\text{Protein Intake}}$ X 100

Statistical analysis

At the end of the experiments data were subjected to a one-way ANOVA test using a Genstat software eight edition, 2005 package for statistical problems. All the means were compared at 5% level of probability with Duncan multiple range tests. Similarly, responsiveness of fingerlings to treatments was evaluated.

RESULTS

The results obtained from the study are shown in Table 3 and 4. Dietary treatments had significant effects on all parameters studied. Weight gain varied between 3.33g and 40.38g. The highest weight gain was obtained in diet with 35% crude protein (CP) at Digestible Energy (DE) level of 2600 $kcalkg^{-1}$. The lowest weight gain was obtained on the diet with 25% CP and DE of 3000 $kcalkg^{-1}$. At each of the DE level employed, the weight gain obtained on the 35% CP diet was significantly higher than those fed diets with 25, 30 or 40%CP except at DE level of 2800 $kcalkg^{-1}$ where the difference between 35% and 40% CP were not significant. Generally, at each DE level, weight gain increased significantly with increase in dietary crude protein level up to 35% and thereafter

decreased except for the case of diet with 2800 kcalkg^{-1} diet. The relative growth rate (RGR), % relative weight gain (RWG) and specific growth rate (SGR) followed similar trend as the body weight gain.
At dietary CP levels of 25% and 30%, weight gain decreased with increase in dietary DE level. At 35% CP level, weight gain increased slightly (not significantly) when the level was 2600 kcalkg^{-1} compared to 2400 kcal kg^{-1} diet and thereafter decreased significantly when the DE level was raised to 2800 and 3000 kcal kg^{-1} diet. It can be concluded from these observations that 35% CP level gave the highest weight gain while 2400 and 2600 kcalkg^{-1} had similar and highest weight gains when compared to the other DE levels. Data in this study showed that the highest weight gain was obtained from the 35% CP diet. Concurrently, the highest weight gain was obtained in the diet containing 2400K kcal kg^{-1} diet.

DISCUSSION

The difference between these two diets in terms of weight gain,, absolute growth rate (AGR) and specific growth rate (SGR) was not significant ($p>0.05$). The highest weight gain, AGR and SGR were obtained at 35% crude protein. Therefore, for *H. bidorsalis* dietary crude protein level of 35% could be recommended as the optimum level. Feed intake increased with increase in dietary crude protein up to 35% and decreased thereafter. Similar responses were reported by Cowey *et al.* (1972) for carp, *Cyprinus carpiio,* Jauncey (1982 for *Oreochromis mossambicus*), De Silva *et al.* (1989: from *Oreochromis niloticus*) and Obasa and Faturoti (2004:in *Chrysichthys walker*). This observed value fell within the acceptable concentration ranges for channel catfish (Boyd, 1979). The SGR peaked at the optimum crude protein level and decreased thereafter. This observation can be attributed to the fact that as the dietary CP increases, the nutritional quality of feed also improves and this encourage enhanced feed intake. The recommended CP level of 35% also falls within the CP recommended levels for *H. longifilis* fingerlings (FAO, 2012). On the basis of feed conversion ratio (FCR), the differences between the 35% and 40%CP diet, at all DE levels tested were not significant.
A major reason why the results of energy requirements obtained in this study cannot easily be compared with other energy requirements recommended elsewhere in Nigeria, is due to the differences in measured energy used. While some researchers used metabolizable energy as their measure (Obasa and Faturoti, 2004), Dada *et al.* (2001) and Ovie *et al.* (2005) used gross energy. The present requirements were based on digestible energy. It is therefore proposed that a standard be established for the measure of energy in fish studies. Gross energy is not a practical measure or indicator of usable energy because of differences in the digestibility of certain ingredients. The general trend of decreasing PER and NPU with increasing dietary CP is in agreement with previous findings for fish species (Cowey *et al*., 1974; Davies and Stickney, 1978; Jauncey, 1982; Martinez-Palacios *et al.,* 1986). Considering the DE level alone, PER and NPU increased with increase in dietary DE; the probable explanation is that as the energy level in the diet increases, and less or no protein is diverted for use as energy. Protein is thus available for use in growth determinations and this means higher protein efficiency in promoting weight gain. However, when the crude protein levels were considered alongside the DE levels, the linear relationships between PER/NPU and CP level as well as between PER/NPU and dietary DE level were not clear-cut. The result of the present study suggests that PER and NPU are probably not decisive enough for determining the optimum level of crude protein in fish diets.

CONCLUSION

The predicted crude protein and DE levels for *Heterobranchus bidorsalis* are of 35% and 2600 kcalkg^{-1} respectively.

Table 2: Ingredient Composition and Proximate Analysis of Experimental Diets (%).

Ingredient	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
Maize	29.79	27.29	24.79	22.29	18.29	19.20	19.79	17.79	24.44	22.94	20.44	17.44	3.44	6.14	13.44	10.94
Fishmeal	7.00	7.00	7.00	7.00	14.50	14.50	14.50	14.50	21.40	21.40	21.40	21.40	26.40	26.40	26.40	26.40
Soybean meal	16.77	16.77	16.77	16.77	18.77	18.77	18.77	18.77	20.20	20.20	20.20	20.20	23.20	24.20	24.20	24.20
Brewer's yeast	12.77	12.77	12.77	12.77	20.20	17.86	14.77	14.77	16.40	16.40	16.40	16.40	27.40	25.40	18.10	18.40
Wheat bran	27.58	27.58	27.58	27.58	23.60	23.08	22.58	22.58	14.10	14.10	14.10	14.10	16.10	14.40	14.40	14.10
Soybean oil	2.63	5.13	7.63	10.13	1.18	3.13	6.13	8.13	0.00	1.50	4.00	7.00	0.00	0.00	0.00	2.50
Bonemeal	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00
Vit. Premix	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40
Vitamin E	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06	0.06
Analyses																
DE calculated	2400	2600	2800	3000	2400	2600	2800	3000	2400	2600	2800	3000	2400	2600	2800	3000
CP calculated	25.0	25.0	25.0	25.0	30.0	30.0	30.0	30.0	35.0	35.0	35.0	35.0	40.0	40.0	40.0	40.0
CP Analysed	24.92	24.68	24.44	24.20	32.51	31.54	31.54	30.02	35.54	35.33	35.15	34.86	43.47	43.10	40.54	40.38
Moisture (%)	8.08	8.14	8.05	8.31	8.16	8.09	8.06	9.06	9.07	8.82	8.91	8.69	8.71	8.84	9.01	9.02
Lipid (%)	3.51	6.03	8.09	11.01	2.05	3.56	6.01	8.57	1.59	3.41	4.91	7.53	2.05	2.31	2.45	4.70
Crude fibre(%)	7.69	7.81	7.07	7.41	7.43	7.38	7.49	7.47	5.64	5.61	5.60	6.01	5.07	4.91	4.03	4.01
Ash (%)	8.01	8.03	8.41	8.50	8.09	8.61	8.19	8.08	7.72	7.69	7.71	7.81	7.70	7.57	7.49	7.53
Lysine calculated	5.68	6.54	6.54	6.51	7.83	7.90	9.30	8.41	7.49	7.46	7.43	7.37	8.30	8.65	8.30	6.17
Methionine calculated	2.76	3.40	3.40	3.50	3.77	3.76	5.33	4.02	3.59	3.54	5.52	4.25	3.83	4.20	4.20	3.58

Table 3: Effect of dietary protein and energy levels on growth performance and feed utilization by *H. bidorsalis* fingerlings

Parameter	Protein (%)					DE Kcal kg^{-1}			
	25	30	35	40	SEM	2400	2600	2800	3000
Total Weight gain	6.26d	8.63c	34.98a	30.88b	0.65	24.25a	22.42b	18.75c	15.31d
Relative weight gain	123.00b	203b	799.00a	747.00a	0.30	677.00a	671.00a	447.41a	263.00b
Absolute growth rate ($gfish_{-1}day^{-1}$)	0.02d	0.04c	0.13a	0.11	0.01	0.09a	0.08a	0.68a	0.59a
Specific growth rate (SGR)	0.49c	0.67b	1.21a	1.27a	0.07	1.11a	1.04a	0.75b	0.73b
Relative growth rate ($gday^{-1}$)	0.02b	0.03b	0.10a	0.11a	0.03	0.08a	0.10a	0.03b	0.04b
Feed intake (g)	14.93d	16.28c	50.10b	53.02a	0.14	47.63a	39.68b	28.73c	18.45d
Feed conversion ratio (FCR)	2.38d	1.89c	1.43a	1.72b	0.06	1.96d	1.77d	1.54b	1.21a
Crude protein intake (g)	3.73	4.84	17.53	21.28	0.15	5.48	4.65	3.48	2.20
Protein efficiency ratio (PER)	2.93a	2.26ab	2.13b	1.83b	0.37	1.62c	1.94c	2.42b	3.24a
Net protein utilization (%)	32.37a	30.19b	31.96ab	23.38c	0.86	23.93c	24.70c	31.96b	37.41a

Within protein or energy levels, values in a column with similar superscripts are not significantly different (p>0.05)

Table 4: Effect of varying dietary levels of protein and energy on the growth performance and feed utilization of *H.bidorsalis* fingerlings

Dietary Treatment		TWG	RGR	AGR	RWG	SGR	FI	FCR	FE	PI	PER	NPU
A	2400 KcalKg^{-1}											
	25% protein	10.00g	0.02cd	0.04ef	161de	0.60ef	30.60g	3.10f	0.33h	7.65b	1.31h	8.70ij
	30% protein	13.33a	0.04cd	0.05e	412bcde	1.01cd	26.70j	2.00d	0.50fg	8.01b	1.66fg	50.72ef
	35% protein	39.16a	0.07bcd	0.13ab	799abcd	1.37abc	64.80c	1.79c	0.55cde	22.68a	1.73f	23.13efg
	40%protein	34.57b	0.16ab	0.13b	837ab	1.46ab	68.40a	1.98d	0.51efg	27.36a	1.26g	18.64j
B	2600 KcalKg^{-1}											
	25% protein	6.72h	0.02cd	0.02fg	133de	0.49ef	16.20k	2.41c	0.42g	4.05d	1.66fg	8.40fgh
	30% protein	10.01g	0.04cd	0.04ef	181.7cde	0.66ef	19.50i	1.95d	0.51ef	5.85c	1.71fg	12.31efg
	35% protein	40.38a	0.21a	0.15a	987a	1.59a	65.40b	1.62bc	0.62cd	22.89a	1.75f	24.90ghi
	40%protein	32.57bc	0.13abc	0.12bc	892abc	1.43ab	57.60d	1.77c	0.57def	23.04b	1.41gh	19.49hij
C	2800 KcalKg^{-1}											
	25% protein	5.00hi	0.02d	0.02fg	130de	0.50ef	8.10m	1.62bc	0.62cd	2.03m	2.46bc	9.85bc
	30% protein	6.34hi	0.02cd	0.02fg	133de	0.53ef	11.10j	1.75c	0.57def	3.33k	1.90ef	8.11e
	35% protein	30.97cd	0.07bcd	0.11bc	457bcde	0.84de	38.40e	1.24a	0.81b	13.44d	2.30b	26.12cd
	40%protein	32.70bc	0.08bcd	0.12bc	524bcde	1.14bcd	27.30i	1.75c	0.57def	10.92b	2.99a	37.91hij
D	3000 KcalKg^{-1}											
	25% protein	5.33c	0.01d	0.01g	76e	0.35f	4.80n	1.44b	0.69c	1.20n	2.78a	12.50a
	30% protein	14.84hi	0.02d	0.04ef	103e	0.44f	7.80m	1.61bc	0.64cd	2.34i	2.07de	67.52d
	35% protein	29.40d	0.06bcd	0.10c	453bcde	1.06cd	32.40f	1.10a	0.91a	11.34f	2.59ab	32.36b
	40%protein	23.69e	0.07bcd	0.08d	440bcde	1.06cd	28.80h	1.22a	0.82ab	11.52e	2.06de	24.74e

Within protein or energy levels, values in a column with similar superscripts are not significantly different (P>0.05)
TWG = Total Weight gain, AGR = Absolute growth rate (g/fish/day), RWG = Relative weight gain, SGR = Specific growth rate, RGR= Relative growth rate (gday^{-1}), FI = Feed intake (g), FCR = Feed conversion ratio, CP = Crude protein intake (g), PER = Protein efficiency ratio, and NPU = Net protein utilization (%)

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