PJN
ISSN 1680-5194

Pakistan Journal of Nutrition

ANSInet
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan
Mob: +92 300 3008585, Fax: +92 41 8815544
E-mail: editorpjn@gmail.com

OPEN ACCESS Pakistan Journal of Nutrition

ISSN 1680-5194
DOI: 10.3923/pjn.2018.627.633

Research Article
Effect of Dietary Protein and Energy Levels on Growth, Feed Utilization and Body Composition of Juvenile African bonytongue (*Heterotis niloticus*)

[1,3]Tra Florent Goure-Bi, [1]Celestin Melecony Ble, [2]Assoi Olivier Etchian, [1]Yao Laurent Alla and [3]Nahoua Issa Ouattara

[1]Center for Research of Oceanology, Department of Aquaculture, P.O. Box V 18, Abidjan, Côte d'Ivoire
[2]Laboratory of Biology and Animal Cytology, UFR-SN, University Nangui Abrogoua, 02 P.O. Box 801 Abidjan 02, Côte d'Ivoire
[3]Laboratory of Hydrobiology and Water Ecotoxicology, University Felix Houphouet-Boigny, 01 P.O. Box V 34 Abidjan 01, Côte d'Ivoire

Abstract
Background and Objective: In Cote d'Ivoire, the tilapia *Oreochromis niloticus* is the major species in fish farming, but its production remains low because of the high cost of feed for this fish. Several species with aquaculture potential exist but are produced on a small scale by a few fish farmers. The diversification of fish species through large scale farming of African bonytongue *Heterotis niloticus* (*H. niloticus*) is an option to increase fish production and ensure the sustainable development of aquaculture. The aim of this study was to determine the level of dietary protein and energy for optimal growth of *H. niloticus* juveniles to produce feed based on local agricultural by-products for the intensive production of this species. **Methodology:** An 80 day feeding trial was conducted in hapas installed in earthen ponds to evaluate the effects of dietary protein and energy levels on growth, feed utilization and body composition of *H. niloticus* fingerlings. Fingerlings with an initial body weight of 17.83±0.45 g were fed 16 experimental diets formulated to contain 4 protein levels (25, 30, 35 and 40%) and 5 levels of gross energy (17, 18, 19, 20 and 21 kJ g^{-1}) with three replicates per treatment. **Results:** Final body weight (FBW), specific growth rate (SGR) and feed conversion ratio (FCR) varied significantly with dietary protein level. For the same dietary protein level (25% and 30%), FBW and SGR increased with the increase in dietary energy content. Beyond 40% protein level, further increase in energy level (18 to 21 kJ g^{-1}) caused a decline in FBW. FCR increased with increasing protein and decreasing energy level in their diet, whereas protein efficiency ratio (PER) decreased with increasing dietary protein and increasing energy level. The whole-body protein and lipid contents were significantly affected by dietary protein level, but whole body composition was not significantly affected by energy level. The results indicate that the best total production (3.94±0.43 t ha^{-1} $year^{-1}$), SGR (3.72±0.05% day^{-1}), FCR (0.75±0.01) and PER were obtained with fish fed on a diet containing 30% dietary protein and a gross energy level of 19 Kj g^{-1}. **Conclusion:** We suggest that a diet with 30% dietary protein and 19 kJ g^{-1} is recommended for the best growth at this specific stage of *H. niloticus* fingerlings.

Key words: *Heterotis niloticus*, dietary protein, optimal growth, fish farmers, fingerling

Received: May 15, 2018 Accepted: August 02, 2018 Published: November 15, 2018

Citation: Tra Florent Goure-Bi, Celestin Melecony Ble, Assoi Olivier Etchian, Yao Laurent Alla, Nahoua Issa Ouattara, 2018. Effect of dietary protein and energy levels on growth, feed utilization and body composition of juvenile African bonytongue (*Heterotis niloticus*). Pak. J. Nutr., 17: 627-633.

Corresponding Author: Celestin Melecony Ble, Center for Research of Oceanology, Department of Aquiculture, P.O. Box V 18, Abidjan, Côte d'Ivoire

Competing Interest: The author has declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Aquaculture production has increased sharply in the last decade[1]. This increased production is aimed to satisfy the global demand for fish protein, due to the decline in capture fisheries. However, intensification of aquaculture production requires the use of good quality feed. The quality of fish feed depends on the quality of the ingredients and nutrients and their ability to satisfy nutritional requirements. Fish and other animals need nutrients and energy to perform their vital functions. Dietary protein provides amino acids that constitute the preferential nutrients used for energy production in fish[2]. Protein is used for somatic growth and to satisfy maintenance energy requirements[3]. Several studies have shown that growth (specific growth rate) and feed utilization (feed conversion ratio, protein efficiency) are influenced by dietary protein and energy level[4,5,6,7]. However, the major sources of protein, such as fishmeal, remain among the most expensive ingredients used in the formulation of farmed fish feed[8,7]. Therefore, a good balance between protein and energy level in the diet to achieve optimal fish growth at a minimal production cost is necessary[3]. Protein requirements depend on fish size, age, physiological status, species and environmental conditions[9,7].

The African bonytongue (*Heterotis niloticus*) is present in river basins in sub-Saharan Africa, where it is exploited by fisheries. It has interesting potential for fish farming, such as a high growth rate, omnivorous diet, a commercial size of up to 7 kg and being valued for human consumption[10]. In Côte d'Ivoire, *H. niloticus* is exclusively reared in polyculture in association with the Nile tilapia (*Oreochromis niloticus*) in extensive fish farming systems. Better knowledge of its nutritional needs is necessary to promote intensive production of this fish. This study was initiated to determine the effect of dietary protein and energy levels on growth, feed utilization and body composition of *H. niloticus* juveniles.

MATERIALS AND METHODS

Experimental fish rearing system: The study was carried out at the experimental fish station of the "Association Fish Farming and Rural Development in Humid Tropical Africa" (APDRACI) located in Daloa city (Central-West of Cote d'Ivoire). *H. niloticus* juveniles, with an average initial weight of 17.83±0.45 g, were used for the experiments and taken from the station stock. The fish were stored in a 400 m² pond for seven days before the start of the feeding trial to acclimate them to the experimental conditions.

The experimental system consisted of 48 rectangular hapas in fine mesh nets installed in 16 earthen ponds. Each hapa was 20 m² (8×2.5 m) and had a water level of 0.75 m. The fish were randomly distributed in hapas for feeding trials. During the feeding trial, the edges and bottoms of the hapas were brushed every week to minimize algal growth and promote natural feeding.

Water temperature 3C(T°), pH and dissolved oxygen were measured every week (at 9:00 and 14:00 h) using multiparameter BANTE 900P. The recorded water physio-chemical parameters were relatively constant in the hapas and were not significantly affected by the different treatments: pH value ranged between 6.19±0.11 and 6.63±0.43; temperature was between 26.18±2.00 and 28.67±0.77°C; and dissolved oxygen was between 6.11±1.04 and 6.91±1.63 mg L^{-1}.

Experimental diets and feeding: Sixteen diets were formulated to contain four protein levels (25, 30, 35 and 40%) and energy levels ranged from 17 to 20 kJ g^{-1}. The diets were prepared with local ingredients (fish meal, soybean meal, cotton flour, corn flour, low rice flour) available on the market. These ingredients were previously analyzed to determine their chemical composition (Table 1). Before mixing, the solid ingredients were finely ground and sieved using a 400-micron sieve. For each experimental diet, the powdered ingredients were weighed and mixed until a homogeneous powder was obtained. Oil, minerals and premix vitamins were added gradually, followed by the addition of water until a consistent dough was achieved. The diet mixtures were pelleted using a kitchen meat grinder with a 3 mm diameter grinder plate (Panasonic MK-G 1800P). The formulation and proximate composition of the diets are given in Table 2. The fish in the hapas were fed experimental diets at 5% of their biomass, twice daily (9 am and 4 pm) for a period of 90 days. The treatments were made in triplicate for each experimental diet. At the beginning of the feeding trial, 5 fish were taken and

Table 1: Proximate composition of ingredients (dry matter %)

	Proximate composition				
Ingredients	Moisture	Protein	Lipid	Ash	NFE[2]
Fish meal	3.20	60.3	8.75	20.35	8.67
Soybean meal	7.59	42.25	4.30	5.90	39.28
Cotton flour	4.50	40.00	3.10	7.10	61.28
Corn flour	9.08	10.85	5.23	1.67	68.57
Low rice flour	7.80	18.81	3.90	5.56	62.80

[2]NFE: Nitrogen-free extract = 100-(Protein (%)+Lipid (%)+Moisture (%)+Ash (%)+Fiber (%))

Table 2: Formulation and proximate composition of experimental diets (% dry matter basis)

Experimental diets	D1-1	D1-2	D1-3	D1-4	D2-1	D2-2	D2-3	D2-4	D3-1	D3-2	D3-3	D3-4	D4-1	D4-2	D4-3	D4-4
Protein level	25%				30%				35%				40%			
Energy level (KJ g^{-1})	17	18	19	20	17	18	19	20	17	18	19	20	18	19	20	21
Ingredient (%)																
Fishmeal	14.0	16.0	17.0	16.0	21.0	22.0	23.0	23.0	29.0	29.0	33.0	34.0	36.0	38.0	40.0	43.0
Corn flour	36.0	33.0	30.0	25.0	30.0	29.0	23.0	19.0	20.0	19.0	12.0	9.0	9.0	5.0	4.0	2.0
Cotton flour	12.0	12.0	11.0	11.0	16.0	15.0	15.0	15.0	16.0	16.0	14.0	14.0	18.0	17.0	15.0	12.0
Soybean meal	10.0	10.0	10.0	11.0	14.0	14.0	14.0	14.0	16.0	17.0	14.0	14.0	20.0	20.0	21.0	21.0
Low rice flour	20.0	17.0	17.0	19.0	12.0	12.0	13.0	12.0	14.0	12.0	14.0	13.0	10.0	10.0	5.0	3.0
Palm oil	4.0	8.0	11.0	15.0	3.0	5.0	9.0	14.0	2.0	4.0	10.0	13.0	4.0	7.0	12.0	16.0
Vitamin premix[1]	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Mineral premix[2]	2.8	2.8	2.8	28.0	2.8	2.8	2.8	28.0	2.8	2.8	2.8	28.0	2.8	2.8	2.8	28.0
Proximate composition (%)																
Protein	25.14	25.45	25.32	24.98	30.49	30.58	30.72	30.10	35.45	35.39	35.35	35.44	40.22	40.59	40.37	40.38
Lipid	8.69	12.59	15.49	19.26	7.97	9.98	13.79	18.54	7.31	9.23	15.10	17.99	9.43	12.36	17.27	21.26
Ash	6.07	6.25	6.33	6.22	7.45	7.56	7.72	7.60	9.14	9.07	9.56	9.66	10.53	10.80	10.81	11.06
Moisture	10.39	13.32	13.30	9.87	11.68	13.16	11.48	9.53	9.34	11.04	9.18	10.51	12.25	13.28	10.18	11.34
Fiber	6.80	6.33	5.98	5.97	6.67	6.42	6.20	5.90	6.45	6.25	5.54	5.28	6.08	5.67	4.77	3.87
NFE	49.74	45.97	43.39	41.52	45.23	44.01	40.62	37.25	41.10	39.56	33.96	31.36	34.46	31.28	26.79	22.59
Gross energy (KJ g^{-1})	17.52	18.45	19.11	20.18	17.64	18.24	19.17	20.30	17.78	18.24	19.56	20.25	18.52	19.19	20.28	21.11
Protein/Energy (mg kJ^{-1})	18.45	17.28	16.33	15.00	21.38	20.92	19.77	17.91	24.30	25.28	21.90	20.89	26.88	25.72	23.62	22.28

[1]Vitamine premix (UI): Vitamin A 1 760 000 IU, vitamin D3 880 000 IU, vitamin E 22 000 mg, vitamin B1 4 400 mg, vitamin B2 5 280 mg, vitamin B6 4 400 mg, vitamin B 1 236 mg, vitamin C 151 000 mg, vitamin K 4 400 mg, vitamin P 35 200 mg, folic acid 880 mg, choline chloride 220 000 mg, pantothenic acid D 14 080 mg. [2]Mineral premix (Per kg premix): Cobalt 20 mg, iron 17 600 mg, iodine 2 000 mg, copper 1 600 mg, zinc 60 000 mg, manganese 10 000 mg, selenium 40 mg

kept frozen (-20°C) for subsequent whole-body proximate analysis. At the end of the experiment, each fish was weighed and three individuals from the same hapa were taken and kept frozen (-20°C) for further determination of whole-body composition.

Biochemical analysis: Biochemical analyses (moisture, crude proteins, crude lipids, fibre and ash) of the diets and whole-body fish were performed in duplicate using the standard AOAC[11] methods.

Growth parameters: Growth performance and feed utilization were described using the following parameters:

$$\text{Survival rate (\%)} = 100 \times \frac{\text{Final number of fish}}{\text{Initial number fish}}$$

$$\text{Daily weight gain (DWG) (g d}^{-1}\text{)} = \frac{\text{FBW- IBW}}{\text{d}}$$

$$\text{Specific growth rate (SGR) (\% d}^{-1}\text{)} = \text{ ln FBW-ln IBW} \times \frac{100}{\text{d}}$$

$$K = \frac{100 \times \text{FBW}}{\text{Lt}^3}$$

$$\text{Voluntary Intake (VI) (\% d}^{-1}\text{)} = 100 \times \text{D}/[(\text{Bi+Bf})/2)]/\text{d}$$

where, IBW (g) is the initial mean body weight, FBW (g) is the final mean body weight, Lt (cm) is the total length of the fish, D (g) is the cumulative amount of feed distributed, Bi and Bf (g) were the initial and final biomass and d is the duration of the experiment. Biomass = IBW or FBW x initial or final fish number.

$$\text{Feed conversion ratio (FCR)} = \frac{\text{Dry feed intake (g)}}{\text{Body weight gain (g)}}$$

$$\text{Protein efficiency ratio (PER)} = \frac{\text{Body weight gain (g)}}{\text{Protein intake (g)}}$$

Statistical analysis: Data were analyzed using one way analysis of variance (ANOVA) after prior verification of the homogeneity of the variances and the normality of the data to be analyzed. When significant differences were found, a Tukey HSD test was used for multiple comparisons at the 5% level of significance. All statistical analyses were performed using Statistica® software version 7.1 for windows.

RESULTS

The growth performance results are shown in Table 3. Fish survival rate varied slightly between treatments and no significant differences were found due to the feeding. The condition factor (K) ranged from 0.51 to 1.15, with the highest

Table 3: Growth performance of *H. niloticus* juvenile fed experimental diets containing different levels of protein and energy for 80-days trial

			Growth parameters						
Experimental diets	Protein level	Energy level (kJ g^{-1})	IBW (g)	FBW (g)	Survival (%)	DWG (g d^{-1})	SGR (% d^{-1})	Yield (t ha^{-1} $year^{-1}$)	k
D1-1	25%	17	17.69 ± 0.90^{a}	146.10 ± 5.10^{g}	82.22 ± 9.62	1.61 ± 0.07^{g}	2.64 ± 0.09^{e}	1.20 ± 0.13^{c}	0.51 ± 0.06^{c}
D1-2		18	17.67 ± 0.29^{a}	153.99 ± 3.69^{fg}	87.78 ± 9.62	1.70 ± 0.05^{fg}	2.71 ± 0.05^{e}	1.40 ± 0.17^{c}	0.59 ± 0.07^{c}
D1-3		19	17.70 ± 0.75^{a}	164.28 ± 12.05^{f}	87.78 ± 9.62	1.83 ± 0.16^{f}	2.78 ± 0.14^{e}	1.52 ± 0.33^{cd}	0.58 ± 0.01^{c}
D1-4		20	17.78 ± 0.75^{a}	229.22 ± 7.30^{e}	86.22 ± 9.62	2.64 ± 0.10^{e}	3.19 ± 0.08^{d}	2.02 ± 0.22^{bcd}	0.78 ± 0.10^{c}
D2-1	30%	17	17.66 ± 0.67^{a}	281.35 ± 3.84^{d}	86.67 ± 16.67	3.30 ± 0.05^{d}	3.46 ± 0.04^{c}	2.33 ± 0.67^{bcd}	0.90 ± 0.07^{b}
D2-2		18	17.99 ± 0.46^{a}	295.56 ± 1.64^{d}	83.33 ± 16.67	3.47 ± 0.03^{d}	3.50 ± 0.03^{bc}	3.12 ± 0.66^{ab}	0.91 ± 0.03^{b}
D2-3		19	17.50 ± 0.5^{a}	343.30 ± 2.56^{a}	88.89 ± 9.62	4.08 ± 0.04^{a}	3.72 ± 0.05^{a}	3.94 ± 0.43^{a}	1.15 ± 0.05^{b}
D2-4		20	17.99 ± 0.35^{a}	338.58 ± 2.20^{a}	87.78 ± 2546	4.01 ± 0.03^{ab}	3.67 ± 0.03^{ab}	3.35 ± 1.17^{ab}	1.06 ± 0.04^{a}
D3-1	35%	17	17.99 ± 0.35^{a}	337.20 ± 3.36^{ab}	87.78 ± 9.62	3.99 ± 0.04^{ab}	3.66 ± 0.01^{ab}	3.34 ± 0.44^{ab}	0.99 ± 0.04^{a}
D3-2		18	17.94 ± 0.09^{a}	338.54 ± 1.61^{a}	82.22 ± 9.61	4.00 ± 0.02^{ab}	3.67 ± 0.02^{ab}	3.10 ± 0.46^{ab}	1.01 ± 0.03^{a}
D3-3		19	17.94 ± 0.09^{a}	339.54 ± 1.69^{a}	82.22 ± 9.61	4.02 ± 0.02^{a}	3.68 ± 0.01^{a}	3.11 ± 0.44^{ab}	0.98 ± 0.06^{a}
D3-4		20	17.94 ± 0.10^{a}	336.49 ± 4.78^{ab}	82.22 ± 9.61	3.98 ± 0.06^{ab}	3.66 ± 0.02^{ab}	3.08 ± 0.42^{abc}	0.97 ± 0.003^{a}
D4-1	40%	18	17.95 ± 0.40^{a}	330.76 ± 2.79^{abc}	82.22 ± 9.61	3.31 ± 0.03^{abc}	3.64 ± 0.02^{ab}	3.03 ± 0.45^{abc}	0.94 ± 0.04^{b}
D4-2		19	17.93 ± 0.36^{a}	323.24 ± 4.65^{bc}	87.77 ± 9.61	3.82 ± 0.06^{bc}	3.62 ± 0.03^{abc}	3.20 ± 0.44^{ab}	0.85 ± 0.08^{b}
D4-3		20	17.86 ± 0.68^{a}	319.33 ± 6.86^{c}	86.67 ± 16.67	3.77 ± 0.09^{c}	3.60 ± 0.06^{abc}	2.67 ± 0.74^{abc}	0.89 ± 0.07^{b}
D4-4		21	17.76 ± 0.51^{a}	281.29 ± 1.81^{d}	86.67 ± 0.05	3.29 ± 0.03^{d}	3.45 ± 0.04^{c}	2.33 ± 0.03^{bcd}	0.84 ± 0.04^{b}
Test			ns	0.00000	ns	0.00000	0.00000	0.000002	0.00000

Values are presented as average±standard deviation. Significance of ANOVA 1 test: The values of the same column having at least one letter in common are not significantly different ($p>0.05$), ns = not significant effect

Table 4: Feed utilization of *H. niloticus* juvenile fed experimental diets containing different levels of protein and energy for 80-days trial

			Feed utilization parameters			
Experimental diets	Protein level	Energy Level (g kJ^{-1})	FCR	FER	PER	VI (%/j)
D1-1	25%	17	1.27 ± 0.14^{c}	0.79 ± 0.09^{d}	3.58 ± 0.16^{cde}	2.26 ± 0.25^{bc}
D1-2		18	1.29 ± 0.07^{c}	0.77 ± 0.04^{d}	3.39 ± 0.04^{cde}	2.39 ± 0.15^{c}
D1-3		19	1.27 ± 0.25^{c}	0.81 ± 0.15^{cd}	3.43 ± 0.05^{cde}	2.36 ± 0.28^{c}
D1-4		20	1.13 ± 0.12^{bc}	0.89 ± 0.09^{bcd}	3.85 ± 0.33^{abcd}	2.27 ± 0.27^{bc}
D2-1	30%	17	0.97 ± 0.01^{abc}	1.03 ± 0.01^{abcd}	3.71 ± 0.11^{bcde}	1.99 ± 0.13^{abc}
D2-2		18	0.87 ± 0.15^{ab}	1.17 ± 0.19^{abc}	4.05 ± 0.40^{abc}	1.87 ± 0.26^{abc}
D2-3		19	0.75 ± 0.01^{a}	1.34 ± 0.02^{a}	4.58 ± 0.16^{a}	1.66 ± 0.05^{a}
D2-4		20	0.79 ± 0.09^{ab}	1.27 ± 0.14^{a}	4.43 ± 0.27^{ab}	1.70 ± 0.10^{ab}
D3-1	35%	17	0.79 ± 0.06^{ab}	1.33 ± 0.01^{a}	3.87 ± 0.01^{bcd}	1.71 ± 0.10^{ab}
D3-2		18	0.80 ± 0.04^{ab}	1.25 ± 0.06^{ab}	3.77 ± 0.10^{bcd}	1.72 ± 0.05^{ab}
D3-3		19	0.84 ± 0.10^{ab}	1.20 ± 0.14^{ab}	3.65 ± 0.27^{cde}	1.81 ± 0.20^{abc}
D3-4		20	0.86 ± 0.09^{ab}	1.17 ± 0.12^{abc}	3.65 ± 0.16^{cde}	1.84 ± 0.18^{abc}
D4-1	40%	18	0.94 ± 0.13^{abc}	1.08 ± 0.17^{abcd}	3.00 ± 0.25^{e}	2.02 ± 0.26^{abc}
D4-2		19	0.84 ± 0.06^{ab}	1.19 ± 0.08^{ab}	3.11 ± 0.15^{de}	1.81 ± 0.09^{abc}
D4-3		20	0.98 ± 0.21^{abc}	1.05 ± 0.23^{abcd}	3.00 ± 0.31^{e}	2.04 ± 0.33^{abc}
D4-4		21	0.95 ± 0.09^{abc}	1.05 ± 0.10^{abcd}	2.99 ± 0.23^{e}	1.97 ± 0.20^{abc}
Test			0.000002**	0.000004**	0.000000**	0.000183**

Values are presented as average±standard deviation. Significance of ANOVA 1 test : The values of the same column having at least one letter in common are not significantly different ($p>0.05$), ns = not significant effect, *$p<0.05$, **$p<0.01$, ***$p<0.001$

value obtained in the fish fed diet D2-3 (30% protein level, 19 kJ g^{-1} energy level). Increasing dietary protein and energy level, up to 30% protein content and 19 kJ^{-1} of energy level, significantly ($p<0.05$) increased final body weight (FBW). Beyond that, a decrease in growth appeared when protein and energy level increased in the diet. The highest FBW mean (343.30±2.56g) was found in fish fed with diet D2-3. The same trend was observed in daily weight gain (DWG) and specific growth rate (SGR). Fish biomass at the end of feeding trial was affected by diet composition ($p<0.05$). However, the Tukey HSD test showed that the yields of fish fed with diets containing a protein level greater than 30% were not significantly different ($p>0.05$). Fish fed with diet D2-3 showed greater production (3.94±0.43 t ha^{-1} $year^{-1}$) than those fed with the other experimental diets.

The effects of dietary protein and energy levels on feed utilization efficiency are presented in Table 4. Voluntary intake (VI), with statistically identical overall values, ranged from 1.66±0.05 to 2.39±0.15% d^{-1}. In regard to the feed conversion ratio (FCR), the results showed that this

Table 5: Whole-body composition of *H. niloticus* juvenile fed experimental diets containing different levels of protein and energy for 80 days trial

Experimental diets	Protein level	Energy Level (g kJ^{-1})	Whole-body composition			
			Moisture	Protein	Lipid	Ash
Initial			82.22	16.83	8.84	4.37
D1-1	25%	17	78.54±0.39[c]	18.86±0.90[a]	11.20±2.91[a]	4.67±0.40
D1-2		18	76.99±0.4[b]	19.05±1.10[a]	12.09±0.69[a]	3.07±1.53
D1-3		19	77.41±1.38[b]	19.25±1.06[b]	12.02±0.02[a]	3.65±0.97
D1-4		20	74.30±1.02[b]	19.50±1.30[b]	14.63±0.72[b]	2.28±0.43
D2-1	30%	17	75.92±0.24[b]	19.92±0.96[b]	17.77±2.95[c]	3.17±0.25
D2-2		18	77.10±2.69[b]	20.00±0.39[b]	17.72±1.30[c]	3.67±0.12
D2-3		19	73.72±1.2[a]	20.05±0.07[c]	17.11±0.19[b]	3.75±0.28
D2-4		20	74.04±0.83[b]	20.05±0.67[c]	16.87±0.37[b]	2.97±1.13
D3-1	35%	17	75.17±0.46[b]	20.05±0.83[c]	16.21±1.93[b]	4.70±2.39
D3-2		18	76.89±0.67[b]	19.80±1.05[b]	12.52±1.21[a]	4.37±0.83
D3-3		19	77.03±0.05	19.86±0.96[b]	15.71±1.54[b]	3.67±0.69
D3-4		20	78.30±1.58[b]	20.15±1.20[c]	14.12±0.30[b]	2.97±1.13
D4-1	40%	18	75.75±1.0[b]	20.35±0.97[c]	14.95±1.59[b]	3.65±0.97
D4-2		19	78.30±0.04[b]	19.76±0.58[b]	14.17±1.24[b]	2.37±0.56
D4-3		20	74.09±1.10[b]	19.75±1.37[b]	14.68±1.60[b]	2.37±0.83
D4-4		21	75.67±1.97[b]	19.35±1.90[b]	14.53±1.29[b]	2.48±1.54
Test			0.006	0.0024	0.00014	ns

Values are presented as average±standard deviation. Significance of ANOVA 1 test: The values of the same column having at least one letter in common are not significantly different (p>0.05), ns = not significant effect

nutritional parameter was significantly affected by dietary protein and energy level interaction. For the same dietary protein level, FCR decreased with increasing energy content up to 30% protein level. Beyond a 30% dietary protein level, FCR did not vary significantly regardless of the energy level in the diet.

Dietary protein intake showed two trends in the protein efficiency ratio (PER). For dietary protein levels ranging from 25-30%, PER increased with increasing energy content, whereas PER decreased with increasing energy content for 35-40% protein levels. The highest value (4.58±0.16) of PER was obtained in fish fed with diet D2-3.

Fish carcass analysis showed significant differences ($p<0.05$) in whole-body composition (Table 5). Whole-body protein levels tended to increase with increasing dietary protein and energy contents and this was observed in fish fed with dietary protein levels ranging from 25-30%. In contrast, whole-body protein content appeared to be relatively variable in fish fed with dietary protein levels greater than 30%. The lowest body protein content (18.86±0.90%) was observed in fish fed with diet D1-1 and the highest (20.35±0.97%) was obtained in fish fed with diet D4-1. The same trend was observed with body lipid levels; the highest values (approximately 17%) were found in fish fed with 30% dietary protein levels.

The fish body moisture content at the end of the experiment was lower compared to the initial state. However, fish fed with experimental diets showed significant differences in body moisture content. The highest value (78.54±0.39) of whole-body moisture was obtained in fish fed with diet D1-1 and the lowest (73.72±1.2) in those fed with diet D2-3. However, the Tukey HSD test revealed that fish fed with diet D2-3 had the lowest body moisture content, significantly different from the others.

Table 5 shows that the ash whole-body content ranged from 2.37±0.83 to 4.67±0.4 and was not significantly different in fish subjected to the different dietary protein and energy levels.

DISCUSSION

Survival rates obtained in this study were similar to those found by Monentcham *et al.*[10], in *H. niloticus* juveniles with an average weight between 2 and 62 g. Additionally, values of condition factors (k) indicate that the fish were overweight and in good condition during the experiment as suggested by Bagenal and Tesch[12]. Overall, "k" values were similar to those determined in *H. niloticus* in the Amassoma flood plain in the Niger Delta[13].

Results of growth performance and feed utilization showed that increasing dietary protein and energy level increased final body weight (FBW) up to 30% protein content and 19 kJ^{-1} energy level. At 35% of protein intake and beyond, the growth appeared stable or even decreased. Based on these results, 30% dietary protein with 19 kJ^{-1} energy is considered as the optimum dietary protein and energy level for the optimal growth of *H. niloticus* juveniles. Similar growth patterns have been reported in previous studies. In juveniles of *H. niloticus* (3-15 g), the highest weight gain was obtained with fish fed 30% dietary protein and 18 kJ g^{-1} energy level[10]. In juveniles of Pirarucu, a species close to *H. niloticus*, an increase in weight gain with increasing dietary digestible protein (up to 36.7%) was shown[14]. Results of the present study are also consistent with findings reported for other species[15,16,17]. The previous studies have shown that increasing the protein content in the diet led to an increase in fish growth up to a threshold where growth reached a plateau; this could be explained by the physiological role of proteins in fish growth[3,7]. At adequate levels, proteins and amino acids are used to improve growth performance and to satisfy energy requirements, whereas in excess, the energy cost resulting from the catabolism of these nutrients does not allow for further increase in fish growth[3].

Results of feed utilization showed that FCR and PER were influenced by dietary protein and energy levels. FCR (1.27-0.75) improved with increasing energy content up to 30% dietary protein level, and, in this range of protein level, PER (2.99-4.58) increased with increasing energy content. Beyond 30% dietary protein, regardless of the energy level in the diet, PER decreased with increasing proteins level. These results suggested that the level of proteins for fish optimal growth would be approximately 30%; this is in agreement with results reported for other carnivorous fish species[3,7]. According to Kim *et al.*[7], the decrease in PER indicates an efficient use of dietary protein by fish for body protein synthesis. This was consistent with the observation that an increase in the level of dietary protein led to an increase in fish body protein content. Several previous studies[7,14,18,19] on fish have shown that increasing body protein content are associated with increasing dietary protein level. Kim and Lee[20], explained that body proteins were dependent on dietary protein intake and their increase was related to the dietary protein level that allowed maximum growth in fish. On the other hand, dietary energy and protein levels affected body lipid content in fish fed with a diet containing protein levels between 25 and 30%, whereas no significant difference was observed between fish fed with 35 and 40% dietary protein, regardless of the energy level in the diet. Similar results have been reported for body lipids in *Cyprinus carpio* juveniles fed with gradual levels of dietary protein[21]. These results are in agreement with those found in juvenile parrot fish (*Oplegnathus fasciatus*) fed with diets containing protein levels ranging from 35 to 60%[7]. Body moisture and ash contents varied slightly between treatments and this trend was similar to those obtained in other studies for juvenile *H. niloticus* and Pirarucu[10,13].

CONCLUSION

The results of this study showed that *H. niloticus* juveniles were well adapted to breeding conditions in the hapas with better survival rates. It has been shown that dietary protein and energy levels affected growth and feed utilization. The suitable dietary protein and energy levels for optimal growth were found to be 30% and 19 mg kJ^{-1} respectively.

SIGNIFICANCE STATEMENT

This research evaluated the dietary protein and energy level for optimal growth of juvenile *Heterotis niloticus* with an initial average weight of 23 g, which could benefit feed manufacturers and fish farmers by allowing them to formulate quality foods for intensive production of this fish. The results of this study will enable researchers to better understand the nutritional requirements of this species with high aquaculture potential. Thus, a new theory can be used in the formulation of fish feed.

ACKNOWLEDGMENTS

The experiments of this study were carried out at the experimental fish station of the NGO APDRACI. The authors thank the heads of the NGO and all the technicians who participated in the follow-up of the experiments.

REFERENCES

1. FAO., 2016. The State of World Fisheries and Aquaculture 2016: Contributing to Food Security and Nutrition for All. Food and Agriculture Organization, Rome, Italy, ISBN: 9789251091852, Pages: 200.
2. Medale, J. and J. Guillaume, 1999. Nutrition Energtique. In: Nutrition et Alimentation des Poissons et Crustaces, Guillaume, J., S. Kaushik, P. Bergot and R. Metailler (Eds.)., INRA., Paris, pp: 87-111.
3. Hecht, T., A. Irish and J. Sales, 2003. Effect of protein level and varying protein-lipid concentrations on growth characteristics of juvenile spotted grunter *Pomadasys commersonnii* (Haemulidae). Afr. J. Mar. Sci., 25: 283-288.
4. Sweilum, M.A., M.M. Abdella and S.A. Salah El Din, 2005. Effect of dietary protein energy levels and fish initial sizes on growth rate, development and production of Nile tilapia, *Oreochromis niloticus* L. Aquacult. Res., 36: 1414-1421.
5. El-Saidy, D.M.S.D. and M.M.A. Gaber, 2005. Effect of dietary protein levels and feeding rates on growth performance, production traits and body composition of Nile tilapia, *Oreochromis niloticus* (L.) cultured in concrete tanks. Aquacult. Res., 36: 163-171.

6. Zuanon, J.A.S., A.L. Salaro, S.S.S. Moraes, L.M.D.O. Alves, E.M. Balbino and E.S. Araujo, 2009. Dietary protein and energy requirements of juvenile freshwater angelfish. Rev. Bras. Zootec., 38: 989-993.
7. Kim, K.W., M. Moniruzzaman, K.D. Kim, H.S. Han, H. Yun, S. Lee and S.C. Bai, 2016. Effects of dietary protein levels on growth performance and body composition of juvenile parrot fish, *Oplegnathus fasciatus*. Int. Aquatic Res., 8: 239-245.
8. Mohseni, M., M. Pourkazemi, M.R. Hosseni, M.H.S. Hassani and S.C. Bai, 2013. Effects of the dietary protein levels and the protein to energy ratio in sub-yearling persian sturgeon, *Acipenser persicus* (Borodin). Aquacult. Res., 44: 378-387.
9. Guillaume, J., S. Kaushik, P. Bergot and R. Metailler, 1999. Nutrition et Alimentation des Poissons et Crustaces. INRA., Paris, Pages: 485.
10. Monentcham, S.E., V. Pouomogne and P. Kestemont, 2010. Influence of dietary protein levels on growth performance and body composition of African bonytongue fingerlings, *Heterotis niloticus* (Cuvier, 1829). Aquacult. Nutr., 16: 144-152.
11. AOAC., 1995. Official Methods of Analysis of Association of Analytical Chemist International. 16th Edn., AOAC, Washington DC., Pages: 1094.
12. Bagenal, T.B. and A.T. Tesch, 1978. Conditions and Growth Patterns in Fresh Water Habitats. Blackwell Scientific Publications, Oxford.
13. Ezekiel, E.N. and J.F.N. Abowei, 2013. Length-weight relationship and condition factor of *Heterotis niloticus* from Amassoma flood plain, Niger Delta, Nigeria. Applied Sci. Rep., 4: 164-172.
14. Magalhaes Junior, F.O., M.J.M. Santos, I.B. Allaman, I.J. Soares Junior, R.F. Silva and L.G.T. Braga, 2017. Digestible protein requirement of pirarucu juveniles (*Arapaima gigas*) reared in outdoor aquaculture. J. Agric. Sci., 9: 114-122.
15. Lee, H.Y.M., K.C. Cho, J.E. Lee and S.G. Yang, 2001. Dietary protein requirement of juvenile giant croaker, *Nibea japonica* Temminck & Schlegel. Aquacult. Res., 32: 112-118.
16. Giri, S.S., S.K. Sahoo, A.K. Sahu and P.K. Meher, 2003. Effect of dietary protein level on growth, survival, feed utilisation and body composition of hybrid Clarias catfish (*Clarias batrachus* x *Clarias gariepinus*). Anim. Feed Sci. Technol., 104: 169-178.
17. Yang, S.D., T.S. Lin, C.H. Liou and H.K. Peng, 2003. Influence of dietary protein levels on growth performance, carcass composition and liver lipid classes of juvenile *Spinibarbus hollandi* (Oshima). Aquacult. Res., 34: 661-666.
18. Kim, K.W., X. Wang and S.C. Bai, 2002. Optimum dietary protein level for maximum growth of juvenile olive flounder *Paralichthys olivaceus* (Temminck et Schlegel). Aquacult. Res., 33: 673-679.
19. Khan, I.A. and A. Maqbool, 2017. Effects of dietary protein levels on the growth, feed utilization and haemato-biochemical parameters of freshwater fish, *Cyprinus carpio* var. *specularis*. Fish. Aquacult. J., Vol. 8. 10.4172/2150-3508.1000187
20. Kim, S.S. and K.J. Lee, 2009. Dietary protein requirement of juvenile tiger puffer (*Takifugu rubripes*). Aquaculture, 287: 219-222.
21. Choi, J., Z. Aminikhoei, Y.O. Kim and S.M. Lee, 2015. Effects of dietary protein and lipid levels on growth and body composition of juvenile fancy carp, *Cyprinus carpio* var. Koi. World Acad. Sci. Eng. Technol. Int. J. Bioeng. Life Sci., 9: 31-37.