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Optimum Phosphorus Requirement of Heterobranchus Bidorsalis Using Purified Diets

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Abstract The dietary phosphorus requirement of Heterobranchus bidorsalis fingerlings was investigated by feeding six purified diets containing varying levels of phosphorus inclusion within a range of 0.4 to 1.8% and control using casein and gelatin as dietary protein sources and dextrin as energy source over a 56 day feeding period. The experiment was conducted in a static rearing system consisting of 12 plastic aquaria of 70L capacity. Each aquarium was stocked with 10 fingerlings of Heterobranchus bidorsalis with initial mean weight of 3.33+ 0.02 g/fish. Each treatment had two replicates and fish were fed twice daily to satiation. The results of the study showed that the Mean Weight Gain (MWG) and Specific Growth Rate (SGR) were significantly highest (4.87±0.14; 1.71 ± 0.02) respectively at 0.7%P inclusion level. The Feed Conversion Ratio (FCR) and Protein Efficiency Ratio (PER) among treatments ranged between 1.10±0.05 - 2.80±0.01 and 1.20±0.0 – 2.70+0.14 respectively. Whole body calcium and phosphorus were significantly ($p< 0.05$) highest (6.54+0.01g; 2.75 +0.01g) respectively in fish fed diet with 1.8%P. That showed that body mineralization continue beyond the requirement for optimal growth. Based on observed growth performance, it could be stated that the dietary phosphorus requirement of Heterobranchus bidorsalis fingerlings was 0.7%P.

Keywords Growth Performance, Body Mineralization, Catfish, Fingerlings

1. Introduction

It has been reported clearly that there is dietary need for 22 minerals in one or more animals' species (McDowell, 2003). Fish require a dietary source of phosphorus to meet their relatively high metabolic requirements because level of dissolved phosphorus in natural waters is relatively low. The dietary requirement for phosphorus ranges between 0.45 -1% (Bury *et al*, 2003). Approximately 85% to 90% of the phosphorus in fish is in bone and scales. Unlike calcium which can be gotten from the surroundings fresh water, phosphorus has to come from food, because freshwater is deficient in Phosphate (Goda, 2007).

(De Silva 1999) stated that inability of aquatic animal to absorb inorganic elements from their external surrounding is one of the major problems among many factors in deciding the mineral requirements quantitatively. There is a general problem encountered in mineral nutrition research such as formulating minerals-free-diet and overcoming tissues stores of minerals. Feed management determines the viability of aquaculture as its accounts for at least 60percent of the cost of fish production.

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Many features influence phosphorus requirement of fish (Lall, 2003). According to Adriano and Shin, (2004), almost 80% of phosphorus in the body of fishes are in the bones and other skeletal structures. It is expected that *Heterobranchus bidorsalis* would be in high need of phosphorus for metabolic activities because of its fast growth and weight at maturity.

In addition, due to the low concentration of phosphorus in natural water, the absorption of significant amount of phosphorus from fresh water is unlikely making a dietary source of phosphorus potentially more critical. Developing nutrition strategies such as the use of inorganic phosphorus to complement natural source will make nutrients more available to the fish.

In determining the inorganic minerals source to fish, phosphorus availability is important. In order to balance the available nutrient in the diet, addition of mineral(s) to the feed is required. Therefore, the objectives of this study were to establish optimum phosphorus requirement for *Heterobranchus bidorsalis* using purified diets and the effect on mineral composition of the fish carcass.

2. Materials and Method

2.1. Experimental Diet Preparation

Six experimental diets were formulated from the combination of Casein, Gelatin, Dextrin, Cod Liver oil, Vitamin premix, starch and $CaHPO_4$ to contain diverse percentage of phosphorus (0, 0.4, 0.7, 1.0, 1.4, 1.8) by replacing starch with $CaHPO_4$. The supplementation levels were confirmed by analysis. Crude protein in all diets was designed to be 40% which have been shown to support the optimal growth of *Heterobranchus bidorsalis* fingerlings.

Ingredients were mixed in a Hobart mixer according to (AOAC, 2000) methods and then palletized to 2.5mm diameter and sun dried until moisture was ≤11.30%. Dry pellets were placed in sealed plastic bags and stored at -20℃ prior to use. Total phosphorus was determined with the molybdovanadate method (AOAC, 1995).

2.2. Experimental Diets

The feed ingredients used for the formulation of each diet were purchased from a reliable sales outlet in Lagos, Nigeria.

The prepared diets used for the experiment were divided into six treatments. The formulation of prepared diets was done in such a way that it gave 40% crude protein.

Treatment 1 (T_1), Control diet, 0% P inclusion level
Treatment 2 (T_2), 0.4%P inclusion level
Treatment 3 (T_3), 0.7%P inclusion level
Treatment 4 (T_4), 1 %P inclusion level
Treatment 5 (T_5), 1.4% P inclusion level
Treatment 6 (T_6), 1.8% P inclusion level

2.3. Experimental Fish

The experimental fish for this study *Heterobranchus bidorsalis*, with average initial mean weight of (3.33.00 ± 0.10) g/fish were obtained from Success Fish Breeding Farms, Akure, Ondo State, Nigeria.

2.4. Experimental Procedure

The experiment was carried out in a plastic tank 70cmx45cmx30cm each of which was of a static rearing system. It was an artificial confinement where natural food was absent. Fish were acclimatized for a week, and later replicated twice for each treatment. Fish were fed twice daily for the period of the experiment. The exchange of water was by siphoning.

2.5. Area of Analysis

The fish samples (whole body) before and after the experiment and the six diets were analyzed for their gross and proximate compositions as described by (AOAC, 2000) methods.

2.6. Mineral Analysis

The six experimental diets, fish carcass were analyzed for phosphorus content according to AOAC (1995) methods.

2.7. Growth Parameters

The following growth parameters were estimated.

Mean weight gain (MWG) g = $W_2 - W_1$

Where W_1 = is the initial mean weight of fish at the beginning of the experiment (T_1)

W_2 = Final mean weight of fish at day 56 (T_2)

Specific growth rate (SGR) (%day) = $(Log^{eWt} - Log^{eWi})/ T \times 100$

Where: Wt is weight of the fish at time t

W_i is weight of the fish at time 0

T is the culture period in days

Feed conversion Ration (FCR): Total dry feed fed (g)

Total weight gain (g)

2.8. Statistical Analysis

Data (mean weight gain, SGR, FCR) resulting from the experiment were subjected to analysis of variance (ANOVA) using completely randomized design. Individual (sample means) difference ($P < 0.05$) among treatment means were separated using Duncan's multiple range test.

3.Results

3.1. The Gross and Proximate Composition of the Experimental Diet

The gross and proximate composition of the experimental diets is presented in table 1 below. The values for crude protein, ether extracts, ash, crude fibre, nitrogen free extract and different phosphorus inclusion level were also reported

Table 1. Gross and proximate composition of Experimental Diets

Ingredients	T_1	T_2	T_3	T_4	T_5	T_6
Casein %	30	30	30	30	30	30
Gelatin%	10	10	10	10	10	10
Dextrin%	35	35	35	35	35	35
Cold liver oil %	10	10	10	10	10	10
Vitamin remix %	5	5	5	5	5	5
Starch %	10	8	6	4	2	0
Dicalcium phosphate ($CaHP0_4$) %	0	2	4	6	3	10
% total	100	100	100	100	100	100
Proximate Analysis						
% Moisture content	11.26	11.25	11.23	11.23	11.28	11.27
% Crude Protein	40.13	40.11	40.12	40.10	40.12	40.11
% Ether Extract	5.91	5.93	5.90	5.91	5.91	5.90
% Ash	4.13	4.18	4.21	4.26	4.30	4.32
%CHO	38.57	38.53	38.54	38.50	38.39	38.40
Total Phosphorus (%)	0	0.4	0.7	1.0	1.4	1.8

$T_1 - T_6$ = Treatments

Table 2. The proximate composition of whole body of *Heterobranchus bidorsalis* fingerlings fed diets containing different phosphorus content

Composition (%)	T_1	T_2	T_3	T_4	T_5	T_6
Moisture	10.45±0.04	10.35±0.05	10.77±0.05	10.87±0.02	9.02±0.03	10.10±0.04
Protein	75.19±0.02	75.91±0.01	79.81±0.01	76.33±0.02	75.32±0.01	75.25±0.03
Lipid	4.58±0.10	5.30±0.05	4.71±0.04	4.14±0.03	2.68±0.02	2.60±0.04
Ash	10.96±0.06	12.40±0.02	12.46±0.01	13.36±0.01	15.41±0.02	15.56±0.05

Table 3. Growth performance of *Heterobranchus bidorsalis* fed with experiment diets for 56 days

	Diets					
Parameters	(T_1) 0%P	(T_2) 0.4%P	(T_3) 0.7%P	(T_4) 1%P	(T_5) 1.4%P	(T_6) 1.8%P
Initial weight (g)	3.33± 0.02	3.33 ±0.01	3.33 ±0.03	3.33 ±0.01	3.33 ±0.02	3.33 ±0.02
Final weight (g)	6.90^{a}±0.14	7.40^{b}±0.12	8.20^{c}±0.20	7.50^{b}±0.14	7.10^{ab}±0.11	6.70^{a}±0.10
Mean weight gain (g)	3.57^{b}±0.03	4.07^{c}±0.20	4.87^{d}±0.14	4.17^{c}±0.11	3.77^{b}±0.02	3.37^{a}±0.01
SGR	1.41^{bc}±0.01	1.50^{c}±0.09	1.71^{d}±0.02	1.49^{c}±0.01	1.33^{b}±0.01	1.11^{a}±0.01
FCR	1.79^{C}±0.02	1.41^{b}±0.02	1.10^{a}±0.05	1.39^{b}±0.02	2.80^{d}±0.01	2.50^{e}±0.03
PER	1.83^{a}±0.01	1.80^{c}±0.02	2.70^{d}±0.14	1.80^{c}±0.12	1.20^{d}±0.01	1.40^{a}±0.02

Means of triplicate values with similar superscripts are not significantly different (P>0.05)

Table 4. Mineral composition of *Heterobranchus bidorsalis* fingerlings fed experimental diets with different phosphorus concentration for 56 days

(Whole body)	Initial	0%P (T_1)	0.4%P (T_2)	0.7%P (T_3)	1%P (T_4)	1.4%P (T_5)	1.8%P (T_6)
Ca (mg/g)	4.80±0.01	4.89^{c}±0.01	5.23^{c}±0.01	5.63^{b}±0.01	5.96^{b}±0.04	6.33^{a}±0.02	6.54^{a}±0.01
P (mg/g)	1.77±0.02	1.83^{d}±0.01	2.01^{c}±0.03	2.20^{c}±0.01	2.39^{b}±0.01	2.60^{a}±0.01	2.75^{a}±0.01
Mg (mg/g)	1.64±0.01	1.69^{c}±0.02	1.71^{c}±0.02	3.12^{a}±0.01	3.14^{b}±0.01	2.75^{b}±0.03	2.75^{b}±0.01
Zn (mg/g)	0.07±0.01	0.07^{c}±0.01	0.11^{c}±0.01	0.11^{c}±0.02	0.12^{b}±0.02	0.14^{a}±0.01	0.15^{a}±0.04

Means of triplicate values with similar superscripts are not significantly different (P>0.05)

3.2. Growth Performance of the Heterobranchus Bidorsalis Fed with Experimental Diets for 56 days

The growth parameters of the fish fed with experimental diets at different inclusion level of dietary phosphorus is presented in table 3 below. Fish fed diet 3 (T_3) had the highest ($p< 0.05$) mean weight gain (4.87± 0.14g), followed by fish in treatment T_4 (4.17^{c}±0.11g) and T_2 (4.07^{c}±0.20g) respectively. The least growth performance was recorded in fish fed diet 6 (T_6) with 1.8% phosphorus inclusion level.

4. Discussion

The present experiment showed that phosphorus supplement significantly improved growth and feed efficiency of *Heterobranchus bidorsalis* fingerlings. The result further indicated that suboptimal levels of dietary phosphorus can negatively affect the growth performance of the fish. The result indicated that phosphorus requirement of *Heterobranchus bidorsalis* fingerlings is 0.7%. The relative availability of phosphorus varies greatly with fish species, diet composition and form of phosphorus (Schwarz, 1995). Dietary phosphorus requirements ranging from 0.5 to 0.8 percent have been reported for rainbow trout (Ogino and Takeda, 1978).

Wilson *et al* (1982) reported values of 0.42-0.50% phosphorus as the requirement for channel catfish (*Ictalurus punctatus*), while Andrews *et al* (1973) reported 1.5%. The differences between these values reported for channel catfish and that from the present study could be attributed to species differences.

Reported phosphorus requirements vary from about 0.25-1.0% (Chavez-Sanchez *et al*, 2000) and this range explains that phosphorus requirement is species specific. The present study also showed fish fed diets without phosphorus supplement had significantly lower Zinc and Magnesium levels in whole body. This indicates dietary phosphorus supplement was necessary for magnesium and zinc deposition. It has also been reported that rainbow trout fed low phosphorus diets had significantly lower magnesium content of whole body and skin (Sugiura *et al*, 2007).

5. Conclusions

Phosphorus is a growth promoter when supplied at optimal concentration in animal feeds. The result of the feeding trial revealed that the dietary phosphorus level required for maximum growth of *Heterobranchus bidorsalis* was 0.7%. The study validated some of the earlier work done on warm water fresh fishes on the use of external sources of phosphorus in fish diets. Under practical situation, it is suggested that inorganic phosphorus should be carefully supplemented to replace the quality lost to pelleting and other forms of feed formulation. This will prevent malformation often experienced in artificially raise cat fishes.

It is recommended that better knowledge of bioavailability and appropriate indices of toxicity of this mineral in aquatic nutrition will increase its uses as well as addressing major concerns for the negative impact of minerals on aquatic environment.

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