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Influence of Dietary Protein on the Growth Performance and Chemical Composition of the Snake Head *Par-Ophiocephalus obscurus* Fed with Formulated Feeds

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Abstract
Studies were carried out to identify suitable protein sources and the level of their concentrations in formulated feeds for *Par-ophiocephalus obscurus* fingerlings reared in artificial culture systems for 120 days. Determinations were made with standard procedures recommended by AOAC for assessing the proximate composition of the animals at the beginning and end of the investigation. The feeds compounded for the animals contained each 28%, 28% and 28% protein derived from blood meal (F_1), fishmeal (F_2) and palm kernel cake (F_3) respectively. The proximate composition of *P.obscurus* at commencement of the study was as follows: crude protein= 19.0 ± 0.06 %, carbohydrate = 2.0 ± 0.23, fat = 4.02 ± 0.02%, ash = 5.0 ± 0.00%, moisture = 70.0 ± 8.0%, NFE = 3.50 ± 0.1% with the caloric value of 249 Kj/kg. This result at the end of the study showed that the crude protein content of the fish was 18.00 ± 0.17 %, carbohydrate= 2.00± 0.0 %, moisture was 72.2 ± 2.00%, fat = 3.00%, while ash and caloric value of the fish were 5.00 ± 0.00 % and 253 Kj/kg respectively. There were no significant differences in the food value of the other food components in these fishes. The growth rates of $0.008 gm.d^{-1}$; $0.005 gm.d^{-1}$ and $0.004 gm.d^{-1}$ were calculated for specimens that fed on F_1, F_2 and F_3 respectively, implying that F_1 was the best out the three compounded feeds. Survival rates were high giving 75%, 87% and 62% respectively were significantly different from each other ($P<0.05$). These results are discussed considering the response of *P.obscurus* to formulated feeds in artificial culture systems.

1. Introduction

The ultimate goal of feeding in fish farming is to obtain a fast turnover of fish biomass within a short time by feeding the cultured animals with balanced food (Parker and Schroeder, 1979). The growth rate of a fish under culture, therefore, reflects indirectly the quantity and quality of food available to the fish either as natural or supplemental (Schroeder, 1974, Page and Andrew. 1977).The response of fish to a particular food item and the unit cost of the food are the two most crucial factors that are considered in assessing the economy of a fish farming operation. Farmed animals must be provided with feeds that are capable of providing essential nutrients in quantities sufficient enough to sustain normal growth and reproduction.

Nutritionally, the best feed ingredients for any fish species are those whose biochemical compositions approximates that of the animal body composition (Coates et al 1977,ADCP,1987,Coates and Phillips,1993,Udo,2008).The preparation of complete diets in accordance with known nutritional requirements is very essential to the success of any intensive animal husbandry(Lovell,1977). Many environmental parameters(e.g. water quality) and certain technical and economic demands of the total husbandry system also have powerful influence on the choice of the feed, feeding rate and schedule of feeding (Halver, 1972).Feed and feeding therefore represent about half of the operating cost in a wide variety of aqua-farming situations (Berlin, 1979,Arazu and Udo.2012)

Preliminary studies on the growth of *P.obscurus* under mono-and polyculture situations for 180 days demonstrated the potential for the cultivability of the species (Udo, 2005). The result of this study is intended to provide data on suitable formulated feeds that could encourage rapid and economical growth of the species in culture ponds. It is expected that the identification of a suitable artificial feed for the rearing of *P. obscurus* will encourage the farmers to adopt the species for large scale culture. *P. obscurus* has been associated with suitablecultural qualities similar to those found in other cultivable species (Udo, 2005).

2. Materials and Methods

Fingerlings of *Par-ophiocephalus obscurus* used in this experiment were obtained from fishermen at Itu swamp in Nigeria. Itu swamp is a natural nursery ground for the species and it is located along the Cross River channel in the rain forest belt of Nigeria. This swamp, located in the middle course of this channel between the Cross River estuary and the upper course of the Cross River, receives its flood water from the upper Cross River during the wet seasons. Similar to the characteristics of all middle courses of rivers, this section is very rich in fish food and its nutrients. The caught juveniles were transported in oxygenated bags to the Institute of Oceanography fisheries laboratory/hatchery in the University of Calabar, Calabar, Nigeria,75 km away from Itu. In the laboratory, the fishes were acclimatized in a tank for 48 hours after which they were stocked in six (6) 32 m^2 earthen ponds at a stocking rate of 20 ind/m^2. The initial mean weight of the fingerlings and water quality parameters (pH, dissolved oxygen and temperature) of the culture systems were measured.

3. Feed Composition and Feeding

The stocked specimens were fed each with the already prepared compounded feeds labeled as F_1,F_2 and F_3 with protein sources originating from blood meal, fish meal and palm kernel cake respectively (Table I). The protein quality of the different raw food materials were read from the guide as given by Onyenuga (1968).The proximate composition of both the feeds and the fish were determined with standard methods: crude protein by Micro-Kjeldahl, fat by Soxhlet extraction, ash by furnace ashing Nitrogen Free Extracts(NFE) (that is sugars and amino acids which are not bonded to protein structures and ammonia related sugars) and moisture (by difference)(AOAC, 2000).The dietary protein levels of each set of compounded food were 28%(F_1), 28%(F_2) and 28%(F_3) respectively(Table I). The formulated dry feeds were fed ad libitum to the fish twice daily at a feeding rate of 20% body weight shared into two rations. At the end of the 120 days of study, the final weights of the animals were measured and data computed for the growth rates (GR), Food Conversion Ratios (FCR), Production Efficiency (PE) and Specific Growths Rates (SGR) as follows:

The growth rates were calculated from the following formula (Bowser and Roosemark, 1981):

$$\text{GR (gm/dy.)} = W1 - W0 \Big/ t$$

GR was the average daily weight gain (gm/dy.); 'W_1'the final weight at end of study and 'W_o' the initial weight of specimen at the commencement of study while 't'is the length of the experimental study.

The specific growth rates (SGR %) of the animals were calculated using :

$$\text{SGR (\%)} = InW1 - InW0x100 \Big/ t$$

$\ln W_t$= log.of final weight
$\ln W_o$ = log.of initial weight
t= Duration of study.

The formula used by Sandifer and Smith, (1979) in the calculation of Production Efficiency (PE) was adopted in this study as follows:

$$\text{Production efficiency (PE) =Weight of Fish stocked (gm.)} \Big/ \text{Weight of fish produced (gm.).}$$

The efficiency of the feeds (Food conversion ratio) was determined using the following formula:

$$\text{FCR= Weight of food fed (gm.)} \Big/ weight`of`animal`produced(gm.)$$

One-way analysis of variance (ANOVA) was applied in testing the significant differences between the various sets of data from each feeding trial(Sokal and Rohlf, 1968).

4. Determination of the Proximate Composition of Fish and Feeds

The dried fillets of the stocked fish at the commencement and at the end of thestudy were powdered and stored in an airtight container as stock sample for analysis. In a similar manner another stock sample was also prepared from the three types of the tested feed for analysis. Methods extant inAOAC (2000) were used for the determinations of the proximate composition and mineral contents of the homogenized fillets and the feeds. Crude protein was determined using the Micro-Kjeldahl method, fat by Soxhletextraction, ash by furnace ashing at $600^{O}C$ for 12 hours while NFE was by difference (AOAC, 2000).The differences in proximate composition of the fish at the beginning and end of the study were compared with t-statistic while ANOVA (One way analysis of variance)was applied to test for significant differences in the feeds (Sokal and Rohlf, 1969).

5. Results

Growth rates of fishes changed with the quality of the sources of protein in the feeds such that feeds containing protein of animal origin produced bettergrowths in *P. obscurus* fingerlings (Table III).The growth rates of the fishes that fed on F_1were significantly different from that calculated for animals that were fed on F_2 and F_3 ($P > 0.05$)respectively. But these growth rates were not different from the one calculated in specimens that fed on feed $F_{2.}$

Table I. *Proximate composition of the compounded feeds and P. obscurus at the beginning and end of study*

Feeds	Protein (%)	Carbohydrate (%)	Moisture (%)	Fat (%)	Ash (%)	NFE	Energy (Kj/Kg)
F1	28.0±2.11	19.78 ±0.33	40.00	7.02	4.2 ±0.14	-	232 ±28.10
F2	28.0±2.00	18.80 ±1.22	40.00	7.22	5.9 ± 0.62	-	238 ±21.09
F3	28.0 ±1.80	19.00 ±0.13	40.00	9.00	4.0 ±0.12	-	242 ±11.16
Fa	19.0 ±2.02.	2.0± 0.23	70.0 ±8.00	4.00	5.0 ± 0.00	50±0.1	249 ±0.80
Fb	18.0 ±0.17	2.0± 0.00	72.2 ± 2.00	3.00	5.0 ± 0.00	50.0±0.1	252± 0.60

F_a and F_b represents data on *P.obscurus* at the beginning and at the end of study reared for 120 days.

Statistics show that ash and energy levels in the feeds were not significantly different from that determined in the fish while fat differed significantly from those in the fish ($P>0.05$) at the beginning and end of study (Table I). However, the compounded food with fishmeal produced the highest growth from species than the one compounded with PKC(Palm kernel cake) (Table II). But the survival rates in the study from the groups were high, giving 75%,87% and 62% respectively for fishmeal, blood meal and palm kernel cake(Table II). The food conversion ratio (FCR's) for F_1 was better than those obtained for specimens that used the other feeds (Table II); the highest weight gain was measured from specimens that fed on F_1 compared to others.

Water quality average throughout the experiment giving dissolved oxygen as $6.8 mg.l^{-1}$, pH = 7.5and temperature at $28.0 \pm 3.0\ ^{O}C$.

Analysis of variance (ANOVA) showed that the energy / calories available in the feeds were similar to each other and to that determined in the fish ($P< 0.05$) (Table I).

Table II. *Growth records of P. obscurus fed with compounded feeds derived from three major protein sources.*

Growth parameters	F_1	F_2	F_3
Initial weight of fish (gm.)	1.06	1.07	1.08
Final weight of fish (gm.)	1.48	1.26	1.23
Growth rate($gm.d^{-1}$)	0.008	0.005	0.004
No. of animals stocked	200	200	200
No. of animals harvested	150	175	124
Area of culture tank (m^2)	36	36	36
PE	0.83	0.84	0.88
SGR (%)	0.47	0.41	0.33
FCR	0.45	0.61	0.68
Survival Rates (%)	75.00	87.50	62.00

6. Discussion

Literature reportson channel catfish reared under similar environmental conditions as those of this study but with a fish based meal showed that it is possible for *P.obscurus* to gain higher weightswithin a short growth period of 50 days (up to 1.01 gm) (Wee, 1982, Tan and Stif, 1993). Allison *et al* (1976) also reported that groundnut meal was more conducive for the production and growth of young *Channa striata* (Bloch) at high density. The result of the present study shows the growth rates obtained for *P.obscurus* fed with F_1compared favorably with the results reported by Allison *et al* (1976) for *C. striata*. The result of this study and that of

Allison *et al* (1976) does not agree with that reported for *Channa striata* (Bloch) by Tan and Stif (1993). The later demonstrated that growth and survival rates of *C.striata* reared in rice farm were extensively better. In addition, Parameswaram and Murugersan (1976) asserted that Ophiocephalidae sp produced artificially and reared extensively on natural food grew better than those fed with formulated feeds. The growth rates determined in this study were lower than those reported for Hetero-Clarias (a cross breed between *Heterobranchus* and *Clarias* species) that fed on natural food containing 50 – 60% protein (Udo, 2008). Those measured from *Heterobranchus longifilis* which accepted formulated feed and grew from 1.80 gm. to 2.60 gm. in 62 days (0.012gm/dy.) (Huisman, 1976) also performed slightly better than the one fed with fishmeal in this study .Also, Udo (2005) produced better growth rates from an earlier study on *Parachanna obscurus* reared with IOC Feed - I under mono- and polyculture situations in earthen ponds. However, statistics indicate that F_1 was the best food for *P. obscurus* in this study having produced the highest growth rate in the species (Table II).

The growth rate determined from animals reared with meal $F_2$was 0.0047 gm.d^{-1}(Table II). The protein content of F_2 was also 28% made from fishmeal. The minimum dietary protein level giving optimal weight gain in fish was first studied in Chinook salmon *Onchorhynchus sp.* and was reported to be the one close to the proximate protein level of the fed fish (ADCP,1987, Garling and Wilson, 1979).Maximum growth rate appears to have been achieved in fish diet with crude protein level varying between 220–440 gm/kg (Garling and Wilson, 1979). Sin (1973) used fishmeal as a test meal to define the food requirement of trout which also confirmed the results earlier given by Garling and Wilson (1979).Page and Andrew (1977) used fishmeal diet (11 to 17 gm/kg of protein) to feed Channel catfish and achieved a significant increase in growth in 148 days. We could not adducereasonsfor the low growth rates in the specimens that fed on F_2.It could be speculated that the source of the fishmeal could have been the reason for the differences. The fishmeal used in our study was made from Tilapia while our specimen is a bottom feeder possibly with a protein profile different and unsuitable for Ophiocephalidae species or the fish did not accept meal. As earlier reported, Ophiocephalidae species reared in a rice field performed better in growth than those fed with protein rich compounded feeds.

Animals grown on F_3 with palm kernel cake base exhibited the lowest growth rate (Table III). The origin of this protein is plant with protein quality clearly different from what is in fishes or animals. This meal produced the lowest survival rates of 62.2 %(Table III).This outcome also contradicts Kaushik (1977) and ADCP (1987) who asserted that food with protein contents close to that of the fed fish produce better growth rates or alternatively, it might be the amino acid quality of the protein in palm kernel cake is low or lacking the species of amino acids that assist in growth. Animals fed on F_3 could not show encouraging growth rates in the fed fish because the protein quality of the food and fed fish are different (Garling and Wilson 1979).

Garling and Wilson (1979) reported that increases in the scope of activity due to high food conversion rates corresponded with the increase in body size of a species. High FCR values show that the feed is less efficient and vice versa. Also based on FCR of F_1, it can be stated categorically that it is the best meal for the species, Food Conversion Ratios (FCR) of species is known to be influenced (among other factors) by the age and weight of the organism under investigation, taking into consideration that young organisms need more food as a percentage of their body weight than older individuals (Huisman, 1976, Halver and Tiews, 1978, Coates and Phillips 1993).

Smith *et a*l. (1981) and Bowser and Roosemark, (1981) stated that the mortality rates of cultured aquatic organisms can also be used in assessing their performance in captivity. A high mortality rate in cultured organisms indicates poor performance and vice-versa. Mortalities were not high in the study which suggests that all the meals tested here can be used in rearing the fish. This probably also suggests that other unknown factors could have hindered fast growth of *P.obscurus*(Table II).

The production efficiency (PE) of *P. obscurus* in this study was high, giving0.82, 0.84, and 0.88 for the species under the different feeding regimes (Table III). Smith *et al* (1981) discovered that the production structure of culture fish affects their final production level.

The proximate composition of the specimens at the beginning and end of investigation were not significantly different (P>0.05), however, the energy content of fish at the beginning and end of study showed significant differences (P>0.05) indicating that the older specimens at end of study contained more calories than when specimens were younger. Muhammad *et al* (2011), comparing the energy content of cultured and wild silver carp and grass carp, reported that, notwithstanding the source of fish(wild or reared), the energy content does not change. Also, the chemical composition of species at different stages of their life history changes with age, with some components being higher at youth and at senescence (Schiroerbel,1970). In this study, the energy content of all specimens was different at the end of the investigation (Table III). The reason for this discrepancy in energy content cannot be immediately established but reasons probably linked to differences in sizes of specimens at beginning and end of study could be implicated.

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