# DIETARY PROTEIN REQUIREMENTS OF MALE AND FEMALE BROODSTOCK: AN ECONOMIC FACTOR FOR INCREASED SUSTAINABILITY OF PRIVATE CATFISH HATCHERIES.

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**ABSTRACT**

Two separate feeding experiments were carried out concurrently to determine the optimum and thus the most economical dietary protein levels for maximum growth, survival and gonadal maturation/ripeness of male and female Heterobranchus longifilis broodstock. Seven diets of varying crude protein levels (20% to 50% at 5% protein intervals) were formulated and fed to "spent" male and female brooders of Heterobranchus, longifilis (230.0g 248.3g body weight) stocked in concrete tanks at 10 fish per tank. The fish were fed at 3% of their body weights twice daily (08.00 09.00h and 17.00 18.00h) for 70 days.

No fish mortality was recorded in any treatment or replicate of the two experiments and the robustness/general well-being of the brooders expressed by the condition factor (K) was high with all the dietary protein levels. The best economy of feed was achieved with the 35% c.p. diet.

For the male broodstock, the greatest increase in body weight (219.2g) as well as the best specific growth rate (0.89%) was recorded in the diet containing 35% crude protein level. 100% ripeness was observed in diets containing 25%, 30%, 35% and 40% crude protein levels while the best economy of feed was obtained in the 35% crude protein diet.

In the female broodstock, the greatest increase in body weight (270g) and best specific growth rate (1.07%/d) were recorded in fish fed 40% crude protein diet. The highest number (100%) of ripe females was also observed in brooders fed 40% c.p. diet. Feed conversion ratio and economy of feed were best at 40% c.p.

Based on this study, 35% and 40% c.p. are recommended as the most economical dietary protein levels for the diets of male and female Heterobranchus longifilis broodstock respectively. This will help to minimise

## INTRODUCTION

The catfishes ( Heterobranchus sp, Clarias sp. and the hybrids) are highly esteemed in Nigeria. and the sub-saharan Africa as both culture and table fish (Ayeni, 1995, De Graaf and Janssen, 1996). They are hardy, fast growing, readily adaptable to culture conditions and can grow to large sizes of 20kg and above (Holden and Reed, 1972, Olaosebikan and Raji, 1998) which are very attractive to both farmers and consumers in Nigeria. However, the fingerlings are usually very scarce and one of the major problems encountered by hatchery operators (fingerling producers) is the non-availability of suitable feeds for the broodstock. According to Huet (1994), the success of any breeding programme begins with adequate broodstock management and food is considered by Bromage and Robert (1995), SEAFDEC, (2000) as one of the most important determinants of the reproductive potentials of a fish. Presently, no practical/commercial feed has yet been developed specifically for the broodstock of most of the cultured catfishes in Nigeria (Eyo, 1995). This calls for an immediate attention especially of the private hatchery operators because fish feed is generally considered as the most expensive cost item in intensive/commercial fish farming and could constitute 40% - 60% of the recurrent expenditure. This high cost of feed input could negate the economic viability of a hatchery or even scare away prospective fish farmers and hatchery operators if suitable feeds are not used profitably..

In this study, as in most nutrition studies, high priority is given to protein requirement because it is the single nutrient that is required in the largest quantity for growth and development (NRC, 1993) and also the most expensive ingredient in diet formulation. Thus, fish feed should be carefully formulated so as to ensure that the protein fraction does not exceed the optimum level required by the fish in order to minimize wastage.

The aim of this study was therefore, to determine the optimum and thus the most economic dietary protein levels required for the maximum growth, survival and gonadal maturation/ripeness of male and female Heterobranchus longifilis broodstock as a working guide to fish feed millers and hatchery operators.

## MATERIAL AND METHODS

The study was carried out at the fish hatchery complex of the National Institute for Freshwater Fisheries Research (NIFFR), New Bussa. Two different experiments were conducted concurrently one for the male and the other for the female broodstock.

### The Experimental System

Forty two outdoor concrete tanks located at the fish hatchery complex were used for the experiment. Twenty one tanks were used for the males while the remaining twenty one anks were used for the females. Each tank measured 2m x 2m x 1m and had provisions for water inlet and drainage. Filtered reservoir water (water passed through a biological filter of three compartments - stone, small gravels and beach sand) was used for the experiment. All the tanks were also covered with mosquito mesh nylon screens to prevent fish from jumping out and possible predation by birds.

### Experimental diets

Seven artificial diets of varying crude protein levels (from 20% to 50% at 5% protein intervals) were formulated for the experiments, as indicated in Table 1. All the ingredients were separately milled and mixed with warm water to form a consistent dough which was then pelleted, sun-dried, packaged in cellophane bags and stored at -20oC until when needed..

### Experimental Fish and Acclimation.

The experimental fish were "spent" male and female broodstock of Heterobranchus longifilis (mean:238.4 0.08gfor the females and 240.7 32 g). Spent brooders (which had been used for induced breeding) were used such that the gonads relatively at the same stage of development. The fish were selected from the "spent" broodstock tanks and stocked in the experimental tanks for acclimation for three days during which period they were fed to satiation with a commercial catfish fingerling feed (NIFFR catfish feed containing 40% crude protein) twice daily, 08.00h - 09.00h and 17.00h - 18.00h.

### Stocking and Feeding of Fish.

On the third day of acclimation, the fish were not fed over-night in order to empty their guts and

Table 1: Percentage Composition of the Experimental Diets.

| Feed stuff | Diets and Crude Protein Levels (%) [20] D1 | [25] D2 | [20] D3 | [35] D4 | [40] D5 | [45] D6 | [50] D7 |
|---|---|---|---|---|---|---|---|
| *Fish Meal* | 10.7 | 13.5 | 16.1 | 18.8 | 21.4 | 24.2 | 26.8 |
| **Blood Meal** | 3.6 | 4.6 | 5.4 | 6.4 | 7.3 | 8.2 | 9.1 |
| **Groundnut cake** | 2.2 | 5.4 | 8.8 | 12.1 | 15.5 | 18.7 | 22.1 |
| **Soyabean Meal** | **3.3** | **8.2** | **13.3** | **18.1** | **23.2** | **28.1** | **33.2** |
| **Yellow Maize** | 73.2 | 61.3 | 49.4 | 37.6 | 25.6 | 13.8 | 1.8 |
| ***Vit/Min. premix** | 2.0 | 2.0 | 2.0 | 2.0 | 2.0 | 2.0 | 2.0 |
| **Cod liver oil** | 1.0 | 1.0 | 1.0 | 1.0 | 1.0 | 1.0 | 1.0 |
| **Vegetable oil** | 1.0 | 1.0 | 1.0 | 1.0 | 1.0 | 1.0 | 1.0 |
| **Common salt** | 0.5 | 0.5 | 0.5 | 0.5 | 0.5 | 0.5 | 0.5 |
| **Bone meal** | 0.5 | 0.5 | 0.5 | 0.5 | 0.5` | 0.5 | 0.5 |
| **Cassava starch** | 2.0 | 2.0 | 2.0 | 2.0 | 2.0 | 2.0 | 2.0 |
| **Calculated crude protein level (%)** | 20.0 | 25.0 | 30.0 | 35.0 | 40.0 | 45.0 | 50.0 |
| **Analysed crude protein level (%)** | 20.3 | 24.8 | 30.5 | 35.6 | 39.7 | 45.2 | 49.4 |
| **Analysed energy content ($KJ.g^{-1}$)** | 17.8 | 18.0 | 17.9 | 18.2 | 18.4 | 18.6 | 18.5 |
| Cost of feed (₦/kg) | 38.0 | 41.0 | 45.0 | 50.0 | 54.0 | 60.0 | 68.0 |

* According to Fagbenro *et al.*, (1992)

prepare their appetite for the new feed. The number of fish per tank was then, adjusted to 10 and the total weights, mean weights and mean standard lengths of fish in each tank were taken and recorded. The fish were anaesthetized using MS 222 (Tricaine methanosulfonate, Sandoz Ltd, Switzerland) at a dose of 300mg/l before the measurements to reduce handling stress. The 21 tanks per experiment were randomly allocated the seven diet treatments in triplicates to form a randomised complete block design for each experiment. Feeding commenced one hour after the weighing exercise and the fish were fed daily at 3% of their body weights per day, administered in two equal rations, one in the morning (08.00 h - 09.00 h) and the other in the evening (17.00 h -18.00 h).

**Sampling for Growth and Gonadal Maturation/Ripeness.**

Individual body weight and standard length measurements of the experimental fish in each treatment were taken fortnightly and the rations fed to the fish were adjusted according to the new body weights. Gonadal maturation was assessed by "ripeness" (ready-to-spawned condition) and a female was considered "ripe" if the abdomen was distended and eggs oozed out on a gentle manual pressure (antero-posteriorly) on the abdomen. The genital papilla of a ripe male was reddish in colour. The number of ripe fish per tank was observed and recorded during each sampling period. Fish mortality was also noted and water in the experimental tanks was renewed during each sampling period. The experiments lasted 70 days cocurrently.

**Water Quality Monitoring**

Water temperature records for each tank were taken daily, (8.00 h-9.00 h) using the mercury-in-glass thermometer (-10.0°C to 110°C). Dissolved oxygen (D.O) concentration was determined using the Jenway D.O. meter (Model 3050, England) and the pH taken with Jenway pH meter (model 9070, England). Both D.O. and pH measurements were taken every morning before the feeding exercise.

**Analytical Procedure**

Samples of the experimental diets were analysed for proximate composition and energy content using the standard AOAC methods (AOAC, 1995). Growth response, feed utilization and survival data obtained from the different treatments and their replicates were subjected to analysis of variance (ANOVA) test and the treatment means were compared with each other for significant differences (P? 0.05). All statistical analyses were done using the Statistical Package for Social Scientists (SPSS 6.0). for windows on an IBM-compatible PC.

## RESULTS AND DISCUSSION.

### Survival, Growth and Feed Utilization

The survival, growth and feed conversion ratios of the male and female brooders fed the experimental diets containing varying crude protein levels for 70 days are shown in Table 2 and 3. The fish showed good appetite to all the diet treatments as revealed by the increases in body weight and standard lengths and no fish mortality was recorded in any of the treatments and their replicates.

However, for the males, the greatest increase in body weight (218.4g) was achieved with the diet containing 35% crude protein (c.p.) followed by the 40% c.p. diet (208.5g). The least increase (111.3g) was recorded with the 50% c.p. diet. A similar trend was observed for the specific growth rate (S.G.R). The least SGR (0.90%) was recorded with 35% c.p. diet followed by the 40% c.p. diet (0.89%) while the least (0.54%) was by 50% c.p. diet. The values for 35% c.p. and 40% c.p. were, however, not significantly different (P > 0.05). Feed conversion ratio was also best (3.0) with the 35% c.p. diet while the robustness and general well-being of the fish expressed as the condition factor was best (0.94) at 40% c.p. level but the value was not significantly different from 0.93 obtained with the 35% c.p. Diet.

**Table 2: Growth, survival and feed conversion ratio of spent male brooders of *H. longifilis* fed experimental diets containing varying crude protein levels for 70 days.**

| | Dietary protein level (%) | | | | | | |
|---|---|---|---|---|---|---|---|
| **Parameter** | **20** | **25** | **30** | **35** | **40** | **45** | **50** |
| Mean Initial body weight (g) | 230.4 | 251.6 | 241.5 | 250.2 | 244.2 | 232.0 | 242.5 |
| Mean Initial stand. length (cm) | 32.6 | 34.5 | 33.6 | 33.7 | 32.6 | 32.8 | 33.6 |
| Mean Final body weight (g) | 387.6 | 438.2 | 421.7 | 468.6 | 452.7 | 358.3 | 355.8 |
| Mean Final stand. length (cm) | 34.8 | 36.5 | 35.7 | 37.0 | 35.9 | 35.3 | 34.8 |
| Weight gain (g) | 153.2 | 186.6 | 180.2 | 218.4 | 208.5 | 126.3 | 111.3 |
| Increase in stand. length (cm) | 2.2 | 2.0 | 2.1 | 3.3 | 3.3 | 2.5 | 1.2 |
| *Specific growth rate (S.G.R) | 0.72 | 0.80$^{b}$ | 0.80$^{b}$ | 0.90$^{a}$ | 0.89$^{a}$ | 0.63 | 0.54 |
| **Feed conversion ratio (FCR) | 4.0 | 3.8 | 4.0 | 3.0 | 3.2 | 4.3 | 4.6 |
| ***Condition Factors (K) | 0.90 | 0.90 | 0.94$^{a}$ | 0.93$^{a}$ | 0.90 | 0.82 | 0.87 |
| Survival (%) | 100 | 100 | 100 | 100 | 100 | 100 | 100 |
| No.of ripe fish (%) | 80 | 100 | 100 | 100 | 100 | 90 | 90 |
| Economy of feed/ Economy of wt. Gain (N/g.wt.Gain) **** | 0.13 | 0.13 | 0.13 | 0.12 | 0.15 | 0.24 | 0.31 |

a Figures with similar superscrips are not significantly different (P > 0.05).

**Table 3** : Growth, survival, feed conversion ratio and gravid status of spent female brooders of *Heterobranchus longifilis* fed experimental diets containing varying crude protein levels for 70 days.

| | **Dietary protein level (%)** | | | | | | |
|---|---|---|---|---|---|---|---|
| **Parameter** | **20** | **25** | **30** | **35** | **40** | **45** | **50** |
| **Mean Initial body weight (g)** | 236.7 | 236.0 | 240.0 | 233.5 | 243.3 | 240.7 | 238.7 |
| **Mean Initial standard. length (cm)** | 33.7 | 33.5 | 33.1 | 32.5 | 33.2 | 32.1 | 32.2 |
| **Mean Final body weight (g)** | 380.0 | 379.3 | 402.5 | 460.0 | 513.3 | 426.0 | 455.3 |
| **Mean Final standard. length (cm)** | 36.3. | 36.2 | 35.3 | 36.9 | 37.9 | 34.7 | 36.2 |
| **Weight gain (g)** | 143.3 | 143.3 | 162.5 | 226.7 | 270.0 | 185.3 | 216.6 |
| **Increase in standard. lengths (cm)** | $2.6^a$ | $2.7^a$ | 2.2 | 4.4 | 4.7 | 2.6a | 4.0 |
| ***Specific growth rate (S.G.R)** | $0.67^a$ | $0.69^a$ | 0.74 | 0.97 | 1.07 | 0.81 | 0.91 |
| ****Feed conversion ratio (FCR)** | $3.5^a$ | $3.4^a$ | $3.3^a$ | 2.9 | 2.6 | $3.3^a$ | 3.1 |
| *****Condition Factors (K)** | 0.79 | 0.80 | $0.92^a$ | $0.92^a$ | $0.92^a$ | $0.92^a$ | $0.92^a$ |
| **Survival (%)** | 100 | 100 | 100 | 100 | 100 | 100 | 100 |
| **No. of ripe fish (%)** | $50^a$ | $50^a$ | $80^b$ | $80^b$ | $100^c$ | $100^c$ | $80^b$ |
| **Economy of feed/ Economy of wt. Gain ( ₦/g.Wt.Gain) ****** | 0.13 | 0.14 | 0.14 | 0.11 | 0.10 | 0.16 | 0.16 |

a Figures with similar superscripts are not significantly different ($P > 0.05$).

* **S.G.R. (%)** = $100 (L_n W_2 - L_n W_1)/T$
<u>where</u>: $W_1$ = Initial weight, $W_2$ = Final weight, T = Period of experiment (days)

** **F.C.R.** = Feed consumed/Weight gained

*** **Condition Factor (K)** = $100W/L^3$
<u>where</u>: W = Weight of fish (g), L = Length of fish (cm).

**** **Economy of Wt. Gain or Economy of feed (N/g. Wt. Gain).**
**<u>= Cost of feed fed(₦) / Body Weight Gain (g)</u>**

The above results suggest that 35% was the optimum dietary protein level for maximum growth and feed utilization in spent males of *H. longifilis* and, therefore, the most suitable dietary protein level for the fattening of spent *H. longifilis* male broodstock.

For the females, the greatest increase in body weight (270.0g) was achieved with the diet containing 40% crude protein (c.p.) followed by the 35% c.p. diet (226.7g). The least increase (143.3g) was recorded with the 20% and 25% c.p. diets . Also the best SGR (1.07%) was recorded with 40% c.p. diet followed by the 35% c.p. diet (0.97%) while the least (0.67%) was by 20% c.p. diet. The values for 20% c.p. and 25% c.p. were, however, not significantly different ($P > 0.05$). Feed conversion ratio was also best (2.6) with the 40% c.p. diet while the robustness and general well-being of the fish expressed as the condition factor was 0.93 at 30%,35%,40%, 45%,and 50% dietary protein levels.

**Ripeness or "Ready-to-Spawn" Conditions.**
Tables 2 & 3 also show the percentage of ripe fish in the various experiments. For the males 111% of the fish fed 25%,30%,35%,and 40% were ripe while in the females, it was observed that 100% of fish fed the 40% and the 45% c.p. diets were ripe while only 50% of the fish fed 20% and 25% c.p. diets were ripe at the end of the 70-day period of the experiment. Fish fed the diets containing 30%, 35% and 50% crude protein levels had 80% of their population in the ripe condition.

**Economy of Feeds/ Economy of Weight Gain.**
Tables 4a&b show the economic analysis of the feeds in terms of economy of body weight gain-ie the cost of feed used to raise the fish to gain one gram of body weight. For the males the best economy of feed was achieved with the 35% crude protein feed while the most uneconomical feed was the 45% c.p. diet. For the females, the most economical feed was the 40% c.p. diet while the worst feeds in terms of economy of weight gain

**Table 4 (a)** Economy of feeds/diets of varying crude protein levels fed to male *H. longifilis* broodstock for 70 days.

| Diets | C.P. of diet/feed (₦/kg) | Cost of diet/feed (₦/kg) | Body Wt of fish (g) | Quantity of feed fed (g) | Cost of feed fed (₦) | Body Wt Gain (g) | * Econ. of feed (₦/g Wt Gain) |
|---|---|---|---|---|---|---|---|
| 1. | 20 | 38.0 | 233.3 | 509.9 | 19.4 | 153.4 | 0.13 |
| 2. | 25 | 41.0 | 250.0 | 568.0 | 25.0 | 186.7 | 0.13 |
| 3. | 30 | 45.0 | 240.0 | 504.0 | 22.6 | 180.0 | 0.13 |
| 4. | 35 | 50.0 | 248.3 | 521.4 | 26.6 | 219.2 | 0.12 |
| 5. | 40 | 54.0 | 243.3 | 560.9 | 30.3 | 207.4 | 0.15 |
| 6. | 45 | 60.0 | 230.0 | 508.0 | 30.5 | 128.3 | 0.24 |
| 7. | 50 | 68.0 | 240.0 | 514.0 | 34.9 | 111.3 | 0.31 |

**Table 4 (b) Economy of feeds/diets of varying protein levels fed to female *H. longifilis* brood stock for 70 days.**

| Diets | C.P. of diet/feed | Cost of diet/feed (₦/kg) | Body Wt of fish (g) | Quantity of feed fed (g) | Cost of feed fed (₦) | Body Wt Gain (g) | * Econ. of feed (₦/g Wt Gain) |
|---|---|---|---|---|---|---|---|
| 1. | 20 | 38.0 | 236.7 | 502.7 | 19.1 | 143.3 | 0.13 |
| 2. | 25 | 41.0 | 236.0 | 504.7 | 20.7 | 143.3 | 0.14 |
| 3. | 30 | 45.0 | 240.0 | 520.8 | 23.4 | 162.5 | 0.14 |
| 4. | 35 | 50.0 | 233.5 | 515.3 | 25.8 | 226.7 | 0.11 |
| 5. | 40 | 54.0 | 243.3 | 525.6 | 28.4 | 270.0 | 0.10 |
| 6. | 45 | 60.0 | 240.7 | 520.3 | 31.2 | 185.3 | 0.16 |
| 7. | 50 | 68.0 | 238.7 | 519.8 | 35.4 | 216.6 | 0.16 |

*** Economy of Wt. Gain or Economy of feed (₦/g. Wt. Gain).**
**= Cost of feed fed(₦) / Body Weight Gain (g)**

were the 45% c.p. and the 50% c.p. diets. Fig. 1 shows the economy of weight gain in relation to varying crude protein levels.

The excellent survival (100%) of fish in all the treatments and replicates for the two experiments could be attributed to the fact that the experimental conditions were good. Thus, the water quality records for the various treatments fell within the limits of good quality water for aquaculture as recommended by Boyd and Lichtkoppler (1985). Dissolved oxygen was never below 4.5 mg/l and ranged between4.5-5.6 mg/l. while pH was between 7.5 and 8.5. Water temperature ranged between 27oC and 28.5oC with a mean daily temperature of 28.O + 0.82 oC. The high survival of fish recorded in this experiment may, therefore, not be wholly in response to the feed treatments but could be as a result of good culture and handling techniques. It is also known that at this size and age Heterobranchus is usually hardy.

It is clear from Tables 2 & 3 that the best body weight gain, specific growth rates and feed conversion ratios for the male and female broodstock were recorded with the diets containing 35%and 40% crude protein levels respectively. These results suggest that 35% & 40% were the optimum dietary protein levels for maximum growth and feed utilization in spent males and females of Heterobranchus longifilis and, therefore, the most suitable dietary protein levels for the fattening of male and female broodstock of Heterobranchus longifilis.

Though all the dietary protein levels caused ripeness in the fish, (Tables 2 & 3) 35% and 40% c.p. for males and females respectively, produced the greatest enhancement of ripening in all the fish fed the diets. This result corroborates the work of De Silva and Anderson(1995) who observed that the dietary protein requirement of most broodstock fish are similar to those for their optimal growth. According to Bromage and Robert (1995) final ovarian development in fish involves major physiological and biochemical changes which result in a massive incorporation of proteins and lipids into the growing oocytes. These nutrients must be drawn from the body of the fish. The authors also observed that in females, feeding level determines the total number of oocytes being highest in the better-fed fish. The use of natural food alone such as trash fish, squid meal as reported by Huet (1994) or fingerling feeds as substitutes for broodstock feeds as practiced by many hatchery operators in Nigeria (Madu, 1995) are not adequate. The diet with the highest crude protein content (50%) did not produce the best growth and gonadal maturation because according to Ufodike and Ekokotu (1986) the energy which would have been used for growth and gonadal development would have been diverted and used for the de-amination and excretion of the excess protein which would not be stored by the fish.

The economy of feed for the male broodstock was best in the 35% c.p. diet while in the females the best economy of feed was achieved with the 40% c.p. diet. The results go further to suggest 35% c.p. and 40% c.p. as the most economical crude protein level for the feeds of male and female broodstock respectively of H.longifilis.

Based on the records obtained from these experiments, 35% protein is considered as the optimum dietary level for the maximum growth, survival and gonadal maturation of male H. longifilis brooders while 40% c.p. is ideal for the female brooders.. This result should, therefore, become a basic information and working guide extendable to fish feed millers and fish hatchery operators for economic and sustainable broodstock feeds.

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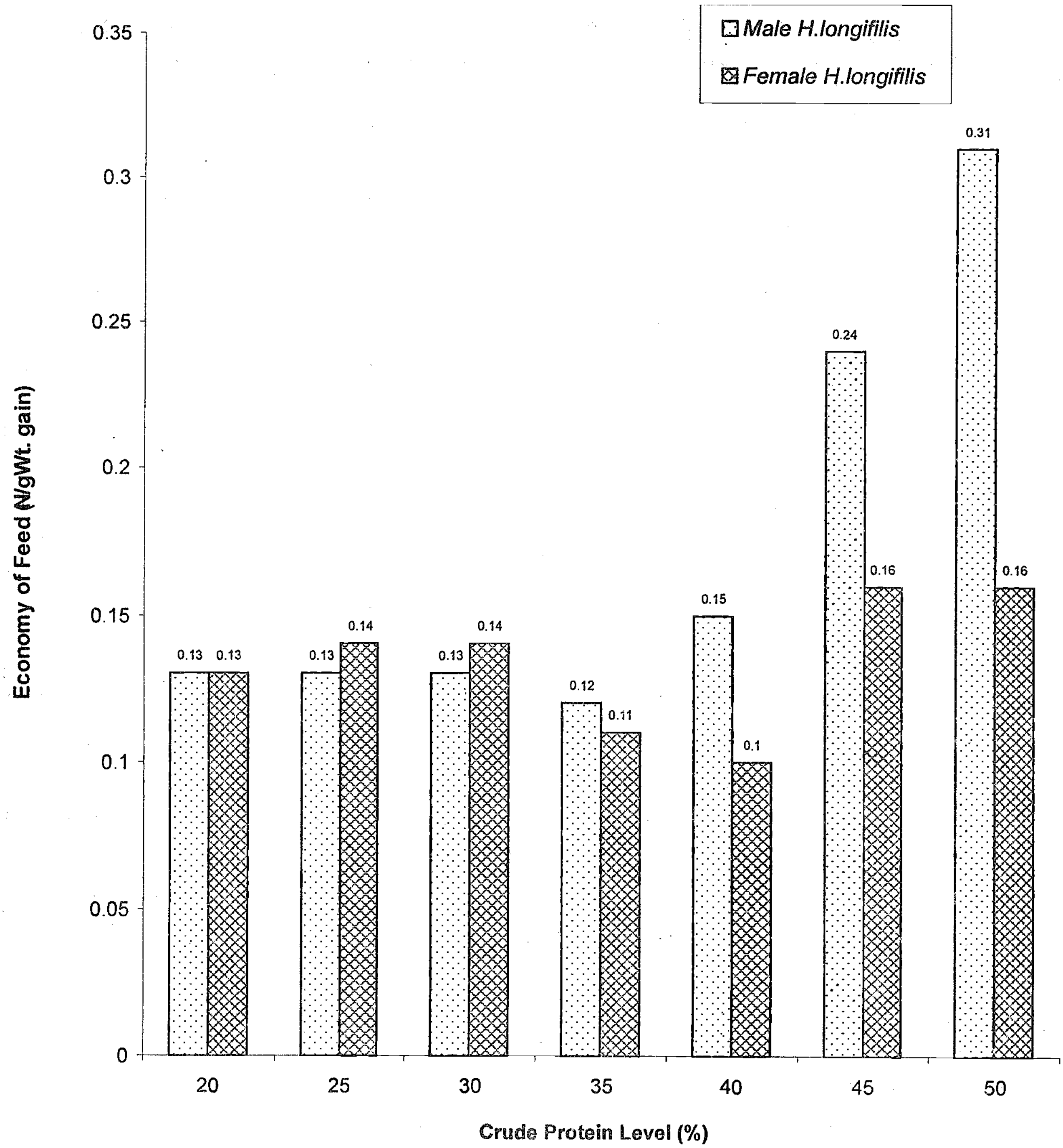


**Fig 1: Economy of feed of varying crude protein levels fed to male and female H.longifilis brood stock for 70 days.**