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# Assessment of the growth response of African catfish *Clarias gariepinus* fed varying levels of mice (*Mus musculus*) meal by substituting fishmeal

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**Abstract**
A 56-day growth research was carried out to assess the suitability of adding processed micemeal (*Mus musculus*) as the protein source in the diet of *Clarias gariepinus* fingerlings (average weight 2.05±0.11). Ten fish were placed in circular pools with a capacity of sixteen liters each and designated as treatments one to five, with one replication for each treatment. Four test diets were created with micemeal inclusion levels of 30%, 50%, 70%, and 100%, respectively, replacing equal proportion of fishmeal from the control diet with 35% crude protein. Standard procedures were used to determine water quality parameters such as temperature, pH, total alkalinity, free carbon dioxide, and dissolved oxygen. Except for dissolved oxygen, which ranged from 4.10 to 4.37 mg/l, all metrics fell within an acceptable tolerance range for fish culture. The fingerlings accepted the experimental meals. Significant differences ($p<0.05$) were seen in the test diets (D2-D5) and the control diet (D1). When compared to other treatments, fish fed the control diet showed a significant ($p<0.05$) increase in live weight gain. As micemeal content in the food rose to 100%, live weight gain decreased. According to the study, compared to other percentages, a diet containing up to 30% micemeal produced superior live weight gain. Consequently, micemeal at 30% inclusion offers a potential to be added to *Clarias gariepinus* diets as a fallback when diets prepared with fishmeal are unavailable.

**Keywords:** *Clarias gariepinus,* fish feed, diet, fingerlings, fishmeal, mice meal

## 1. Introduction

The fish feed industry has grown tremendously as a result of fish farming activity, which is the fastest-growing animal production sector in the world, with an average annual rate of about 10.3% since 2010, producing 89% of the world's total in volume terms over the last 20 years (FAO, 2020) [10]. Protein is one of the important dietary elements that affects fish growth, survival, and yield. It offers both required and non-essential amino acids for body protein synthesis and energy. The choice of protein source has a significant impact on fish growth and development, as well as production costs (Barroso *et al.,* 2014) [5]. Because of its high level of readily digestible proteins, composition of amino acids, highly digestible lipids, and long-chain polyunsaturated fatty acids, fishmeal is one of the most significant constituents in fish diets (Trushenski & Rombenso, 2020) [21]. Despite being a vital component of fish diets, fishmeal is the most expensive ingredient to produce in fish feed, which drives up feed prices. This has thus made it necessary to conduct numerous experiments in an effort to lessen the dependency of fish feed on fishmeal (Shekarabi *et al.,* 2022) [19]. The effectiveness of several alternative protein sources as a partial or total replacement for fish meal has been investigated individually in fish diets. Fishmeal has been replaced by other protein sources, such as animal byproduct meal (Kenge & Ofojekwu, 2022) [12] Composite Meal (Ojewole *et al.,* 2022) [16] mealworm, grasshopper meal (Michael, & Kolapo, 2017) [14], among others. African catfish are an important freshwater fish produced in many countries due to their high resilience to disease and high production abilities.

African catfish, *Clarias gariepinus*, is a popular and accepted freshwater fish species with great qualities such as short-term production cycle, excellent taste, rapid-growth and high tolerance to stocking density (Wei *et al.*, 2024) [22]. There is a need to enhance the production of African catfish at a reduced cost of production due to the increasing demand for this species and its desirable quality. The aim of this study was to evaluate the growth of African catfish *Clarias gariepinus* fingerlings fed varying levels of mice (*Mus musculus*) meal as the protein source.

## 2. Materials and Methods

### 2.1 Study Area

The study was conducted in the Zoology Department Research Laboratory at the University of Jos in Plateau State, Nigeria. The location of the laboratory is between latitudes 9.949560, or 90 56' 58" North, and longitude 8.889560, or 80 53' 22" East.

### 2.2 Collection and Acclimatization of Experimental Fish

African catfish (*Clarias gariepinus*) fingerlings of mixed sex, measuring 2.05g±0.11g were obtained from Artee Aquatic Konsult, Dogon Dutse, Jos and Plateau State, Nigeria. The fingerlings spent seven days in the laboratory to get used to the laboratory environment. The fish were fed 3% of their body weight with 3 mm commercial feed (Coopens®) every day during the acclimation period. Before each daily feeding session, leftover food and excrement were siphoned out to prevent ammonia accumulation in the water, hazardous waste buildup, and the decrease of dissolved oxygen.

### 2.3 Collection of Ingredients for the Experimental Diet

Precisely fifty (50) mice were captured using local traps in and around Kopjel Jing hamlet under the Pankshin Local Government Council. The mice were knocked unconscious with a forceful punch to the head. They were then gutted, skinned, and placed in the sun under a screen to dry out and prevent insects like flies from laying their eggs on them. The powdered dried mice were ground using a blender and sieved through a 90µm mesh sieve before being weighed with an electric sensitive balance. Additional feed ingredients, such as fishmeal and vitamin premix (Vitalyte) were obtained at Abdulaziz Agrovet Enterprise situated on Bauchi Road in Jos Plateau State, Nigeria. Additionally, groundnut cake, groundnut oil, and maize flour were acquired for use in feed.

### 2.4 Determination of Proximate Composition of Micemeal

The AOAC (2019) procedures were utilized to ascertain the nutritional composition of micemeal (*Mus musculus*).

### 2.5 Crude Protein

In order to quicken the process, a catalyst combination consisting of K2SO4 and CuSO4 was used. One gram (1 g) was introduced to a 100 cm3 Kjeldahl digestion flask containing two grams (2 g) of the sample. Concentrated sulfuric acid (25 milliliters) was added to the flask. The Kjeldahl digestion flask's contents were heated gradually at first in the Kjeldahl heating unit until frotting stopped, and then move vigorously with irregular flask rotations to guarantee even digestion and prevent overheating of the contents. Heat was applied incrementally until a translucent solution was obtained. Following cooling, the solution was transferred into a 100 cm3 volumetric flask and the necessary amount of distilled water was added to dilute it. A Markham semi-macroscopic nitrogen container was pipetted with 10 mL of the digest or diluted solution. Distillation was continued until the pink color of the indicator became green. The conical flask's contents were titrated with 0.1M of HCl. A shift in color from green to pink signified the conclusion point. The amount of acid used for the distillate and the blank was recorded. It is equal to {(0.014 × M × (V1-V0)}/ {test sample weight} × 100} for percentage nitrogen. The molarity of the acid is denoted by M, and the volumes of HCl required for the 10 mL sample solution and the blank are represented by V0 and V1, respectively. Atomic weight of nitrogen is equal to 0.014% Nitrogen (N2) time's 6.25 x crude.

### 2.6 Crude Fat Content

A clean, dry 500 mL round-bottom flask with a few anti-bumping granules was filled with four grams of the sample (W1), which was then weighed (W2) along with 300 mL of petroleum ether (40-60°C) for the extraction. The flask was then sealed with a Soxhlet extraction device. The soxhlet extractor was linked to the round-bottom flask, a condenser, and cold-water circulation was turned on. After turning on the heating mantle and adjusting the heating rate, the solvent began to reflux steadily. An extraction took six hours to complete. After recovering the solvent, the oil was dried for an hour at 70 degrees Celsius in the oven. The oil and round-bottom flask were allowed to cool before being weighed (W3) % Crude Content is equal to {(W2-W3)/(W2-W1)} x 100.

### 2.7 Ash Content

Following ten minutes of cooling at 100 °C in a desiccator, the porcelain crucible was weighed and then dried in an oven (W1). A finely ground sample weighing two grams was added to a clean, previously weighed crucible (W2). After that, the crucible was lit at 550 $0^C$ for an hour in a muffle furnace and allowed to cool in a desiccator. The crucible and its contents were placed inside the muffle furnace, and the temperature was gradually increased until it reached 550 $0^C$. The sample was fired for eight hours in order to ensure sufficient ashing. This crucible was removed and allowed to cool in a desiccator until it attained a constant weight (W3) after the ash had been allowed to cool to 200 °C. 100% of the ash content is (W2-W3)/(W2-W1)} × 100.

### 2.8 Moisture Content

A sterilized crucible was weighed after cooling in a desiccator and drying to a constant weight at 110 °C (W1). In the crucible, 2 g of finely powdered sample was weighed twice (W2). The crucible and its contents were oven-dried to a certain weight (W3). The formula for estimating percentage moisture content is {(W2-W3)/(W2-W1)} × 100.

### 2.9 Nitrogen Free Extract

Total carbohydrate content was determined by addition of the percentage moisture, ash, crude fat, and crude protein. The total was deducted from 100. NFE = 100 (% ash +% crude fat +% crude protein +% moisture).

### 2.10 Formulation of Experimental Diet

Different ratios of fish meal to mice meal were used to produce five test meals with varying levels of crude protein. The control diet (D1) contained fishmeal as the only source of protein. Different percentages of mice meal were made for use as the protein source in test diets 30%, 50%, 70%, and 100%. Using Pearson's square method, the different ratios of feed ingredient inclusion levels were calculated. Each calculated portion was weighed precisely. After that, they were thoroughly blended using a food mixer. After cautiously adding boiling water, mixing was continued until full homogeneity was achieved. A mechanically driven pelleting machine was used to pellet the semi-moist paste diet via a 2mm dice. The pelleted meals were gathered into trays that were flat and sun-dried under screens to a consistent weight. The diets were then manually divided into crumps, carefully labeled, and placed in a polythene bag for storage. Proximate analysis was used on each sample of the dried test food to determine the percentage composition of the various diet components.

### 2.11 Experimental Design

The investigation was carried out by utilizing a totally randomized design. Following the acclimation phase, the fish were separated into five feeding regimens, denoted as D1-D5. The fish were randomly assigned to 10 plastic baths (16-liter capacity) that were filled with dechlorinated water to the eighth litter level. They were divided into five feeding treatments with replicate.

### 2.12 Stocking, Feeding and Weighing of the Experimental Fish

Ten (10) fingerlings were weighed before being placed in each of the designated plastic pools. Each plastic bath was then covered with a square wire mesh put on a wooden frame to keep the fingerlings from jumping out of the tanks while also avoiding the introduction of undesirable objects. To prevent toxicant buildup, the experimental tanks were gently washed by hand on a daily basis. For 56 days, the fish were fed twice a day, from 7:00 to 8:00 a.m. and 5:00 to 6:00 p.m. Fish were weighed once a week using an electronic balance.

### 2.13 Determination of Growth Performance and Nutrient Utilization Parameters

Data was collected and processed to assess fish growth performance. Ten fish were gathered from each tank, weighed weekly using a digital Scout Pro sensitive scale (Model: KD-200-110, USA), and an average was calculated. At the end of the feeding trial, the growth performance metrics were determined as follows:

Feed conversion ratio (FCR)=feed intake (g)/weight gain (g)
Weight gain (%)=(Final weight-initial weight)/Initial weight) x 100
Specific growth rate (%)=(Final Mean weight-Initial Mean weight)/Length of feeding trial (days)
Apparent Net Protein Utilization (ANPU)=Fish protein gain / protein consumed × 100
Protein Efficiency Ratio (PER)=Gain in weight of fish (g) / Protein intake × 100
Mean Weight Gain=Mean Final Body Weight-Mean Initial Body Weight.

### 2.14 Determination of Water Quality Parameters of Fingerlings of *Clarias gariepinus* Fed Mice Meal

Weekly measurements of temperature, free carbon dioxide, dissolved oxygen, and total alkalinity were made of the water quality parameters using a thermometer, in accordance with the procedures outlined in APHA (1998) [4]. The pH meter (EIL 7045/46) was used in the laboratory at room temperature to measure the concentration of hydrogen ions.

### 2.15 Data Analysis

R-console (version 2.9.2) was used to evaluate the data that was obtained. To determine whether there were any significant differences between the parameters, a one-way analysis of variance (ANOVA) was used for the growth, nutrient consumption, and water quality measures. The significance level was set at the ($p<0.05$) level.

## 3. Results

### 3.1 Percentage proximate composition of the experimental diet

The findings of the percentage proximate analysis of the experimental meals are presented in Table 1. The table shows that the percentage of crude protein content of the experimental diets varied. The reference diet (control) had the highest protein value (35.63%), whereas D5 had the lowest (33.48%). Diets (D1) one and two (D2) had the highest crude fat content (19.05% and 19.12%, respectively, whereas diet five (D5) had the lowest at 15.60%. The test diet's percentage ash content was highest in D5 (12.10%) and lowest in D1 (6.60%). The moisture content was highest in D5 (8.78%), and lowest in D3 (7.84%). The nitrogen-free extract of the diet varied, with the maximum value in the reference diet (30.62%) and the lowest value in diet D4 at 29.26%.

**Table 1:** Percentage proximate composition of the experimental diets (Micemeal-based diet) fed to *Clarias gariepinus* for 8 Weeks

| Composition | | | Diet | | |
|---|---|---|---|---|---|
| | D1(Ctrl) | D2 (30%) | D3 (50%) | D4 (70%) | D5 (100%) |
| Crude Protein (%) | 35.63 | 35.44 | 35.20 | 34.37 | 33.48 |
| Crude fat (%) | 19.05 | 19.12 | 18.61 | 18.56 | 15.60 |
| Ash Content (%) | 6.60 | 7.27 | 9.08 | 9.55 | 12.10 |
| Moisture (%) | 8.11 | 8.12 | 7.84 | 8.27 | 8.78 |
| NFE (%) | 30.62 | 29.96 | 29.27 | 29.26 | 29.55 |

Where NFE=Nitrogen free extract

### 3.2 Water Quality Parameters of Fingerlings of *Clarias gariepinus* Fed Micemeal

The results of the mean water quality metrics that were tracked during the eight-week trial are shown in Table 2. According to the dissolved oxygen data, D5 had the highest value at 4.37 mg/l, while D3 had the lowest value at 4.10 mg/l. The highest and lowest levels of free carbon dioxide were found in D3 (7.50 mg/l) and D4 (7.20 mg/l), respectively. The hydrogen ion concentration (pH) value varied between 7.14 and 7.4, with D2 having the highest value (7.4) and D4 having the lowest (7.14). Throughout the investigation, the mean temperature (240 °C) was constant throughout all experimental time.

**Table 2:** Mean water quality parameters monitored during the experimental period of eight (8) Weeks

| Parameter | $D_1$ (Ctrl) | $D_2$ (30%) | $D_3$ (50%) | $D_4$ (70%) | $D_5$ (100%) |
|---|---|---|---|---|---|
| Temperature (ºC) | 24±0.00 | 24± 0.00 | 24± 0.00 | 24±0.00 | 24±0.00 |
| pH (mg/1) | 7.32±0.06 | 7.40±0.06 | 7.24±0.02 | 7.14±0.10 | 7.28±0.03 |
| Dissolved oxygen (mg/1) | 4.34±0.05 | 4.24±0.05 | 4.10±0.30 | 4.20±0.02 | 4.37±0.02 |
| Free $CO_2$ (mg/1) | 7.40±0.40 | 7.45±0.05 | 7.50 ± 0.20 | 7.20 ± 0.30 | 7.25±0.05 |
| Total Alkalinity (mg/1) | 70.30±0.20 | 71.35±0.15 | 71.20±0.40 | 71.45±0.25 | 70.70±0.10 |

$CO_2$ = Carbon dioxide, pH = Hydrogen ion concentration

**Table 3:** Growth performance and nutrient utilization of African catfish fed varying levels of Micemeal

| Parameter | $D_{1(Ctrl)}$ | $D_{2\ (30\%)}$ | $D_{3\ (50\%)}$ | $D_{4\ (70\%)}$ | $D_{5\ (100\%)}$ |
|---|---|---|---|---|---|
| Mean growth(MG) | 14.68±0.11 | 14.15±0.06 | 13.68±0.12 | 13.15±0.06 | 12.36±0.18 |
| Specific growth rate(SGR) | 1.56±0.01 | 1.50±0.01 | 1.44±0.01 | 1.38±0.01 | 1.29±0.02 |
| Cumulative Weight Gain | 9.93±0.59 | 9.58±0.56 | 9.38±0.54 | 9.67±0.54 | 9.70±0.52 |
| Feed Conversion Ratio | 1.59±0.05 | 1.45±0.06 | 1.16±0.04 | 1.15±0.06 | 1.11±0.03 |
| Protein Efficiency Ratio | 1.25±0.03 | 1.24±0.02 | 1.23±0.01 | 1.14±0.01 | 1.13±0.01 |
| Mean live weight gain | 8.10±0.10 | 7.65±0.05 | 7.30±0.10 | 7.15±0.05 | 6.75±0.15 |
| ANPU | 67.27±2.29 | 53.41±1.94 | 47.37±0.49 | 39.38±0.92 | 37.91±0.21 |

ANPU = Apparent Net Protein Utilization

The control group had the highest mean growth (14.68), while D5 had the lowest mean growth (12.36), according to the mean growth rate calculated from the weekly growth performance of the *Clarias gariepinus* fed on diets containing varied amounts of mouse meal and fishmeal. Statistical analysis indicated that the mean growth rate of *Clarias gariepinus* fed with different percentages of mice meal differed significantly ($p<0.05$), (Table 3). Fish fed D5 (1.29) had the lowest specific growth rate (SGR) and fish fed D1 (1.56) had the highest (Table 3). The Cumulative Weight Gain for the control diet D1 (9.93%) is the largest, while D3 (9.38%) was the lowest There was no significant difference ($p>0.05$) in cumulative weight gain of *Clarias gariepinus* fed with experimental diets (Table 3, Figure 1). D5 (1.11) had the lowest Food Conversion Ratio (FCR) while D1 (1.59) had the highest. Between the control and treatment groups, statistical analysis revealed a significant difference ($p<0.05$). D1 exhibited the highest Protein Efficiency Ratio (1.25), whereas D5 displayed the lowest (1.13). The Protein Efficiency Ratio (PER) indicated a significant difference ($p<0.05$) between the control diet and the various treatments. The fish fed the control diet had the highest live weight gain (8.10g), while the fish fed diet D5 had the lowest live weight gain (6.75g), (Table 3, Figure 2). The weight gain of the fish decreased as the dietary level of mice meal increased in the following order: 8.10, 7.65, 7.30, 7.15, and 6.75g. The statistical analysis showed a significant difference ($p<0.05$) between the test diets and the control diets. The highest apparent protein utilization was recorded in the control diet D1 (67.27) while the lowest is recorded in diet D5 (37.91). Apparent protein utilization declined as the dietary level of mice meal increases. There was a higher significant difference ($p<0.05$) across apparent protein utilization of *Clarias gariepinus* fed with varying percentages of mice meal and the control.

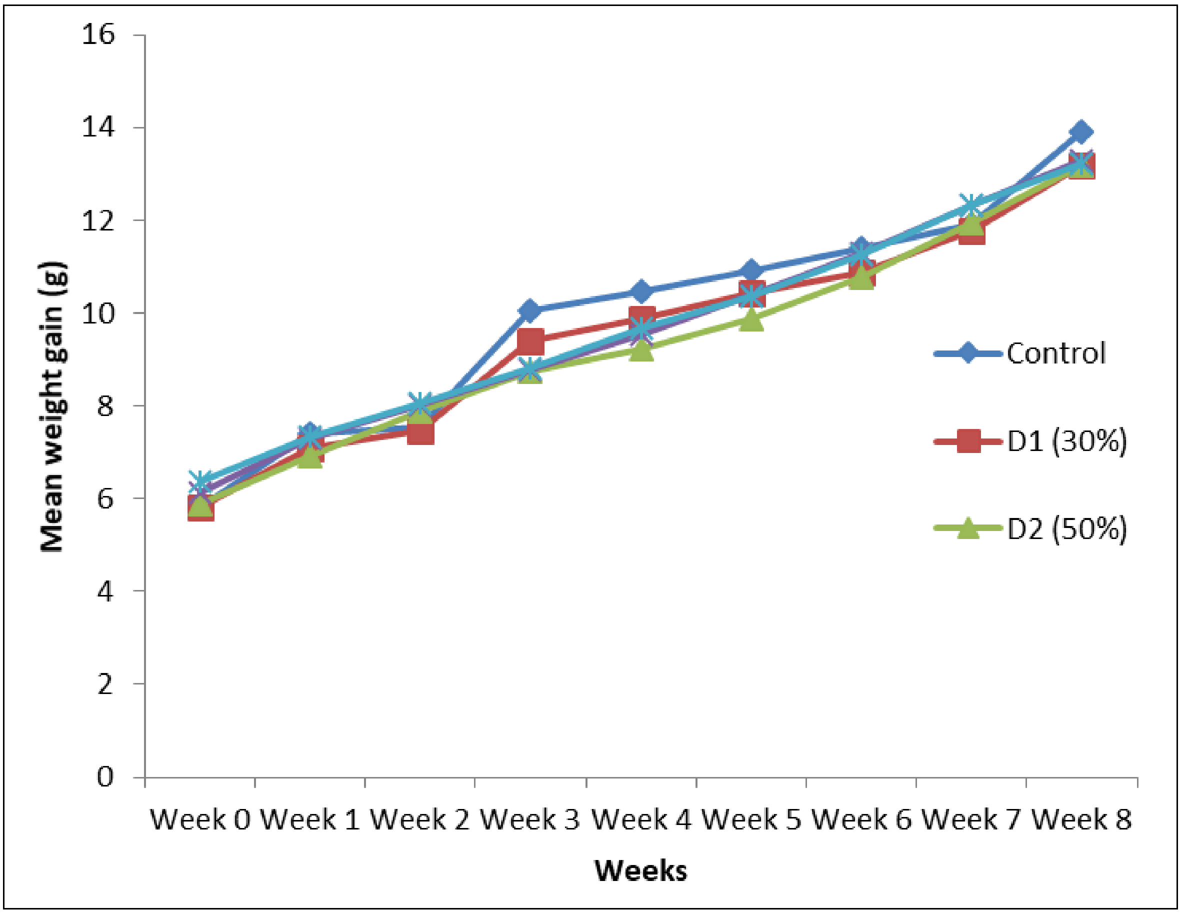


**Fig 1:** Mean of cumulative weight gain of *C. gariepinus* fingerlings fed with varying percentages of mice meal and reference diet for 8 weeks

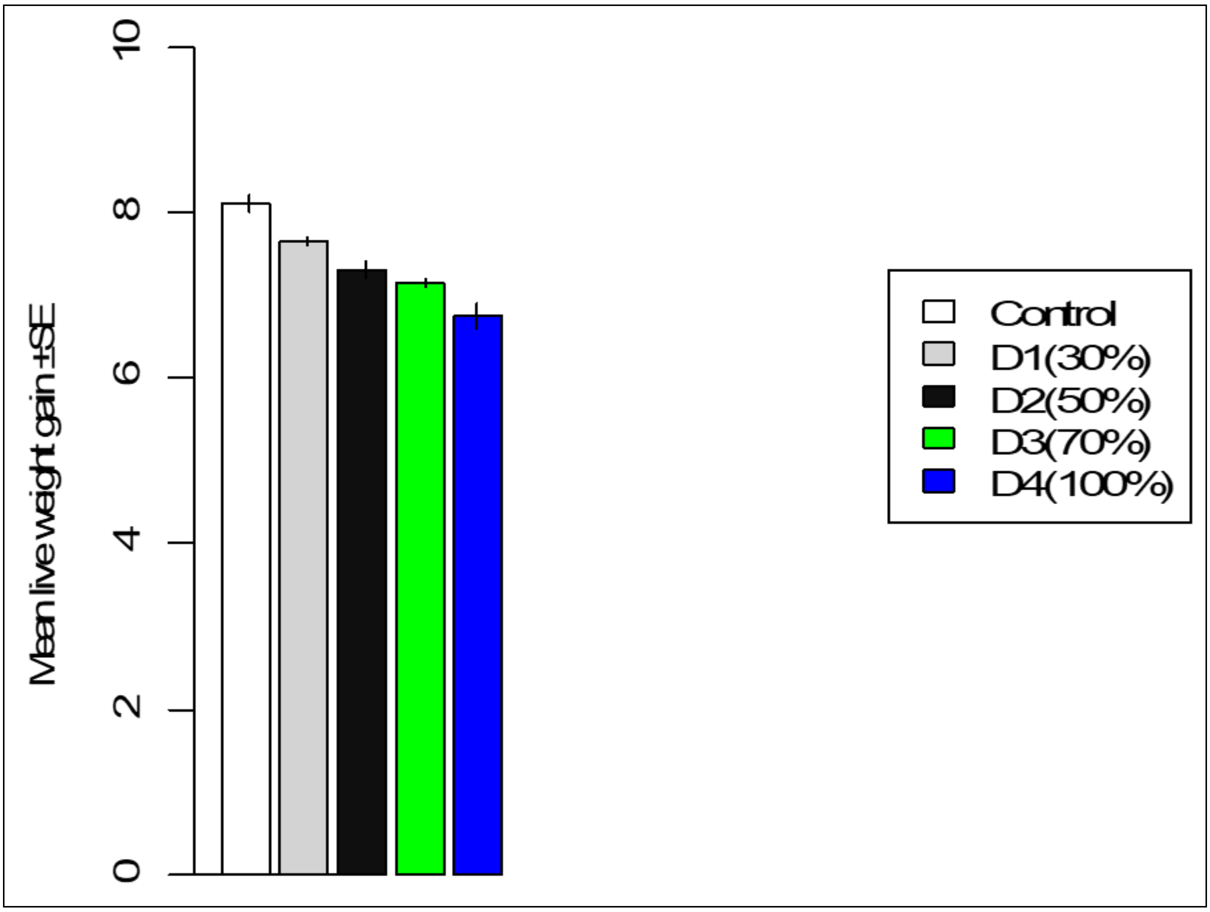


**Fig 2:** Comparison across live weight gain of *Clarias gariepinus* fingerlings fed varying percentages of mice meal and reference diet for 8 weeks

## 4. Discussion

Proximate analysis revealed nutritional makeup of a diet. The proximate analysis revealed that the experimental diet's crude protein level ranged from 33.4% in D5 to 35.4% in D2, with 35.6% in D1, the control diet. This was somewhat in line with the findings of Kenge and Ofojekwu (2022) [12], who found that all diets aside from diet D1, which had 36.79% crude protein contained between 31-35% crude protein. Moreover, Faturoti (2003) [8] proposed that fingerlings and juvenile *Clarias gariepinus* require 35 percent crude protein. Water quality is very important to aquaculture since it increases fish productivity. The test and reference diets did not affect the physicochemical parameters, based on the fact that the values of the water quality parameters under observation did not change substantially. The temperature of water has a significant impact on the metabolism of a fish. All of the study's observed temperatures were the same in the test and reference diets, which were 24 °C. It was below the findings of Adeosun *et al.* (2017) [1], who recorded 28.2°C-29.1°C as temperature for raising African catfish *Clarias gariepinus* and above the finding of Kenge and Ofojekwu (2022) [12] who reported the average temperature of 22.82±0.33°C to be within tolerant range. The result of dissolved oxygen monitored showed a range of 4.10-4.37 mg/L, this value falls below the findings of Afia and David, (2019) [3] who reported the range of 5-5.4 mg/L for aquaculture. Free carbon dioxide content was in the range of 7.20mg/L-7.5mg/L. The values were within the range of what was obtained by Kausar, and Salim, (2017) [11], who reported an optimal range of free Carbon dioxide in aquaculture water to be 5-8 mg/l. Total alkalinity range from 70.70-71.45mg/L. These total alkalinity values were within the optimum range of Faturoti (2004), who reported 20-300mg/L alkalinity was ideal for fresh water fishes. However, it was slightly different from the findings of Kirya (2011) [13], who reported 75 mg/l-200 mg/l to be the optimal range of alkalinity in water quality parameters in aquaculture systems. The result of pH obtained in this study showed that pH ranges from 7.1 to 7.4 mg/l. Bhatnagar and Devi (2013) reported the pH range between 7.0 and 8.5 to be optimum and suitable to fish. pH of 7.0 to 8.5 is good for biological productivity as fish become stressed in water with pH ranging from 4.0 to 6.5 and 9.0 to 11.0 (Ekubo and Abowei, 2011) [7]. According to Food and Agricultural Organization (2018), growth performance is one of the most important factors for aquaculture economics, considering that the operational costs for aqua feed are between 50 and 70%. In this study, fish fed D5 (100% mice meal inclusion) showed lower growth performance than the control group with fishmeal as the source of protein. This confirms previous observation by Rosenau, *et al.* (2021) [17] that a total replacement of fishmeal with Spirulina (*Arthrospira platensis*) resulted in reduced growth performance in African Catfish (*Clarias gariepinus*).Contrary to this present findings, Djissou *et al.* (2016) [6] reported improved growth performances and feed utilization in the diet containing the highest earthworm and maggot in the ratio of 2.5 compared to the fishmeal based diet which produced the lowest growth performance. The major growth indices such as the Mean growth, Specific growth rate and live weight gain decreased with an increase in the concentration of the mice meal. This report is in line with the finding of Michael, and Kolapo (2017) [14], who reported a decrease in growth indices of African catfish *Clarias gariepinus* with an increase in the concentration of the grasshopper meal in place of fishmeal and then recommended the lowest percentage of the grasshopper meal in the fish diet as an alternative for fishmeal in case the fishmeal is not available. Food Conversion Ratio helps in evaluating the wise use of feed (Afia *et al.*, 2019), [2, 3]. Knowledge of FCR is essential for feeding fish the precise amount of feed needed to prevent stress and produce high-quality meat suitable for human consumption (Shabir *et al.*, 2003) [18]. FCR in this study were slightly below the recommended limit of 2 for commercial culture for D2-D5 and approximately 2 for D1. This is in line with what was obtained by Ojewole (2022) [16]. Protein Efficiency Ratio (PER) and Apparent Net Protein Utilization (ANPU), results were used as an indicator for the protein productive value and were also within a good range. PER values ranged from (1.25) in fish fed diet D1 to (1.13) in fish fed diet D5.This findings is relatively similar to the findings of Kenge and Ofojekwu (2022) [12]. For Apparent Net Protein Utilization (ANPU), values ranged from (67.27%) in fish fed diet D1 to (37.91%) in fish fed diet D5. Fish growth and development, as well as production costs, are significantly impacted by the choice of protein source (Barroso *et al*., 2014) [5].

## 5. Conclusion and Recommendations

The study found that there was a significant difference ($p$<0.05) between the control and treatment groups, indicating that the control diet (D1) had the most growth performance. Fish that were fed with 30% mice meal inclusion, however, showed good growth performance. Less weight gain was observed when the dietary mice meal was increase above 30%.Thus where fishmeal-formulated diets are not readily available, adding 30% mice meal to the diet of African catfish may act as a backup plan.

Since there is limit to how big fish can grow in plastic baths, especially the size used in this investigation. There is need for additional investigation in larger water bodies like concrete tanks, earthen pond respectively. It would also be beneficial to examine the efficacy of mice meal as a source of protein in fish feed with different cultural fish species in Nigeria. Further investigation should be done on mice meal's tendency against growth.

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