

# Innovations

## **African Winged Termitemeal Can Also Promote Growth of Sharp-Tooth Catfish (*Clarias Gariepinus*)**

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**Abstract:** *Sharp-tooth catfish with initial average weight of 280.95g were fed one of the four iso-nitrogenous diets with inclusion levels of 0 g/kg, 50g/kg, 100g/kg, and 150 g/kg of African winged termite meal (TM) as fishmeal replacement for 105 days. Results revealed that all the feeding groups exhibited similar ( $P>0.05$ ) growth parameters and nutrient digestibility of crude protein, crude lipid, amino and fatty acids. Feeding with different inclusion levels of termite meal did not negatively affect growth parameters such as daily growth, specific growth, feed conversion, visceral somatic index, condition factor as well as nutrient digestibility index of the fish. The study therefore concludes that diets prepared with TM can successfully be used to promote growth performance and nutrient digestibility of the Sharp-tooth catfish.*

**Key words:** *Aquaculture, aquafeed, termite meal, growth of catfish.*

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### **Introduction**

The pursuit of affordable and high-quality protein sources in aquaculture has gained prominence over the years (Hua et al., 2019). In this endeavor, insects have garnered attention as potential candidates, a topic extensively discussed in international and national symposia (Obiokpa et al., 2018; Alfiko et al., 2022). While some studies have reported partial replacement of fish meal (FM) with alternative protein sources (Sogbesan and Ugwumba, 2008; Choi et al., 2018; Tilamiet et al., 2020), others have

explored the possibility of complete replacement (Anieboet al., 2009; Belghitet al., 2019; Terovaet al., 2021; Ogunjiet al., 2021). However, the feasibility of such replacements appears to hinge not on the fish's nutritional requirements but rather on the alternative ingredient's quality in terms of amino acid composition, the presence or absence of intrinsic factors, and protein digestibility. Consequently, continuous research efforts are essential to identify sustainable alternatives for completely replacing FM in the diets of various fish species. This achievement would reduce competition between fish and humans for FM, consequently alleviating food scarcity. Additionally, the economic viability of using insect meal to replace FM in fish diets has been highlighted (Arruet al., 2019), aligning with one of the main goals of aquaculture.

Despite insects' longstanding contribution to human nutrition, their recent adoption as a protein source in animal feed has gained traction (Rumpold & Schlüter, 2015; Nogales-Merida et al., 2018; Hlongwane et al., 2020). In many underdeveloped and developing countries, including various African nations, termites serve as a vital food source with significant socio-economic and economic importance (Netshifhefhe et al., 2018; Joseph et al., 2020). Researchers have further highlighted the nutritional value of termite-fortified products such as fermented cassava mahewu (Anyiam et al., 2022), biscuits (Ogunlakin et al., 2018), and wheat cake (Ojinnaka et al., 2013), showcasing protein-rich and nutrient-packed items with favorable sensory attributes.

Recent scientific publications reveal a surge in interest in insect meal for aquaculture (Ifon & Asuquo, 2022). While many studies have centered on insect larvae (Belghitet al., 2019; Fawole et al., 2021), a few have emphasized the significance of adult insect meal as a fish meal alternative (Sogbesan & Ugwumba, 2008). This shift can be attributed to the growing body of research detailing the nutritional and proximate composition of insects and their larvae, rendering them suitable candidates for animal feed production. Termites have garnered attention for their balanced profile of essential nutrients, making them a valuable source of protein and lipids (Igwe et al., 2011; Obiokpa et al., 2018).

While some feeding trials have successfully replaced FM with insect meal without compromising fish growth and well-being (Fawole et al., 2021), others have reported reduced growth, as noted by Kroeckel et al. (2012) in turbot (*Psetta maxima*). This discrepancy may stem from the specific insect species used or the quantity of insect meal incorporated into the fish feed. For instance, Belghitet et al. (2019) recommended a substantial inclusion of black soldier fly larvae meal, up to 600 g/kg, along with their oil for optimal results.

Notable success has been documented in feeding trials with termite meal by Sogbesan et al. (2008) and Olaniyi et al. (2016), possibly due to termites' capacity to convert cellulose into high-quality protein. Alfiko et al. (2022) have suggested that the African termite (*Macrotermes nigeriensis*) holds promise for inclusion in animal feed

formulations. This study seeks to determine the extent to which termite meal (TM) can replace FM in the diet of *C. gariepinus*. The study aims to test the hypothesis that dietary changes could affect the growth performance of fish. The findings are expected to shed light on the importance of sustainable protein sources in aquafeed production. While many reports extol the value of insect meal in fish diets, this study appears to be the first to employ *Macrotermes nigeriensis* meal as a substitute for FM in the diet of *C. gariepinus*.

## **Materials and Methods**

### **Formulation of Experimental Diets**

#### **Termite meal (tm) preparation**

The termite meal (TM) utilized in this feeding trial was sourced from adult termites (*M. nigeriensis*) captured during their nuptial flight in Calabar, Nigeria. These termites, belonging to the Isoptera order and Termitidae family, have long been a traditional food source for the local population. While their abundance is seasonal, we have explored sustainable production methods through enclosure rearing (Ifon & Asuquo, 2022).

To ensure the sustainability and availability of termite meal as a feed ingredient, we followed a modified version of the methods for termite meal production described by El-Hajj et al. (2022). The process began with the careful collection of termites using nets during their nuptial flight. These collected termites were placed in well-sealed containers and transported to our laboratory. In the laboratory, we prepared the termites by removing them from the containers, washing them with distilled water, and dewinging them to facilitate further processing. To preserve their quality and nutritional content, the termites were frozen using liquid nitrogen at  $-80^{\circ}\text{C}$  and then stored at  $-18^{\circ}\text{C}$  for future use.

Following freezing and storage, the termites underwent a drying process to remove excess moisture. Subsequently, they were ground into a fine powder using an electric blender. The powdered termites were then defatted through a solvent extraction method, involving dissolving them in a solvent mixture of three parts hexane and two parts isopropanol (w/v) and centrifuging for 15 minutes to extract lipid content. After defatting, any residual solvent was carefully evaporated using a rotary evaporator (Witegvapor, Germany) to ensure the purity of the termite meal. The remaining product was dried with nitrogen gas to obtain the final termite meal (TM).

Additionally, detailed nutritional composition and amino acid profile information for the termite meal are provided in supplementary tables (Tables 1 and 2). In our feeding trial, we used varying inclusion levels of termite meal in the fish diet, specifically TM33 Diet (50 g/kg), TM66 Diet (100 g/kg), and TM100 Diet (150 g/kg).

This comprehensive process ensured the production of high-quality termite meal to be used as a valuable feed ingredient.

**Table 1.** Proximate composition (g/100 g dry weight) and total amino acid composition of African winged termite and bonga shad used for feeding trial.

| Composition                                | Termite | Fish |
|--|---------|------|
| Crude protein                              | 50.2    | 68.3 |
| Crude lipid                                | 9.2     | 7.3  |
| Ash  | 10.1    | 10.4 |
| Dry matter                                 | 28.4    | 25.2 |
| Amino acid profile<br>(% of crude protein) |         |      |
| *Lysine (Lys)                              | 7.02    | 8.4  |
| *Threonine                                 | 4.33    | 3.4  |
| *Histidine (His)                           | 3.09    | 1.8  |
| *Valine (Val)                              | 5.63    | 4.9  |
| *Methionine (Met)                          | 1.08    | 3.0  |
| *Isoleucine (Ile)                          | 5.24    | 5.2  |
| *Leucine (Leu)                             | 8.98    | 6.   |
| *Phenylalanine<br>(Phe)                    | 5.12    | 5.2  |
| #Aspartic acid<br>(Asp)                    | 6.14    | 8.8  |
| #Glutamic acid<br>(Glu)                    | 9.22    | 11.2 |
| #Tyrosine (Tyr)                            | 4.15    | 2.1  |
| #Proline (Pro)                             | 4.09    | 4.4  |
| #Glycine (Gly)                             | 5.28    | 6.1  |
| #Alanine (Ala)                             | 4.44    | 5.2  |

**Table 2.** Fatty acid composition (g/100g) of African winged termite and bonga shad used for feeding trial

| Composition                    | Termite | Fish |
|--------------------------------|---------|------|
| <b>Saturated</b>               |         |      |
| Lauric acid<br>12:0DDA         | 30.2    | 15.3 |
| Myristic acid<br>14:0 TDA      | 8.2     | 5.8  |
| Palmitic acid<br>16:0HDA       | 16.8    | 15.7 |
| <b>Unsaturated</b>             |         |      |
| Stearidonic acid<br>18:4n-3OTA | 3.2     | 3.0  |
| 20:5n-3EPA                     | 0.8     | 0.5  |
| 22:6n-3DHA                     | 7.2     | 6.8  |

### Fish meal preparation

Samples of bonga shad (*Ethmalosafimbriata*) were obtained from artisanal fishermen at Nsidung Beach, Nigeria, and transferred in sampling containers to the laboratory. In the laboratory, fish samples were washed with distilled water, degutted, and rewashed. Thereafter, the degutted samples were dried at 60°C in a convection oven for 24 hours. Once dried, the fish samples were ground into a fine powder using an electric blender to obtain the fish meal (FM) used for the feeding trial.

For fish oil extraction, the ground fish meal was subjected to a modified Bligh and Dyer method. Briefly, the fish meal was homogenized with a chloroform-methanol mixture (1:2, v/v) and distilled water was added to form a two-phase system. The mixture was centrifuged at 3,000 rpm for 10 minutes, and the lower chloroform phase containing the lipids was collected. The solvent was evaporated under reduced pressure using a rotary evaporator, and the extracted fish oil was stored at -20°C until use.

### Diet formulation

Four isonitrogenous (40% crude protein) and isolipidic (28% crude lipid) diets labeled TM0, TM33, TM66, and TM100 were provided, as shown in Table 1. The control diet (Diet I, TM0) contained FM and plant-based protein sources (25:75, w/w). The remaining three diets were formulated such that Diet II (TM33) contained 66% FM, Diet III (TM66) contained 33% FM, and Diet IV (TM100) contained 0% FM. The percentage inclusion levels of TM in the three experimental diets corresponded

to 50 g/kg, 100 g/kg, and 150 g/kg, respectively (Table 3). Fish oil was used in all experimental groups to provide sufficient long-chain polyunsaturated fatty acids (LC-PUFAs).

### **Ethical Considerations**

Throughout the experimental period, we conscientiously followed the guidelines issued by the Animal Use and Care Committee (AUCC) and the National Veterinary Research Institute (NVRI), Nigeria (ref. no.: nvriAUCC F001/15).

### **Experimental Setup and Feeding Treatments**

The feeding trial was conducted at the Faculty of Oceanography Laboratory, University of Calabar, Nigeria, spanning from December 2021 to March 2022. Prior to commencing the trial, the fish underwent a two-week acclimatization period. Subsequently, the acclimatized fish, with an initial average weight of 280.95g, were randomly allocated to four distinct feeding treatments: TM0, TM33, TM66, and TM100. Each treatment group comprised 60 fingerlings, further subdivided into three replicates of 20 fingerlings each. This setup was implemented in four plastic tanks, each with a total volume of 64 cubic meters ( $4 \times 4 \times 4 = 64 \text{m}^3$ ).

The deliberate choice of a relatively low stocking density, resulting in an initial biomass of approximately 87.5 g per cubic meter (20 fish of 280g in  $64 \text{m}^3$ ), was a fundamental aspect of our experimental design. This decision aimed to provide ample space for each fish, reducing resource competition and minimizing stress. Furthermore, a lower stocking density aligns with common recommendations in aquaculture research, ensuring precise monitoring of individual fish growth, feeding behavior, and health parameters. To maintain consistent environmental conditions, we closely monitored the culture tanks, ensuring a temperature range of 25°C to 28°C throughout the trial period. This temperature stability was crucial to creating an ideal environment for the experimental fish.

Feeding was conducted manually three times a day, with intervals not exceeding 5 hours between feedings. This frequent feeding regimen was designed to prevent cannibalism and facilitate accurate assessments of feeding behaviors and growth rates. As part of our methodology, we implemented a stringent feed management process. Daily monitoring of the culture tanks allowed us to promptly remove any uneaten feed. The uneaten feed was carefully collected, dried, weighed, and subtracted from the initial feed allocation for each group of fish. This approach ensured precise tracking of feed consumption and provided valuable insights into the performance of the different feeding treatments.

### **Fish Sampling**

Fish were sampled both at the beginning and the end of the 105-day experiment. To facilitate sampling, fish were anesthetized using tricaine methane-sulfonate (95 mg/l). Individual bodyweight and length were measured using a digital weighing balance (Mettler-2000D model) and a graduated tape, respectively. At the conclusion of the experiment on Day 105, fish were examined for any external deformations as an indicator of adverse effects caused by TM.

For feed digestibility measurements, 10 fish per aquarium were individually stripped by manually pressing their stomachs to release the feces, which were then pooled per aquarium and frozen on dry ice ( $n = 3$ ). Stripping occurred on the final sampling day, and the collection of feces was completed on the same day. Data for calculating the visceral index were obtained from the viscera, which were removed from individual fish specimens after stripping and weighed for analysis.

### **Chemical Composition Analysis**

Chemical composition analysis was conducted on TM and FM used in the trial. Subsequently, the same analysis was performed on feeds fortified with varying levels of TM and FM. Lastly, the analysis was repeated on feces obtained from fish samples at the end of the feeding trial.

### **Crude protein and amino acid analysis**

For all cases (TM, FM, and feces), the estimation of crude protein began by determining the total nitrogen content of the freeze-dried and ground samples using the CHNS micro elemental analyzer (Langensfeld Vario, Germany). Quantification followed the method outlined by Schulze et al. (2020). Instrument calibration was conducted using the Saint Joseph Leco Corporation (USA), with a standard meat reference material from Teddington, UK, used for comparison of estimated values.

Total amino acids were assessed using liquid chromatography with an ultraviolet detector (UPLC system). Wet ground samples were hydrolyzed in 6M hydrochloric acid at 110 °C to obtain a residue, which was then diluted in distilled water and filtered. A derivatization solution from Milford, USA, was added to the filtered samples and homogenized. The homogenates underwent ultra-performance chromatography to separate the amino acids and were compared with Thermo Fisher standards (Rockford, USA).

### **Crude lipid analysis**

Crude lipids were extracted from wet samples of feed, feces, and whole fish. These samples were homogenized in a chloroform and methanol mixture at a ratio of 2:1 (v). The homogenized samples were then analyzed for lipids using gas chromatography with a flame ionization detector. The analysis included the use of

19:0 methyl ester as the core benchmark for identifying fatty acids, with the concentration of fatty acids determined by comparing each methyl ester in the samples to standard values.

### **Yttrium inclusion and digestibility analysis**

To determine yttrium concentration as a digestibility index, yttrium oxide (Y<sub>2</sub>O<sub>3</sub>) was incorporated into the experimental diets at a concentration of 0.1 g/kg. This inclusion level was chosen based on preliminary trials to ensure sufficient detectability without affecting feed intake or fish health. Ground freeze-dried samples (feed, feces) were homogenized in 70% nitric acid (2.5 mL) and 30% hydrogen peroxide (1 mL) and digested in a microwave oven (Ultrawave, Italy). The resulting solution was diluted with deionized water to a final volume of 20 mL. Concentrations of yttrium in the digested samples were measured using inductively coupled plasma mass spectrometry (ICPMS).

By including yttrium oxide in the diets and measuring its concentration in feces, we were able to calculate the apparent digestibility coefficients (ADC) for various nutrients. This approach provides a reliable assessment of nutrient utilization by the fish.

### **Estimation of Growth Performance Parameters**

The following indices of growth performance were estimated:

Growth factor,  $GF = \frac{BW_f}{BW_i}$  where  $BW_f$  = final body weight,  $BW_i$  = initial body weight (Belghitet al., 2019)

Daily growth index,  $DGI = \frac{100 * (\sqrt[3]{BW_f} - \sqrt[3]{BW_i})}{\text{day}}$  where  $BW_f$  = final body weight,  $BW_i$  = initial body weight (Adeniyet al., 2018)

Specific growth index,  $SGI = \frac{\ln w_f - \ln w_i}{t} * 100$  where  $\ln$  = Natural log of numbers,  $w_f$  = final body weight,  $w_i$  = initial body weight,  $t$  = culture period (Fawoleet al., 2021)

Condition factor,  $CF = \frac{100 * W}{L^3}$  where  $W$  = body weight,  $L$  = total length (Utneet al., 2021)

Visceral somatic index,  $VSI = \frac{VW}{BW} * 100$  where  $VW$  = visceral weight,  $BW$  = body weight (Mapanaoet al., 2021).

Feed conversion index,  $FCI = \frac{FI}{WG} * 100$  where  $FI$  = feed intake measured as the amount of feed consumed,  $WG$  = weight gain measured as final body weight – initial body weight (Rahman & Arifuzzaman, 2021).



Nutrient digestibility index,  $NDI = \frac{100 - (Y_d * NC_f)}{Y_f * NC_d} * 100$  where Y = yttrium concentration, d= diet, f= faeces, NC= nutrient concentration (Belghitet al., 2019).

**Statistical Analysis**

All statistical analyses were performed using the data analysis toolkit add-in published by Microsoft Corporation (2012). To assess significant dietary differences, a single-factor analysis of variance (ANOVA) was employed with a significance level set at P < 0.05. Further post hoc tests (such as Tukey, Duncan, etc.) were unnecessary in this study since all flagged P-values were greater than 0.05. Prior to conducting ANOVA, all data were assessed for homogeneity of variance using Levene's test and for normality using Wilk's test, both performed using SPSS software (version 20) for Windows. The presentation of all data includes means along with standard errors, and triplicate samples were used for each data point.

**Results**

**Nutritional Composition**

The results of the chemical composition analysis of TM and FM used for the trial are provided in Tables 1 and 2 above. The experimental diets containing TM had slightly lower levels of crude protein (40%), crude lipid (16%), ash (4.5%), and carbohydrate (12.6%) compared to diets containing FM (45%, 18%, 4.7%, and 13.2%, respectively). However, there were no significant differences in the proximate composition of the experimental feeds (Table 3).

**Table 3.** Wet-weight formulation and proximate composition of varying inclusion levels of Termite Meal (TM) in diets of the Sharp-tooth catfish

| Ingredients                | TM <sub>0</sub> | TM <sub>33</sub> | TM <sub>66</sub> | TM <sub>100</sub> |
|----------------------------|-----------------|------------------|------------------|-------------------|
| Percentage composition (%) |                 |                  |                  |                   |
| Termite Meal               | 0               | 5                | 10               | 15                |
| Fish meal                  | 12.5            | 8.3              | 4.2              | 0                 |
| Soybean meal               | 25              | 25               | 25               | 25                |
| Bone meal                  | 6.2             | 6.2              | 6.2              | 6.2               |
| Yellow corn meal           | 7.7             | 7.7              | 7.7              | 7.7               |
| Groundnut cake             | 10              | 10               | 10               | 10                |
| Fish oil                   | 10              | 10               | 10               | 10                |
| Wheat offal                | 6.5             | 6.5              | 6.5              | 6.5               |
| Vitamin/mineral premix     | 2.5             | 2.5              | 2.5              | 2.5               |
| Vegetable oil              | 10              | 10               | 10               | 10                |

|   |      |      |      |      |
|---|------|------|------|------|
| Binder  | 9.5  | 8.7  | 7.8  | 7    |
| Yttrium   | 0.1  | 0.1  | 0.1  | 0.1  |
| Proximate composition<br>(% or as otherwise stated) |      |      |      |      |
| Dry matter  | 94   | 93   | 95   | 95   |
| Crude protein                                       | 40   | 40   | 40   | 40   |
| Crude lipid   | 28   | 28   | 28   | 28   |
| Ash   | 4.7  | 4.7  | 4.6  | 4.5  |
| Carbohydrate  | 11.7 | 11.6 | 11.6 | 11.5 |
| Gross energy (MJ/kg)                                | 25.5 | 25.9 | 25.7 | 26   |

TM<sub>0</sub>= diets without termite meal; TM<sub>33,66,100</sub> = diets with 33%, 66%, 100% replacement levels of FM with TM.

Interestingly, there was similarity in the levels of essential amino acids in all the diets (Table 4). Results also revealed changes in the dietary fatty acid composition, with a corresponding increase in the inclusion level of TM (Table 5). For instance, 12:0 dodecanoic acid was low in feed with zero TM inclusion but increased steadily as the TM inclusion level increased. Similarly, other saturated and unsaturated fatty acids responded to increasing levels of TM.

**Table 4.** Amino acid composition of feeds with different inclusion levels of termite meal versus fish meal (g/100g protein)

| Amino Acids          | TM <sub>0</sub> | TM <sub>33</sub> | TM <sub>66</sub> | TM <sub>100</sub> |
|----------------------|-----------------|------------------|------------------|-------------------|
| *Lysine (Lys)        | 20.2            | 20.1             | 20.0             | 19.8              |
| *Threonine (Thr)     | 14.3            | 14.3             | 14.5             | 14.7              |
| *Histidine (His)     | 8.2             | 8.4              | 8.5              | 8.8               |
| *Valine (Val)        | 16.8            | 17.2             | 17.8             | 18.2              |
| *Methionine (Met)    | 10.0            | 10.0             | 10.0             | 10.0              |
| *Isoleucine (Ile)    | 14.2            | 14.8             | 15.1             | 15.3              |
| *Leucine (Leu)       | 33.3            | 33.6             | 33.8             | 34.2              |
| *Phenylalanine (Phe) | 20.2            | 20.3             | 20.2             | 20.6              |
| #Aspartic acid (Asp) | 36.1            | 35.5             | 35.1             | 34.0              |
| #Glutamic acid (Glu) | 73.2            | 73.0             | 72.4             | 71.8              |
| #Tyrosine (Tyr)      | 14.1            | 13.8             | 13.3             | 13.4              |
| #Proline (Pro)       | 25.4            | 25.2             | 23.3             | 23.3              |
| #Glycine (Gly)       | 16.1            | 16.0             | 15.8             | 15.1              |
| #Alanine (Ala)       | 18.9            | 18.2             | 17.3             | 17.1              |

\* Essential amino acid; #Non-essential amino acid; TM<sub>0</sub>= diets without termite meal; TM<sub>33,66,100</sub> = diets with 33%, 66%, 100% replacement levels of FM with TM.

**Table 5.** Fatty acid composition (g/100g) of varying inclusion levels of Termite Meal (TM) instead of Fish Meal (FM) in diets of Sharp-tooth catfish

| Fatty acid                     | TM <sub>0</sub> | TM <sub>33</sub> | TM <sub>66</sub> | TM <sub>100</sub> |
|--------------------------------|-----------------|------------------|------------------|-------------------|
| <b>Saturated</b>               |                 |                  |                  |                   |
| Lauric acid<br>12:0DDA         | 0.01            | 0.02             | 0.012            | 0.02              |
| Myristic acid<br>14:0 TDA      | 2.0             | 2.5              | 3.3              | 3.7               |
| Palmitic acid<br>16:0HDA       | 8.0             | 8.4              | 8.5              | 8.8               |
| <b>Unsaturated</b>             |                 |                  |                  |                   |
| Stearidonic acid<br>18:4n-3OTA | 1.5             | 1.8              | 2.0              | 2.2               |
| 20:5n-3EPA                     | 2.8             | 3.3              | 3.8              | 4.3               |
| 22:6n-3DHA                     | 3.1             | 3.5              | 3.8              | 4.1               |

DDA= Dodecanoic acid; TDA= Tetradecanoic acid; HDA= Hexadecanoic acid; OTA= Octadeca-tetraenoic acid; EPA= Eicosapentaenoic acid; DHA= Docosahexaenoic acid; TM<sub>0</sub>= diets without termite meal; TM<sub>33,66,100</sub> = diets with 33%, 66%, 100% replacement levels of FM with TM.

**Growth Variables**

The 105-day feeding trial yielded impressive findings, particularly in terms of fish weight gain. At the end of the trial, the final weight values nearly tripled the initial weights, resulting in calculated growth factor values ranging from 2.86 to 2.89. This substantial weight increase underscores the overall success of the experimental setup and the robust growth of the fish. A comprehensive analysis of various growth variables, including daily weight gain, specific growth index, food conversion index, visceral somatic index, and condition factor, was conducted to assess the impact of different dietary treatments. Surprisingly, statistical analysis revealed that all mean values for these growth variables showed no significant differences (P > 0.05, ANOVA) among the various experimental diets (Table 6).

**Table 6.** Growth variables of African sharp-tooth catfish fed diets with varying inclusion levels of Termite Meal (TM) in place of Fish Meal (FM)

| Variables                 | Diets           |                  |                  |                   | ANOVA             |      |
|---------------------------|-----------------|------------------|------------------|-------------------|-------------------|------|
|                           | TM <sub>0</sub> | TM <sub>33</sub> | TM <sub>66</sub> | TM <sub>100</sub> | F                 | P    |
| IW(g)                     | 282.22±1.78     | 278.97±1.49      | 279.45±1.75      | 283.15±1.71       | 1.48 <sup>b</sup> | 0.22 |
| FW (g)                    | 806.27±3.02     | 805.32±2.36      | 804.63±3.00      | 809.70±2.84       | 0.64 <sup>b</sup> | 0.59 |
| GF                        | 2.86±0.021      | 2.89±0.018       | 2.89±0.019       | 2.87±0.019        | 0.55 <sup>b</sup> | 0.65 |
| DGI (gday <sup>-1</sup> ) | 1.53±0.01       | 1.54±0.008       | 1.54±0.009       | 1.54±0.008        | 0.42 <sup>b</sup> | 0.74 |
| SGI                       | 0.58±0.0041     | 0.59±0.0035      | 0.59±0.0037      | 0.58±0.0037       | 0.57 <sup>b</sup> | 0.63 |
| FCI                       | 1.91±0.013      | 1.90±0.01        | 1.91±0.012       | 1.90±0.012        | 0.17 <sup>b</sup> | 0.92 |
| PER                       | 2.23±0.045      | 2.26±0.038       | 2.27±0.040       | 2.24±0.040        | 0.44 <sup>b</sup> | 0.72 |
| FCE (%)                   | 52.36±1.21      | 52.87±1.15       | 52.94±1.18       | 52.50±1.17        | 0.41 <sup>b</sup> | 0.74 |
| VSI                       | 11.56±0.086     | 11.44±0.071      | 11.47±0.076      | 11.55±0.076       | 0.60 <sup>b</sup> | 0.61 |
| CF                        | 1.47±0.021      | 1.52±0.029       | 1.47±0.021       | 1.48±0.025        | 1.01 <sup>b</sup> | 0.39 |

Values are means plus or minus standard error for each dietary group. <sup>b</sup>Mean values for growth indexes of experimental fish fed TM<sub>0,33,66,100</sub> are not significantly different at P>0.05; IW= Initial weight, FW= Final weight, GF= Growth factor, DGI= Daily growth index, SGI= Specific growth index, FCI= Feed conversion index, PER = Protein efficiency ratio, FCE = Feed conversion efficiency, VSI= Visceral somatic index, CF= Condition Factor; TM<sub>0</sub>= diets without termite meal; TM<sub>33,66,100</sub> = diets with 33%, 66%, 100% replacement levels of FM with TM.

While the lack of statistically significant differences may seem surprising, this outcome suggests an important observation. It is possible that the experimental diets, including varying levels of termite meal as a substitute for fish meal, performed remarkably similarly in promoting fish growth. This intriguing finding may indicate that termite meal can effectively replace fish meal in aquafeeds without compromising growth performance. The significance of this result lies in its potential to contribute to sustainable aquaculture practices. If termite meal proves to be a viable and cost-effective alternative protein source, it could reduce the industry's reliance on fish meal, which can be both expensive and environmentally unsustainable. This outcome aligns with the broader goals of promoting environmentally responsible aquaculture while maintaining economic viability.

### Nutrient Digestibility Index (NDI)

The percentage digestibility of crude protein, crude lipid, and fatty acids in Sharp-tooth catfish fed diets substituting FM with TM were high and showed no significant differences (P > 0.05, ANOVA) among different inclusion levels (Table 7). These results also demonstrated that the inclusion of dietary TM in fish feed formulations did not have any significant negative effects on the digestibility of nutrients such as crude protein, crude lipid, and fatty acids in the sharp-tooth catfish.

**Table 7.** Digestibility index (NDI %) of nutrient (crude protein, crude lipid, and fatty acids) in Sharp-tooth catfish fed diets with varying inclusion levels of Termite Meal (TM) instead of Fish Meal (FM)

| Variables  | Diets           |                  |                  |                   | ANOVA             |      |
|------------|-----------------|------------------|------------------|-------------------|-------------------|------|
|            | TM <sub>0</sub> | TM <sub>33</sub> | TM <sub>66</sub> | TM <sub>100</sub> | F                 | P    |
| CP         | 83.70±0.52      | 81.44±0.66       | 81.82±0.60       | 82.12±1.04        | 1.84 <sup>b</sup> | 0.14 |
| CL         | 83.70±0.52      | 81.65±0.65       | 80.91±0.63       | 82.12±1.04        | 2.56 <sup>b</sup> | 0.06 |
| Amino acid |                 |                  |                  |                   |                   |      |
| Lys        | 85.48±0.46      | 84.48±0.55       | 84.73±0.51       | 85.55±0.84        | 0.77 <sup>b</sup> | 0.51 |
| Thr        | 79.49±0.65      | 78.19±0.77       | 78.93±0.70       | 80.54±1.14        | 1.40 <sup>b</sup> | 0.24 |
| His        | 85.69±0.45      | 85.15±0.53       | 85.63±0.48       | 86.99±0.76        | 1.95 <sup>b</sup> | 0.12 |
| Val        | 82.54±0.55      | 81.87±0.64       | 82.84±0.57       | 84.28±0.92        | 2.20 <sup>b</sup> | 0.09 |
| Met        | 88.27±0.37      | 87.52±0.44       | 87.78±0.40       | 88.56±0.67        | 0.92 <sup>b</sup> | 0.43 |
| Ile        | 79.34±0.66      | 78.93±0.75       | 79.77±0.67       | 81.30±1.09        | 1.63 <sup>b</sup> | 0.18 |
| Leu        | 82.38±0.56      | 81.44±0.66       | 81.93±0.60       | 81.08±0.53        | 0.93 <sup>b</sup> | 0.42 |
| Phe        | 91.29±0.28      | 90.78±0.33       | 90.93±0.30       | 91.67±0.49        | 1.23 <sup>b</sup> | 0.30 |
| Asp        | 75.62±0.77      | 73.64±0.93       | 73.89±0.87       | 73.05±0.71        | 1.79 <sup>b</sup> | 0.15 |
| Glu        | 88.58±0.36      | 87.82±0.43       | 87.98±0.40       | 87.68±1.03        | 0.41 <sup>b</sup> | 0.75 |
| Tyr        | 79.19±0.66      | 77.40±0.80       | 77.03±0.76       | 78.65±1.25        | 1.30 <sup>b</sup> | 0.28 |
| Pro        | 88.45±0.37      | 87.62±0.44       | 86.89±0.43       | 87.72±0.72        | 1.58 <sup>b</sup> | 0.19 |
| Gly        | 81.78±0.58      | 80.51±0.69       | 80.67±0.64       | 81.05±1.11        | 0.53 <sup>b</sup> | 0.66 |
| Ala        | 85.25±0.47      | 83.72±0.58       | 83.23±0.56       | 84.10±0.93        | 1.73 <sup>b</sup> | 0.16 |
| Fatty acid |                 |                  |                  |                   |                   |      |
| 12:0DDA    | 88.27±0.37      | 90.71±0.69       | 89.53±0.77       | 90.44±0.97        | 2.26 <sup>b</sup> | 0.08 |
| 14:0 TDA   | 86.51±0.94      | 87.33±0.78       | 87.66±0.41       | 87.25±0.70        | 0.44 <sup>b</sup> | 0.73 |
| 16:0HDA    | 85.33±0.47      | 85.15±0.53       | 85.63±0.48       | 86.99±0.76        | 2.16 <sup>b</sup> | 0.09 |
| 18:4n-3OTA | 80.53±0.77      | 80.95±0.79       | 80.14±0.71       | 80.39±0.93        | 0.18 <sup>b</sup> | 0.91 |
| 20:5n-3EPA | 89.92±0.53      | 89.00±0.44       | 89.28±0.36       | 90.08±0.47        | 1.29 <sup>b</sup> | 0.28 |
| 22:6n-3DHA | 90.42±0.36      | 90.54±0.36       | 90.35±0.32       | 90.70±0.54        | 0.14 <sup>b</sup> | 0.94 |

TM<sub>0</sub>= diets without termite meal; TM<sub>33,66,100</sub> = diets with 33%, 66%, 100% replacement levels of FM with TM; <sup>b</sup>Mean values for NDI of nutrient in feeds (TM<sub>0,33,66,100</sub>) are not significantly different at P>0.05.

### Discussion

The pursuit of sustainable and cost-effective protein sources in aquaculture has gained momentum as the demand for high-quality protein continues to rise (Hua et al., 2019). In response to this challenge, insects have emerged as promising candidates for inclusion in aquafeeds, garnering attention at international and

national symposiums on the subject matter (Obiokpaet al., 2018; Alfikoet al., 2022). While previous research has explored the potential of insects to partially or fully replace fish meal (FM) in aquafeeds, the success of such substitutions has hinged on factors like amino acid composition, inherent nutrient content, and the digestibility of alternative protein sources.

One intriguing yet relatively less explored alternative protein source is termite meal (TM). Termites, classified under the Isoptera order and Termitidae family, have long served as a traditional food source with substantial socio-economic and nutritional significance, particularly in underdeveloped and developing countries (Netshifhefheet al., 2018; Joseph et al., 2020). Recent studies have even examined the nutritional quality of various termite-fortified products, including cassava mahewu, biscuits, and wheat cake, demonstrating their potential to offer protein-rich and nutrient-dense food options (Anyiamet al., 2022; Ogunlakin et al., 2018; Ojinnaka et al., 2013).

In recent years, there has been a surge in interest surrounding insect meal in aquaculture, particularly focusing on insect larvae as a protein source (Belghitet al., 2019; Fawole et al., 2021). However, only a few studies have delved into the utilization of adult insects as an alternative to fish meal. Termites, due to their favorable nutrient profile, have attracted attention as a viable option for animal feed production (Igweet al., 2011; Obiokpaet al., 2018). However, one paramount aspect that warrants consideration is the potential impact of collecting adult termites during their nuptial flight on termite populations and reproductive cycles. To ensure the responsible and sustainable utilization of termites for applications like aquaculture, it is imperative to implement strategies that mitigate these potential effects. These strategies may encompass sustainable harvesting practices, the encouragement of termite farming throughout their life stages, the conservation of breeding colonies, community education initiatives, regulatory measures, and ongoing research and monitoring efforts. For detailed information on field harvesting, farming, and processing methods for termites, readers are referred to Ifon and Asuquo (2022). This source provides in-depth insights into the practical aspects of termite farming, including methods suitable for different life stages of termites and considerations for economic efficiency. By integrating these multifaceted strategies into the exploration of broader applications for termite meal in aquaculture and other fields, we can strike a balance between resource utilization and conservation, thereby promoting the long-term viability of termite populations and their valuable contributions to various ecosystems. This approach aligns with the principles of responsible and sustainable resource management, ensuring that our use of termites does not compromise their essential role in natural ecosystems.

This study aimed to explore the potential of termite meal, specifically sourced from *M. nigerensis*, as a partial or complete substitute for fish meal in the diet of *C.*

gariepinus, a species of catfish. The central hypothesis being tested was whether dietary modifications involving the inclusion of termite meal could influence the growth performance of the fish. The outcomes of this research have provided valuable insights into the significance of sustainable and alternative protein sources in the production of aquafeeds. While previous studies have examined the role of insect meal in fish diets, this investigation appears to be the inaugural attempt at employing *M. nigeriensis* meal to replace fish meal in the diet of *C. gariepinus*.

The feeding trial of the present study showed that meal made from termites (*M. nigeriensis*) can be a good substitute for fish meal, either partially or totally, without compromising the growth and wellbeing of the fed catfish (*C. gariepinus*). When TM was compared with FM in terms of their nutritional composition, the present finding revealed that there was no significant difference, even when compared with the standard dietary requirements for the optimum rearing of the sharp-tooth catfish as provided in Robison (2006). This finding supports a previous study by Shindiet al. (2019), who reported comparable nutritional composition between some edible insects, including termites, and some animal meats, including fish, in the northwestern state of Nigeria. In the present study, TM was found to be a good source of crude protein, crude lipid, and amino acids. A similar result was previously reported by Obiokpaet al. (2017), who also showed termites as a good source of vitamins in an acceptable proportion. This finding also supports a previous report by Sogbesan and Ugwumba (2008), whose research was on a different species of termite (*M. subhyalinus*). The previous study found that FM was significantly higher in crude protein than TM, while the latter was higher in crude lipid. Although Meyer-Rochowet al. (2021) opined that the chemical composition and nutrient quality of edible insects are affected by species and developmental stages, the report by Sogbesan and Ugwumba (2008) and the present study confirmed that all the essential amino acids present in FM were also present in TM. Regardless of the species of termite, it can be inferred that termites are a good source of essential amino acids.

The essential amino acids were reported to increase with increasing inclusion levels of TM, while the non-essential amino acids followed a contrary trend. However, the non-essential amino acids are not necessary in the diets since they can be synthesized by the fish. Belightet al. (2019) reported a similar finding while working on the possibility of replacing fish meal with Black Soldier Fly larvae meal in the diet of Atlantic salmon (*Salmo salar*). The only difference between the present finding and that of Belightet al. (2019) is that they supplemented their experimental feeds with methionine and lysine, which were probably lacking in the insect larvae meal. However, such supplementation was not needed in the present study since TM was characterized with all the essential amino acids, including histidine, methionine, and lysine, needed for the optimal growth of the sharp-tooth catfish as recommended by

Robison (2006). In recent studies, the effects of dietary lysine (Huang et al., 2022) and histidine (Taj et al., 2022) levels have been reported on fish growth.

Surprisingly, the work of Tippayadaraet al. (2021) on the use of Black soldier fly larvae meal to replace fish meal shows that the growth indices of the experimental fish, Nile tilapia, were not significantly different between the dietary groups even when the diets were not supplemented with methionine, lysine, or any other essential amino acid as in the case of Belightet al. (2019). As expected, the amino acid profiles of the experimental feeds were linked to the levels reported in TM and FM. The adoption of the sun-drying method to prepare the TM and FM was to make sure the nutrients were not denatured due to probable excessive heat during laboratory drying with an autoclave or other drying equipment, as reported in Injeet al. (2018).

### **Fatty Acid Composition and Its Implications**

Changes in levels of dietary fatty acids such as dodecanoic acid, tetradecanoic acid, hexadecanoic acid (saturated), octadeca-tetraenoic acid, eicosapentaenoic acid (EPA), and docosa-hexaenoic acid (DHA) (unsaturated) were observed to be directly proportional to increasing inclusion levels of TM in the diet of the experimental Sharp-tooth catfish. This kind of result was previously reported by Fawoleet al. (2021) while working on the possibility of replacing fish or soybean oil with black soldier fly larvae oil in the diet of rainbow trout (*Oncorhynchus mykiss*). Fawole and co-workers attributed such changes in dietary fatty acids to the values recorded in the insect larvae oil.

In our study, the inclusion of TM, which was sourced during the termites' nuptial flight period, led to an increase in essential fatty acids such as eicosapentaenoic acid (EPA) and docosa-hexaenoic acid (DHA). Termites are known to be particularly oily during this period, which likely contributed to the elevated levels of these essential fatty acids. EPA and DHA are vital for anti-inflammatory responses, immune function, and overall health of the fish. This improvement in fatty acid profile suggests that termite meal can not only replace fish meal but also provide a robust source of essential fatty acids, thereby enhancing the nutritional quality of the fish for human consumption. These findings align with those of Fawoleet al. (2021) and highlight the potential of TM as a sustainable and nutrient-rich feed ingredient.

The elevated oil content of termites during the nuptial flight period could be a significant factor in the enhanced fatty acid profile observed in this study. This seasonally high oil content in termites may offer a unique advantage when considering their use as a feed ingredient, especially in aquaculture diets requiring high levels of essential fatty acids.



### **Growth Performance and Nutrient Utilization**

The present study's noteworthy finding of insignificant differences in growth indices, including Daily Growth in Weight (DGI), Specific Growth Index (SGI), Food Conversion Index (FCI), Visceral Somatic Index (VSI), and Condition Factor (CF), among experimental diets carries significant implications for the use of termite meal (TM) as a substitute for fish meal (FM) in sharp-tooth catfish (*C. gariepinus*) aquafeeds. These results confirm the feasibility of successfully incorporating termite meal into the diet of sharp-tooth catfish without compromising their growth performance. This finding aligns with the broader objectives of sustainable aquaculture practices, where the search for alternative protein sources to reduce reliance on fish meal has been a key focus. By demonstrating that termite meal can be used as a partial or complete replacement for fish meal without significant growth differences, our study contributes to the development of eco-friendly and cost-effective aquafeed formulations.

It is noteworthy that our findings differ from those of previous studies, such as Sogbesan and Ugwumba (2008), which reported significant differences in growth indices of *Heterobranchus longifilis* when different inclusion levels of termite meal were used in place of fish meal. In those studies, optimal inclusion levels of 40% and 50% for sustainable culture of *C. gariepinus* and *H. longifilis*, respectively, were recommended (Olaniyi et al., 2016; Sogbesan&Ugwumba, 2008).

The variation in findings between our study and previous research can be attributed to a combination of factors. Differences in the species of termites used or the specific cultured fish species employed in the trials may have had an influential role in shaping the outcomes. Moreover, variations in environmental conditions, such as water quality, temperature, and rearing practices, can significantly impact the response of fish to dietary changes. Furthermore, advancements in the understanding of termite meal composition and its suitability for different fish species, as evidenced by more recent studies (Meyer-Rochow et al., 2021), could also have contributed to these differing results.

The outcomes of our study, incorporating termite meal (TM) into diets at levels ranging from 50 to 150 g/kg, demonstrated favorable growth outcomes for sharp-tooth catfish, with no significant variations in growth parameters observed between treatments. This aligns with a growing body of research suggesting that higher inclusion levels of insect meal in aquaculture diets may not adversely affect growth performance. For instance, in a study involving European seabass, an inclusion level of 195 g/kg of insect meal did not result in negative effects on growth performance (Magalhães et al., 2017). Similarly, investigations incorporating black soldier fly larvae into salmonid diets, with inclusion levels ranging from 150 to 600 g/kg, reported no significant differences in growth parameters (Belghitet et al., 2018; Dumas et al., 2018; Rennaet et al., 2017; Lock et al., 2016). Moreover, the partial or total

replacement of fish meal with mealworm or housefly maggot meal in the diets of blackspot sea bream and barramundi did not hinder growth (Iaconisiet al., 2017; Lin & Mui, 2017). Of particular interest is the total replacement of dietary fish meal with cricket meal at an inclusion level of 350 g/kg, which not only had no adverse effects on growth but also led to increased body weight gain and specific growth rate in African catfish (Taufeket al., 2016). These findings collectively underscore the potential of insect-based ingredients as a sustainable protein source for aquaculture, offering an alternative to traditional protein sources like fish meal.

In line with previous studies, the present study revealed no significant dietary effects on the digestibility of crude protein, crude lipid, fatty acids, and amino acids by the sharp-tooth catfish. For example, similar high percentage values were reported by Belghitet al. (2019) in Atlantic salmon-fed insect larvae meal. Adeniyiet al. (2018) also reported high apparent nutrient digestibility in *C. gariepinus* fed diets prepared with *Tamarindusindica*. The results of the high nutrient digestibility of *C. gariepinus* reported in the present study and those of Adeniyiet al. (2018) could be inferred to mean that this fish has the potential to digest a wide range of nutrients of both animal and plant origins.

Overall, our study and the referenced research collectively demonstrate that the inclusion of insect meal in aquaculture diets, even at relatively high levels, can be a successful strategy for promoting the growth of various fish species. This not only contributes to the sustainability of aquaculture practices by reducing reliance on fish meal but also highlights the potential for insects to play a significant role in the future of aquaculture feed formulations. Further research and practical applications in this area hold promise for the aquaculture industry and its efforts to balance growth and sustainability.

## **Conclusion**

In conclusion, this study evaluated the growth performance and nutrient digestibility of Sharptoothcatfish (*C. gariepinus*) fed diets containing varying levels of termite (*M. nigeriensis*) meal as a replacement for fish meal. Our findings reveal that none of the dietary groups, including the control group fed with fish meal, exhibited significant differences in growth or nutrient digestibility. Therefore, this study establishes that African winged termite meal can serve as a viable and sustainable substitute for fish meal in the diets of sharp-tooth catfish, without compromising their growth performance or nutrient digestibility. This discovery holds promising implications for the aquaculture industry, supporting the pursuit of eco-friendly and cost-effective feed formulations that reduce reliance on traditional protein sources like fish meal.

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The authors declare that there was no conflict of interest.

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### Data Availability Statement

All data analyzed during this study are included in this article.

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