Egyptian Journal of Aquatic Biology & Fisheries
Zoology Department, Faculty of Science,
Ain Shams University, Cairo, Egypt.
ISSN 1110 – 6131
Vol. 29(5): 2139 – 2161 (2025)
www.ejabf.journals.ekb.eg

Comparative Effects of Plant-Based and Synthetic Premixes on Nutrient Digestibility and Whole-Body Composition in *Clarias gariepinus* Fingerlings

Emmanuel Oluwamuyiwa Oje[1, 2]*, Jeremiah Kang'ombe[1], Wilson L. Jere[1]

[1] Department of Aquaculture and Fisheries, Faculty of Natural Resources, Lilongwe University of Agriculture and Natural Resources, Malawi

[2] Department of Agriculture Biotechnology, National Biotechnology Development and Research Agency, Nigeria

*Corresponding Author: muyiwaoje@gmail.com / oje.oluwamuyiwa@nabrda.gov.ng

ARTICLE INFO

Article History:
Received: July 7, 2025
Accepted: Sep. 3, 2025
Online: Oct. 10, 2025

Keywords:
Nutrient utilization,
Plant-based premix,
Feed efficiency,
Aquaculture sustainability

ABSTRACT

This study addressed the challenge of reducing dependence on imported synthetic vitamin–mineral premixes (SVMP) in aquaculture by evaluating baobab (*Adansonia digitata*) and tamarind (*Tamarindus indica*) pulp powders as plant-based alternatives. While previous work has reported their effects on growth performance in *Clarias gariepinus*, little is known about their influence on nutrient digestibility and whole-body composition. Five isonitrogenous and isoenergetic diets were prepared: a control diet with 3% SVMP and four test diets with baobab or tamarind pulp powders incorporated at 5 and 7% (BPPVM and TPPVM). A total of 300 fingerlings (initial weight 5.54 g) were reared in triplicate groups and fed to satiation. Digestibility was determined using chromic oxide as an inert marker. Apparent digestibility coefficients (ADCs) for protein, lipid, dry matter, energy, ash, and fiber varied significantly across diets ($P< 0.001$). BPPVM diets achieved protein digestibility values (91.4–91.5%) comparable to the control, while TPPVM diets, particularly at 7%, showed reduced digestibility (protein ADC 73.6%). Whole-body composition patterns reflected these results: fish fed BPPVM diets had crude protein (62.4–62.9%) and lipid (7.35–7.42%) levels similar to the control, whereas TPPVM-fed fish recorded lower values. Result from a 90-day trial, growth performance and feed conversion ratio (FCR) supported these findings: BPPVM diets sustained growth and FCR comparable to the synthetic premix, while TPPVM groups showed reduced growth efficiency. These results indicate that baobab pulp powder is a viable alternative to synthetic premixes, while tamarind pulp requires further processing to reduce the impact of anti-nutritional factors. Future studies should explore mechanisms underlying nutrient–anti-nutrient interactions and assess micronutrient bioavailability over extended culture periods.

INTRODUCTION

Feed formulation plays a central role in aquaculture operations, accounting for approximately 60–70% of total production costs **(Naylor *et al.*, 2021)**. Among the critical dietary additives in aquafeeds are vitamin–mineral premixes, which support enzymatic activity, nutrient metabolism, immune responses, and tissue development in cultured fish **(Jimoh *et al.*, 2021)**.

However, the high cost and inconsistent availability of imported synthetic premixes constrain their regular use, particularly in resource-limited aquaculture sectors. Recent studies have focused on locally available, plant-based feed ingredients as alternative micronutrient sources for aquaculture. These natural materials supply essential vitamins and minerals and contribute bioactive compounds such as polyphenols, flavonoids, and organic acids that influence digestive function, nutrient absorption, and oxidative metabolism in cultured fish **(Samtiya *et al.*, 2020)**. Among these, baobab (*Adansonia digitata*) and tamarind (*Tamarindus indica*) pulps, valued for their regional availability and nutrient profiles, have gained particular interest as potential dietary additives. Baobab pulp is especially rich in ascorbic acid, calcium, and potassium, while tamarind pulp provides significant amounts of phenolic compounds and dietary acids, components known to affect digestive processes and nutrient assimilation in fish **(Hassan *et al.*, 2015)**.

While earlier studies have reported the effects of these plant materials on growth performance and hematological parameters in *Clarias gariepinus* **(Oje *et al.*, 2025)**, limited information exists regarding their influence on apparent nutrient digestibility and whole-body composition in this commercially important species. Apparent digestibility coefficients (ADCs) are essential indicators for evaluating how effectively fish utilize dietary protein, lipids, and energy **(Muin & Taufek, 2024)**. Likewise, whole-body composition analysis provides valuable insight into nutrient deposition, flesh quality, and market acceptability in aquaculture production **(Ochang *et al.*, 2007)**.

This study was conducted to investigate the replacement of synthetic vitamin–mineral premixes with baobab and tamarind pulp powders as plant-based alternatives, and assess their effects on the apparent nutrient digestibility and whole-body composition of *Clarias gariepinus* fingerlings. A conventional synthetic premix diet served as the control to evaluate the nutritional efficacy and suitability of these plant-based additives as sustainable alternatives for micronutrient supplementation in African catfish culture.

MATERIALS AND METHODS

Experimental site

The experiment was conducted at the Bunda Aquaculture Fish Farm and the wet laboratory of Lilongwe University of Agriculture and Natural Resources (LUANAR), Bunda Campus, Malawi (14°35′S, 33°50′E). The location was selected for its consistent water supply and capacity to maintain controlled experimental conditions. The trial lasted for 56 days (8weeks). A map of the study site is provided in Fig. (1) for geographic reference.

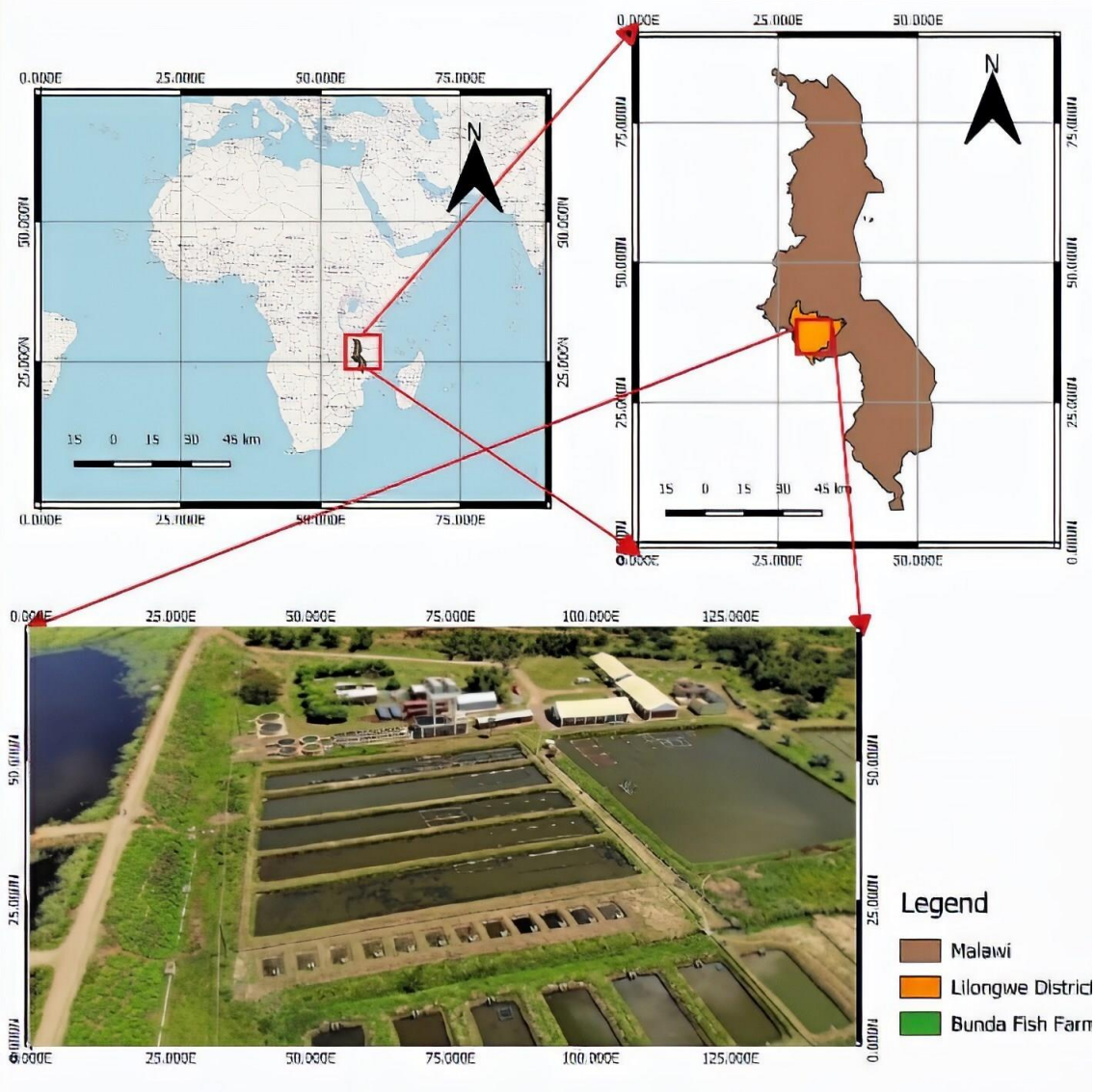

Fig. 1. A Map showing the geographical location of the study area

Feed ingredient sourcing, preparation, and processing

Diets were formulated using both conventional and plant-based ingredients: fishmeal, soybean meal, rice bran, maize meal, baobab pulp powder, and tamarind pulp powder. Baobab and tamarind powders were used as natural sources of vitamins and minerals in specific formulations. A synthetic vitamin–mineral premix (SVMP) was included only in the control diet. All ingredients were procured from licensed suppliers in Lilongwe, Malawi.

Soybeans were dry roasted at 110 °C for 15 minutes to inactivate heat-labile anti-nutritional factors, primarily trypsin inhibitors, and to enhance protein digestibility. Rice bran, maize meal, and fishmeal were finely milled and sieved to achieve uniform particle size and ensure consistent nutrient distribution. Baobab pulp powder was passed through a 0.5mm mesh to eliminate coarse fibers and impurities.

Tamarind pulp was pre-treated by soaking in warm water (40 °C) for 15 minutes to separate the edible mesocarp from seeds and fibrous tissue. The softened pulp was air-dried at room temperature (25–27 °C) for 48 hours in a clean, shaded, and well-ventilated area. This method aimed to preserve heat-sensitive micronutrients and to reduce moisture content, thereby improving storage stability. All processed ingredients were stored in airtight, moisture-free containers to prevent microbial contamination and nutrient degradation prior to diet formulation.

Proximate and micronutrient analysis of feed ingredients

Proximate composition of all feed ingredients was analyzed at the Aquaculture Nutrition Laboratory, Lilongwe University of Agriculture and Natural Resources (LUANAR), Bunda Campus, using standard methods outlined by the Association of Official Analytical Chemists **(AOAC, 2016)**. All analyses were performed in triplicate. Moisture content was determined by drying samples at 105°C in a hot-air oven for 24h. Crude protein was measured by the Kjeldahl method, involving digestion with concentrated sulfuric acid (H_2SO_4) and a copper catalyst, followed by distillation and titration. Total nitrogen was converted to protein using a factor of 6.25. Crude lipid was extracted using the Soxhlet method with analytical-grade hexane; 2g of each ground sample was refluxed for 6h in a pre-weighed extraction thimble. Ash content was determined by incinerating samples in a muffle furnace at 550°C for 4h. Crude fiber was measured using sequential acid alkali digestion. Samples were boiled with 1.25% sulfuric acid (H_2SO_4), followed by 1.25% sodium hydroxide (NaOH). The residue was filtered, oven-dried at 105°C, and incinerated at 550°C to quantify the indigestible fiber fraction, composed mainly of cellulose and lignin.

Micronutrient composition of baobab (*Adansonia digitata*) and tamarind (*Tamarindus indica*) pulp powders was analyzed for vitamin C, calcium, and phosphorus content on a dry weight basis (per 100g). Vitamin C was determined using the 2,6-dichlorophenolindophenol titration method **(AOAC, 2016)**. Phosphorus was measured using the molybdenum blue colorimetric method after acid digestion, with absorbance read at 880nm. Calcium was analyzed by atomic absorption spectrophotometry at 422.7nm.

Diet formulation and experimental treatments

Five isonitrogenous (35% crude protein) and isoenergetic diets were formulated, consisting of a control diet containing 3% synthetic vitamin–mineral premix (SVMP) and four experimental diets incorporating baobab pulp powder (BPPVM) and tamarind pulp powder (TPPVM) at two inclusion levels (5% and 7%) as plant-based premixes. All other dietary components, including macronutrient sources and energy content, were maintained constant across treatments to isolate the effects of premix type and inclusion level. Chromic oxide was incorporated at 0.5% in all diets as an inert marker for apparent digestibility determination. The detailed ingredient composition of the experimental diets is presented in Table (1).

Table 1. Ingredient composition (%) of experimental diets used in the nutrient digestibility trial for *Clarias gariepinus* fingerlings

Diet Composition	SVMP (3%)	BPPVM (5%)	TPPVM (5%)	BPPVM (7%)	TPPVM (7%)
Fish meal	26.8	26.3	26.3	25.8	25.8
Soybean meal	26.8	26.3	26.3	25.8	25.8
Rice bran	21.2	20.7	20.7	20.2	20.2
Maize meal	21.2	20.7	20.7	20.2	20.2
SVMP	3.00	0.00	0.00	0.00	0.00
BPPVM	0.00	5.00	0.00	7.00	0.00
TPPVM	0.00	0.00	5.00	0.00	7.00
Starch	0.80	0.80	0.80	0.80	0.80
Chromic oxide	0.50	0.50	0.50	0.50	0.50
Total	100	100	100	100	100

NB: BPPVM (Baobab pulp powder vitamin mineral) and TPPVM (Tamarind pulp powder vitamin mineral), +++ Synthetic vitamin and mineral (Vitamin A 15,000,000 i.u, Vitamin D3 4,000,000 i.u, Vitamin E 200,000 i.u, Vitamin B1 10,000mgr, Vitamin B12 1000mgr, Vitamin C300gr, Phosphorus 4000gr and Calcium 800gr),++ Plant based natural micro-nutrients (BPPVM and TPPVM), Diet composition: Diet 1 = 3% SVMP; Diet 2 = 5% Baobab pulp powder; Diet 3 = 5% Tamarind pulp powder; Diet 4 = 7% Baobab pulp powder; Diet 5 = 7% Tamarind pulp powder, Chromic oxide (Cr_2O_3) was added at 0.5% of the diet as an inert digestibility marker.

Experimental fish and holding conditions

African catfish (*Clarias gariepinus*) fingerlings used in the study were obtained from a single batch produced via controlled spawning to standardize genetic background, age, and physiological condition. From the post-hatch cohort, 300 uniform individuals (mean weight: 5.54 ± 0.5 g) were selected for the digestibility trial. Fish were maintained in a recirculating flow-through system (1.2 L/min exchange rate) and acclimated for 14 days under consistent photoperiod (12L:12D) and temperature conditions. During acclimation, fish were fed a commercial basal diet (35% crude protein) at 3% body weight per day in two equal portions. Individuals showing signs of stress, deformity, or abnormal feeding response were excluded. Water quality parameters were monitored daily and maintained within optimal limits for *Clarias. gariepinus* culture: temperature (26.5 ± 1.0 °C), dissolved oxygen (>6.0 mg/L), and pH (7.1–7.4). A digestibility trial was conducted over 21 days to determine the apparent digestibility coefficients (ADCs) of dry matter, crude protein, lipid, crude fiber, ash, and gross energy in *Clarias gariepinus* fingerlings. This trial was carried out independently following the completion of a 90-day growth study reported previously.

A total of 300 fingerlings (mean initial weight: 5.54 g) were randomly distributed into 15 plastic tanks (capacity: 50L), arranged in a completely randomized design with five dietary treatments and three replicates per treatment. Each tank was stocked with 20 fish. Prior to fecal collection, fish were acclimated to their assigned diets for 7 days.

Experimental diets were formulated to be isonitrogenous and isolipidic. Chromic oxide (0.5%) was included as an inert marker. All feed ingredients were weighed using a precision balance (±0.01 g), thoroughly dry-mixed using a ribbon blender for 10 minutes, and preconditioned with warm water (45°C) to facilitate pelleting. The mash was extruded using a 2 mm die, oven-dried at 60°C for 24 hours, and crushed to 0.8– 1.2mm to match fish size. Feeds were stored in airtight containers at 4°C until use. Each dietary batch was processed separately to avoid inter-batch variability.

Feeding and sampling protocol

Fish were fed the experimental diets by hand twice daily at 09:00 and 16:00 hours in two equal portions, at a rate equivalent to 3% of the total biomass per tank. The feeding rate was adjusted weekly based on collective biomass determined by bulk weighing of fish in each tank. To prevent contamination of fecal material with residual dietary matter, feed was withheld for 24 hours before sample collection, following the protocol described by **Hussain *et al.* (2011)**.

Fecal collection and processing

Fecal matter was collected using the modified settling-stripping technique validated for *Clarias gariepinus* digestibility studies **(Adewolu & Adeoti (2010)**. Experimental tanks were fitted with detachable bottom outlets to facilitate fecal settling. Uneaten feed was carefully siphoned from each tank 1h post-morning feeding to prevent mixing with fecal material. Feces were recovered 6 hours after feeding corresponding to peak intestinal evacuation in juvenile *Clarias gariepinus* **(Leenhouwers *et al.*, 2006)** using a narrow polyethylene tube to minimize water agitation.

Collected fecal material was gently rinsed with distilled water to remove extraneous debris and tank water residues, then immediately stored at –20°C to prevent nutrient degradation For each replicate, samples were collected daily for 10 consecutive days, pooled, and oven-dried at 70°C for 24 hours until weight variation between measurements was less than 0.05g. Dried fecal matter was finely ground using a laboratory mill and passed through a 0.5mm mesh sieve to obtain homogeneous samples for chemical and digestibility analyses.

Chemical and digestibility analysis

Chromic oxide (Cr_2O_3) content in diets and fecal samples was analyzed following the acid digestion protocol described by **Furukawa and Tsukahara (1966)**. Approximately 100mg of dried, ground sample was digested using a nitric–perchloric acid mixture (4:1 v/v), and absorbance was measured at 440nm using a UV–visible spectrophotometer (UV-1800, Shimadzu, Japan). Calibration was performed using certified chromic oxide standards before each batch of readings.

Proximate composition of diets and feces was determined according to **AOAC (2016)** official methods. Moisture content was measured by oven-drying at 105°C to constant weight. Crude protein was determined using the Kjeldahl method, while crude lipid was extracted with a Soxhlet apparatus using petroleum ether. Ash was measured by incineration in a muffle furnace at 550°C, and crude fiber was estimated using sequential acid–alkali digestion. Gross energy content was analyzed using an adiabatic bomb calorimeter (IKA C2000, Germany), calibrated with benzoic acid standards. All analyses were conducted in triplicate using analytical-grade reagents and properly calibrated instruments.

Apparent digestibility coefficients

Apparent digestibility coefficients (ADCs) for dry matter, crude protein, crude lipid, and gross energy were calculated using the indirect marker method described by **Furukawa and Tsukahara (1966)**, with chromic oxide (Cr_2O_3) included in the diets as an inert, indigestible marker at a known concentration.

The ADC for dry matter was computed as:

$$ADC_{DM}(\%) = 100 - (100 \times \frac{Cr_2O_3 \; in \; diet}{Cr_2O_3 \; in \; feces})$$

For nutrients (crude protein, crude lipid, gross energy), ADCs were calculated as:

$$ADC_{Nutrition}(\%) = 100 - (100 \times \frac{Cr_2O_3 \; in \; diet}{Cr_2O_3 \; in \; feces} \times \frac{Nutrient \; in \; feces}{Nutrient \; in \; diet})$$

All concentrations were expressed on a dry matter basis. Chromic oxide content in diets and fecal samples was determined by acid digestion and UV–visible spectrophotometry at 440nm, using certified standards for calibration, and nutrient concentrations were determined through proximate analysis. Digestibility was calculated for each replicate, with all measurements conducted in analytical triplicates.

Whole-body proximate composition analysis

At the end of the 90-day feeding trial, five fish were randomly selected from each replicate tank (n = 15 fish per dietary treatment) for whole-body proximate composition analysis. Fish were euthanized by immersion in an overdose of tricaine methane sulfonate (MS-222; 200mg/ L) in accordance with approved animal ethics guidelines. Each fish was rinsed with distilled water, blotted dry, and weighed to the nearest 0.01g using a calibrated precision balance (Model HL-200i, A&D Weighing, Japan).

The five fish from each replicate tank were pooled to form a composite sample (n = 3 per treatment), homogenized using a laboratory blender (Philips HR-7769, Netherlands), and stored in labeled polyethylene bags at −20°C until analysis. Proximate composition was determined in triplicate per treatment using standard **AOAC (2016)** procedures. Moisture content was measured by oven-drying at 105°C to constant weight. Crude protein was determined by the

Kjeldahl method using a nitrogen-to-protein conversion factor of 6.25. Lipid content was extracted using a Soxhlet apparatus with petroleum ether. Ash content was obtained by incineration in a muffle furnace at 550°C for 6 hours. Crude fiber was analyzed by sequential acid–alkali digestion, and gross energy was measured using a bomb calorimeter (IKA C2000, Germany) calibrated against benzoic acid standards.

Certified analytical-grade reagents were used in all assays. Equipment was calibrated daily using certified reference standards to ensure analytical precision and reproducibility.

Statistical analysis

All data analyses were conducted using R statistical software (version 4.5.1; R Core Team, 2025). Raw data were screened for missing values, outliers, and data consistency prior to formal analysis. Apparent digestibility coefficients and whole-body proximate composition variables were analyzed using a one-way analysis of variance (ANOVA) to assess the effects of dietary treatments. Diet was considered as a fixed categorical factor, while all response variables were continuous. Assumptions of normality, homogeneity of variance, and independence of residuals were verified using the Shapiro–Wilk test, Bartlett's test, and the Durbin–Watson statistic, respectively. When assumptions were satisfied, treatment means were compared using Tukey's Honest Significant Difference (HSD) test at a significance level of 0.05 to identify pairwise differences among treatments.

In addition to *P*-values, effect sizes were estimated as partial eta-squared (η^2) using the effect size package in R **(Ben-Shachar *et al.*, 2020)**. Partial eta-squared expresses the proportion of variance in each response variable explained by dietary treatments and thus reflects the magnitude of treatment effects. Effect sizes complement p-values by indicating the biological relevance of observed differences. Benchmarks for interpreting η^2 values followed **Cohen (1988)**, where 0.01, 0.06, and 0.14 denote small, medium, and large effects, respectively. All measured parameters in this study exhibited large effect sizes ($\eta^2 \geq 0.87$), confirming that dietary treatments exerted strong influences beyond statistical significance.

RESULTS

Apparent digestibility coefficients of experimental diets

Significant differences ($P < 0.001$) were observed in apparent digestibility coefficients (ADCs) for protein, lipid, dry matter, gross energy, ash, and fiber among the dietary treatments (Table 2, Fig. 2). Effect size analysis confirmed that treatment effects were biologically substantial, with η^2 values ranging from 0.93 to 0.99 across all parameters.

Protein and lipid digestibility

Apparent protein digestibility coefficients (ADCs) were the highest in fish fed the synthetic premix (91.7 ± 0.10%), baobab at 5% (91.4 ± 0.12%), and baobab at 7% (91.5 ±

0.08%), with no significant differences among these groups ($P > 0.05$). Tamarind-based diets produced significantly lower values (87.4 ± 0.23% and 73.6 ± 0.02% for 5% and 7%, respectively; $P < 0.05$), with the 7% tamarind group showing the lowest digestibility. The effect size for protein digestibility was very large ($\eta^2 = 0.96$), indicating that diet type accounted for nearly all variation. A similar trend was observed for lipid digestibility. Diets containing synthetic premix or baobab yielded values between 91.5–91.8 ± 0.05–0.08%, while tamarind diets were significantly lower (87.4 ± 0.21% and 85.6 ± 0.12%; $P < 0.05$). The corresponding effect size was also very large ($\eta^2 = 0.93$).

Dry matter and energy digestibility

Dry matter digestibility varied significantly among treatments ($P < 0.001$). Fish fed baobab at 7% recorded the highest ADC (79.4 ± 0.08%), significantly higher than all other groups ($P < 0.05$). Diets with synthetic premix (78.7 ± 0.10%) and baobab at 5% (78.4 ± 0.02%) were statistically similar ($P > 0.05$). Tamarind-based diets yielded the lowest values (72.3 ± 0.25% and 69.6 ± 0.12%), with the 7% inclusion group significantly lower than the 5% group ($P < 0.05$). The effect size for dry matter digestibility was extremely large ($\eta^2 = 0.99$). Gross energy digestibility followed the same pattern ($P < 0.001$). The highest value was observed in the baobab 5% group (92.4 ± 0.09%), followed by baobab 7% (92.0 ± 0.85%) and the synthetic premix (91.9 ± 0.13%), which did not differ significantly from each other ($P > 0.05$). Tamarind diets were significantly lower (89.4 ± 0.08% and 88.4 ± 0.06%; $P < 0.05$). The effect size for energy digestibility was $\eta^2 = 0.96$.

Ash and fiber digestibility

Ash digestibility coefficients differed significantly among treatments ($P < 0.001$). Diets containing synthetic premix (88.4 ± 0.08%), baobab 5% (88.6 ± 0.10%), and baobab 7% (88.7 ± 0.12%) were statistically similar ($P > 0.05$). Tamarind diets produced significantly lower values (83.7 ± 0.25% and 73.6 ± 0.12%; $P < 0.05$), with the 7% tamarind group showing the lowest digestibility. The effect size was extremely large ($\eta^2 = 0.99$). Fiber digestibility followed a similar statistical pattern ($P < 0.001$). Diets with synthetic premix and baobab (91.4–91.7 ± 0.22–0.24%) recorded the highest ADCs, while tamarind-fed groups exhibited significantly reduced values (83.0 ± 0.90% and 84.5 ± 0.83%; $P < 0.05$). No significant difference was found between the two tamarind diets ($P > 0.05$). The effect size for fiber digestibility was $\eta^2 = 0.99$. Effect size analysis confirmed that treatment effects were very large across all parameters ($\eta^2 = 0.93–0.99$), showing that dietary treatments accounted for nearly all observed variation in apparent digestibility coefficients

Table 2. Apparent digestibility coefficients (%) of nutrients in *Clarias gariepinus* fingerlings fed diets with synthetic and plant-based premixes

Parameter (%)	SVMP (3%)	BPPVM (5%)	TPPVM (5%)	BPPVM (7%)	TPPVM (7%)	p-values	Es (η^2)
Protein	91.70 ± 0.10^{a}	91.40 ± 0.12^{a}	87.40 ± 0.23^{b}	91.50 ± 0.08^{a}	73.60 ± 0.02^{bc}	< 0.001	0.96
Fat	91.80± 0.08^{a}	91.8±0.05^{a}	87.40 ± 0.21^{b}	91.50 ± 0.08^{a}	85.60±0.12^{b}	< 0.001	0.93
Dry matter	78.70± 0.10^{a}	78.40 ± 0.02^{a}	72.30 ± 0.25^{b}	79.40 ± 0.08^{a}	69.60±0.12^{c}	< 0.001	0.99
GE	91.90 ± 0.13^{a}	92.40 ± 0.09^{a}	89.40± 0.08^{b}	92.00± 0.85^{a}	88.40 ± 0.06^{b}	< 0.001	0.96
Ash	88.70 ± 0.01^{b}	88.40 ± 0.12^{ab}	83.70 ± 0.25^{c}	88.4 ± 0.08^{a}	73.60 ± 0.12^{c}	< 0.001	0.99
Fibre	91.70± 0.22^{a}	91.70± 0.24^{a}	83.00 ± 0.90^{b}	91.40± 0.23^{a}	84.50± 0.83^{b}	< 0.001	0.99

Values are means ± SE of triplicate groups (n = 3). Different superscript letters within a row indicate significant differences among treatments (Tukey's HSD, P < 0.05). P-values are from one-way ANOVA. Es = effect size, reported as partial eta-squared (η^2), which expresses the proportion of variance in each parameter explained by dietary treatments. Benchmark values of η^2 are 0.01 (small), 0.06 (medium), and ≥ 0.14 (large) (Cohen, 1988), with larger values indicating stronger treatment influence.

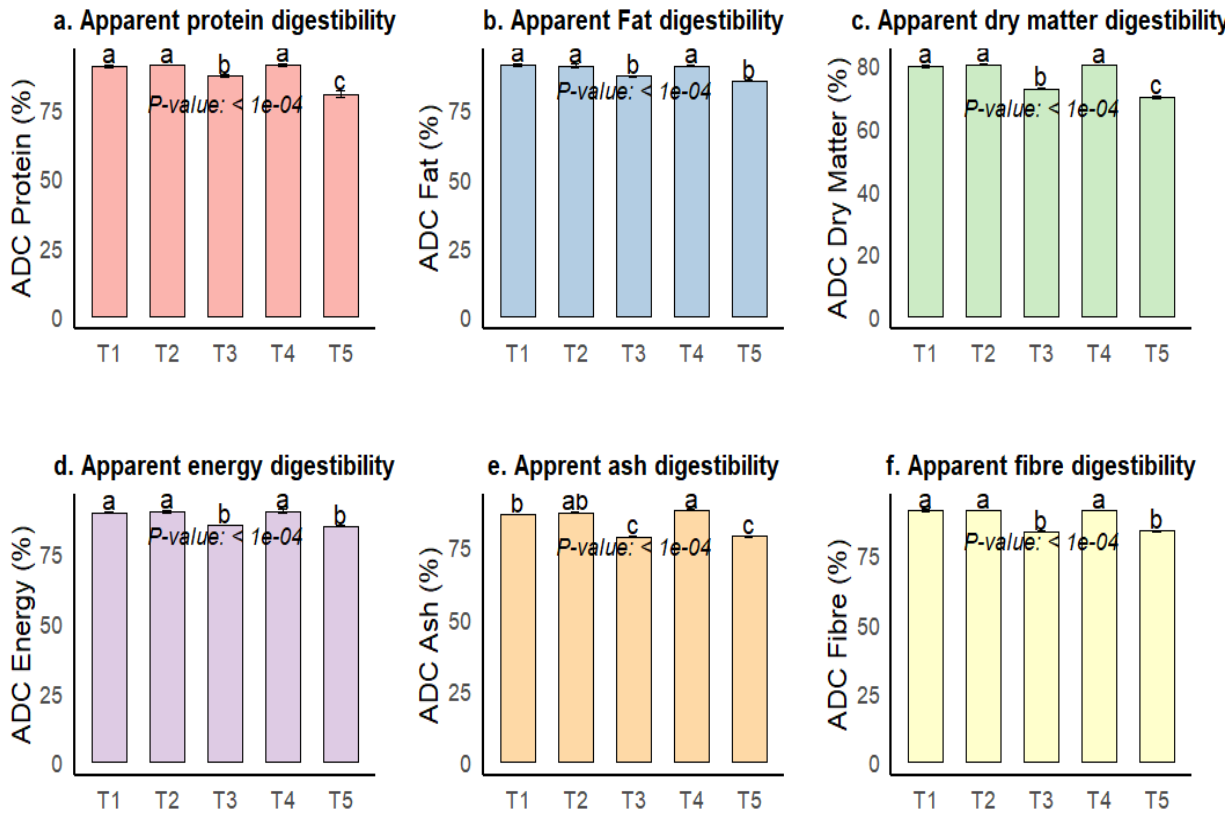

Fig. 2 Apparent digestibility coefficient of fish fed different diets (Bars represent mean + SE) Different letters indicate significant difference (Tukey's HSD, $P< 0.05$).

Whole-body composition

Significant differences ($P< 0.001$) were observed in the whole-body composition of *Clarias gariepinus* fingerlings across dietary treatments (Table 3 & Fig. 3). Effect sizes were consistently large ($\eta^2 \geq 0.87$), indicating that dietary treatments explained a substantial proportion of the variance in all measured parameters.

Fish fed diets containing baobab pulp (BPPVM at 5% and 7%) and the synthetic vitamin–mineral premix (SVMP) exhibited comparable crude protein contents (62.4–62.9%), which were significantly higher than those recorded in tamarind-based groups (TPPVM), where values ranged from 58.5 to 58.9%. Moisture content also varied significantly among treatments. The highest moisture levels were recorded in fish fed BPPVM (5%) and SVMP diets (72.9% and 72.7%, respectively), while BPPVM (7%) produced an intermediate value (72.1%). Both tamarind-based diets yielded significantly lower moisture levels (70.3–70.7%).

Crude lipid content was highest in fish fed BPPVM diets (7.35–7.42%), followed by the SVMP group (6.62%). Fish fed TPPVM diets recorded the lowest lipid deposition (5.10–5.27%).

Ash content followed a similar pattern. Higher values were observed in fish fed BPPVM diets (3.72–3.77%), intermediate values in the SVMP group (3.52%), and the lowest in the TPPVM-fed groups (3.15–3.21%).

Gross energy content ranged from 4.72 to 4.78 kcal/g in fish fed BPPVM and SVMP diets, which were significantly higher than those observed in tamarind-based diets (4.22–4.28 kcal/g). No significant differences were detected between the 5% and 7% inclusion levels within each premix type.

Comparative Effects of Plant-Based and Synthetic Premixes on Nutrient Digestibility and Whole-Body Composition in *Clarias gariepinus* Fingerlings

Table 3. Whole-body proximate composition (%) of *Clarias gariepinus* fingerlings fed diets with synthetic and plant-based premixes

Parameter	SVMP (3%)	BPPVM (5%)	TPPVM (5%)	BPPVM (7%)	TPPVM (7%)	p-values	Es (η^2)
Crude protein%	62.37 ± 0.25^{a}	62.92 ± 0.04^{a}	58.53 ± 0.15^{b}	62.36 ± 0.14^{a}	58.87 ± 0.03^{b}	< 0.001	0.99
Moisture%	72.71 ± 0.11^{a}	72.85 ± 0.02^{a}	70.71 ± 0.09^{c}	72.13 ± 0.05^{b}	70.32 ± 0.13^{c}	< 0.001	0.99
Crude lipids%	6.62 ± 0.20^{b}	7.42 ± 0.09^{a}	5.27 ± 0.04^{c}	7.35 ± 0.10^{a}	5.10 ± 0.03^{c}	< 0.001	0.98
Ash %	3.52 ± 0.07^{b}	3.77 ± 0.04^{a}	3.15 ± 0.02^{c}	3.72 ± 0.03^{a}	3.21 ± 0.03^{c}	< 0.001	0.95
Energy (kJg-1)	4.72 ± 0.09^{a}	4.73 ± 0.09^{a}	4.28 ± 0.03^{b}	4.78 ± 0.06^{a}	4.22 ± 0.04^{b}	< 0.001	0.87

Values are means ± SE of triplicate groups (n = 3). Different superscript letters within a row indicate significant differences among treatments (Tukey's HSD, P < 0.05). P-values are from one-way ANOVA. Es = effect size, reported as partial eta-squared (η^2), which expresses the proportion of variance in each parameter explained by dietary treatments. Benchmark values of η^2 are 0.01 (small), 0.06 (medium), and ≥0.14 (large) (Cohen, 1988), with larger values indicating stronger treatment influence.

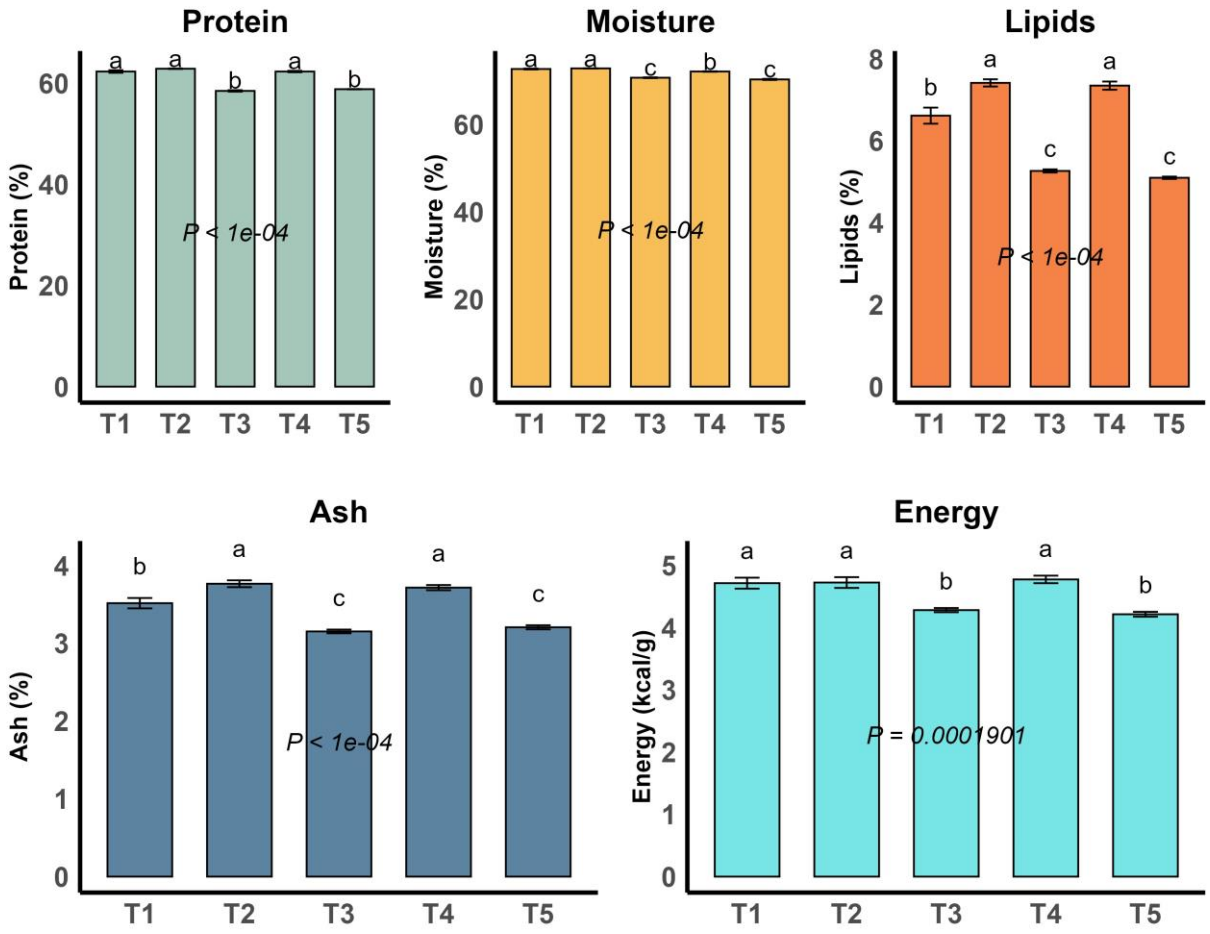

Fig. 3 Whole body composition of fish fed different diets (Bars represent mean + SE) Different letters indicate significant difference (Tukey's HSD, $P< 0.05$).

DISCUSSION

Apparent digestibility coefficients

The apparent digestibility coefficients (ADCs) for crude protein, lipid, dry matter, gross energy, ash, and fiber in *Clarias gariepinus* fingerlings were significantly influenced by dietary treatments ($P< 0.001$), with very large effect sizes across all parameters ($\eta^2 = 0.93–0.99$). The magnitude of these effect sizes confirms that the observed statistical differences correspond to strong biological effects, with diet type explaining the majority of variation in nutrient utilization.

Protein digestibility was significantly affected by diet type ($p< 0.0001$, $\eta^2 = 0.96$). Fish fed baobab (BPPVM) and synthetic premix (SVMP) diets showed higher ADC values (>91%), whereas tamarind-based diets, particularly at 7% inclusion, recorded markedly lower values (as low as 73.6%). The higher digestibility observed with baobab inclusion may be linked to its favorable amino acid profile and relatively low levels of protease inhibitors, which facilitate efficient proteolysis and amino acid absorption **(Hassan *et al.*, 2015)**. In contrast, tannins in tamarind pulp have been shown to reduce proteolysis and impair protein utilization **(Mandal & Ghosh, 2010)**. The very large effect size, together with the highly significant *P*-value, indicates that these differences represent a clear nutritional limitation of tamarind inclusion in catfish diets.

Lipid digestibility differed significantly among diets ($P< 0.0001$, $\eta^2 = 0.93$). Fish fed baobab- and synthetic premix diets showed higher ADC values (91–92%) compared with tamarind-based groups (85–87%). The higher digestibility in baobab-fed fish may reflect its relatively low fiber content, which facilitates bile salt emulsification and micelle formation, supporting efficient lipid absorption **(Bayon *et al.*, 2019)**. The markedly lower lipid digestibility observed in tamarind-based diets (ADC 68.4%) may be linked to their high insoluble fiber and polyphenol content, which are known to bind bile salts and interfere with micelle formation during lipid absorption **(McClements, 2018)**

Dry matter digestibility was significantly influenced by diet type ($p < 0.0001$, $\eta^2 = 0.99$). Fish fed baobab- and synthetic premix diets recorded higher ADC values (≈79%) compared with tamarind-fed groups (≤72%). The higher digestibility in baobab-based diets likely reflects greater availability of carbohydrates and other digestible fractions **(Hamed *et al.*, 2019)**. In contrast, the lower ADCs observed in tamarind-fed fish may be attributed to fiber and polyphenol interactions that interfere with enzymatic hydrolysis and reduce substrate availability for absorption **(Magalhães *et al.*, 2018)**.

Gross energy digestibility differed significantly among diets ($P< 0.0001$, $\eta^2 = 0.96$), showing that diet type explained almost all of the variation in energy retention. Fish fed baobab and synthetic premix diets achieved higher digestibility values (≈92%), while tamarind-based diets produced lower values (88–89%). The reduced energy utilization in tamarind-fed groups likely reflects cumulative effects of indigestible fiber fractions and phenolic compounds that interfere with metabolic efficiency, as reported by **Tran-Ngoc *et al.* (2019)**.

Ash digestibility was also significantly influenced by diet type ($P< 0.0001$, $\eta^2 = 0.99$). Baobab and synthetic premix groups recorded higher mineral digestibility compared with tamarind diets, with baobab's advantage likely reflecting its soluble calcium, potassium, and magnesium content **(Assogbadjo *et al.*, 2012; Hossain *et al.*, 2024)**. Conversely, the lower values in tamarind-fed fish are consistent with the presence

of phytates and oxalates that form insoluble mineral complexes, thereby impairing absorption **(Araújo *et al.*, 2018).**

Fiber digestibility differed significantly among diet groups ($P < 0.0001$, $\eta^2 = 0.99$), indicating that diet type accounted for almost all the observed variation. Fish fed baobab- and synthetic premix-based diets achieved higher ADC values (91%), consistent with the lower levels of resistant polysaccharides and anti-nutritional compounds in baobab pulp **(Assogbadjo *et al.*, 2012)**. In contrast, tamarind-based diets produced lower digestibility values (83–85%), which can be attributed to tannins and oxalates that restrict enzymatic access and limit fermentative breakdown **(Nwanna *et al.*, 2004; Muzaffar & Kumar, 2015)**.

Overall, the combination of highly significant *P*-values ($P < 0.001$) and large effect sizes ($\eta^2 > 0.90$) across all digestibility parameters indicates that the observed differences among treatments were both statistically robust and biologically relevant. Effect size analysis confirmed that diet formulation particularly the inclusion of baobab pulp was the primary determinant of nutrient digestibility variation in *Clarias. gariepinus*.

Whole body composition

Whole-body crude protein content was higher in fish fed the synthetic premix (SVMP) and baobab-based diets (BPPVM) compared to tamarind-based formulations (TPPVM). Values ranging from 62.4–62.9% in the SVMP and BPPVM groups are consistent with improved nitrogen retention, likely reflecting more efficient amino acid assimilation and reduced interference from anti-nutritional compounds. The very large effect size for protein ($\eta^2 = 0.99$) shows that dietary treatment explained almost all of the variance in carcass protein deposition, underscoring the critical role of premix type in supporting nitrogen accretion. This observation aligns with carcass protein values reported by **Ahmad (2012)** and supports the findings of **Hendam *et al.* (2024)**, who demonstrated improved protein deposition in *Clarias gariepinus* with baobab supplementation. In contrast, the lower protein content in TPPVM-fed groups (58.5–58.9%) is consistent with the presence of tannins and phytates, which impair proteolysis and amino acid absorption **(Adeniyi *et al*., 2018)**.

Energy content followed a similar trend, with higher gross energy retention in SVMP and BPPVM-fed fish (4.72–4.78 kcal/g) compared to those fed TPPVM diets (4.22–4.28 kcal/g). The large effect size for energy ($\eta^2 = 0.87$) indicates that diet composition accounted for most of the observed variance in carcass energy. These results corroborate previous reports that baobab pulp enhances energy availability due to its digestible carbohydrate profile and relatively low concentration of anti-nutritional factors **(Hamed *et al.*, 2019)**. The reduced energy values in tamarind-fed groups likely reflect

impaired nutrient absorption caused by fibrous cell wall components and phenolic compounds **(Adeniyi *et al.*, 2018)**.

Whole-body lipid content was also strongly influenced by diet, with the highest values recorded in fish fed BPPVM diets (7.35–7.42%), followed by SVMP (6.62%), and the lowest in TPPVM treatments (5.10–5.27%). The very large effect size ($\eta^2 = 0.98$) indicates that premix type was the dominant factor regulating carcass lipid deposition. Similar outcomes have been reported in studies showing enhanced lipid retention in fish fed baobab-based diets, attributed to improved lipid metabolism and digestibility **(Bayon *et al.*, 2019; Nosrati Movafagh *et al.*, 2019)**. The elevated lipid deposition may reflect baobab's favorable carbohydrate-to-lipid ratio, which promotes a lipid-sparing effect, as well as its low fiber content that allows efficient bile salt emulsification and micelle formation. By contrast, the reduced lipid values in TPPVM-fed fish are consistent with the high levels of insoluble dietary fiber and phenolic compounds in tamarind pulp, which interfere with emulsification and micellar solubilization, thereby restricting lipid absorption **(Nwanna *et al.*, 2004; Muzaffar & Kumar, 2015)**.

Moisture content was significantly higher in SVMP- and BPPVM-fed fish (72.7–72.9%) compared to TPPVM-fed fish (70.3–70.7%). The associated effect size ($\eta^2 = 0.99$) indicates that dietary treatment explained almost all of the variance in carcass moisture. Although higher moisture content is often associated with reduced nutrient density in fish tissues **(Shearer, 1994)**, the present findings contrast with that expectation. In this study, elevated moisture coincided with higher crude protein and energy retention, suggesting that diets containing baobab and synthetic premixes supported efficient nutrient assimilation alongside effective tissue hydration. Such outcomes are consistent with mechanisms described by **Seiliez *et al.* (2011)**, where improved amino acid uptake and reduced anti-nutrient interference promoted intracellular water retention.

Ash content also varied significantly among treatments, with higher values in fish fed baobab-based diets (3.72–3.77%), intermediate values in SVMP (3.52%), and the lowest in TPPVM-fed groups (3.15–3.21%). The very large effect size ($\eta^2 = 0.95$) confirms that diet strongly influenced mineral retention. The elevated ash values in BPPVM-fed groups are consistent with the high mineral density of baobab pulp, which contains appreciable levels of calcium, phosphorus, potassium, and magnesium **(Ajibade *et al.*, 2021)**. In contrast, the reduced ash values in tamarind-fed groups can be attributed to phytates and oxalates, which form insoluble complexes with minerals, reducing solubility and absorption efficiency **(Adeniyi *et al.*, 2018; Lall & Kaushik, 2021)**.

The combined use of *P*-values and effect sizes strengthens the interpretation of these results. While ANOVA detected highly significant differences among treatments,

the consistently large η^2 values (≥ 0.87) show that dietary formulation accounted for the majority of variance in carcass composition traits. Such high values are reasonable in the present context, given the strong nutritional contrasts between baobab- and tamarind-based premixes and the controlled experimental setting, which minimized background variability. This emphasizes that differences between baobab-, tamarind-, and synthetic premix diets were not only statistically detectable but also large in effect, with diet type emerging as the main determinant of nutrient retention and tissue composition in *Clarias gariepinus*

The lower digestibility recorded in tamarind-based diets can be partly explained by the presence of anti-nutritional factors such as tannins, phytates, and oxalates, which are known to interfere with protein and mineral utilization in fish **(Nwanna *et al.*, 2004; Muzaffar & Kumar, 2015)**. Although these compounds were not quantified in the present study, their reported abundance in tamarind pulp provides a plausible explanation for the observed trend. Processing may also have contributed. The mild pre-treatment applied to tamarind pulp (soaking and shade drying) likely helped preserve heat-sensitive vitamins but was insufficient to substantially reduce tannins or phytates. Soaking has been shown to lower some water-soluble anti-nutrients, but more intensive approaches such as roasting or fermentation achieve greater reductions, albeit with possible nutrient losses **(Samtiya *et al.*, 2020)**. These considerations highlight the trade-off between nutrient preservation and anti-nutrient reduction when processing plant-based premix ingredients.

In contrast, baobab pulp supported digestibility and carcass composition outcomes comparable to the synthetic premix, reflecting its favorable nutrient composition and relatively low levels of anti-nutritional factors. Baobab pulp is also widely available in sub-Saharan Africa and remains underutilized, suggesting potential for incorporation into aquafeeds.

From a practical standpoint, baobab pulp represents a cost-effective and locally accessible resource for feed formulation. Its seasonal abundance and traditional use as a food material suggest that reliable supply chains could be developed to support aquaculture applications. However, large-scale adoption would require standardization of processing methods and quality control to ensure consistency across batches. Incorporating such underutilized local resources could reduce reliance on imported synthetic premixes and improve the economic sustainability of small- and medium-scale aquaculture enterprises.

The patterns observed in digestibility and whole-body composition are consistent with growth performance data from a related 90-day trial **(Oje *et al.*, 2025)**, where baobab-based diets produced growth rates and feed conversion ratios comparable to the synthetic premix, whereas tamarind-based diets impaired growth efficiency. These

complementary findings strengthen the interpretation that higher nutrient digestibility in baobab diets translates into improved nutrient retention and growth.

A limitation of this study is the absence of direct quantification of anti-nutritional factors in the tested ingredients and diets. Their contribution to reduced digestibility in tamarind diets was inferred from literature and known compositional characteristics. Future work incorporating direct measurement of tannins, phytates, oxalates, and related compounds would enable a more comprehensive evaluation of plant-based vitamin–mineral premixes in aquafeeds.

CONCLUSION

Baobab pulp powder supported nutrient digestibility and whole-body composition of *Clarias gariepinus* fingerlings at levels comparable to the synthetic premix. Tamarind pulp inclusion reduced digestibility and nutrient retention, consistent with the effects of residual anti-nutritional factors. These findings identify baobab pulp as a potential plant-based premix alternative, while highlighting the need for improved processing of tamarind pulp.

Declaration of competing interest

The authors declare no conflict of interest.

Acknowledgement

The authors gratefully acknowledge the financial and institutional support provided by the German Academic Exchange Service (DAAD), National Biotechnology Development Agency (NABDA), Nigeria, Kenya Marine and Fisheries Research Institute (KMFRI), the Department of Aquaculture and Fisheries Science, and the African Center of Excellence in Aquaculture and Fisheries Science (AquaFish), Lilongwe University of Agriculture and Natural Resources (LUANAR), Malawi**.**

Quality assurance and ethical compliance

All chemical analyses were conducted in triplicate. The UV–visible spectrophotometer was calibrated daily using certified chromic oxide standards to ensure analytical precision. Diets were formulated and processed following Good Laboratory Practice (GLP) protocols to maintain consistency and reproducibility.

Ethical approval for animal handling and experimental procedures was obtained from the LUANAR Research Ethics Committee, in accordance with the National Research Ethics Guidelines of Malawi.

REFERENCES

Adeniyi, O. V.; Olaifa, F. E. and Emikpe, B. O. (2018). Growth performance and nutrient digestibility of *Clarias gariepinus* (Burchell, 1822) fed diets fortified with *Tamarindus indica* pulp and leaf meal. *Asian Fisheries Science*, *31*(1), 17–31.

Adewolu, M. A. and Adoti, A. J. (2010). Effect of mixed feeding schedules with varying dietary crude protein levels on the growth and feed utilization of *Clarias gariepinus* (Burchell, 1822) fingerlings. *Journal of Fisheries and Aquatic Science*, *5*(4), 304–310.

Ajibade, A. O.; Adeosun, O.; Sule, O. S.; Adedokun, M. A. and Adesina, B. T. (2021). Growth efficiency and profitability indices of African catfish *Clarias gariepinus* fingerlings fed with different levels of *Adansonia digitata* (Baobab) seed meal. *African Journal of Agricultural Research*, *17*(7), 1008–1015.

Ahmad, M.; Qureshi, T. A. and Singh, A. B. (2012). Effect of dietary protein, lipid and carbohydrate contents on the carcass composition of *Cyprinus carpio communis* fingerlings. *African Journal of Biotechnology*, *11*(33), 8353–8360.

Araújo, J. G.; Nebo, C.; Pádua, D. M. C.; Souto, C. N. and Guimarães, I. G. (2018). Apparent digestibility of minerals from several ingredients for Tambaqui, *Colossoma macropomum*, juveniles. *Journal of the World Aquaculture Society*, *49*(6), 1026–1038.

Association of Official Analytical Chemists (AOAC) (2016). *Official Methods of Analysis of AOAC International* (20th ed.). AOAC International, Rockville, MD, USA. Retrieved from http://www.worldcat.org/title/official-methods-of-analysis-of-aoac-international/oclc/476032693.

Assogbadjo, A. E.; Chadare, F. J.; Kakaï, R. G.; Fandohan, B. and Baidu-Forson, J. J. (2012). Variation in biochemical composition of baobab *(Adansonia digitata)* pulp, leaves and seeds in relation to soil types and tree provenances. *Agriculture, Ecosystems and Environment*, *157(3)*, 94–99.

Bayon, M. S.; Kpundeh, M. D.; Ikenweiwe, N. B.; Ngegba, P. M. and Ndoko, E. J. (2019). Effects of *Adansonia digitata* (Baobab) bark meal additive on growth performance and haematological parameters of *Clarias gariepinus* (Burchell, 1822) fingerlings. *International Journal of Fisheries and Aquatic Studies*, *7*, 280–286.

Ben-Shachar, M. S.; Lüdecke, D. and Makowski, D. (2020). Effectsize: Estimation of effect size indices and standardized parameters. Journal of Open-Source Software, 5(56), 2815. https://doi.org/10.21105/joss.02815

Cohen, J. (1988). *Statistical Power Analysis for the Behavioral Sciences* (2nd ed.). Hillsdale, NJ: Lawrence Erlbaum Associates. https://doi.org/10.4324/9780203771587

Dougall, H. W. (1956). Crude fibre: Its determination and its place in the analysis of animal feeding-stuffs. *The East African Agricultural Journal*, *21*(4), 225–229.

Food and Agriculture Organization (FAO) (2020). The state of food security and nutrition in the world 2020: transforming food systems for affordable healthy

diets. FAO, Rome, Italy. DOI: 10.4060/ca9692en.

Furukawa, A. and Tsukahara, H. (1966). On the acid digestion method for the determination of chromic oxide as an index substance in the study of digestibility of fish feed. *Bulletin of the Japanese Society of Scientific Fisheries*, *32*(6), 502–508.

Hamed, A. M.; Dungos, F. A. and Yagob, G. A. (2019). Effect of replacing fishmeal with baobab seed meal *Adansonia digitata* on growth, feed conversion and carcass composition for Nile Tilapia Fry *(Oreochromis niloticus*). *Egyptian Academic Journal of Biological Sciences, B. Zoology*, *11*(3), 97–105.

Hassan, M.; Abba, A. and Wakili, U. B. (2015). Effects of Replacing Soybean Meal with Baobab (*Adansonia digitata*) Seed Meal in the Diets of *Clarias gariepinus* (Burchell, 1822) Fingerlings. *Nigerian Journal of Fisheries and Aquaculture*, *3*(1 and 2), 42–48.

Hendam, B. M.; Baromh, M. Z.; Khafaga, A. F.; Shukry, M.; El-Son, M. A. M. and Abdel-Latif, H. M. R. (2024). Effects of dietary baobab, *Adansonia digitata* on growth, haemato-immunological status, antioxidant biomarkers, intestinal histomorphometry, gene expression responses, and disease resistance in Nile tilapia, *Oreochromis niloticus. Aquaculture*, *581*, 740473.

Henken, A. M.; Lucas, H.; Tijssen, P. A. T. and Machiels, M. A. M. (1986). A comparison between methods used to determine the energy content of feed, fish and faeces samples. *Aquaculture*, *58*(3–4), 195–201.

Hossain, M. S.; Small, B. C.; Kumar, V. and Hardy, R. (2024). Utilization of functional feed additives to produce cost-effective, ecofriendly aquafeeds high in plant-based ingredients. *Reviews in Aquaculture*, *16*(1), 121–153.

Hussain, S. M.; Afzal, M.; Salim, M.; Javid, A.; Khichi, T. A. A.; Hussain, M. and Raza, S. A. (2011). Apparent digestibility of fish meal, blood meal and meat meal for *Labeo rohita* fingerlings. *Journal of Animal and Plant Science, 21*(4), 807–811.

Jimoh, S. O.; Ishiaku, Y. M.; Burnett, T.; Amisu, A. A. and Adebayo, R. A. (2021). Potentials of leys or pasture-based forage production in Nigeria. *African Journal of Range and Forage Science*, *38*(3), 191–205.

Lall, S. P. and Kaushik, S. J. (2021). Nutrition and metabolism of minerals in fish. *Animals*, *11*(09), 2711–2726.

Leenhouwers, J. I.; Adjei-Boateng, D.; Verreth, J. A. J. and Schrama, J. W. (2006). Digesta viscosity, nutrient digestibility and organ weights in African catfish *Clarias gariepinus* fed diets supplemented with different levels of a soluble non-starch polysaccharide. *Aquaculture Nutrition*, *12*(2), 111–116.

Magalhaes, S. C. Q.; Cabrita, A. R. J.; Valentao, P.; Andrade, P. B.; Rema, P.; Maia, M. R. G.; Valente, L. M. P. and Fonseca, A. J. M. (2018). Apparent digestibility coefficients of European grain legumes in rainbow trout (*Oncorhynchus mykiss*) and Nile tilapia *Oreochromis niloticus*. *Aquaculture Nutrition*, *24*(1), 332–340.

Mandal, S. and Ghosh, K. (2010). Inhibitory effect of Pistia tannin on digestive enzymes of Indian major carps: an in vitro study. *Fish Physiology and Biochemistry*, *36*, 1171–1180.

McClements, D. J. (2018). Enhanced delivery of lipophilic bioactives using emulsions: a review of major factors affecting vitamin, nutraceutical, and lipid bio accessibility. *Food & Function*, *9*(1), 22–41.

Muzaffar, K. and Kumar, P. (2015). Parameter optimization for spray drying of tamarind pulp using response surface methodology. *Powder Technology*, *279(1)*, 179–184.

Muin, H. and Taufek, N. M. (2024). Evaluation of growth performance, feed efficiency and nutrient digestibility of red hybrid tilapia fed dietary inclusion of black soldier fly larvae (*Hermetia illucens*). *Aquaculture and Fisheries, 9*(1), 46–51.

Naylor, R. L.; Hardy, R. W.; Buschmann, A. H.; Bush, S. R.; Cao, L.; Klinger, D. H.; Little, D. C.; Lubchenco, J.; Shumway, S. E. and Troell, M. (2021). A 20-year retrospective review of global aquaculture. *Nature, 591*(7851), 551–563.

Nosrati Movafagh, A.; Keramat Amirkolaei, A. and Oraji, H. (2019). Effect of different levels of Spirulina meal on growth performance, survival feed efficiency, body composition and apparent digestibility in Oscar fish (*Astronotus ocellatus*). *Journal of Aquaculture Development*, *13*(3), 119–133.

Nwanna, L.; Fagbenro, O. and Olanipekun, S. (2004). Evaluation of tamarind (*Tamarindus indica*) seed meal as a dietary carbohydrate for the production of Nile tilapia *Oreochromis niloticus* (L). *Animal Research International, 1*(3), 164–168.

Ochang, S. N.; Fagbenro, O. A. and Adebayo, O. T. (2007). Growth performance, body composition, haematology and product quality of the African catfish (*Clarias gariepinus*) fed diets with palm oil. *Pakistan Journal of Nutrition*, *6*(5), 452–459.

Oje, E. O.**; Kang'ombe, J. and Jere, W. L.** (2025) Performance of *Clarias gariepinus* fingerlings fed baobab and tamarind pulp powders as sustainable alternatives to synthetic vitamin-mineral premix. *Egyptian Journal of Aquatic Biology and Fisheries*, 29(3), 2449–2472.

Samtiya, M.; Aluko, R. E. and Dhewa, T. (2020). Plant food anti-nutritional factors and their reduction strategies: an overview. *Food Production, Processing and Nutrition*, *2*, 1-14.

Seiliez, I.; Panserat, S.; Lansard, M.; Polakof, S.; Plagnes-Juan, E.; Surget, A.; Dias, K.; Larquier, M.; Kaushik, S. and Skiba-Cassy, S. (2011). Dietary carbohydrate-to-protein ratio affects TOR signalling and metabolism-related gene expression in the liver and muscle of rainbow trout after a single meal. *American Journal of Physiology-Regulatory, Integrative and Comparative Physiology*, *300*(3), R733–R743.

Shearer, K. D.; Åsgård, T.; Andorsdöttir, G. and Aas, G. H. (1994). Whole body elemental and proximate composition of Atlantic salmon *(Salmo salar)* during the life cycle. *Journal of Fish Biology, 44*(5), 785–797.

Shimizu, G.; Marubayshi, R. and Goncalves, I. (2024). *AgroR: Experimental statistics and graphics for agricultural sciences* [R package version 1.3.6]. Retrieved from https://CRAN.R-project.org/package=AgroR on 4th December, 2024.

Tran-Ngoc, K. T.; Haidar, M. N.; Roem, A. J.; Sendão, J.; Verreth, J. A. J. and Schrama, J. W. (2019). Effects of feed ingredients on nutrient digestibility, nitrogen/energy balance and morphology changes in the intestine of Nile tilapia (*Oreochromis niloticus)*. *Aquaculture Research, 50*(9), 2577–2590.