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Performance of *Clarias gariepinus* Fingerlings Fed Baobab and Tamarind Pulp Powders as Sustainable Alternatives to Synthetic Vitamin-Mineral Premix

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ABSTRACT

This study assessed the potential of baobab (*Adansonia digitata*) and tamarind (*Tamarindus indica*) pulp powders as sustainable replacements for synthetic vitamin-mineral premixes (SVMPs) in *Clarias gariepinus* fingerling diets. A completely randomized design was used to assess growth performance, hematological indices, and water quality across five dietary treatments: 3% Synthetic vitamin-mineral premix (SVMP) (control), 5% and 7% baobab pulp powder, and 5% and 7% tamarind pulp powder. A total of 300 fingerlings were randomly assigned to 15 experimental units and fed at 3% of body weight twice daily for 12 weeks. Fish fed baobab-based diets (5% and 7%) exhibited comparable weight gain and feed efficiency compared to those fed SVMP-based diets, while lower growth rates and reduced feed conversion efficiency were recorded for fish fed tamarind-based diets, suggesting poor nutrient utilization. Hematological analysis revealed no significant differences in red blood cell counts, hemoglobin concentration, or packed cell volume between fish fed baobab-based diets and those fed on SVMP diets, indicating adequate oxygen transport and immune function. However, fish fed tamarind-based diets exhibited significantly lower hematological values, suggesting anemia and physiological stress, possibly due to tannins and phytates impairing nutrient absorption. Water quality varied significantly among treatments. Diets containing SVMP (3%) and baobab pulp powder (5% and 7%) maintained stable pH and dissolved oxygen levels. In contrast, tamarind-based diets led to increased total dissolved solids and reduced dissolved oxygen, likely due to higher rate of organic matter decomposition and increased microbial oxygen demand. Baobab pulp powder demonstrated potential as an effective SVMP alternative, maintaining growth performance, hematological stability, and favorable water quality. However, tamarind pulp powder requires additional processing to mitigate its anti-nutritional factors. Future research should explore fermentation and enzymatic treatments as strategies to enhance nutrient bioavailability and feed efficiency in aquaculture.

INTRODUCTION

Feed constitutes the largest cost component in aquaculture, accounting for 60–70% of total production expenses **(Naylor *et al.*, 2021)**. A key contributor to this cost is the inclusion of essential micronutrients, particularly synthetic vitamin-mineral premixes (SVMPs), which are crucial for supporting growth, immunity, and feed efficiency.

However, the high cost and inconsistent availability of SVMPs, particularly in resource-limited regions, pose significant challenges to sustainable fish production **(Jimoh *et al.*, 2021)**. These challenges highlight the need for cost-effective, locally available feed additives that enhance fish performance while reducing dependence on synthetic formulations.

Plant-derived additives have been explored as cost-effective alternatives to synthetic feed components due to their bioactive compounds that enhance growth, immunity, and feed efficiency. Among these, baobab (*Adansonia digitata*) and tamarind (*Tamarindus indica*) pulps are rich sources of vitamins, minerals, and antioxidants, which could potentially replace SVMPs in aquafeeds. Baobab pulp is particularly high in vitamin C, calcium, and potassium, which support immune function and metabolic homeostasis **(Hassan *et al.*, 2015)**. Tamarind pulp, on the other hand, contains polyphenols, organic acids, and essential minerals that contribute to antioxidant defense and digestive health **(Samtiya *et al.*, 2020)**. While various parts of these plants have been studied in animal nutrition, their efficacy as SVMP replacements in fish diets remains underexplored, particularly concerning their effects on growth, health, and water quality.

Diet composition not only affects fish growth and health but also influences water quality dynamics in aquaculture systems. Nutrient leaching and metabolic excretion from different feed formulations can alter critical parameters such as pH, alkalinity, and ammonia levels, impacting overall system stability **(Almazán-Rueda *et al.*, 2004)**. While SVMPs are formulated to minimize fluctuations in water quality, there is limited research on how plant-based vitamin-mineral premixes affect aquaculture water dynamics. Furthermore, hematological indices such as red blood cell (RBC) count, hemoglobin (Hb), and packed cell volume (PCV) serve as important indicators of fish health and metabolic function **(Hrubec *et al.*, 2020)**, yet their response to plant-based premixes remains poorly documented.

Given the need for cost-effective and locally available alternatives to SVMPs, this study evaluated the potential of baobab and tamarind pulp powders as sustainable replacements in the diets of *Clarias gariepinus* fingerlings. Specifically, it investigated their effects on growth performance, hematological indices, and water quality parameters, providing insights into their viability for practical application in aquafeed formulation.

MATERIALS AND METHODS

Experimental site

The study was conducted at the Bunda Aquaculture Fish Farm and the wet laboratory at Lilongwe University of Agriculture and Natural Resources (LUANAR), Bunda campus, Malawi (latitude 14°35′S, longitude 33°50′E). LUANAR, recognized as the Africa Center of Excellence in Aquaculture and Fisheries Science, offers well-equipped research facilities and technical expertise. The site was chosen for its well-structured aquaculture facilities, reliable water supply, and controlled conditions suitable for experimentation. Fig. (1) illustrates the map of Bunda where the research was conducted. The experiment lasted 90 days (12 weeks).

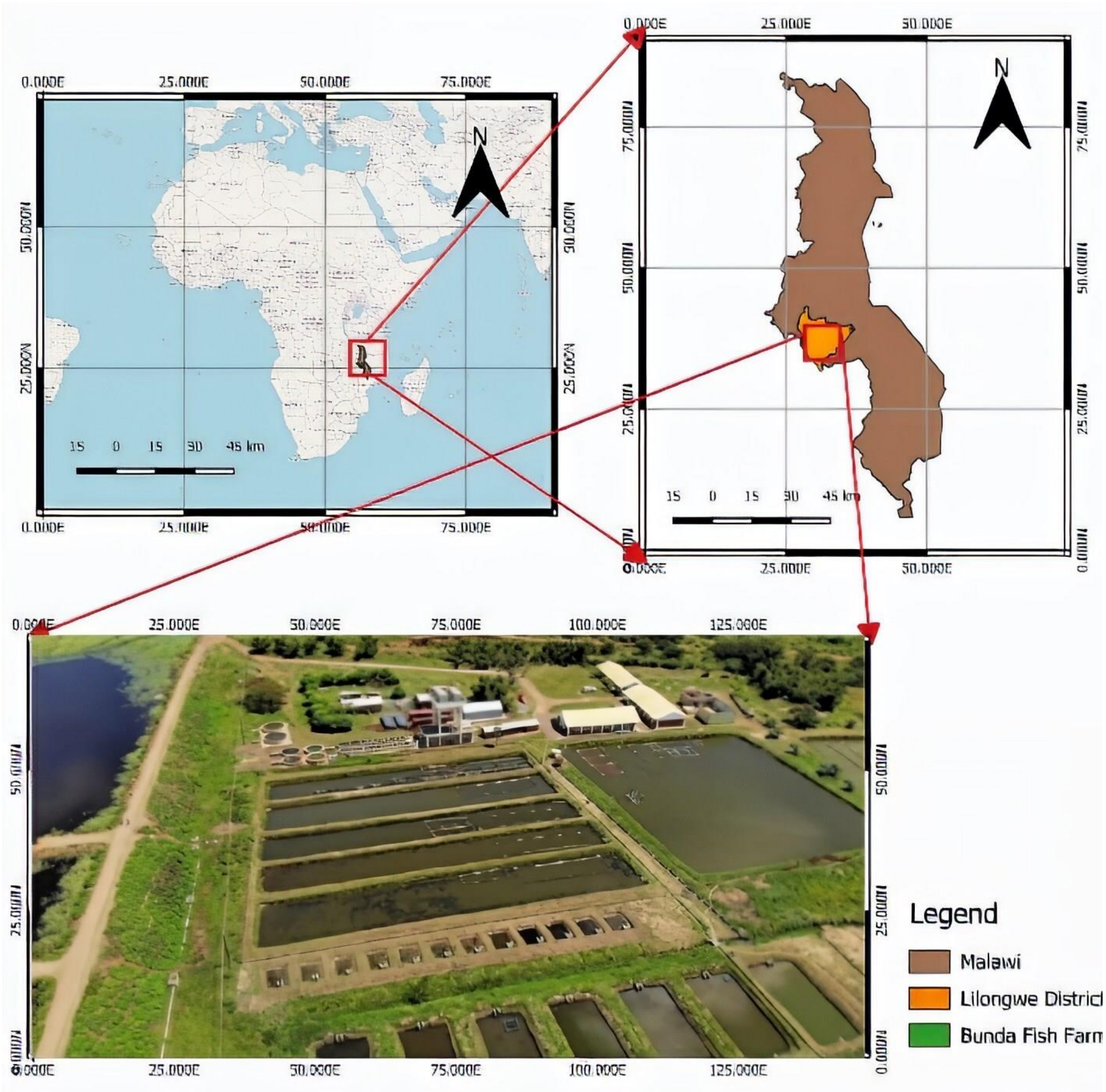

Fig. 1. Map showing the geographical location of the study area

Broodstock selection, acclimatization and spawning

Two sexually mature *Clarias gariepinus* females (average weight per fish: 1.5kg) and two sexually mature males (average weight per fish: 1kg) were obtained from the National Aquaculture Center in Zomba and Bunda Aquaculture Farm, respectively, to ensure genetic diversity and reduce inbreeding bias. Broodstock were selected based on morphological uniformity, absence of visible deformities, and external signs of good health to minimize phenotypic variability that might influence experimental outcomes. The selected fish were acclimatized for two weeks at the Bunda farm solar hatchery section under standard conditions.Throughout this period, water quality parameters such as temperature, dissoved oxygen, and ammonia were monitored and maintained within the recommended range for *Clarias gariepinus* culture. Fish were fed a commercial diet (Skretting, 40% crude protein, 6mm pellet size) twice daily.

Spawning was induced through hypophysation, following the method described by **Agbakwuru and Osuji (2024)**. Females broodstock received intramuscular injections of pituitary gland extract at a dosage of 0.5ml per kg of body weight and were maintained at 27°C under a continuous aeration. After a latency period of 12 hours, approximately 250g of eggs per female were manually stripped and fertilized with milt extracted from males broodstock.The fertilized eggs were incubated on standardized meshhatching subtrates at 26-28^{O}C. Hatching occurred within 18 to 24hours under controlled conditions.

Nursery phase and fingerling acclimatization

Newly hatched larvae were reared in nursery tanks for 45 days and fed a commercial starter diet (German Wean, 40% crude protein, 0.1–0.8mm pellet size). Growth was assessed weekly through measurements of total length and wet weight. Upon the completion of the nursery phase, 300 uniform and healthy fingerlings (8–10cm, 4–5.5g) were selected based on morphological uniformity and active swimming behavior for the feeding trial. Prior to the trial, fingerlings were acclimatized for six days in aerated rectangular plastic tanks (0.50m x 0.25m x 0.40m; 50L capacity) under controlled laboratory conditions. Water quality parameters such as temperature, dissolved oxygen, pH and ammonia were monitored daily to ensure optimal rearing conditions.

Feed ingredient sourcing, preparation and processing

Raw materials used for diet formulation included both conventional and plant based feedstuffs: fishmeal, soybean meal, rice bran, maize meal, baobab pulp powder, and tamarind pulp powder. Baobab and tamarind pulp powders were considered as natural sources of vitamins and minerals in selected formulations. A synthetic vitamin-mineral premix (SVMP) was included only in the control and comparison diets. All ingredients were sourced from licenced suppliers in Lilongwe Malawi. Soybeans were dry roasted at 110°C for 15 minutes to denature heat-labile anti-nutritional factors, primarily trypsin inhibitors, and to enhanced protein digestibility. Rice bran, maize meal and fishmeal were finely milled and sieved to ensure uniformity in particle size and homogeneous nutritional distribution of nutrients during mixing. Baobab pulp powder was filtered through a 0.5mm mesh to remove coarse fibers and debris. Tamarind pulp was pre-treated by soaking in warm water (40^{o}C) for 15 minutes to facilitate separation of edible mesocarp from the seeds and fibrous tissue. The softened pulp was air -dried at room temperature (25-27^{o}C) for 48 hours in a clean, shaded and well ventilated area. This technique was adopted to retain heat-sensitive micronutrients and reduced moisture content, thereby enhancing storage stability.

All processed ingredients were stored in airtight, moisture-free containers to prevent microbial contamination and nutrient degradation prior to formulation.

Proximate and micronutrient analysis of feed ingredients

The proximate composition of all raw feed ingredients was determined in triplicate in the Aquaculture Nutrition Laboratory, Lilongwe University of Agriculture and Natural Resources (LUANAR), Bunda Campus, using standard analytical procedures decribed by **AOAC (2005)**.

Moisture content was determined by drying the sample at 105°C in a hot-air oven for 24 hours until a constant weight was obtained. Crude protein was analyzed using the Kjeldahl method, which involved acid digestion with concentrated sulfuric acid (H_2SO_4) and a copper catalyst, followed by distillation and titration. Total nitrogen content was converted to crude protein using a factor of 6.25. Crude lipid was determined through Soxhlet extraction, using analytical-grade hexane as the solvent. Approximately 2g of each finely ground sample was refluxed for 6 hours in a pre-weighed extraction thimble. Ash content was determined by incinerating the sample in a muffle furnance at 550^{o}C for four hours.

Crude fiber was determined by sequential acid-alkali digestion, beginning with 1.25% sulfuric acid (H_2SO_4), followed by 1.25% sodium hydroxide (NaOH). The residue was filtered, oven-dried at 105^{o}C, and incinerated in a muffle furnace at 550^{o}C to quantify the indigestible fiber fraction, primarily composed of cellulose and lignin, following the procedure described by **Dougall (1956)**.

In addition to proximate analysis, baobab and tamarind pulp powders were subjected to micronutrient profiling to evaluate their suitability as natural substtutes for synthetic vitamin-mineral premixes. Vitamin C, phosphorus, and calcium were quantified and reported per 100g of dry weight. Vitamin C content was determined using the 2, 6-dichlorophenolindophenol titrimetric method. Phosphorus content was determined colorimetrically after acid digestion, following the molybdenum blue method, with absorbance measured at 880nm. Calcium was determined using atomic absorption spectrophotometry (AAS) at 422.7nm. The quantified levels of vitamin C, phosphorus, and calcium served as foundational data for evaluating the suitability of baobab and tamarind pulp powders as functional micronutrient sources in experimental diet formulation.

Feed formulation and diet composition

Five isonitrogenous (35% CP) experimental diets were formulated using the Pearson Square method, incorporating baobab pulp powder vitamin-mineral premix

(BPPVM) and tamarind pulp powder vitamin-mineral premix (TPPVM) at 5% and 7% inclusion levels, with a 3% synthetic vitamin-mineral premix (SVMP) serving as the control, as shown in Table (1). Micronutrients are essential in aquafeeds, typically required at inclusion levels between 3-5% to support optimal physiological functions **(Hoque *et al.*, 2023)**.

Table 1. Ingredient composition of the experimental diets

	Replacement levels				
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	0.0	7.00
Starch	1.00	1.00	1.00	1.0	1.0
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.

Feed processing and pelleting

All feed ingredients were ground using a dry grinder (Model ASEFAC, ASEFAC Technologies, Malawi) and weighed according to pre-calculated dietary proportions using a precision electronic balance (Model OHAUS-LS 200, OHAUS Corporation, USA). Ground ingredients were manually mixed for 15 minutes in a sanitized stainless steel tray to ensure initial uniformity. Pre-gelatinized cassava starch was incorporated at 1% (w/w) as a binder. The mixed feed dough was conditioned with warm water to reach approximately 30% moisture content and pelleted using a mechanical pelleting machine (Model INTRA PRO 600, 2.5mm die size). Pellets were spread in a single layer and air-dried at ambient temperature (25-28°C) under a screened shelter for 48 hours until a constant weight was achieved. Dried pellets were subsequently ground to crumbles using a clean mortar and pestle to match the mouth size of fingerlings. After cooling to ambient temperature, all the feed samples were stored in airtight, clearly labeled containers and kept at room temperature (approximately 25ºC) in a dry, well ventilated feed storage cabinet to prevent nutrient degradation and contamination.

To maintain experimental integrity, all batches were prepared following identical protocols to ensure consistency across dietary treatments.

Experimental design and feeding trial

The feeding experiment was conducted from August 14, 2024 to November 30, 2024 utilizing a completely randomized design (CRD) with five dietary treatments. Experimental diets included a control group with 3% synthetic vitamin-mineral premix, and four treatment groups comprising 5% baobab pulp powder vitamin-mineral, 5% tamarind pulp powder vitamin-mineral, 7% baobab pulp powder vitamin-mineral and 7% tamarind pulp powder vitamin-mineral. Three experimental replicates, each consisting of 20 fish with an initial weight ranging from 5.52–5.54g totaling 300 fingerlings, were randomly assigned to fifteen experimental units. Fish were hand-fed twice daily at 09:00 and 16:00 hours (local time) at a feeding rate of 3% of their wet body weight, divided equally between the two feeding sessions **(Almazán-Rueda *et al.*, 2004)**. The feeding rate was adjusted fortnightly based on fish growth. Uneaten feed and fecal waste were siphoned out daily, and 50% of the water in each aquarium was replaced. Complete cleaning of the aquaria and refilling with fresh water were conducted bi-daily to ensure optimal rearing conditions.

Growth performance, survival assessment, and feed utilization parameters

At the beginning of the experiment, the initial body weight of *Clarias gariepinus* fingerlings in each treatment group were recorded using a digital precision balance (Model HL-200i, A&D weighing, Japan) with an accuracy of 0.1g. This served as the baseline for evaluating growth performance over the trial period. Fingerlings were weighed weekly throughout the feeding trial to monitor growth response to the dietary treatments. Final body weights were measured at the end of the 12-week period under the same conditions.

Survival was determined by counting the number of fish remaining in each treatment group at the conclusion of the experiment. Survival rate (%) was calculated by determining the ratio of fish that survived to the total number of fish stocked at the commencement of the experiment expressed as percentage (survival%) according to the formula provided below. Growth performance and feed utilization parameters which include weight gain, specific growth rate, feed conversion ratio, and protein efficiency ratio were calculated according to standard procedures described by **Tacon (1995)** as outlined below:

Weight Gain (WG) (g) = Final weight - Initial weight

Specific Growth Rate (SGR) (% day^{-1}) = [(ln Final weight - ln Initial weight) / Duration (days)] × 100

Feed Conversion Ratio (FCR) = Feed intake (g) / Weight gain (g)

Protein Efficiency Ratio (PER) = Weight gain (g) / Protein intake (g)

Survival Rate (%): = (Final fish count / Initial fish count) × 100

Water quality monitoring

Water quality parameters were monitored daily to maintain optimal rearing conditions for *Clarias gariepinus* fingerlings. *In situ* measurements of water temperature and dissolved oxygen (DO) were taken at 09:00 and 16:00 hours using OxyGuard Handy Polaris meter (OxyGuard International A/S, Denmark). pH was measured using a YSI Ecosense pH10A pH meter (YSI inc., USA). Total dissolved solids (TDS) were determine gravimetrically using the evaporation method. Ammonia concentrations were assessed using an API Ammonia Test Kit (Mars Fishcare, USA), which provides a semi-quantitative estimate of total ammonia nitrogen (TAN). Water samples were collected from each tank, and ammonia levels were estimated by matching the color of test strips to a standardized color chart provided with the kit.

Blood collection and haematological analysis

Blood samples were collected following the procedure of **Blaxhall and Daisley (1973)** and **Wedemeyer and Yasutake (1977)**. Thereafter, the samples were taken to the Lilongwe University of Agriculture and Natural Resources Health Center (Bunda campus) for haematological analysis. Hematological parameters such as packed cell volume (PCV), haemoglobin (Hb), and red blood cell (RBC) mean cell volume (MCV), mean cell hemoglobin (MCH) mean cell hemoglobin concentration (MCHC), and white blood cell (WBC) were assessed.

The direct measurement of PCV, Hb, and RBC, along with the calculation of absolute erythrocyte indices, followed the method described by **Adebayo *et al.* (2007)** using the following formulas:

Mean cell volume (fL) = (PCV (%) × 10) / RBC (10^6/mm^3)

Mean cell haemoglobin (pg) = (Hb (g/dL) × 100) / RBC (10^6/mm^3)

Mean cell haemoglobin concentration (g/dL) = (Hb (g/dL) × 100) / PCV (%)

White blood cell and differential counts were determined according to the method described by **Penttilä *et al.* (1985).**

Statistical analysis

All statistical analyses were conducted in R (version 4.5.0; R Core Team, 2025). Initial data entry and preliminary screening for missing values and outliers were performed in microsoft excel, while all subsequent data cleaning, assumption testing, and statistical modeling were carried out in R to ensure reproducibility and transparency. A one-way analysis of variance (ANOVA) was used to assess the effects of dietary treatments on growth performance, hematological indices, and water quality parameters. Diets was treated as a fixed factor, and all response variables were continuous. Model diagnostics and analysis of variance (ANOVA) computations for the completely randomized design (CRD) were performed using the Descriptive Inference for CRD (DIC) function from the AgroR package **(Shimizu, *et al.*, 2024)**. The function generates a diagnostic summary that includes the Shapiro-Wilk test for normality, Bartlett's test for homogeneity of variance, and the Durbin-Watson statistic for assessing residual independence. When the overall F-test indicated a significant treatment effect ($P< 0.05$), Tukey's Honest Significant Difference (HSD) test was automatically applied for pairwise comparisons among treatment means.

RESULTS

Growth performance, nutrient utilization, and survival of *Clarias gariepinus* Fingerlings

The growth performance, nutrient utilization, and survival of *Clarias gariepinus* fingerlings fed diets supplemented with synthetic vitamin-mineral premix (SVMP), baobab pulp powder vitamin-mineral premix (BPPVM), and tamarind pulp powder vitamin-mineral premix (TPPVM) are summarized in Table (2). The weight gain pattern over the 12-week feeding trial showed a steady increase across all dietary groups, as indicated from the data presented in Fig. (2). Fish fed SVMP (3%), BPPVM (5%), and BPPVM (7%) exhibited significantly higher weight gain ($P< 0.05$) compared to those fed on TPPVM (5% and 7%). However, there was no significant difference ($P> 0.05$) among BPPVM (5%), BPPVM (7%), and SVMP (3%), indicating comparable growth performance in these groups. The specific growth rate (SGR) followed a similar trend, with fish fed SVMP (3%), BPPVM (5%), and BPPVM (7%) achieving higher SGR values, while fish fed on TPPVM-based diets had significantly lower SGR ($P< 0.05$). The feed conversion ratio (FCR) varied significantly among dietary treatments ($P< 0.001$). Fish fed SVMP (3%), BPPVM (5%), and BPPVM (7%) had lower FCR values, indicating better feed utilization efficiency, whereas fish fed on TPPVM-based diets had higher FCR values. The protein efficiency ratio (PER) also differed significantly ($P= 0.001$). Fish fed BPPVM (5%) and BPPVM (7%) had PER values comparable to those of SVMP-fed fish, while those fed on TPPVM (5% and 7%) recorded lower PER values. The survival rate remained high across all dietary groups, with SVMP (3%), BPPVM (5%), and BPPVM (7%) achieving 100% survival. Fish fed TPPVM (5%) and TPPVM (7%) had slightly lower survival rates, though the difference was not statistically significant.

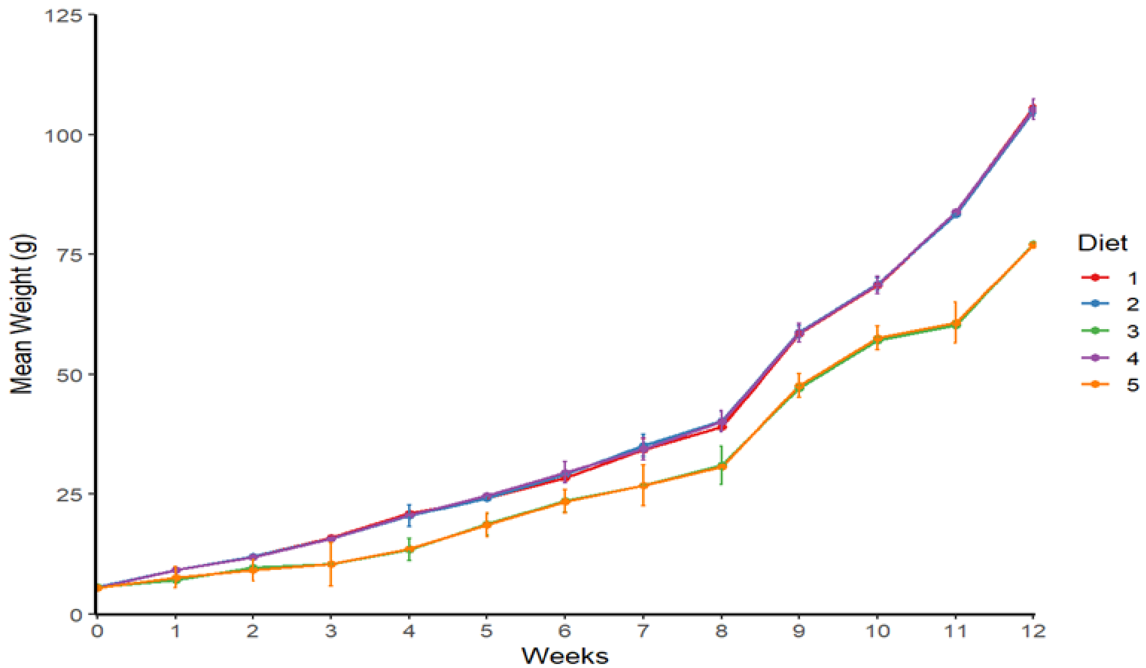

Fig. 2. Growth curve of *Clarias gariepinus* fingerlings fed different experimental diets. Mean body weights(g) are presented at each point, with error bars indicating standard error (SE)

Table 2. Growth performance and feed utilization of *Clarias gariepinus* fingerlings fed different experimental diets

	Experimental diets					
Parameter	**SVMP (3%)**	**BPPVM (5%)**	**TPPVM (5%)**	**BPPVM (7%)**	**TPPVM (7%)**	***P*-values**
Initial weight (g)	5.54±0.01	5.53±0.06	5.54±0.02	5.52±0.12	5.53±0.11	0.275
Final weight(g)	105.60±0.09[a]	104.86±0.13[a]	77.14±0.04[b]	105.24±0.21[a]	77.05±0.42[b]	< 0.001
Weight gain (g)	100.06±0.08[a]	99.33±0.27[a]	71.60±0.02[b]	99.72±0.09[a]	71.52±0.11[b]	< 0.001
SGR(%/day)	3.27±0.03[a]	3.27±0.22[a]	2.93±0.2[b]	3.27±1.08[a]	2.93±0.21[b]	< 0.001
FCR	1.09±0.21[a]	1.12±0.14[a]	3.81±0.33[b]	1.17±0.53[a]	3.24±0.31[b]	< 0.001
PER	1.90±0.82[a]	1.90±0.14[a]	1.63±1.21[b]	1.90±0.41[a]	1.68±0.21[b]	< 0.001
Survival(%)	100±0.23	100±0.04	98.33±0.12	100±0.16	96.67±0.11	0.552

Values are presented as means ± standard error. Within the row, means with the same letter are not significantly different at 5% level of significance. = Synthetic vitamin mineral premix (3%), BPPVM (5% and 7%) = Baobab pulp powder vitamin mineral, TPPVM (5%and 7%) = Tamarind pulp powder vitamin mineral, SGR = Specific growth rate, PER =Protein efficiency ratio.

Hematological responses of *Clarias gariepinus* to experimental diets

The hematological response of *Clarias gariepinus* fingerlings to dietary treatments is summarized in Table (3). Red blood cell (RBC) counts were significantly higher ($P < 0.05$) in fish fed SVMP (3%), BPPVM (5%), and BPPVM (7%) compared to those fed on TPPVM-based diets. Similarly, fish in these groups exhibited higher white blood cell (WBC) counts than those fed TPPVM (5%) and TPPVM (7%).

Hemoglobin (Hb) concentration and packed cell volume (PCV) varied significantly among dietary groups ($P < 0.05$). The highest Hb and PCV values were recorded in fish fed SVMP (3%), BPPVM (5%), and BPPVM (7%), while fish fed on TPPVM-based diets exhibited significantly lower values.

Mean corpuscular volume (MCV) was significantly higher ($P < 0.05$) in fish fed TPPVM (5%) and TPPVM (7%), whereas those fed on SVMP (3%) and BPPVM-based diets had lower MCV values. A similar trend was observed for mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC), with SVMP (3%) and BPPVM-fed fish exhibiting higher values than those fed on TPPVM-based diets.

Table 3. Haematological parameters of African catfish (*Clarias gariepinus*) fed different diets

	Experimental diets					
Parameter	**SVMP (3%)**	**BPPVM (5%)**	**TPPVM (5%)**	**BPPVM (7%)**	**TPPVM (7%)**	**p-values**
RBC (Cells$\times10^{12}$)	2.72 ± 0.39^{b}	2.86 ± 0.21^{a}	2.38 ± 0.32^{c}	2.80 ± 0.20^{a}	2.40 ± 0.10^{c}	< 0.001
WBC	23.21 ± 0.52^{a}	22.50 ± 0.21^{b}	22.62 ± 0.17^{b}	23.05 ± 0.13^{a}	22.83 ± 0.43^{b}	0.002
Hb(gldl)	9.28 ± 0.17^{b}	9.83 ± 0.20^{a}	8.07 ± 0.48^{c}	9.62 ± 0.31^{a}	8.73 ± 0.38^{c}	< 0.001
PVC(%)	24.90 ± 0.89^{a}	25.22 ± 0.29^{a}	22.20 ± 0.05^{b}	25.75 ± 0.78^{a}	22.80 ± 0.07^{b}	< 0.001
MCV(fl)	91.54 ± 0.33^{b}	88.18 ± 0.26^{b}	93.27 ± 0.28^{c}	91.96 ± 0.44^{b}	95.00 ± 0.46^{a}	< 0.001
MCH(pg)	34.11 ± 0.25^{a}	34.37 ± 0.33^{a}	33.90 ± 0.50^{b}	34.35 ± 0.23^{a}	36.38 ± 0.55^{b}	< 0.001
MCHC(%)	37.26 ± 0.18^{b}	39.00 ± 0.50^{a}	36.35 ± 0.20^{c}	37.35 ± 0.49^{b}	36.71 ± 0.33^{c}	< 0.001

Means within each row having different superscripts are significantly different (P>0.05).

Effect of water quality parameters in response to dietary treatments

Water quality parameters such as temperature, pH, dissolved oxygen (DO), total dissolved solids (TDS), and ammonia concentration, varied among dietary treatments but remained within the optimal range for the culture of *Clarias gariepinus*, as shown in Table (4). Water temperature followed a typical diurnal pattern across all treatments, with no significant differences observed.

pH was at its highest value in TPPVM-based diets, indicating increased alkalinity, whereas SVMP and BPPVM-fed fish maintained stable pH levels. DO levels were the lowest in TPPVM-fed fish, suggesting increased microbial oxygen demand, whereas SVMP and BPPVM diets supported more stable oxygen levels. TDS levels were higher in fish fed TPPVM-based diets, suggesting greater leaching of organic matter, while SVMP and BPPVM-fed fish exhibited lower concentrations. Ammonia levels were at their highest in TPPVM-fed fish, indicating greater nitrogenous waste accumulation, whereas SVMP and BPPVM-fed fish maintained lower ammonia concentrations, suggesting better water quality stability.

Table 4. Water quality parameters measured in the experimental tanks for the culture of *C. gariepinus* fry on different diets

		Experimental Diets					
Parameter	**Time**	**SVMP (3%)**	**BPPVM (5%)**	**TPPVM (5%)**	**BPPVM (7%)**	**TPPVM (7%)**	**p-values**
Temp (0˚C)	07:00	25.69±0.13	25.45±0.14	25.71±0.12	25.42±0.14	25.33±0.12	0.15
	16:00	28.84±0.05	28.77±0.05	28.01±0.05	28.55±0.04	28.01±0.06	0.130
Ph	07:00	6.85-7.30	7.03-7.40	7.65-7.92	7.10-7.31	7.73-8.05	-
	16:00	7.10-7.25	7.21-7.42	7.72-7.85	7.28-7.32	7.69-7.82	-
DO (mg/L)	07:00	4.12±0.03[a]	3.84±0.02[a]	3.03±0.25[b]	3.64±0.12[a]	2.14±0.81[c]	< 0.001
	16:00	5.14±0.05[a]	4.93±0.07[a]	2.89±0.03[b]	4.85±0.06[a]	2.84±0.08[c]	< 0.001
TDS (mg/L)	07:00	527.21±0.43[a]	516.04±0.01[a]	1013.05±0.26[b]	532.78±0.07[a]	1246.52±0.04[c]	< 0.001
Ammonia (mg/L)	07:00	0.07±0.10[a]	0.07±0.15[a]	0.31±0.01[b]	0.07±0.08[a]	0.32±0.01[b]	< 0.001

Values are expressed as Mean ± Standard Error (SE) based on triplicate measurements (n = 3). Parameters include temperature (Temp), pH, dissolved oxygen (DO), total dissolved solids (TDS), and ammonia (NH_3). A dash (-) denotes parameters not anlysed , especially those presented only as ranges, Diets: SVMP = synthetic vitamin-mineral premix; BPPVM = baobab pulp powder vitamin-mineral premix; TPPVM = tamarind pulp powder vitamin-mineral premix.

DISCUSSION

Growth performance and feed utilization in *Clarias gariepinus*

Growth performance differed significantly (*P*<0.05) across dietary treatments, with fish fed SVMP (3%) and BPPVM (5% and 7%) exhibiting higher weight gain, specific growth rate (SGR), and improved feed conversion efficiency (FCR), suggesting more efficient nutrient utilization. The enhanced growth in BPPVM-fed fish is attributed to improved nutrient utilization and metabolic efficiency, supported by higher antioxidant activity and micronutrient availability. Baobab pulp is a rich source of ascorbic acid, which facilitates iron metabolism, collagen synthesis, and protein turnover, contributing to enhanced metabolic function and nutrient retention. These findings are consistent with the reports of **Bayon *et al.* (2019)** and **Hamed *et al.* (2019)**, who demonstrated that plant-derived antioxidants enhance nutrient absorption and promote growth efficiency in fish.

In contrast, fish fed TPPVM (5% and 7%) exhibited significantly lower weight gain and higher FCR values, reflecting reduced nutrient utilization efficiency. This trend aligns with the inhibitory effects of tannins and phytates on protein digestion and mineral bioavailability, as reported by **Adeniyi *et al.* (2022)**. Tannins have been shown to form complexes with dietary proteins, limiting enzymatic hydrolysis, while phytates chelate essential minerals such as calcium, zinc, and iron, impairing their bioavailability. These interactions have been reported to contribute to impaired protein utilization and metabolic inefficiency, ultimately affecting growth performance. A comparable trend was observed by **Khumujam *et al*. (2024)**, who demonstrated that high tannin levels in fish diets negatively affected feed palatability, protein metabolism, and overall physiological stability.

Survival rates were highest in SVMP and BPPVM-fed fish, suggesting greater physiological resilience in these groups. The presence of bioactive compounds and antioxidants in baobab pulp has been reported to support cellular homeostasis by reducing oxidative stress and improving metabolic stability **(Oyetunji *et al.*, 2021).** However, since immune markers were not directly assessed in this study, further research is needed to confirm the immunomodulatory effects of BPPVM. In contrast, the slightly lower survival rates observed in TPPVM-fed fish are consistent with findings by **Chabite *et al.* (2015)**, who reported that anti-nutritional factors in plant-based diets interfere with digestion and metabolic regulation, leading to physiological stress.

Feed utilization efficiency, as indicated by FCR values, varied significantly among dietary groups. The lower FCR values in SVMP and BPPVM-fed fish indicate improved feed-to-biomass conversion efficiency, attributed to higher protein digestibility and enhanced metabolic processing of nutrients. The presence of ascorbic acid and polyphenols in baobab pulp has been reported to enhance nutrient absorption and enzymatic function, contributing to improved feed efficiency. These findings are consistent with the results of **Guluwa *et al.* (2017)**, who observed improved carcass quality and nutrient retention in poultry fed baobab-based diets, highlighting its potential as a functional feed additive across species.

Higher feed conversion ratio (FCR) values observed in fish fed TPPVM-based diets suggest reduced feed digestibility. This observation aligns with the findings of **Kanfon *et al.* (2023)**, who reported that tannin-rich diets inhibit protein hydrolysis and mineral bioavailability, leading to lower nutrient utilization and metabolic inefficiency. These results suggest that tamarind pulp at the tested inclusion levels is not a suitable substitute for a synthetic vitamin-mineral premixes (SVMPs) unless its anti-nutritional factors are reduced through appropriate processing.

Hematological indices

Hematological parameters serve as key indicators of fish health, providing insights into oxygen transport efficiency, metabolic function, and immune responses. Variations in red blood cell (RBC) count, white blood cell (WBC) count, hemoglobin (Hb), packed cell volume (PCV), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC) reflect the physiological impact of dietary formulations.

Fish fed synthetic vitamin-mineral premix (SVMP) and baobab pulp powder vitamin-mineral premix (BPPVM) at 5% and 7% inclusion levels exhibited significantly higher RBC, Hb, and PCV values ($P< 0.05$), indicating improved oxygen transport capacity. This finding is consistent with hose of **Bayon *et al.* (2016)** and **Sönmez *et al.* (2022)**, who reported that plant-derived antioxidants support erythrocyte integrity and enhance hematological parameters in fish. The higher hematological values observed in BPPVM-fed fish are attributed to the micronutrient content of baobab pulp, particularly vitamin C, which is known to facilitate iron metabolism and antioxidant defense.

The increase in WBC counts in these groups aligns with findings by **Minářová *et al.* (2021)**, who demonstrated that plant-based antioxidants enhance leukocyte activity and immune resilience in fish. However, since no immune assays were conducted in this study, additional research is required to confirm these immunomodulatory effects. In contrast, fish fed tamarind pulp powder vitamin-mineral premix (TPPVM) at 5% and 7% exhibited significantly lower RBC, Hb, and PCV values ($P < 0.05$), reflecting a decline in oxygen-carrying capacity. The presence of anti-nutritional factors (ANFs), particularly tannins and phytates, in tamarind pulp has been reported to interfere with nutrient absorption, contributing to reduced hematological values. This observation is consistent with that of **Silva *et al.* (2020)**, who found that tannin-rich diets impair iron bioavailability, leading to hematological suppression. Similarly, **Lima *et al.* (2021)** reported that high tannin levels negatively impact erythrocyte function, reducing overall hematological parameters in fish.

MCV, MCH, and MCHC values followed similar trends across dietary treatments. Higher MCV values in TPPVM-fed fish suggest erythrocyte enlargement, a condition associated with altered erythrocyte morphology. However, since no hematological index analysis was conducted to assess erythrocyte structure, further studies are necessary to confirm this observation. This trend concurs with **Neamat-Allah *et al.* (2021)**, who reported that diets containing ANFs altered erythrocyte characteristics in fish. In contrast, BPPVM-fed fish exhibited stable MCV, MCH, and MCHC values, indicating better erythrocyte stability and hemoglobin synthesis.

The relationship between hematological indices and growth performance was evident, as higher RBC, Hb, and PCV values correlated with improved weight gain, specific growth rate (SGR), and feed conversion efficiency (FCR). The higher hematological values observed in BPPVM-fed fish are congruent with **Abdel-Aziz *et al.* (2024)**, who reported that optimal hematological function enhances metabolic efficiency and growth in fish. The lower hematological values observed in TPPVM-fed fish were associated with reduced oxygen transport and limited nutrient utilization, which likely contributed to impaired growth performance. This observation aligns with **Odioko and Daniel (2016)**, who linked declining hematological parameters to compromised nutrient metabolism and poor feed efficiency.

The improved hematological profile in BPPVM-fed fish has been attributed to its high vitamin C content, which enhances erythrocyte stability and iron metabolism **(Sola-Ojo *et al.*, 2016)**. In contrast, the reduced RBC and Hb levels in TPPVM-fed fish indicate potential micronutrient limitations. **Barrow *et al*. (2008)** found that tannin-induced iron chelation significantly reduces iron absorption, leading to lower hemoglobin synthesis. **Isiyaku *et al*. (2023)** reported that low dissolved oxygen levels intensifies the physiological impact of anti-nutritional factors (ANFs), particularly by compromising hematological function in fish. Tamarind pulp powder vitamin mineral fed groups exhibited higher pH and lower dissolved oxgen levels which may have contributed to the observed reductions in hematological indices.

Water quality parameters

Water quality plays a critical role in fish health and growth, influencing metabolic function, physiological stability, and overall performance. **Boyd and Tucker (2014)** emphasized that maintaining optimal water quality parameters is essential for sustaining fish growth and survival in aquaculture systems. In this study, diurnal temperature fluctuations (25–30°C) remained within the optimal range for *Clarias gariepinus*, consistent with natural variations reported by **Ochang *et al.* (2007)**. Since temperature did not exceed acceptable limits, it was unlikely to have significantly impaired fish performance.

The high pH levels observed in Diet 5 (TPPVM 7%) was likely due to microbial decomposition of organic matter, leading to alkalinity buildup. **Boyd (2014)** reported that pH values above 9.0 can induce osmoregulatory stress, impair gill function, and negatively affect feed utilization. In this study, high pH in Diet 5 may have compromised metabolic efficiency by disrupting ion exchange and acid-base balance, which are essential for homeostasis in fish. This could have contributed to lower growth rates and feed efficiency in fish fed tamarind-based diets.

Dissolved oxygen (DO) levels were significantly lower in tamarind-based diets, especially in Diet 5, likely due to increased microbial oxygen demand and organic matter accumulation. **Mariu *et al.* (2023)** reported that diets rich in plant tannins and fiber promote microbial activity, increasing biochemical oxygen demand (BOD) and reducing DO availability. This oxygen depletion likely impaired aerobic metabolism and reduced energy availability, potentially contributing to suboptimal fish growth in TPPVM-fed groups. In contrast, fish fed baobab-based diets maintained relatively higher DO levels, suggesting a lower organic load and better oxygen utilization, which may have promoted more efficient metabolic function and improved growth performance.

Total dissolved solids (TDS) concentrations were significantly higher in tamarind-based diets, likely due to increased leaching of organic compounds, including tannins and soluble carbohydrates. High TDS levels have been associated with osmoregulatory dysfunction, leading to physiological stress in fish **(Boyd, 2014)**. In this study, high TDS may have disrupted ion balance and interfered with nutrient absorption, which could explain the reduced growth performance observed in fish fed TPPVM diets. These findings align with **Portz *et al.* (2006)**, who reported that high TDS levels negatively affect osmoregulatory function, leading to poor feed conversion efficiency.

The high ammonia levels recorded in Diets 3 and 5 indicated incomplete protein metabolism and nitrogenous waste accumulation, which likely contributed to water quality deterioration and increased physiological stress. **Islam (2005)** reported that ammonia accumulation impairs gill function and oxygen intake, which can reduce feed efficiency. In this study, the combination of high ammonia and low DO levels likely aggravated metabolic inefficiencies, further compromising fish health and reducing growth performance. This observation aligns with **Boyd and Tucker (2014)**, who reported that ammonia buildup is a major contributor to water quality degradation, often leading to stress-induced health issues and lower feed efficiency in fish.

CONCLUSION

This study demonstrated that baobab pulp powder vitamin-mineral premix (BPPVM) at 5–7% inclusion supported comparable growth performance and physiological stability to synthetic premixes in *Clarias gariepinus* fingerlings. In contrast, fish fed tamarind pulp powder vitamin-mineral premix (TPPVM) exhibited lower growth and hematological indices, suggesting potential challenges in nutrient utilization. These findings suggest that BPPVM could serve as a sustainable alternative to synthetic premixes in aquafeeds, while further research is needed to optimize the nutritional potential of TPPVM.

Declaration of competing interest

There is no conflict of interest to declare between the authors of this manuscript.

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