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Effect of Dietary *Parquetina nigrescens* Leaf Extract on Growth Performance, Activities of Digestive Enzymes, Serum Biochemical Profile and Antioxidative Status of African Catfish (*Clarias gariepinus*)

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Abstract

The current study sought to determine how dietary *Parquetina nigrescens* leaf (PNL) affected the performance of *Clarias gariepinus*. Ethanol extract of PNL was included at 0.0 (control), 0.5, 1.0, and 1.5 g/kg diet for *C. gariepinus* juveniles. The fish (n = 240) were fed the assigned diets for eight weeks to apparent satiation, twice daily in three replicates to determine the growth performance, nutrient utilization and activities of intestinal digestive enzymes. Thereafter, samples of blood were withdrawn from the fish to determine the serum biochemical and anti-oxidative parameters. One-way analysis of variance was used analyze the data generated at $P < 0.05$. In comparison to the fish fed others diets, fish fed 1.0-1.5 g PNL showed considerable increase in weights, specific growth rate, protein efficiency ratio and reduced feed conversion ratio. Higher fish survival and fish productivity index were obtained at 1.0 g PNL. The fish fed 1.0-1.5 g PNL had higher activities of maltase, lipase, chymotrypsin, and pepsin. Additionally, blood proteins were enhanced, while liver enzymes reduced with dietary PNL. The activities of serum catalase, glutathione peroxidase, glutathione-s-transferase, superoxide dismutase were also boosted in the fish on 1.0-1.5 g PNL. Ultimately, the growth performance of *C. gariepinus*, as well as its serum biochemical and antioxidative status were enhanced with dietary PNL at 1.0-1.5 g. Hence, PNL inclusion at 1.0 g/kg diet is recommended, as organic supplement, for healthy production performance of the fish.

Keywords: *Clarias gariepinus, Parquetina nigrescens, growth performance, serum biochemical, antioxidant enzymes*

Introduction

Production of aquatic animals is increasing due to their high nutritional value and contribution to healthy diet, food security and economic development. Globally, inland aquaculture contributes about 84% to global inland fish production (FAO, 2024). African catfish, *Clarias gariepinus*, is one of the highly reared and priced inland food fishes in Africa and other continents. The fish is intensively, widely and mostly cultured in Nigeria because of its attributes, including high consumers' acceptance, market value, fast growth rate, as well as its high adaptability adverse rearing, water quality and environment conditions (Adeniyi et al., 2025). The use of natural additives with growth and health-promoting effects are nonnegotiable and fundamental to economic viability, sustainable production and food security without compromising quality diets to meeting the requirements of the fish for essential nutrients and safety of the consumers. One category of such natural additives is herbs.

Medicinal herbs are recognized for possession of diverse bioactive compounds, which are the bases for their wide usage. Additionally, it is opined that herbs as natural additives are ecofriendly, renewable, sustainable and relatively safe for animal use. Investigations on the usage of herbs in the culture of aquatic animals are rising owing to its diverse roles. The roles of herbal additives in aquafeeds, including promotion of growth, antioxidant capacity, fish immunity and resistance against disease-causing organisms have been attested in many recent studies (Garg et al., 2021; Promprom et al., 2024; Adeniyi et al., 2023a, 2025; Paray et al., 2025).

Parquetina nigrescens is a woody perennial shrub with smooth twining stem and wide leaves, in the family Apocynaceae and subfamily Periplocoideae. It is rich in flavonoids (Ayoola et al., 2019; Kayode et al 2022), while many compounds such as octadecatrienoate, hexadecanoic acid, octadecanoate, squalene, mannos, iophene, phytol, decanoic acid, 5-ethyl-2-furaldehyde have been identified in reasonable quantities from the leaves (Adeyomoye and Adeoye, 2018; Adase et al., 2022). The plant possess several biological properties, including; antibacterial, antioxidant, antidiabetic, anti-helminthic, anti-inflammatory, anti-ulcerative, antipyretic, hepatoprotective and immmuno-stimulating properties (Saba et al., 2010; Banwo et al., 2020; Ojuade et al; 2021; Adeniyi et al., 2023a; Akinduko et al., 2024). The rich phytochemical compounds and diverse biological properties of *P. nigrescens* leaf could have led to its wide and effective utilization for ethnomedicinal purposes (Daramola, *et al*, 2022; Kayode *et al*., 2022; Olumide *et al*., 2022). Despite the potentials of this plant, there is limited information on its usage for farmed aquatic animals. Therefore, this study purposed to evaluate how dietary *P. nigrescens* leaf extract would affect the growth performance, digestive enzymes, serum biochemical indices and antioxidative capacity of *Clarias gariepinus*.

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Materials and Methods

Source and preparation Parquetina nigrescens leaf extract

Fresh *P. nigrescens* leaves were obtained, cleaned with tap water, allowed to air-dry at ambient temperature and ground into meal using an electric blender. Ethanol was used as the solvent in a ratio of 1:5 (Plant: ethanol, weight/volume) to extract the herbal meal. To ensure sufficient homogenization and extraction, the *P. nigrescens* leaf meal was dissolve in ethanol using a glass rod and shaken on electrical orbital shaker (DLAB, SK033pro, UK) at 250 rpm intermittently within the 48 hours of extraction. After that, muslin cloth and filter paper were used to sieve and filter the mixture, respectively. The solvent was removed from the filtrate, using rotary evaporator, to produce concentrated *P. nigrescens* leaf extract (PNL), which was preserved in refrigerator (-4 °C) until when it was included in the diet (Adeniyi et al., 2023a).

Experimental fish and feeding

Juveniles of *Clarias gariepinus* were acquired from a reliable farm and given a commercial feed (Bluecrown, Nigeria; 2 mm size; 42% crude protein, 12% lipid) for 2 weeks during which the fishes adjusted to the rearing conditions. The juveniles (240; 17.74 ± 0.23) were then divided at random into four treatments of three replicates. The fish were reared in twelve experimental plastic aquaria of 80 L-capacity, using a renewal method of 72-hour water replenishment. The ingredients listed on Table 1 were used to produce four 2 mm diets, with PNL {0.0 (control), 0.5, 1.0 and 1.5 g / kg diet} added by spraying. The pellets of each diet were immediately air-dried and put in labeled polythene bags. During the eight-week feeding trial, the designated diets were offered to the fish daily, at 08:30-9:30 am and 4:30 – 5:30 pm, till they appeared satiated.

Growth performance

The fish were batch-weighed prior and after the feeding trial to estimate the initial and final weights. The sum of the weights of feed offered and consumed by the fish in each aquarium was taken as feed intake. The fish weight gain (WG), specific growth rate (SGR), feed conversion ratio (FCR), protein efficiency ratio (PER), fish survival (FS) and fish productivity index(FPI) were calculated from the data obtained as shown below:

i. WG (g) = Final weight (FW) – Initial weight (IW)
ii. SGR (%/day) =100 x (Ln FW – Ln IW) / Time of feeding trial (days)
iii. FCR = Feed intake (FI) / WG
iv. PER = WG / Protein intake

(Protein intake = Crude protein in diet × FI)

v. FS (%) = 100 (Number of survived fish / number of fish stocked)
vi. FPI = (Weight gain × Fish survival) / (FCR × 10)

Analysis of digestive enzymes

To avoid contamination with endogenous enzymes, feeding was stopped 24 hours before the removing the guts. Thereafter, guts of two fishes were removed, from each replicate and labeled appropriately in a sample bottle. The guts were cleansed with ice-cold and then homogenized with ice-cold deionized water (1:10); centrifugation of the homogenate was performed at 5,000 *g* for 15 min; and the supernatants of the samples were used as enzyme extracts for the digestive enzymes assay on the activities of lipase, chymotrypsin, pepsin, and maltase, following the standard procedures described by Ogunbiyi and Okon (1976) and Fagbenro *et al*. (2005).

For lipase, mixture of 1.0 mL each of enzyme extract and 25% olive oil emulsion (at pH 7.0) were simultaneously incubated with the control (blank) in a water bath at 37 °C for an hour. After incubation, 3.0 mL of 95% ethanol and two drops of phenolphthalein were added to produce a reaction mixture that was titrated against 0.05N sodium hydroxide. The difference in titre values of control and extract was recorded as lipase activity (Ogunbiyi and Okon, 1976). To estimate the activities of pepsin and chymotrypsin, mixture 0.5 mL of enzyme extract, 10 mg of hide powder azure as substrate, and 2.0 mL phosphate buffer (pH 8.0 for chymotrypsin and pH 2.0 for pepsin) were incubated at 37 °C for an hour along with control samples. Thereafter, 3.0 mL of ice-cold phosphate buffer was added, the mixture was filtered and the absorbance read at 595 nm (Fagbenro et al., 2005). Maltase activity was analyzed by heating mixture of 0.2 mL of the intestinal enzyme extract, 0.4 mL of 1 % substrate (0.112 µM maltose), 0.2 mL phosphate buffer (pH 7.0), and 1.6 mL of alkaline 3, 5-dinitrosalicyclic acid reagent (DNSA) for 30 minutes in a water bath at 100 °C. The mixture was made up to 4.0 mL by adding distilled water and absorbance was measured at 540 nm (Fagbenro et al., 2005).

Blood collection, biochemical and antioxidant analyses

Following the feeding trial, samples of blood were collected from the fish's caudal vein using a 2 mL plastic syringe in plain bottles, left at ambient temperature for 30 minutes allowed to allow it to clot and then transported to the laboratory on ice, where the serum were separated using centrifuge, properly labeled in eppendorf tubes and store in freezer for serum biochemical and antioxidant analyses. Blood proteins, cholesterol, triglycerides, total

bilirubin, creatinine, blood urea nitrogen (BUN), aspartate aminotransferase (AST), alanine aminotransferase (ALT), and alkaline phosphatase (ALP) were determined with Randox kit, following the standard procedure of the manufacturer. The concentration of malondialdehyde (MDA), hydrogen peroxide, reduced glutathione (GSH) and activities of glutathione peroxidase (GPx), glutathione-s-transferase (GST), catalase, and superoxide dismutase (SOD) were also evaluated using commercial assay kit (Cayman).

Table 1: Gross composition (g/kg diets) of experimental diets containing *Parquetina nigrescens* leaf (PNL) extract for *Clarias gariepinus*

Ingredients	*Levels of inclusion (g/kg)*			
	0.0	0.5	1.0	1.5
Fish meal	280	280	280	280
Soya bean meal	256	256	256	256
Groundnut cake	191	191	191	191
Maize flour	179.5	179.5	179.5	179.5
Fish oil	50	50	50	50
Di-calcium phosphate	6	6	6	6
Premix	20	20	20	20
Starch	15	14.5	14.0	13.5
PNL extract	0.0	0.5	1.0	1.5
Methionine	0.5	0.5	0.5	0.5
Table salt	2	2	2	2

*Premix (per kg premix): Vitamin A, 22,000IU; vitamin B1, 20mg; vitamin B2, 25mg; vitamin B6, 10mg; vitamin B12, 0.05mg; vitamin C, 300mg; vitamin D3, 5,000IU; vitamin E, 300mg; vitamin K3, 10mg; niacin, 120mg; folic acid, 5mg; biotin, 1mg; inositol, 50mg; calcium pantothenate, 60mg; choline chloride, 500mg; manganese, 30mg; iron, 35mg; zinc, 45mg; copper, 3mg; cobalt, 2mg; iodine, 5mg; selenium, 0.15mg

Statistical analysis

Using one-way analysis of variance, the data from the study were presented as mean ± standard error (SE). The differences among means of the treatments were analyzed with Duncan multiple range tests at $P < 0.05$, using statistical package for social sciences (SPSS), version 25.

Results

Fish growth performance and nutrient utilization

Table 2 presents the growth performance of *C. gariepinus* fed different levels of PNL extract and the control diet. In comparison to the control treatment, the final weight, weight gain, specific growth rate of the fish fed 1.0 - 1.5 g PNL extract/kg increased ($P<0.05$). The feed intake of the fish did not vary significantly among the treatments, but lower ($P<0.05$) feed conversion ratio and higher protein efficiency ratio were obtained in *C. gariepinus* fed 1.0 and 1.5 g PNL extract, in relation to the fish fed other diets. Fish survival (100%) was significantly higher at 1.0 g PNL, than the control treatment. On the overall, dietary PNL at 1.0 g PNL/kg diet enhanced fish productivity index significantly.

Activities of digestive enzymes

The effect of dietary PNL extract on activities of digestive enzymes in *C. gariepinus* is shown on Table 3. Higher ($P<0.05$) lipase activity was recorded in treatments fed PNL extract, reaching the peak at 1.0 g inclusion level. Also, the activities of pepsin, chymotrypsin and maltase in the fish rose as the levels of dietary PNL extract increased.

Table 2: Growth performance of *Clarias gariepinus* juveniles fed diets containing *Parquetina nigrescens* leaf extract for eight weeks

Parameters	*PNL inclusion level (g/kg diet)*			
	0.0	0.5	1.0	1.5
Initial weight (g/fish)	17.72 ± 0.14^{a}	17.76 ± 0.15^{a}	17.63 ± 0.07^{a}	17.87 ± 0.07^{a}
Final weight (g/fish)	120.28 ± 1.80^{c}	130.96 ± 0.65^{b}	135.38 ± 1.62^{a}	138.38 ± 0.70^{a}
Weight gain (g/fish)	102.57 ± 1.69^{c}	113.20 ± 0.61^{b}	117.76 ± 1.56^{a}	120.51 ± 0.54^{a}
SGR (%/day/fish)	3.42 ± 0.02^{c}	3.57 ± 0.02^{b}	3.64 ± 0.02^{a}	3.66 ± 0.01^{a}
Feed intake (g/fish)	140.22 ± 0.49^{bc}	143.70 ± 0.84^{a}	139.75 ± 0.33^{c}	142.35 ± 1.12^{ab}
Feed conversion ratio	1.37 ± 0.01^{a}	1.27 ± 0.02^{b}	1.19 ± 0.01^{c}	1.18 ± 0.02^{c}
Protein efficiency ratio	1.83 ± 0.03^{c}	1.97 ± 0.02^{b}	2.11 ± 0.02^{a}	2.12 ± 0.02^{a}
Fish survival (%)	95.00 ± 0.00^{b}	96.67 ± 1.67^{ab}	100.00 ± 0.00^{a}	96.67 ± 1.67^{ab}
Fish productivity index	713.10 ± 22.83^{c}	862.61 ± 26.78^{b}	992.65 ± 16.91^{a}	986.22 ± 27.08^{a}

Means ± SE along a row with different superscript differ significantly at $P < 0.05$. SGR - Specific growth rate

Table 3: Activities of digestive enzymes in *Clarias gariepinus* juveniles fed diets containing *Parquetina nigrescens* leaf extract (PNL) for eight weeks

Parameters	*PNL inclusion levels (g/kg diet)*			
	0.0	0.5	1.0	1.5
Lipase	38.58 ± 0.24^{c}	40.18 ± 0.5^{b}	44.53 ± 0.91^{a}	42.59 ± 0.84^{a}
Pepsin	0.05 ± 0.00^{c}	0.06 ± 0.00^{bc}	0.07 ± 0.00^{b}	0.09 ± 0.01^{a}
Chymotrypsin	0.63 ± 0.01^{c}	0.65 ± 0.00^{bc}	0.71 ± 0.00^{b}	0.88 ± 0.03^{a}
Maltase	0.33 ± 0.01^{d}	0.45 ± 0.01^{c}	0.51 ± 0.01^{b}	0.73 ± 0.03^{a}

Means ± SE along a row with different superscript differ significantly at $P < 0.05$.

Fish biochemical profile

Table 4 shows the result of serum biochemistry profile of *C. gariepinus* fed with PNL extract. Dietary PNL extract enhanced the blood proteins ($P<0.05$). Concentration of AST and ALP reduced in all the fish fed with PNL-supplemented diets, with the lowest values obtained at 1.0 g inclusion level. The treatment fed 1.0 g PNL had the lowest ($P<0.05$) ALT, compared to others. There were no significant variations ($P>0.05$) in the concentrations of BUN, creatinine, and total bilirubin. Lower ($P<0.05$) cholesterol and triglyceride were obtained at 1.0-1.5 g PNL and 1.5 g PNL, respectively, compared to other treatments. Similarly, dietary PNL also reduced glucose levels at 1.0-1.5 g PNL dosage.

Antioxidative status

The effect of PNL extracts on the anti-oxidative status of *C. gariepinus* infected with *A. hydrophila* is shown in Table 4. There was significant reduction in the concentration of serum MDA and hydrogen peroxide (H_2O_2) of *C. gariepinus* fed 1.0 g PNL / kg diet, compared to those fed the control diet. Significantly higher GSH (Reduced glutathione) was exhibited in fish on 1.0 g PNL extract, compared to those fed other diets. The activities of antioxidant enzymes _CAT, GPx, and GST_ were also boosted in the fish fed PNL-based diets, especially at 1.0-1.5 g inclusion levels, compared to the fish on control diet.

Table 4: Serum biochemical profile of *Clarias gariepinus* juveniles fed diets containing *Parquetina nigrescens* leaf extract for eight weeks

Parameters	*PNL inclusion levels (g/kg diet)*			
	0.0	0.5	1.0	1.5
Total Protein (g/dL)	4.88 ± 0.05^{b}	5.06 ± 0.04^{a}	5.11 ± 0.04^{a}	5.12 ± 0.04^{a}
Albumin (g/dL)	1.08 ± 0.02^{b}	1.04 ± 0.03^{a}	1.18 ± 0.03^{a}	1.11 ± 0.04^{a}
Globulin (g/dL)	3.80 ± 0.04^{b}	4.01 ± 0.03^{a}	3.93 ± 0.03^{a}	4.01 ± 0.03^{a}
AGR	0.28 ± 0.00^{ab}	0.26 ± 0.01^{b}	0.30 ± 0.01^{a}	0.28 ± 0.01^{ab}
AST (UI/L)	316.67 ± 1.92^{a}	307.00 ± 3.62^{b}	282.81 ± 2.69^{c}	301.56 ± 2.52^{b}
ALT (UI/L)	50.44 ± 0.87^{a}	51.44 ± 1.02^{a}	45.11 ± 0.89^{b}	49.78 ± 1.00^{a}
ALP (UI/L)	601.56 ± 1.39^{a}	562.67 ± 8.77^{b}	544.78 ± 1.85^{c}	564.67 ± 2.36^{b}
BUN (mg/dl)	6.41±0.11	6.43±0.08	6.21±0.07	6.44±0.03
Creatinine (mg/dl)	0.56±0.02	0.58±0.01	0.53±0.02	0.57±0.02
Cholesterol (mg/dL)	185.89 ± 1.33^{a}	188.00 ± 1.87^{a}	178.78 ± 1.21^{b}	171.00 ± 0.83^{ab}
Triglycerides (mg/dL)	60.11 ± 0.96^{a}	62.78 ± 0.36^{a}	63.11 ± 0.68^{a}	50.11 ± 0.70^{b}
Glucose (mg/dL)	321.33 ± 1.13^{a}	317.78 ± 2.37^{a}	298.00 ± 0.71^{c}	307.00 ± 1.49^{b}

Means ± SE along a row with different superscript differ significantly at $P < 0.05$. AGR – Albumin globulin ratio; AST - Aspartate aminotransferase; ALT - alanine aminotransferase; ALP - Alkaline phosphatase (ALP); BUN – Blood urea nitrogen

Table 5: Antioxidative status of *Clarias gariepinus* juveniles fed diets containing *Parquetina nigrescens* leaf extract for eight weeks

Parameters	*PNL inclusion levels (g/kg diet)*			
	0.0	0.5	1.0	1.5
MDA (µmol/mg protein)	3.04 ± 0.02^{a}	1.05 ± 0.01^{b}	1.02 ± 0.03^{b}	1.06 ± 0.01^{b}
H_2O_2 (µmol/mg protein)	7.39 ± 0.13^{a}	5.85 ± 0.06^{b}	4.60 ± 0.07^{c}	4.09 ± 0.05^{d}
GSH (U/mg protein)	516.28 ± 3.75^{d}	618.07 ± 1.92^{b}	626.37 ± 1.28^{a}	607.08 ± 1.34^{c}
GPX (U/mg protein)	12.36 ± 0.42^{b}	12.90 ± 0.58^{b}	16.09 ± 0.36^{a}	15.61 ± 0.25^{a}
GST (U/mg protein)	55.75 ± 0.34^{c}	57.91 ± 0.18^{c}	63.82 ± 1.43^{b}	85.31 ± 0.81^{a}
Catalase	2.94 ± 0.13^{d}	3.31 ± 0.06^{c}	3.60 ± 0.05^{b}	3.86 ± 0.06^{a}
SOD (U/mg protein)	3.92 ± 0.06^{b}	6.47 ± 0.04^{a}	6.41 ± 0.06^{a}	6.34 ± 0.04^{a}

Means ± SE along a row with different superscript differ significantly at $P < 0.05$. MDA - Malondialdehyde; H_2O_2 . Hydrogen peroxide; GSH - Reduced glutathione; SOD - Superoxide dismutase; GPx - glutathione peroxidase; GST - Gluthathione-s-transferase

Discussion

Dietary PNL has been shown to enhance feed efficiency, growth performance, survival, productivity index, and the activities of the digestive enzymes of African catfish in this study. Similarly, many phyto-additives have been reported to promote growth in diverse farm aquatic animals, including Chenopodium album (Amhamed et al., 2018), *Apium graveolens* (Sutthi et al., 2020), *Mitracarpus scaber* (Adeshina et al., 2021), moringa (Adeniyi et al., 2023b), paulownia (El-Refiae et al., 2024), ivy (Fadaei et al., 2025) leaf extracts. Digestives enzymes _amylases, proteases and lipases_ catalyze the breakdown of carbohydrates, protein, and lipids into absorbable nutrients that are utilize for growth, repairs of worn out cells and tissues and energy for diverse fish's metabolic activities. Hence, the growth-promoting effects of PNL diets in the current study could be linked with the enhanced digestion of feed by the digestive enzymes and nutrients' absorption. Similar observations on the enhanced activities of digestive enzymes in fishes on dietary herbs have been published in previous studies (Amhamed et al., 2018; Naiel et al., 2021; Fadaei et al., 2025; Li et al., 2026).

Blood biochemical parameters are vital indicators for the evaluation and screening of dietary supplements on the physiological, nutritional benefit, health and general welfare of farm fishes (Adeniyi et al., 2023b). The significant increase in the blood proteins and lower liver enzymes (AST, ALT, and ALP) suggest that PNL enhanced protein metabolism and promote healthy liver function. Liver enzymes are common biomarkers for assessing the impact of extrinsic factors and their effects on the animal and higher level of these enzymes could indicate pathological stress due to hepatic injuries detrimental to the hepatocytes (Tennant and Center, 2008). Similarly, the lower concentrations of cholesterol, triglycerides and glucose recorded at 1.0 or 1.5 g PNL could be related the roles PNL in improving fat and carbohydrate metabolism at these dosages. The current observations on the biochemical profiles of fish align with the recent similar studies (Adeniyi et al., 2023b; Fadaei et al., 2025; Li et al., 2026).

Plant-based natural antioxidants have been proven to be efficacious agents that mitigate peroxidation of lipids in feeds and other substrates, and scavenge free radicals. They provide effective defense against oxidative stress and damage by boosting endogenous non-enzymatic (GSH) and enzymatic antioxidants (catalase, GPx, GST, and SOD) for sustainable production of healthy aquatic species (Adeshina et al., 2021; Adeniyi et al., 2024, 2025; Hu et al., 2025). The significant lower concentration of serum MDA and H_2O_2 with corresponding increase in GSH, catalase, GPx, GST and SOD in this study could be associated with the antioxidant activity of dietary PNL mitigating against oxidative stress in the fish. Previous in vitro (Ayoola et al., 2011; Banwo et al., 2020) and recent in vivo studies in rat (Akinduko et al., 2024; Bamisaye et al., 2026) also demonstrated antioxidant activities of PNL and this might be attributed to its flavonoid content (Ayoola et al., 2019; Kayode et al 2022). In agreement with the present study, many herbal leaves have been reported as effective natural antioxidants in different aquatic species (Adeniyi et al., 2018; Maldonado-Garciaa et al., 2019; Naiel et al., 2021; Adeniyi et al., 2024,2025; Paray et al., 2025).

In conclusion, the study determined the effects of *P. nigrescens* leaf extract performance of *C. gariepinus*. The extract promoted the fish weight gain, specific growth rate, protein efficiency ratio, fish productivity index, activities of lipase, pepsin and chymotrypsin and maltase, reduced feed conversion ratio, as well as improved the serum biochemical and antioxidative status of the fish at 1.0-1.5 g. Hence, PNL inclusion at 1.0 g/kg diet is recommended for healthy production performance of the fish.

References

Adase, E., Ankutse, P., Kumadoh, D., Archer, M-A., Kyene, M. O., Yeboah, G.N., & Agyare, D. O. S. (2022). A Review of *Parquetina nigrescens* (Afzel.) Bullock, a plant for traditional medicine: Phytochemical and pharmacological properties. Evidence-Based Complementary and Alternative Medicine , 6076707, https://doi.org/10.1155/2022/6076707

Adeniyi, O. V., Magaji, I. M., Oladimeji, Z. A., Adekola, B. I., & Ibiyo, L.M.O. (2023a). Immune response and resistance to *Aeromonas hydrophila* infection in C*larias gariepinus* fed diets supplemented with P*arquetina nigrescens* leaf extract. Proceedings of the 38th Annual Conference of Fisheries Society of Nigeria (FISON), Dr. Kariye E. Lelei (Ed.), 348-352.

Adeniyi, O. V., Setufe, S. B., Ajao, H. A., & Shabako, G.M. (2023b). Influence of dietary *Moringa oleifera* leaf extract on growth performance, blood profile and immune status of African catfish (*Clarias gariepinus*). Badeggi Journal of Agricultural Research and Environment, 05(02), 36-50. https://doi.org/10.35849/BJARE202302/101/004

Adeniyi, O. V., Adedayo, M. R., & Afe, A. I. (2024). Effects of dietary *Jatropha gossypiifolia* leaves extract on growth performance, antioxidant capacity, immune response, and disease resistance of African sharptooth catfish (*Clarias gariepinus*) infected with *Aeromonas hydrophila*. Journal of Applied Aquaculture, 1–26. https://doi.org/10.1080/10454438.2024.2382431

Adeniyi, O. V., Pourmozaffar, S., Setufe, S. B., & Adeshina, I. (2025). Effects of dietary supplementation of *Euphorbia hirta* on the growth performance, serum biochemistry, antioxidative status, and immune response of *Clarias gariepinus*. Aquaculture Research, 5208865. Published by Wiley-Blackwell. Available online at https://doi.org/10.1155/are/5208865

Adeshina, I., Tiamiyu, L. O., Akpoilih, B. U., Jenyo-Oni, A., & Ajani, E. K. (2021). Dietary *Mitracarpus scaber* leaves extract improved growth, antioxidants, non-specific immunity, and resistance of Nile tilapia, *Oreochromis niloticus* to *Gyrodactylus malalai* infestation Aquaculture 535: 736377 https://doi.org/10.1016/j.aquaculture.2021.736377

Adeyomoye, O. I., & Adewoye, E. O. (2018). Preliminary assessments and renoprotective e4ects of methanol extract of *Parquetina nigrescens* (African *Parquetina*) in diabetic wistar rats, Asian Journal of Research in Medical and Pharmaceutical Sciences, 3(4), 1–10.

Akinduko, A. A., Salau, S. O., Akinmoladun, A. C., Akindahunsi, A. A., & Osemwegie, O. O. (2024). Assessment of the anxiolytic, antidepressant, and antioxidant potential of *Parquetina nigrescens* (Afzel) bullock in wistar rats. Journal of Ethnopharmacology, 322, 117597. https://doi.org/10.1016/j.jep.2023.117597

Amhamed, I. D., Mohamed, G. A., Almabrok, A. A., Altıef, T. A. S., & Bilen, S. (2018). Efficacy of dietary *Chenopodium album* extract on some health parameters, digestive enzymes and growth performance in juvenile *Cyprinus carpio*. Alinteri Journal of Agricultural Science, 33(2), 165-176. https://doi/10.28955/alinterizbd.412455

Ayoola, A. O., Akinloye, O., Oguntibeju, O. O., Oke, J. M., Odetola, A. A. (2011). Antioxidant activities of *Parquetina nigrescens*. African Journal of Biotechnology, 10(24), 4920-4925. https://doi.org/10.5897/AJB10.1622

Bamisaye, F. A., Orji, E. E., Ajiboye, B. O., & Ayoola, A. O. (2024). Antioxidant activities of aqueous leaf extract of *Parquetina nigrescens* in high-fat-diet-streptozotocin-induced diabetic male albino rats. Comparative Clinical Pathology, 33, 239–246. https://doi.org/10.1007/s00580-023-03545-3

Banwo, K., Alao, M. B., & Sanni, A. I. (2020). Antioxidant and antidiarrhoeal activities of methanolic extracts of stem bark of *Parkia biglobosa* and leaves of *Parquetina nigrescens*, Journal of Herbs, Spices, and Medicinal Plants, 26(1), 14–29. https://doi.org/10.1080/10496475.2019.1663770

Daramola, O. O., Oyeyemi, W. A., Akinola, A. O., & Raji, Y. (2022). Haematoprotective and hepatoprotective effects of methanolic leaf extract of *Parquitina nigrescens* on arsenic trioxide-induced toxicity in male rats. Nigeria Journal Physiological Science, 37, 235-246.

El-Refiae, N. M., Ayyat, M. S., Mahmoud, H. K., & Naiel, M. A. E. (2024). The effects of *Paulownia* leaf extract dietary administration on growth, redox status, immune responses, and modulate intestinal microbial content in Nile tilapia. Aquaculture International, 32, 1857–1877. https://doi.org/10.1007/s10499-023-01247-9

Fadaei, R., Noori, A., Akbarzadeh, A., Hoseinifar, S. H., & Paolucci, M. (2025). Effects of Ivy (*Hedera helix*) leaf extract on growth, digestive enzyme activity, hematological parameters, innate immunity, and the expression of immune-related genes in rainbow trout (*Oncorhynchus mykiss*). Aquaculture Nutrition, **https://doi.org/10.1155/anu/4030680**

Fagbenro, O., Adedire, O., Fateru, O., Owolabi, I., Ogunlanna, O., Akanbi, B., Fasanmi, T. & Ayo-Amu, P. (2005). Digestive enzymes assays in the gut of *Oreochromis niloticus* Linnaeus 1757, *Parachanna* (*Channa*) *obscura* Gunther 1861 and *Gymnarchus niloticus* Cuvier 1829. Animal Research International, 2, 292-296.

Kayode, O. T., Ohanaka, N. J., Femi-Olabisi, F. J., Agboola, A. O., Oyebanji, E. O. (2022). *Parquetina nigrescens* species: A concise review of phytochemistry and pharmacology. Tropical Journal of Natural Product Research, 6(4), 454-461. **https://doi.org/10.26538/tjnpr/v6i4.1**

FAO (Food and Agriculture Organization) (2024). The state of the world fisheries and aquaculture: Blue transformation in action, 227. Rome: FAO of the United Nations. https://digitallibrary.in.one.un.org/TempPdfFiles/28661_1.pdf

Garg, C. K., Sahu, N. P., Maiti, M. K., Shamna, N., Deo, A. D., & Sardar, P. (2021). Dietary *Houttuynia cordata* leaf extract and meal enhances the immunity and expression of immune genes in *Labeo rohita* (Hamilton, 1822). Aquaculture Research, 52, 381–394. https://doi.org/10.1111/are.14901

Li, N., Shang, X., Ding, Z., Sun, H., Li, M., Wang, J., Ma, L., Xia, S., & Zhang, X. (2026). Effect of compound Chinese herbs on growth performance, digestion and serum biochemical indexes of Mandarin fish (*Siniperca chuatsi*). Israeli Journal of Aquaculture - Bamidgeh. 78(1):26-32. https://doi.org/10.46989/001c.151286

Naiel, M. A. E., Khames, M. K., Abdel-Razek, N., Gharib, A. A., El-Tarabily, K. A. (2021). The dietary administration of miswak leaf powder promotes performance, antioxidant, immune activity, and resistance against infectious diseases on Nile tilapia (*Oreochromis niloticus*). Aquaculture Reports, *20*, 100707.https://doi.org/10.1016/j.aqrep.2021.100707.

Ojuade, F. I., Olorundare, O. E., Akanbi, O. B., Afolabi, S. O., Njan, A. A. (2021). Antidiabetic and antihyperlipidemic effects of aqueous extract of *Parquetina nigrescens* in streptozotocin–nicotinamide induced type 2 diabetic rats. Heliyon, 7(6), e07363.

Olumide, M. D., Adeyinka, A. O., Shobo, B. A., Oreagha, T. (2022). Nutritional and ethnomedicinal potentials of *Parquetina nigrescens* leaf extracts in livestock production. Tropical Animal Production Investigations, 25(1), 15-16.

Paray, B. A., Bhat, E. A., Fawole, O. O., Umma, S. B., Adeshina, I. (2025). Dietary herbal leaves mixture extract enhances growth, antioxidant status, and resistance to *Gyrodactylus malalai* in heteroclarias catfish, *Clarias gariepinus* ♀ × *Heterobranchus longifilis* ♂. Aquaculture Nutrition. https://doi.org/10.1155/anu/1989752

Promprom, W., Khunchalee, J., Chatan, W., & Munglue, P. (2024). Effects of dietary *Boesenbergia rotunda* (L.) Mansf. rhizome extract on the growth of hybrid catfish (*Clarias macrocephalus* × *Clarias gariepinus*). Fisheries and Aquatic Sciences, 27(10), 665 - 676. https://doi.org/10.47853/FAS.2024.e63

Saba, A. B., Oyagbemi, A. A. & Azeez, O. I. (2010). Antidiabetic and haematinic effects of *Parquetina nigrescens* on alloxan induced type-1 diabetes and normocytic normochromic anaemia in wistar rats. African Health Sciences, 10(3), 276–282.

Sutthi, N., Panase, A., Chitmanat, C., Sookying, S., Ratworawong, K., & Panase, P. (2020). Effects of dietary leaf ethanolic extract of *Apium graveolens* L. on growth performance, serum biochemical indices, bacterial resistance and lysozyme activity in *Labeo chrysophekadion* (Bleeker, 1849). Aquaculture Reports, 18, 100551. https://doi.org/10.1016/j.aqrep.2020.100551

Tennant, B. C., & Center, S. A. (2008). Hepatic function. In J. J. Kaneko, J. W. Harvey, & M. L. Bruss (Eds.), Clinical Biochemistry of Domestic Animals (13, 378–412). Amsterdam: Elsevier.