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Determination of Biochemical Parameters and Growth Response of *Clarias gariepinus* Fingerlings Fed with *Cirina forda* Larvae and *Telfaria occidentalis* Leaves and their Proximate Composition

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ABSTRACT

The mainpurpose of this research is to investigate the biochemical parameters and growth response of Clarias gariepinus fingerlings fed with Cirina forda larvae and Telfaria occidentalis and the proximate composition of the two leaves. Grounded meals of C. forda larvae and T. occidentalis leaves were administered on C. gariepinus fingerlings at percentage ratio labelled A – H for ten weeks. Each treatment had three replicates and stocked with 10 fingerlings per replicate in 50-litres plastic tank and were fed daily. Blood samples from three randomly selected fish per treatment were obtained and were used to determine biochemical parameters such as Glucose, Cholesterol, Creatinine, Total protein, Alkaline Phosphatase (ALP), Albumin, Serum Glutamic Oxaloacetic Transaminase (SGOT) and Serum Glutamic Pyruvic Transaminase (SGPT) respectively using standard laboratory procedures. Blood samples for the analysis were collected in Lithium heparinized bottles to avoid clotting. Seven growth parameters; Final body weight (FBW), Body weight gain (BW), Specific growth rate (SGR), Feed Conversion Ratio (FCR), Growth rate (GR), Relative growth rate (RGR), and % Survival rate were determined using their respective formulas. The mean ranges of 23.00 - 60.00 mg/dl, 96.67 – 113.67 mg/dl, 1.10 – 1.60 mg/dl, 3.5 – 5.5 mg/dl and 2.03 - 2.70 mg/dl were obtained for glucose, cholesterol, creatinine, total protein, and albumin. Each mean range did not exceed its respective control value and also fell within the normal physiological range for fish. Diets (A, C, D, E, F and G) had lower SGOT mean range of 238.00 – 360.33 Mmol/l was observed as compared to their control values. Diets that lack C. forda were (E, F, G and H) and each group recorded higher SGPT values than the control. Growth parameters values were highly significant ($p<0.05$) to those of their respective control values. The present study showed that the compounded diet stand a better chance of replacing conventional fish meal due to their impact on fish performances. However, there were significant differences ($p<0.05$) in moisture, ash and carbohydrate contents of experimental diets. This study has shown that compounded diets containing C. forda larvae and T. occidentalis leaves elicited comparable values to the conventional feeds.

Keywords

Cholesterol Creatinine, SGR, Albumin, Total protein, SGPT, Moisture, Carbohydrate.

Introduction

The importance of fisheries sector to individuals and the economy of many developed and developing countries cannot be overlooked. It is noticeable that fish provides more than 60% of the world supply of protein, particularly in developing countries like Nigeria. Its impact could be directly or indirectly felt among rural dwellers and urban residents in Nigeria [1]. Fisheries is a pertinent subsector that particularly contributes about 3-5% to the agriculture share of the Gross Domestic Product (GDP) in Nigeria. Fish is an important protein source in the diets of Nigerians over

time. Protein from fish is highly digestible and of high nutritional value and comprise of amino acids, vitamins, and minerals. Asides the high-quality protein contents of the fish compared to other protein sources like beef, pork, chicken and goat meat, it is also cheaper and easily accessible products (Akinrotimi *et al.* [2].

One of the key problems faced by Nigerian aquaculture industry includes inadequate supply and prohibitive cost of quality fish feeds. As a result of this, most of the fish farmers particularly in the rural areas still depend on agricultural wastes including poultry litters to feed their fishes Aderemi *et al.* [3]. Attempts were being made continually to produce fish meal of high quality in other to reduce the level of anti-nutritional factors in some feed inclusions to the barest minimum Baruah *et al*. [4]. Aquaculture can return to its greatest potential of supplying the national fish requirement of Nigeria if the high cost of fish meal is surmounted using locally available resources in order to reduce the overall cost of production. However, sustainable fish production and fish feed increment is inevitable in order to boost the protein source of Nigerian citizens especially during this period of highly cost of daily usable commodities Adesina *et al.* [5]. Therefore, in order to achieve more economically sustainable, and viable fish feed production, this research work have been directed towards using *Cirina forda* and *Telfaria occidentalis* as part of feed inclusion for *Clarias gariepinus* for improved biochemical qualities and its growth response.

Materials and Methods

Description of the Experimental Site

The experiment was conducted in 2014 and was carried out in the Fishery Laboratory of the Department of Pure and Applied Biology, Ladoke Akintola University of Technology, Ogbomoso, Oyo State, Nigeria. The coordinates of the research location is latitude 8° 8′0″N and Longitude 4° 16′ 0″ E. Ogbomoso is in Southern Guinea savanna zone. The climate is equatorial, notably with dry and wet seasons with relatively high humidity. The rainy season span through April to October while the dry season lasts from November to March. The average daily temperature ranges between 25-35°C almost throughout the year. The annual rainfall is about1200 mm with average range of 786.2 to 1513 mm [6].

Sample Collection

Three hundred and ninety (390) fingerlings of *Clarias gariepinus* used for the experiment were obtained from the Fish farm of Oyo State Ministry of Agriculture located at Ogbomoso. The fingerlings were acclimatized in twenty seven bowls each of 50 liters capacity. For a period of two weeks, fingerlings were fed with conventional feed two times a day based on 5 % of their body weight, the fish were weighed in group of fifteen on weekly basis.

Fluted pumpkin (*Telfaira occidentalis*) leaves, were collected from a household in the study area and were identified at the herbarium of the Department of Pure and Applied Biology, Ladoke Akintola, University of Technology, Ogbomoso, Oyo State, Nigeria. The leaves were thoroughly washed with water to remove dirt, drained properly and later air dried for five days. Thereafter, the leaves were ground into fine powder and analyzed for proximate composition according to AOAC [7].

Cirina forda larvae were sourced from Akande market in Ogbomoso. Dirts were removed from the larvae and later sun-dried for 3 days. Thereafter, the larvae were grind into fine powder and analyze for proximate composition using AOAC [7]. All other feed ingredients like maize, groundnut cake, rice bran, fish meal and soybean used during the investigation were obtained from Oyo State Ministry of Agriculture Fish farm Ogbomoso.

Fish Diets Formulation and Processing

Eight different diets were prepared using Pearson's method of fish feed formulation to contain 35 % crude protein. Prior to processing, the two feed ingredients, the pumpkin leaf, *T. occidentalis* and *C. forda* larvae were grinded separately to a fine powder using hammer mill machine; they were then weighed and properly mixed together with adequate water added to ensure smooth pelleting. The conventional feed is the control diet which contained neither of the two ingredients, the diet compositions are shown on Table 1.The two ingredients were incorporated into each of the eight diets in the proportion presented in Table 2. The pelleted meals were sun-dried for 3 days to remove moisture. Gross composition of experimental diets Containing *Telfaria occidentalis* leaves and *Cirina forda* larvae are shown on Table 3.

Experimental Design

The experiment was set in a complete randomized design. Twenty seven (27) bowls were randomly allocated to nine treatment diets (A, B, C, D, E, F, G, H and Ct) in triplicate and fingerlings were randomly distributed into the bowls at a stocking density of thirteen fingerlings per bowl. Feeding was generally carried out twice daily (8.00 – 9.00 am) and (5.00 – 6.00 pm).

Table 1: Composition of Conventional Feed with 35 % Crude protein Content.

Ingredient	Quantity %
Fish Meal	16.00
Wheat bran	13.00
Maize grain	10.00
Groundnut cake	21.00
Rice bran	18.00
Soya beans	22.00
Telfaria occidentalis	0.00
Cirina forda	0.00

Table 2: Percentage Substitutions of the Compounded Feeds.

Treatment	Percentage substitution
A	25% *C. forda*, 25% *T.occidentalis*, 50% Conventional feeds
B	50% *C. forda*, 25% *T.occidentalis*, 25% Conventional feeds
C	75% *C. forda,* 25% *T.occidentalis*, 0% Conventional feeds
D	100% *C. forda*, 0% *T. occidentalis*, 0% Conventional feeds
E	0% *C. forda,* 25% *T. occidentalis*, 75% Conventional feeds
F	0% *C. forda,* 50% *T. occidentalis*, 50% Conventional feeds
G	0% *C. forda*, 75%*T. occidentalis*, 25% Conventional feeds
H	0% *C. forda*, 100% *T. occidentalis* , 0% Conventional feeds
CT	0% *C. forda*, 0% *T. occidentalis*, 100% conventional feeds

Table 3: Percentage Gross Composition of Experimental Diets Containing *Telfaria occidentalis* leaves and *Cirina forda* Larvae.

Parameters	A	B	C	D	E	F	G	H
Fish meal %	8.00	4.00	0	-	12.0	8.00	4.00	-
Wheat bran %	6.50	3.25	0	-	9.75	6.50	3.25	-
Maize grain %	5.00	2.50	0	-	7.50	5.00	2.50	-
Ground cake %	10.50	5.25	0	-	15.75	10.5	5.25	-
Rice bran %	9.00	4.50	0	-	13.50	9.00	4.50	-
Soybean powder %	11.00	5.50	0	-	16.50	11.00	5.50	-
***T. occidentalis* %**	25	25	25	-	25	50	75	100
***C. forda* %**	25	50	75	100	-	-	0	0

Blood Samples Collection

After 70 days of feeding, three fishes per treatment were randomly selected for biochemical analysis, 5 ml blood samples from each treatment were collected by cardiac puncture using 5 ml disposable syringe and was collected into the lithium heparium bottles and they were spined in a centrifuge at 100 nm revolutions per minute for 5 minutes. Following the standard methods of collection.

Determination of Biochemical Parameters

Serum Total protein and Albumin were determined colorimetrically by Biuret and Bromocresol Green binding procedure [8] [9]. Glucose, Cholesterol, Creatinine, Serum glutamic oxaloacetic transaminase (SGOT/AST), and Serum glutamic pyruvate transaminase (SGPT/ALT) were estimated colorimetrically using standard procedure described by Reitman and Frankel [10]. Alkaline phosphatase was measured with phenolphthalein monophosphate method Klein *et al.* [11] and Babson *et al.* [12].

Growth Measurement: Growth was determined using the following parameters.

$$\textbf{Specific Growth Rate (SGR) \%} = \frac{\ln(Wl1)-\ln(Wl2)}{l1-l2}x100$$

Where $W1_1$ and WI_2 are weights at respective times I_1 and I_2 and the differences of I_1-I_2 is duration period (in days) considered between W_2 and W_1

Mean Weight Gain (MGW) = Mean final weight-Mean initial weight

$$\textbf{Growth Rate} = \frac{Fw-Iw}{Fw}x100$$

Where Fw is final weight and Iw is the Initial weight

$$\textbf{Absolute Growth Rate} = \frac{Fw}{Iw}x100$$

$$\textbf{Feed Conversion Ratio (FCR)} = \frac{Total\ Dry\ Diet\ Fed}{\text{Total Weight Gain}}$$

Determination of Proximate Analysis

The *Clarias gariepinus* were analysed for proximate composition such as crude protein, crude fibre, lipid, Ash and moisture contents according to the procedure described below

Determination of Crude Protein

The total nitrogen (crude protein) was determined using the Kjeldahl method. About 0.5 g of the fish sample was weighed on a Nitrogen-free paper. The paper was wrapped round the sample and dropped at the bottom of the Kjeldahl digestion flask together with 6-8 glass beads, 4-5 spatulaful of granular mixture of CuSO4 and K_2SO4 as catalyst and 20 ml of concentrated H_2SO4 was carefully added. The flask was gently heated on a Gerhardt heating mantle in an inclined position in a fume cupboard until full digestion was achieved (when the liquid changed from brown to colourless). The contents of the flask were then transferred to a clean 100 ml volumetric flask, and 25 ml aliquot was used for the distillation and total nitrogen was determined calorimetrically according to the procedure of Huda *et al.* [13].

Determination of Crude Fibre and Carbohydrate

Crude fibre and carbohydrate of the experimental diets were determined by the use of AOAC [14] procedure. Carbohydrate was calculated by subtracting the sum (g/100g dry matter) of crude protein, crude fat, ash and fibre from 100g. The calorific value was also determined.

Determination of Fat Content

The percentage lipid content in the experimental diets was determined using the Soxhlet extraction method. An empty extraction thimble was weighed and noted as W1, about 5 g of the ground powder was measured into the empty thimble, the weight of the extraction thimble plus the sample was recorded as W2. The extraction thimble and its content was placed gently in the extractor, 110 ml of petroleum ether at 40 – 60 °C boiling point was turned into the round bottom flask and then placed in the heating mantle, the extractor was fitted onto the round bottom flask followed by the reflux condenser and connected to tapped water inlet tube with the outlet emptied in the sink. The heating mantle was switched on and adjusted so that the solvent in the round bottom flask boils gently. During the heating process, water was allowed to run constantly through the reflux condenser to cool and condense the evaporating solvent which then collects in the extractor thus extracting the lipid from the sample in the extraction thimble of the extractor, when the extractor is filled with solvent it is then siphoned back into the round bottom flask, this process goes on continuously for 6 hours. At the end of 6 hours, the extraction thimble and its content was removed from the extractor and oven dried at 50°C to a constant weight which was recorded as W3 following the procedure described by Bolawa *et al.* [15]. The percentage lipid was calculated as follows:

$$\textbf{\% Lipid Content} = \frac{W2-W3}{W2-W1}X100$$

Where W1 = Weight of empty extraction thimble W2 = Weight of extraction thimble plus sample extraction W3 = Weight of extraction thimble plus sample residue after extraction.

Determination of Ash Content

Ash content of feed samples was determined by incineration in a carbolated Sheffield LMF3 muffle furnace at 5000°C AOAC

[14]. The difference in weight of the feed samples before and after heating was taken as the ash content, the formula is as follows:

$$\textbf{\% Ash Content} = \frac{W2-W0}{W1-W0} X100$$

Where; W0 = Empty crucible, W1 = Dry sample; and W2 = Ash sample

Determination of Moisture Content

The moisture content of feed samples was determined using the oven dry method. 5g of the samples were placed in weighed crucibles maintained at 80°C in an oven until constant weights were obtained. The samples were transferred into desiccators to cool to ambient temperature and reweighed according to the procedure of Bolawa *et al.* [15]. The difference in weights indicates the dry matter and was calculated as follows:

$$\% \text{ Moisture Content} = \frac{W1-W2}{W1} X100$$

Where; W1 = Wet sample, W2 = Dry sample

Statistical Analysis

All data collected were subjected to Analysis of variance (ANOVA). The means were separated using Duncan's multiple Range Test (DMRT).

Results

The present result revealed that fish fed with sample A had highest mean value of glucose (60.00 ± 0.58) least value was (23.00 ± 0.67) fed with sample G. Though, fish fed with sample C, E, F, also revealed high glucose content in their body tissues and are significantly differ (p<0.05) with those fed with control (70mg/dl). The mean values were significantly lower (p<0.05) in all other groups. The fish fed with conventional feed revealed highest cholesterol level (119.67 ± 0.88) mg/dl and significantly differ from the mean values of experimental diets that ranged between (98-113) mg/dl mg/dl. Similarly, Fish fed with experimental diets had lower level of creatinine, total protein and significantly differ from the mean value for control. Albumin level of fish fed with experimental diets C (2.63 ± 1.67) g/dl and F (2.70 ± 0.10) g/dl were higher compared to mean value of control, while other diets produce lower albumin to the fish under study. *Clarias gariepinus* fed with B has high amount of SGOT (Serum glutamic oxaloacetic transaminase) and B(369.33 ± 0.67) Mmol/1 and H(370.67 ± 0.33) Mmol/1 while other groups including control samples produced lesser SGOT in fish samples examined. The serum glutamic pyruvate transaminase (SGPT/ALT) were higher in the fish fed with feed inclusion E, F, G and H, even more than the value recorded for the control. Alkaline phosphatase reported for this study were higher in fish fed with feed inclusion C, D, G and H compared to the control.

There were significant differences (p<0.05) in the body weight of the fish samples across the experimental diet groups. Growth rate and relative growth rate were significantly differ (p<0.05) across the groups. Survival rates were very high in fish samples fed with 50% *Telfaria occidentalis* and 50% conventional feeds. However, there were significant differences (p<0.05) in the initial and final weights of the fish samples examined respectively. Highest growth response was reflected in *C. gariepinus* fed with group B and C that have high percentage feed inclusion of *Cirina forda* and *T. occidentalis*.

High percentage of moisture contents of all experimental diets more than the control implies that the herbal leaves has good amount of water which can satisfy the fish. There were significant variations (p<0.05) in the crude protein values across the experimental diets. Group G with 75% *T. occidentalis* and 25% conventional feeds have high value of crude protein (23.65%) and F (19.06%) with 50% *T. occidentalis* and 50% conventional feed. Crude fibres of experimental diets were significantly varied (p<0.05) while Fat values were not significantly differ (p>0.05) across diet groups except group H and control. High percentage of moisture contents of all experimental diets more than the control implies that the herbal leaves has good amount of water which can satisfy the fish The present study revealed significant variation (p<0.05) in values of Ash, Moisture, and Carbohydrates contents of experimental feeds and the values were higher compared to the control values.

Discussion

The highest glucose level reported in sample A could be attributed to high percentage composition of *C. forda* in the diet formulation that gives needed nutrient to the fish examined. Similar observation was reported by Akintayo *et al.* [16]. Departed report was observed by Ali *et al.* [17] who reported decrease glucose in the serum of rainbow trout examined for all treatments. Lesser values of Cholesterol in fish fed with all the experimental diets indicated low fat which makes the fish healthier instead of fat accumulation. This may be due to active ingredients contained in *C. forda* and *T. occidentalis*. The same observation was noted by Obaroh *et al.* [18] in sampled culture *Clarias gariepinus* in Kebbi State. Contrary opinion was reported by Ali *et al.* (2021) who noted high cholesterol levels in the control. Decreased level of creatinine in the *C. gariepinus* presently studied was also observed by Adesina *et al.* [5]. Higher levels of Protein in the body of fish examined revealed that high percentage diet inclusion produced necessary amount of protein to *C. gariepinus*. Contrary observation was reported by Yadav *et al.* [19] and Adesina *et al.* [5]. The increase level of Albumin in diets C and F given to the fish may be due to high percentage inclusion and beneficial ingredients in the diets. Similar report was made by Yadav *et al.* [19] and Ali *et al.* [17] who observed increased albumin with increasing percentage of plant extract than the control and ascribed it to boosting of immune responses of fish by supplemented diets. Deviated observation was reported by Adesina *et al.* [5]. Increase values of ALT and AST in the serum of *C. gariepinus* for this study could be ascribed to stress undergone as a result of additional levels of anti-nutrients even at high percentage of *C. forda* and *T. occidentalis* inclusion. Same report was observed by Dienye and Olumuji [20] in their study of fish fed with Moringa oleivera leaf meal. Contrary opinion

Table 4: Mean Values of Plasma Biochemical Parameters of *Clarias gariepinus* fingerlings Fed with the Experimental Diets and the Control.

Parameters/Diets	Control	A	B	C	D	E	F	G	H
Glucose mg/dl	$70.00^{a} \pm 0.57$	$60.00^{b} \pm 0.58$	$31.30^{e} \pm 0.67$	$55.00^{bc} \pm 5.03$	$31.00^{e} \pm 1.00$	$44.00^{d} \pm 0.88$	$49.00^{d} \pm 3.18$	$23.00^{f} \pm 1.16$	$30.00^{e} \pm 0.67$
Total chol mg/dl	$119.67^{a} \pm 0.88$	$100.33^{c} \pm 0.33$	$98.67^{c} \pm 0.33$	$100.00^{c} \pm 1.00$	$113.67^{b} \pm 3.67$	$98.67^{c} \pm 0.33$	$96.67^{c} \pm 0.67$	$100.33^{c} \pm 0.67$	$100.33^{c} \pm 0.33$
Creatinine mg/dl	$1.73^{b} \pm 0.03$	$1.53^{bc} \pm 0.09$	$1.10^{f} \pm 0.07$	$1.47^{cd} \pm 0.17$	$1.60^{bc} \pm 0.06$	$1.37^{cde} \pm 0.07$	$1.17^{ef} \pm 0.09$	$2.03^{a} \pm 0.03$	$1.27^{def} \pm 0.03$
Total protein g/dl	$5.33^{a} \pm 0.07$	$5.00^{bcd} \pm 0.06$	$4.53^{bc} \pm 0.15$	$5.20^{ab} \pm 0.15$	$4.33^{f} \pm 0.03$	$5.13^{ab} \pm 0.07$	$5.03^{bc} \pm 0.03$	$4.73^{de} \pm 0.07$	$4.83^{cde} \pm 0.07$
Albumin g/dl	$2.53^{ab} \pm 0.07$	$2.23^{cd} \pm 0.03$	$2.20^{cd} \pm 0.06$	$2.63^{a} \pm 1.67$	$2.03^{d} \pm 0.03$	$2.37^{bc} \pm 0.03$	$2.70^{a} \pm 0.10$	$2.03^{d} \pm 0.03$	$2.17^{cd} \pm 0.03$
SGOT Mmol/l	$365.00^{ab} \pm 2.52$	$360.33^{b} \pm 0.67$	$369.33^{a} \pm 0.67$	$250.33^{e} \pm 0.33$	$238.00^{f} \pm 6.55$	$300.33^{d} \pm 0.03$	$352.67^{c} \pm 0.33$	$359.33^{bc} \pm 1.20$	$370.67^{a} \pm 0.33$
SGPT Mmol/l	$182.67^{d} \pm 0.67$	$175.67^{d} \pm 2.85$	$180.00^{de} \pm 0.58$	$167.33^{a} \pm 3.18$	$174.00^{e} \pm 3.00$	$190.33^{c} \pm 0.67$	$207.67^{b} \pm 3.33$	$199.67^{b} \pm 0.33$	$201.00^{b} \pm 0.58$
Alkphos Mmol/l	$35.33^{c} \pm 0.67$	$33.33^{c} \pm 0.67$	$34.00^{c} \pm 0.58$	$38.00^{bc} \pm 1.00$	$42.00^{b} \pm 1.00$	$43.30^{b} \pm 0.33$	$43.33^{b} \pm 5.17$	$51.33^{a} \pm 0.67$	50.67a ± 0.33

Mean with different superscripts are significantly different (P<0.05) along the rows

Table 5: Growth Response of *Clarias gariepinus* fingerlings Fed with the Experimental Diets and the Control.

Parameters	A	B	C	D	E	F	G	H	Control	SEM
Initial body weight (g)	108.0^{abc}	118.90^{a}	115.13^{ab}	108.87^{abc}	100.40^{bc}	99.57^{cd}	99.03^{cd}	86.70^{d}	107.9^{abc}	2.13
Final body wt (g)	149.83^{a}	154.67^{a}	152.60^{a}	147.07^{a}	142.13^{a}	147.63^{a}	148.57^{a}	141.47^{a}	137.97^{a}	3.47
Bodyweight gain (g)	41.83^{ab}	35.90^{b}	37.47^{b}	38.20^{b}	41.73^{ab}	48.06^{a}	49.54^{a}	54.77^{a}	30.90^{c}	2.83
Specific Growth Rate	0.44^{ab}	0.39^{b}	0.40^{ab}	0.41^{b}	0.48^{ab}	0.56^{ab}	0.58^{ab}	0.67^{a}	0.36^{b}	0.29
FCR	2.81^{a}	1.44^{a}	2.44^{a}	2.58^{a}	1.33^{a}	1.44^{a}	1.12^{a}	1.17^{a}	2.00^{a}	.239
Growth rate	25.92^{ab}	23.62^{ab}	23.44^{ab}	24.53^{ab}	28.25^{ab}	31.82^{a}	33.18^{a}	36.68^{a}	23.39^{ab}	1.49
Relative growth rate	38.01^{ab}	31.14^{ab}	32.75^{ab}	38.04^{ab}	39.88^{ab}	47.89^{ab}	49.84^{a}	58.55^{a}	21.32^{b}	2.80
Survival rate %	69	77	82	72	80	90	82	80	77	

Mean with different superscripts are significantly different (P<0.05) along the rows

Table 6: Proximate Composition of Experimental Diets in Percentages.

Parameters	A	B	C	D	E	F	G	H	Control
Crude protein	16.48^{d}	17.09^{c}	16.19^{e}	16.28^{e}	15.51^{f}	19.06^{b}	23.65^{a}	11.96^{h}	12.73^{g}
Crude fibre	6.21^{d}	6.44^{b}	6.12^{e}	6.37^{c}	6.14^{e}	6.42^{b}	6.12^{e}	7.40^{a}	6.12^{e}
Fat	3.37^{d}	3.36^{d}	3.29^{d}	3.41^{d}	3.42^{d}	3.61^{c}	3.60^{c}	4.04^{b}	4.64^{a}
Ash	9.14^{d}	9.77^{c}	8.70^{e}	9.50^{c}	10.03^{b}	11.02^{a}	10.24^{b}	10.01^{b}	9.50^{c}
Moisture	12.14^{a}	12.11^{a}	12.02^{a}	11.64^{b}	11.65^{b}	10.49^{bc}	10.42^{bc}	10.17^{bc}	9.82^{c}
Carbohydrates	52.68^{h}	51.29^{g}	53.70^{c}	52.82^{e}	53.26^{d}	49.36^{g}	46.23^{h}	56.44^{b}	57.21^{a}

Mean with different superscripts are significantly different along the row

was reported by Mousa and Khattab [21]. Okechukwu and Auta [22] also observed significant decreased values of AST and ALT in their study. Alkaline phosphatase that showed high values in the serum biochemistry of *C. gariepinus* could be associated with hepatic cellular damage that leads to their leakages into the blood stream. Similar report was made by Dienye and Olumiji [20], Mousa *et al.* [23] and Adesina *et al.* [5]. Contrary observation was observed by Mousa and Khattab [21].

The significant differences and increased in body weight of the *C. gariepinus* fed with different diets could be associated with varieties of nutrients present in the diets. Similar report was observed by Fahimeh *et al.* [24] and Yuli *et al.* [25] who reported improved weight gain of *C. gariepinus* fed with marshmallow and herbs. This report is also in line with the report of Turan [26] who examined *Oreochromis aureus* fed with *Trifolium protense.* However, decrease in weight gain of rainbow trout fed with plant meal diet was also observed by Pierce *et al.* [27].

The survival rate of the fish sample fed with 50% *T. occidentalis* and 50% conventional diet that were very high implied that active ingredients such as bioflavonoids is an active chemical that provide necessary ingredients for the survival rate of fish studied. Similar observation was reported by Yuli *et al.* [25] who opined that the survival rate of *C. gariepinus* fed with herbal feeds were higher compared to the control.

The highest crude protein, moisture and carbohydrate contents observed in fish fed with high percentage *T. occidentalis* is an indication of active ingredients present in its leaves. This report is in line with the report of Mahboob *et al.* [28]. High percentage of moisture contents of all experimental diets more than the control implies that the herbal leaves has good amount of water which can satisfy the fish. Contrary observation was reported by Ojewuyi *et al.* [29] who opined that moisture content were low in *Polyalthia longifolia* leaves used for their study. Significant variation reported for Moisture, Ash and Carbohydrate contents of different feed composition could be ascribed to different ingredients and some unknown factors in the medicinal leaves which favours the fish growth since their values were higher than the control feed. The same report was observed by Adekunle [30] who also examined the *T. occidentalis* powder as additive to *C. gariepinus* feed. Similar report of significant diffrences (p<0.05) in moisture, ash and carbohydrate was also observed by Zim *et al.*

[31]. However, Fat that was reported lower in values compared to control feed could be due to herbal nature of the experimental diets which may help in reducing fat levels of the fish. Contrary report was observed by Zuraini *et al.* [32] who observed higher lipids contents on *Synodontis schall*, *Clarias anguillaris* and *Lates niloticus in Malaysia* and Obaroh *et al.* [18] who also reported low fat in cultured *C. gariepinus* compared to wild ones in Kebbi State.

Conclusion

This study revealed that herb inclusion in fish feed improved the serum composition of *Clarias gariepinus*, due to increased proximate composition of the experimental diets and also showed that these herb inclusions is a promising fish feed for better growth of *C. gariepinus.*

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