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Serum biochemistry profile of *Clarias gariepinus* (Burchell 1822) fingerlings fed varying levels of dietary maca (*Lepidium meyenii,* Walp.) root powder

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Abstract

The serum biochemistry profile of Clarias gariepinus fingerlings fed varying levels of dietary maca (Lepidium meyenii, Walp.) root powder was investigated. Dietary maca root powder was included at: 0.0g/100g (TM_1 i.e. control)), 0.25g/100g (TM_2), 0.5g/100g (TM_3,) 0.75g/100g (TM_4) and 1.0g/100g (TM_5) in triplicate, fed to C. gariepinus fingerlings (n = 300, 10.0±0.00g) in *fifteen (15) plastic tanks (n = 20) at a fixed feeding rate of 3% body weight twice daily for twelve (12) weeks. Blood samples were collected for serum biochemical analysis. Data collected were analysed using one-way analysis of variance (ANOVA) at P = 0.05. Serum Total protein (4.15±0.7 – 4.85±0.1 g/dL), albumin (2.55±0.7 – 2.75±0.5 g/dL), globulin (1.60±0.03 – 2.10±0.07 g/dL), albumin/globulin (A: G) ratio (1.30±0.9 – 1.60±0.7), aspartate aminotransferase i.e. AST (32.60±2.7 – 33.65±1.4 U/L), alanine aminotransferase i.e. ALT (53.67±2.5 – 54.80±2.8 U/L), glucose (313.10±1.8 – 320.30±1.6 mmol/L), potassium (2.58±0.4 – 3.00±0.3 mmol/L), sodium (136.10±1.8 – 142.20±1.2 mmol/L) and chlorine (106.20±1.2 – 110.50±1.7 mmol/L) respectively were obtained. The serum aspartate transaminase (AST), alanine aminotransferase (ALT) and glucose values recorded significant increases (P<0.05) with inclusion of maca root powder while potassium values decreased (P<0.05). Total serum protein, albumin, globulin, albumin/globulin (A: G) ratio, sodium and chlorine values were not significantly influenced (P>0.05) with the inclusion of maca root powder. Therefore, further study may be required to assess the suitability of dietary maca root powder at higher levels into formulated feed, its correlation with the various biological impacts, and the possible implication for human consumers.*

Keywords: *Albumin, Chlorine, Blood Samples, Globulin, Potassium.*

Introduction

The serum biochemical parameters of cultured fish are crucial for monitoring their health during cultivation (Abdel - Hay *et al.*, 2021). Serum biochemical parameters indicate the suitability of feed, physiology, and environment of cultured fish (Rawung *et al.*, 2019). Responses to feed, pollution, and stress, as well as ecological and physiological factors, affect the serum biochemical parameters of fish (Okey *et al.*, 2022). Changes in serum biochemical parameters depend on fish species, age, sexual maturity cycle, and health condition (Rawung *et al.*, 2019). The key variations that occur in serum biochemistry related to feed are fluctuations in protein, glucose, cholesterol, and other serum biochemical components (Ndatsu *et al.*, 2022). Serum biochemistry profile are particularly useful in assessing a fish's physiological and pathophysiological status since morphological and biometric parameters alone do not always

provide a complete picture (Abdel - Hay *et al.*, 2021). However, reliable reference ranges for comparison are necessary for the data on serum biochemistry profile of fish species to be meaningful (Ndatsu *et al.*, 2022). There is still a scarcity of published literature establishing normal serum biochemical values and reference ranges for numerous fish species. Normal values exist for only a handful of serum biochemical parameters, and the established values tend to have a wide range due to a lack of standardization between methods (Abdel - Hay *et al.*, 2021). Research on supplementation of feed additives such as maca (*Lepidium meyenii*, Walp.) root powder into fish feed may have effects on fish serum biochemistry. Maca (*L. meyenii*) is a vegetable root plant which can be used in its dry powder form as additive in animal feeds (Garba *et al.*, 2023). Plants are natural source of safer and cheaper chemicals. Beneficial effects of bioactive plant substances in animal nutrition may include the stimulation of appetite and feed intake, the improvement of endogenous digestive enzyme secretion, activation of immune responses and antibacterial, antiviral and antioxidant actions (Ikwor, 2025). Maca is an adaptogen, which helps the body adapt to stress, contains plant sterols which help the body produce the appropriate levels of hormone and also help in the digestion of some vital nutrients (Gaddafi *et al.*, 2023). Maca has been used as a food health - promoting food material, stamina builder, and fertility promoter due its various biological effects, including the regulation of metabolism and hormonal secretion, improvement in memory and digestion, and also antidepressant activity in humans (Zhang *et al.*, 2017). Maca possess the ability to improve, digestion, energy, modulate the response against oxidative stress and improve carcass quality (Tang *et al.*, 2017). Maca comprises various classes of bioactive compounds, including saponins, alkaloids, steroid hormones, and polyphenol compounds, Maca is also rich in crude protein, crude lipid, some essential acids (Ali *et al.*, 2024). *Clarias gariepinus* (Burchell 1822) is the most cultured and economically important freshwater fish species in Nigeria, economic benefits of this fish species are due to its hardiness, fast growth, large size attainable and ability to withstand and tolerate harsh environmental conditions (Ali 2022). This study investigated the serum biochemistry profile of *C. gariepinus* fingerlings fed varying levels of dietary maca (*L. meyenii*,) root powder.

Materials and Methods

Study Area

The study was carried out at Lay – Joy Fish Farm, Gombe – Yola road, Billiri local government area (LGA), Gombe State Nigeria. Billiri LGA which lies within Latitude 9°50'N; 11°09´E and Longitude 9.833°N 11.150°E, covers an area of 737km² (285 sq. m) and is 50 km away from Gombe the State capital.

Experimental Fish

Three hundred (300) *C. gariepinus* fingerlings with mean initial weight (10.0±0.00g) were stocked at twenty (20) fingerlings per tank in triplicate per treatment after one (1) week of acclimatization, the study lasted for twelve (12) weeks.

Experimental Feed

The formulated feed contained; fish meal (FM), soybean meal (SBM), yellow maize meal (YMM), groundnut cake meal (GNCM) and dietary maca root powder. All ingredients were grounded into a fine powder using a hammer mill and sieved by a 0.25 mm sieve. Fish meal, soybean meal, groundnut cake meal and yellow maize meal were obtained from commercial suppliers in Gombe, the vitamin/mineral premix, fish oil and chromic oxide (Cr_2O_3) were purchased from TTS Integrated Farms Lagos, while the dietary maca root powder naturally derived from dry maca (*L. meyenii*) root was obtained from Bon – Amour Pharmacy Limited Lagos, Nigeria. The dietary maca root powder was included at varying levels for each treatment feed and was diluted into 100 ml of warm water to form a solution. The solution was sprayed onto the pelleted feed. Fish oil 15ml/kg feed was added

to preserve dietary maca root powder. The feed was prepared in one kg each time, the feed was mixed by a feed mixer machine to achieve homogeneity, pelleted using a pelleting machine, air – dried and kept at room temperature until feeding. Experimental feed was prepared by inclusion of dietary maca root powder at varying levels into formulated feed with 40% crude protein as recommended by Ali *et al.* (2024) at 0.0g/100g ($TM_{1)}$), 0.25g/100g (TM_2), 0.5g/100g ($TM_{3,}$) 0.75g/100g (TM_4) and 1.0g/100g (TM_5) inclusion levels, feed without dietary maca root powder (TM_1), served as the control as shown in Table 1.

Table 1: Ingredient Percentage Composition (g/100g) of Experimental Feed with Varying Levels of Dietary Maca Root Powder

Ingredients (%)	TM_1	TM_2	TM_3	TM_4	TM_5
Fish meal	20.00	20.00	20.00	20.00	20.00
Soybean meal	21.50	21.25	21.00	21.00	21.00
Groundnut meal	23.00	23.00	23.00	23.00	23.00
Yellow maize	30.00	30.00	30.00	29.75	29.50
Maca root powder	**0.00**	**0.25**	**0.50**	**0.75**	**1.00**
Fish Oil	1.00	1.00	1.00	1.00	1.00
Vegetable oil	1.00	1.00	1.00	1.00	1.00
Starch	1.00	1.00	1.00	1.00	1.00
Lysine	0.25	0.25	0.25	0.25	0.25
Methionine	0.25	0.25	0.25	0.25	0.25
*Vitamin/premix	1.00	1.00	1.00	1.00	1.00
Salt	0.50	0.50	0.50	0.50	0.50
Cr_2O_3	0.50	0.50	0.50	0.50	0.50
Total	100.0	100.0	100.0	100.0	100.0

Key: TM_1 – Maca root powder (0.0g/100g), TM_2 – Maca root powder (0.25g/100g), TM_3 – Maca root powder (0.5g/100g), TM_4 – Maca root powder (0.75g/100g), TM_5 – Maca root powder (1.0g/100g).

Experimental Design

The *C. gariepinus* fingerlings were cultured in fifteen (15) rectangular plastic tanks (flow-through system) with a water holding capacity of one thousand litres (1,000 L) each in a completely randomized design (CRD). Each tank was washed thoroughly with salt, filled to just a little over 1/3 (350 litres) capacity, and stocked with twenty (20) fingerlings of *C. gariepinus* with a mean initial weight of (10.0±0.00 g). The *C. gariepinus* fingerlings were fed the experimental feed at 3% body weight two (2) times daily for twelve (12) weeks. The quantity of feed was adjusted accordingly after every two (2) weeks of sampling for growth performance and survival rate (mean body weight and mortality). Water temperature, pH, dissolved oxygen was measured using the Horiba U-22 XD multi-parameter water quality checker, while ammonia was measured using a freshwater aquaculture test kit (Model AQ-2, Code 3633-03, Lamotte U.S.A.) at the beginning of the experiment, after which they were measured weekly throughout the period of the experiment.

Determination of Serum Biochemistry Profile

Blood samples were collected from three (3) fish at the start and again from three (3) fish from each treatment and the control at the end of the feeding trial, following the method described by Dauda *et al.* (2023) for serum biochemical analysis. The fish blood samples were analysed at the Specialist Hospital Gombe, Gombe State for serum biochemical contents such as; total protein (TP), albumin, globulin, the albumin/globulin (A: G) ratio, aspartate aminotransferase (AST), alanine aminotransferase (ALT), glucose, potassium, sodium, and chlorine. A 2ml

plastic syringe needle was inserted at the ventral midline just posterior to the anal fin of the fish at angle 450 until it penetrated the caudal vessel lying between adjacent haemal arches. This was accomplished by inserting the needle until it stopped at the backbone. Blood was then drawn slowly into the syringe and preserved in labelled ethylene diminitetra acetate (EDTA) bottles. The non – heparinized blood samples were centrifuged at 3000 rpm for 10 minutes to extract serum as described by Dauda *et al.* (2023). The serum samples were stored at -80°C for the biochemical analysis. Total serum protein was determined by the biuret method, albumin content was estimated by the bromocresol green binding method, globulin content was calculated by subtracting albumin values from Total protein, while albumin/globulin ratio was estimated by dividing albumin values by those of globulin (Abdel - Hay *et al.*, 2021), aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were determined calorimetrically as described by Dauda *et al.* (2023), while the glucose, potassium**,** sodium and chlorine concentrations were all determined by using flame photometer (Ali *et al.*, 2025).

Statistical Analysis

Data obtained were analysed using one – way analysis of variance (ANOVA) at P = 0.05, where significant differences were detected, mean values were separated using least significant difference (LSD).

Results and Discussion

The serum biochemistry profile of *C. gariepinus* fingerlings fed varying levels of dietary maca root powder is presented in Table 2. The serum aspartate transaminase (AST), alanine aminotransferase (ALT) and glucose values recorded significant increases ($P<0.05$) with inclusion of maca root powder while potassium values decreased ($P<0.05$). The Total serum protein, albumin, globulin, albumin/globulin (A: G) ratio, sodium and chlorine were not significantly influenced ($P>0.05$) with the inclusion of maca root powder.

Table 2: Serum Biochemistry Profile of *C. gariepinus* Fingerlings Fed Varying Levels of Dietary Maca Root Powder

Parameters	Initial	TM_1	TM_2	TM_3	TM_4	TM_5
Total protein (g/dL)	4.00±0.3	4.15±0.7	4.30±0.9	4.40±0.6	4.85±0.1	4.50±0.5
Albumin (g/dL)	2.45±0.4	2.55±0.7	2.60±0.2	2.60±0.3	2.75±0.5	2.65±0.8
Globulin (g/dL)	1.55±0.06	1.60±0.03	1.70±0.08	1.80±0.05	2.10±0.07	1.85±0.09
A: G ratio	1.59±0.1	1.60±0.7	1.53±0.4	1.44±0.5	1.30±0.9	1.43±0.3
AST (U/L)	$25.00±2.3^b$	$32.60±2.7^a$	$33.30±3.6^a$	$33.40±1.7^a$	$33.65±1.4^a$	$33.45±2.4^a$
ALT (U/L)	$31.60±1.9^b$	$54.80±2.8^a$	$54.60±2.1^a$	$54.50±2.7^a$	$53.67±2.5^a$	$54.30±2.4^a$
Glucose (mmol/L)	$298.10±1.1^b$	$320.30±1.6^a$	$318.00±2.7^a$	$316.00±2.3^a$	$313.10±1.8^a$	$315.20±1.4^a$
Potassium (mmol/L)	$5.11±0.6^b$	$3.00±0.3^a$	$2.93±08^a$	$2.89±0.9^a$	$2.58±0.4^a$	$2.83±0.5^a$
Sodium (mmol/L)	133.40±2.9	136.10±1.8	138.30±1.6	139.70±2.7	142.20±1.2	140.00±1.4
Chlorine (mmol/L)	104.10±1.9	110.50±1.7	106.20±1.2	106.60±1.8	107.30±1.6	107.00±1.3

Mean values in each row with similar superscripts are not significantly different ($p>0.05$).

Key: TM_1 – Maca root powder (0.0g/100g), TM_2 – Maca root powder (0.25g/100g), TM_3 – Maca root powder (0.5g/100g), TM_4 – Maca root powder (0.75g/100g), TM_5 – Maca root powder (1.0g/100g).

The aminotransferase enzyme is an enzyme that catalyses the transamination reaction (Okey *et al.,* 2022). There are two types of serum transaminase enzymes namely aspartate aminotransferase (AST) enzyme and alanine aminotransferase (ALT) enzyme. The main source of AST enzyme is in liver, whereas ALT enzyme are found in tissues especially in heart, skeletal muscle, kidney and brain (Hastuti and Subandiyono, 2018). The final serum AST values, 32.60±2.7 – 33.65±1.4 U/L were lower than 53.49 – 59.29 U/L reported by Okey *et al.* (2022) for *C. gariepinus* juveniles fed sesame seed meal as replacer for soybean meal. Elezuo (2016) reported that the recommended range values for ALT was 10 U/L – 55 U/L. An increase in the final serum AST values observed in all the fish fed feed with varying levels of dietary maca root powder when compared with the control feed indicated the efficient utilization of dietary amino acid by fish for growth or high energy demand. Analyses of serum AST values give proper situations on the status of hepatocytes and diseases, which could be damaging to the liver caused by the reactive oxygen species generated (Ali *et al.,* 2025). However, all the values of serum AST evaluated were within the recommended range, which signify the sound healthy status of the fish.

The final serum ALT values, 53.67±2.5 – 54.80±2.8 U/L were higher than the values, 11.24 – 22.43 U/L reported by Ndatsu *et al.* (2022) for *C. gariepinus* juveniles fed fish meal containing different inclusion levels of *Sesame indicum* seed. Elezuo (2016) stated that the recommended range values for ALT was 7 U/L – 56 U/L. An increase in the final serum ALT values observed in the fish may be related to the immune – potentiating effect of the feed which increase the enzymes to the range of normal levels which indicated the high rate of phosphorylation and transport of substances across the cell membrane that may be leading to elevated kidney detoxification effects, elevations of the serum ALT may reflect an alteration in the pathway ways of biosynthesis and mixed – function oxidase (Ndatsu *et al.,* 2022).

Glucose concentrations are used as an indirect stress indicator in fish, since high stress levels on fish are reflected on higher cortisol amounts in circulating blood, which resulted to corresponding higher glucose levels thereby increasing susceptibility to disease (Ali *et al.,* 2025). The final serum glucose values, 313.10±1.8 – 320.30±1.6 mmol/L were comparable with the values, 307.00 – 361.00 mmol/L reported by Elezuo (2016) for *C. gariepinus*. The increase may signify the efficacy of the rapid secretion of glucocorticoids and catecholamine's from the adrenal tissues of the fish due to some anti – metabolites. This might enhance the process of glycolysis which causes the increase in the serum glucose final values.

Electrolytes such as potassium, sodium and chlorine are molecules found throughout the blood tissues and cells of fishes including *C. gariepinus*, these molecules which could be either positive or negative ions conduct electric current and help to balance pH and acid base levels in the fish, they also facilitate the passage of fluid between and within cells through a process known as osmosis and also play a part in regulating the functioning of neuro-muscular, endocrine and excretory system of fishes, electrolyte concentration is indicative of the ability of the fish to osmoregulation (Ali *et al.,* 2025). The final serum potassium values, 2.58±0.4 – 3.00±0.3 mmol/L were lower than the values, 6.92 – 11.60 mmol/L reported by George and Uedeme – Naa (2020) for *C. gariepinus* juveniles in response to treatment with detergent (linear alkylbenzene sulfonate). These values were within the recommended range values, 2.30 – 5.2 mmol/L reported by Elezuo (2016). This indicated that the feed stabilized the serum potassium values within the recommended range. The final serum sodium values, 136.10±1.8 – 142.20±1.2 mmol/L were lower than the values, 157.33 – 259.02 mmol/L reported by Ike – Obasi and Ukazu (2018) for *C. gariepinus* juvenile treated with paraquat

dichloride. These values were within the normal range values, 130.00 – 150.00 mmol/L reported by Elezuo (2016). This signify that the feed stabilized the serum sodium values of the fish to the optimal levels. The final serum chlorine values, 106.20±1.2 – 110.50±1.7 mmol/L were comparable with the values, 107.30 – 110.70 mmol/L reported by Ali *et al.* (2025) for *C. gariepinus.* The findings indicated that the feed was suitable in maintaining the fish serum chlorine values.

The measurement of Total serum proteins and albumin are of considerable value in fish, as it relates to general nutritional status as well as the integrity of the vascular system and liver function (Ahmad *et al.,* (2015). The final Total serum protein values, 4.15±0.7 – 4.85±0.1 g/dL were comparable with the values, 4.20 – 4.75 g/dL reported by Ali *et al.* (2025) for *C. gariepinus* fingerlings fed feed co – supplemented with betaine and β – glucan. The improvement in the final Total serum protein values might be as a result of the anabolic effect of the varying levels of dietary maca root powder in improving the nutritional value of the feed. The final serum albumin values, 2.55±0.7 – 2.75±0.5 g/dL were comparable with the values, 2.60 – 2.75 g/dL reported by Ali. (2022) for *C. gariepinus* fingerlings fed β – glucan Additive diet. The improvement in the final serum albumin values could be attributed to the actively coordination of the function of various endocrine glands in the body by the varying levels of dietary maca root powder. Serum globulin is a component of serum protein. The final serum globulin values, 1.60±0.03 – 2.10±0.07 g/dL were lower than the values, 1.60 – 3.10 g/dL reported by Adesina (2017) for *C. gariepinus* juveniles fed mechanically extracted sunflower (*Helianthus annuus*) seed meal. The improvement in the final serum globulin values indicated a stronger innate immunity by the varying levels of dietary maca root powder. The final serum albumin/globulin (A: G) ratio values, 1.30±0.9 – 1.60±0.7 were comparable with the values, 1.38 – 1.63 reported by Ali *et al.* (2025) for *C. gariepinus* fingerlings. A lower A: G ratio in the serum is an indication that there is a greater globulin concentration in the Total protein compared to the albumin content, which was the situation in this present study. However, since the gamma globulin fraction constitutes the largest part of the overall globulin content, it could be concluded that the varying levels of dietary maca root powder have stimulatory effects on the serum biochemical profile of the *C. gariepinus* and hence the immune response.

Conclusion

The dietary maca root powder inclusion at varying levels indicated that the serum aspartate transaminase (AST), alanine aminotransferase (ALT) and glucose values recorded significant increases ($P<0.05$) with inclusion of maca root powder while potassium values decreased ($P<0.05$). The Total serum protein, albumin, globulin, albumin/globulin (A: G) ratio, sodium and chlorine values were not significantly influenced ($P>0.05$) with the inclusion of maca root powder. Therefore, further study may be required to assess the suitability of dietary maca root powder at higher levels into formulated feed, its correlation with the various biological impacts, and the possible implication for human consumers.

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