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**Abstract:** Seasonal variations in serological profiles and growth status of farmed and wild *Clarias gariepinus* were investigated. Serological profiles of *Clarias gariepinus* can be an effective tool for proper monitoring of stress induced by management practices in intensive fish culture, which may elicit devastating effect on fish. Spectrophotometry procedures were used to analyze serum parameters of *Clarias gariepinus*. No different seasonal patterns were observed for mean values of albumin, total protein, urea, cholesterol, glucose and alanine amino-transferase. However, aspartate amino-transferase showed different seasonal pattern. Levels of cholesterol and alanine amino-transferase were highly significant in farmed and wild fish, separately. Juvenile fish had high level of urea irrespective of season. Seasonal variations in water quality parameters were observed except for pH which had no seasonal pattern. The growth exponential shows *b*-values between -0.048 and 7.434 for *Clarias gariepinus*. Adult female and juvenile fish from the wild had the highest *b*-value and the least *b*-value, separately. In this study, *b*-values were higher in the wild fish than the farmed fish. The condition factor for *Clarias gariepinus* ranged from 0.422 to 0.698, and was observed to be high in juvenile fish. With a condition factor less than 1, fish may not be doing well, probably due to environmental stress. Some serological parameters varied according to season and environment of fish. Thus, serological profile of fish is an effective and sensitive tool to monitor fish response to stress factors in the environment.

Key words: Serology, Clarias gariepinus, farmed fish, wild fish, Asaba, Nigeria.

## **1. Introduction**

Fish is one of the most valuable sources of cheap protein available to Nigerians. This has led to a high demand for fish, which has necessitated an increase in pisciculture with management practices that could cause a lot of stress to farmed fish [1]. Intensive fish culture has been associated with effects of chronic stress, such as reduction of growth rate, alteration in physical condition, health of fish and activation of stress responses [2]. Management procedures as crucial as they are, produce some level of disturbances, which can elicit a stress response leading to the decreased fish performance, alterations of the blood parameters [3, 4], the increased susceptibility to diseases [5] and in extreme cases leading to mortality [6]. Environmental degradation resulting in aquatic pollution has also taken its toll on wild fish populations. Fish therefore is exposed to stress and frequent disease outbreaks [7]. Fish is disposed to a number of diseases, and unfortunately there are few diagnostic tools available to fish health professionals to evaluate these diseases. Haematological parameters are considered as patho-physiological indication of the whole body, and therefore are important in diagnosing the structural and functional diseases of fish and assessing the status of fish heath [8]. Haematological studies on most tropical fishes have focused on haematological parameters rather than on blood serum, thereby creating an imbalance between available research information in haematological and serum characteristics of fish. Serological parameters have been reported to be closely related to fish response to

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the environment in which it lives [9]. Serological values of fish are generally used as effective and sensitive tool to monitor physiological and pathological changes in fish with respect to different environmental conditions. Thus, serology is a valuable diagnostic tool in evaluating the health status of fish under different environmental conditions, as the normal composition of serum and other tissues of the fish may be changed by the various conditions prevailing in the environment including diseases [10, 11]. Management procedures with and without apparent warning may elicit very distressing effect on fish. Thus, baseline information on the serum profile of Clarias gariepinus is necessary for proper monitoring of stress in fish in order to reduce to the negative effect of management procedures. This study examined the serum characteristics of farmed and wild Clarias gariepinus-a common catfish, which has been widely adopted as a choice culture species in Nigeria.

# 2. Materials and Methods

## 2.1 Collection of Fish Samples

One hundred and eighty *Clarias gariepinus* samples were collected in six months between November, 2012 and January, 2013, representing the dry season, and between May and July, 2013, representing the rainy season.

Ninety Farmed fish samples were collected from Obiora farms in Asaba and 90 wild fish samples were collected from local fisher folks who caught the fish with traps at Ona Lake in Delta state (Fig. 1). Thirty *Clarias gariepinus* fish—15 from farm and 15 from wild, with each 15 made up of five male, five female and five juvenile fish, were bought once every month. Fish samples were obtained on the same day and were separately transported live in 30 L open plastic bucket, containing little amount of water from the different sources of procurement to the laboratory.

#### 2.2 Biometric Measurements

Fish samples were measured for weight (g),

standard length (cm) and total length (cm) using the lark model (LP 302A) electric weighing balance and measuring board, respectively. Adult and juvenile fish were used approximately of the same size ranges. Fish weight ranged from 480.2 g to 501.5 g and fish standard length ranged from 41 cm to 55 cm for adult fish; while fish weight ranged from 65.14 g to 211.91 g and total length ranged from 21 cm to 31 cm for juvenile fish for both farmed and wild fish samples.

#### 2.3 Collection of Fish Blood Serum Samples

To avoid mixing the farmed and wild fish samples together, fish samples were maintained in separate glass tanks ( $45 \times 45 \times 90$  cm) and handled separately. Physical stress on fish was avoided by handling each fish gently as the blood samples were drawn from the pectoral caudal vein with the aid of a sterile disposable clinical 5 mL syringe and needle. The needles were inserted at right angle of the vertebral column of each fish and were gently aspirated during penetration. The needles were pushed down until blood started entering the syringe. The needle was withdrawn after 3.0 mL of blood was drawn and immediately transferred into a microcentrifuge test tube.

## 2.4 Experimental Procedure

The blood collected from each fish sample was transferred into separate sterile tubes and allowed to clot for a few minutes (approximately 3-5 min) before centrifugation at 4,000 rpm for 5 min at 4 °C to isolate the serum using a table centrifuge model 800. Serum samples were transferred to another labelled sterile tube. The labelled sterile tubes were placed in ice and stored at 70 °C prior to analyses.

#### 2.5 Serological Analyses

The frozen serum samples were left at room temperature to thaw. Serum sample for each individual fish from both farmed and wild sources was analyzed for the following parameters: albumin (ALB, g/L), total protein (TP, g/L), urea (UR, mmol/L), cholesterol (TC, mmol/L), glucose (GLU, mmol/L) alanine amino-transferases (ALT, U/L) and aspertate amino-transferases (AST, U/L), which were determined by spectophotometry using serological kit (biosystem, Barceiona, Spain), 2ID PEC (medical USA) according to Fatih et al. [12] and Gul et al. [13].

## 2.6 Water Quality Parameters

Water quality parameters, such as temperature (°C), pH, dissolved oxygen (mg/L), transparency (cm) and suspended matter (g/100 mL), were determined according to American Public Health Association (APHA) [14].

## 2.7 Length Weight Relationship (LWR) of Fish

The LWR was determined from Eq. (1):

$$W = aL^b \tag{1}$$

The parameters a and b in the formula were estimated through logarithmic transformation in Eq. (2):

$$\log W = \log a + b \log L \tag{2}$$

where,

W = body weight of fish (g);

L =total length of fish (cm);

b = growth exponent or regression coefficient;

Loga = intercept on the *Y*-axis.

# 2.8 Condition Factor (K) of Fish

The condition factor (K) was estimated from the

relationship in Eq. (3):

$$K = \frac{100W}{L^3} \tag{3}$$

where, K is condition factor, L is standard length of fish (cm) and W is weight of fish (g).

# 2.9 Statistical Analysis

A one-way analysis of variance was used to determine variability in serological parameters. Paired *t*-test was used to compare serological values between the farmed and wild *C. gariepinus* and for seasonal variation in serological values.

## **3. Results**

3.1 Seasonal Variations in Serological Profiles of Farmed and Wild Fish

Seasonal variations in mean values of serological profiles of farmed and wild adult and juvenile *Clarias gariepinus* are presented in Tables 1 and 2. Mean values of albumin obtained for adult male, adult female and juvenile *C. gariepinus* did not show any seasonal pattern. Wild juvenile fish had high mean values of albumin irrespective of season (Fig. 2). Fig. 3 shows that juvenile fish had high mean values of total protein in both dry and rainy seasons.

These values were, however, not significantly (P > 0.05) higher than values obtained for adult female and adult male fish. The level of urea was higher in farmed fish than wild fish, and higher in juvenile fish

Table 1 Seasonal variation (dry season) in mean values of serological parameters of farmed and wild Clarias gariepinus.

Serological parameters	Farmed fish			Wild fish		
Serological parameters	Adult male	Adult female	Juvenile	Adult male	Adult female	Juvenile
Albumin (g/dL)	$3.46\pm0.68$	$2.51\pm0.45$	$2.32\pm0.76$	$3.03\pm0.30$	$3.40\pm0.34$	$2.87\pm0.34$
Total protein (g/dL)	$3.50\pm0.37$	$4.35\pm0.53$	$3.40\pm0.35$	$4.59\pm0.60$	$3.71\pm0.44$	$6.08\pm0.58$
Urea (mmol/L)	$2.04\pm0.15$	$2.56\pm0.13$	$4.11\pm0.54*$	$1.30\pm0.06$	$1.27\pm0.03$	$1.65\pm0.19$
Cholesterol (mmol/L)	$27.70\pm0.29*$	$25.48\pm0.38^{\ast}$	$30.13 \pm 1.7 *$	$9.33 \pm 1.94$	$18.32\pm4.83$	$13.53\pm0.99$
Glucose (mmol/L)	$4.17\pm0.61$	$5.20\pm0.49$	$5.13\pm0.51$	$3.00\pm0.31$	$3.37\pm0.32$	$6.88\pm0.78$
Alanine amino-transferase (U/L)	$1.88\pm0.28$	$1.92\pm0.35$	$1.40\pm0.04$	$3.03\pm0.49$	$3.01\pm0.24$	$2.31\pm0.45$
Aspertate amino-transferase (U/L)	$1.28\pm0.12$	$2.01\pm0.55$	$1.61\pm0.12$	$1.88\pm0.12$	$1.49\pm0.10$	$1.41\pm0.23$
Weight (g)	$494.16\pm3.73$	$494.22\pm3.97$	$143.03\pm29.3$	$494.18\pm3.84$	$494.36\pm3.79$	$114.30\pm25.52$
Total length (cm)	$47.20 \pm 1.65$	$48.40\pm2.56$	$27.00\pm1.58$	$47.40 \pm 1.69$	$47.40 \pm 1.93$	$25.00\pm1.78$

\*Means significant values at 95% confidence level.

Serological parameters	Farmed fish			Wild fish		
Serological parameters	Adult male	Adult female	Juvenile	Adult male	Adult female	Juvenile
Albumin (g/dL)	$3.61\pm0.23$	$3.68\pm0.11$	$2.26\pm0.14$	$3.01\pm0.09$	$3.54\pm0.17$	$2.96\pm0.19$
Total protein (g/dL)	$3.68\pm0.14$	$4.70\pm0.21$	$3.56\pm0.38$	$3.35\pm0.14$	$4.57\pm0.21$	$4.69\pm0.38$
Urea (mmol/L)	$2.21\pm0.08$	$2.26\pm0.10$	$3.73\pm0.13$	$2.12\pm0.09$	$2.10\pm0.11$	$3.57\pm0.13$
Cholesterol (mmol/L)	$28.07 \pm 1.39 *$	$31.65 \pm 3.39*$	$32.30\pm2.47*$	$14.47 \pm 1.86$	$20.29 \pm 1.25$	$15.8\pm1.19$
Glucose (mmol/L)	$4.81\pm0.37$	$6.39\pm0.47$	$6.26\pm0.39$	$4.96\pm0.24$	$5.94\pm0.32$	$5.68\pm0.31$
Alanine amino-transferase (U/L)	$1.98\pm0.30$	$2.23\pm0.12$	$2.60\pm0.32$	$3.66\pm0.30$	$3.53\pm0.12$	$2.60\pm0.32$
Aspertate amino-transferase (U/L)	$5.50\pm0.91$	$5.23\pm0.75$	$1.62 \pm 0.17$	$4.28\pm0.68$	$4.12\pm0.59$	$1.74 \pm 0.11$
Weight (g)	$487.05\pm3.33$	$466.44\pm10.61$	$115.35\pm7.84$	$492.13\pm3.27$	$493.03\pm6.09$	$122.08\pm13.62$
Total length (cm)	$44.79\pm0.62$	$46.17\pm0.66$	$25.35\pm0.79$	$48.45\pm0.57$	$69.69 \pm 12.67$	$26.91\pm0.93$

Table 2 Seasonal variation (rainy season) in mean values of serological parameters of farmed and wild Clarias gariepinus.

\*Means significant values at 95% confidence level.

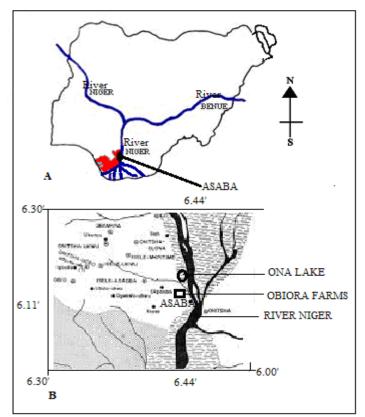


Fig. 1 Map of Nigeria showing Delta State at Asaba (A) and map of Asaba and environs showing sampling locations, Ona Lake and Obiora Farms (B).

Adapted from www.Nigeria.com/map.

than in the adult farmed and wild male and female fish (Fig. 4). Urea was highly significant (P < 0.05) in juvenile fish sourced from the farm in the dry season. Mean values of cholesterol showed a seasonal pattern of variation. Cholesterol levels were significantly (P < 0.05) higher in fish sourced from the farm than fish from the wild irrespective of season, size and sex of

fish (Fig. 5). The level of glucose was high in adult female and juvenile fish in farm and wild in both the rainy and dry seasons (Fig. 6). Mean values of alanine amino-transferase were generally higher in the wild fish than farmed fish irrespective of season (Fig. 7). Mean values of aspartate amino-transferase were higher (P <0.05) in rainy season than dry season (Fig. 8). Adult

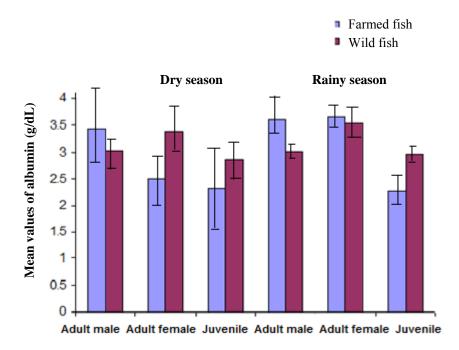
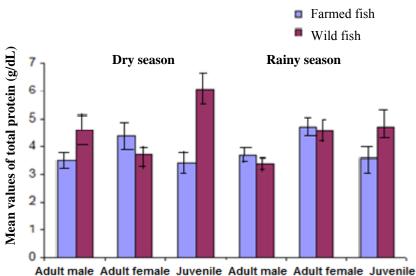


Fig. 2 Mean values of albumin in farmed and wild Clarias gariepinu (with standard error as vertical bars).



#### Fig. 3 Mean values of total protein in farmed and wild *Clarias gariepinus* (with standard error as vertical bars).

male and female had higher (P > 0.05) mean values of aspartate amino-transferase than juvenile fish.

## 3.2 Seasonal Variation in Water Quality Parameters

Seasonal variation in water quality parameters is presented in Table 3. Temperature was highest in the culture pond during the dry season and lowest in the culture pond in the rainy season. Dissolved oxygen was highest in the wild and least in the culture pond during the rainy season. pH values were high in the wild during the rainy season. Secchi disc transparency was observed to have high values in the rainy season. This shows that the water was more transparent in the dry season than in the rainy season. The water was also clearer in the wild than in the culture water. Suspended matter was higher in the rainy season than

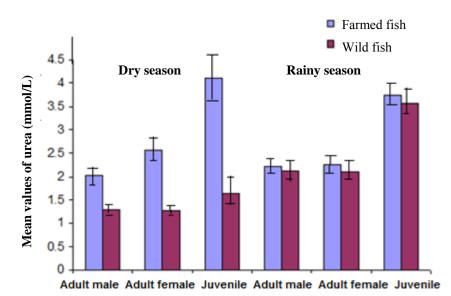


Fig. 4 Mean values of urea in farmed and wild *Clarias gariepinus* (with standard error as vertical bars).

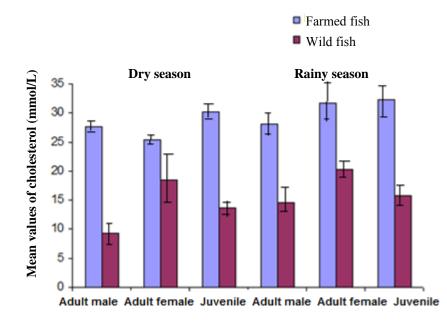


Fig. 5 Mean values of cholesterol in farmed and wild Clarias gariepinu (with standard error as vertical bars).

in the dry season. The water from the wild was generally lower in suspended matter than the water sourced from farmed source.

## 3.3 LWR

Parameters of LWR obtained in this study were presented in Table 4. The *b*-values for this species for adult male, adult female and juvenile *C. gariepinus* ranged from -0.048 to 7.434. Adult female fish

sourced from the wild during the dry season had the highest *b*-value, while juvenile fish also sourced from the wild during the dry season had the least *b*-value. The *b*-values obtained in this study were observed to be generally higher in fish sourced from the wild than in fish sourced from the farm.

## 3.4 Condition Factor (K)

Results of analysis for condition factors (K) of fish

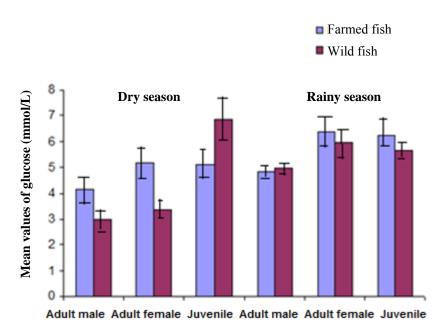


Fig. 6 Mean values of glucose in farmed and wild *Clarias gariepinus* (with standard error as vertical bars).

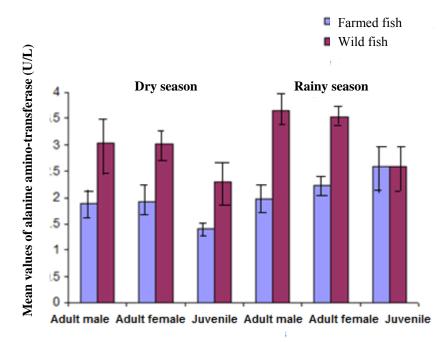


Fig. 7 Mean values of alanine amino-transferase in farmed and wild *Clarias gariepinus* (with standard error as vertical bars).

are presented in Table 5. The condition factor for *C*. *gariepinus* in farmed and wild female fish ranged from 0.422 to 0.4640, and ranged from 0.6673 to 0.698 in wild juvenile fish. Seasonal variation in mean condition factor (*K*) shows that the condition factor of *C. gariepinus* was slightly but not significantly (P > C

0.05) higher during the dry season than in the rainy season. Juvenile *C. gariepinus* generally had higher *K* than the male and female *C. gariepinus*. The female fish had the least *K*. Farmed fish had slightly higher *K* than the wild fish. The maximum value of *K* was obtained in adult female in the dry season.

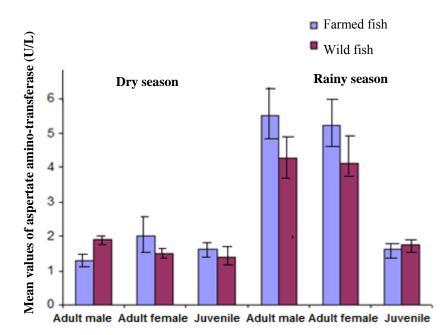


Fig. 8 Mean values of aspertate amino-transferase in farmed and wild *Clarias gariepinus* (with standard error as vertical bars).

Parameter	Dry s	eason	Rainy season		
	Farmed	Wild	Farmed	Wild	
Temperature (°C)	$30.10\pm0.30$	$29.70\pm0.10$	$26.40\pm0.60$	$26.60\pm0.30$	
Dissolved oxygen (mg/L)	$4.20\pm0.1$	$4.50\pm0.20$	$3.80\pm0.20$	$4.9\pm0.50$	
pH	$6.40 \pm 1.0$	$6.10\pm0.40$	$6.90\pm0.10$	$7.1\pm0.60$	
Transparency (cm)	$21.00\pm0.5$	$19.00\pm0.20$	$26.50\pm0.40$	$24.13\pm0.50$	
Suspended matter (g/100 mL)	$1.32 \pm 0.2$	$0.99\pm0.70$	$1.10\pm0.00$	$1.73 \pm 0.10$	

 Table 4
 Length-weight relationships and related statistics of farmed and wild Clarias gariepinus.

Fish samples	BWT (g)	TL (cm)	R	А	В	F	P value
DAFM	$500.15\pm2.83$	$49.25\pm0.76$	0.000	1.390	0.356	0.000	1.000
DAFF	$497.86\pm1.93$	$50.29\pm0.92$	0.125	-0.763	1.156	0.096	0.909
DFJ	$143.18\pm14.81$	$27.45\pm0.78$	0.563	1.159	0.196	1.703	0.224
DAWM	$497.19\pm2.02$	$49.46\pm0.85$	0.678	-11.630	3.209	2.122	0.153
DAWF	$496.03\pm1.54$	$49.32\pm0.71$	0.737	-29.738	7.434	2.145	0.151
DWJ	$110.76 \pm 13.20$	$24.69\pm0.91$	0.743	-30.766	-0.048	1.644	0.252
RAFM	$487.05\pm3.33$	$44.79\pm0.62$	0.765	-19.694	-1.853	1.412	0.330
RAFF	$466.44 \pm 10.62$	$46.13\pm0.66$	0.776	-22.298	-0.259	1.136	0.451
RFJ	$115.35\pm7.85$	$25.35\pm0.79$	0.840	-53.001	0.911	1.331	0.395
RAWM	$492.13 \pm 3.27$	$48.45\pm0.57$	0.144	-0.443	0.694	0.277	0.608
RAWF	$491.03\pm6.53$	$48.37 \pm 1.09$	0.206	0.724	-0.410	0.267	0.770
RWJ	$122.09 \pm 13.62$	$26.91 \pm 0.93$	0.891	0.556	0.287	14.064	0.000

R: correlation coefficient, A: intercept, B: regression coefficient (slope), F: covariance.

DAFM: dry season adult farmed male; DAFF: dry season adult farmed female; DFJ: dry season juvenile; DAWM: dry season adult wild male; DAWF: dry season adult farmed; DWJ: dry season wild juvenile; RAFM: rainy season adult farmed male; RAFF: rainy season adult farmed female; RFJ: rainy season juvenile; RAWM: rainy season adult wild male; RAWF: rainy season adult wild female; RWJ: rainy season wild juvenile; BWT: body weight; TL: total length.

Fish	Dr	y season	Rainy season		
F ISH	Farmed	Wild	Farmed	Wild	
Male	$0.4820\pm0.02$	$0.4780\pm0.02$	$0.4450\pm0.02$	$0.4427 \pm 0.17$	
Female	$0.4640\pm0.03$	$0.4480\pm0.03$	$0.4227\pm0.03$	$0.4227\pm0.03$	
Juvenile	$0.6920\pm0.02$	$0.6980\pm0.01$	$0.6790\pm0.02$	$0.6673\pm0.02$	

Table 5 Seasonal variation in mean (± SE) condition factor (K) of farmed and wild *Clarias gariepinus*.

# 4. Discussion

Although no seasonal variations in mean values of albumin, total protein, urea and cholesterol were observed, farmed fish had higher mean values of albumin, total protein and cholesterol than wild fish. High levels of albumin have been attributed to environmental stress. Gras [15] observed relatively high albumin level in cultured Northern snakehead, which indicated that its concentration in serum is significantly influenced by environmental stress. Gabriel and Akinrotimi [1] noted that as aquaculture world over is becoming more and more intensive, manipulation of fish and other farm management procedures could produce some level of disturbances, which can elicit a stress response leading to decreased fish performance. Juvenile fish had high mean values of total protein and urea irrespective of seasons, which shows high protein in diet or increased protein catabolism. The elevation of serum urea may be due to the correlation between urea and increased protein catabolism or from more efficient conversion of ammonia to urea as a result of increased synthesis of involved enzyme in urea production [16]. Significantly higher values of cholesterol were observed in adult male, adult female and juvenile fish sourced from the farm than fish from the wild irrespective of season, size and sex of fish. Bender et al. [17] reported that small differences in body size do not result in significant changes in cortisol production and metabolism, which are related to elevated levels of cholesterol. Xu and Cao [18] and Coz-Rakovae et al. [19] also reported the elevated levels of cholesterol in farmed fishes. Furthermore, Yousafzai and Shakoori [20] reported that aquatic pollution leads to the elevated tissue cholesterol in fish. The high values

in glucose observed in farmed fish are in line with the findings of Barton and Iwama [21] and Coz-Rakovae et al. [19], who reported the differences in glucose concentration as a secondary response to stress. Adeyemo et al. [22] noted that fishes are very susceptible to physical and chemical changes in the environment, which may be reflected in their blood components. Alanine amino-transferases and aspartate amino-transferases are among the most important indicators of innate immune response to stress [23]. Mean values of aspertate amino-transferase were generally higher in adult farmed fish than in the wild fish. Svoboda et al. [23] reported the elevated levels in farmed fish, which is indicative of impairment of liver function.

Seasonal pattern was observed in water temperature. Temperature has been reported to have effects on blood sugar, urea, uric acid and protein levels [24]. Like the observations of Egborge et al. [25] and Odum [26], there was no discernible seasonal pattern in the pH. The water was more transparent in the dry season than rainy season. The dissolved oxygen in the wild water environment was better aerated than the culture medium.

Wild adult male and female studied exhibited positive allometric growth pattern with *b*-values > 3. All other fish samples had negative allometric growth pattern. According to Sangun et al. [27], values of the exponent "*b*" provide the information on fish growth, fish growth being isometric when "*b*" = 3 and allometric when "*b*"  $\neq$  3, which could be negative (when "*b*" < 3) or positive (when "*b*" > 3). In the present study, the condition factor for *C. gariepinus* ranged from 0.422 to 0.4640 in farmed and wild female fish, and ranged from 0.6673 to 0.698 in farmed and wild juvenile fish, respectively. Condition factor less than 1, could indicate that the fish is not doing well. Bagenal [28] documented that the condition factor of mature freshwater fish must to be in the range of 2.9-4.8. The mean "*K*" obtained in this study disagrees with that in Ref. [29], which reported positive allometric growth but with high "*K*" range of 0.91-8.66 and "*K*" could be influenced by factors including prevailing environmental condition, availability of food, feeding intensity, density or population changes, the period and duration of gonadal maturation among others.

# 5. Conclusions

Albumin, total protein alanine and amino-transferase values were higher in wild fish than farmed fish, while urea, cholesterol, glucose and aspartate amino-transferase values were lower in wild fish compared to farmed fish. Stressors, including water quality parameters and seasons of the year, may have affected the serum biochemical parameters of Clarias gariepinus. The significant differences in some serological parameters in farmed and wild Clarias gariepinus could therefore attributed to different habitat conditions and the type of management practices.

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