Survival, growth, and tissue biochemical profile of African catfish (*Clarias gariepinus)* juveniles fed diets supplemented with Country onion (*Afrostyrax lepidophyllus)* bark powder

YEMDJIE MANE Divine Doriane[1]*, NGOUANA TADJONG Ruben[1], ZANGO Paul[1], EBILE DAYAN Agwah[2], NANHOU Raïssa Linda[1], TSAMBOU MEGNIMEZA Astride Martine[2], POUOMOGNE Victor[1], TOMEDI EYANGO Minette [1]

[1]*Department of Aquaculture, Institute of Fisheries and Aquatic Sciences, University of Douala, P.O. Box: 7236-Douala, Cameroon.*

[2]*Department of Animal Science, Laboratory of Animal Nutrition, University of Dschang, P.O. Box: 96-Dschang, Cameroon.*

**Corresponding author's Email: dyemdjie5@yahoo.fr*

Key words: *Afrostyrax lepidophyllus*, Growth, Biochemical parameters, *Clarias gariepinus*, Fish diet, Survival

Submitted 21/01/2025, Published online on 30th April 2025 in the *Journal of Animal and Plant Sciences (J. Anim. Plant Sci.) ISSN 2071 – 7024*

1 ABSTRACT

To promote growth and maintain fish healthy, this study examined the effects of nutritional supplementation using *Afrostyrax lepidophillus* (Country onion) on survival, growth and biochemical parameters of African catfish, *Clarias gariepinus*. Juveniles weighing 9.29 ± 0.15 g were split up into four treatments in triplicate before being administered diets that contained 0, 10, 15 and 20 (T_0, T_1, T_2, and T_3, respectively) g/kg *A. lepidophyllus* for 56 days. Adding *A. lepidophyllus* bark in the diets enhanced significantly the survival rate, growth performances and tissue biochemical components of *C. gariepinus*. Fish fed with T_3 diet demonstrated the most favourable influence on tissue biochemical components and reduced the cost of production. This study demonstrated that *A. lepidophyllus* bark powder can be added to the diet of *C. gariepinus* at 2% inclusion level as a feed additive without any negative impact on the physiological function of the fish.

2 INTRODUCTION

In Cameroon's fish farms, the African catfish, or *Clarias gigipinus*, is one of the most often grown and well-liked fish species. It could be either polycultured together with Nile, tilapia and carp in fertilized ponds or monocultured in concrete and fast tanks, installed and supplied in a closed or open circuit, supplied with borehole water. *C. gariepinus* has several favourable traits for cultivation, including quick growth, strong survival in high density culture, disease resistance, immunity to low oxygen levels, and resilience to pH changes (Beingana *et al*., 2016). However, the unavailability of good quality feed at low cost remains the major constraint for the development of fish farming in Africa. *C. gariepinus* must be raised on premium foods with a high protein content and some dietary supplements or additives to maintain fish healthy and growth more (Sayed *et al*., 2011; Adegbesan *et al*., 2019). Several studies have demonstrated that the active ingredients in herbs, spices, and their byproducts (phytobiotics) include alkaloids, flavonoids, pigments, polyphenols, terpenoids, and steroids that have a number of uses and qualities, including immunostimulant, growth promoter, appetite stimulant, antioxidant, anti-stress, and antibacterial (Kumar *et al*., 2012). Among the plant species with phytobiotic potential in Cameroon, there is *Afrostyrax lepidophyllus*. Typically found in Tropical and Equatorial Africa, *Afrostyrax lepidophyllus* belongs to the Huaceae family (Cronquist, 1981; Moukette *et al*., 2015; Namkona *et al*., 2017). This herb is used in traditional medicine and as an antiseptic in the Congo to treat gastroenteric diseases (Bouquet, 1969). This shrub's seeds and bark have long been used as a spice in the Central African Republic and Cameroon. Numerous researchers have also conducted pharmacological experiments. Fogang *et al.* (2014) demonstrated that the phenylpropanoid (eugenol) and sulfur found in *A. lepidophyllus* bark can increase the activity of the digestive enzyme in the stomach mucosa and promote the action of pancreatic enzymes (lipases, amylases, and proteases). Furthermore, the bark includes polyphenolic compounds with strong antioxidant qualities (Oben *et al*., 2010; Fogang *et al.* 2014) with numerous additional characteristics such as lipid metabolism, stimulation of digestive enzymes, and regulation of microbial populations (Muneendra *et al*., 2014). The research conducted by Moukette *et al.* (2015) on the bark of *A. lepidophyllus* revealed remarkable antioxidant and free radical scavenging qualities. Additionally, they showed greater potential for protection against certain oxidative stress-related liver indicators. Fogang *et al.* (2014) also observed that A. lepidophyllus bar contains eugenol methyl, limonene, β-ocimene, apinene, trithiapentane, methyl-trithiahexane, dimethyl tetrathiaoctane, and pentathiaundecane. Prior research has examined the impact of *A. lepidophyllus* bark powder, which greatly improved the growth performance, feed nutrient utilization, and nutrient retention in the whole-body composition of the juvenile African catfish (Yemdjie *et al*., 2023). However, little is known about its physiological functions and activity in cultured fish. Therefore, the purpose of the current study was to examine the impact of *A. lepidophyllus* powder on the survival, tissue biochemical profile, and production cost of *C. gariepinus* juveniles.

3 MATERIALS AND METHODS

3.1 Study Area: This research was conducted between April and May 2022 at the Agro-ecological Farm of Bilone, located in the Obala, Lekié Division of the Center Region in Cameroon. This farm is positioned at 4°10' North and 11°31' East, with an elevation of 557 m above sea level. The yearly average temperature is 25°C. The average annual rainfall is 1577 mm, with a rainy season lasting 9 months (from March to October).

3.2 Experimental design, housing and equipment: Three hundred (300) juveniles of *C. gariepinus*, averaging a weight of 9.29 ± 0.15 g, were sourced from artificial reproduction performed at the farm hatchery. They were divided into four treatments, each of which had 75 fish and was reproduced three times, using a fully randomized methodology. The juveniles were contained in 12 experimental hapas, each measuring 0.5 x 0.5 x 1.0 m³, with 25 individuals placed in a concrete rectangular tank with a volume of 13 m³ and a height of 1.5 m. Water was supplied through 32 mm diameter PVC pipes, while drainage was facilitated by 90 mm diameter PVC pipes. The various feed rations were distributed manually three times daily: in the morning at 6 a.m., at noon at 12:00 p.m., and in the evening at 5 p.m., with each feeding amounting to 5% of the fish biomass. The growth and quantity of feed provided during each period were monitored using a landing net, and control fishing was conducted after 14 days during the cooler hours of the day at 6:00 a.m. Individual weights and lengths of the fish were measured using a precision electronic scale with 1 g accuracy and an ichthyometer, respectively. Furthermore, physico-chemical parameters such as water temperature (°C) using a maximum-minimum thermometer, dissolved oxygen (D.O.) using JBL Test Kits, pH, nitrite ($NO2^-$), and nitrate (NO3−) using Test strips (JBL Easy Test 6in1) were recorded daily prior to feeding. Table 1 displays the values of the water's physico-chemical properties that were noted throughout the experiment.

Table 1. Water quality parameters (Mean ± SD) for the 56-day testing period

Parameters	**Rearing period (days)**			
	0-14	**14-28**	**28-42**	**42-56**
T (°C)	28.25±1.08	28.65±1.13	28.67±0.92	28.7±0.84
pH	6.68±0.29	7.05±0.18	7.2±0,29	7.2±0.19
D.O (ppm)	7.85±0.02	7,89±0.55	7.63±0.59	7.5±0.13
NO_2^- (mg/L)	0±0	0.02±0.01	0±0	0.1±0.02
NO_3^- (mg/L)	0±0	0.3±0.01	0±0	0.25±0.03

Temperature (T°C); Hydrogen potential pH; dissolved oxygen (D.O); nitrite (NO_2^-); nitrate (NO_3^-)

3.3 Dietary experimentation: The bark of *A. lepidophyllus* was procured from the local market, processed by grinding and sieving, and the resulting powder was mixed into locally produced feed at varying proportions. Four experimental diets were created to be isoproteinic, isolipidic, and isoenergetic; they were called T_0, T_1, T_2, and T_3. These diets were made by adding 1%, 1.5%, and 2% of *A. lepidophyllus* to the baseline ratio (T0 or control). Table 2 provides information about the basic ratio (T0).

Table 2. Experimental diet formulation and proximate composition

Ingredients	**Quantities (g)**
Fish meal	27
Soybean meal	15
Peanut meal	20
Cotton meal	8
Wheat bran	8
Maize meal	16
Premix 5%	5
Palm oil	1
Total	**100**
Biochemical composition (%)	
Protein	38.02±1.71
Energy (kcal/kg DM)	283.25±3.17
Lipid	8.73±0.64
Ash	18.67±0.57
Moisture	9±1.00
Fiber	7.18±0.63
Dry matter	91±1.00

*Premix 5%: Crude protein =40%; Lysine =3.30; Methionine= 2.40; Calcium= 8; Phosphorus= 2.05; Metabolized Energy = 2078 kcal/kg.

3.4 Preparation of diets and proximate composition: The raw material was finely milled. *A. lepidophyllus*, which had been previously crushed, was added to the mixtures in accordance with the proportions outlined in Table 2. For each treatment, cold water was incorporated and mixed to achieve the desired consistency. An electric pelleting machine with a 150 kg/hour capacity was then used to pelletize the resultant mixture, creating pellets with a 2.5 mm diameter. After three days of sun drying, the pellets were placed in marked plastic containers and kept dry until they were needed. Ten grams of the prepared diet were analyzed using AOAC guidelines (1990). The sample was dried overnight at 105°C in an air convection oven to ascertain its moisture content. Crude protein was assessed using a KJELTEC SYSTEM 1002 Distilling Unit from Belgium, employing the Kjeldahl method following acid digestion (percentage crude protein = % nitrogen × 6.25). Crude lipid content was measured through extraction with petroleum ether using the Soxhlet method. It was determined how much ash was in the diets by burning the samples for 12 hours at 550°C in a muffle furnace.

3.5 Growth characteristics and survival rate: At the end of the feeding trial, the survival rate, and growth parameters for each treatment were evaluated by calculating mortality rate (MR), weight gain (WG), condition factor (K) and hepato-somatic index (HSI). The assessments were conducted using the specified formulae:

1) $\mathrm{MR}\ (\%) = \frac{\text{initial number of fish} - \text{final number of fish}}{\text{initial number of fish}}\ x\ 100$
2) WG (g) = Wf – Wi; Where: Wf = final weight; Wi = initial weight.
3) $\mathrm{K} = \frac{\text{Weight}}{\text{Length}^3}\ X\ 100$
4) $\mathrm{HSI}\ (\%) = \frac{\text{Liver weight}}{\text{Total fish weight}}\ x\ 100$

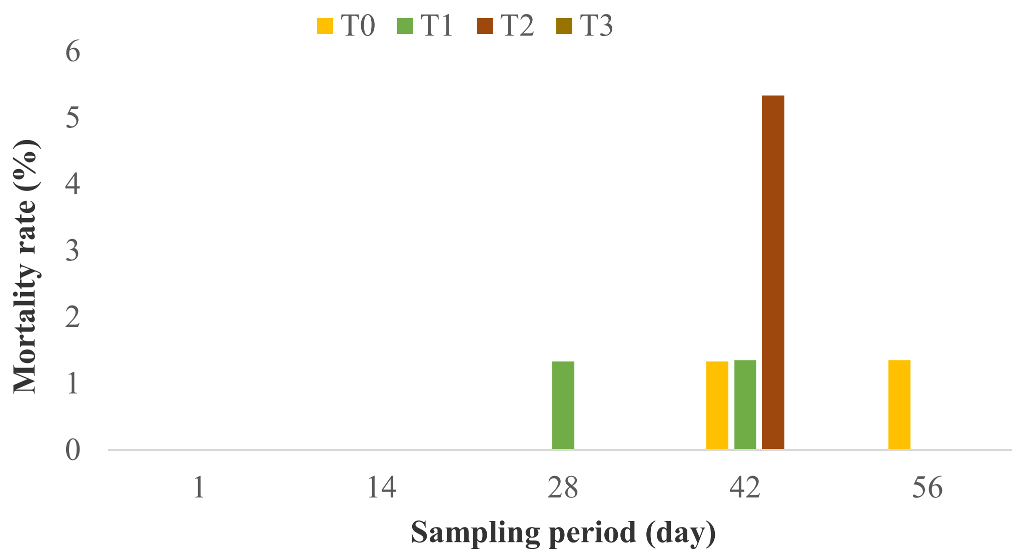
3.6 Tissue biochemical parameters*:* At the end of the experiment, 5 fish were sampled randomly from each hapa. For each fish, 0.6 g of flesh was removed before the caudal fin, then ground in a porcelain mortar placed on a block of ice with 3.4 ml of Tris buffer (10 Mm and pH 7.4), so as to obtain 15% homogenates. The ground material thus obtained was then centrifuged at 3000 rpm for 30 min using a cold centrifuge. For the assessment of biochemical parameters, the resulting supernatant was gathered in labeled Eppendorf microtubes and kept at 20°C. As directed by the LABKIT® (Espagne) kit, the colometric method was used to assess the level of total protein, albumin, aspartate aminotransferase (ASAT), alanine aminotransferase (ALAT), total cholesterol, HDL and LDL cholesterol, triglycerides, urea, and creatinine. Albumin content was subtracted from total protein content to determine globulin content.

3.7 Cost of production: The price of a kilogram of feed was determined by assessing the cost of each ingredient as per local market standards. To ascertain the feed intake cost, the average feed consumption was multiplied by the price per kilogram of the specific diet. The production cost for one kilogram of body weight was then calculated by multiplying the feed cost per kilogram by the relevant feed conversion ratio.

3.8 Statistical analysis: Each replicate's tissue biochemical profiles, growth characteristics, and mortality rates were averaged and used in statistical analysis. The Statistical Package for Social Sciences (SPSS 20.0) software's General Linear Model function was used to perform a one-way Analysis of Variance on the data. Duncan's multiple range tests were used to identify significant differences between treatment means, and probability values below 0.05 were deemed significant.

4 RESULTS

4.1 Mortality rate: The mortality rate observed across various treatments during the study illustrated in figure 1 did not correlate with the levels of *A. lepidophyllus* incorporated into the diet of *C. gariepinus*. At the end of the feeding period, the only mortalities registered were observed with *C. gariepinus* fed T_0 diet without supplement. Moreover, throughout the trial, *C. gariepinus* fed T_3 treatment supplemented with 2% *A. lepidophyllus* did not record any mortality.



T_0= control ration, T_1= T_0+1% *A. lepidophyllus*, T_2= T_0+1.5% *A. lepidophyllus*, T3= T_0+ 2% *A. lepidophyllus*

Figure 1. Mortality rate of *C. gariepinus* juvenile fed with different diets of *A. lepidophyllus* bark powder for 56 days.

4.2 Growth parameter and hepatosomatic index: Results showed that fish fed diet supplemented with 2% *A. lepidophyllus* obtained significantly (P < 0.05) high values of final weight, weight gain and condition factor. On the other hand, *C. gariepinus* fed T_2 diet (supplemented with 1.5% of *A. lepidophyllus)* recorded significantly (P < 0.05) the highest liver weight and hepatosomatic index (Table 3).

Table 3. Growth parameter and hepatosomatic index of *Clarias gariepinus* fed with *A. lepidophyllus* bark powder for 56 days.

Parameters	Treatments				
	T_0	**T_1**	**T_2**	**T_3**	**P. value**
Wi (g)	9.29 ± 0.51^a	9.11 ± 0.29^a	9.47 ± 0.51^a	9.41 ± 0.29^a	0.725
Wf (g)	65.85 ± 1.91^b	61.12 ± 6.29^b	67.08 ± 3.65^b	86.67 ± 6.27^a	0.0001
WG (g)	56.56 ± 2.4^b	52.01 ± 6.47^b	57.62 ± 3.62^b	77.25 ± 6.54^a	0.001
SGR (%/day)	$2,09 \pm 0,27^b$	$2,58 \pm 0,23^b$	$2,50 \pm 0,48^b$	$4,94 \pm 0,99^a$	0,001
K factor	0.76 ± 0.02^b	0.77 ± 0.56^b	0.78 ± 0.001^b	1.06 ± 0.01^a	0.0001
LW (g)	0.79 ± 0.24^b	0.72 ± 0.34^b	1.22 ± 0.32^a	0.79 ± 0.12^b	0.004
HSI (%)	1.09 ± 0.1^b	1.29 ± 0.24^{ab}	1.57 ± 0.27^a	1.21 ± 0.28^b	0.031

Values are mean ± standard deviation of three replicates of 25 fish each. Means in the same row with distinct superscripts differ significantly at P < 0.05. Wi, initial weight; Wf, final weight; WG, weight gain; K, condition factor; LW, liver weight; HSI, hepatosomatic index.

T_0= basal diet; T_1 = basal diet + 1% *A. lepidophyllus*; T_2 = basal diet + 1.5% *A. lepidophyllus*; T_3= basal diet + 2% *A. lepidophyllus*;

4.3 Tissues biochemical parameters: The data shown in table 4 revealed that, there was no significantly different ($P > 0.05$) for total proteins, total cholesterol, cholesterol LDL, triglyceride, urea and creatinine among the various treatments. The albumin and HDL-Cholesterol of fishes fed on the T_2 diet were significantly high ($P < 0.05$) compared to those fed T_0 (control) but was comparable ($P > 0.05$) with the other treatment groups. Except for those fed T_2 diet (1.5% *A. lepidophyllus)* the tissue content of ALAT was not significantly different ($P > 0.05$) as compared to the control group. ASAT significantly ($P < 0.05$) increased in the control group (T_0) from that of T_2 and T_3 but was similar ($P > 0.05$) to those fed T_1 diet. Globulin was significantly low ($P < 0.05$) in T_2 diet compared to the other treatment groups.

Table 4: Tissue biochemical parameters of *Clarias gariepinus* fed on diets supplemented with *A. lepidophyllus* for 56 days.

Biochemical parameters	Treatments				
	T_0	**T_1**	**T_2**	**T_3**	***P.* value**
Total proteins (g/dl)	0.60 ± 0.06^{a}	0.61 ± 0.03^{a}	0.58 ± 0.04^{a}	0.64 ± 0.08^{a}	0.420
Albumin (g/dl)	0.22 ± 0.02^{b}	0.24 ± 0.03^{ab}	0.29 ± 0.04^{a}	0.27 ± 0.04^{ab}	0.054
Globulin (g/dl)	0.38 ± 0.04^{a}	0.37 ± 0.02^{a}	0.29 ± 0.02^{b}	0.37 ± 0.05^{a}	0.004
A/G	0.59 ± 0.06^{b}	0.67 ± 0.10^{b}	1.01 ± 0.20^{a}	0.73 ± 0.14^{b}	0.001
ALAT (U/L)	120.56 ± 42.61^{b}	162.03 ± 33.38^{b}	222.09 ± 51.03^{a}	154.69 ± 27.49^{b}	0.008
ASAT (U/L)	74.93 ± 58.38^{a}	41.47 ± 22.70^{ab}	14.53 ± 12.04^{b}	23.67 ± 8.56^{b}	0.043
ASAT/ALAT	0.86 ± 0.93^{a}	0.27 ± 0.19^{ab}	0.07 ± 0.06^{b}	0.16 ± 0.07^{b}	0.074
Total cholesterol (mg/dl)	36.72 ± 13.86^{a}	53.60 ± 19.35^{a}	57.57 ± 14.30^{a}	57.57 ± 16.68^{a}	0.173
HDL (mg/dl)	9.83 ± 3.66^{b}	18.02 ± 10.68^{ab}	31.12 ± 14.65^{a}	19.65 ± 9.34^{ab}	0.037
LDL (mg/dl)	19.33 ± 14.32^{a}	26.74 ± 23.77^{a}	17.60 ± 14.79^{a}	29.80 ± 13.62^{a}	0.637
Triglyceride (mg/dl)	41.96 ± 10.65^{a}	43.64 ± 11.21^{a}	45.02 ± 10.21^{a}	40.80 ± 3.72^{a}	0.900
Urea (mg/dl)	23.84 ± 7.36^{a}	23.35 ± 4.67^{a}	28.34 ± 9.14^{a}	27.53 ± 7.65^{a}	0.630
Creatinine (mg/dl)	0.29 ± 0.11^{a}	0.28 ± 0.13^{a}	0.29 ± 0.46^{a}	0.26 ± 0.1^{a}	0.948

a, b: means in the same row with distinct superscripts differ considerably ($P < 0.05$).
T_0= basal diet; T_1 = basal diet + 1% *A. lepidophyllus*; T_2 = basal diet + 1.5% *A. lepidophyllus*; T_3= basal diet + 2% *A. lepidophyllus* ;

4.4 Cost of production: Throughout the study period, the highest ($P < 0.05$) costs of feed intake and production per kilogram of fish were recorded with *C. gariepinus* fed on a diet supplemented with 1.5% *A. lepidophyllus* (T_2). Although comparable with the other treatments, the lowest production cost per kilogram of fish ($P < 0.05$) was recorded with *C. gariepinus* fed on a diet supplemented with 2% *A. lepidophyllus* bark powder (Table 5).

Table 5. Cost of production of *Clarias gariepinus* fed on diets supplemented with *A. lepidophyllus* for 56 days.

Parameters (FCFA)	Treatments				
	T_0	T_1	T_2	T_3	P. value
CKF	394	426	442	458	
FI (g)	70.22	61.07	68.88	61.59	
CFI	27.67	26.01	30.44	28.21	
FCR	1.24 ± 0.05^a	1.18 ± 0.14^a	1.20 ± 0.08^a	0.80 ± 0.07^b	0.001
CPKF	489.75 ± 20.75^b	505.05 ± 58.78^b	529.80 ± 33.48^a	366.83 ± 30.27^c	0.004

a, b: means in the same row with distinct superscripts differ considerably ($P < 0.05$). FCFA refers to Francs CFA (1 US$= 600 CFA). CKF, cost of kilogram of feed; FI, feed intake; CFI, cost of feed intake; FCR, feed conversion ratio; CPKF, cost of production of kilogram of fish.
T_0= basal diet; T_1 = basal diet + 1% *A. lepidophyllus* ; T_2 = basal diet + 1.5% *A. lepidophyllus*; T_3= basal diet + 2% *A. lepidophyllus*.

5 DISCUSSION

The present study showed that, at the end of feeding period, the only mortalities registered were observed in *C. gariepinus* fed on a diet without supplement (T_0). When *C. gariepinus* were fed a food supplemented with 2% *A. lepidophyllus* (T_3), no mortality was seen. Nonetheless, it was found that the experimental water tanks' physico-chemical characteristics fell within the range suggested for the culture of freshwater fish (Iheanacho *et al.*, 2017). The lack mortality recorded after feeding juveniles with diet containing 2% of *A. lepidophyllus* compared to the juvenile fed with the control diet could be attributed to the antioxidant properties of the phyto-additive contained in the feed. Fogang *et al.* (2014) demonstrated that the bark of *A. lepidophyllus* is rich in polyphenolics compounds that exhibit notable antioxidant properties. Shahidi and Hossain (2018) indicate that spices play a significant role in the aquaculture sector, enhancing not only the taste of feed and serving as flavouring agents but also due to their rich antioxidative properties. The primary bioactive compounds found in spices include a wide variety of elements, such as terpenoids, flavonoids, phenolic compounds, saponins, glycosides, and other bioactive substances (Parthasarathy et al., 2008). So, the lack of mortality recorded in T_3 treatment supplemented with 2% *A. lepidophyllus* compared to other treatments could be attributed to the high concentration of the bioactive molecules mentioned contained in the Phyto-additive used. The increase in the quantity of bark of *A. lepidophyllus* in the feed increases the concentration of bioactive compounds such as polyphenolics compounds contained in these barks, which implies an increase in antioxidant properties, free radical scavenging and reduce oxidative stress (Moukette *et al.*, 2015). The nutritional condition consequently enhances health and enables the fish to perform more effectively. This observation corroborates the previous research works conducted by Nyadjeu *et al.* (2021) whereby it was noted that the fry fed the experimental food containing the highest amount of *Allium Sativum* (2%) had the highest survival rate. The physical and biological conditions and fluctuations resulting from the interaction of feeding conditions, parasitic infestations, and physiological parameters are indicated by a fish's condition factor (K) (Le Cren, 1951). This also reflects the variations in food reserves, serving as a measure of the overall health of the fish population. Additionally, K is an index that assesses fish health, grounded in the premise that fish of greater weight at a specific length are in superior condition (Bagenal and Tesch, 1978). Results in this study showed that, fish fed diet supplemented with 2% *A. lepidophyllus* obtained significantly ($P < 0.05$) high values of K (1.06 ± 0.01), compared to the other diets. According to Fulton (1902), $K \geq 1$ expresses the wellbeing of a population during the varying stages of its life cycle; while K<1 signifies that the fish is not in good health in its biotope. The wellbeing of *C. gariepinus* was better express with juveniles fed T_3 diet (2% *A. lepidophyllus*) compared to the other treatments groups. This result is in contradiction with that obtained by Stanley *et al.* (2018) who recorded K<1 with *C. gariepinus* juvenile fed on diet containing ginger at 0% (control), 0.5% (T_1), 1.0% (T_2), 1.5% (T_3) and 2.0% (T_4) on 70 day of rearing period. This result contradicts those reported by Nyadjeu *et al.* (2021a; 2021b) who found K<1 for *C. gariepinus* fry fed on diet containing ginger, garlic and ginger-garlic blend respectively at 0% (control), 1% (T_1), 2% (T_2) (ginger), 1% (T_3), 2% (T_4) (garlic) and D_0 or control, 50mg (D_1), 100mg (D_2) and 200mg (D_3). This conflicting results of adding dietary ginger, garlic, ginger-garlic blend and *A. lepidophyllus* on the condition factor K of *C. gariepinus* during two stages of their life cycle could be attributed on feeding conditions. The concentration of bioactive compounds in *A. lepidophyllus* bark did, in fact, raise the levels of sulfur and phenylpropanoid (eugenol), which can activate pancreatic enzymes (lipases, amylases, and proteases) and boost the activity of gastric mucosal digestive enzymes (Fogang *et al.*, 2014). Furthermore, polyphenolic substances with antioxidant qualities have been shown to increase lipid metabolism and digestive enzymes (Muneendra *et al.*, 2014; Oben *et al.*, 2010; Fogang *et al.*, 2014). Therefore, these properties of bark of *A. lepidophyllus* could improve feed nutrient utilization of fish, induce overweight, improved "well-being" and general conditions of fish in their biotope. Performance of *C. gariepinus* juvenile, body weight and body weight gain were enhanced with increasing *A. lepidophyllus* levels in the diets. These findings are consistent with those of Adeshina *et al.* (2018), Soosean *et al.* (2010), and Abbasi *et al.* (2017), who showed that the highest levels of dietary clove, *Eugenia caryophyllata*, buds extract in *C. gariepinus*, *Garcinia mangostana* in African catfish, and *Zingiber officinale* powder in common carp and *Cyprinus carpio* diets, respectively, produced the highest final weight and weight gain in fish. In addition to stimulating the digestive enzymes, the antibacterial qualities of their particular active compounds and their effect on gut function may have contributed to the greatest increase in body weight observed in *C. gariepinus* administered a food supplement at 2%. The increased incorporation of *A. lepidophyllus* into the diet has been associated with higher levels of flavonoids and phenolic compounds, which are recognized for enhancing animal performance by modifying the intestinal ecosystem through their antimicrobial properties (Odoemelam *et al.*, 2013). These compounds function by forming complexes with various proteins, disrupting bacterial membranes, rendering certain substrates inaccessible to bacteria, and inactivating bacterial enzymes (Frankič *et al.*, 2009). The alterations in the intestinal ecosystem resulting from their antimicrobial effects may lead to improved nutrient availability for the host, thereby enhancing body weight gain and feed efficiency. This observation aligns with the findings of Frankič *et al.* (2009), who noted that the growth-promoting effects of many herbs and spices stem from their ability to eliminate parasites that impede digestibility and overall growth performance in animals. Hepatosomatic Index (HSI) is associated with the liver energy reserve. High HSI value implies large amount of food availability and favourable conditions (Ogunji *et al.*, 2008). Highest liver weight and hepatosomatic index were recorded with fed T_2 diet (1.5% *A. lepidophyllus*) followed by fish fed diet T_1 (1.0% *A. lepidophyllus*). The HSI values reported in this study differ significantly between treatment groups compared to the control and treatment fed with 2% *A. lepidophyllus*. This signifies that the liver conditions were not stable. It also implies that there was high fat deposition in the liver of *C. gariepinus* fed T_2 diet (1.5% of *A. lepidophyllus*). This result is in contradiction with those of Stanley *et al.* (2018) who reported no significant difference between treatment groups compared to the control. However, they observed the highest HSI value with fish fed 2.0% ginger (T4) and was closely followed by fish fed T_2 diet (1.0% ginger). Ogunji *et al.* (2008) reported HSI values ranging from 3.08g – 3.14%

when they used housefly maggot meal to replace fish meal in diets fed to *Oreochromis niloticus*, but no significant difference between treated groups and the control. ASAT and ALAT enzymes are used as indices of liver damage. Increased enzyme levels in fish have been associated with liver dysfunction or inflammation, leading to the release of these enzymes into the bloodstream due to cellular leakages (Akrami *et al*., 2015; Fawole *et al*., 2016; Ajima *et al*., 2019). Such elevations also signify liver degeneration, necrosis, and cellular destruction resulting from damage (Bhardwaj *et al*., 2010). These enzymes serve as indicators of liver health and help determine whether fish that have been fed supplemented diets may experience hepatotoxicity or harm to liver cells. We found in this study that except for the T_2 treatment (1.5% of *A. lepidophyllus*), the tissue content of ALAT was not significantly different ($P > 0.05$) as compared to the control group. Furthermore, the ASAT levels in the tissues of fish that were given a dietary supplement of *A. lepidophyllus* showed a significant decrease compared to the control group. This suggests that the addition of the powder did not cause any liver dysfunction in the fish. This result could be explained by the bioactive compounds found in *A. lepidophyllus* bark that prevented the fish from infection by triggering immune system and its administration might prevent lipid peroxidation of cell membranes and inhibit the release of foresaid enzymes. Consistent with our findings, Anene *et al.* (2022) documented that the reactions of the above enzymes were significantly reduced in *C. gariepinus* fingerlings fed on dietary supplement of turmeric powder compared with the control. Cholesterol is essential for the absorption of fatty acids from the intestine and their subsequent transportation in the bloodstream or haemolymph. In our experiment, fish fed with *A. lepidophyllus* bark had no significant difference on triglyceride, total cholesterol and cholesterol LDL compared with the control. These findings align with those of Binaii *et al.* (2014) who observed there were no change in the cholestrol and triglyceride levels between treated groups and control group on week 4, whereas they were significantly decreased in H. huso fed on dietary 6% and 12% nettle compared to the other group on week 8. In contradiction, Anene *et al.* (2022) reported reduction in the level of cholesterol in *C. gariepinus* fingerlings fed on dietary supplement of turmeric powder for 60 days. Hypocholesterolaemia has also been reported with Abdel-Tawwab *et al.* (2018) who revealed decreased level of serum cholesterol with elevated *Ocimum gratissimum* leaf extract in the feed of *C. gariepinus*. These contradictory findings may be due not only to the various additives employed but also to the duration of the feeding period. Some authors claim that total proteins are the most importantly indicators of the biochemical nutritional and health status of the fish (Patriche *et al*., 2009). In the present study, at the end of rearing period, total proteins of *C. gariepinus* juveniles not significantly change after feeding with different doses of *A. lepidophyllus*. This result corroborates finding of Vahedi *et al.* (2017) and Anene *et al.* (2022) who observed through the trial no significant variation of total protein respectively on juvenile beluga after feeding with different doses of ginger extract and in *C. gariepinus* fingerlings fed dietary supplement of turmeric powder for 60 days. An albumin blood test is conducted to assess overall health and evaluate the functioning of the liver and kidneys. If the liver is compromised or if an individual is poorly nourished, it may produce insufficient albumin. Decreased levels of albumin can indicate liver or kidney disease, or other underlying medical issues. This paper show that Supplementing *C. gariepinus* with *A. lepidophyllus* increase level of albumin in the tissue. Significant high value of albumin was obtained by fish fed of 1.5% *A. lepidophyllus* (T_2). Moreover, except for fishes fed T_2 diets that obtained significantly low level of globulin in the tissue, no significant different was observed for globulin level between other treatments and control group. The conflicting result have been carried out by Gholipour *et al.* (2014) who reported that, globulin significantly increased in serum, but no significant difference was found in albumin in *Huso huso*. Previous study by Binaii *et al.* (2014) revealed that albumin level was not affected in beluga juvenile fed nettle. Similarly, albumin and globulin had no significant difference in fish fed diet containing ginger extract when compared with the control (Vahedi *et al*., 2017). An increase in urea and creatinine levels suggests impaired kidney function, commonly referred to as renal failure. While the kidneys filter urea into urine, a portion of this filtered urea is reabsorbed and utilized by the body. The analysis of creatinine and urea levels in fish revealed no significant differences between the control group and those receiving dietary supplement *A. lepidophyllus* powder, indicating that the addition of *A. lepidophyllus* powder to the diet does not impact the kidney function of the fish. These findings align with the research conducted by Anene et al. (2022), which also found no significant differences between the control group and *C. gariepinus* fingerlings fed a turmeric powder dietary supplement. In contrast, a reduction in creatinine levels was observed in *C. gariepinus* that were fed a diet containing *Zingiber officinale*, as reported by Olaniyi *et al.* (2020).

6 CONCLUSION

The current study demonstrated that, the inclusion of *A. lepidophyllus* bark powder in the diets of *C. gariepinus* favourably influenced tissue biochemical components, survival rate, growth performances, condition factor and reduced the cost of production. The findings also indicated that incorporating *A. lepidophyllus* bark powder as a feed additive in the diet of *C. gariepinus* at a 2% inclusion rate does not negatively impact the physiological functions of the fish.

7 ACKNOWLEDGMENTS

The authors thank Agro-ecological Farm of Bilone at Obala Lekié Division, Center Region of Cameroon for providing the technical installations.

Disclosure statement : No potential conflict of interest was reported by the author(s).

Author's Contributions : YEMDJIE MANE Divine Doriane, ZANGO Paul, NGOUANA TADJONG Ruben, NANHOU Raïssa Linda, went to the field to carry out the research and collect the samples. YEMDJIE MANE Divine Doriane, POUOMOGNE Victor, TOMEDI EYANGO Minette supervised the overall research work. YEMDJIE MANE Divine Doriane and NANHOU Raïssa Linda wrote the first draft before being revised by EBILE DAYAN Agwah, TSAMBOU MEGNIMEZA and NGOUANA TADJONG Ruben and approved by all the authors.

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