

Original Research Article

DIFFERENT DRYING METHODS AFFECTS THE PROXIMATE, NUTRITIONAL AND MINERAL COMPOSITION OF *Clarias Gariepinus* (CATFISH)

ABSTRACT

Aim: This study investigated the effect of modern drying methods on the proximate, nutritional, and mineral composition of *Clarias gariepinus* in Port Harcourt, Nigeria. **Methods:** Fresh *C. gariepinus* were purchased from Mile 3 Market in Port Harcourt, Rivers State. Samples were dissected using a sharp knife, intestinal components, and gills were removed and the edible portion was washed properly and rinsed with distilled water. Samples were treated in four groups: fresh, smoking using firewood, electric drying (200°C), and non-electric drying (200°C for 30 minutes) methods. Samples were blended using mortar and pestle into powder and kept in an air-tight container prior to analyses. Proximate composition was determined by standard methods, mineral concentration was done by Atomic Absorption Spectrophotometry and antinutrient composition was determined by Ultra Violet (UV) spectrophotometry. **Results:** Proximate composition of the sample had the following ranges: crude protein-23.01± 0.00% to 52.16±0.01% (fresh and electric drying respectively); lipid content-8.18±0.00 to 19.87±0.00 (non-electric drying and electric drying methods respectively); Carbohydrates-0.59±0.01% to 0.90±0.00 (non-electric drying and smoking respectively); ash-0.76±0.00% to 1.95±0.00% (electric drying and smoking respectively); Moisture-38.84±0.00% to 74.03±0.01 (electric drying and fresh respectively) and fibre-25.70±0.66% to 65.16±0.01% (electric drying and fresh respectively). Anti-nutrients ranged as follows: flavonoids-2.63±0.00% to 3.252±0.00% (electric drying and smoking respectively); oxalate-0.34±0.03% to 1.147±0.01% (non-electric drying and fresh respectively) and phytate-1.98x10⁻⁴±0.01 to 2.03x10⁻³±0.00% (non-electric drying and smoking respectively). Mineral composition ranged as follows: potassium-247.9±4.086mg/kg to 744.4±11.22mg/kg and sodium-131.9±1.515mg/kg to 938.8±15.12mg/kg for smoking and non-electric drying respectively while calcium was 19.83±0.29mg/kg to 184.1±8.19mg/kg (fresh and electric drying respectively). **Conclusion:** Modern drying methods had a significant effect on the proximate component, flavonoids, and oxalate composition of *C. gariepinus*.

Keywords: *Clarias Gariepinus*, Catfish, Proximate, drying methods, antinutrient, minerals Composition

INTRODUCTION

All over the world, fish has been widely used as an excellent source of animal protein and other nutrients [1]. Fish is regarded as the cheapest source of protein, essential vitamins and minerals among the urban and rural populace. The demand for fish as a source of protein increases as a source of protein increase as the human population grows. In an attempt to improve fish supply as protein source, there has been a tremendous increase in the development of fish farming [2]. Many people in developing nations depend on fish for both their food and their income. According to Adeyeye *et al.* [3], Nigeria's aquaculture fish production has witnessed a steady

increase since the 1950s feeding and employing millions of citizens. The availability of food in the ecosystem, fertilization, feed composition, feeding rates, aquatic environment water quality, and, of course, the method of processing and preservation all have an impact on fish quality, which is an important factor in the fish supply value chain [4]. Fish is subjected to various processing methods including cooking, canning, drying (electric and non-electric), smoking, freezing, and drying. Proteins, fats, vitamins, minerals, and sensory qualities like color, flavor, texture, and overall appearance are the most important components of fish that can be altered by processing methods. The treatment temperature and duration determine the extent of these changes. The final quality of fish and fishery products is impacted by all of these changes, including chemical, physical, and nutritional factors [1]. In a study Al-Reza *et al.* [5] showed that sun drying, smoking, freezing, and canning affected the proximate, biochemical, and microbiological characteristics of fish.



Figure 1: Fresh *Clarias gariepinus* (Catfish)

Clarias gariepinus (Catfish) is a primarily freshwater fish that is well-adapted to confined environments and are resistant to manipulation and disease. The African catfish, *Clarias gariepinus* belongs to a family of air-breathing catfishes, *claridae* which is divided into two genera, *Clarias* and *Heterobranchus*. There are over a hundred species in this family occurring naturally throughout most of Africa and the Southern half of Asia. *C. gariepinus* is characterized by naked skin and fairly long dorsal and anal fins (Figure 1). It has string pectoral fins with spines that are serrated on the outer side. It possesses nasal and maxillary barbells and smallish eyes. *C. gariepinus* is extremely nutritious, containing a high concentration of unsaturated fatty acids, vitamins, proteins, and minerals [6]. *C. gariepinus* is widely distributed throughout Africa,

inhabiting tropical swamps lakes, and rivers, some of which are subject to seasonal typing [7]. Consumption of *Clarias gariepinus* has increased rapidly in recent years as a result of its availability, affordability, consistency, and health benefits [6]. *C. gariepinus* is widely consumed in Port Harcourt city and occupies a central position in the cuisines around the city. Due to the likely effects of various processing methods on the final quality of the fish, this study was aimed at determining the effect of traditional smoking, electric drying, and non-electric drying methods compared to fresh *C. gariepinus* samples.

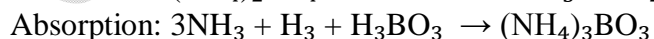
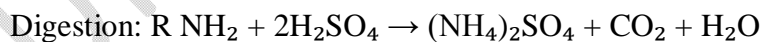
MATERIALS AND METHODS

Sample collection and preparation

Fresh *Clarias gariepinus* (Catfish) species were purchased from Mile 3 Market in Port Harcourt, Rivers State. These were conveyed to the laboratory in Zip-lock bags for further analysis. Samples were processed (by placing on a dissecting board, dissecting with a sharp knife and removing intestinal components and gills, washing with tap water and rinsing with distilled water) and divided into four groups, one group was assessed fresh, while the second group was dried by local drying method (using firewood). The third group was dried using electric air fryer at temperate 200°C and the fourth group was dried using the nonelectric air dryer at temperature of 200°C for 30 minutes. The samples were blended using mortar and pestle into powdery form and kept in an air tight container, carefully labeled and stored pending the analysis.

Determination of protein composition

The compositions of protein in the samples were determined by Kjeldahl Method. The principle of the method is based on the following reactions:



The Kjeldahl method starts with digestion: Sample was weighed into a clean conical flask 250mL capacity, 3grams of digestion catalyst was added into the flask and 20mLs concentrated sulphuric acid was also added and the sample was heated to digest, the content from black to

sky- blue-coloration. The digest was cooled to room temperature and was diluted to 100mL with distilled water. This was followed by distillation: 20mLs diluted digest was measured into a distillation flask and the flask was held in place on the electro thermal heater or hot plate. The distillation flask was attached to a Liebig condenser connected to a receiver containing 10mLs of 2% boric acid indicator 10mLs of 41% sodium hydroxide was injected into the digest via a syringe attached to the mono-arm steelhead until the digest became strongly alkaline. The mixture was heated to boiling and the distilled ammonia gas through the condenser attached to the receiver beaker. The color of the boric acid change from purple to greenish as ammonia distillate was introduced into the boric acid. Finally, titration concludes the process: the distillate was titrated with standard 0.1N Hydrochloric acid solution back to purple from greenish. The volume of hydrochloric acid added to effect this change was recorded as titer value. Percent organic nitrogen was calculated by:

$$\% \text{ Nitrogen} = \frac{\text{Titer value} \times 1.4 \times \text{total volume of digest} \times 100}{1000 \times \text{weight of sample (g)} \times \text{aliquot volume(mL)}}$$

The percent crude protein was then calculated from the percent nitrogen obtained as follows:

$$\% \text{Crude Protein} = \% \text{nitrogen} \times 6.25$$

Where;

Titer value = the volume of HCl used in titrating the ammonium distillate; 1.4 = Nitrogen equivalent to the normality of HCl used in the titration 0.1N; 100 = percentage factor; 1000 = conversion factor from gram to milligram; 6.25= nitrogen factor [8]

Determination of Carbohydrate

Carbohydrates composition was determined by Cleg Anthrone method. 0.1g of the sample was weighed into a 25mLs volumetric flask, 1mL distilled water and 1.3mLs of 62% perchloric acid was added and shaking for a period of 20mms to homogenize completely. The flask was made up to 25mLs mark with distilled water and stopper. The solution formed: was filtered through a glass filter paper or allowed to sediment and decanted, 1mL of the filtrate was collected and transferred into a 10mL test tube this was diluted to volume with distilled water, 1mL of working solution was pipette into a clean test tube and 5mLs Anthrone reagent was added, 1mL distilled water and 5mLs Anthrone reagent was mixed. The whole mixtures were read at 630nm wave length using the 1mL distilled water and the 5mLs Anthrone reagent prepared as blank. Solution

glucose of 0.1mL was also prepared and was treated as the sample with Anthrone reagent. Absorbance of the standard glucose was read and the value of carbohydrate as glucose was calculated using the formula below:

$$\% \text{ Carbohydrate (as \% glucose)} = \frac{\text{Dilution factor} \times \text{Absorbance of sample}}{\text{Absorbance of standard glucose} \times \text{Weight of sample}}$$

Determination of Moisture (Air Oven Method)

1 gram of the sample was weighed into a clean dried porcelain evaporating dish. This was placed in an oven to maintain a temperature of 105 for six hours. The evaporating dish was cooled in desiccators room temperature then it was re-weighed and recorded. Percentage moisture was determined by the formula below:

$$\% \text{ Moisture} = \frac{\text{Weight of fresh sample} - \text{Weight of dried sample}}{\text{Weight of sample}} \times \frac{100}{1}$$

Determination of Lipid (Soxhlet Extraction Method)

2g of sample was inserted into a filter paper and was placed into a Soxhlet extractor. The extractor was placed into a pre-weighed dried distillation flask. Then the solvent (acetone) was introduced into the distillation flask via the condenser end attached to the Soxhlet extractor. The set-up was held in place with a retort stand clamp. Cooled water jet was allowed to flow into the condenser and the heated solvent was refluxed as a result. The lipid in the solvent chamber was extracted in the process of continuous refluxing. When the lipid was observably extracted completely from the sample under test, the condenser and the extractor was disconnected and the solvent was evaporated to concentrate the lipid. The flask was the fluffed in the air oven to constant weight and re-weighed to obtain the weight lipid. Percent lipid was calculated as follows:

$$\% \text{ Lipid} = \frac{(\text{Weight of lask} + \text{Extract}) - \text{Weight of empty lask}}{\text{Weight of sample extracted}} \times \frac{100}{1}$$

Determination of Ash

The percentage ash was determined by Furnace Method. One gram (1g) of the dried sample was weighed into a porcelain crucible which was previously preheated and weighed. The crucible was inserted into a muffle furnace and regulated to a temperature of 630°C for three hours and allowed to cool to room temperature and reweighed. Percent ash was calculated thus:

$$\% \text{ Ash} = \frac{(\text{weight of crucible} + \text{ash sample}) - \text{weight of empty crucible}}{\text{Weight of sample}} \times 100$$

Determination of Crude Fibre

One gram (1g) of the sample was extracted with petroleum ether. A weighted amount of sample (W1) was boiled under reflux for 30 minutes with 200mL of dilute hydrochloric acid and filtered. The residue was properly washed with water until it becomes acid-free. The residue was then transferred into a beaker and boiled for 1 hour and 30 minutes. The residue was then washed 3 times with 10mLs ether. The residue was dried in an oven and cooled then weighed (W2). The dried residue was transferred into a furnace and ignited, cooled, and weighed (W3). Percent crude fibre was calculated as follows:

$$\% \text{ Crude Fibre} = \frac{W2 - W3}{W1} \times 100$$

Determination of Anti-Nutrients

The anti-nutrients were determined by UV-spectrophotometry.

Phytate concentration: Phytate was extracted using 0.5M HCl (100mL) and the extract was made up to 50mL with distilled water. Drops of phenolphthalein were added to 15mL of extract and neutralized with 0.5mL NaOH (purple colouration is observed) and made slightly acidic with 0.17mL HCl (mixture turns colourless). The mixture was then made up to 50mL with distilled water. 4mL FeCl₃ was added to an aliquot (10mL) of the extract and added into a centrifuge tube and heated for 15mins at 100°C. The mixture was cooled, and centrifuged and the residue was washed with 2mL of 0.5M HCl, centrifuged and the supernatant decanted. 2mL water was added to the residue and boiled for 3 minutes at 100°C. This was followed by the addition of 2mL 0.5M NaOH and heated for 15 minutes and filtered into a conical flask. Thereafter, the filter paper was washed with hot water and retained in the flask. 0.5mL H₂SO₄ was added and the setup was boiled down to white fume. After cooling to room temperature, 3mL of conc. nitric acid and 1mL perchloric acid was added to the white fume and further heated to a colourless solution in an electrothermal heater and cooled to room temperature after digestion. This digestion was made up to 50mL of distilled water. The procedure for colour development was as follows: 1mL of the diluted digest was pipetted into a 10mL test-tube and 0.58mL of combined reagent was added. This was made up to 5mL with distilled water. A blank solution of distilled water and combined reagent was prepared as was above. The solution was allowed to develop a blueish colour within

3mins intervals A standard phosphorous conc. Was also prepared in the same manner, and developed as in the analysis of phosphorous in the sample. These solutions were read as 700nm in a spectrophotometer (Jenway 6305) using blank as a calibrating solution. The concentration of phytate was extrapolated from the standard phosphorous graph plotted.

Oxalate: Five grams (5g) of the sample was weighed and 100mL of distilled water was added to it. It was heated for 1 hour and allowed to cool. The solution was made up to 100mL mark with distilled water. 25mL of the sample was transferred into a clean distillation flask. 20mL of 2M H_2SO_4 was added to it. The sample was heated at a temperature of 70°C after which it was allowed to cool. After cooling, the sample was titrated with $KMnO_4$ until the colour changed. 25mL of oxalic acid was measured into another flask and was treated as Oxalate standard. The absorbance of the solution was read at 600nm in a spectrophotometer (Jenway 6305) using blank as a calibrating solution.

Total flavonoid: The total flavonoid content was measured by aluminum chloride ($AlCl_3$) according to the spectrophotometric method using quercetin as a standard. 0.5 mL of the extract was added in the test-tube followed by the addition of 1.5mL methanol, 0.1mL of 10% aluminum chloride, 0.1 mL of 1M potassium acetate and 2.8mL of distilled water respectively. The test-tube tube was left at room temperature for 30 minutes after which the absorbance of the reaction mixture was measured at 415 nm using a UV/ Visible spectrophotometer [9].

Determination of mineral composition

The mineral contents were determined using Atomic Absorption Spectrophotometry. Calcium, potassium, and sodium ion concentration were determined using Atomic Absorption Spectrophotometry. Samples were digested following standard procedure. The digested samples were made up to 50mL and the appropriate wavelength was selected (422nm for calcium, 766nm for potassium, and 589nm for sodium). The concentration of the particular metal was automatically displayed on the equipment screen after following all machine procedures.

Ethical Consideration

This study was carried out in accordance with the guidelines for the care and use of animals and in compliance with the University Research Ethics Committee's fundamental principles for animal-based research.

Statistical analysis

All the values are expressed as mean \pm standard error of the mean (mean \pm SEM). The data obtained from the analyses were subjected to statistical analysis of variance (ANOVA) using SPSS[®] (Version 21) and the means were separated and compared at $p < 0.05$.

RESULTS AND DISCUSSION

The results of the study are shown in table 1-3 below.

Table 1: Proximate composition (%) of *Clarias gariepinus*

Parameter	Processing method			
	Fresh	Smoking	Electric drying	Non-electric drying
Protein	23.01 \pm 0.00 ^a	32.72 \pm 0.00 ^b	52.16 \pm 0.01 ^c	43.40 \pm 0.00 ^d
Lipid	9.58 \pm 0.00 ^a	16.56 \pm 0.15 ^b	19.87 \pm 0.00 ^c	8.18 \pm 0.00 ^d
Carbohydrates	0.70 \pm 0.01 ^a	0.90 \pm 0.00 ^b	0.84 \pm 0.01 ^c	0.59 \pm 0.01 ^d
Ash	1.51 \pm 0.01 ^a	1.95 \pm 0.00 ^b	0.76 \pm 0.00 ^c	0.89 \pm 0.00 ^d
Moisture	74.03 \pm 0.01 ^a	69.84 \pm 0.00 ^a	38.84 \pm 0.00 ^b	61.29 \pm 0.01 ^c
Fibre	65.16 \pm 0.01 ^a	48.02 \pm 0.00 ^b	25.70 \pm 0.66 ^c	46.93 \pm 0.01 ^d

Values are expressed as mean \pm Standard Error of the Mean (n=3); Values in the same row bearing different superscripts letters are significantly different ($p < 0.05$).

Table 2: Antinutrient composition (%) of *Clarias gariepinus*

Antinutrient	Processing method			
	Fresh	Smoking	Electric drying	Non-electric drying
Flavonoids	3.11 \pm 0.00 ^a	3.252 \pm 0.00 ^a	2.89 \pm 0.33 ^a	2.63 \pm 0.00 ^a
Oxalate	1.147 \pm 0.01 ^a	1.046 \pm 0.01 ^b	0.88 \pm 0.02 ^c	0.34 \pm 0.03 ^d
Phytate	4.54x 10 ⁻⁴ \pm 0.01 ^a	2.03x10 ⁻³ \pm 0.00 ^b	7.77x10 ⁻⁴ \pm 0.01 ^c	1.98x10 ⁻⁴ \pm 0.01 ^d
Phytate(mg/kg)	4.46.51 \pm 0.24 ^a	20.28 \pm 0.62 ^b	7.44 \pm 0.70 ^c	1.98 \pm 0.00 ^d

Table 3: Mineral composition (mg/kg) of *Clarias gariepinus*

Mineral(mg/kg)	Processing method			
	Fresh	Smoking	Electric drying	Non-electric drying
Potassium	269.8±28.70 ^a	247.9±4.09 ^a	359.1±2.24 ^b	744.4±11.22 ^c
Sodium	306.0±1.89 ^b	131.9±1.52 ^a	370.6±0.90 ^c	938.8±15.12 ^d
Calcium	19.83±0.29 ^a	42.24±1.69 ^b	184.1±8.19 ^d	59.11±1.47 ^c

The proximate composition of *C. gariepinus* (Table 1) showed that crude protein concentration was as follows: 23.01±0.00% (fresh), 32.72±0.00% (smoking), 52.16±0.01% (electric drying and 43.40±0.00% non-electric drying methods. An increase in protein content was due to the aggregation of proteins after the removal of water molecules present in proteins during drying [10]. This result is in agreement with that of Immaculate *et al.* [11] and Chukwu [12] who both reported that protein content increased when dried with various methods. The variation in protein content observed may be due to the methods of processing fish. The lipid content of smoked and electric-dried *C. gariepinus* was significantly higher compared to the fresh sample. However, there was no significant difference between the lipid content of fresh *C. gariepinus* sample and non-electric dried sample. A decrease in the lipid content observed when dried with the non-electric method is in consonance with the report of Chili and Saha, [13] as they concluded that lipids may exude with moisture evaporation and extended heat treatment may lead to the removal or decrease in lipid content. There was a significant difference in the carbohydrate content of the fish. The carbohydrate content increased in the smoking and electric drying methods but decreased in the non-electric drying method. The concentration of carbohydrates was generally low compared to other nutrients and this could be attributed to the fact that fish is considered a proteinous food [14]. The ash content increased only in the smoking method but decreased in both the electric dryer and non-electric dryer methods. The increase in ash content in the smoker fish could be due to the accumulation of dust or other foreign bodies. High level of ash could be a reflection of the level of salt uptake. Kumolu-Johnson *et al.* [15] reported that during smoking, organic matter is removed and inorganic material will remain as ash. This result is in agreement with that of Mustapha *et al.* [16] who reported that an increase in ash content in smoked fish could be due to a substantial loss of moisture. Expectedly, fresh samples had the highest moisture content (74.03±0.00%) while the least was observed in the electric dried sample (38.84±0.00%).

The decrease in moisture content may be due to the loss of free water present in the fish due to the ingress of heat and salt [17]. This result is in tandem with the research of Immaculate *et al.* [11] as they reported a decrease in moisture content when dried with an electric device. An increase observed in the smoking method could be that the catfish sample was not properly smoked or no proper heat was applied.

The percent anti-nutrient composition of *C. gariepinus* subjected to different methods of processing (Table 2) smoked samples had the highest flavonoid concentration ($3.252 \pm 0.00\%$) which was not significantly higher ($p > 0.05$) than the flavonoid content of electric dried *C. gariepinus* (2.89 ± 0.33). Electric and non-electric drying methods did not affect the flavonoids composition of *C. gariepinus* samples. According to Karamać [18] and Adamczyk *et al.* [19] flavonoids can chelate metals like zinc and iron, reducing their absorption, inhibiting digestive enzymes and possibly precipitating proteins.

The results of this study showed that there was a significant decrease ($p < 0.05$) in percentage of oxalate in *C. gariepinus* samples after subjection to various drying methods compared to the fresh fish. Oxalates have been implicated in causing salts insolubility by interfering with divalent cations like calcium [20]. Study by Lo *et al.* [21], revealed that oxalate compounds are a powerful organic acid that can bind to minerals like sodium or potassium to form salts that dissolve in water, as well as calcium, iron, or zinc to form salts that do not dissolve in water. According to Popova and Mihaylova [22], calcium oxalate can have a deleterious effect on human nutrition and health by accumulating kidney stones. From the findings of this study, a significant increase ($p < 0.05$) in percent phytate concentration percentage was observed in samples subjected to the various processing methods with the exception of non-electric drying method which resulted in a significant decrease ($p > 0.05$) in phytate as compared with the fresh fish. Study by Raboy *et al.* [23], revealed that fish, pigs, and poultry are unable to digest phytate, and feeding grain-based diets causes phytate to be excreted, which in turn contributes to the accumulation of phytate in water and soil. Due to its strong chelation of Ca, Fe, and Zn into insoluble complexes that are not absorbed and can contribute to Fe and Zn deficiency, phytate has antinutritional effects in human and monogastric animal diets [23].

The levels of selected minerals (Table 3) showed varying concentrations of potassium, sodium and calcium for the various processing methods. Concentration of potassium in fresh and smoked *C. gariepinus* were low compared to samples dried using electric dryer and non-electric dryer.

There was no significant difference ($p < 0.05$) between fresh and smoked fishes. Potassium is good for health and deficiency can lead to muscle weakness, paralysis and mental confusion, and acidosis [24]. Concentration of sodium in fresh *C. gariepinus* samples was highest in non-electric dried samples and lowest in smoked samples; concentration of sodium differed significantly ($p < 0.05$) across the various processing methods. Sodium is a major cation of extracellular fluids which aids in maintenance of osmotic pressure, water balance, acid-base balance and a buffer constituent; sodium deficiency can lead to dehydration, acidosis whereas excess sodium leads to edema and hypertension [24]. Concentration of calcium differed significantly ($p < 0.05$) across the various processing methods however the highest concentration of calcium was observed in electric dried samples and the lowest was in fresh sample. According to Vansudevan, *et al.* [24] calcium is a major constituent of bone and teeth, blood clotting, regulation of nerve, muscle and hormone function. Regulation of enzyme activity, lack of calcium can cause tetany, muscle cramps, convulsion, osteoporosis and rickets so normal level of calcium in the body is very much needed.

CONCLUSION

The findings of this study indicate that proximate composition *C. gariepinus* samples subjected to various methods of drying still had standard proximate composition with minimal effects especially on protein, carbohydrates and lipid composition. Antinutrient composition also showed no significant effect based on the modern methods of processing with the exception of oxalates. The mineral composition of *C. gariepinus* showed that it is a good source of potassium, sodium and calcium and the various processing methods of this study had no significant effect on mineral concentration.

REFERENCES

1. Abraha, B., Admassu, H., Mahmud, A., Tsighe, N., Shui, X. W., and Fang, Y. (2018). Effect of processing methods on nutritional and physicochemical composition of fish: a review. *MOJ Food Process Technology*, 6(4), 376-382.
2. Abdel-Gaber, R., El Garhy, M., and Morsy, K. (2015). Prevalence and intensity of helminth parasites of African catfish *Clarias gariepinus* in Lake Manzala, Egypt. *Advances in Bioscience and Biotechnology*, 6(07), 464.

3. Adeyeye, S.A.O., Oyewole, O.B., Obadina, A.O., Omemu, A.M., Adeniran, O.E., Oyedele, H.A., and Abayomi, S.O. (2015). Quality and safety assessment of traditional smoked fish from Lagos State, Nigeria. *International Journal of Aquaculture*, 5. 1927-5773
4. Abeni, A. A., Ibiyinka, O., and Funmilayo, C. F. (2015). Effect of processing on the nutritive value of *Clarias gariepinus* from Isinla fish pond, Ado Ekiti, Nigeria. *American Journal of BioScience*, 3(6), 262-266.
5. Al-Reza, S., Karmaker, S., Hasan, M., Roy, S., Hoque, R., and Rahman, N. (2015). Effect of traditional fish processing methods on the proximate and microbiological characteristics of *Laubuka dadiburjori* during storage at room temperature. *Journal of Fisheries and Aquatic Science*, 10(4), 232.
6. Hesham, A.E.L., Kaur, T., Devi, R., Kour, D., Prasad, S., Yadav, N., ... and Yadav, A. N. (2021). Current trends in microbial biotechnology for agricultural sustainability: conclusion and future challenges. In *Current trends in Microbial Biotechnology for Sustainable Agriculture* (pp. 555-572). Springer, Singapore.
7. Olojo, E. A. A., Olurin, K. B., Mbaka, G., and Oluwemimo, A. D. (2005). Histopathology of the gill and liver tissues of the African catfish *Clarias gariepinus* exposed to lead. *African Journal of Biotechnology*, 4(1), 117-122.
8. Horwitz, W. (2010). Official methods of analysis of AOAC International. Volume I, agricultural chemicals, contaminants, drugs/edited by William Horwitz. Gaithersburg (Maryland): AOAC International, 1997.
9. Win, A., Nyo, A. M. T. and Lwin, K. M. (2019). Determination of Total Flavonoid Content of Commonly Consumed Commercial Tea. *International Journal of Scientific and Research Publications*, 9(5): 802-805.
10. Ninaw, U. and Ratnakumar, U. (2008). Traditional drying methods. *Research and Food Science*, 10(5):148-156.
11. Immaculate, J., Sinduja, P., and Jamila, P. (2012). Biochemical and microbial qualities of *Sardinella fimbriata* sun dried in different methods. *International Food Research Journal*, 19, 1699–1703.

12. Chukwu, O., and Shaba, I. M. (2009). Effects of Drying Methods on Proximate Compositions of Catfish (*Clarias gariepinus*). *World Journal of Agricultural Science*, 5, 114–116.
13. Chili, O. and Saha, B. (2009). Fish processing technology. *Indian Journal of Fish.* 9(1):156161
14. Olusanya, J. O. (2008). Essentials of food and nutrition. 2nd Edn., Nigeria: Apex Books Limited. ISBN-9789784804554
15. Kumolu-Johnson C.A, Aladetohun N.S and Ndimele P.E (2010). The effects of smoking on the nutritional qualities and shelf-life of *Clarias gariepinus* (LACEPEDE). *African Journal of Biotechnology*, 9 (1), 073-076
16. Mustapha, P., Howgate, P. A., Johnson, P. and Whittle, K. J. (2012). Multilingual guide to EC preservation method of fish drying augment the quality and the shelf life. *Academic Press*, 7(1), 30-42.
17. Guan, W., Tarrio J, Jafor, M. and Ashra, M. (2013). Methods of preserving fish. *Journal of Food Science*, 7(3), 78-83.
18. Karamać, M. (2009). Chelation of Cu (II), Zn (II), and Fe (II) by tannin constituents of selected edible nuts. *International journal of molecular sciences*, 10(12), 5485-5497.
19. Adamczyk, B., Simon, J., Kitunen, V., Adamczyk, S., and Smolander, A. (2017). Tannins and their complex interaction with different organic nitrogen compounds and enzymes: old paradigms versus recent advances. *ChemistryOpen*, 6(5), 610-614.
20. Gemedede, H. F., and Ratta, N. (2014). Antinutritional factors in plant foods: Potential health benefits and adverse effects. *International journal of nutrition and food sciences*, 3(4), 284-289.
21. Lo, D., Wang, H. I., Wu, W. J., and Yang, R. Y. (2018). Anti-nutrient components and their concentrations in edible parts of vegetable families. *CAB Rev*, 13(15), 10-1079.
22. Popova, A., and Mihaylova, D. (2019). Anti-nutrients in plant-based foods: A review. *The Open Biotechnology Journal*, 13(1).
23. Raboy, V., Young, K. A., Dorsch, J. A., and Cook, A. (2001). Genetics and breeding of seed phosphorus and phytic acid. *J. Plant Physiol.* 158:489-497.

24. Vansudevan, D. M., and Sreekumari, S. Kannan Vaidyanathan. 2011. Textbook of Biochemistry for Medical Students. 6th edition. *Jaypee Brothers Medical Publishers (P) Ltd*, New Delhi–110002, India.

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