Nutrients, Inorganic Substances And Microbial Evaluations Of Some Commercially Important Coastal Fish Species

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Abstract
Mineral, proximate, amino acids, microbial and chemical compositions of *Pomadasy Jubelini, Coptodon zillii, Mugil cephalus* and *Cyprinus carpio* were assessed to ascertain the wholesomeness and health risk associated with consuming the most common sort after fish species in Lagos coastal water. A total of twelve fish samples (three specimens each) were collected from different landing sites with the samples iced and taken to the Biochemistry and Pharmaceutical laboratory sections of the College of Medicine University of Lagos, Idi-Araba, Lagos, Nigeria for analysis. The iced samples were maintained in alternate layers of ice until spoilage was noticed. Bligh and dye standard method as described by AOAC was used to evaluate the nutritional and mineral compositions while chemical analysis was conducted using Atomic Absorption Spectrophotometer and Soxhlet Extraction methods. Thiobarbituric Acid (TBA), Acid Value (AV) and Peroxide Value (PV) were also determined. Statistical difference between species was done using one way ANOVA with Significance established at ($p<0.05$). The result indicates higher Trimethylamine value Total Enterprise Value (TEV) (11.23±0.13 mg N/kg), PV (5.49±0.01 mg N/kg), Total Volatile Nitrogen (TVN) (27.99±0.25 mg/kg) for *M. cephalus* than other fish samples except for TBA (7.89±0.26 mg/kg) content which was highest in *C. zillii* but within the standard limits. Higher mean mould load recorded were (5.71±0.20), (9.58±0.00), (7.07±0.07) and (5.04±0.02) for *P. Jubelini, C. zillii, M. Cephalus, C. carpio* respectively. The results indicated a notable quality improvement leading to a high quality fish in terms of shelf life, quality and market value. None of the fish samples exceeded the mould load limit for fish thereby exhibited no risk on public health. Therefore the fish are wholesome and safe for human consumption.

Keywords: Amino acids, Chemical, *C. carpio, C. zillii, M. cephalus, ,* Mineral, *P. jubelini* , Proximate,

INTRODUCTION

The increasing rate of contamination and pollution in our environment in recent times has brought about an increased concern about the health status of food consumed. Food products which include sea foods such as fish and fisheries products are the most vulnerable to this effect. One big challenge menacing the human health is the problem of malnutrition and under nutrition particularly in underdeveloped and developing countries.

Fish being one of the most diverse groups of animal known to man have more species than all other vertebrates. It has remain as one of the most important sources of animal protein due to its availability, affordability being relatively cheap compared with other sources of protein such as meat, excellent taste, easy digestibility, lack of cultural or religious taboos associated to its consumption by any particular ethnic group, high content of essential nutrients and unsaturated fatty acids which are relevant for functionality of protein in the body (Elaigwu, 2019). Fish lipids act as important sources of energy for the biochemical activities of cell membranes. It is composed of mono and poly unsaturated fatty acids such as omega three (ω-3) and omega six (ω6) that can help reduce the cholesterol content of the body and the risk of cardiovascular diseases (Sidiq *et al.,* 2021). From the nutritive point of view, studies conducted on proximate composition of fish shows that fish is composed of between 30 to 90% water, 60-75 % protein, 30 to 50% lipids, 0.1 – 1% carbohydrates (Soetan *et al.,* 2017), essential minerals such as sodium, magnesium, calcium, phosphorus, potassium, iodine and appreciable quantity of vitamins such as A, D, E, K (Galtan *et al.,* 2017). These reports may vary depending on the geographical location, season of the year, the feed intake, sex, species, age and maturity or size of the fish (Bouis 2013). One of the most common fish species in Nigeria is *Clarias gariepinus* (African sharp-tooth catfish) which belongs to the family of Claridae found in freshwaters, lakes, rivers and swamps. Other species are Tilapia, which belong to the Cichlidae family, *Hyperopisusbebe occidentallis* (Elephant fish) of the family of Mormyridae, Synodontis of the genus Mochokidae and

Gymnarchus niloticus (Trunk fish) (Jolaoso *et al.,* 2016). This study was aimed at determining the nutritional composition vis-à-vis mineral, proximate, microbial, chemical and amino acids of *P. jubelini, C. zillii, M. cephalus, C. carpio* fish spp found in coastal market of Kosofe in Lagos Nigeria and come up with data on the nutritional composition of *P. jubelini, C. zillii, M. cephalus, C. carpio* of fish spp. found in this location which contributes to the relevance of this study. These species were selected due to their economic importance and consumers' demand thus, make the detailed information about their nutritional composition very important.

MATERIALS AND METHODS Sample collection, preparation and pre-treatment
The fish species used were procured from fishermen at Kosofe landing site in Lagos State, Nigeria. The fish were taken to the laboratory, washed with water and lacerated. The non-edible parts were removed and properly washed again with tap water and rinsed with distilled water. The samples were oven dried at 90°C until a constant weight was obtained. The dried samples were ground and homogenized

Mineral content determination
Procedure for digestion: Five (5) gram of the air-dried and finely pulverized sample was weighed into a 100ml of digestion flask. Five milliliters (5ml) of 6M hydrochloric acid were added. The mixture was heated to dryness on a hot plate set at 150ºC. The dry mass produced was leached with 10ml of 4M HCl, and thereafter filtered into a flask and the residue washed with more HCl and made up to the mark. The blank was prepared in the same manner as described above, but without any fish sample. The extracts and the blanks were analyzed at the Biolife Consults, Lagos State using the atomic absorption spectrophotometer (SCHIMAZU-AA7000).

Atomic Absorption Spectrometry (AAS): The determination of essential mineral was achieved for potassium (K), sodium (Na), Magnesium (Mg), Iron (Fe) and Zinc (Zn) using an Atomic Absorption Spectrophotometer (AAS), Unicam Atomic Absorption – M Series), Unicam Limited, U.K.

Proximate Analysis
For proximate composition, moisture content was determined using the hot air oven, by drying the sample at 105 °C ± 2 °C until a constant weight was obtained. Total lipid was determined by Bligh and Dyer method using chloroform/methanol (1/1, v/v). Crude protein content was determined by converting the nitrogen content obtained by Kjeldahl's method (Nx6.25). Ash content was determined after combustion for 20 hours at 550°. Total carbohydrate was determined by subtracting the sum of fat content, protein content, ash content and moisture from 100. All analyses were carried out on ten different fish.

Determination of Total Carbohydrate by Anthrone Method: For the determination of carbohydrates (CHO), 100 mg of each sample was weighed into a boiling tube. It was then hydrolyzed by keeping it in a boiling water bath for three hours with 5 mL of 2.5 N HCl and cool to room temperature. Solid sodium carbonate was used to neutralize the sample until the effervescence ceases. However 100 ml of the sample was centrifuged. The supernatant was collected, (adding 1 mL of aliquots) for analysis. The volume of the sample was made up to 1ml, distilled water and 4 mL of anthrone reagent was added to it. It was then heat for eight minutes in a boiling water bath. The substance was allowed to cool rapidly before the green to dark green colour was read at 620 nm. A standard graph showing the concentration of the standard on the Xaxis versus absorbance on the Y-axis was plotted. From the graph, the amount of carbohydrate present in the sample tube was calculated.
Amount of carbohydrate present in 100 mg of the sample:

$$= \frac{\text{mg of glucose}}{\text{Volume of test sample}} \quad X\ 100$$

Extraction and measurement of total lipids (Bligh and Dyer method): 10g of wet sample was weighed in a pre-weighed 100 mL- conical flask, 20 mL methanol (MeOH) and 10 mL chloroform ($CHCl_3$) was added. The sample was then homogenized for 2 mins with an UltraTurrax mixer. 10 ml was added a second time. However the mixture was shaked vigorously for 1 min. 18 mL of distilled water was added (including the water already in the sample). The mixture was vortex again for 1 min. The two layers were separated by centrifugation for 10 min at 450 g in a thermostatic centrifugation at 20°C. The lower layer was transferred to a pear-shaped flask with a Pasteur pipette. A second extraction was also done with 20 mL 10% (v/v) MeOH in by vortexing for 2 min. Aftercentrifugation, the lower phase was added to the first extract. A rotavapor was used to evaporate the sample to dryness. Theresidue was further dried at 104°C for 1 h. The extracted weight was recorded hence the lipid content was calculated as described by Breil *et al.,* (2017).

Determination of crude fibre: This method was previously described and used by AOAC (2019). 1.0 g of the finely ground sample was weighed out into a round bottom flask, 100ml of 1.25% Sulphuric acid solution was added and the mixture boiled under a reflux for 30 min. The hot solution was quickly filtered under suction. The insoluble matter was washed several times with hot water until it was acid free. It was quantitatively transferred into the flask and 100 ml of hot 1.25% sodium hydroxide (NaOH) solution was added and the mixture boiled again under reflux for 30 min and quickly filtered under suction. The soluble residue was washed with boiling water until it was base free. It was dried to constant

weight in the oven at 105°C, cooled in a desiccator and weighed (C1). The weighed sample (C1) was incinerated in a muffle furnace at 300˚C for about 30 minutes, cooled in the desiccator and reweighed (C2).

The loss in weight of sample on incineration = C1 - C2 x 100
Weight of original sample% Crude fibre = C1 - C2

Moisture content: The method described by AOAC (2019) was adopted. The method is based upon the removal of water from the sample and its measurement by loss of weight. A clean crucible was weighed and dried in the oven (W1); 1.0 g of each of the samples was weighed into the crucible (W2) and was dried at 105°C, for twenty four hours. The crucible was then transferred from the oven to desiccator, cool and reweighed (W3). The % moisture content was calculated from:

$$\% \text{ Moisture content} = 100 - \frac{w3-w1}{w2-w1} \text{ x } 100)$$

Total ash: The AOAC (2019) method was used. The porcelain crucible was dried in an oven at 100°C for 10 min, cooled in a desiccator and Weighed (W1). Two grams of the sample was placed into the previously weighed porcelain crucible and reweighed (W2) and then placed in the furnace for four hours at 600°C to ensure proper ashing. The crucible containing the ash was removed cooled in the desiccator and weighed (W3).
The % ash content was calculated as:

$$\% \text{ Ash content} = \frac{W2 - W1}{W3 - WI} \quad X\ 100$$

Crude Protein Determination: The micro Kjeldahl method described by AOAC (2019) was used. Two grams of each of the samples was mixed with 10ml of concentrated H_2SO_4 in a heating tube. One tablet of selenium catalyst was added to the tube and mixture heated inside a fume cupboard. The digest was transferred into distilled water. Ten millimeter portion of the digest mixed with equal volume of 45% NaOH solution and poured into a Kjeldahl distillation apparatus. The mixture was distilled and the distillate collected into 4% boric acid solution containing 3 drops of methyl red indicator. A total of 50ml distillate was collected and titrated as well. The sample was duplicated and the average value taken. The Nitrogen content was calculated and multiplied with 6.25 to obtain the crude protein content.
This is given as percentage Nitrogen = $\frac{(100 \text{ x } N \text{ x } 14 \text{ x } VF)\ T}{100 \text{ x } Va}$

Where,
N = Normality of the titrate (0.1N)
VF = Total volume of the digest = 100ml
T = Titre value
Va = Aliquot Volume distilled

Determination of amino acids: The amino acid analyzes of the samples were made duplicate (n = 2) in TUBITAK MAM according to Dimova (2018), and Gheshlaghi *et al*., (2008), using the high-performance liquid chromatography (HPLC) method. This method is based on the reading of ultra-fast liquid chromatography– ultraviolet (UFLC-UV) detector by derivatization with phenyl isothiocyanate and acetonitrile: methanol: triethylamine solution after acidic hydrolysis applied to disassociate the constituent proteins into amino acid components. Tryptophan is completely disappeared as a result of acid hydrolysis; for that reason, tryptophan analyzes were made by base hydrolysis method. The sulfur-containing amino acids immediately expose to degradation when hydrolyzed with a strong acid solution, so sulfur-containing amino acids did not determine. Totally, 20 amino acids (aspartic acid, glutamic acid, serine, glycine, arginine, histidine, threonine, lysine, alanine, proline, leucine, isoleucine, tyrosine, phenylalanine, valine, methionine, and tryptophan) were determined as mg/100 g.

Determination of microbial and chemical profile
Peroxide Value (AOAC Method 965.33): To 1.00 g of the extracted oil in a clean boiling tube was added, 1g of potassium iodide and 10 ml of acetic acid-chloroform (2:1) mixture. The mixture was placed on a hot plate and allowed to boil for 30 sec. The tube was washed twice with 25 ml portions of distilled water and the washings were added to the titration flask. This was then titrated with 0.01N Sodium thiosulphate, using starch as indicator. A blank was carried through the procedure. The peroxide value is calculated as;

$$\text{Peroxide value (mEq/kg)} = Titre(ml) \times 0.01N \times 1000 \div weight of sample extracted$$

Thiobarbituric Acid Reactive Substances (TBARS): 1.0 g of a homogeneous sample was extracted with 5 ml of 50% glacial acetic acid in distilled water, by shaking for 1 h and, thereafter filtered. The filtrate was centrifuged, if necessary, and was used for analyses. The standard MDA solution (1 mL) was taken in a 10 mL test tube and mixed with TBA (1 mL). The mixture was heated in a boiling water bath at 95°C for 60 minutes. The test tubes were cooled at room temperature and absorbance was measured at 532 nm using a spectrophotometer. The extract of each sample (1 mL) was

mixed with 1 mL TBA reagent and the above procedure was repeated. The TBARS was calculated using the calibration curve, from standard solutions of MDA. (Mohammed *et al*., 2019)

Total Phenolic Content: Total phenolic content was determined as gallic acid equivalents (GAE), mg/100 g20µL aliquot of sample extract or gallic acid standard (10-50 mg/L) was mixed with 1.58 ml of water followed by 100 µL of Folin-Ciocalteau's reagent. After vortexingand incubating at room temperature for 8 min, 300 µL of 20% aqueous sodium carbonate solution was added. Samples were vortexed and held at room temperature for 2 h. Absorbance of the blue-colored solution was recorded at 765 nm on a UV-Visible spectrophotometer, using 1-cm cuvets (Umari, 2016).

Total Volatile Basic Nitrogen (TVBN): 5 g of each sample was homogenized in 90 ml of 6% (w/v) trichloroacetic acid solution. The solution was centrifuged at 3000 rpm for 10 minutes and the homogenate filtered through Whitman No. 1 filter paper, and then steam distilled in presence of 5 ml of 10% NaOH solution, using a Kjeldahl-type distillation unit. The distillate was collected in a beaker containing 10 ml of a 4% aqueous boric acid solution and 0.04 ml of methyl red and bromocresol green indicator. TVB-N was then determined by titration of the ammonia, in an aliquot of the distillate.

The quantity of TVBN was determined from the titre of standard 0.1 N sulfuric acid added as follows:

$$\text{TVBN (mg/100g)} = Titre(ml) \times t \times c \times a \times 14 \times 100 \div weight of sample extracted$$

Where:
t = titre for standard solution, a = aliquot (ml) titrated,
c = equivalent concentration of standard
14 = factor for recalculating N,
100 = factor unit (mg/100g) (Mashood *et al.,* 2018)

Microbiological Tests

Sample Pre-treatment: Microbiological tests were carried out on all fish samples using standard methods, briefly 5 g of fish sample were homogenized for 4 min in 90 ml of sterile (0.1%) peptone water. Serial dilutions were made, and 1 ml of all dilutions was plated in a sterile petri dish using sterile pipette and then inoculated in appropriate bacteriological media. The plates were incubated for 1- 3 days at 35-37 °C. (Amarimala *et al.,* 2016).

Enumeration Total Viable Count (TPC) / Aerobic Plate Count (APC): For evaluating total viable counts of the microorganisms, standard pour plate technique was used, with inoculations at dilutions at 10-3, 10-4 and 10-5. Using pour plate method, 0.1 ml aliquots of 10-3, 10-4 and 10-5 dilutions of the inoculum was aseptically transferred, in duplicates, into sterile labelled Petri dishes, with Nutrient Agar. The media and the inoculum were mixed and allowed to solidify, and then incubated at 37 °C for 24-48 h. Thereafter, the colonies were counted for total plate count, and expressed as cfu/g (Signh, 2016).

Statistical Analysis: Data were analyzed and presented using Microsoft Excel 2010. All data collected were analysed for significant differences (p □ 0.05) (ANOVA) on Graph Pad Prism V. The results were expressed as mean ± SD. The determined differences among treatments were partitioned by the Least Significant Difference (LSD) and the Duncan' s New Multiple Post Hoc Test (Duncan, 1955).

RESULTS

The result of the mean concentration obtained from four fish samples. The first section as presented in table 1 showed the proximate composition (Moisture, Carbohydrates, crude fat, crude fibre, Ash and Protein content) from the fish samples. Presented in table 2 is the mineral profile of the fish samples. Table 3 showed the amino acids content of the fish samples. Lastly, table 4 showed the biochemical properties (Trimethylamine TMA, Total volatile basic nitrogen TVBN), lipid oxidation (peroxide, thiobarbituric acid (TBA) contents and microbial contents of the fish samples.

Table 1: Proximate analysis of the fish Samples (n=3)

Fish Species	% Moisture	% CHO	% Protein	% Crude Fiber	% Crude Fat	% Ash
P. jubelini	74.57±0.29^{b}	8.47±0.18^{b}	12.05±0.14^{c}	3.07±0.70^{a}	1.79±0.09^{b}	1.05±0.23^{bc}
C. zillii	69.12±0.20^{ab}	7.20±0.19^{ab}	17.25±0.56^{a}	1.66±0.12^{b}	1.23±0.34^{ab}	3.54±0.02^{b}
M. cephalus	66.45±0.78^{a}	9.66±0.79^{c}	15.21±0.18^{bc}	1.05±0.16^{ab}	5.17±0.19^{b}	2.46±0.67^{c}
C. carpio	71.05±0.10^{ac}	5.82±0.89^{b}	17.85±0.80^{b}	1.92±0.69^{b}	2.28±0.34^{a}	1.08±0.40^{a}

Values in the same column followed by the same letter are not significantly different at p<0.05). Values represent pooled means vertically of replicate determination

While Moisture content is the highest in all the species, crude fibre is found to be generally low in all the species. The table 1 above shows the proximate analysis of the *P. Jubelini, C. zillii, M. cephalus and C. carpio*. The concentration of moisture content, sodium, carbohydrates (CHO), protein, crude fibre, crude fat and Ash content were determined. The

concentration these parameters ranges as follows, Moisture content (66.12±0.78[a] - 74.57±0.29[b]) CHO (5.82±0.89[b] - 9.66±0.79[c]), protein (12.05±0.14[c] - 17.85±0.80[b]), Crude fibre (1.05±0.16[ab] - 3.07±0.70[a]), Crude fat (1.23±0.34[ab] - 5.17±0.19[b]) and Ash content (1.05±0.40[a] - 3.54±0.02[b]).

Table 2: Mineral contents of the fish samples (n=3)

Fish Species	Calcium mg/kg	Sodium	Potassium	Magnesium	Iron	Zinc
P. jubelini	126.55±0.12[c]	125.35±0.24[a]	17.44±0.19[ab]	219.56±0.80[c]	1.57±0.30[b]	2.05±0.00[ac]
C. zillii	57.70±0.15[b]	272.48±0.45[b]	47.09±0.20[c]	352.45±0.48[ab]	4.30±0.97[a]	1.10±0.26[b]
M. cephalus	157.01±0.18[a]	39.05±0.01[c]	250.21±0.12[ab]	401.19±0.20[b]	11.62±0.26[c]	6.90±0.37[c]
C. carpio	102.44±0.34[ab]	66.56±0.06[a]	349.73±0.40[b]	31.85±0.38[c]	1.45±0.29[bc]	7.53±0.28[a]

Values in the same column followed by the same letter are not significantly different at p<0.05).
values represent pooled means vertically of replicate determination

The table 2 above also shows the mineral content of *the P. Jubelini, C. zillii, M. cephalus* and *C. carpio*. The concentration of Calcium, sodium, Potassium Magnesium, Iron and Zinc concentration ranges as follows, Ca (57.70±0.15[b] - 126.55±0.12[c] mg/kg) Na (39.05±0.01[c] - 125.35±0.24[a] mg/kg), K(17.44±0.19[ab] - 349.73±0.40[b] mg/kg), Mg (31.85±0.38[c] - 401.19±0.20[b] mg/kg), Fe (1.45±0.29[bc]-11.62±0.26[c] mg/kg) and Zn (1.10±0.26[b] - 7.53±0.28[a] mg/kg). While Magnesium is the highest in all the species, zinc content is found to be relatively low in all the species.

Table 3: Amino acid contents

Amino acides	*P. jubelini*	*C. zillii*	*M. cephalus*	*C. carpio*
Cysteine	0.45±0.23[a]	1.52±0.59[b]	0.95±0.54[c]	1.89±0.29[a]
Histidine	3.62±0.01[ab]	2.95±0.74[a]	2.90±0.38[b]	3.05±0.66[bc]
Aspartic acid	9.67±0.24[b]	14.05±0.64[c]	12.05±0.19[a]	7.68±0.40[ab]
Methionine	2.81±0.17[c]	4.62±0.39[a]	2.67±0.56[c]	3.23±0.17[b]
Asparagine	3.52±0.16[a]	2.06±0.60[c]	2.30±0.75[b]	5.39±0.68[a]
Threonine	1.25±0.98[b]	5.57±0.21[a]	4.98±0.34[ab]	7.20±0.11[c]
Serine	8.94±0.08[c]	6.12±0.01[a]	4.34±0.47[b]	5.22±0.08[c]
Glutamic acid	10.75±0.20[ab]	14.50±0.90[b]	16.83±0.50[ab]	8.78±0.34[a]
Glycine	5.02±0.03[a]	7.45±0.23[c]	5.70±0.39[b]	12.09±0.00[ab]
Alanine	3.23±0.09[a]	6.20±0.89[b]	6.95±0.26[ac]	4.32±0.38[bc]
Valine	1.81±0.10[c]	10.45±0.20[ab]	7.01±0.22[b]	1.79±0.27[a]
Leucine	5.89±0.50[a]	4.27±0.30[bc]	9.95±0.69[a]	6.50±0.54[b]
Tyrosine	2.98±0.03[b]	3.89±0.18[ab]	4.52±0.21[c]	4.15±0.46[c]
Isoleucine	0.70±0.34[c]	1.11±0.25[bc]	5.34±0.29[b]	4.89±0.31[a]
Glutamine	9.56±0.25[c]	6.12±0.10[b]	8.09±0.39[a]	9.11±0.20[b]
Lysine	5.05±0.19[a]	10.57±0.27[ac]	12.65±0.10[b]	3.00±0.90[a]
Arginine	0.78±0.25[b]	7.25±0.76[a]	7.80±0.76[ab]	3.58±0.50[a]
Proline	0.95±0.26[c]	3.50±0.54[b]	3.18±0.72[b]	1.56±0.04[b]
Phenylalanine	2.40±0.98[a]	7.88±0.17[c]	4.10±0.00[c]	2.50±0.29[c]
Taurine	0.79±0.29[b]	2.10±0.13[ab]	1.03±0.06[a]	5.04±0.26[c]

Values in the same column followed by the same letter are not significantly different at p<0.05). Values represent pooled means vertically of replicate determination.

The total amount of amino content is shown in the table above, the total amino acid content ranged from 80.17±0.05[ab] - 127.18±0.12[c]. *M. cephalus* was found to content the highest content of 127.68±0.12[c] amino acid amongst other species. *P. jubelini* had the lowest amino acid Content of 80.17±0.05[ab]. Both *C. zillii* and *C. carpio* had total amino acids content as 122.18±0.02[b] and 100.97±0.54[a] respectively.

Table 4: Microbial load and chemical content of the fish samples (n=3)

Biochemical Content	*P. jubelini*	*C. zillii*	*M. cephalus*	*C. carpio*
Mould load of Fish (ML)	5.71±0.20[a]	9.58±0.00[ab]	7.07±0.07[c]	5.04±0.02[b]
Thiobarbituric Acids (TBA)	6.50±0.45[ab]	7.89±0.26[b]	7.15±0.19[a]	2.88±0.02[ab]
Peroxide value (PV) mE/kg	5.71±0.10[c]	5.49±0.01[c]	9.65±0.14[a]	7.56±0.18[b]
Total volatile nitrogen mg/100g (TVN)	21.40±0.30[c]	27.78±0.25[a]	27.99±0.11[ab]	12.14±0.70[c]
Trimethylamine value (TEV)	5.72±0.55[b]	7.58±0.20[b]	11.23±0.13[c]	2.70±0.69[a]

Values in the same column followed by the same letter are not significantly different at p<0.05).
Values represent pooled means vertically of replicate determination

The mould load of the fishes ranged from 5.04±0.02[b] - 9.58±0.00[ab] mg/100g, *C. zillii* with the highest mould load of 9.58±0.00[ab] mg/100g, and least mould was found in *C. carpio*. Thiobarbituric acids content was high in *C. zillii* (7.89±0.26[b]mg/100g) but low in *C. carpio* (2.88±0.02[ab]mg/100g). While *C. zillii* show lower value of peroxide values of 5.49±0.01[c] mE/kg, *M. cephalus* showed a high value of the same acid. The total volatile nitrogen content ranged from 12.14±0.70[c] - 27.99±0.11[ab]mg/100g, this parameter showed the higher presence than other parameters (ML, TBA and TEV). TEV was very low in *C. carpio*.

DISCUSSION

The moisture content of *P. Jubelini, C. zillii, M. cephalus, C. carpio* were 74.57%, 69.12% 66.45% and 71.05 respectively. Moisture content is considered the amount of moisture available for microbial activities (Ajai *et al*., 2019), an indication that *P. jubelini* was most probably prone to spoilage than other fish species which could lead to low shelf life and making *M. cephalus* the better species with the longest shelf live. The result of the current study was higher than the result obtained by Agbugui *et al.,* (2013), for *M. cephalus* (63.66%), but was lower than *M. cephalus* (74.74%) reported by Ogundiran *et al.,* (2014). *M. cephalus* (9.66%) showed the highest carbohydrate content and *C. carpio* (5.82%) with lowest carbohydrate content. Carbohydrates provides energy and plays an important role in the human body. They act as an energy source, help control blood glucose and insulin metabolism, participate in cholesterol and triglyceride metabolism, and help with fermentation (Holesh *et al.,* 2021). The carbohydrates result obtained was higher than that reported by Rishiraj *et al.,* (2020). *M. cephalus* can be recommended as a source of energy due to its high carbohydrate concentration. *C. Carpio* on the other hand showed high content of protein (17.85%), while *P. jubelini*, *C. zillii* and *M. cephalus* have 12.05%, 17.25% and 15.21% respectively. Protein repair and build body's tissues, allows metabolic reactions to take place and coordinates bodily functions. In addition to providing body with a structural framework, proteins also maintain proper pH and fluid balance, development and replacement of worn out cells. Protein value of *C. carpio* was higher than value (16.9%) obtained in the research of Skibniewsk *et al.,* (2013). Crude fibre content was discovered to be high in *M. cephalus* (5.17%). High fibre content in the diet aid easy digestion, hence *M. cephalus* was considered suitable for such purpose. The crude fat ranges between 1.23% - 5.17%, the lowest percentage crude fat was obtained in *C. zillii* (1.23%), and the specie containing a highest concentration was *M. cephalus* (5.17%). Crude fat is the term used to refer to the crude mixture of fat-soluble material present in a sample. Crude fat also known as the ether extract or the free lipid content, is the traditional measure of fat in food products. Fish fat contains a high proportion of polyunsaturated fatty acids, which may help to reduce the incidence of atherosclerosis and heart related diseases. Fish reduces vulnerability to hunger by providing a complementary food source as part of diversified livelihood strategies. Ash content refers to the remaining inorganic residue after being ignited or oxidized of organic matter in a given food sample. It is also an important attribute for determining the quality of food ingredients (Ismail, 2017). The ash content of the study ranges from 1.05% to 3.54%. The highest percentage ash content was obtained in *C. zillii* (3.54%). Ash is an important attribute for determining the quality of food ingredients (Ismail, 2017). A significant relationship ($p < 0.05$) exists in all the proximate contents across the samples. The concentration of calcium in fish species ranged from 57.70 to 157.01mg/kg. The concentrations of Calcium in *M. cephalus* (157.01mg/kg), *P. jubelini* (126.55mg/kg), *C. carpio* (102.44mg/kg) were significantly higher than that of *C. zillii* (57.70mg/kg). Alfa *et al.,* (2019) reported a lower concentration of calcium (0.03 -0.14 mg/kg) in freshwater fish sold in Bida Markets (Ajai *et al.,* 2019) than that obtained in this study. The magnesium concentration in the fish species ranged from 31.85 – 352.45mg/kg, this falls within the recommended range of 45-4520 mg/kg (Oko, 2019). The order of magnesium content in different fish species were *M. cephalus* (401.19mg/kg) > *C. zillii* (352.45mg/kg) > *P. jubelini* (219.56mg/kg) > *C. carpio* (31.85mg/kg). These results were similar to those obtained by Hammed and Amosu (2020); Hammed *et al.,* (2020); Hammed *et al.,* (2022a); Hammed *et al.,* (2022b); Amosu *et al.,* (2023); Hammed *et al.,* (2023) which also corroborates the findings obtained by Alfa *et al.,* (2019); Holesh *et al.,* (2022). The order of zinc content in different fish species were *C. carpio* (7.53mg/kg) > *M. cephalus* (6.90mg/kg) > *P. jubelini* (2.05mg/kg) > *C. zillii* (1.10mg/kg). The concentration of iron in fish species ranged from 11.62 to 1.45mg/kg. The concentration of iron in *M. cephalus* (4.30mg/kg), *P. jubelini* (1.57mg/kg), *C. carpio* (1.45mg/kg*)* were significantly lower than that of the *C. zillii,* (11.62mg/kg) while the order of sodium and potassium content in different fish species were *C. carpio* (349.73mg/kg) > *M. cephalus* (250.21mg/kg) > *C. zillii* (47.09mg/kg) > *P. jubelini* (17.44mg/kg) and *M. cephalus* > (157.01mg/kg) > *P. jubelini* (126.55mg/kg) > *C. carpio* (102.44mg/kg) > *C. zillii* (57.70mg/kg) respectively.

The most abundant essential amino acids from the study were lysine and leucine, while aspartic acid, glutamic acid, alanine and glycine were non-essential amino acids. The highest value of amino acid was found in *M. cephalus* and the lowest in *P. Jubelini*. These changes could be as a result of seasonal conditions, age, size, catching area, spawning season and feeding conditions. Fish are known to have high protein content and the abundant amino acids are glutamic acid, aspartic acid, and lysine in aquatic organisms (Jolaoso *et al.,* 2016). In the study conducted by Mohammed and Ahmed (2016), the amino acid contents of three kinds of marine flounder fish (*Hippoglossus hippoglossus*, *Pleuronectes ferruginea*, and *Paralichthys olivaceus*) were studied and they found that the most abundant amino acids are aspartic acid, glutamic acid, glycine, leucine and lysine. Galtan *et al.,* (2017) studied the amino acid contents of *Channa striats*, *Channa micropeltes* and *Channa lucius*, and found that the most abundant amino acids were glutamic acid, aspartic acid, and lysine, thus indicated that these species are rich in essential amino acids for human health and evolution. Report also has it that aspartic acid and glutamic acid are important in enzyme solubility and protecting ionic character in enzyme activity (Ajai *et al.,* 2019). Ajai *et al.,* (2019) also examined the amino acid contents of *Clarias angullaris*, Oreochromis niloticus, and *Cynoglossus senegalensis* species, and identified that glutamic acid, aspartic acid, and leucine were most common

amino acids. They reported that these species are valuable amino acid sources. Sidiq *et al.,* (2021) examined the amino acid content of *Trichurus trichurus* and found that the most abundant amino acids were aspartic acid, glutamic acid, and lysine. Elagwu (2019) studied about the amino acid contents of *Engraulisen crasicolus*, *Pomatomus saltatrix*, *Sarda sarda*, *M. surmelutus*, and *Merlangius merlangus* and found that lysine, leucine, arginine, glutamic acid, and aspartic acid were the most abundant amino acids. Bouis (2013) studied about the amino acid contents of *Clupea harengus*, *Scromber scrombus*, *Trichurus trichurus*, and *Urophycis tenuis* species. They reported that the most abundant amino acids are glutamic acid, aspartic acid, lysine, and leucine. Oko (2019) studied the amino acid contents of some fishery products and found that the amino acid values changes according to species. Baki *et al.,* (2015) compared the amino acid contents of natural and cultured sea bass (*Dicentrarchus labrax*) and they found that the most abundant amino acids were aspartic acid, glutamic acid, leucine, and lysine. Suseno (2015) reported that the most abundant amino acids were glutamic acid, arginine, leucine, and lysine in tuna fish (*Thunnus sp*.). Salma *et al.,* (2016) found that the most abundant amino acids found in mackerel (*Scromber scrombus*) were glutamic acid, aspartic acid, and lysine. The mould load of the fish species shown in Table 4 ranged between 5.04 -9.58. The mould load determined followed the order of *C. zillii* (9.58) > *M. cephalus* (7.07) > *P. jubelini* (5.71) > *C. carpio* (5.04) in order of increasing concentration. Mold (Aflatoxicosis) is a disease that can affect many species of fish, and results when feed contaminated with aflatoxins is eaten by the fish. The aflatoxins are chemicals produced by some species of naturally occurring fungi (*Aspergillus flavus* and *Aspergillus parasiticus*). Factors that increase the production of aflatoxins in feeds include environmental temperature above 27°C (80°F), humidity levels greater than 62%, and moisture levels in the feed above 14% (Juli-Anne, *et al.,*2010). These molds have the potency to cause cancer-causing agents in animals and human. From the result obtained*, C. carpio* has the lesser mold load thereby recommended more for consumption than *C. zillii* with greater mold load. The thiobarbituric acids content in the fish species was observed to be high in *C. zillii* (7.89 mg of malondialdehyde) and low in *C. carpio* (2.88 mg of malondialdehyde) and the other two species values had 7.15mg of malondialdehyde (*M. cephalus*) and 6.50 mg of malondialdehyde (*P. jubelini*). The thiobarbituric acid content is the measure of the oxidation degree of the fish species. In the broader sense, the thiobarbituric acid (TBA) test measures Malonaldehyde (MDA) produced due to the oxidation of fatty acids with three or more double bonds, and it measures other TBA reactive substances such as 2alkenals and 2, 4-alkadienals. Therefore, TBA is also referred to as TBARs (TBA reactive substances). High TBA value in *C. zillii* is an indication of the presence of off-flavors and loss of nutrients, which might be occur even during frozen storage. Hence, *C. carpio* is better recommended for consumption due to it low TBA value. Peroxide value (PV) determined the concentration of hydro-peroxide, the primary oxidation products which are capable of influencing rancidity in fishes during storage. A lower number of peroxide values indicate a good quality and a good preservation status. From the result shown in Table 4, the PV was represented as follows in other of decrease as: *M. cephalus* (9.65) > *C. carpio* (7.56) > *P. jubelini* (5.71) > *C. zillii* (5.49). These suggest that *C. zillii* content low TBA value a proof of a good quality and good preservative status than *M. cephalus* with high TBA value. But *C. carpio* and *P. jubelini* contains a moderately TBA value. The total volatile nitrogen content in *P. jubelini, C. zillii, M. cephalus, and C. Carpio fish* were 21.40, 27.78, 27.99, and 12.14 respectively. *M. cephalus* (27.99) was observed to be the specie with the highest content of total volatile nitrogen, TVN and *C. carpio* (12.14) having the least value of TVN. Total volatile nitrogen (TVN) is a chemical method used to measure fish spoilage. It is a good indicator of fish freshness and used for quality control of fish and these TVNs result in the destructive activities of microorganisms (Castro *et al.,* 2012). Therefore, high TVN value noticed in *M. cephalus* suggests that this specie is prone to spoilage and can lost freshness easy, but presenting *C. carpio* as less susceptible to spoilage and can stay fresher than other species. The trimethylamine (TMA) value ranges from 5.72, 7.58 11.23 and 2.70 representing *P. Jubelini, C. zillii, M. cephalus, and C. carpio*. Trimethylamine (TMA), the metabolic precursor to TMAO, is formed in fish during bacterial spoilage. Trimethylamine is a pungent volatile amine often associated with the typical "fishy" seafood. Its presence in spoiling fish is due to the bacterial reduction of trimethylamine oxide (TMAO) which is naturally present in the living tissue of many marine fish species. From the result of the study, it can be observed that *C. carpio* has less tendency of bacterial spoilage than *M. cephalus* which have more tendency of producing higher pungent volatile amine leading to bacterial spoilage than in the other species

CONCLUSION

The study shows that *M. cephalus* contains more essential minerals, amino acid content, and good microbial and chemical profiles with promising nutritional advantages which can serve as future treatment in combating malnutrition. The results obtained were similar to those obtained from previous works. More so, result obtained for mineral content in *M. cephalus* was found to be within permissible limit than those from *C. carpio, C. zillii* and *P. jubelini*. Thus it is recommended that *M. cephalus* species be adopted as a rich nutritional source for healthy body building and development in animal and human food. It is recommended for food production as an important source of energy, protein and its good shelf life in food industries. *M. cephalus* is highly recommended for consumption to consumers because of the less content of pollutants.

AUTHORSHIP CONTRIBUTION STATEMENT

Amosu A.O and Hammed A.M: Conceptualization, Methodology, Software, Validation, Writing – original draft. **Dalmeida L.O and Sunnuvu T.F**: Formal Analysis, Investigation, Writing – review & editing, Project administration. **Joseph O.O and Oshinowo K. T**: Methodology, Resources, Supervision, Review & editing.

DECLARATION OF COMPETING INTEREST

The authors declare that they have no known competing financial interests or conflict of interest.

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