

Fatty acids composition of two commonly consumed freshwater fishes (*Clarias gariepinus* and *Chrysichthyes nigrodigitatus*) from the Cross River at Ahaha, Obubra, Nigeria.

Abstract

Fish oils are good sources of essential fatty acids beneficial to human health by helping the body in preventing diseases and enhancing proper growth and development. This study is aimed at investigating the quantitative and qualitative composition of fatty acids in two commonly consumed and most abundant freshwater fishes (*Clarias gariepinus* and *Chrysichthyes nigrodigitatus*) from the Cross River at Ahaha, Obubra. The oil was converted to fatty acids methyl esters (FAME) and the fatty acids evaluated by gas chromatography – mass spectrometry using retention time method. The fatty acids profiles include saturated fatty acids (SFAs), monounsaturated fatty acids (MUFAs) and polyunsaturated fatty acids (PUFAs). The composition varied from 0.06 – 24.00% of SFAs, 0.23 – 21.40% of MUFAs and 0.23 – 12.22% of PUFAs in the species. The major SFAs were palmitic (C16:0) and stearic (C18:0) acids while the most abundant MUFA was oleic (C18:1) acid. Dominant PUFAs in the both specie were linoleic (C18:2), eicosapentaenoic (EPA, C20:5) and docosahexaenoic (DHA, C22:6) acids. Quantitatively the composition of various fatty acids was in the order SFAs < MUFAs < PUFAs although not significantly different ($p < 0.05$) in the species. *Chrysichthyes nigrodigitatus* had higher total fatty acids (TFA), PUFAs and omega -3 / omega -6 ratio than *C. gariepinus* with higher levels omega -6 fatty acids. The ratios of PUFA/SFA were higher in *C. nigrodigitatus* although both were greater than the minimum value (0.4) recommended by World Health Organization. The result indicates that the species had high quality essential fatty acids especially DHA and EPA although relatively higher in *C. nigrodigitatus*. Hence these species have the potentials to serve as the natural dietary supplement for all important omega -3 fatty acids.

Keywords: Fatty acids, freshwater, *Clarias gariepinus*, *Chrysichthyes nigrodigitatus*, Ahaha River, Nigeria

Introduction

Fish is highly nutritious, tasty, easily digested and constitute an important source of protein and lipids for mankind throughout the World (Keriko *et al.* 2010). Fish is lower in total and saturated fats than red meats and is the major source of omega – 3 polyunsaturated fatty acids (PUFA), which lower total serum cholesterol levels (Lands 1986). It is estimated that around 60 percent of people in many developing countries depend on fish for over 30 percent for their animal protein supplies, whereas 80 percent in most developed countries obtain not less than 20 percent of their animal protein from fish (Tenyang *et al.*, 2016). Fish consumption has increased in importance among health conscious people because it provides a healthy, low cholesterol source of protein and other nutrients (Knuth *et al.*, 2003). Fish contains significant amounts of all essential fatty acids, which are relatively poor and lacking

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in cereals. Fish oils is also reported to have significant amount of excellent dietary sources of highly unsaturated fatty acid (HUFA) and polyunsaturated fatty acid (PUFA), essentially the linoleic acids, lenolenic acids, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). Fresh fish contains saturated fatty acids (SFAs), monounsaturated fatty acids (MUFAs), and long-chain unsaturated fatty acids that play vital role in human health. Diet rich in PUFA, especially the omega -3 fatty acids has been reported to have beneficial effects on human health. According to Vanderwesthuyzen *et al* (1984) linoleic and linolenic acids found in fish lipids are important for preventing skin diseases and are considered essential as they cannot be synthesized by the organism. Fish omega-3 and omega -6 oils are known to reduce cholesterol levels, incidence of coronary heart diseases (CHD), stroke and pre-term delivery (Daviglius *et al.*, 2002). Cholesterol is a fatty – like waxy substance which is found naturally in all part of animal body and is the main constituents of saponifiable lipid fraction which serves as biosynthesis of vitamin D, steroid hormone and bile. The low density lipoproteins (LDL) known bad cholesterol are transported from liver and deposited in the arteries causing arteriosclerosis (Jeyasenta and Patterson 2013). Food and Agricultural Organisation (FAO, 2010) reported that lauric, myristic and palmitic acids increase the levels of LDL in the blood stream thereby increasing cholesterol level. Studies have also shown that while omega – 3 fatty acids prevents arteriosclerosis and autoimmune diseases (Simopoulos, 2004), omega – 6 have pro inflammatory properties and enhance immune reactions like fever and pain (Calder, 2008). Monounsaturated fatty acids (MUFAs) have been reported to reduce LDL cholesterol level hence reducing the incidences of coronary heart disease (CHD) and arteriosclerosis (Grundy, 1987). According to Bowman and Rand (1980) fish and fish oils plays significant role in decreasing the risk of developing cardiovascular diseases, improving foetal brain development, regulates prostaglandins syntheses and induce wound healing. Polyunsaturated fatty acids (PUFAs) are essential due to their ability to prevent certain disorders and diseases like arterial sclerosis, thrombogenesis, high blood pressure, cancer, and skin diseases (Arts *et al.*, 2001; Kris-Etherton *et al* 2002). European Food Safety Authority (EFSA, 2010), recommended consumption of between 250 and 500 mg/day of EPA and DHA for primary cardiovascular prevention. Eicosapentaenoic acids reduces the concentration of cholesterol and triglycerides in the plasma by lowering the rate of synthesis of LDL and very low density lipoprotein (VLDL) by the liver and vascular tissues (Illingworth *et. al.*, 1984). Similar studies have showed docosahexaenoic acid (DHA), to be effective in skin disorders, aids brain development and also forms the greater part of the retina (Lee *et. al.*, 1985). However deficiency in EPA and DHA causes' neurobehavioural

disorder known as attention deficit hyperactivity disorder and most commonly occurred in childhood and adolescence that can be prevented by regular consumption of omega-3 PUFAs (Conner 1997). Studies have also shown that freshwater fish oil contains relatively higher level of omega – 6 than marine which has more of omega – 3 (Gunstone, 1996; Osibona 2011; Kaliyamurthi *et al* 2019). Tenyang *et al.* (2016) reported the higher levels of EPA, DHA and arachidonic (ARA) acids in four freshwater fishes of Maga lake North Cameroon. Higher proportions of palmitic (32.2%), palmitoleic (13.2%), oleic (8.1%), stearic (9.5%) and linoleic (12.3%) acids were reported for *C. gariepinus* (Osibona *et al* 2009). Saturated, monounsaturated and polyunsaturated fatty acids were detected to be higher in *C. gariepinus* > *Pseudotolithus typus* > *Pentanemus quinquairius* > *Tilapia zilli* from Lagos lagoon (Osibona, 2011). Sarkar *et al* (2019) reported that the most abundant fatty acids usually found in ten freshwater fishes of Hel river of Assam, India are myristic, palmitic, oleic, linoleic EPA and DHA acids. The major SFA (palmitic, Stearic and behenic acids), MUFA (oleic acid) and PUFA (linoleic acids) were reported four fishes of Hooghly estuary, West Bengal, India (Nath *et al* 2014). Gutierrez and Silva (1993) had also reported that palmitic and oleic acids were the most predominant saturated and monounsaturated fatty acids of commercially important fishes from Atlantic coast of Brazil. Dhaneesh *et al.* (2012) reported higher levels of PUFAs in some commercially important fish species of Lakshadweep Archipelago that ranges from 30.32 to 35.11 %. Ljubojevic *et al.* (2013) stated that PUFAs in some fishes from Inland waters which varied from 17.07 to 28.15 %. Linoleic acid (omega -6) detected in nine fish species and was found to be the most abundant PUFAs and varied from 0.32 % in *T. putitora* to 27.21 % in *N. hexagonolepis* (Sarkar *et al* 2019).

Studies on lipid composition of animal origin foods have shown that the fatty acid composition of animal products is significantly influenced by a number of factors such as diet, species, breed, age and gender of the animals (Sargent *et al.* 1999, Zanardi *et al.* 2000, Estevez *et al.* 2006). The existing inter and intra species variability in the composition of FA of fish lipids (and of the specific PUFA in particular) is usually explained by the existence of a large number of external factors (environment, culturing method, tropic effects) and internal factors (fish species, feeding regime and digestion, life cycle stage, quantitative and qualitative characteristics of lipid and heat). Fish have the ability to change the composition of their cell membranes throughout the year replacing saturated fats with unsaturated ones as temperature drop. Saturated fatty acids and MUFAs are storage lipids preferentially used as energy sources (Daniela, 2005), MUFA are good substrates for oxidation in fish (Sidell *et al.*,

1995; Stabhaug *et al.*, 2005). Zenebe *et al.* (1998) have argued that variation in tissue lipid and fatty acid in herbivorous fish is greater than in those of carnivore fish and is due to the diversity of food habit. Proportion of plant and animal food will influence the accumulation of fatty acid (Domaizon *et al.*, 2000).

In view of the fact that demand for fish is globally increasing and that fish consumption is also generally recommended by dieticians, surprisingly, current overview of FA profiles and FA content in different fish species and in food products derived from marine and freshwater fish is lacking in the literature. Although few studies have been conducted to investigate the composition of fatty acids in some African fish species (Tenyang *et al.* 2016; Mohammed *et al.* 2014; Ugoala *et al.* 2009). *Clarias gariepinus* and *Chrysichthys nigrodigitatus* were considered for this study because of their economic importance and most cherish species by consumers in the study area. The study was undertaken to determine the differences in the compositions of fatty acids in the amounts of saturated, monounsaturated and polyunsaturated in two commercially important freshwater fishes of Cross River, Obubra Nigeria.

MATERIAL AND METHODS

Collection of samples

Fresh samples of African catfish (*Clarias gariepinus*) and Silver catfish (*Chrysichthys nigrodigitatus*) were obtained from Fishermen at Ahaha Obubra, Cross River State. The fishes are most predominant of their catches and commonly consumed in Obubra and its environs. Once collected, they were immediately transported to the laboratory in ice boxes for identification and analyses.

Preparation of samples

Fresh fish were washed with tap water several times to remove adhering blood and slime. The fishes were weighed, beheaded, eviscerated and cleaned prior to freezing. In an attempt to obtain a homogeneous sample from each species, their flesh were removed from their backbones, minced, blended and immediately extracted.

Extraction of Lipid and Fatty acids

Lipids were extracted from the powdered fish and fatty acid composition of oil were investigated after conversion of their Fatty Acid Methyl esters (FAME) using boron trifluoride-methanol method. The lipids were saponified and esterified for fatty acid analysis by the method of Christie (1993). The fatty acid methyl esters (FAMES) were analyzed on a Hewlett-Packard (HP) 5880 gas chromatograph (GC) with a flame ionisation detector (FID).

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The esters were separated on a 50 m × 0.20 mm. Wall-coated open tubular fused silica capillary column coated with Carbowax 20 M. Column injector and detector temperatures were 200 and 300°C, respectively with Helium gas at a ratio of 100:1. Identification and quantification was achieved by gas chromatography according to (AOAC 2005).

Statistical Analyses

Data on the fatty acids composition of fish samples were computed for mean and standard deviation using Statistical Package for Social Science (SPSS 20.0 version). Differences among the mean values was computed using t- test and graph plotted to show level of fatty acids composition among the species (Zar 1999).

RESULTS

Saturated Fatty Acids (SFAs)

The fatty acid composition of *C. gariepinus* and *Chrysichthys nigrodigitatus* is presented in Table 1. The result showed that 10 SFAs were detected in the study with *C. gariepinus* having nine (09). The SFA in *C. gariepinus* ranged from 0.06 – 26.00% and 0.17 – 21.50% in *C. nigrodigitatus* for lignoceric and palmitic acids respectively. The composition of individual SFA was higher in *C. nigrodigitatus* except lauric, myristic and palmitic acids which were lower. Palmitic acid was the most abundant of the SFA followed by stearic acid in the both species while lignoceric acid was the least detected in this study. However quantitative composition of SFA showed that *C. gariepinus* (45.18%) had significantly higher ($p < 0.05$) levels than for *C. nigrodigitatus* (42.24%).

Table 1 Saturated Fatty Acid (SFA) composition (%) of two freshwater species *C. gariepinus* and *C. nigrodigitatus*

Name	Carbon atoms	<i>Clarias gariepinus</i>	<i>Chrysichthys nigrodigitatus</i>
Lauric acid	C12:0	3.33± 0.54 ^{cd}	2.75 ±0.31 ^c
Myristic	C14:0	4.06± 0.19 ^c	2.23 ±0.24 ^{cd}
Pentadecanoic	C15:0	2.26±3.16 ^{cde}	1.13 ±0.17 ^{cde}
Palmitic	C16:0	26.00 ±1.83 ^a	21.50 ±2.08 ^a
Heptadecanoic	C17:0	0.70±0.09 ^e	2.18 ±0.17 ^{cd}
Stearic	C18:0	8.40 ±0.22 ^b	10.60 ±0.89 ^b
Arachidic	C20:0	0.24 ±0.04 ^e	0.58 ± 0.06 ^{de}
Behenic	C22:0	0.13 ±0.03 ^e	0.82 ±0.02 ^{de}

Tricosanoic	C23:0	ND	0.30 ±0.04 ^c
Lignoceric	C24:0	0.06 ± 0.02 ^e	0.17 ±0.22 ^e
Total $\Sigma n = 10$		45.18±1.34^a	42.24±3.54^b

Mean with the same superscript under the same column are not significant (p< 0.05)

Monounsaturated Fatty Acids (MUFA)

The result of monounsaturated fatty acid (MUFA) of *C. gariepinus* and *C. nigrodigitatus* is shown in Table 2. A total of eight (8) MUFA were detected during the study with *C. gariepinus* having seven (07) of the MUFA. The acids ranged from 0.23 – 21.40% and 0.26 – 15.25% in myristoleic and oleic acids for *C. gariepinus* and *C. nigrodigitatus* respectively. Oleic acid was the most abundant followed by palmitoleic acid while Nervonic acid was not detected in *C. gariepinus*. Similar trends was observed in *C. nigrodigitatus* with oleic acid (15.25%) having the highest concentration followed by palmitoleic acid (7.47%) whereas myristoleic acid (0.26%) had the least concentration. The composition of MUFA in the species showed that pentadecanoic, oleic, gadoleic and cetoleic acids were higher in *C. gariepinus* than *C. nigrodigitatus*. The quantitative composition also shows that *C. gariepinus* (31.35%) have a higher concentration of than *C. nigrodigitatus* (30.63%) although the difference was not significant (p> 0.05).

Table 2. Monounsaturated Fatty Acid (MUFA) composition (%) of two freshwater species *C. gariepinus* and *C. nigrodigitatus*

Name	Carbon atoms	<i>C. gariepinus</i>	<i>C. nigrodigitatus</i>
Myristoleic acid	C14:1	0.23± 0.07 ^f	0.26 ±0.03 ^c
Pentadecanoic acid	C15:1	1.48 ±0.17 ^d	0.34± 0.03 ^c
Palmitoleic acid	C16:1	3.70 ±0.18 ^b	7.47 ±4.33 ^b
Heptadecenoic acid	C17:1	0.70 ±0.08 ^{ef}	2.43 ± 0.17 ^c
Oleic acid	C18:1	21.40 ±0.79 ^a	15.25 ±0.44 ^a
Gadoleic acid	C20:1	2.48±0.17 ^c	1.88 ±0.10 ^c
Cetoleic acid	C22:1	1.38 ±0.17 ^{de}	0.86 ±0.05 ^c
Nervonic acid	C24:1	ND	2.15 ±0.24 ^c
Total $\Sigma n = 8$		31.35±2.44	30.63±3.55

Mean with the same superscript under the same column are not significant (p< 0.05)

Polyunsaturated Fatty Acids (PUFA)

The result of the polyunsaturated fatty acid (PUFA) of *C. gariepinus* and *C. nigrodigitatus* is presented in Table 3. The result shows a total of 10 PUFA were detected in the both species. In *C. gariepinus* PUFA ranged from 0.58 (C22:4) – 12.22% (C18:2) and 0.19% (C20:3) – 10.78% (C22:6) for *C. nigrodigitatus*. In *C. gariepinus* linoleic acid was the most abundant followed by docosahexaenoic acid (3.38%) and eicosadienoic acid (0.65%) was the least abundant. The composition of linolenic, octadecatetraenoic, arachidonic and decosahexaenoic acids were significantly different ($p < 0.05$) in the both species. Quantitative composition of PUFA in the species shows it was higher in *C. nigrodigitatus* (26.30%) than *C. gariepinus* (24.01%). A total of 7 omega – 3 and 3 omega – 6 fatty acids were detected in the both species with *C. nigrodigitatus* having a significantly higher ($p < 0.05$) level of omega – 3 (18.70%). The ratio of PUFA/ SFA and omega – 3/omega – 6 were also higher in *C. nigrodigitatus* (0.62 and 3.37) than *C. gariepinus* (0.54 and 0.79) although both species are favourable for consumption.

Table 3. Polyunsaturated Fatty Acid (PUFA) composition (%) of two freshwater species *C. gariepinus* and *C. nigrodigitatus*

Name	Carbon atoms	<i>C. gariepinus</i>	<i>C. nigrodigitatus</i>
Linoleic acid	C18:2 (n-6)	12.22 ± 0.27 ^a	5.46 ± 0.23 ^b
Alpha-Linolenic acid (ALA)	C18:3 (n-3)	1.50 ± 0.08 ^{cd}	1.60 ± 0.08 ^d
Octadecatetraenoic acid	C18:4 (n-3)	1.70 ± 0.26 ^c	0.38 ± 0.10 ^{ef}
Eicosadienoic acid	C20:2 (n-6)	0.65 ± 0.13 ^f	0.19 ± 0.13 ^f
Eicosatrienoic acid (Omega3)	C20:3 (n-3)	0.73 ± 0.10 ^{ef}	0.23 ± 0.17 ^f
Arachidonic acid (Omega3)	C20:4 (n-3)	0.74 ± 0.18 ^{ef}	1.93 ± 0.15 ^d
Eicosapentaenoic acid	C20:5 (n-3)	1.38 ± 0.10 ^{cd}	3.50 ± 0.08 ^c
Docosatetraenoic acid	C22:4 (n-6)	0.58 ± 0.10 ^f	0.38 ± 0.10 ^{ef}
Docosapentaenoic acid	C22:5 (n-3)	1.15 ± 0.13 ^d	1.88 ± 0.22 ^d
Docosahexaenoic acid	C22:6 (n-3)	3.38 ± 0.39 ^b	10.78 ± 0.17 ^a
Total $\sum n =$	10	24.01 ± 2.40^b	26.30 ± 2.03^a
Ratio of PUFA = PUFA/SFA		0.54^b	0.62^a
Omega – 3 (n-3) = 7		10.58	20.30
Omega – 6 (n-6) = 3		13.45	6.03
Ratio of Omega -3/Omega -6		0.79^b	3.37^a

Mean with the same superscript under the same column are not significant ($p < 0.05$)

The overall comparison of the fatty acids composition in the two species revealed that the levels of total fatty acids (TFA), PUFA and omega -3 were higher in *C. nigrodigitatus* than *C. gariepinus* (Figure 1). However, the composition of MUFA, SFA and omega -6 in the species were not significant ($p < 0.05$). The ratio of omega -3 to omega -6 showed that omega -3 fatty acids was higher in *C. nigrodigitatus* (3.37) than *C. gariepinus* (0.79), figure 2.

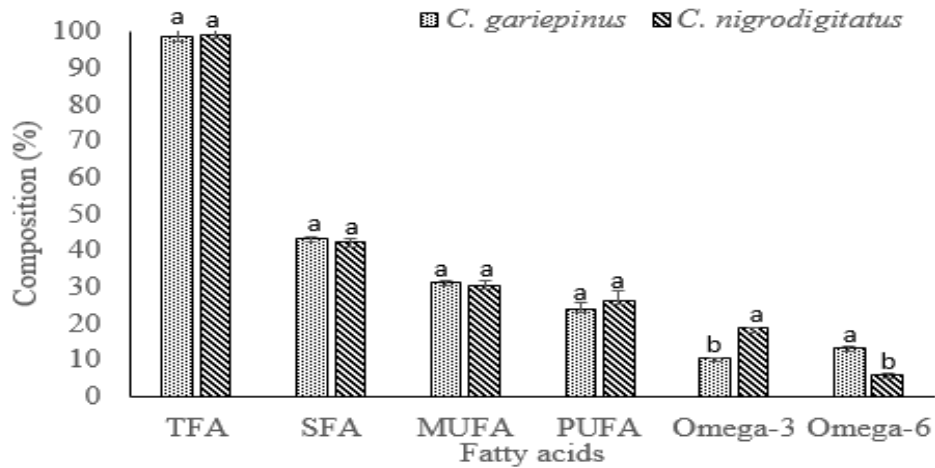


Figure 1: Fatty acids composition in *C. gariepinus* and *C. nigrodigitatus*

Where **TFA** = Total fatty acids, **SFA** = Saturated fatty acids, **MUFA** = Monounsaturated fatty acids and **PUFA** = Polyunsaturated fatty acids. Mean with the same alphabet are not significant ($p < 0.05$).

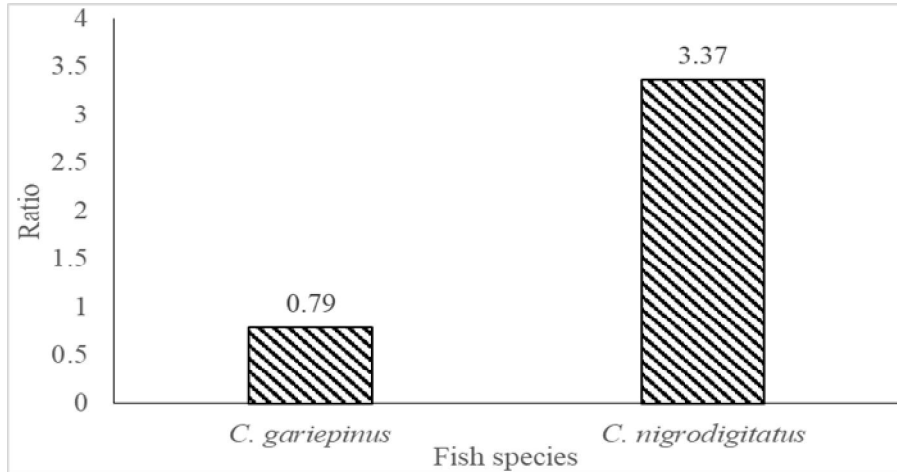


Figure 2: Comparison of omega -3/ omega -6 fatty acids in *C. gariepinus* and *C. nigrodigitatus* from Cross River at Ahaha, Obubra, Nigeria.

Discussion

Fatty acids profile analysis provide information about the essential fatty acids requirements of fish which would aid the compounding of adequate protein-to-fat ratios feed that would balance energy requirements with caloric intake. The incorporation of fish fatty acids as healthy additives in foods and as formulations in different forms of health promoting capsules or oils is currently increasing (Pickova 2009). This makes fish oils extremely valuable and explains the continuous increase in their prices. Fish oils are rich in highly saturated and unsaturated long chain fatty acids which plays vital roles in animal and human nutrition reflecting their characters in various critical biological processes. The fatty acid composition of these fish species decrease in the order saturated, > monounsaturated > and polyunsaturated fatty acids which was in agreement with available information on fatty acid composition of similar fish species as reported in previous work (Gutierrez and Silva, 1993; Osibona, *et al.*, 2009; Osibona, 2011; Tenyang *et al.*, 2016; Salaudeen *et al.*, 2017). The most abundant of the SFAs were palmitic and stearic acids for the both species. Palmitic and myristic acids have been reported as the predominant SFA in freshwater channel catfish, *Oncorhynchus mykiss*, (Haliloglu *et al* 2004), while Kaliyamurthi *et al* (2019) reported the abundant of palmitic, myristic and stearic acids in two loach species (*Biota lohachata* and *B. geto*) of Brahmaputra river, India. Similar results were obtained for African fishes as Ugoala *et al* (2009), Osibona *et al.*, (2009), Mohamed and Al-Shabali (2014) and Salaudeen *et al* (2017) all reported palmitic, myristic and stearic acids as the most abundant SFAs in fishes. Tricosanoic acid in addition to palmitic, myristic and stearic acids were found to be the most

abundant in African Lungfish, *Protopterus annectens* (Mohamed and Al –Shabali 2014) while Kaliyamurthi et al (2019) also found behenic acid to be abundant in loach species. Kaya and Erdem (2009) also reported that palmitic and stearic acids were major SFA in wild and farmed trout. According to Henderson et al. (1984) palmitic and stearic acids are the preferred substrates for mitochondrial β – oxidation and are heavily catabolized through the tricarboxylic acid cycles (TCA) to generate metabolic energy in fish. Ackman, (1980) reported that high levels of palmitic acids is a characteristic of freshwater fish hence corroborates the findings in this study. The total of 43.18 and 42.24% of SFA recorded for *C. gariepinus* and *C. nigrodigitatus* respectively in this study were higher than those reported for fresh (35.79%), smoked with skin (39.62%) and smoked without skin (38.43%) croaker, *Pseudotolithus senegalensis* (Salaudeen et al 2017). This study showed that *C. gariepinus* had a higher composition of saturated fatty acids than *C. nigrodigitatus*. This is also in agreement with the findings of several researchers who studied fatty acids profile of different freshwater and marine fishes (Osibona, 2011; Ugoala et al 2009; Mohammed and Al – Sabahi 2011). Saturated fatty acids speed up atherogenesis whereas MUFAs and PUFAs, reduces coronary artery disease (Kinsella et al., 1990). Decreasing SFAs and increasing MUFAs in the diet decrease diastolic blood pressure. The replacement of SFA with MUFA and omega 3- linolenic acids seems to stimulate beneficial health effects in humans with cardiovascular disease (Rasmussen et al., 2006). According to FAO (2010) replacing SFA with PUFA decreases the risk of coronary heart disease and also effect the concentration of plasma lipoprotein cholesterol fractions. Lauric, myristic and palmitic acids have been reported to increase LDL “bad cholesterol” whereas stearic (C18:0) has no identifiable effect LDL (FAO 2010).

Monounsaturated fatty acids (MUFAs) plays a vital role in human health by reducing the level of LDL in the body thereby reducing the risk of coronary heart disease (Grundy, 1994; Mensik and Katan, 1987). In this study a total of 7 and 8 MUFAs were detected in *C. gariepinus* and *C. nigrodigitatus* respectively. Uguoala et al (2008) had earlier reported similar number of MUFAs in freshwater fish species of Kainji Lake, Niger State Nigeria while less MUFAs (5) were detected by Osibona (2011) in four commercially important fishes in Lagos. The most abundant MUFA recorded in this study was oleic acid and its corroborates the findings of several researchers in both freshwater and marine fishes (Osibona et al 2009; Osibona, 2011; Mohammed and Al – Sabahi 2014; Kaliyamurthi et al 2019). The ranged of 0.23 to 21.40% detected *C. gariepinus* in this study was slightly lower

than the 0.2 to 26.0% and 0.25 to 32.96% detected for the same species by Osibona et al (2009) and Tenyang et al (2016) respectively. The high levels of oleic acid in *C. gariepinus* and *C. nigrodigitatus* in this study was consistent with that of other species of freshwater fishes. Steiner – Asiedu *et al* (1991) found that freshwater tilapia had significantly higher level of oleic acid the sardine and sea bream while American freshwater channel catfish had higher level of oleic acid the marine sardine and sea mullet (Ackman, 1994). Kaliyamurthi *et al.* (2019) reported different oleic acid composition in two loach species, *Botia lohachata* (43.26%) and *Botia geto* (27.96%) which was higher than the 26.0% and 21.40% recorded for *C. gariepinus* by Osibona (2011) and in this study respectively. However, Momen *et al* (2010) reported a higher value of palmitoleic than oleic acids in eight fish species of Indus River, Pakistan. Clement and Lovell (1994) have also reported the dominance of palmitoleic and oleic MUFAs in the tissues of freshwater fishes, *Oreochromis niloticus* and *Ictalurus punctatus*. The higher value of MUFA, 31.35% in *Clarias gariepinus* than 30.36% in *C. nigrodigitatus* although not significant ($p > 0.05$) agrees with various researchers who recorded differences in the composition of MUFAs in various fish species (Osman *et al* 2001; Osibona 2011; Ugoala *et al* 2009). These values were similar to the 31.50% and 30.70% reported *C. gariepinus* and *T. zillii* respectively by Osibona (2011) but were however higher than those reported for *Pseudotolithus senegalensis* (26.61%) by Salaudeen *et al* 2017) and *Pseudotolithus typus* (24.90%) by Osibona (2011). Monounsaturated fatty acids and SFAs can be synthesized de novo. De novo fatty acid synthesis increases when diets rich in carbohydrate and protein is the preferred carbon source for energy provision in fish (Sargent *et al.*, 2002). MUFAs are good storage lipid preferentially used as energy sources and substrates for β – oxidation in fish (Daniela, 2005; Stabhaug *et al* 2005). According to Dutta and Dutta, (2013) palmitoleic acid increases insulin sensitivity by suppressing inflammation and inhibits the destruction of pancreatic beta-cells which are known to secrete insulin (Dutta and Dutta, 2013). High MUFAs recorded in these fish species denotes that they are better sources of energy to both the fish and the consumer.

Polyunsaturated fatty acids are known as important chemical constituents for the human diet which can provide several health benefits. The lower values of PUFA than MUFA and SFA obtained in these species corroborates the findings of many researchers (Ugoala *et al.*, 2009; Osibona *et al* 2009; Dhaneesh *et al* 2012; Salaudeen *et al* 2017). Guler *et al.* (2017) reported higher values of PUFAs than those of MUFAs and SFAs in Wild brown Trout and cultured Rainbow Trout. Quantitatively, the amount of PUFAs recorded in this study for *C.*

garipepinus (24.01%) and *C. nigrodigitatus* (26.30%) were higher than those of *C. gariepinus* (22.30%) and *P. typhus* (19.70%) reported by Osibona (2011) and *Johnius gangeticus* (16.20%) and *Clupisoma garua* (14.64%) by Nath et al. (2014). Similar values of 24.48% for *Psilorhynchus nudithoracicus* and 26.84% for *Labeo pangusia* were reported for selected fishes of Assam, India (Sarker et al 2019). However, higher values of 30.64% and 41.87% was reported in the tissues of wild and cultured female broodstock of greater amberjack, *Seriola dumerili* respectively. Salaudeen et al. (2017) and Ugoala et al (2009) have earlier reported higher values PUFAs of ranging from 29.03 – 41.34% in fish species of Nigerian inland waters. Guler et al. (2017) reported highest values of 44.77 and 45.01% in wild brown trout and farmed Rainbow trout in comparison with those recorded for this study. The number of omega – 3 (seven) and omega -6 (three) fatty acids recorder in this study were higher than those reported for two Loach species (Kaliyumurthi et al 2019), some economically important fish species in Lagos (Osibona, 2011) and croacker purchase from Lekki beach in Lagos (Salaudeen et al 2017). Guler et al (2017) reported a total value of 35.53 and 27.43% omega – 3 and 9.25 and 17.58% omega – 6 fatty acids for wild brown trout and cultured Rainbow trout respectively. These values were higher than those recorded for this study except omega – 6 in wild brown trout which was lower than the 13.45% recorded for *C. gariepinus* in the present study. Higher levels of omega – 3 than omega – 6 fatty acids were recorded in this study for both species corroborates the studies of Steffens (1997), Osibona *et al* (2009), Tenyang *et al* (2014), Nath *et al* (2014) and Sarkar *et al* (2019) for fresh water fish species of the world. The most dominant PUFAs in *C. gariepinus* was linoleic acid (12.22%) followed by docosahexaenoic acids (3.38%) while in *C. nigrodigitatus* was docosahexaenoic acid (10.78%) and linoleic acid (5.46%). This was in agreement with several researchers who reported linoleic, docosahexaenoic acid (DHA) and eicosapentaenoic (EPA) as principal fatty acids in fishes (Memon *et al* 2010; Osibona 2011; Guler *et al* 2019; Sarkar *et al.*, 2019). Tenyang *et al.* (2016) reported that the predominant PUFA present in some freshwater fish species of Cameroon were α -linolenic acid (C18:3), eicopentaenoic acid (C20:5; EPA) and docosahexaenoic acid (DHA).The omega -3 PUFAs such as EPA and DHA are essential biomolecules which on consumption can improve the quality of life and reduce the risk of premature death of human beings. EPA is influential on mood and behavior whereas DHA is essential for brain growth and development of infants and is also needed for the normal functioning of brain in adults (Mohanty *et al* 2016; Karuppasany *et al* 2013). Eicosapentaenoic acids and DHA can assist in reducing the risk of coronary heart disease, hypertension, cancer, atherosclerosis, rheumatoid arthritis, lung diseases, old age disease such

as Alzheimer's disease, dementia and age related macular degeneration (Mahanty *et al.*, 2014). Deficiency of EPA and DHA mainly causes neurobehavioral disorder known as attention deficit hyperactivity disorder and most commonly occurred in childhood and adolescence that can be prevented by regular consumption of omega -3 PUFAs (Feldman and Reiff, 2014). According to Muhamad and Mohamad, (2012) PUFAs can help in reducing ones risk of developing an abnormal heartbeat that can lead to heart problems and even sudden death and prevents asthma, hypertension, diabetes, cancer and kidney dialysis and also tend to inhibit the development or metabolism of these diseases in the body. *Clarias gariepinus* and *C. nigrodiditatus* contained appreciated amount of very important PUFA, arachidonic acid which is a precursor for prostaglandin and thromboxane biosynthesis (Pompeia *et al* 2002). Arachidonic acid also interfere with the blood clotting process and attach to endothelial cells during wound healing Rahman *et al* (1995). This, however, was contrary to the result obtained in three local Malaysian *Channa* spp fish which recorded very high levels of arachidonic acids (Zuraini *et al* 2006). The high proportions of essential omega -3 fatty acids such as EPA and DHA acid makes them to be a better source of essential fatty acids which are of greater pharmaceutical importance since they are linked to reduced risk of cancer (Steffens, 1997). According to the European Food Safety Authority (EFSA, 2010), recommended consumption of EPA and DHA for the primary cardiovascular prevention is reported as between 250 and 500 mg/day. Many studies have demonstrates that low consumption of omega 3 polyunsaturated fatty acids is related to the incidence of coronary heart disease (Hu and Willett, 2002; Lee and Lip, 2003). Consumption of fish and fish oils appears to reduce the risk of coronary heart disease (Kris- Etherton *et al* 2002). In addition, these omega 3 fatty acids are important to prevent of cancer, hypertension, diabetes, depression, allergy and some other disease (Connor, 2000)

Dietary intake of omega -3 fatty acids is known to influence the expression of several genes (Price *et al.* 2000). The omega -3/ omega -6 ratio has been suggested to be the best index when comparing relative nutritional values of fish oils from different species. An omega -6/omega -3 ratio of $\leq 5:1$ is suggested by many studies to be the evolutionary developed optimum in the human diet (Simopoulos 2011). Previous studies have shown that the omega -6/ omega -3 PUFA ratio ranged from 1 to 4 for freshwater fish species. The UK Department of HMSO (2001) recommends an ideal relationship of omega-3/ omega -6 of ratio of $\leq 4:1$. The values recorded in these species were within the recommended Ideal range and were higher than those reported in croaker and tilapia (Salaudeen *et al* 2017; Osibona *et al* 2009).

The ratios in these species *C. gariepinus* (1:1) was better than in *C. nigrodigitatus* (1:3) although were within the recommended range by FAO (2005). Similar ratio of 3.84 was recorded in cultured rainbow trout higher than 1.57 in cultured brown trout (Guler et al., 2019) while very high value of 22.27 was reported for fresh croaker (Salaudeen et al 2017). Omega – 3 and -6 fatty acids are critical to fighting the invisible inflammation that occurs inside the body thereby preventing adverse reaction.

The ratio of PUFA/SFA indicates the quality of fats. According to Wood et al. (2008), the fish species would be the most favourable if the ratio of PUFA/SFA is above 0.4 (FAO, 2005). In the present investigation, the value of PUFA/SFA varied from 0.54 to 0.62 hence were found favourable to provide optimum dietary essential fatty acids requirement needed for efficient growth and development. Salaudeen *et al* (2017) reported a higher value of 0.81 in fresh croaker and similar values of 0.55 and 0.60 in smoked skinned and skinless croaker. A minimum value of PUFA/SFA ratio recommended is 0.45, (HMSO, 1994) which was lower than those obtained from the freshwater fish species investigated in present study, indicating that they are a good source of PUFA, particularly EPA, DHA, LA and ALA, hence suitable for inclusion in highly unsaturated low-fat diets.

Conclusion

The fish oil isolated from two different fish species provided interesting data regarding the fatty acid composition of the total lipid classes. In these fish species, different fatty acid groups (saturated, monounsaturated and polyunsaturated) were identified. Palmitic fatty acid was found to be the most abundant fatty acid with the highest concentration in *C.gariepinus* which is a common characteristic for freshwater fish. Essential fatty acids such as eicosapentaenoic (EPA) and docosahexaenoic (DHA) were identified and found to be at significant concentrations in both species. The distribution of fatty acids composition was found to be in accordance with observations in similar fish species from most freshwater fishes of the world. The fish species studied were good sources of essential unsaturated fatty acids required to reduce the risk of cardiovascular diseases. However, *Chrysichthys nigrodigitatus* appears to be the best as diet for humans due to its relatively high levels of omega –3 to omega –6, PUFA particularly DHA, EPA and low SFA making its very essential for human growth and development.

Comment [s5]: conclusion is too long

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly used in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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Comment [s6]: add the latest reference

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