#### FATTY ACIDS COMPOSITION IN SOME TISSUES OF COMMERCIALLY SELECTED FRESHWATER AND MARINE FISHES OF THE KENYAN WATERS

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#### Abstract

Fatty acid composition analysis in some tissues of commercially available freshwater and marine fishes in the Kenyan waters was conducted. Four (4) fish species from Lake Naivasha; Largemouth bass or black bass (Micropterus salmoides), Common carp (Cyprinus carpio), Mirror carp (Cyprinus specularis) and Tilapia (Oreochromis leucostictus) and three (3) species from the Indian Ocean; Red snapper (Lutjanus campechanus), White snapper (Macolor niger) and Rabbit fish (Siganus ludridus)] were sampled and analyzed. GC-MS analysis was performed using a GC Voyager-800 series with Trio-01 MS detector in electron ionization (EI) mode to determine qualitatively the fatty acids composition in fish oils. The study revealed that freshwater fish contain essentially omega-6 ( $\omega$ -6) fatty acids series of the polyunsaturated fatty acids (PUFA) while the marine fishes have more omega-3 ( $\omega$ -3) fatty acids series. The linoleic acid (LA, C18:2) was the prominent omega-6  $(\omega-6)$  fatty acid while the prominent omega-3  $(\omega-3)$  fatty acid was docosahexaenoic acid (DHA, C22:6) series. This may suggest that the dietary essential fatty acids available for marine fishes was the omega-3 polyunsaturated fatty acids which may be absent and hence unavailable for freshwater fishes Thus, the marine fish species are better providers of omega-3 fatty acids such as DHA (C22:6n-3) while the freshwater species are better providers of omega-6 fatty acids such as the linoleic acid (C18:2n-6) as well as the arachidonic acid (C20:4n-6). This study reveals that marine fish species contain appreciable levels of  $\omega$ -3 polyunsaturated fatty acids and would therefore be suitable for the provision of highly unsaturated low-fat diet containing omega-3 fatty acids while freshwater fishes will provide the  $\omega$ -6 fatty acids. This study however, may not explain whether the  $\omega$ -3 fatty acids observed in marine fishes are derived directly from the fish diet or the fish species are good converters of the short chain  $\omega$ -3 fatty acids like linolenic acid (18:3n-3) into EPA and DHA through enzyme controlled de-saturation followed by chain elongation processes.

#### Key words: Fish lipids, fatty acid composition, freshwater and marine fishes, Kenyan waters, docosahexaenoic acid (DHA)

#### 1.0 Introduction

Over the last few decades, a considerable number of experiments have indicated that consumption of fish oils rich in  $\omega$ -3 polyunsaturated fatty acids (PUFA) has health implications (Hu, et al., 2002). Fish lipids, especially those of marine origin, are the natural source of plenty of these fatty acids. It is documented that, the consumption of lipids rich in saturated fatty acids and cholesterol increases atherogenesis, while lipids rich in monounsaturated fatty acids (MUFA) and PUFA reduces atherogenesis and thrombogenesis and the risk of cardiovascular diseases (Fujimoto, et al., 1990). These fatty acids, particularly eicosapentaenoic acid (20:5 n-3 or EPA) and docosahexaenoic acid (22:6 n-3 or DHA) have been proved to have beneficial effects in cardiovascular diseases and control of blood lipid levels (Kinsella, et al., 1990a), diabetes mellitus (McManus, et al., 1996), depression (Edwards, et al., 1998), autoimmune disorders, rheumatoid arthritis and other inflammatory disorders. EPA and DHA are also known to play a major role in modulating the biosynthesis of eicosanoids (Kinsella, et al., 1990b). Moreover, DHA is found in high concentrations in membranes of important organs, possibly influencing membrane-lipid dependent functions, especially in retina and brain (Hoffman, et al., 1993) and the eye acuity in the elderly and therefore, the nutritional importance of fish consumption is highly associated with the n-3 PUFA contents. These findings have created a new market for fish oil as a food and dietary supplement globally (Hjaltason, 1990). Fatty acid composition data are needed by food scientists and nutritionists to aid them in dietary formulations, processing and product development leading therefore, to several aqua culture studies focused on the determination of the optimum levels of dietary-lipid ratios in relationship to enhanced fish body lipids with growth in different fish species globally which have revealed interesting results (Peres and Oliva-Teles, 1999; Takeuchi, et al., 2002, Morais et al, 2006; Lopez et al., 2009; Zhang et al., 2010; Chatzifotis et al., 2010; Xu, et al, 2011, Güroy, et al., 2011, Arslan, et al., 2012, Mercan, et al., 2013, Aimin et al., 2014, Ali, et al., 2014).

Thus, fish being a major source of proteins for many human populations, has stimulated many studies geared towards collecting information about the quality of fish species spread all over the World. Fatty acid compositional data for different marine, freshwater and aqua cultured fish species, especially originating from the United States, Europe, Canada and Japan are available in literature (Hornung, et al., 1994; Peres and Oliva-Teles, 1999; Takeuchi, *et al.*, 2002, Morais et al, 2006; Lopez et al., 2009; Zhang et al., 2010; Chatzifotis et al., 2010; Xu, *et al*, 2011, Güroy, et al., 2011, Arslan, et al., 2012, Mercan, et al., 2013, Aimin et al., 2014, Ali, et al., 2014). However, published data on the composition of fatty acids in fishes found in developing nations especially in Africa is unavailable although factors determining fish feeding habits in some Lake's inhabitants have been reported (Keriko, et al. 2010). The aim of this study was to collect data on fatty acids composition in some

selected fish species that represent an important protein source in Kenya's marine and fresh waters.

## 2.1 Materials and Methods

#### 2.1.1 Study Sites and Sampling Method

Fish species were collected from two sites namely; the waters of Lake Naivasha, Kenya and the Kenyan Indian Ocean, along the Coastal town of Mombasa (at Shimoni and Wasini Island) (Fig. 1).



Figure 1. Map of Kenya showing the fish sampling locations (L. Naivasha and Shimoni in Kwale County)

#### 2.1.2 Sampling at Lake Naivasha

Lake Naivasha is located within the Kenyan Rift Valley with the entire watershed covering parts of both the Rift Valley and the Central Provinces located 0°46'S and 36°21'E. The watershed is a unique ecosystem because Lake Naivasha is the only fresh water lake within the Rift Valley. Its watershed is mainly a semi-arid environment with scarce surface and underground water resources. Lake Naivasha ecosystem plays a very important role in Kenya's national development. The area contributes about 70% of Kenyan flower export, 15% of Kenyan geothermal power generation and is home to attractive tourist sites (Republic of Kenya, 2002). The mean temperature around the Lake is approximately 25°C with a maximum temperature of 30°C, with December–March as the hottest period. June-July is the coldest period of the year with a mean temperature of 23°C. The Lake Naivasha

watershed is served by only two perennial rivers namely; the Malewa and the Gilgil Rivers with a total catchment area of 1,700 Km<sup>2</sup> and 400 Km<sup>2</sup> respectively. The rivers and ground water sources are a key to the provision of water to the Naivasha and Nakuru municipalities as well as other adjoining anthropogenic activities (LNMC, 2004). There are virtually no native fish species in the system, and the introduced aquatic species include; Common carp (Cyprinus carpio, Plate 1), Mirror carp, (Cyprinus specularis, Plate 2), Largemouth bass or black bass (Micropterus salmoides, Plate 3), Tilapia (Oreochromis leucostictus, Plate 4), and the Crayfish (Procambarus clarkia) support the local fisheries (Hickley, et al., 2003). In this study, only the first four (4) species were sampled. Sampling were conducted in October 2007 and in May 2008 after the onset of both the short and the heavy rainy seasons respectively at the fisher's sales and landing bay in Kihoto area along the Eastern shore of the Lake and a few hundred meters from the Nakuru-Nairobi highway. Through physical measurements, approximate equal sizes of each fish species were sampled and placed in cool boxes containing super-cooled ice that was maintained at below zero degrees celcius by a regular spread of salt. The samples were then transported to our laboratory in Juja where they were refridgerated. Guts extraction and other procedures were conducted later on.

# 2.1.3 Sampling in the Indian Ocean

In a similar manner and through physical measurements equal sizes of each of the three species of fish from the Indian Ocean; White snapper (*Macolor niger*, Plate 5), Red snapper (*Lutjanus campechanus*, Plate 6) and Rabbit fish (*Siganus luridus*, Plate 7) were purchased at the fisher's sales and landing bay/market at Shimoni and Wasini Island, South of Mombasa City (Fig. 1) in the months of September 2007 and April 2008. Shimoni is a historical slave trade port in South Eastern Kenya. It is known for its Swahili ruins and slave caves where slaves were kept vigil waiting shipping. Wasini Island lies off the Southern Indian Ocean Coast of Kenya in Msabweni Sub-district of Kwale County. The Island is sparsely populated, undeveloped with neither roads nor vehicles. The island is next to the Kisite-Mpunguti Marine National Park which is a site of early Swahili civilization. Its attractions include exposed coral reefs. The island is occupied by the Fuba people who number about 1,500. Sampled fish were handled in the same manner as the L. Naivasha samples.

#### 3.0 Data Analysis

The analysis of the fish specimen physical data (height, width and the breadth) were done using the SPSS analytical tool.

#### **Plates:**



Plate 1. Common carp (Cyprinus carpio)



2. Mirror carp (Cyprinus specularis)



Plate 3. Largemouth bass (Micropterus salmoides)



Plate 4. Tilapia (Oreochromis leucostictus),



Plate 5. White snapper (Macolor niger)



Plate 6. Red snapper (Lutjanus campechanus)



Plate 7. Rabbit fish (Siganus luridus)

# 3.1 Fish sampling and handling procedure

Fish species were sampled at random in open fish landing bays as boats arrived from fishing. Almost equal sizes (weights, lengths and widths) samples of each fish species were purchased and subsequently placed into super-cooled ice boxes ready for transport to Juja the same day. The samples from Indian Ocean were transported the following day after sampling. On arrival in JKUAT (Juja), all samples were rinced with clean-distilled water severally, placed in labeled polythene bags and stored in a refrigerator before gutting until when required.

# 3.2 Lipid Extraction

Each individual organ tissue namely; the muscles, pyloric caecum, the liver and the orbital region of each fish specimen was separated, weighed and then minced. The minced tissue was then solvent extracted with chloroform/methanol (2:1). After dispersion, the whole mixture was agitated for 20 min. in an orbital shaker at room temperature. The homogenate was filtered to recover the liquid phase. The interface was rinsed twice with methanol/chloroform (1:1). After centrifugation and siphoning of the upper phase, the lower chloroform phase containing lipids was filtered off and the water was removed from the extract by passing it through a filter paper containing anhydrous sodium sulphate (Na<sub>2</sub>SO<sub>4</sub>), (Sigma-Aldrich, Germany). The individual lipids were recovered after evaporating the solvent under vacuum in a rotary evaporator as described by the Folch protocol (Hiroaki *et al.*, 2002; Keriko *et al.*, 2010). The lipids extracted were weighed and transferred into vials and any remaining solvent was left to evaporate in a fume chamber. The dry extracts were covered with Aluminium foil papers and finally refridgerated. This procedure was repeated for each separate organ.

# 3.3 Separation of Fish Lipid Classes

Each extracted lipid class (TAG, Wax, DAGE, Sterols, FFA, PE and PC) were obtained in fractions from silica gel columns as described in Hiroaki, *et al.*, (2002) and Keriko *et al.*, (2010).

# 3.4 Preparation of Methyl Esters for GC-MS Analysis

Individual lipid components of triacylglycerides (TAG) fraction was converted into fatty acid methyl esters through direct trans-esterification by refluxing each component using methanol (10 ml) containing 1% concentrated hydrochloric acid for a period of three hours. Each resulting crude product was poured into saturated brine (200 ml) containing 10 ml of saturated sodium hydrogen carbonate (NaHCO<sub>3</sub>) in a separating funnel after allowing it to cool down to room temperature. The upper organic layer was then extracted with *n*-hexane (50 ml), purified by passing it through a short column of silica gel after dehydrating it overnight using anhydrous sodium sulfate (Na<sub>2</sub>SO<sub>4</sub>). Elution was carried out using dichloromethane: *n*-hexane (2:1) solvent system (Hiroaki et al. 2002). The fatty acid methyl esters thus prepared (1  $\mu$ ), was injected into the Gas column.

# 3.5 Peak Identification of Fatty Acid Methyl Esters

Fatty acids methyl esters prepared above were identified using marine lipid methyl esters as reference standards (PUFA-1, Marine source Cat. No. 47033 and PUFA-3, Menhaden oil Cat. No. 47085-U, Supelco UK) and comparison of the mass spectral data obtained by the GC-MS to that of the NIST Library. The fatty acids were identified by comparison of the retention times with those of standard purified fatty acids as described by Shirai, *et al.*, (2001).

## 3.6 GC-MS analysis

Gas Chromatography - Mass Spectroscopic (GC-MS) analysis was performed using a GC Voyager-800 equipment series connected to a Trio-01 MS detector in electron ionization (EI) mode which is stationed at IEET building in JUKAT. Separations were achieved with a fused silica capillary column Omega-wax (30 m x 0.25 mm I.D, 0.25  $\mu$ m film). The carrier gas was helium (flow rate 1 ml/min) with a split injection of 50:1. The temperature profiles were as follows: initial temperature, 140°C for 4 min; ramped at 10°C/min; final temperature, 240°C and held for 10 min (final time, 50 min.); injector temperature, 250°C; and detector temperature, 280°C. EI-MS data was obtained under the following conditions. The transfer line temperature was set at 280°C and the mass range was 50 – 450 amu. Solvent cut was set at 8 min. full scan mode.

# 4.0 Results and Discussion

# 4.1 Physical Measurements

Physical parameters namely; weight, width and length of each fish specimen were recorded as shown in Table 1. Specific organs such as the; muscles, liver, orbital and pyloric caecum were removed separately and each individual part was subjected to the Folch protocol as reported with some slight modifications (Keriko et al., 2010).

Fish species	Ν	Weigh (g)	Length (cm)	Width (cm)
Common carp (Cyprinus carpio)	6	618.27 ± 40.02	36.93 ± 2.42	5.37 ± 1.05
Mirror carp (Cyprinus specularies)	6	549.14 ± 22.12	34.9 ± 1.74	4.7 ± 0.17
Largemouth bass ( <i>Micropterus salmoides</i> )	6	589.02 ± 4.52	32.5 ± 0.58	4.85 ± 0.55
Tilapia (Oreochromis leucostictus)	6	180.78 ± 8.84	21.22 ± 0.22	2.62 ± 0.35
White snapper ( <i>Macolor niger</i> )	6	351.13 ± 48.86	28.08 ± 1.54	4.47 ± 1.29
Red snapper (Lutjanus campechanus)	6	331.41 ± 29.58	28.38 ± 2.14	4.18 ± 1.32
Rabbit fish (Siganus luridus)	6	234 ± 27.79	25.6 ± 1.02	4.53 ± 1.28

Table 1. Physical parameters of selected freshwater fishes of L. Naivasha and marine fishes of Indian Ocean

Key: N = Number (n) of specimen for each fish species

The fish specimen physical measurements (Table 1) indicate that, three fish specimens from L. Naivasha (*M. salmoides, C. carpio* and *C. specularis*) were of comparable average sizes with all parameter (wt, wd, I) differing slightly from each other. However, the tilapia (*O. leucostictus*) had all measurements differing significantly from those of the three species. The tilapia specimens were nearly one third lighter and much smaller in all mean sizes. This growth pattern in Tilapia fish species was described as dwarfism growth characteristics as a result of environmental degradation and water pollution. For the Coastal marine specimen, rabbit fish (*S. luridus*) was slightly lighter than the other two species despite being of comparable size to the other two species. However, all the specimen of fishes in this study, were considered lean fishes.

4.2 Qualitative Analysis of Fatty Acids Composition in the TAG Deposit Lipids

*Micropterus salmoides, Cyprinus carpio, Cyprinus specularis* and *Oreochromis leucostictus* are commercially important to freshwater fisheries in L. Naivasha, Kenya while *Lutjanus campechanus, Macolor niger* and *Siganus ludridus* are just but a few commercially important marine fishes along the Kenyan Coast of the Indian Ocean. They constitute a major part of landing, aquaculture and sources of animal protein for the inhabitants of these regions. It was observed that the fatty acid compositions of freshwater fishes (Table 2) vary greatly from that of marine fishes (Table 3). As seen in the two Tables for instance, the major saturated fatty acids included: methyl myristate (25.572), methyl pentadecanoate (28.106), methyl palmitate (30.406), methyl margarate (31.895) and methyl stearate (34.174) while the major unsaturated fatty acids included: methyl palmitoleate (31.275), methyl oleate (34.640), methyl linoleate (35.657), methyl linolenate (41.625), methyl arachidonate (44.810), methyl eicosapendaenoate (49.144) and methyl docosahexaenoate (49.594).

	erus salmiod			us carpio			us specula			romis leucosti	
Fatty acids	Muscles	P. ceacum	liver	Muscles	P. ceacun	liver	Muscles	P. ceacum	liver	Muscles	liver
Myristic acid (C14:0)	+	+	-	-	+	-	+	+	-	+	-
Pentadecanoic acid(C15:0)	+	+	+	-	+	+	-	+	+	+	+
Palmitic acid (C16:0)	+	+	-	+	+	-	+	+	+	+	+
Palmitoleic acid (C16:1)	+	+	-	-	+	-	-	-	-	+	+
Stearic acid (C18:0)	-	-	+	+	-	-	-	+	-	-	-
Oleic acid (C18:1)	+	-	+	+	-	+	-	-	-	-	-
Linoleic acid (C18:2)	+	+	+	-	+	+	-	+	+	-	+
Linolenic acid (C18:3)	+	-	+	-	+	-	+	-	-	-	+
Margaric acid (C19:0)	-	-	-	-	+	-	-	-	-	+	-
Arachidonic acid (C20:4)	-	-	+	-	+	+	+	-	-	-	+
Eicosapendaenoic acid (C20		-	-	-	-	-	-	-	-	-	-
Docosahexaenoic acid (C22:	E -	-	-	-	-	-	-	-	-	-	-

Table 2. Fatty acids detected in the lipids of some tissues of freshwater fishes of Lake Naivasha

<u>*Key*</u>: The positive (+) sign indicates the presence of the fatty acid, while the negative (-) sign indicates the absence of the fatty acid

Table 3. Fatty acids detected in the lipids of some tissues of marine water fishes of the Indian Ocean

	Red snapper		White sna	ıpper	Siganus ludridus		
Fatty acid	Muscles	Orbital	Muscles	Orbital	Muscles	Orbital	
Myristic acid (C14:0)	+	+	+	+	+	+	
Pentadecanoic acid( C15:0)	-	+	-	-	+	-	
Palmitic acid (C16:0)	+	+	+	+	+	+	
Palmitoleic acid (C16:1)	-	+	-	-	-	-	
Stearic acid (C18:0)	-	-	-	-	-	-	
Oleic acid (C18:1)	-	-	-	+	-	-	
Linoleic acid (C18:2)	-	+	+	+	-	-	
Linolenic acid (C18:3)	-	-	-	+	+	-	
Margaric acid (C19:0)	-	-	+	-	-	-	
Arachidonic acid (C20:4)	-	-	-	-	-	+	
Eicosapendaenoic acid (C20:5)	+	-	+	+	+	-	
Docosahexaenoic acid (C22:6)	+	-	+	-	+	+	

<u>Key</u>: The positive (+) sign indicates the presence of the fatty acid, while the negative (-) sign indicates the absence of the fatty acid

**4.3 Comparison of Fatty Acids Composition of Freshwater and Marine Fishes** The major fatty acids in all fish specimen in our study (Tables 2 and 3) were palmitic (16:0), palmitoleic (16:1), stearic (18:0) and oleic (18:1n-9) acids in all lipid classes, but their amounts and ratios differed significantly. The fatty acids composition of marine fish species (Table 3) contain higher levels of saturated fatty acids, particularly the C14:0 and C16:0, and a relatively lower levels of monounsaturated fatty acids (C16:1 and C18:1). The prominent omega-3 PUFA being docosahexaenoic acid (C22:6) while the linoleic acid (C18:2) is the major acid in the omega-6 series.

Our results indicate that marine fishes contain high levels of highly unsaturated *n*-3 PUFA [EPA (C20:5) and DHA (C22:6) which are absent in freshwater fishes of L. Naivasha. Hence, this study points out that, marine fish species contain appreciable levels of *n*-3 PUFA (Table 3) that would be suitable for the provision of this highly unsaturated low-fat oil in the diet. Both EPA and DHA are very important essential fatty acids for growth and development in humans. Freshwater fishes on the other hand, contain the omega-6 PUFA (Table 2) such as the C18:2n-6 as well as in C20:4n-6 contents. The latter fatty acid is a major constituent of membrane lipids (phospholipids) and is the principal precursor by enzymatic action of eicosanoids including the prostaglandins (prostanoids, isoprostanes, and isofurans). The eicosanoids produced from C20:4n-6 cause the strongest inflammatory response in humans. Inflammation is one of the body defense mechanisms that reduce the spread of infection.

This study however, may not explain whether the omega-3 PUFA observed in marine fishes are derived directly from the fish diet or the fish species are good converters of the short chain omega-3 PUFA such as the  $\alpha$ -linolenic acid (ALA, 18:3n-3) into EPA and DHA through enzyme controlled de-saturation followed by chain elongation processes. This variation in the composition of fatty acids in fishes is not unique to this study as there is an *inter-* and *intra-* species variability in the composition of fatty acids of fish lipids (and of the specific polyunsaturated fatty acids in particular) as observed by Buchtova, et al., (2004). This is further explained by the existence of a large number of external and internal factors that may be responsible for this observation. The external factors are environment, culturing method, and tropic effects. The interior factors include; fish species, feeding regime and digestion, life cycle stage, quantitative and qualitative characteristics of lipids (triacylglycerols, phospholipids) and their topographical origin (dorsal and/or ventral) part of muscle tissue (Buchtova, et al., 2004).

It has been observed that, the fatty acid composition of marine fish lipids is even more complicated by the fact that, it also varies remarkably with seasons, fishing ground, age and gender (Hazra, *et al.*, 1998; Mayzaud, *et al.*, 1999) and sea temperature (Saito *et al.*, (1997). Several studies confirmed that the assimilation patterns of dietary fatty acids reflect the content of the dietary lipid sources and that the accumulation of dietary lipids and fatty acids in several fish species was comprehensively observed in a variety of aquacultures (Peres and Oliva-Teles, 1999; Montero *et al.*, 2001; Takeuchi, *et al.*, 2002, Morais et al, 2006; Lopez et al., 2009; Zhang et al., 2010; Chatzifotis et al., 2010; Xu, *et al.*, 2011, Güroy, et al., 2011, Arslan, et al., 2012, Mercan, et al., 2013, Aimin et al., 2014, Ali, et al., 2014).

# 4.4 Mass Spectrum of Some Polyunsaturated Fatty Acids (PUFAs)

The mass spectra of methyl eicosapentaenoate (C20:5) of marine fishes is indicated in Fig. 2. Although the molecular ion ( $M^+$ ) peak at m/z 316 is extremely low, the diagnostic peaks at m/z 273 ( $M^+$ -43) and 242 ( $M^+$ -74) are evidence in the spectra. The position of the double bond could not be deduced from the mass spectrum of any unsaturated compound because the isomeric compound having double bond at different positions gives identical spectrum. One common feature to most of the PUFAs is the observance of C<sub>3</sub>H<sub>7</sub> (m/z 43), C<sub>4</sub>H<sub>7</sub> (m/z 55), C<sub>5</sub>H<sub>7</sub> (m/z 67), C<sub>6</sub>H<sub>7</sub> (m/z 79), C<sub>7</sub>H<sub>7</sub> (m/z 91) etc. which emerged by a difference of 12 unit.



Figure 2. Mass spectrum of methyl eicosapntaenoate present in marine fishes

Figure 3 indicates the mass spectra of methyl docosahexaenoate (C22:6) found in marine fish lipids. The compound did not show the mass peak at m/z 342 but as mentioned in the case of eicosapentaenoate, the common diagnostic peaks such as; m/z 55, 67, 79, and 91 were prevalent in this compound with the peak at m/z 79 being the base peak of the spectrum. The ions at m/z 105 (91 + CH<sub>2</sub>), 119 (105 + CH<sub>2</sub>), 133 (119 + CH<sub>2</sub>) and 147 (133 + CH<sub>2</sub>) were also observed, the regular difference being the methylene (14 units) between each subsequent peak. At m/z 99 (M<sup>+</sup> - 243), a small intensity peak can be seen in the spectrum.



Figure 3. Mass spectrum of methyl docosahexaenoate present in marine fishes

The identities of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) in marine fish oils by co-chromatography with reference standards as well as through their mass spectra clearly indicates that these fatty acids are present in marine fish oils samples extracted from the muscle and the orbital organs but are absent in freshwater fishes of L. Naivasha. The diagnostic fragments of the identified fatty acids methyl esters are summarized in Table 4.

Saturated Fatty Acids						
Systemic name	Common name	Molecular formular	Molecular weight	Mass, fragmentation pattern and the % abundance		
Methyl tetradecanoate	Methyl myristate	C <sub>15</sub> H <sub>30</sub> O <sub>2</sub>	242	GC-MS, m/z 242 (M <sup>+</sup> C <sub>15</sub> H <sub>30</sub> O <sub>2</sub> , 13%), 211 (M <sup>+</sup> - 31, 14%), 199 (M <sup>+</sup> -43, 11%), 185 (2%), 157 (2%), 143 (M <sup>+</sup> -99, 12%), 129 (5%), 111 (2%), 107 (7%), 87 (65%), 74 (100%), 55 (22%)		
Methyl <i>n</i> -pentadecanoate	Methyl <i>n</i> -pentadecanoate	C <sub>16</sub> H <sub>32</sub> O <sub>2</sub>	246	GC-MS, m/z 256 (M <sup>+</sup> C <sub>16</sub> H <sub>32</sub> O <sub>2</sub> , 12%), 225 (M <sup>+</sup> - 31, 7%), 213 (M <sup>+</sup> -43, 10%), 185 (2%), 171 (1%), 157 (5%), 143 (14%), 129 (4%), 115 (2%), 101 (5%), 87 (77%), 74 (100%)		
Methyl hexadecanoate	Methyl palmitate	C17H34O2	270	GC-MS, m/z 270 (M <sup>+</sup> C <sub>17</sub> H <sub>34</sub> O <sub>2</sub> , 56%), 239 (M <sup>+</sup> - 31, 29%), 227 (M <sup>+</sup> -43, 39%), 199 (11%),185 (17%), 171 (14%), 157 (7%), 143 (C <sub>8</sub> H <sub>15</sub> O <sub>2</sub> , 62%), 129 (26%), 101 (20%), 87 (100%), 74 (C <sub>3</sub> H <sub>6</sub> O <sub>2</sub> , 92%), 55 (73%)		
Methyl heptadecanoate	Methyl margarate	C <sub>18</sub> H <sub>36</sub> O <sub>2</sub>	284	$ \begin{array}{l} & \text{GC-MS, m/z 284 (M^+ C_{18}\text{H}_{36}\text{O}_2, 32\%), 253 (M^+ - \\ & 31, 10\%), 241 (M^+ -43, 17\%), 227 (2\%), 213 \\ & (1\%), 199 (1\%), 185 (7\%), 171 (1\%), 157 (1.5\%), \\ & 143 (24\%), 129 (10\%), 115 (2.5\%), 101 (9\%), 87 \\ & (100\%), 74 (C_3\text{H}_6\text{O}_2, 99\%), 55 (42\%) \end{array} $		

Methyl Hexadecanoate	Methyl palmitoleate	C <sub>17</sub> H <sub>32</sub> O <sub>2</sub>	268	GC-MS, m/z 268 (M <sup>+</sup> C <sub>17</sub> H <sub>32</sub> O <sub>2</sub> , 7%), 236 (M <sup>+</sup> -32, 25%), 194 (M+ -74, 17%), 180 (2.5%), 166 (4%), 152 (17%), 138 (10%), 124 (13%), 110 (22%), 96 (33%), 82 (47%), 74 (C <sub>3</sub> H <sub>6</sub> O <sub>2</sub> , 64%), 55 (100%)
Methyl Octadecanoate	Methyl oleate	C <sub>19</sub> H <sub>36</sub> O <sub>2</sub>	296	GC-MS, m/z 296 (M <sup>+</sup> C <sub>19</sub> H <sub>36</sub> O <sub>2</sub> , 6%), 264 (M <sup>+</sup> -32, 25%), 222 (M <sup>+</sup> -74, 12.5%), 180 (10%), 166 (7%), 152 (3.5%), 138 (8%), 124 (8.5%), 110 (22%), 110 (17.5%), 97 (40%), 83 (42%), 69 (76%), 55 (100%)
Methyl- 9,12,15-cis Octadecatrienoate	Methyl linolenoate	C <sub>19</sub> H <sub>32</sub> O <sub>2</sub>	292	GC-MS, m/z 292 (M <sup>+</sup> C <sub>19</sub> H <sub>32</sub> O <sub>2</sub> , 61%), 261 (M <sup>+</sup> - 31, 18%), 236 (M <sup>+</sup> -56, 20%), 223 (M <sup>+</sup> -69, 7%), 191 (8%), 175 (7%), 161 (9%), 147 (27%), 133 (17%), 119 (32%), 105 (73%), 91 (94%), 79 (100%), 67 (53%), 55 (43%)
Methyl- 5,8,11,14,17-cis Eicosapentaenoate	Methyl eicosapentaenoate	C <sub>21</sub> H <sub>32</sub> O <sub>2</sub>	316	GC-MS, m/z 316 (M <sup>+</sup> C <sub>21</sub> H <sub>32</sub> O <sub>2</sub> , 2%), 273 (M <sup>+</sup> -43, 0.5%), 242 (M <sup>+</sup> -74, 0.2%), 217 (5%), 203 (4.5%), 189 (0.2%), 175 (8%), 161 (7%), 147 (13.5%), 133 (25%), 119 (43%), 105 (38%), 91 (75%), 79 (100%), 67 (62%), 55 (33%)
Methyl-4,7,10,13,16,19-cis Docosahexaenoate	Methyl- Docosahexaenoate	C <sub>23</sub> H <sub>34</sub> O <sub>2</sub>	342	GC-MS, m/z 342 (M <sup>+</sup> C <sub>23</sub> H <sub>34</sub> O <sub>2</sub> ), 311 (M <sup>+</sup> -31, 0.2%), 236 (M <sup>+</sup> -106, 0.5%), 222 (1%), 208 (5.5%), 194 (1%), 180 (2.5%), 175 (5%), 161 (12%), 147 (12%), 133 (22%), 119 (43%), 105 (43%), 91 (86%), 79 (100%), 67 (65%), 55 (33%)

### 5.0 Conclusion

- Saturated fatty acids (SFA) and monounsaturated fatty acids (MUFA) were the major fatty acids in all freshwater and marine fish specimens in this study including palmitic (16:0), palmitoleic (16:1), stearic (18:0) and oleic (18:1n-9) acids in all lipid classes with their amounts and ratios differing significantly.
- This study reveals out that, marine fish species contain appreciable levels of omega-3 ( $\omega$ -3) polyunsaturated fatty acids (PUFA) which are absent in fresh water fishes of L. Naivasha
- Freshwater fishes contain high levels of omega-6 ( $\omega$ -6) fatty acids.
- It is not clear from these results whether the omega-3 fatty acids observed in marine fishes are derived directly from the fish diet or the fish species are good converters of the short chain omega-3 fatty acids like the α-linolenic acid (18:3n-3) into higher level PUFA.

#### 6.0 Recommendations

Further studies are recommended in order to unravel the cause of the differences in the composition of the fatty acids between the freshwater and the marine fishes taking into considerations both the internal and the external factors and/or whether, the marine fishes are better converters of short chain PUFA into long chain PUFA through enzyme-controlled processes.

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