FATTY ACIDS COMPOSITION AND NUTRITIONAL QUALITY OF SOME FRESHWATER FISHES

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ABSTRACT

The aims of this paper were to investigate the component fatty acids of some freshwater fish species by comparing the levels of essential fatty acids present. The FAME were determined by GC-MS and were identified using retention time locked methods and retention time databases. The fatty acids profiles include minor amounts of odd-number, branched-chain, and even-number fatty acids as well as saturated components, the MUFA and PUFA. The major SFA were C14:0 and C16:0. The C18:1 was the prominent MUFA. The dominant PUFA are of the omega-6 series and are found chiefly in C18:2 fatty acids. The essential fatty acids compositions showed prominence in C18:3n-3 and C18:2n-6. The branched chain fatty acids identified C15:0, C16:0, C17:0, C18:2 and C20:0. The overall significance of this study is the revelation that freshwater fishes have irregular pattern of FA which are good sources of omega-6 EFA. The high percent of branched and saturated FA in freshwater fish gives them an advantage in curing processing.

KEYWORDS: Fatty acids composition, Freshwater fish, Essential fatty acids, Nutritional quality

INTRODUCTION

Sarotherodon galilaeus, *Oreochromis niloticus* and *Tilapia zilli* as well as *Clarias gariepinus*, *Clarias anguillaris* and *Heterobranchus longifilis* which belong to the families, Cichlidae and Clariidae respectively are among the species of freshwater fish that are mostly utilized in aquaculture, especially in the developing world. In almost every country in sub-Saharan Africa where these species are cultured, they are cultivated for food. Since their introduction into aquaculture, many aspects of their biology have been investigated including food habits, potential as cultivated food fish, and in some cases, potential as biological control for nuisance aquatic vegetation (Bakir *et al.*, 1993).

Fawole *et al* (2007) have studied the proximate and mineral compositions of some freshwater fish species and observed that freshwater fishes are good in mineral compositions. Oladapo *et al* (1984) observed that the amino acid compositions of traditionally smoked (TS), traditionally solar dried (TD), oven dried (OD), Ife solar dried (ISD) of *Clarias lazera*,

Sarotherodon niloticus, *Sarotherodon galilaeus*, *Tillapia zilli* and *Hemichromis fasciatus* decreased in percent available lysine in the order OD $\approx$ ISD>TS>TD. However no systematic variation in the fatty acid compositions was observed. Analysis of nucleotides and nucleotide related products revealed no systematic variation in the individual nucleotide content. However a pooled percent of the total nucleotides shows that TD has the highest value of hypoxanthine. Abdullahi and Balogun (2006) studied the amino acid and protein quality of *Oreochromis niloticus*, *Tillapia zilli*, *Sarotherodon galilaeus*, *Hemichromis fasciatus*, *Clarias gariepinus* and *Clarias anguillaris*. Analysis of the whole fish indicated high protein content in the *Clariidae* than in the *Cichlidae*. Eighteen amino acids were noted in all species with glutamic acid being the highest in concentration in the pool, followed by lysine. Achionye-Nze and Omoridion (2002) studied the lipid composition of *Heterotis niloticus*, *Brycones nurse*, *Gnathonemus cyprinoids* and *Sarotherodon galilaeus* and observed that the common neutral lipids were cholesterol, free fatty acids and cholesterol esters, while diphosphatidyl glycerol, phosphatidyl glycerol and phosphatidyl ethanolamine were the most predominant phospholipids.

Recently, the lipids in fish muscle have received much interest as a source of EPA and DHA fatty acids in human diets. Lipid and fatty acids compositions of many marine fish and shell fish as well as the effect of different diets on lipid compositions of these marine species have been investigated (Ackman and Takeuchi, 1986; Viswanathan and Gopakumar, 1984; Halver, 1980; IFFO Bulletin No. 18). Suzuki *et al* (1986), Viola *et al* (1988), Bieniarz *et al* (2000) have investigated some of the factors causing changes in the composition of fatty acids in various fish species. Tothmarkus and Sasskiss (1993), and Fajmonova *et al* (2003) studied the impact that various types of heat treatment will have on the fatty acids composition.

The lipid in fish muscle can influence product quality through interaction with other components and through degradative changes after death. These changes are lipolysis and auto-oxidative deterioration of unsaturated fatty acids, resulting in product deterioration and undesirable aroma and flavours (Ackman, 1967; German and Kinsella, 1985). The effect produced by these changes is a major problem in the frozen storage of some fish species. Therefore, lipid composition of fish and fish products is of practical importance, particularly in relation to the effects of lipid components on deterioration during frozen storage and consumer acceptance. Detailed information about lipid components and their fatty acids constituents is needed to understand how to diminish oxidative or hydrolytic factors which

affect quality of fish. Also, fatty acids composition is the surest method of determining the selectivity of a hydrogenation reaction since fatty acids profile will aid in determining oils suitable for the production of solid fats for industrial uses (Buckley *et al*, 1989).

The present paper reports on the differences in the quantitative and qualitative compositions of fatty acids in the amounts of saturated, monounsaturated, and polyunsaturated Omega-3 and Omega-6 fatty acids between some freshwater fishes.

MATERIALS AND METHODS

Sample Collection and Preparation

Fresh captured fishes were sorted and identified. They include *Mormyridae* (*Hyperopisus bebe occidentalis*, *Mormyrops deliciosus* and *Mormyrus rume*), *Cichlidae* (*Oreochromis niloticus* and *Sarotherodon galileus*), *Clariidae* (*Clarias gariepinus* and *Clarias anguillaris*), and *Heterobranchus bidorsalis*, *Centropomidae* (*Lates niloticus*), *Clariheterobranchus* and *Characidae* (*Hydrocynus forskalii*) were obtained from Fishermen at the Kainji Lake Dam site. The fishes were weighed, beheaded, eviscerated and cleaned prior to freezing. In an attempt to obtain a homogeneous sample from each species, their fleshs were removed from their backbones, minced, blended and immediately extracted using chloroform-methanol mixture in the ratio of 2:1.

Lipid Extraction

Lipid extractions were performed on minced fish samples (10g each) using the extraction methods of Folch *et al* (1957). In this method, chloroform-methanol was used in the ratio of 2:1.

Fatty Acids Analyses

Fatty acids can be analyzed directly on polar stationary phases. However robust and reproducible chromatographic data are obtained if the fatty acids are derivatised (hydrolysis of the glycerol and methylation of the resulting fatty acids) to the fatty acids methyl esters. The method used for derivatisation in this work is that of Park and Goins, (1994). In this method, FAME is prepared by heating lipids with large excess of either acid- or base-catalyzed reagents. Acid-catalyzed reagents form FAME from free fatty acids and o-acyl lipids while the base reagents cause only transesterification, i.e. conversion of o-acyl lipids to FAME. Methylene chloride (100 μ L) and 1 mL 0.5M NaOH in methanol were added to oil extracts in a test-tube and heated in a water bath at 90°C for 10 min to prepare fatty acids methyl esters. The test tubes were removed from the water bath and allowed to cool before

the addition of 1 mL 14% BF₃ in methanol. The test tubes were heated again in a water bath for 90°C for 10 min, and cooled to room temperature. One mL of distilled water and 200µL hexane was added to the test tubes and then fatty acid methyl esters were extracted by vigorous shaking for one minute. After centrifugation, the top layer which is the fatty acids methyl esters was collected and transferred into a sample bottle for analysis. The fatty acids profiles were determined using an Agilent Gas Chromatograph, Model 6890N fitted with an Agilent Mass Selective Detector, 5973 series. Separation was carried out in a capillary column (30 x 0.25mm id x 0.25µm DB wax). The starting temperature was 150°C maintained for 2minutes at a heating rate of 10 °C/minute. The total running time was 22 minutes. Helium was the carrier gas while the injection volume was 1µL. The fatty acids peaks were identified using Agilent Technologies software 5988-5871EN.

RESULTS AND DISCUSSION

The fish oils Mormyridae(*Hyperopisus bebe occidentalis*, *Mormyrops deliciosus* and *Mormyrus rume*), Cichlidae (*Oreochromis niloticus* and *Sarotherodon galileus*), Clariidae (*Clarias gariepinus* and *Clarias anguillaris*), and Heterobranchus *bidorsalis*, Centropomidae (*Lates niloticus*), *Clariheterobranchus* and Characidae (*Hydrocynus forskalii*) are unique in their variety of fatty acids (Table 1) of which they are composed and their degree of unsaturation (Figure 1). There are high levels of Omega-6 polyethylenic than Omega-3 polyethylenic fatty acids. The most common fatty acids presented below have even number of carbon atoms per molecule and seldom contain functionalities other than cis and trans olefinic unsaturation, which usually occurs in a methylene-interrupted pattern in polyenes. Long chain fatty acids (Table 1) are ubiquitous constituents noticed. In a given species, saturated and unsaturated fatty acids occur generally side by side, their structures varying widely in chain length and in degree of unsaturation.

Table 1: Fatty acids compositions of some freshwater fish species

FATTY ACIDS	Clarias anguillaria ss	Lates niloticus	Mormyrops deliciosus	Mormyrus rume	Hydrocynus forskali	Heterobranchius bidorsalis	H. bebe occidentalis	Clarias gariepinus	Clarihetero branchus	Sarotherodon galileus	Oreochromis niloticus
4:0			*	*			*		*		
6:0	*										
10:0	*			*					*		
11:0	*					*					
12:0	*		*	*		*	*		*		
13:0	*			*		*	*				
14:0	*					*		*		*	*
14:1	*							*			
15:0	*			*		*	*	*		*	*
15:1	*			*		*	*	*	*		
16:0		*								*	*
16:1										*	*
17:0										*	*
17:1		*									
18:0	*	*						*		*	*
18:1cis		*						*		*	*
18:1 trans	*	*						*			
18:2cis		*	*			*					
18:2 trans		*	*				*	*	*	*	*
18:3n-3		*	*			*	*	*		*	*
18:3n-6		*	*					*			
18:4										*	*
20:0		*	*						*		
20:1n-9		*	*						*		
20:2		*	*						*		
20:3n-6		*	*					*	*		
20:3n-3		*	*			*					
20:4n-6		*	*					*	*	*	*
20:5n-3		*	*							*	*
21:0		*	*						*		
21:1						*					
22:0	*										
22:1			*	*			*				
22:2		*	*		*	*					
22:5n-3										*	*
22:6n-6										*	*
23:0	*		*		*						
24:0		*									

• * indicates fatty acid 2-piperidinone also found in *Mormyrops deliciosus*

Figure 1: Major fatty acids groups of studied fishes

The fatty acids profiles (Table 1) include minor amounts of odd-number, branched-chain, and even-number fatty acids. These varieties as well as the quantity and quality of fatty acids noticed are due to differences in sub-species, diet, spawning cycle, season and environment. The saturated components ranged from 9% to 76%. Within these components the major fatty acids were C14:0 and C16:0. The mole percent of each fatty acid seems to vary (Figures 1 and 2). The monoene contents ranged from 10% to 90% with C18:1 the prominent monounsaturated fatty acids (MUFA) (Figure 1). Polyunsaturated fatty acids (PUFA) attained the highest value (90%). The branched chain fatty acids identified are C15:0, C16:0, C17:0, C18:2 and C20:0 (Table 1). These fatty acids were 51%, 44% and 31% in *Sarotherodon galilaeus*, *Oreochromis niloticus* and *Lates niloticus* respectively and were the highest noticed. This high level of branched chain fatty acids in these species has an important advantage. Branched chain fatty acids influence lower melting point, lower cholesterol levels, provide energy, and form an integral part of biomembranes. Branched fatty acids because of their high temperature stability play an important role in the finished product of hydrogenated fish oils. Esterification of branched chain fatty acids

to cholesterol causes the fatty acids to stimulate protein synthesis. The branched chain esters influence some ribosomal functions which are necessary for peptide elongation (Hradec *et al*, 1974).

Figure 2: Essential fatty acids of studied fishes

In all the fish species analysed, the dominant PUFA are of the Omega6series and are found chiefly in C18:2 fatty acids. The essential fatty acids compositions showed prominence in C18:3n-3 and C18:2n-6. C22:6n-3 was noted in the tilapia species. *Clarias anguillaris* have more of saturated fatty acids than *Clarias gariepinus* and their hybrid *Clariheterobranchus*. *Clarias gariepinus* have more MUFA while *Clariheterobranchus* is more of PUFA (Figure 1). *Mormyrus rume* and *Hyperopisus bebe occidentalis* contain more of MUFA while *Mormyrops deliciosus* is highest in PUFA contents. The tilapia species (*Oreochromis niloticus* and *Sarotherodon galileus*) contain more of saturated lipids but comparable amounts of

MUFA, PUFA as well as omega-3 fatty acids. *Lates niloticus* and *Hydrocynus forskali* have their lipid content being more of PUFA. *Heterobranchus bidorsalis* is about 50% of saturated fatty acids (SFA).

Clarias anguillaris, *Mormyrus rume*, *Hydrocynus forskalii* and *Clariheterobranchus* have undetectable levels of omega-3 fatty acids while omega-6 fatty acids were not detected in *Clarias anguillaris*, *Mormyrus rume*, *Hydrocynus forskalii*, *Heterobranchus bidorsalis*, and *Hyperopisus bebe occidentalis*. Wild fish has low omega-3: omega-6 ratios. This is needed to reduce high levels of omega-6: omega-3 in most human diets. The essential fatty acids (figure 2) were lacking in *Clarias anguillaris*, *Mormyrus rume* and *Hydrocynus forskalii*. The Omega-3 fatty acid, 4,7,10,13,16,19-docosahexaenoic acid (DHA) were absent in all the species except the tilapias. The tilapia species contain all the essential fatty acids although they vary in composition. The degree of unsaturation of fish oils vary with seasons. It rises as the water temperature falls and vice versa (IFFO BULLETIN No.18). 9,12-octadecdienoic acid contents in *Lates niloticus* and *Clariheterobranchus* as well as *Mormyrops deliciosus* are high in that order. The oils were characterized by low levels of omega-3 PUFA.

Detailed information about lipid components and their fatty acids constituents is needed to understand how to diminish oxidative or hydrolytic factors which affect quality of fish. The nature, proportion, and degree of unsaturation of the fatty acids in the lipids are all closely related to the oxidation of the oils. However, the fatty acids composition of the muscle cell membranes are especially important factors in determining the stability because oxidative changes are initiated from the membrane components of muscle (Buckley *et al*, 1989).

Rancidity development is a vital concern to the food industry because it may be used for indexing and assisting in technology development. Fatty acids profile analysis also 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.

CONCLUSION

The aims of this thesis was to investigate the component fatty acids of some freshwater fish species as well as compare the nutritional quality of freshwater fish with that of marine fish by comparing the levels of essential fatty acids present. A systematic review of the scientific-medical literature was included, in order to appraise, and synthesizes the evidences for the economic potentials accruable from these natural resources as regard their contents of 5,8,11,14-eicosatetraenoic acid (EPA) and 4,7,10,13,16,19-docosahexaenoic acid (DHA). The overall significance of this study based on the fatty acid composition of the fish species revealed that although the freshwater fish have irregular content of fatty acid, they are however, good source of omega-6 EFA. The high percent of branched and saturated fatty acid in freshwater fish gives them an advantage in curing processing. The freshwater fish have also good oil quality because of the high content of 18:2n-6 and 20:4n-6 Fatty acids.

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