FATTY ACID COMPOSITION OF *Heterobranchus* GEOFFROY SAINT-HILAIRE, 1808 SPECIES FROM RIVER GALMA, ZARIA, KADUNA STATE, NIGERIA

***[1]YASHIM, E. Y., [1]T. O. L. AKEN'OVA, [2]J. AUTA, [3]I. A. ADEYINKA, [4]S. E. ATAWODI AND [1]S. J. ONIYE**

[1] Department of Zoology, Ahmadu Bello University, Zaria
[2] Department of Biology, Ahmadu Bello University, Zaria
[3]National Animal Production Research Institute, Ahmadu Bello University, Zaria
[4] Department of Biochemisty, Ahmadu Bello University.
Corresponding Author: nmana2007@yahoo.com

ABSTRACT

The fatty acid profiles of Heterobranchus species were investigated from River Galma, Zaria Kaduna State, Nigeria. Fresh samples of H. bidorsalis and H. longifilis obtained from the water body were subjected to Soxhlet method of oil extraction using n-hexane as solvent and Gas Chromatography Mass-Selective Detection (GC-MSD; Agilent Technology) for the detection of fatty acids present in the fish species. The resulting profiles revealed 17 fatty acids in H. bidorsalis and 19 fatty acids in H. longifilis, whereas monounsaturated fatty acid (MUFA) forms the bulk of the fatty acid in both species, saturated (SFA), as well as polyunsaturated (PUFA) fatty acids, were also identified. More PUFA was found to be present in H. bidorsalis than in H. longifilis with the reverse occurring in MUFA. Dominant among the identified and qualitatively measured fatty acids include Myristic C14:0, Lauric C12:0, Pentadecylic C15:0, Palmitic C16:0, Margaric C17:0, Palmitoleic acid C16:1(n-7), Oleic Acid C18:1(n-9), Fumaric acid C18:1(n-11), and Linoleic C18:2(n-6). Essential fatty acids which are important sources of nutrients promoting good health, prevention, and healing of diseases were identified from the species.

Keywords: Freshwater, *H. bidorsalis, H. longifilis,* SFA, MUFA, PUFA

INTRODUCTION

Clariid catfishes occur in most freshwater bodies of Africa where they constitute a significant component of the catches and are of great economic importance as food fish. African catfish, *Clarias*, and *Heterobranchus* species are widely cultured in Africa and beyond. They are especially readily acceptable among fish farmers and consumers in Nigeria, and so have a high commercial value (Aluko and Shaba, 1999; Offem, et al., 2008, Froese and Pauly, 2017). Quality and nutritional value of food are important since there is an increasing awareness of the need for consumption of healthy diet among the public. The nutritional properties of fish and fish products generally make them valuable foodstuffs that are beneficial for human health, nevertheless, lipid composition and thus fatty acid composition in fish differs depending on various factors: some of these usually include their natural (aquatic) environment (marine water, freshwater, and cold or warm water) as well as the biological, physical, and chemical properties of that environment. Also, seasonal changes, migration, sexual maturity and spawning period, species, feeding habits, and whether reared in aquaculture or grown in natural habitats affect the lipid/fatty acid composition (Lee, 2013). Fish are the most important sources of these fatty acids, especially fatty fish like sardines, mackerel, anchovies, and some salmon species, which are high in EPA and DHA and have a high ratio of n3 to n6 fatty acids. Although fish cannot synthesize these fatty acids, they can obtain them from the food they eat (algae and planktons) (Falk-Petersen, et al., 1998). In recent years, increasing attention has been focused on the significance of polyunsaturated fatty acids (PUFAs) in human nutrition, particularly eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) (Graham *et al*., 2007).

Fatty acids are fundamental biomolecules and have been used as trophic biomarkers in food web analysis. The interest in fat, which holds an important place in human nutrition, has increased with the recently increased interest in, and awareness of, human health. Fats are important components of hormones, cell membranes, and signaling molecules, as well as being important energy sources, when ingested into the body, it is first stored in the liver, hypodermic connective tissues, mesentery, and muscles and used when necessary. The human body can produce some of these fatty acids, but others, some of which also contain n-3 and n-6 PUFA, cannot be produced by the body. These essential fatty acids (EFAs) need to be obtained through food intake (Tocher, 2010).

Seafood remains a rich source of high-quality proteins, vitamins, mineral elements which contribute a significant source in the human diet and most importantly ω-3 polyunsaturated fatty acids (PUFA), that play a beneficial and protective role towards cardiovascular, chronic, and inflammatory diseases. This study, therefore, investigates the fatty acids present in the clariid freshwater species.

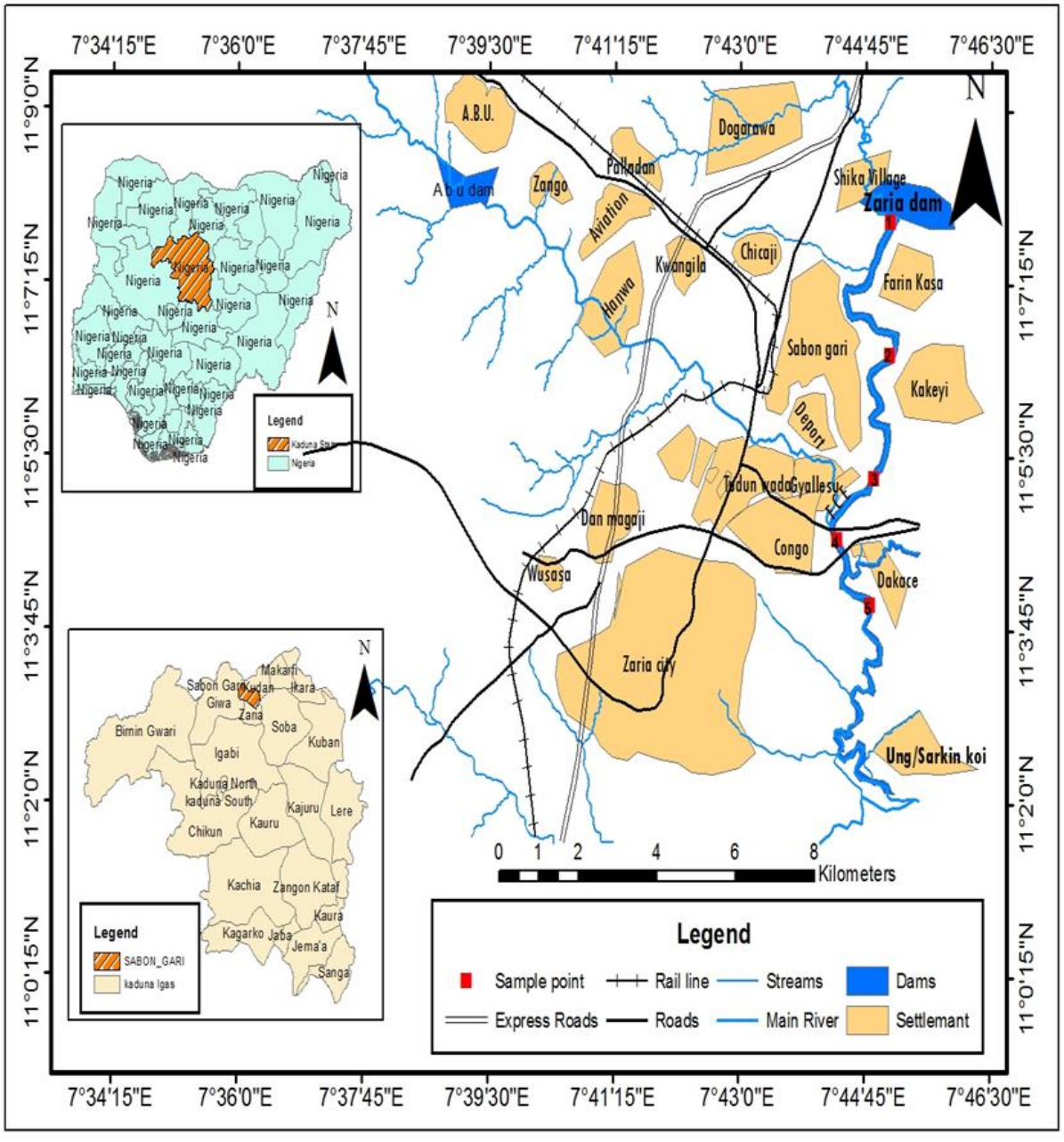
METHODOLOGY

Study area

Species of catfish *Herobranchus* species were collected from River Galma, Zaria, Kaduna State. River Galma is one of the main tributaries of River Kaduna with coordinates 10°37'60" N and 7°42'0" E (Figure 1).

Sample collection and preparation

Various sizes of the fish species *Heterobranchus bidorsalis* Geoffroy Saint-Hilaire, 1809 and *Heterobranchus longifilis* Valenciennes, 1840 were sampled from the water body and identified using taxonomic keys (Teugels, 1986; Idodo-Umeh, 2003). The fish species were thoroughly washed and the moisture content of the fish was reduced by oven drying at 65°C. The samples were further ground and packaged in labeled polythene bags for oil extraction.



Source: Satellite Image (2013).

Figure 1: The study area showing the course of River Galma

Fatty Acid Determination

Fish oil extraction and Gas Chromatography Mass-Selective Detector (GC-MSD) analysis

The fish oil was extracted using Soxhlet extractor using n-hexane as solvent after oven drying and crushing of the fish samples. The Soxhlet extraction operates continuously as the solvent is re-circulated through the sample (Garcia, 2003); the

fish oil was collected and used for analysis. The identification and qualitative measurement of fatty acid methyl esters (FAMEs) were carried out by gas chromatography coupled with mass spectrometry (GC-MSD) using Agilent Technologies GC-7890B MSD-5977A machine, equipped with auto-sampler, oven, and flame ionization detector. The injection volume was 1ml. The separation was carried out with helium carrier gas flow (0.7 mL/min), the pressure of 4.4867psi, and average velocity (30.641cm/sec). A fused silica capillary column (Agilent HP 5ms ultra insert 350°C, 30m × 250µm×0.25µm) was used. Oven profile: the temperature was set at 60°C for 1min, maximum oven temperature 350°C equilibrating time 1min, maximum temperature 350°C, initial 60°C; hold time 3min, Ramp 1 at 7°C/min finally held at 290°C for 35.857min. The column temperature was programmed starting at a constant temperature of 180 ºC for 20 minutes, heated to 200 ºC at 1.0 ºC/min, held at 200 ºC for 1 minute, heated again to 220 ºC at 5 ºC/min and finally held at 220 ºC for 20 minutes. A split injector (20:1) at 250 ºC, the pressure of 4.4897psi, and total flow (17.7ml/min) with a split flow of 14ml/min was used. The flame ionization detector was also heated to 250 ºC. MS parameters: tune type-E1, Tune EMV-806, MS source-230, MS quad-150, Acquisition type-scan, start mass-50, stop mass-550 at 150 thresholds with frequency (scan/sec) of 2.9.

Peak identification of fatty acids in the analysed samples was carried out by comparing with the retention time and molar mass of mass spectra of standard, obtained from the NIST library of the GC-MS machine and also confirmed from the Mass Spectrometric Fragmentation Pattern.

RESULTS

The fatty acid profiles of *H. bidorsalis* and *H. longifilis* shows an array of saturated, monounsaturated, and poly-unsaturated fatty acid composition (Table 1).

Table 1: Fatty acid composition of *Heterobranchus bidorsalis* and *Heterobranchus longifilis* from River Galma

Species	Fatty acid	Trivial name	Abbreviation	Level of Saturation	Peak No.	Retention time	% Area	Quality
Heterobranchus bidorsalis	Tetradecanoic acid	Myristic	C14:0	SFA	98	20.095	0.05	92
	Dodecanoic acid	Lauric	C12:0	SFA	98	20.095	0.05	76
	Undecanoic acid	Undecyleic	C11:0	SFA	98	20.095	0.05	76
	Pentadecanoic acid	Pentadecylic	C15:0	SFA	106	25.283	0.26	96
	Hexadecanoic acid	Palmitic	C16:0	SFA	111	26.85	5.15	99
	Heptadecanoic acid	Margaric	C17:0	SFA	115	28.331	0.3	97
	9-Hexadecenoic acid	Palmitoleic acid	C16:1(n-7)	MUFA	109	26.553	2.56	99
	9-Octadecenoic acid	Oleic Acid	C18:1(n-9)	MUFA	119	29.435	4.71	99
	7-Octadecenoic acid	Fumaric acid	C18:1(n-11)	MUFA	119	29.435	4.71	99
	9-Eicosenoic acid	Gadoleic Acid	20:1(n-11)	MUFA	123	30.728	0.44	59
	11-Hexadecenoic acid	Palmitvaccenic Acid	C16:1(n-5)	MUFA	114	28.018	0.19	94
	10,13-Octadecadienoic acid	None	C18:2 (n-10)	MUFA	118	29.358	0.89	99
	trans-13-Octadecenoic acid	None	C18:1(n-5)	MUFA	130	32.08	1.71	45
	7,10,13-Hexadecatrienoic acid	Roughanic acid	16:3 (n-3)	PUFA	117	29.252	0.31	94
	9,12-Octadecadienoic acid	Linoleic	C18:2(n-6)	PUFA	118	29.358	0.89	99
	5,8,11,14-Eicosatetraenoic acid	Arachidonic	C20:4(n-6)	PUFA	126	31.637	0.84	99
	5,8,11,14,17-Eicosapentaenoic acid (EPA)	Timnodonic Acid	C20:5(n-3)	PUFA	127	31.74	0.84	95
Heterobranchus longifilis	Dodecanoic acid	Lauric	C12:0	SFA	106	20.088	0.03	91
	Tridecanoic acid	Tridecylic Acid	C13:0	SFA	108	23.63	0.09	95
	Pentadecanoic acid	Pentadecylic	C15:0	SFA	109	24.689	0.06	95

Hexadecanoic acid	Palmitic	C16:0	SFA	122	27.792	0.09	97
Heptadecanoic acid	Margaric	C17:0	SFA	122	27.792	0.09	91
9-Hexadecenoic acid	Palmitoleic acid	C16:1(n-7)	MUFA	114	26.544	0.29	99
11-Octadecenoic acid	cis-Vaccenic Acid	C18:1(n-7)	MUFA	118	27.31	0.03	86
6-Octadecenoic acid	Petroselinic Acid	18:1(n-12)	MUFA	120	27.575	0.02	84
13-Docosenoic Acid	Erucic acid	C22:1(n-9)	MUFA	121	27.688	0.01	94
7-Octadecenoic acid	Fumaric acid	C18:1(n-11)	MUFA	161	35.362	0.13	25
cis-10-Heptadecenoic acid	None	C17:1	MUFA	124	28.008	0.14	93
10,13-Octadecadienoic acid	None	C18:2 (n-10)	MUFA	128	29.349	0.66	99
9-Octadecenoic acid	Oleic Acid	C18:1(n-9)	MUFA	129	29.425	2.56	99
cis-13-Octadecenoic acid	None	C18:1(n-5)	MUFA	129	29.425	2.56	99
11-Octadecenoic acid	cis-Vaccenic Acid	C18:1(n-7)	MUFA	129	29.425	2.56	99
cis-11-Eicosenoic acid	Gondoic	C20:1 (n-9)	MUFA	143	32.203	0.06	66
cis-13-Eicosenoic acid	Paullinic acid	C20:1(n-7)	MUFA	143	32.203	0.06	83
cis-10-Nonadecenoic acid	None	C19:1	MUFA	144	32.244	0.15	53
9,12-Octadecadienoic acid	Linoleic	C18:2(n-6)	PUFA	128	29.349	0.66	99

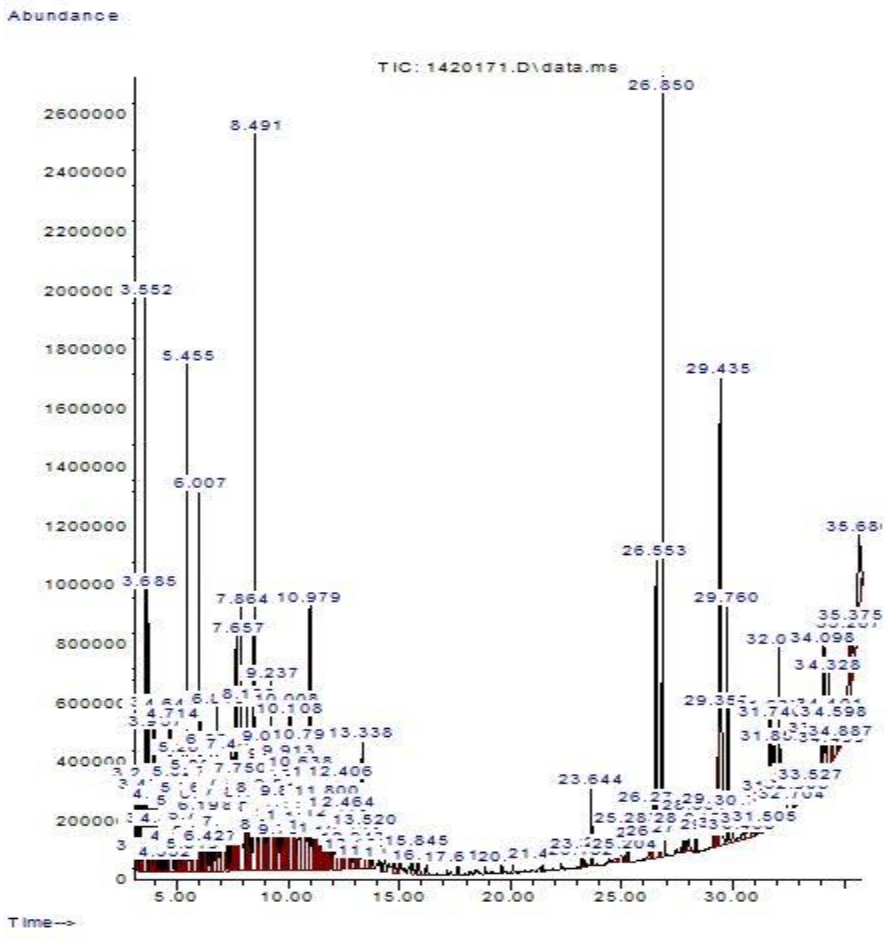


Figure: GC-MS Chromatogram of *Heterobranchus bidorsalis*

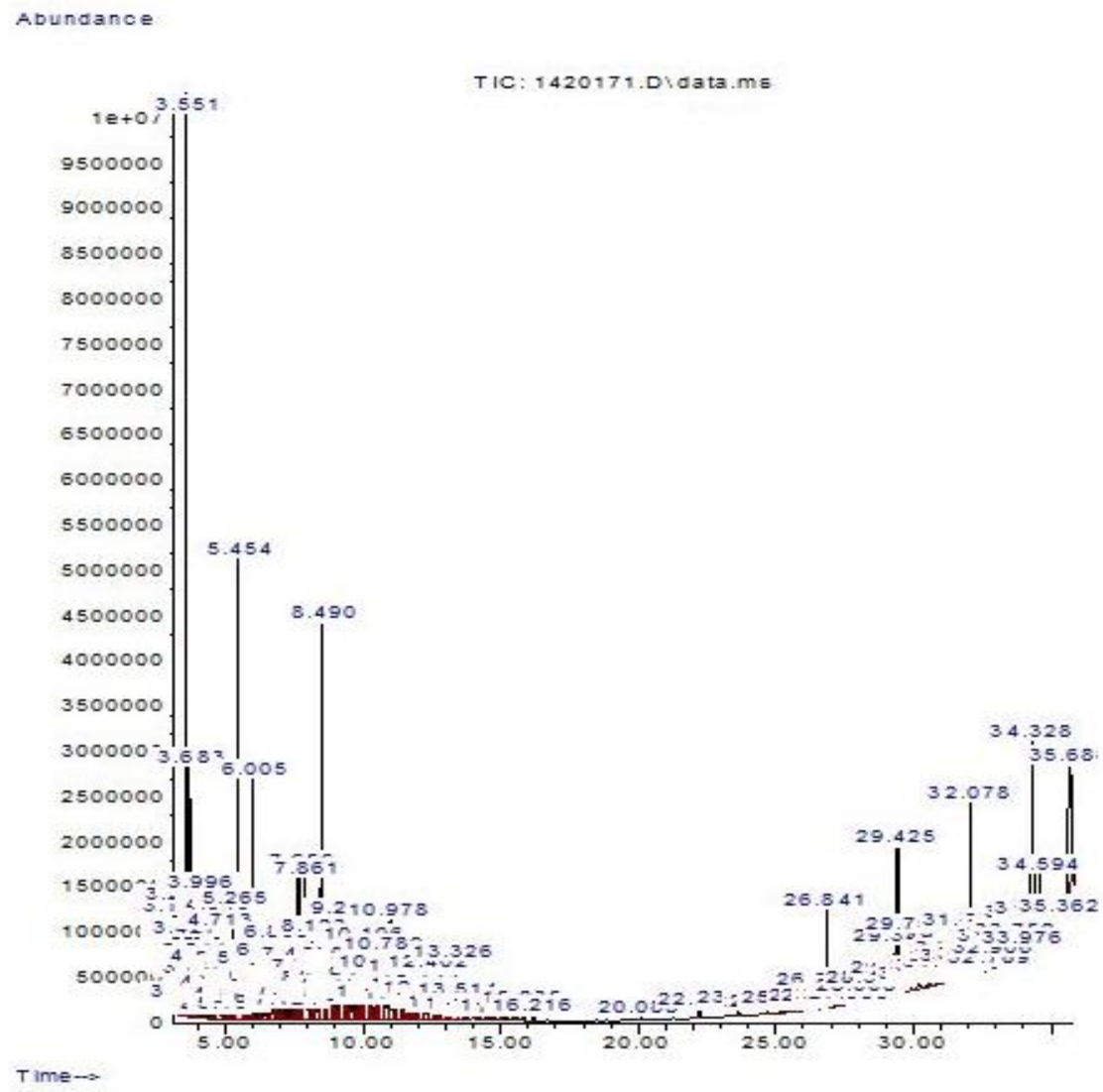


Figure: GC-MS Chromatogram of *Heterobranchus longifilis*

DISCUSSIONS

The nutritional benefits of fish stem for the most part, from its exceptionally advantageous fatty acids profile; this was reflected in this study with various fatty acids being identified. The resulting profiles revealed 17 fatty acids in *H. bidorsalis* and 19 fatty acids in *H. longifilis*, with saturated, monounsaturated, and poly-unsaturated fatty acid composition. Overall, there was little or no variation in fatty acid composition in the species of fish this could be because they are members of the same family from the same freshwater environment. Also, organisms living in aquatic environments with different ecological conditions are an important source of variation in nutritional components. Moreover, the aquaculture conditions and feeds used in fish aquaculture also cause variations in the fatty acid compositions of fish that were supplied to the market using aquaculture (Falk-Petersen, *et al*., 1998).

The resulting profiles revealed 17 fatty acids in *H. bidorsalis* and 19 fatty acids in *H. longifilis*, with monounsaturated fatty acid content being slightly higher than saturated and poly-unsaturated fatty acids. Myristic C14:0, lauric C12:0, pentadecylic C15:0, palmitic C16:0, margaric C17:0 palmitoleic acid C16:1(n-7), oleic acid C18:1(n-9), fumaric acid C18:1(n-11), and linoleic C18:2(n-6) were identified in both species while many other varieties of fatty acids where identified singly in the species (Table 1). Osibona *et al* (2009) in a study of freshwater fish species *Clarias gariepinus* and *Tilapia zillii* purchased from local fishermen in two landing sites in Lagos State identified that the fatty acids occurring in the highest proportions were myristic acid (C14:0, 4.2-5.2%), palmitic acid (C16: 0, 22.0-32.2%), palmitoleic acid (C16:1, 3.6-13.2%), heptadecanoic acid (C17:0, 0.7-3.0%), stearic acid (C18:0, 8.1-9.5%), linoleic acid (C18:2, 1.4-12.3%) and oleic acid (C18:1).

Unique and very important polyunsaturated fatty acids including roughanic, linoleic, arachidonic, and timnodonic acid were identified in *Heterobranchus* species which fall

under the *n*-3 and *n*-6 class of fatty acids. PUFA plays a vital role in alleviating many diseases such as cardiovascular disease, diabetes, inflammatory diseases, and rheumatoid arthritis, EPA and DHA are reported to have a beneficial effect on foetal development, proper neuronal and cellular functions, and healthy aging (Wang *et al.,* 1990). This result also agrees with Dhaneesh *et al*. (2012) with the diet because the human metabolism cannot create them from other fatty acids. DHA (docosahexaenoic acid) and AA (arachidonic acid) are both essential for proper brain and ocular development. Some other studies investigating the effect of the lack of n-6 and n-3 long-chain PUFA show its tremendous benefit on children with attention-deficit hyperactivity. Moreover, recent studies have also suggested that the *n*-6 family and its metabolites show beneficial effects on cardiovascular system health (Graham *et al*., 2007).

Furthermore, herbivorous, omnivorous, and carnivorous animals have varying intestinal lengths and morphologies, which affect nutrient digestion and absorption. Previous research has found that different fish families have distinct habits and traits, such as nutrition, nocturnal/diurnal habits, and deep/shallow water preferences, which lead to major variances in fish anatomy and physiology (Rodrigues et al., 2017).

Based on nutrients availability, when compared to marine microalgae, freshwater microalgae which are the major sources of these biomolecules contain higher levels of 18:3 n-3 fatty acids than EPA and DHA. Moreover, even though there is not a great amount of 18:2 n-6 fatty acids in marine microalgae, freshwater microalgae contain plenty of this fatty acid. This explains why freshwater fish require higher amounts of linoleic acid as compared to marine fish (Tocher, 2010)

Generally, species of catfish had an appreciable composition of fatty acids comprising of the saturated and unsaturated groups with a good quality of representation. These contents identified from freshwater species also reflect their availability which can be comparable with the marine species.

CONCLUSION

The findings of the present study present a good representation of SFA, MUFA, and PUFA fatty acids in *Hetrobranchus* species with palmitic, margaric, oleic, palmitoleic, fumaric, and linoleic dominating which are essential for the maintenance of structural and functional integrity of living beings, hence their useful contribution in global food security and economic development.

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