Fatty acid composition of a fatty fish using GC-FID and GC-MS analysis: comparative study

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ABSTRACT

The aim of this study was to profile the fatty acids in smoked fish oil using two GC-FID and GC-MS. To achieve this, fish (*Polydactilous quadrifilis*) from Youpwe fishermen smoked and cold pressed to extract the oil. The oil obtained was used for fatty acid profiling through methylation using the MeOH/KOH method before injection into a Stabil Wax®-DA GC-FID column and a SP 2560 GC-MS capillary column. GC-FID identified 30 FAMEs, with palmitic acid (C16:0) being the most abundant. Biologically active fatty acids such as docosahexaenoic acid (DHA, C22:6n3) and eicosapentaenoic acid (EPA, C20:5n3) were also identified at levels of 3.73% and 13.36%. Of the identified FAMEs, 48.71% were saturated and 51.79% were unsaturated. Polyunsaturated fatty acids were less abundant than monounsaturated ones, with ω-3 dominating this class. GC-MS detection of FAMEs and other compounds identified 72 FAMEs, 11 methyl ester FAMEs and 5 other compounds. Among the FAMEs identified in this library were non-conventional fatty acids such as C17:3n3, C16:1n10, C17:1n7, C18:2n9 and C23:6n3. Of the identified FAMEs, 17 are saturated and 55 are unsaturated. Comparison of the GC-MS and GC-FID profiles shows a similarity in the proportions of saturated and unsaturated fatty acids.

1. Introduction

Malnutrition and famine continue to wreak havoc around the world, particularly in Africa, where more than 1/3 of the child population is affected (FAO, 2010). These problems are compounded by the demographic boom, which puts pressure on already scarce local resources, leading to malnutrition (Pretty, 1999). Given this situation, seafood products such as fish could help alleviate the problems. Fish is the main source of protein for more than 400 million Africans and provides almost 25% of protein demand in 21 countries across the continent (Tacon & Metian, 2009). Cameroon is not to be outdone, with increasing consumption of several fish species, including *Merluccius merliccius*, *Sardinella aurita* (Khaoula et al., 2013), *Tilapia niloticus*, *Silurus glanis, Aurius parkii* and *Polydactilous quadrifilis* (Ali et al., 2011). *Polydactilous quadrifilis* is highly valued locally, particularly for its cost and fleshiness.

Fish flesh is a vital source of many nutrients, including proteins, minerals (phosphorus, calcium, iron and zinc), vitamins (B_{12}, A, D, E and K), amino acids and essential fatty acids (Ayeloja et al., 2023). It is highly prized throughout the world, where it is eaten either fresh (as in Japan) or after a number of processes such as boiling, frying, stewing and smoking (Ayeloja et al., 2023). Fish consumption is an integral part of the diets of people around the world, where it is considered to be the main source of essential amino acids and, in particular, long-chain unsaturated fatty acids with antimicrobial properties (Ayeloja & Yusuff, 2021). As a result, fish consumption continues to increase globally and in Africa. In fact, Ayeloja et al. (2023) reported in their paper the absence of cholesterol in freshwater fish, making this seafood product more attractive than red meat. Consumption of fish, particularly because of its DHA (docosahexaenoic acid) content, would therefore limit the incidence of coronary heart disease, type 2 diabetes, cancer and inflammatory diseases and promote fetal growth (Innis et al., 1991).

Fish oil is obtained by chemical or physical extraction. Its chemical composition varies and is basically composed of triglycerides, which contain saturated and unsaturated fatty acids of different chain lengths on the carbons of glycerol, as well as phospholipids, cerides and very few cholesterides (Mgbechidinma et al., 2023). The chemical and biological quality of this product depends on a number of parameters, such as size, age, sex, diet, geographical area of growth and cooking treatments (Danae et al., 2010). The latter modify the fatty acid composition of fish oil, which is considered one of

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the most complex due to its composition of ⍵-3 and ⍵-6 unsaturated fatty acids such as DHA and eicosapentaenoic acid (EPA), which have numerous biological properties (Abdulkadir et al., 2010). These long-chain polyunsaturated fatty acids are likely to undergo oxidation reactions during these treatments, resulting in the loss of their properties. Smoking is very often used and recommended cooking methods, as it preserves the fish well, particularly by limiting the oxidation of its lipids (Njinkoue et al., 2017). In fact, chemical modifications lead to the appearance of new fatty acid molecules and the transformation of others (Khaoula et al., 2013). In addition to the well-known conventional fatty acids, this oil contains many other free and esterified fat molecules that would remain unidentified if the methods used did not have a wide range of standards. A lot of work has therefore been done on fatty acid characterization in fish oils, all by GC-FID, where only a small number of FAMEs have been identified. The aim of this study was therefore to identify the fatty acid profile of a smoked fish species using GC-FID and GC-MS.

2. Materials and Methods

2.1. Experimental design

The study is descriptive and quantitative, taking into account acid profiles and proportions.

2.2. Material

The materials used in this study were chemical, physical and biological. The chemical material consisted of heptane (HPLC grade), methanol (HPLC grade) and potassium hydroxide (KOH) obtained from Biosolve Chmie Sarl (57260 Dieuze, France). Sample storage tubes and vials for injection into the various instruments were obtained from SIGMA-ALDRICH Corporation (3050 Saint Louis Missouri, United States of America). The super-vortexer for perfect homogenization of the solutions was supplied by LAB-LINE Instruments, Inc (MELROSE PARK ILL, United States of America). The analytical equipment used was supplied by Interscience Thermo Electron Corporation (Science Park Einstein/1348 Louvain La-neuve, Belgium). The Trace GC-Ultra gas chromatograph coupled to a flame ionization detector (GC-FID) was supplied by Interscience Thermo Electron Corporation (Science Park Einstein/1348 Louvain La-neuve, Belgium) and the SP 2560 gas chromatograph coupled to a mass spectrometer (GC-MS) was supplied by SHIMADZU-EUROPA GmbH (Germany).

The biological material consisted of a species of oily fish (*Polydactilus quadrifilis*) that was purchased fresh from fishermen in Youpwe (Douala, Littoral Region, Cameroon) and then transported under ice in a cooler (fish/ice ratio 1:2, w/w) to the Laboratory of Food Science and Nutrition, Department of Biochemistry, University of Douala. Prior to transport, the fish were inspected by the veterinary services of the Ministry of Wildlife, Fisheries and Animal Industry of Cameroon to ensure that they were of hygienic quality (no chemical, physical or microbiological deterioration).

2.3. Methods

Extraction of fish oil (Polydactilous quadrifilis)

After the fish of the species *Polydactilous quadrifilis* (5 fish) were collected from the Youpwe fishermen and transported to the laboratory in a cool box, they were eviscerated with a stainless steel knife before smoking. The smoking process took place in three stages: in the first stage, the fish were pre-cooked in an automatic smoking chamber (Chokor type smoker, GIC la Compétence, Cameroon). The fish were placed on grids at a distance of 1.30 m from the heat source for 150 min at 40 °C. At the end of this stage, the fish were cooked for 480 min at 85 °C and finally dried at 55 °C for 120 min (Dama et al., 2021). After smoking, the oils were obtained by cold mechanical pressing. For this, 500 g of fish fillets were weighed and ground in an electric blender (Singsung BL-500, Singapore). The resulting grind was bagged in muslin and placed in a hydraulic press where the force generated by the piston was used to extract the soluble components of the fish. The collected mixture was decanted using a separating funnel to separate the oily phase from the non-oily phase. The oil was collected in Eppendorf tubes and stored at -15 °C prior to fatty acid profiling.

Derivatization of fatty acid prior to analysis using MeOH-KOH method

Derivatization aims to convert fatty acids (FAs) into methylated fatty acids (FAMEs), which are more volatile and therefore easier to detect. The MeOH-KOH method described by Cruz-Hernandez et al. (2004) was chosen because it is faster and gives similar results to the long hexane/MeOH/MeOH-BF3 method. A drop of oil equivalent to 10 mg was taken with a Pasteur pipette and placed in a 5 mL test tube to which 2 mL of heptane was added, followed by 200 µL of 2 M MeOH-KOH reagent. The mixture was stirred manually and then mechanically for 1 min at a speed of 900 rpm using a LAB-LINE INSTRUMENTS, Inc. electric super-mixer (MELROSE PARK ILL, United States of America). The mixture was allowed to stand for 15 min and then 1 mL of the upper clear phase containing the FAMEs was collected with a micropipette and transferred to the vials for injection and profiling of these fatty acids.

Fatty acid analysis using Gas Chromatography coupled to a Flame Ionization Detector (GC-FID)

The profiling and quantification of FAMEs initially required a gas chromatography (Trace GC-Ultra) coupled to a flame ionization detector (GC-FID). The instrument was supplied by Interscience Thermo Electron Corporation (Science Park Einstein/1348 Louvain La-neuve, Belgium) and fitted with an AI 3000 auto-injector from the same company. The column used was Stabil Wax®-DA (30 m x 0.25 mmID x 0.25 um inner diameter film) access number 1459753, USA. The oven temperature varied from 50 to 250 °C at a rate of 3 °C/min with isothermal stages of 10 min and a final time of 20 min. The injector and detector temperatures were 250 °C and 270 °C respectively. The mobile phase (carrier gas) had the following characteristics: air flow rate of 300 mL/min and helium transport velocity of 25 cm/s at 250 °C. Once these parameters had been checked and set, 1 µL of solution was automatically injected in split mode at a ratio of 10:1. FAMEs were detected using a flame ionization detector (FID) and each methylated fatty acid peak was acquired at a frequency of 100 Hz. Peak identification was possible by comparing the retention times of the sample FAMEs with those of the SUPELCO standard containing 37 known FAMEs using a Philips desktop (Priminfo, Belgium) equipped with Thermo Scientific Dionex Chromeleon 7 (Chromatography Data System version 7.3) and ChromSpace version 1.5.1 (Markes International Limited) software.

Fatty acid analysis using Gas Chromatography coupled to a Mass Spectrometer (GC-MS)

The identification of FAMEs, esterified FAMEs and other compounds present in the fish oil was carried out using gas chromatography-mass spectrometry (GC-MS). The SHIMADZU Nexis GC-2030 (GC-MS TQ 8050 NX, Europa Gmbl, Germany) was equipped with a low-polarity SP 2560 capillary column (100 m x 0.25 mmID x 0.20 µm inner diameter film) and a SHIMADZU AOC 20i Plus autoinjector. The oven temperature varied from 50 to 250 °C at a rate of 10 °C/min with isothermal steps of 1 min and a final time of 20 min. The temperature of the injector and detector was maintained at 250 °C. The flow rate after injection of the solvent (hexane) and diluted solutions was 1.08 mL/min, while the helium responsible for transporting the molecules did so at a speed of 25 cm/s at 250 °C. After set these parameters, the solvent was self-injected and after 40 min of elution, 1 µL of diluted 1:10 solution was self-injected in split mode at 60 psi at a temperature of 250 °C for 1 min. The molecules were detected on a mass spectrometer of the same make, calibrated at 200 and 250 °C for the electron source and interface temperatures respectively. Over the range m/z 35-550, EI mass spectra were acquired at 70 eV in full scan mode. Peak identification of the different groups of compounds was possible using the NIST17s 4b FAMEs library and the GC-MS post-analysis Chrom Compare T1 (ChromSpace) software installed on a Philips desktop (Priminfo, Belgium). The percentage of similarity applied to the identification of each peak was 90%.

3. Results and Discussion

The FAME profile of smoked fish performed by GC-FID and presented in Table 1 shows the presence of 30 FAMEs of different lengths and types. This number is close to the 29 FAMEs identified by Ayeloja et al. (2024) in three marine fish species. The length of the FAMEs varied from 8C to 24C, with the majority being long chain FAMEs. Ayeloja et al. (2024) also found a FAMEs between 8 and 24C in smoked marine fish. Separating the labelled FAMEs according to retention time shows that the appearance of the peaks of the different FAMEs evolves positively with the carbon number and the number of unsaturations. For an equivalent carbon number, the saturated FAMEs take less time to leave the column. The same observations were made by Ayeloja et al. (2024). Of the FAMEs identified, 13 were saturated and 17 were unsaturated. The unsaturated FAMEs were caproic (C8:0), capric (C10:0), undecanoic (C11:0), lauric (C12:0), myristic (C14:0), palmitic (C16: 0), heptadecanoic (C17:0), stearic (C18:0), arachidic (C20:0), docosanoic (C22:0), tricosanoic (C24:0) and finally lignoceric (C24:0). These unsaturated fatty acids were also identified in *Trachurus trachurus* oil by Ayeloja et al. (2024), with the exception of caproic (C8:0), capric (C10:0), lauric (C12:0) and myristic (C14:0) acids. This variation can be explained by the extraction method, which led to a loss of these fatty acids, and by the derivatization method. Among these saturated FAMEs, palmitic acid was the most abundant (27.75%), followed by myristic (14.93%) and stearic (4.06%) acids. Decanoic and undecanoic acids were the least abundant with a content of exactly 0.01%. Similar observations were also made by Tenyang et al. (2020) in smoked fish oil extracted from *Chrisygtus nigrodigitatus*, where palmitic acid was found to be the most abundant (23.47%) while capric acid was the least

abundant (0.10%). As for the unsaturated fatty acids, 9 were in the polyunsaturated form (PUFAs). The PUFAs identified included trans-linoleic acid (C18:2n6), γ-linolenic acid (C18:3n6), α-linolenic acid (C18:3n3), eicosadienoic acid (C20:2), γ-eicosatrienoic acid (C20:3n6), α-eicosatrienoic acid (C20:3n3), arachidonic acid (C20:4n6), eicosapentaenoic acid or EPA (C20:5n3) and finally docosahexaenoic acid or DHA (C22:6n3) for proportions of 1.04%, 0.45%, 0.35%, 0.03%, 0.28%, 1.58%, 0.02%, 13.36% and 3.73% respectively. The values obtained are higher than those of Tenyang et al. (2020), especially for EPA and DHA, which can be explained by the variation in smoking conditions, season of collection, oil extraction method, fish feed and fish species. These results also suggest good physiological activity, particularly antimicrobial and anti-inflammatory, due to the high levels of EPA and DHA. The results also contradict those of Tenyang et al. (2020) with regard to EPA and DHA levels. In fact, these authors noted the abundance of DHA as opposed to EPA in catfish and red carp oils. This could be explained by the abundance of EPA phospholipids in these fish.

Table 1. Fatty acyl methylated esters of smoking (*Polydactilus quadrifilis*) by GC-FID.

No	Peak Name	Retention Time (min)	Relative Area (%)
1	C8	4.169	0.26
2	C10	5.195	0.01
3	C11	5.734	0.01
4	C12	6.455	0.14
5	C14	8.449	14.93
6	C14:1	8.872	0.05
7	C15:0	9.697	0.67
8	C15:1	10.137	0.03
9	C16:0	11.284	27.75
10	C16:1	11.657	22.34
11	C17:0	12.729	0.38
12	C17:1	13.260	0.04
13	C18:0	14.531	4.06
14	cis and trans C18:1n9	14.862	7.46
15	trans C18:2n6	15.634	1.04
16	C18:3n6	16.172	0.45
17	C18:3n3	16.782	0.35
18	C20:0	18.026	0.18
19	C20:1n9	18.351	0.40
20	C20:2	19.196	0.03
21	C20:3n6	19.680	0.28
22	C20:3n3	20.082	1.58
23	C20:4n6	20.365	0.02
24	C20:5n3	21.316	13.36
25	C22:0	21.523	0.13
26	C22:1n9	21.864	0.05
27	C22:2	22.654	0.08
28	C23:0	23.243	0.06
29	C24:0	24.911	0.13
30	C22:6n3 and C24:1n9	25.276	3.73

The low level of arachidonic acid in this oil is thought to be related to the absence of its precursor, cis-linoleic acid (C18:2n6) (Tenyang et al., 2020). In terms of monounsaturated FAMEs, the most abundant was C16:1 palmitoleic acid (22.34%), followed by cis-C18:1 oleic acid and trans-C18:1 elaidic acid (7.46%). The presence of elaidic acid is not consistent with the work of Tenyang et al (2020) who did not identify this fatty acid in smoked fish. Pentadecenoic acid was the least abundant of the MUFAs with

0.03%. These results agree with those obtained by Tenyang et al. (2020) and Ayeloja et al. (2024), who did not find this fatty acid in the fish oils studied, confirming that it is rare in fish. These analyses show that fish oils are sources of a variety of fatty acids, which require using different methods and standards to study them.

Profiling of FAMEs in smoked fish oil using GC-MS and the NIST17s 4b FAME library identified 72 compounds, including alkanes, FAMEs (saturated, polyunsaturated and monounsaturated), ketones and esterified fatty acids (Table 2). This large number of compounds is thought to be related to the treatments applied, the fish feed and the climatic conditions in the production area. This profile shows the presence of a variety of compounds, such as esters and ketones, which are responsible for the biological and sensory properties of the fish. The presence of alkanes such as heneicosane, 1,3-dichlorobenzene, dotriacontane and nonadecane is evidence of the modification (oxidation) of the oil when the fish is smoked. Although carcinogenic, they protect against microbial attack (Hatab et al., 2012). There is also an alcohol in the form of 2-hexyl-1-decanol, which is thought to result from the breakdown of fatty acids during heat-induced auto-oxidation reactions. 4-Hydroxy-4-methyl-2-pentanone, a ketone compound, was also identified in this oil. The presence of this compound would indicate oxidative activity and degradation in the fish prior to processing. This compound is thought to be responsible for the flavour and antimicrobial activity of the fish oil due to its size (Aponte et al., 2014). Esterified compounds result from the combination of alcohols and fatty acids with the loss of water. The presence of these compounds at the beginning of Undecanoic acid, 2,6,10-trimethyl-, methyl ester; Hexadecanoic acid, methyl ester; Tridecanoic acid, 12-methyl-, methyl ester; Tridecanoic acid, 4,8,12-trimethyl-, methyl ester; Hexadecanoic acid, 3,7,11,15-tetramethyl-, methyl ester; Me. C15:1n5, Pentadec-(10Z)-enoate <methyl->: Hexadecanoic acid, 3,7,11,15-tetramethyl-, methyl ester; 9(E)-Octadecenoic acid, methyl ester; Heneicosapentaenoic acid, methyl ester; Heneicosapentaenoic acid, methyl ester; 5,8,11-Eicosatrienoic acid, methyl ester and omega.-3 arachidonic acid methyl ester. Arachidonic acid methyl ester is responsible for the strong flavour and aroma often attributed to this fish species. What's more, these compounds are presented as antioxidants and, above all, as therapeutic agents, as they are able to create tensions and distortions in the biological membranes of pathogenic micro-organisms (Ali et al., 2011). Their presence thus shows that the activities often attributed in the literature to fish fatty acids alone are not fully verified. It also suggests that special care should be taken when catching, handling and processing fish, as the presence of these compounds may cause them to deteriorate. For FAMEs, all 30 identified by GC-FID except caproic acid (C8) were detected by GC-MS. In addition to these FAMEs (30), more than twenty other FAMEs were identified, including numerous isomers. This is the case for C15:0, where iso and anteiso isomers have also been found in this oil. Palmitoleic acid (C16:1n9) is not to be outdone, with the presence of numerous positional isomers (C16:1n5, C16:1n7, C16:1n10), substituted variants (Me.10-Me C16:0 and Me.9,10-methylene C16:0) and another with more than one unsaturation (C16:2n4). Many other new FAMEs in the C18 class were identified, including Me. C18:1n12, Me. C18:2n9, Me. C18:2n7, Me. C18:1n12, Me. C18:1n6, Me. C18:1n9 and Me. C18:4n3. The presence of this new variety of FAMEs of this class detected by GC-MS demonstrates the need to use these two methods for better profiling of fish oils, which are known to be very complex. It

also suggests that the biological activities of the essential fatty acids (C18:2n6 and C18:3n3) present in fish oils are enhanced by the chemical diversity of the other fatty acids of the same class.

Table 2. Fatty acyl methylated esters of smoking (*Polydactilus quadrifilis*) by GC-MS.

Peak name	Targetting	M-H	Retention time (min)
1-Decanol, 2-hexyl-	Target	57	15.143
Heneicosane	Target	57	15.277
Benzene, 1,3-dichloro-	Target	146	15.427
2-Pentanone, 4-hydroxy-4-methyl-	Target	59	16.523
Nonadecane <n->	Target	57	16.633
Me. C12:0	Target	74	17.983
Me. C13:0	Target	74	19.187
Tridecanoic acid, 12-methyl-, methyl ester	Target	74	19.847
Tridecanoic acid,4,8,12 - trimethyl-, methyl ester	Target	87	20.538
Me. C14:0, Myristate <methyl->	Target	74	20.685
Me. C15:0 iso	Target	74	21.447
Me. C15:0 anteiso	Target	74	21.838
Me C16:1n9	Target	55	21.953
Me. C14:1n5	Target	55	22.24
Me. C15:0	Target	74	22.405
Undecanoic acid, 2,6,10-trimethyl-, methyl ester	Target	88	23.163
Hexadecanoic acid, methyl ester	Target	74	23.367
Me. C18:1n12	Target	55	23.968
Me. C15:1n5, Pentadec-(10Z)-enoate <methyl->	Target	55	24.135
Me. C16:0	Target	74	24.6
Me.10-Me C16:0	Target	74	25.06
Me. C16:1n10	Target	55	25.595
Me. C16:1n7	Target	55	25.755
Me. C16:1n7, Palmitoleate <methyl->	Target	55	25.867
Me. C16:1n10	Target	55	26.085
Hexadecanoic acid, 3,7,11,15 - tetramethyl-, methyl ester	Target	101	26.187
Me. C16:1n9	Target	55	26.423
Me. C16:1n5	Target	55	26.698
Me. C17:0	Target	74	26.93
Me. 9,10-methylene C16:0	Target	55	27.252
Me. 11-Me C13:0	Target	74	28.183
Me. C16:2n4	Target	67	28.315
Me. C18:2n9	Target	67	28.455
Me. C20:2n6	Target	67	28.658
Me. C17:1n7	Target	55	28.888
Me C22:1n11	Target	97	29.22
Me. C18:2n7	Target	67	29.355
Me. C18:0	Target	74	29.753
Me. C18:3n6	Target	67	30.573
Me. C18:1n12	Target	55	31.13
Me. C20:4n3	Target	79	31.663
9-Octadecenoic acid, methyl ester, (E)-	Target	55	31.812
Me. C18:1n9	Target	55	32.073
Me C22:1n11	Target	55	32.265
Dotriacontane <n->	Target	57	32.388

Table 2. Continue

Me. C18:1n6	Target	55	32.497
Me. C19:0	Target	74	32.817
Me. C22:6n3	Target	79	33.627
Me C20:2n6	Target	82	33.707
Me. C18:2n9	Target	67	34.24
Me. C18:3n6	Target	79	34.443
Me. C18:2n6	Target	67	34.62
Me. C18:2n6	Target	67	34.958
Me. C18:2n6	Target	67	35.713
Me. C20:0	Target	74	36.22
Me. C18:3n6	Target	79	37.47
Me. C16:1n10	Target	55	38.228
Me. C17:3n3	Target	79	38.423
Me. C20:1n9	Target	55	38.543
Me. C22:3n4	Target	79	38.915
Me. C25:0	Target	74	39.865
Heneicosapentaenoic Acid methyl ester	Target	79	40.49
Me. C18:1n9	Target	67	41.325
Me. C18:4n3	Target	79	41.69
Me. C20:2n6	Target	81	42.165
Me. C20:2n6	Target	81	43.06
5,8,11-Eicosatrienoic acid, methyl ester	Target	79	43.378
Me. C22:0	Target	74	43.7
Me. C19:3n6	Target	79	44.935
Me. C20:3n6	Target	79	46.022
Me. C22:1n10	Target	55	46.177
Me C22:3n4	Target	91	46.533
Me. C22:1n10	Target	69	46.613
Me. C20:4n6	Target	79	47.088
Me C22:4n6	Target	91	47.433
.omega.-3 Arachidonic Acid methyl ester	Target	79	49.45
Me. C20:5n3	Target	79	50.208
Me. C22:5n7	Target	79	50.832
Me. C20:5n3	Target	79	51.852
trans-Nervonate <methyl->	Target	55	54.203
trans-Nervonate <methyl->	Target	55	54.648
Me. C22:4n6	Target	79	55.603
Me. C21:5n3	Target	79	56.445
Me. C22:5n7	Target	79	57.558
Me. C10:0	Target	74	16.147
Me. C21:5n3	Target	79	60.382
Me. C23:6n3	Target	79	62.452
Me. C24:5n3	Target	79	68.227
Me. C24:6n3	Target	79	71.228

Research and evaluation of the activities of these other fatty acids in the near future would help to better assess their importance for human consumption. The 19C-FAMEs, the trans-nervonate, which is very important for brain membranes, and the substituted 13C-FAME, which was not detected by GC-FID, were also listed among the fats present in this oil. In addition to the fatty acids of the 20C series, such as arachidonic acid, which has been shown to have antimicrobial, cholesterol-regulating and antihypertensive activities, others such as Me. C20:4n3. In fact, this fatty acid of the Ɯ-3 series could be essential for the formation of biological membranes, antimicrobial control, energy source, psychomotor development, gene expression, proper liver function and membrane fluidity (Tenyang et al., 2014). In the series at 21C and above, many other new FAMEs with as yet unproven properties were identified, following the example of Me. C22:6n3, Me. C22:3n4, Me. C25:0, Me. C22:1n10, Me.

C22:1n11, Me C22:3n4, Me C22:4n6, Me. C22:5n7, Me. C21:5n3, Me. C22:5n7, Me. C23:6n3, Me. C24:5n3 et Me. C24:6n3.

The general profile of the compounds identified and quantified by the two methods is presented in Table 3. This table shows that of the 72 compounds identified by GC-MS, 17 were in saturated form, representing 23.61% of the total composition, whereas by GC-FID, 48.71% of the 30 FAMEs were in saturated form. These results are confirmed by the semi-liquid appearance of this oil at room temperature. They are also close to the values (45 to 54%) found by Tenyang et al. (2014) on different fish species from the Cameroonian coast. Unsaturated FAMEs were 51.29% for GC-FID quantification and 55(76.38%) for GC-MS profiling. Of these unsaturated FAMEs, 20.92% were polyunsaturated and 30.37% were monounsaturated. On the other hand, GC-MS profiling revealed more PUFAs, but as the proportions were not determined, it cannot be said that this large number is proportional to the content of this class of FAMEs. Similar observations were also reported by Tenyang et al. (2020), leading to the conclusion that the composition of fish from the Cameroonian coast is similar. The composition of ω-6, ω-3 and the ω-3/ω-6 ratio are used to define the nutritional contribution of fish oils (Tenyang et al., 2014). The higher the ω-3 content, the higher the ratio and, as a result, people who eat these fish have a lower risk of cardiovascular disease (Wardlaw et al., 1992). The proportion of ω-3 is greater than that of ω-6, although GC-MS revealed the presence of more ω-6. Although this proportion of ω-3 is higher than that of Tenyang et al. (2014), these authors also showed that ω-6 levels were lower than ω-3 levels in six fish species. However, the ω-3/ω-6 ratio found in this study is much higher than that of Tenyang et al. (2020), which were 1.3 and 1.60 in smoked fish oils, respectively. The ratio of 10.63 obtained is very high compared to the standard, which is a maximum of 5 for oils intended for human consumption. Consumption of these fish could therefore lead to an imbalance in the fatty acid intake of the consumer. The PUFAs/SFA ratio is close to the 0.45 recommended for edible oils (Wood et al., 2008). GC-MS profiling revealed the presence of 11 fatty acid esters, 27 unsaturated FAMEs of other classes and 5 other compounds, mainly alkanes.

Table 3. Fatty acid profile according to different classes in fish lipid extract

Classification	GC-MS	GC-FID (%)
Methylated fatty acids (n)	**72**	**30**
Saturated Fatty acid (SFA)	17	48.71
Total Unsaturated Fatty Acid	55	51.29
PolyInsaturated Fatty Acid (PUFAs)	34	20.92
MonoInsaturated Fatty Acid (MUA)	21	30.37
ω-6	15	1.79
ω-3	11	19.02
Ratio ω-3/ω-6	/	10.63
Ratio PUFAs/SFA	/	0.43
Others classes of Insaturated Fatty Acid	27	/
Methyl ester	11	/
Other compounds	5	/

n: number of fatty acids.

4. Conclusions

At the end of this study, which aimed to determine the fatty acid profile of a fatty fish oil using two chromatographic methods (GC-FID and GC-MS), it come out that the general

composition of the fatty fish was not really affect by the profiling method. However, the GC-MS analysis revealed the presence of a greater variety of FAMEs (72) and other molecules whose study would reveal a biological interest. Palmitic acid was the most abundant FAMEs in this oil, while unsaturated fatty acids were the more abundant classes of FAMEs. These results also demonstrate the value of combining the two methods when profiling fats.

Data accessibility

Ethical statement

The author have read and followed the ethical requirements for publication in Applied Food Research and confirm that the current work does not involve any human subjects, animal experiments, or any data collected from social media platform.

Credit author statement

Stephano Tambo Tene: Conceptualization, Methodology, Software, Data curation, Writing- Original draft preparation.

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Declaration of interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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