



NUTRIENT COMPOSITION ASSESSMENT AND GC-MS ANALYSIS OF CHUB MACKEREL
(*Scomber japonicus*) MEAL AND OIL

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ABSTRACT

Fish and fish products are widely consumed by most population, and have been associated with a lot of health benefits. The present study was undertaken to evaluate the nutrient composition of *Scomber japonicus* meal and oil. Proximate composition of fish meal, fatty acid profile and physicochemical properties of fish oil were analyzed using standardized methods. Results of proximate analysis revealed that the fish meal contain 8.56% moisture content, 4.05% ash, 26.23% crude fat, 13.18% crude protein and 47.98% total carbohydrate. Fatty acid composition of the fish oil comprises tetradecanoic acid 8.99%, 9-hexadecenoic acid 4.84%, hexadecanoic acid 16.73%, 9,12,15-octadecatrienoic acid 5.66%, 7-octadecenoic acid 15.90%, 8,11,14-docosatrienoic acid 3.50%, octadecanoic acid (stearic acid) 2.51%, 11-eicosenoic acid 11.37%, 5,8,11,14,17-eicosapentaenoic acid (EPA) 13.03%, 4,7,10,13,16,19-docosahexaenoic acid (DHA) 1.07%, 13-docosenoic acid 15.66%, 9-octadecenoic acid 0.74%, saturated fatty acid 28.23%, monounsaturated fatty acids 48.51%, polyunsaturated fatty acid 23.26%. Physicochemical properties of the oil also revealed iodine value of 140.82g/100g, saponification value of 252.46 mg/g, free fatty acid value of 4.89 mg/g, peroxide value of 2.21 meq/kg, acid value of 9.82 mg/g and refractive index of 0.883. In conclusion, chub mackerel's (*Scomber japonicus*) meal can be a good source of energy, protein and mineral elements, with potential health benefits of the oil due to its high stability and predominant unsaturated fatty acids

Keywords: Fish meal, Fish oil, Health benefits, Nutrient composition, *Scomber japonicus*

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INTRODUCTION

Malnutrition which includes protein energy malnutrition, micronutrient deficiencies and over nutrition is a public health problem in developing countries [1]. Fish and fish-derived products are rich in nutrients, providing 15 to 20% protein content, making them an effective addition to complement the cereal and tuber-based diets that are commonly consumed across Africa. [1, 2]. Sea food especially fish are an important part of the human diet across the world. Nutritional importance of fish is well documented and consumption is associated with not only protein and minerals but also largely with *omega-3*-polyunsaturated fatty acid (PUFA) content [3, 4]. Fish is known to be a source of protein rich in essential amino acids (lysine, methionine, cysteine, threonine, and tryptophan). Fish muscle also contains micro- and macro elements and fat-soluble vitamins [5]. Fish oils have long been a natural constituent of human diet, and have been reported to be hypocholesterolemic [6]. They contain essential fatty acids especially oleic and linoleic acids as well as other polyunsaturated fatty acids [7, 8]. Fish oils are the richest known sources of vitamins A and D [4],

while fish muscle is a good source of vitamin D [9, 10]. However, there is limited data on local species in this region as they have not been extensively studied, leaving gaps in nutrient profiles specific to this region. Environmental factors like local water quality, temperature, and pollution could all have significant effects on the nutrient composition and physicochemical properties. Date will also help in comparison with species from other regions. The aim of this study is to investigate the nutrient profile and physicochemical properties of *Scomber japonicus* oil and meal and also to evaluate its fatty acid composition using GC-MS analysis.

MATERIALS AND METHODS

Sample collection and preparation

Mackerel (*Scomber japonicus*) (15-20 cm in length and 350-400 g in weight) were purchased from Sabon Gari market, Zaria. This was thoroughly Washed, degutted and cut into medium sizes prior to oven drying at 40°C for 72 hours. The dried fish sample was manually ground and homogenized into powder. The weight of the sample was taken before and after drying.

Extraction of fish oil

The fish oil was extracted using Soxhlet extractor with n-Hexane as the solvent. The weight of the sample was taken in a porous thimble covered with cotton wool, before it was placed in the inner tube of the apparatus fitted to a round bottom flask. The solvent was boiled for 1 hour, and the oil was collected through reflux. The solvent used was later recovered by applying heat and collected above the round bottom flask into the soxhlet apparatus

Determination of moisture content

Moisture content was determined according to method of Association of Official Analytical Chemists (AOAC) [11]. The sample was placed in a moisture dish and then dried in an oven at 105°C until constant weights was obtained.

Determination of ash content

Ash content was determined according to method of AOAC [11]. Pre-dried samples obtained from moisture content analysis were ashed in a furnace at 550°C overnight.

Determination of crude lipid

A cleaned, heat-resistant conical flask (250 ml) was dried in an oven, which was then transferred into a desiccator and allowed to cool. Two (2 g) of the sample was added into labeled thimbles (filter paper). The heat-resistant flask was filled with petroleum spirit or N-hexane where applicable. The soxhlet apparatus was assembled and then allowed to reflux for 8 hours. The thimble was removed and transferred to an oven for drying and cooling followed by weighing.

% Crude fat was calculated as: $\frac{\text{Weight of fat} \times 100}{\text{Weight of sample}}$

Determination of crude protein

Kjeldhal method of AOAC [12] was used for crude protein determination. Two (2) g of sample was weighed into a Kjeldhal flask and 1 g of CuSO₄ and 25 ml of conc. H₂SO₄ were added. The flask was placed on a Kjeldhal digestion rack and heated in the fume cupboard until a green color solution was obtained. The flask was cooled and the sample transferred into a 250 ml volumetric flask and made up to the mark with distilled water. Ten (10) ml of the digest was pipetted into the Markham distillation apparatus, followed by addition of 10 ml NaOH (40%). The sample was then steam distilled, liberating ammonium sulphate into a 100 ml conical flask containing 10 ml of 4% boric acid, using methyl blue as an indicator. The content of the conical flask was then titrated with 0.01N HCl. Percentage Nitrogen and crude protein content of the sample was calculated using the formulae below.

$$0.014 \times \text{VD} \times \text{N} \times 100 \times \text{TV}$$

$$\% \text{ Nitrogen} = \% \text{ Protein} = \% \text{ N} \times \text{F}$$

Weight of sample x AD

Where, VD = Volume of digest, N = Normality of acid, TV = Titre value, AD = Aliquot of digest,

F = Conversion factor for nitrogen to protein (6.25).

Determination of total carbohydrate

The carbohydrate percentage was calculated by difference, following the method outlined by Pearson and Cox [13] thus;

Percentage total carbohydrate = 100 - (% moisture + % Ash + % Crude fat + % Crude Protein)

Gas Chromatography- Mass Spectrometry (GC-MS) analysis of fish oil

GC-MS analysis was performed in two consecutive steps. First the preparation of fatty acid methyl ester as described by AOAC [14, 12]. Fatty acid methyl ester was produced by extracting lipids from the sample, followed by heating with methanolic sodium hydroxide (NaOH) for saponification, and subsequently using methanolic hydrogen chloride (HCl) or boron trifluoride (BF₃) to complete the esterification process. Five (5) ml hexane was added to recover the methyl ester in organic phase. Saturated NaCl solution was added to the mixture and the aqueous and organic layer were separated using a separating funnel. The upper hexane layer was pipetted and used for GC-MS analysis by injecting into the GC-MS system, where it is vaporized in the injector and passed through a gas chromatograph column, where components are separated based on their volatility and interaction with the column's stationary phase. The separated components enter the mass spectrometer, where they are ionized, fragmented, and detected. Fatty acids were identified by comparison with the retention time and length of side chain.

Determination of iodine value

Iodine Value was determined by method of AOAC [11]. 0.2 g of fish oil sample was weighed into quick fitted conical flasks, then 15 ml of CCl₄ and 25 ml W_{ij}'s reagent were added into each flask and placed in the dark for one hour. Afterwards, 20 ml of 10% Potassium Iodide (KI) and 150 ml distilled water were added. Resulting mixture was titrated against 0.1M Na₂S₂O₃ using 1% starch solution as indicator. A blank test was also carried out simultaneously under the same condition. The iodine value of oil samples was determined in relation to the blank test.

Determination of saponification value

Saponification Value was evaluated according to the method described by AOAC [12]. Two (2) g of fish oil sample was weighed into a 250cm³ quick fitted conical flask and 25ml of freshly prepared 0.5M ethanolic potassium hydroxide was added. The mixture was boiled under reflux for 1 hour and then

titrated with 0.5M hydrochloric acid using phenolphthalein indicator. A blank determination was carried out and the saponification value (SV) determined.

Determination of acid value

A solvent mixture of 95% ethanol (25 ml) and 25 ml diethyl ether were neutralized with 0.1M ethanolic KOH using phenolphthalein as indicator. Four (4) g of sample oil was dissolved in the neutralized solvent mixture and titrated with 0.1M KOH [12].

Determination of peroxide value

Peroxide value was determined according to the procedure described by AOAC [12]. Four grams of oil sample was weighed into 250cm³ conical flask. Ten (10) ml of chloroform and 15ml of acetic acid were added and the mixture stirred after which 1ml of (10% W/V) potassium iodide was added. The flask was covered and shaken for 1 minute and placed in the dark for 5 minutes then 75 ml of water was added. The iodine liberated was titrated with standard (0.1M) sodium thiosulphate. The peroxide value (PV) was determined in relation to reagent blank.

RESULTS AND DISCUSSION

Proximate composition of *Scomber japonicus* (Chub mackerel) meal is shown in Table 1. The meal contained 8.56% moisture, 4.05% ash, 26.23% fat, 13.18% protein and 47.98% Carbohydrate. This is in contrast to the research conducted by [15] which states that mackerel meat fat content was 9.04% and according to report [16] chub mackerel protein content was 20.61 to 22.16%. The present study indicated *Scomber japonicus* possess more energy content, fatty acids and mineral elements but less protein compared to the four species analyzed by Ogundiran et al. [1]. Moon et al. [17] reported that muscle of chub mackerel contained 64.8- 68.3% moisture, 5.7-10.9% lipid, 17.7-18.3% protein and 1.2-1.3% ash, respectively [18]. The present study revealed that *Scomber japonicus* has high carbohydrate, lipid and ash contents but low proteins in comparison to four fish species analyzed by Olagunju [19]. The proximate levels are strongly influenced by the environment and habitat residence of the fish [20, 16, 15], The levels of proximate composition in fish are influenced by factors such as age, feeding rate, and the growth of the species [21, 22, 23]. Ash is a measure of the mineral content of food item, which is the inorganic residue that remains after the organic matter has been burnt off [1]. Low moisture content (8.56%) of *Scomber japonicas* indicated that it has a good storage quality. High moisture content is disadvantageous, because it increases the fishes' susceptibility to microbial spoilage, oxidative degradation of polyunsaturated

fatty acids and consequently decreases in the quality of a fish for longer preservation time [24, 19].

Table 1: Proximate composition of chub mackerel (*Scomber japonicus*) meal

Composition	Percent (%)
Moisture	8.56 ± 0.06
Ash	4.05 ± 0.73
Crude fat	26.23 ± 0.32
Crude protein	13.18 ± 1.14
Total carbohydrate	47.98 ± 1.00

Values are means of triplicate determinations ± standard deviation.

Fatty acid profile of fish oil as presented in Table 2, comprises several fatty acids with, hexadecanoic acid 16.73%, 7-octadecenoic acid 15.90%, 5,8,11,14,17-eicosapentaenoic acid (EPA) 13.03%, 13-docosenoic acid 15.66% among the most abundant. Overall saturated fatty acid was found to be 28.23%, monounsaturated fatty acids 48.51%, polyunsaturated fatty acid 23.26% and PUFA/SFA 0.82, respectively. In the fatty acid composition of the chub mackerel muscle determined by Moon et al. [17], saturated fatty acid comprised 36.8-39.5% of total lipid; monounsaturated fatty acid occupied 31.8-33.4% of total lipid; polyunsaturated fatty acid took 28.7-29.6% of total lipid, respectively. Total lipid content of mackerel muscles contained 23.0-25.2% palmitic acid, 18.3-19.0% oleic acid, 6.1-7.5% EPA and 13.7-14.3% DHA [17].

Table 2: Fatty acid profile of *Scomber japonicus* oil

Fatty acid	% Value
Tetradecanoic acid (myristic acid)	8.99
9-Hexadecenoic acid (palmitoleic acid)	4.84
Hexadecanoic acid	16.73
9,12,15-Octadecatrienoic acid	5.66
7-Octadecenoic acid	15.90
8,11,14-Docosatrienoic acid	3.50

Octadecanoic acid (stearic acid)	2.51
11-Eicosenoic acid	11.37
5,8,11,14,17-Eicosapentaenoic acid (EPA)	13.03
4,7,10,13,16,19-Docosahexaenoic acid (DHA)	1.07
13-Docosenoic acid	15.66
9-Octadecenoic acid (oleic acid)	0.74
Saturated fatty acids (SFA)	28.23
Monounsaturated fatty acids (MUFA)	48.51
Polyunsaturated fatty acids (PUFA)	23.26
PUFA/SFA	0.82

Table 3: Physiochemical properties of Chub mackerel (*Scomber japonicus*) oil

Physicochemical property	Percent (%)
Iodine value (g/100g)	140.82±0.08
Saponification value (mg/g)	252.46±0.029
Free fatty acid (mg/g)	4.89±0.07
Peroxide value (meq/kg)	2.21±0.02
Acid value (mg/g)	9.82±0.02
Refractive index	0.883

Values are expressed as mean ± standard deviation of triplicate determinations

In nutrition and health, fish oil has found tremendous use in the treatment of arthritis, multiple sclerosis, cancer, malaria, non-insulin dependent diabetes, renal diseases, and chronic liver diseases [25, 26, 27, 8]. The uniqueness of fish lipids is the presence of long chain (LC) PUFA, namely eicosapentaenoic acid (EPA), docosahexaenoic acid (DHA) and a low amount of docosapentaenoic acid (DPA). [28] Fish contains most of the essential trace minerals for human metabolism, such as Fe, Cu and Zn [29, 30]. Several studies reported an inverse proportional relation between PUFA/SFA ratio and cardiovascular risks, suggesting the replacement of SFA with PUFA in the feed diet would decrease cardiovascular diseases [31]. In a study conducted by Smichi *et al.* [32], the PUFA/SFA ratio was found to be 0.580, 0.615 and 0.720 for the three studied species. However, these results are lower than what we observed in the present study (Table 2). During the past decades, MUFAs have received increasing attention because of their heart healthy benefits after studies of the olive oil highly consumed by the

populations of the Mediterranean region. In a study conducted by Smichi *et al.* [32], MUFA constituted nearly half of the total fatty acids of sardine and annular sea bream but less than 40% in golden grey mullet fillets. This fact is due to the abundance of palmitoleic acid (C16:1) and oleic acid (C18:1). Annular sea bream fillet contains the higher percentage of oleic acid. The results are coherent with those reported by Alasalvar *et al.* [33] these authors show that oleic acid is the predominant MUFA (60 to 75%) in fish lipids. However, in our present study, Oleic acid constitutes only 0.74% of fatty acid composition in *Scomber japonicus*. Kim and Joo [34] reported that the main constituents of fish oil were palmitic acid, oleic acid, EPA, and DHA. The Chub markrel bone (CMB) showed higher DHA level than the chub mackerel muscle [18].

The physicochemical properties of the oil revealed iodine value of 140.82 g/100g, saponification value of 252.46 mg/g, free fatty acid value of 4.89 mg/g, peroxide value of 2.21 meq/kg, acid value 9.82 mg/g and refractive index 0.883 (Table 3). The value of saponification number indicates that the fish oil contains short chain fatty acids. Acidity values obtained from this study of 9.82 mg KOH/g is higher than International Fish Oil Standards (IFOS) recommended amount of less than 2.25 mg KOH/g. Peroxide value of mackerel fish oil in this study was 2.21 meq / kg. Peroxide value of mackerel fish oil standards based on International Fishmeal and Oil Manufacturers Association (IFOMA), amounting to 3-20 meq / kg, and for IFOS standard is ≤ 3.75. Oxidation values are very important as an indicator of oil quality, the lower the value of primary and secondary oxidation, the higher the oil stability thus, the quality of fish oil. The oxidative stability of fish oils was tested by measuring the peroxide values (PV). Smichi *et al.* [32] found peroxide values of 1.2, 1.6 and 2.6 meq O₂/kg for the freshly extracted oils from annular sea bream, sardine and golden grey mullet, respectively. Thus the peroxide value of chub mackerel still stands relatively low compared to other fish oils commercially available

CONCLUSION

The result of this study shows that *Scomber japonicus* oil and meal have high nutritive value and may be used in various food formulations as a source of energy, protein, essential minerals and fatty acids. Further exploitation and studies on the bioavailability of the nutrients to optimize their use in human diets and establish quality benchmarks for processing and storage can lead to incorporation into diverse food formulations, aquaculture feeds, nutraceutical development and regional applications to address nutritional deficiencies.

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