PRODUCTION AND CHARACTERIZATION OF OIL FROM FISHES

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ABSTRACT
Production and characterization of fish oil were carried out using soxhlet apparatus and n-Hexane as the solvent for extraction. The extraction was carried out at the boiling point of the solvent. Five different species of fishes were used for the experiment. From the result of the extraction, it was observed that the fish A (Mormyrups deliciousus) and fish E (shawa) have a good percentage of oil content of about 30.22% and 24.02% of its dry mass, respectively, while cat fish, tilapia and mud fish have 6.72, 14.52 and 17.92, respectively. [1] reported a similar observation. The iodine and refractive indexes of this study is in agreement with those obtained by [1], and also falls within the specification of the standard value. These values, therefore, show that fish oil is a non drying oil, a good lubricant and a cure for goiter, while saponification and acid values were found to be higher than the standard value. However, the boiling points of the oil were close to the boiling point of water.

Keywords: fish, oil, characterization.

INTRODUCTION
Fish oil is the lipid fraction extracted from fish and fish by-products. Presently, the production of fish oil is becoming more demanding as there is a sizeable and growing world market demand for high quality fish oils. Apart from its various uses as consumable oils, it is also appreciable in both pharmaceuticals and industries.

However, the most frequently used technique in fish oil extraction are fractionation by high speed configurations, low temperature solvent extraction, superficial fluid extraction etc. In this study, solvent extraction was employed during this research. This is because solvent extraction is one of the most efficient methods of oil extraction from oil bearing materials based on the fact solvent can easily be recovered and recycled and it reduces the residual oil in the oil bearing substance to less than 1%.

In view of this, care must be taken when selecting the right solvent for the extraction process.

Practically, all fish species as well as other marine animals may be converted into fish oil and meal. The composition and quality of these fish species are predominant factors in determining the properties and yield of the products [7]. The quality and freshness of the raw material is the factor of great importance in preparation of premium quality fish oil and fish meal [13]. Enzymatic and bacteriology activity in the fish and fish products can rapidly increase, which in turn can substantially decrease the content and quality of the protein and oil as protein decomposes to amines and ammonia, and both reduce the protein value and recovery.

Fish oil is different from other oils mainly because of the unique variety of fatty acids it contains including high level unsaturated fatty acid (OEGA-3FFA and Omega-6FFA) which are essential to the body. This is known as the eicosapentaenoic acid (EPA) and the docosahexaenoic acid (DHA).

Although work has been done on fish oil production but literature has shown that little or no work has been done in terms of characterizing and comparing oil products from different species of fish. The aim of the study therefore is to:

(i) Extract the fish oil from a number of fresh water fishes and a marine fish.
(ii) Evaluate, refine and characterize the extracted oil.
(iii) Recommend types of fish with high nutritional and medicinal value.

Fishes are creatures that live and breathe in water and there are over 25000 different types of fishes in the world and numerous others yet to be discovered [1] and [15]. Five different types of fishes were used for this work of which four are fresh water fishes and one marine fish.

Sample A
Species: Mormyrops deliciousus
Synonyms: Mormyrops angulloides
Common name: Mormyrids or Trunk fish.

Sample B
Species: Bagrus docmac niger
Synonyms: Bagrus Doemak
Common name: Silver cat fish

Sample C
Species: Tilapia dagati
Synonyms: Tilapia melanopleura thys van de
Common name: Chichilid (Tilapia)

Sample D
Species: Clarias anguilloris
Synonyms: Clarias Senegatensis
Common name: Catfish (mud fish)

Sample E
Titus, A marine or frozen type of fish.
Common name: shawa
Fish is a source of protein and proteinous food, are body building foods. Fish oil is the lipid fraction extracted from fish as fish by-products. Apart from the benefit derived from the consumption of fishes, there are numerous other benefits to be derived from the consumption of fishes due to its high nutritional content [15].

The production of fish oil started long ago since the 19th century in Northern Europe and North American, where they utilized the non-edible fishes and other fish by-products to produce oil used in leather tanning and in the production of soap and glycerol [6].

Fish oil composition (Lipids)

Fish oil is very similar to one another in their physical nature. A whole fish consists of protein, fat, Ash and water irrespective of the species however; these compositions are greatly influenced by seasonal changes due to the nature cycle, maturity stage, geographical location, feeding habit etc. Because the more a fish eats, the greater the oil and other chemical composition will be produced. Most fish oil in general is more complex than land animal oils or vegetable oils due to the long chain unsaturated fatty acids. It is generally believed that fish oil odour is due to the unsaturated fatty acids, since hydrogenation causes the oil to lose their colour but fish caught in colder water have a higher degree of instauration than that caught in warm water.

The lipids is the edible part of fish and is important to the food scientist in two respects, firstly any oil deposit noticeably influence the sensation of the cooked flesh and secondly has some medical applications. Fish oil deteriorates very rapidly due to the natural lipase and bacterial in the fat. Both of these hydrolyze fat to free fatty acids. The condition of the fish at the time of processing affects the oil physically, chemically, and nutritionally. Fish of poor quality yields malodorous oil with high contents of free fatty acids and sulphur. These undesirable properties affect the economic values and the application of the oil.

Processing and packaging

The processing and packaging of fish oil are crucial in determining its quality. Low quality oils may be quite unstable and contain significant amounts of mercury, pesticides and undesirable oxidation products. High quality oils are stabilized with adequate amounts of Vitamin E and are packaged in industrial foil pouches or other packaging resisive to light and oxygen. Some very recent researches carried out at the University of Minnesota found out that emulsified fish oil is much better absorbed than the straight oil in gelatin capsules [8].

Properties of fish oil

- Physical properties of fish oil comprises of melting point, the refractive index and the specific gravity, whilst the chemical properties are iodine value, saponification value, and acid value.
- The world demand for vegetable oil is constantly increasing due to increase in the world population. The production of vegetable oils and fats, which is around 30 metric tones, is not enough to meet the needs of people, since fats and oil are required industrially for the manufacturing of soap and other industrial purposes [3, 14]. This research work is part of the efforts of finding ways of increasing fats and oil production and it is shown by considering the extraction of oil from acacia seeds. *Acacia nilotica* is a tree of 5-20 m height with a dense spherical crown stems and branches usually dark to black colored, fissured back, grey-pinkish flask, exuding a reddish low quality gum. The tree has thin, straight, grey spines in auxiliary pairs, usually in 3 to 12 pairs, 5 to 7.5 cm long in young trees, mature trees commonly without thorns. The leaves are bipinnate, with 3-6 pairs of pinnulate. Flowers in globulous heads 1.2 to 1.5 cm in diameter of a bright golden- yellow color. The tree has scented thorns all over the stem and branches, and is found mostly in riverine areas and seasonal flooded areas [2, 10, 11 and 14].

* Acacia nilotica seed contains protein, fat, nitrogen freed extract, acid detergent fiber ADF, crude fiber CF, organic matter digestibility, metabolizable energy, tannin, phosphorus, calcium, magnesium, potassium, silicon, sulphur, chlorine, copper, zinc, manganese and iron. The seed has numerous chemical components and it finds its place in pharmaceutical industries. Bullocks fed 45% oil - extract seeds of *A. nilotica* in their diet showed reduced weight gain (68 g/day to 16 g/day). *A. nilotica* tannin has been used to treat cotton seed cake to prevent rumen degradation of protein. *A. nilotica* seed is made up of shell and kernel. The kernel finds many uses. It can be used as fuel and to make products like flour, starch, and cattle/poultry feed other than oil. The kernel yields good manure for plant. The fat in the kernel is edible and it can be substituted for cocoa - butter. Also, the fat and oil can be used industrially in manufacturing of antiseptic soaps [2, 10 and 11].

MATERIALS AND METHODS

The principle employed involve, pretreatment of sample, extraction of the oil from the fishes, characterization of the extracted oil and subsequent comparison of the oil extracted for the different species fishes used for the experiment.

Raw materials

The raw materials used are the 5 species of fishes earlier named. They include Mormirops deliciosus (Trunk fish) Bagrus docmac (Silver cat fish), Tilapia, Clarias (Catfish or mud fish) and Titus (shawa).

Pre-treatment of raw material

In order to enhance a successful extraction of the oil, the fish underwent some treatment prior to the extraction. These include:

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14.
Refrigeration: The fish when bought was freeze in order to preserve it since the extraction did not commence immediately.

Washing: The fish was thoroughly washed in order to remove dirt that might get stuck to the body after undergoing a de-freezing process.

Size reduction: The fishes were then cut into sizes in order to enhance a speedy oven drying because of their size while removing the gills and intestine which were unwanted.

Drying: The moisture content of the fishes was reduced by oven drying since water is immiscible in oil. Further size reduction: the samples were further reduced in size and later blended into a finer form by pounding in a mortar after undergoing the moisture content elimination in the oven.

Weight: The weight of the samples were taken accordingly noting the difference in weight due to weight lost through evaporation.

The fish oil extraction process

The fish oil was extracted using soxhlet extractor and n-Hexane as the solvent. The solid substance or sample was placed in a porous thimble covered with cotton wool and the weight of the sample taken, before it was placed in the inner tube of the apparatus and then fitted to a round bottom flask of appropriate size that contain the solvent. Heat was applied to heat the solvent to its boiling point for 1 hour.

As the heating continued, the solvent in the flask started boiling just within 5 minute of heating and the water begins to drop from the top to the sample in the thimble.

When the solvent reached the top of the tube, it siphoned over into the flask and thus removes the portion of the oil which has been extracted in the process of refluxing. It was noticed that 18 minutes later, after boiling has started, there was refluxing and this continued at 2 minutes interval.

The solvent used was later recovered by applying heat and collected above the round bottom flask into the soxhlet apparatus while the oil extracted was collected and measured.

Characterization of fish oil

The evaluation of the oil involves the analysis and testing needed for the assessment of the quality, purity and as well as the identification of the oil. A number of physical and chemical “constants” have been established for these purposes. Each of the constituents used in examining the oils and fat is chosen to measure one of the characteristics of the glycerol or fatty acids present in the oil. An assessment of all these are then related to the composition and therefore the identity of the fats being examined [7].

Determination of moisture content of the fish

The method specified by [14] was used. The principle was that a test portion was heated at 105°C until moisture and volatile substances are completely eliminated, and the loss in mass determined.

Procedure: An empty Petri dish was weighed (w₁) the wet sample of the fish was then put into the Petri dish. The weight of the fish and Petri dish was taken (w₂), this was then transferred into the Gallenkamp oven which was set for 105°C this allowed the complete evaporation of the moisture content from the sample.

At the end of the drying, the dried sample in the Petri dish was removed and allowed to cool for a while after which the weight was taken (w₃) and the difference calculated. The percentage moisture removed represents the percentage loss in mass of the sample.

Calculation:

Moisture content removed

\[
\% = \frac{(w₂ - w₁) - (w₃ - w₁)}{w₂ - w₁} \times 100
\]  (1)

Determination of refractive index

Refractive index is the ratio of the speed of light at a definite wave length in a vacuum to its speed in the medium and this varies with the wave length of light and temperature.

Procedure: Abbey refractometer was used in determining the refractive index of the oil. The measuring prism surface was cleaned with solvent and distilled water, and then wiped with a clean towel after which the mode selector was regulated to the desired mode position. A drop of oil was dropped on the prism surface using a glass dropper and covered. The illumination arm was then positioned so that the exposed face of the upper prism will be fully illuminated. The refractometer was used through the eyepiece, the dark position viewed was adjusted to be in line with the cross line. At no parallax error, the pointer to the scale pointed in the refractive index, the reading was then taken. This measurement represents the refractive index of the oil sample.

Determination of acid value

The acid value is the number of milligrams of KOH required to neutralize the free fatty acid present in 1g of fat. Hence acid value gives an indication of the age and quality of the fat.

Procedure: An account weight of 1g of fat sample was taken and dissolved in carbon tetrachloride and the solution was titrated with 0.05m Alkali; using phenolphthalein as indicator with constant shaking until a dark colour was observed and the value noted.
Determination of saponification value

The saponification value is the number of milligram of KOH required to neutralize the fatty acids present as a result of the complete hydrolysis of 1g fat [1].

Procedure: 1.00g of the samples was weighed into 2.5 cm³ of alcohol 10 cm³ of 0.5m alcoholic KOH solution. This was then attached to a reflux condenser; the mixture was allowed to boil for 30 minutes with constant shaking. Similarly 2.5 cm³ of alcohol and 10 cm³ alcohol 0.5 M KOH was treated while adding few drops of phenolphthalein to the warm solution and then titrated against 0.5 HCL until the pink colour of the indicator just disappeared. Same procedure was used for the other samples and the blank solution.

Determination of Iodine value

The amount of iodine consumed is determined by titrating the iodine released (after adding KI) with a standard Thiosulphate.

Procedure: 0.3 g of fats was weighed into a small weighing dish and placed in a 250 cm³ conical flask 10 cm³ of carbon tetrachloride was added to the samples.

To all the flask an equal quantity of about 25 cm³ wigits reagents was added using a burette, this was mixed well and kept in the dark for an hour, after that it was titrated with standard 0.1M sodium thiosulphate solution while adding 15cm³ of 10 % potassium iodide solution and then titrated against 0.5 HCL until the pink colour of the indicator just disappeared. Same procedure was used for the other samples and the blank solution.

RESULTS AND DISCUSSIONS

Experimental analysis was conducted on the oil extracted from each of the samples of fishes and the results in Table-1 shows that the percentage moisture content of species A, B, C, D and E were 22.84, 4.84, 9.54, 4.92 and 25.05 %, respectively. This signifies that species A and E will require higher drying time than species B, C, and D when subjected to the same drying condition. However, specie B has the least moisture content.

Table-1. Moisture and oil content of the five samples.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Moisture content (%)</th>
<th>Oil extracted (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>22.84</td>
<td>30.22</td>
</tr>
<tr>
<td>B</td>
<td>4.84</td>
<td>6.72</td>
</tr>
<tr>
<td>C</td>
<td>9.54</td>
<td>14.52</td>
</tr>
<tr>
<td>D</td>
<td>4.92</td>
<td>17.93</td>
</tr>
<tr>
<td>E</td>
<td>25.50</td>
<td>24.02</td>
</tr>
</tbody>
</table>

Table-2. Comparison of the properties of the oil samples with the standard.

<table>
<thead>
<tr>
<th>Sample</th>
<th>R. index</th>
<th>Acid value (mg)</th>
<th>Iodine value (mg)</th>
<th>Saponification value (mg)</th>
<th>Melting point °C</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>1.6642</td>
<td>8.40</td>
<td>174.41</td>
<td>549.78</td>
<td>100</td>
</tr>
<tr>
<td>B</td>
<td>1.6769</td>
<td>7.84</td>
<td>182.88</td>
<td>367.46</td>
<td>92</td>
</tr>
<tr>
<td>C</td>
<td>1.6240</td>
<td>6.72</td>
<td>182.88</td>
<td>347.82</td>
<td>88</td>
</tr>
<tr>
<td>D</td>
<td>1.5990</td>
<td>5.04</td>
<td>187.11</td>
<td>318.48</td>
<td>91</td>
</tr>
<tr>
<td>E</td>
<td>1.5850</td>
<td>6.44</td>
<td>178.65</td>
<td>398.31</td>
<td>98</td>
</tr>
<tr>
<td>Standard Value</td>
<td>1.473 – 0.40-4.8 mg/ of sample</td>
<td>135 – 190</td>
<td>176 – 195</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

CONCLUSIONS AND RECOMMENDATIONS

The production and characterization of oil from different samples of fishes was carried out and the results showed that the fish oil has a very high percentage of iodine value, hence can be recommended for patient suffering from goiter. Also, the moisture content of a fish is a reflection of its oil content (i.e. the higher the moisture content the higher the oil). From the analysis of the oil content of the different species it can be concluded that sample A (Mormyrups deliciousus) and sample E (Titus) has the highest oil content among the five species analyzed.
REFERENCES


