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Production and characterization of some fishes oil from southern region of Iran

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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. Three different species of fishes were used for the experiment. From the result of the extraction, it was observed that the fish B (Kharoo baleh sefid) and fish C (Soboor) have a good percentage of oil content of about 30.22% and 24.02% of its dry mass, respectively. The iodine and refractive indexes of this study 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 acid value were found to be higher than the standard value. However, the boiling points of the oil were close to the boiling point of water.

Key words: 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

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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 (Lees, 1990). The quality and freshness of the raw material is the factor of great importance in preparation of premium quality fish oil and fish meal (Sarjent, 1997). 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.

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 (Adeyemo, 2004) and (William, 1966). Three different types of fishes were used for this work of which all are marine fishes.

Sample A	**Sample B**	**Sample C**
Family name: Scombridae Scientific name: *Scomberomorus guttatus* Local name: Gobad English name: Indo-pacific king mackerel	Family name: Chirocentridae Scientific name: *Chirocentrus nudus* Local name: Kharoo baleh sefid English name: White fin wolf-herring	Family name: Clupidae Scientific name: *Tnualosa ilisha* Local name: Soboor English name: Hilsa shad

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 (William, 1966). 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 (International Standard Organization, 1988).

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.

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 resistive 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 (Kulli *et al.*, 2007).

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 (Alfred and Patrick, 1985; Charles and Gordon, 2005; Weiss, 2000). This research work is extraction and characterization of some fish oil obtained from Persian Gulf of Iran.

The aims of the study therefore were to (i) extract the fish oil from a number of fresh water fishes and a marine fish, (ii) to evaluate, refine and characterize the extracted oil and (iii) to recommend types of fish with high nutritional and medicinal value.

Materials and methods

The principle employed 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 used raw materials were three species of fishes. They include Indo-pacific king mackerel (*Scomberomorus guttatus*), White fin wolf-herring (*Chirocentrus nudus*) and Hilsa shad (*Tnualosa ilisha*).

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 refrigeration which the fish was freezed in order to preserve it since the extraction did not commence immediately. The fish was thoroughly washed in order to remove dirt that might get stuck to the body after undergoing a de-freezing process.

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.

The moisture content of the fishes was reduced by oven drying since water is immiscible in oil. 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.

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 flash 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 began 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 removed 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 used solvent was later recovered by applying heat and collected above the round bottom flask into the soxhlet apparatus while the oil extract was collected and measured (Macrae *et al.,* 1993).

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" was 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. The assessment was then related to the composition and therefore identified the fats being examination (Intentional Fish Meal and Fish Oil Organization, 1986).

Determination of moisture content of the fish

The method was specified by Weiss (2000). The principle of test portion was heated at 1050C until moisture and volatile substances are completely eliminated, and the loss in mass determined.

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 were taken (w2), then transferred into the Gallenkamp oven which was set for 105^{o}C to allow 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 calculated. The percentage removed moisture represented the percentage of loss in mass sample.

Moisture content removed (%) = $[(w_2-w_1)-(w_3-w_1)] \div [(w_2-w_1) \times 100$

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.

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 noted.

Determination of Iodine value

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

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, 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 100 cm3 of distilled water using starch as an indicator.

Results and discussions

Experimental analysis was conducted on the oil extracted from each of the samples of fishes and the results in Table 1 showed that the percentage moisture content of species A, B and C were 66.69, 66.19 and 55.95 %, respectively. This signifies that species A and B would require higher drying time than species C when subjected to the same drying condition. However, species B was the least moisture content. While their oil content were 9.5, 12.49 and 13.27 %, respectively signifying that species C was a higher amount of oil while species A was the least amount of oil extracted. It can be deduced that moisture content of the fishes was a reflection of their oil content this is due to species with higher moisture content yield high amount of oil when extracted.

The result of the characterization carried out for the samples of the fishes presented in Table 2 showed that samples A, B and C revealed a refractive index of 3.21, 1.21 and 6.72 respectively which were outside the range of standard value of 1.4 - 1.473 for fishes. The iodine index directly depended on ~~to~~ the number of double bonds in unsaturated fatty acids in fishes oils. White fin wolf-herring and Hilsa shad oils gave the highest and lowest iodine indexes respectively. Therefore, it is observed that unsaturated fatty acids with several double bonds and omega-3 and omega-6 fatty acids in white fin wolf-herring oil prevent from heart and brain diseases and Atherosclerosis. It can conclude that white fin wolf-herring oil has highest chemical spoilage, it may be increased with increasing of concentration of oxygen, metal ions, light intensity and temperature as stated by Firestone (1989).

The mixture ratios of chloroform/methanol/water (4:2:1and 2:4:1) both gave higher yields (550%) that was more than results obtained from our study. Bligh and Dyer method has been recognized as the most reliable method currently available for total lipid extraction (Bailey and Wells, 1994). Bligh & Dyer method uses polar solvent, chloroform and methanol mixture to extract the oil from fish by-products. Many researchers who have employed this particular extraction method have also reported higher yields (Tanamati *et al.*, 2005; Ewald *et al.*, 1998; Hole *et al.*, 1996; Undeland *et al.*, 1998).

In order to determine the stability and quality of fish oil extracts, some quality assessment were conducted. These results are shown in Table 2. Undeland *et al.* (1998) indicated that unsaturated character of the lipids and the strong pro-oxidative systems naturally present in fish tissue could cause susceptibility of lipids to oxidize during processing and storing especially for fish selected species. Increase in acid value is generally associated with the lipase activity originating from microorganism or biological tissue (Boran *et al.*, 2006). The acceptable limit for AV was reported to be 7-8 mg KOH/g (Bimbo and Crowther, 1991). That results obtained from our study was less. Abdulkadir *et al*. (2010) reported iodine values of some fish species (174.41, 182.88, 187.11 and 178.65 mg) that more than results obtained from our study (119.23, 141.3 and 38.42).

Table 1. Moisture and oil content of the five samples

Samples	Moisture content (%)	Oil extracted (%)
A	66.69	9.5
B	66.19	12.49
C	55.95	13.27

Data expressed as mean values of three independent experiments.

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

Sample	R. index Acid value (mg)	Iodine value (mg)
A	3.21c	119.23 c
B	1.21d	141.3b
C	6.72 b	38.42e
Standard Value	0.40-4.8 mg/ KOH	135 – 190 I2/ 100 of sample

Data expressed as mean values of three independent experiments.

Conclusion

The production and characterization of oil from different samples of fishes showed that the fish oil gave a very high percentage of iodine value, hence can be recommended for patient suffering from goiter. Also, the moisture content of a fish was 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 B (White fin wolf-herring) and sample C (Hilsa shad) gave the highest oil content among the five species analyzed.

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