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Extraction of Fish Oil from Fsh Viscera

Fatma Abd El-Rahman[1*], Nesreen S. Mahmoud[1], Abo El – Khair Badawy[2] and Samy M. Youns[1]

[1]Department of Agricultural Engineering, Faculty of Agriculture, Cairo University, Giza , Egypt. Postal Code 12613. [2]Algal Biotechnology Unit, Fertilization Technology Department, National Research Center, Giza, Egypt. Postal Code 12622.

IN THIS study, fish oil was extracted from tilapia and mackerel viscera by the wet rendering extraction method. Using water base extraction method with more yield is a green chemistry which is better than using solvent extraction which is hazardous chemicals. The extracted fish oil is intended to be used for the production of biodiesel in further investigation. Increasing the temperature from 80ºC to 100ºC had significant effect on the extracted oil yield. Centrifugation had no significant effect on oil extraction at high cooking temperature (100 ºC) and therefore can be omitted from the extraction process. The results showed the applicability of producing fish oil from Tilapia viscera. The oil yield obtained from Tilapia viscera was about 20% which was higher than that obtained from Mackerel viscera (7% higher). Increasing cooking time in the range of 30 to 60 min had a significant effect on the extracted oil yield. 60 min cooking time was found to be optimum. The extracted fish oil was analyzed for its viscosity, moisture content, free fatty acids, acid value and fatty acids composition. The results indicated the suitability of fish oil for using for biodiesel production.

Keywords: Fish viscera, Fish oil, Wet rendering extraction, Free fatty acids, Kinematic viscosity, Biodiesel.

Introduction

Fish is an excellent source of high quality animal protein, essential fatty acids, and micronutrients, all of which are found at much higher level in fish than in terrestrial animal food sources. The fishing industry is very important not only to ensure food security but also to provide employment and income that help elimination of poverty especially in developing countries. Egypt is the largest aquaculture producer in Africa and the 8th largest aquaculture producer globally. The Egyptian aquaculture production increased gradually from 367,000 tonnes in the year 2002 to 1,018,000 tonnes in the year 2012, to 1.48 million tonnes in the year 2014 and to 1.53 million tonnes in the year 2015 [1].

There are few species of fish used in the Egyptian aquaculture some of which are native while few are imported. However, three fish groups compromise more than 95% of the aquaculture total production. These are Tilapias (75.54%), mullet (12.74%) and carps (6.59%). Tilapia is considered the most popular native fish due to its rapid growth and low selling price. China, Egypt, Indonesia, Philippines and Thailand are the world top producing countries. Mackerel is also a very popular imported fish and has low selling price compared to other native and imported fish species [2].

The fishing and fish processing industries produce large quantities of fish waste which often represent about 20-50% of the total fish weight, depending on the degree of fish processing [3,4]. Fish waste has a bulky nature, perishable and is considered a threat to the environment when it is improperly disposed off. Fish viscera represent from 7.5 - 15% of the total fish body weight and have no commercial value. However, fish viscera have high protein and lipid contents and some minerals but the composition of fish viscera depends on the species, seasonality, age, sex, nutrient intake and environmental factors [5]. Tilapia viscera composition of 14.62% protein, 10.75% lipids, 60.44% moisture and 4.90% minerals [6].

*Corresponding author email: fatma.suiefy2014@yahoo.com
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Fish oil can be extracted from fish viscera by a number of processes including: wet rendering, enzymatic hydrolysis, autolysis, dry rendering, solvent extraction, and supercritical fluid extraction [7-13]. Wet rendering extraction has been used extensively since it does not require the use of harsh chemicals and consequently preserve the natural state of the fish waste components, lower the extraction cost and does not possess harmful effect on the environment [11].

For many years, fish oil was considered a waste product and was often burnt. However, recently the production and availability of fish oil is growing worldwide. Today, fish oil is recognized as a valuable nutrient for both livestock and humans, and is widely used in the food such as (production of margarines, salad oil, mayonnaise, and several types of spreads and pastes in bakery products), the feed of farm-raised animals; such as pigs, poultry, cattle, and sheep, and it is also used in pet foods, and other industries [14]. Biodiesel is a renewable source of energy that could be obtained from fish oil.

The aim of this study was to assess the use of a potent pollutant waste material such as fish viscera for the production of value-added product (fish oil) that could be converted into a renewable source of energy (biodiesel). The specific objectives were : (a) to investigate the applicability of producing fish oil from Tilapia fish viscera (the most popular and least expensive native fish in Egypt) and determine the amount of oil produced from the viscera of tilapia fish as the most popular and least expensive native fish produced in Egypt, (b) to compare the amount of the oil extracted from tilapia viscera with the amount of oil extracted from mackerel viscera (low cost imported fish), (c) to evaluate the effectiveness of two oil extraction schemes adopted from the literature (long heating at high temperature vs. short heating at lower temperature followed by centrifugation) and (d) to determine the characteristics of the extracted Tilapia visceral oil (moisture content, free fatty acid, viscosity, acid value and fatty acid composition) in order to determine the suitability of the extracted oil for conversion to biodiesel and choose the best conditions and procedures for the conversion process in order to be applied in a further study.

Material and Methods

Experimental design

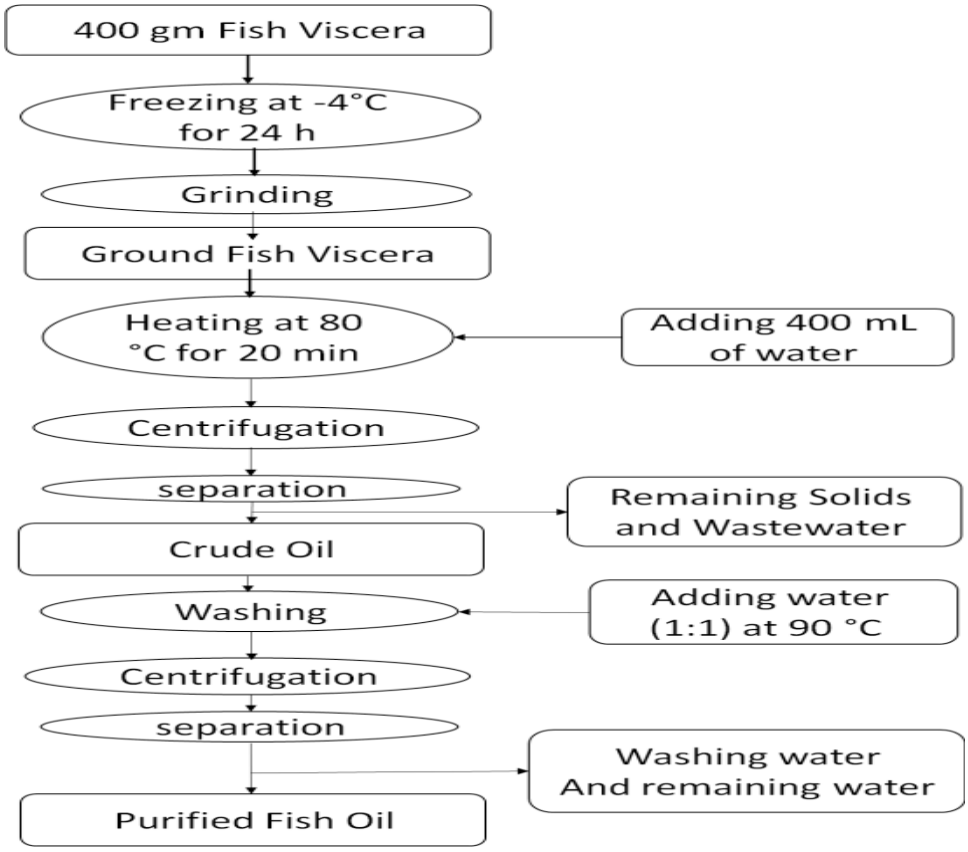
Four sets of experiments were carried out in this study. The first set was performed to study the applicability of producing fish oil from tilapia viscera and to determine the average oil productivity. The second set was performed to compare the amounts of oil produced from the viscera of Tilapia and Mackerel fishes. The third set was performed to compare the oil productivities of two different oil extraction schemes adopted from the literature (long heating at high temperature vs. short heating at lower temperature followed by centrifugation). The effects of two cooking temperatures (80 ºC and 100 ºC) at the same cooking time (50 min) and centrifugation at the same cooking temperature and time were also investigated. The fourth set was performed to study the effect of cooking time (20, 30, 40, 50, 60, 70, 80 and 100 min) at cooking temperature of 100 ºC and without centrifugation on oil extraction yield. The extracted oil was analyzed for its characteristics.

Material

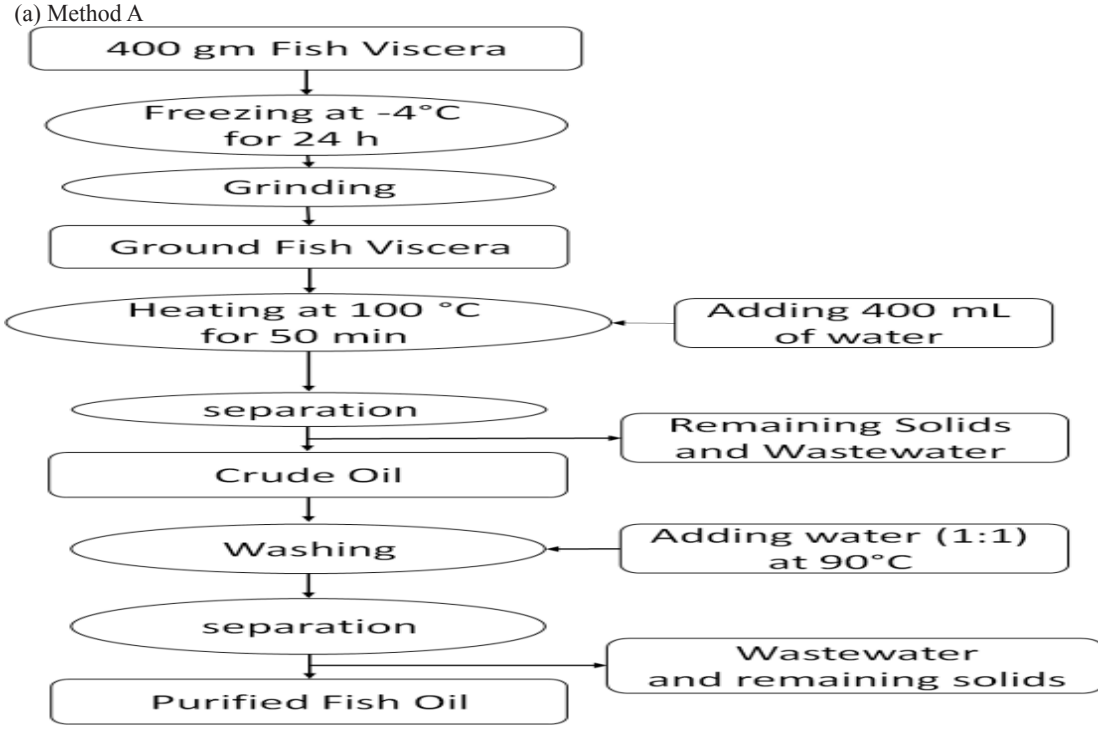
The viscera of the fish used in this study were bought from the local market in Giza (Monuib Market). The fish viscera were extracted from the fish manually by the market vendors. They were bought fresh and then frozen at -4°C in a conventional freezer for 24 hours before use in the experiments. The freezing process was done in order to break down the fat cells and facilitate the extraction process. After freezing, the viscera were then grounded using a laboratory blender to break the tissue into small particles in order to increase the efficiency of the extraction process [12].

Extraction methods

Two wet rendering extraction schemes were applied in this study as shown in the schematic diagram (Figure 1). The first scheme (method A) was adopted from the method [15] who indicated that this extraction method gave optimum results for the extraction of oil from fish waste. In this method, a relatively lower temperature is applied for a shorter time followed by centrifugation. The second method (Method B) was higher temperature than Method A for a longer time but omitted the centrifugation step [11].



(a) Method A



(b) Method B

A: Short heating (20 min) at lower temperature (80°C) followed by centrifugation

B: Long heating (50 min) at high temperature (100°C)

Fig.1. Wet rendering extraction process.

Wet rendering extraction method A

In this method 400 gm of either Tilapia viscera or Mackerel viscera were used. The extraction process was done according to the schematic diagram shown in Figure 2a. First, the 400 fish viscera were put in 400 mL of water in a beaker and heated to 80 °C for 20 min using a hot plate stirrer (WiseStir, Model: MSH-20A, Daihan Scientific Co., Ltd., Korea) in order to rupture the fat cells. The cooked sample was then centrifuged at 1500 rpm for 5 minutes by using a clinical centrifuge (SCILOGEX, Model: DM0412, USA) in order to separate the oil from other solids and wastewater. A separating funnel was used to separate the crude oil from the wastewater. The final step was the addition of water at 90°C to the oil with a ratio of 1:1 followed by centrifugation at 3000 rpm for 10 minutes for washing the oil. The purified fish oil was then separated from the water using the separating funnel.

Wet rendering extraction method B

This method was adopted from the method used by [11] who applied boiling in two stages. The procedure was slightly modified by boiling in one stage to speed up extraction process. The extraction process was done according to the schematic diagram shown in Figure 2b. First, 400g Tilapia viscera were put into 400 mL boiling water (100 ºC) in a beaker and heated for 50 minutes by using an electrical hot plate (FARA Tech, Model: HP - ISSF, China). The cooked waste left over night to cool down and to allow the oil to float on the surface of the cooked mixture. The floating oil was then collected and placed in a separating funnel in order to separate the crude oil from the wastewater. Then the crude oil was washed with warm water in order to clean it from any remaining solids. Finally, the washed oil was separated once again using a separating funnel in order to obtain the purified fish oil.

Characteristics of purified fish oil

The purified fish oil was analyzed for its viscosity, moisture content, free fatty acids and acid value. The analyses were done in the Fuel Analysis Laboratory of the Petroleum Research Center, Cairo, Egypt. GC analysis for fatty acids composition of the extracted fish oil was performed in Oils and Fats Department in the National Research Center, Giza, Egypt.

The FFA determination was performed following two methods. ASTM D664, Standard Test Method for Acid Number of Petroleum Products by Potentiometric Titration, Method A, was first used to determine TAN in the samples, after this, the FFA values were calculated using the mathematical formulas found in the American Oil Chemists' Society (AOCS) Method Ca 5a-40. [16,17]. TITRONIC basic and TitroLine easy (SCHOTT, D- 55122 Mainz, Germany) was used.

The moisture content was determined according to ASTM D 6304 [18], TitroLine Karl Fischer trace (SCHOTT, D- 55122 Mainz, Germany) was used.

The kinematic viscosity determination requires the measurement of the time (t) the fluid takes to go from point A to point B inside the viscometer. The kinematic viscosity (v) is calculated by means of the following equation [19]: v = c. t. ASTM D445, Standard Test Method for Kinematic Viscosity of Transparent and Opaque Liquids (and Calculation of Dynamic Viscosity) was used. The units of kinematic viscosity are centistokes (cSt) or mm^2/s [19]. Viscosity Bath (Tamson Instruments, TV4000, Germany) was used.

Analysis of Fatty Acids composition

Preparation of fatty acids methyl esters was done as followed; about 0.2 g of the oil was mixed with 30 ml sulphuric acid to methanol (4:96 v/v) in A250 ml round bottom flask. The contents of the flask were then heated under reflux for about three hours. The methyl esters were thrice extracted with petroleum ether then it was washed several times with distilled water till the washings were neutral to phenolphthalein. The combined fatty acids methyl esters layers were dried over anhydrous sodium sulphate and filtered. The petroleum ether was then removed using a rotary evaporator and an aliquots of the fatty acid methyl esters were analyzed by gas chromatography [20].

The identification of the components of fatty acids methyl esters was done using gas liquid chromatography on a Hewlett Packard Model 6890 chromatograph equipped under the following conditions:
Separation was done on an INNO wax (Polyethylene glycol) Model No.19095 N-123, 240°C maximum, capillary column 30.0 m × 530 μm×1.0 μm, nominal flow 15 ml / min. With average velocity 89 cm / sec. and pressure 8.2 psi. Column temperature was 240°C with temperature programming: Initial temperature 100°C to 240°C maximum with 10°C rising for each minute and

then hold at 240°C for ten minutes. Injection temperature 280°C, back inlet, with split ratio 8:1, split flow 120 ml/min, gas saver 20 ml / min. Carrier gas was nitrogen with flow rate 15 ml / min, flame ionization detector temperature 280°C, hydrogen flow rate 30 ml / min, air flow rate 300 ml /min.

Results and Discussion

Applicability of extracting fish oil from Tilapia viscera

The results of this study showed the applicability of using tilapia viscera for fish oil production as a value-added product. Of the 400 g viscera mass an average of 40 g oil was extracted using modified wet rendering extraction method (Method A). The productivity of fish oil from Tilapia viscera using Method (A) was about 10%. The productivity of the oil extracted from tilapia viscera in this study was higher than that of the oil extracted from Tilapia viscera reported in previous studies. In this connection, an oil yield of 6.44% using wet rendering extraction at 70 °C for 35 minutes was obtained [13]. Furthermore, oil yield of 8.12% from Tilapia viscera using the solvent extraction method was occurred [21], where the oil yield in the current study increased by 1.98 % than the extraction by using solvent. Also using water instead of chemical solvents offers many advantages because it is a cheap, readily available, non-toxic and non-flammable solvent, thus being very attractive from both an economical and an environmental point of view [22]

Oil production from two fish species

Two different types of fish (Tilapia and Mackerel) were compared for the amount of oil extracted from their viscera. Tilapia and Mackerel are the most popular native and imported fish in Egypt, respectively. Previous studies showed that the tissue of Mackerel fish contains more fat than those of Tilapia fish [23]. However, the lipids are concentrated more in the Mackerel skin. In this study, 400 grams of each type of fish viscera were used and the oil was extracted from the viscera of both fishes using the modified wet rendering extraction method (Method A). Table 1 shows the results of the average of three replicates. The results show that 40±1.35 grams of oil were extracted from Tilapia viscera while 12±1.02 grams were extracted from Mackerel viscera. This means that the oil yields were 10 and 3% for Tilapia and mackerel, respectively. This compared favorably with Jayasinghe et al [12] who reported that the extracted oil from the whole body of mackerel was in the range of 3.7 - 5.2%. Furthermore, extracted oil from different parts of Mackerel using a solvent extraction method and reported average oil yields of 9.18%, 9.2% and 38.1 for viscera, muscles and skin, respectively [24]. The higher productivity of mackerel oil comparing with the current study may be due to the difference in the procedure of performing the extraction process in the two extraction methods.

TABLE 1. Amount of extracted oil

Viscera sample	Sample weight (g)	Oil weight (g)	Oil Productivity (%)
Tilapia	400	40±1.35	10
Mackerel	400	12±1.02	3

In this study the productivity of oil extracted from the viscera of mackerel was much lower than the productivity of oil extracted from the viscera of tilapia. This is due to the Fact that different types of fish may be exposed to different culturing conditions and different diets. In addition, mackerel is an imported fish and hence degradation of fish fat might have taken place during freezing, packaging and storage. Oil in fish can differ due to their feeding habits, climatic condition, age, maturity and type of species [25, 26, 27-28].

Effect of oil extraction method

The effect of two extraction methods on oil extraction from tilapia viscera was investigated. Method B had a higher cooking temperature (100 ºC) and longer cooking time (50 min) than Method A (80 ºC and 20 min, respectively). Also, Method A had a centrifugation step which was omitted in method B. Centrifugation step was mandatory when method A was applied since the oil did not float on the surface at the end of the cooking process. While when method B was applied the oil floated on the surface and applying centrifugation did not result in any extra yield of the extracted oil. The results (Table 2) showed higher oil yield when Method B was used. Method B resulted in 20% oil productivity while Method A resulted in only 10% oil productivity. The higher cooking temperature and longer cooking time applied in Method B had a significant effect on the oil extraction yield. The oil extraction yield was doubled with the increase in cooking temperature from 80 ºC to 100 ºC and cooking time from 20 min to 50 min.

The effect of cooking temperature (80ºC

vs. 100ºC) at 50 min cooking time on the oil extraction yield was studied. By the end of the cooking time, the oil floated on the surface and centrifugation was not needed. The results (Table 3) showed higher oil yield at cooking temperature of 100 ºC. Temperature of 100ºC resulted in 20% oil productivity while temperature of 80ºC resulted in only 11% oil productivity. Higher temperatures causes cell rupture and consequently facilitate oil extraction and increase the extracted oil yield. The yield of extracted fish oil increased with the increase in temperature and reported an optimum temperature of 80 °C and further increase in temperature resulted in a decrease in oil yield [29]. The oil yield in the current study was higher than the study performed by Suseno et al [13], who reported that the optimum cooking temperature of 70 °C at 35 minutes cooking time for wet rendering extraction and achieved highest yield of 6.44%; while the commercial fishmeal plants use cooking temperatures above 90 °C for oil extraction [12].

Effect of cooking time

The effect of cooking time at cooking temperature of 100 ºC on oil extraction yield from tilapia viscera was investigated. Figure 2 shows the results. It was observed that by increasing cooking time up to 60 min, the amount of oil extracted increased, further increase in cooking time (from 60 to 100 min) did not result in any extra extracted oil yield. The yield of extracted oil was first increased slightly (from 15.4 % to 15.6%) when cooking time increased from 20 min to 30 min. Then the extracted oil yield increased significantly when cooking time increased from 30 min to 60 min (15.6, 17.0, 20.0 and 22.0% after cooking time of 30, 40, 50 and 60 min, respectively). Increasing cooking time caused increase in the cell exposure time to heat and consequently increased cell rupture and oil yield. The results obtained in the current study agrees with that of [11] who found that boiling for 50 minutes was good for extracting the oil from the tilapia viscera.

The relationship between cooking temperature (in the range of 30 to 60 min) and extracted oil yield could be described by the following equation

$$(R^2 = 0.985): \quad Y = 10.8\, e^{0.011\, t}$$

Where:

Y is the yield of the extracted oil (%)

t is the cooking time (min)

Characterization of Tilapia visceral oil

Characterization of fish oil is very important for determining the potential uses of the extracted oil and if further treatment is needed before the conversion into biodiesel. The characteristics of the extracted oil from tilapia viscera were determined. The characteristics included kinematic viscosity, free fatty acids content (FFA), acid value and moisture content Table 4 shows the main characteristics of the extracted oil from tilapia viscera. The fatty acid composition was determined using GC analysis.

Kinematic viscosity

Kinematic viscosity is defined as the resistance to flow of a fluid under gravity. Oil viscosity has a significant impact on engine operation, fuel injection, atomization systems, engine wear and the rate of injector fouling, if the oil is to be used for biodiesel. Viscosity increases with the increase in the number of CH_2 moieties in the fatty ester chain and in the degree of saturation [30, 31].

The results showed high kinematic viscosity (33.37 $mm^2.sec^{-1}$) of the extracted oil. The high viscosity of the oil is known to interfere with its direct use in diesel engines (32, 33, 4]. In general, fish oil has higher viscosity compared to petroleum diesel and the kinematic viscosity of 31.09 and 32.40 $mm^2\ sec^{-1}$ at 40 ºC for fish oil obtained from raw and frozen wastes, respectively [15]. However, lower kinematic viscosity (18.44 $mm^2.sec^{-1}$) of fish oil at 40 ºC [34] and 32.1 ($mm^2.sec^{-1}$) at 37 ºC for oil extracted from Tilapia waste were obtained [35]. Therefore, the extracted fish oil must be converted into biodiesel in order to be used in diesel engines. Fish oil high viscosity interferes with its use in diesel engines and can cause problems including: high carbon deposits, scuffing of engine liner, injection nozzle failure, gum formation, lubricating oil thickening and high cloud and pour points [32, 33]. In order to avoid these problems, the oil must be chemically modified to its derivatives which have properties more similar to conventional diesel [32]

Free fatty acids content (FFA)

In the process of converting oil to biodiesel the free fatty acids and triglycerides contained in the oil must be reduced to fatty acid alkyl esters (FAAEs) [36]. Low value of free fatty acid (less than 5%) is required when fish oil is to be used for biodiesel production. Higher concentration of free fatty acids (FFA's) may pose a problem of soap formation and lead to under reacted material, thus

TABLE 2. Oil production from tilapia viscera using two extraction methods.

Parameters	Extraction method	
	Method A	Method B
Sample weight (g)	400	400
Oil weight (g)	40±1.35	80±0.92
Oil yield (%)	10	20

Method A: Short heating (20 min) at lower temperature (80ºC) followed by centrifugation
Method B: Long heating (50 min) at high temperature (100ºC)

TABLE 3. Oil production from tilapia viscera at different cooking temperatures and 50 min cooking time.

Parameters	**Cooking Temperature**	
	80 ºC	100 ºC
Sample weight (g)	400	400
Oil weight (g)	44±.08	80±0.92
Oil yield (%)	11	20

TABLE 4. Characteristics of the extracted oil from tilapia viscera.

Characteristic	**Value**
Kinematic viscosity mm^2/sec @ 40º C	33.37
Free fatty acids (as oleic acid) (%)	3
Acid value (mg KOH / mg)	5.97
Moisture content (%)	0.14

affecting the yield [37]. When the level of FFA is higher than 5%, the soap inhibits separation of the methyl or ethyl esters and glycerol and contributes to emulsion formation during washing with water [11].

The FFA value obtained from this study is 3%. This compared favorably with the value of 3.1% obtained by oliveira et al [11]. On the other hand, FFA in the current study was lower than the study which performed by Suseno et al [13] who found that FFA was in the range of 3.85-7.15% for oil extracted from Tilapia fish. According to the International Association of Fish Meal and Oil Manufacturers (IFOMA) Standards, the standard FFA value for fish oil is in the range of 1-7% measured as oleic acid [38]. The FFA value for fish oil obtained in this study was within the range recommended by the IFOMA standards.

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 [39]. It is used as a guide in quality control as well as in monitoring oil degradation during storage. The acid value obtained in this study was 5.97, this compared favorably with the value of 5.8 of Tilapia viscera obtained by oliveira et al [11]. Acid value in the range of 6.5-12.5 for fish oil extracted from Tilapia obtained by Suseno et al [13] and acid value of 10.04 for oil extracted from fish canning industry wastes [40]. The acid value obtained in this study was comparable to the values reported in the literature.

Fatty acid methyl ester composition

Table 5 shows the fatty acid composition according to the results of GC, The fatty acid methyl ester profile indicated that the most potent fatty acid chains were found in the range of C14 and C22 which in turn determines the promising properties of the biodiesel that might be produced from the extracted oil. In agreement to the current study Santos et al [35], reported that

fatty acids of the Tilapia visceral oil were in the range of C16:0 and C22:6. The major fatty acids were oleic acid, palmitic acid, Linoleic acid and Palmitoleic acid. Also, it was observed that Oleic acid (C18:1) was the major component with 41.54 % of the composition. In agreement to our results, Dias [41] reported that oleic acid (C18: 1) is a major fatty acid present in the composition of oil extracted from the viscera of tilapia.

Moisture content

The moisture content is another factor that strongly influences the performance of the biodiesel production process. Both water and FFA readily react with the catalyst to form soaps which interferes with efficient separation of pure glycerol in the final step of the process. In this study the moisture content of the oil was 0.14, this compared favorably with the value of 0.12% obtained by oliveira et al [11]. On the other hand moisture content was lower than the study obtained by Santos et al [35], who reported that moisture content of oil extracted from Tilapia processing wastes was 0.3% as well as 0.28% obtained for oil extracted from fish canning industry wastes [40]. The low moisture content obtained in this study was comparable to the values reported in the literature. Low moisture content is required for the efficient transformation of fish oil into biodiesel.

Conculosions

This study fish oil was extracted by using wet rendering extraction method; where using water base in the extraction method with more yield is a green chemistry which is better than using solvent extraction which is hazardous chemicals. The extracted fish oil moisture content, acid value, free fatty acids content and kinematic viscosity were 0.14%, 5.97 mg KOH/ mg, 3%, 33.37 mm^2/sec, respectively. The low value of FFA and moisture content obtained in this study are very promising and indicates the applicability of efficient production of biodiesel from the extracted fish oil. The fatty acid methyl ester profile obtained from GC analysis indicated that the most potent fatty acid chains were found in the range of C14 and C22, which also determines the promising properties of the biodiesel that might be produced from the extracted oil.

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TABLE 5. Fatty acid composition of Tilapia visceral oil

Fatty Acids	%
Myristic acid (C14:0)	3.58
Palmitic (C16:0)	24.1
Palmitoleic (C16:1)	6.87
Stearic (C18:0)	0.00
Oleic (C18:1)	41.54
Linoleic (C18:2)	19.86
Linolenic (C18:3)	1.44
Eicosenoic (C20:1)	1.70
Docosahexanoic acid (22:6)	0.91
Total Saturated Fatty Acids (TSF)	27.68
Total Unsaturated Fatty Acids (TUSF)	72.32
TUSF/TSF	2.61

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استخلاص زيت السمك من أحشاء السمك

فاطمة عبد الرحمن عبد الوهاب[١]، نسرين سمير محمود[١]، ابو الخير بدوى السيد[٢] و سامى محمد يونس[١]
[١] قسم الهندسة الزراعية – كلية الزراعة – جامعة القاهرة و [٢] وحدة تكنولوجيا الطحالب ، قسم تكنولوجيا التسميد - المركزالقومى للبحوث - الجيزة - مصر.

فى هذة الدراسة تم استخلاص الزيت من احشاء كلا من سمك البلطى والماكريل بطريقة الفرم الرطب والزيت الكيميائيه لهذا الزيت. وقد وجد أن زيادة درجة الحرارة من ٨٠ درجة مئوى الى çالناتج مع تفدير بعض الخواص ١٠٠ درجة مئوى له تاثير عالى فى زيادة انتاجية الزيت المستخلص. ووجد ايضا ان الطرد المركزى ليس له تأثير فى استخلاص الزيت عند استخدام درجات الحرارة العالية (١٠٠) درجة مئوى ، ولذلك يمكن الغاء الطرد المركزى من عملية الاستخلاص. واظهرت النتائج إمكانية انتاج زيت السمك من أحشاء السمك ونسبة الزيت الناتج من أحشاء البلطى حوالى ٢٠٪ اعلى من انتاجية الزيت الناتج من احشاء الماكريل بحوالى ٧٪. زيادة زمن الاستخلاص من ٣٠ دقيقة اى ٦٠ دقيقة له تأثير عالى على انتاجية الزيت المستخلص . ووجد ايضا ان انسب زمن للاستخلاص هو ٦٠ دقيقة . تم تحليل الزيت المتسخلص من حيث المحتوى المائى ، رقم الحموضة ، الاحماض الدهنية الحرة واللزوجة الكينماتيكية وايضا تحليل الغاز كروماتوجرافى واشارت النتائج الى امكانية استخدام زيت السمك الناتج فى انتاج الديزل الحيوى.