

PRELIMINARY STUDY ON THE CHARACTERIZATION OF OIL FROM NURSE TETRA (*Brycinus nurse*) FISH

M. M. Bello^{1*}, F. A. Oluwole² and B. A. Jajere¹

(¹Department of Fisheries, Faculty of Agriculture, University of Maiduguri, P.M.B.1069, Maiduguri
Nigeria

²Department of Mechanical Engineering, Faculty of Engineering, University of Maiduguri,
P.M.B.1069, Maiduguri Nigeria)

*Correspondence e-mail: mmbello@unimaid.edu.ng Telephone: +234-803-5663-565

Abstract

Fish is an important source of protein providing essential amino acids. Imported fish oil are expensive, scarce and sometimes unavailable. However, extraction of oil from indigenous fish species will provide cheap, abundant and readily available product. This study therefore, aimed at the extraction of fish oil from *Brycinus nurse*. A total of 1368g of *B. nurse* was procured from Lake Alau, Borno State. The fish were divided into four samples A, B, C and D respectively. Sample A was oven dried for a period of 60 minutes, at maximum temperature of 70°C, sample B for 90 minutes at a maximum temperature of 96°C, sample C for 60 minutes at maximum of 96°C, and sample D for 90 minutes at a maximum temperature of 70°C. After oven drying, the samples were immediately transferred to mechanical workshop for oil extraction using hydraulic press. The characterization and the quality of fish oil were measured using the acid value, saponification value and relative density. Results showed that the fish oil from samples A, B, C, and D had acid value of 3.57mg/KOH, 3.59mg/KOH, 2.9mg/KOH, and 2.75mg/KOH respectively, the saponification value of 82.8mg/KOH/g, 94.42mg/KOH/g, 82.8mg/KOH/g, and 70mg/KOH/g respectively while the relative density was found to be 0.04305 for sample A, 0.04301 for sample B, sample C 0.0433 and sample D 0.04307. It can be concluded that the fish oil values falls within the acceptable standard value which are suitable for application in pharmaceutical and food industries. Therefore, *Brycinus nurse* has the potential of producing fish oil for domestic and industrial use.

Keywords: *Brycinus nurse*, mechanical extraction, fish oil, characterization, saponification value.

1. Introduction

Fish are aquatic vertebrate that are typically cool blooded animals covered with scales and equipped with two sets of paired and several un-paired fins. Fish is an important source of cheap first class protein providing essential amino acids (Paul and Southgate, 1978). It is low in fat and cholesterol and rich in calcium, phosphorus and vitamin A and D (Osuji, 1976). It is cheap and highly acceptable with little or no religious bias, which gives it an advantage over pork or beef (Eyo, 2001).

In December 1973, a conclusion was reached by Food and Agriculture Organization of the United Nation (FAO) technical conference, held in Tokyo, that conversion of fish is necessary for preservation of waste while ensuring the beneficial exploitation which cannot be gainfully used for direct human consumption (FAO, 1986). This among other things necessitated various researches into the production of fish oil. Apart from its various uses as consumable oil and other beneficial uses, the production of fish oil is necessary in the utilization of those fish species regarded as un-saleable and unpalatable, and those species too small and which quickly turn rancid for economic storage. Fish oil is the lipid fraction extracted from fish and fish by products. *Scombroids* (mackerel) and *Clupeids* (herring) provides the largest single source of raw material for production of fish oil and fish meal. They are regarded as fatty species, having fat content well distributed throughout the body (FAO, 1986). Generally, fish oils are more complex than terrestrial animal oil or vegetable oil due to long chain unsaturated fatty acid (Hall, 1992). Fish oil is unique in the variety

of fatty acid of which they are composed and their degree of un-saturation (Ackman *et al.*, 1982). Fish oil contains essential long-chain polyunsaturated omega-3 fatty acids such as *Eicosapentaenoic acid* (EPA) and *Docosahexaenoic acid* (DHA) which are essential in human diet (Sidhu, 2003). These omega-3 fatty acids are believed to have health benefits ranging from reducing the risk of heart attack and coronary heart disease to combating depression bipolar disorder and schizophrenia. There is evidence from multiple studies supporting intake of recommended amounts of DHA and EPA in form of dietary fish or fish oil supplement, reduces the risk of death, heart attack, lowers triglycerides, dangerous abnormal heart rhythms and strokes in people with known cardiovascular disease, slows the buildup of atherosclerotic plaques (hardening of the arteries), and lowers blood pressure slightly (Lam, 2002). Fish oil comes from cold water fish such as mackerel, tuna, salmon, cod and other fishes, it is recommended for a heart healthy diet. The importance of fish oils has increased greatly during recent years as the nutritive values of certain components is being emphasized. The consumption of fish oil supplement can aid in the treatment of some inflammatory condition such as rheumatoid arthritis, ulcerative colitis, psoriasis, asthma, lupus and cystic fibrosis (Cleland *et al.*, 2003; Ruxton *et al.*, 2004). Fish oil supplementation does not appear to induce any clinically significant side effect, with majority of research not reporting an adverse effect at all. Some adverse effects from consuming fish include a fishy after taste, gastrointestinal disturbance and nausea (Kris-Ethertan *et al.*, 2002; Peet and Stokea, 2005). Current medical research suggests that these fatty acids have a unique role to play in prevention of coronary artery disease and the growth of different types of cancers. Presently, the production of fish oil is becoming more demanding as there is a growing world market demand for high quality fish oils. In order to meet the demand of the society there is the need to locate new oil fish and do further research to know their characteristic and usefulness. This study was aimed at the extraction and characterization of fish oil from an indigenous species of *Brycinus nurse*.

2. Material and Methods

2.1 Experimental Station

This study was carried out at the Faculty of Engineering Teaching Workshop, University of Maiduguri, Nigeria.

2.2 Sample Collection and Preparation

A total of 1368.4g of *Brycinus nurse* was bought from Alau Lake, Borno State, Nigeria. The fish was stored in an insulated flask during transportation. The fish was beheaded, degutted, descaled and the fins were removed. It was washed thoroughly with water to remove slime and blood and then transferred into a basket for proper draining prior to oven drying.

2.3 Oven Drying

The samples were oven dried in an electric oven at different temperatures and time to observe the effect of such factors on acid value, saponification value and relative density of fish oil. The fish were divided into four samples of 342.1g each and labeled samples A, B, C and D. Sample A was oven dried for a period of 60 minutes at maximum temperature of 70°C, sample B for 90 minutes at maximum temperature of 96°C, sample C for 60 minutes at maximum temperature of 96°C and sample D for 90 minutes at maximum temperature of 70°C. They dried samples were immediately transferred into a clothing material for the oil extraction.

2.4 Oil Extraction Method

Each of the fish samples was packed inside a clothing material and placed inside a perforated cylindrical cage. The perforated cylindrical cage with the sample was placed on the hydraulic press shown in Figure 1 and the lever of the press operated manually to compress the fish. As the compression increases oil starts gushing out, at a pressure of 135KN/m^2 as indicated on the pressure gauge on the hydraulic press, the compression was stopped and maintained for about 15 minutes as was observed during the preliminary investigation. This was replicated 3 times and the average value recorded. The expressed oil was collected in a measuring cylinder and recorded as the oil yield.

The oil yield (O_Y) was calculated using the equation below:

$$O_Y = \frac{M_{oil}}{M_{Fish}} \times 100 \% \quad (1)$$

where: M_{Oil} = mass of expressed oil (g), M_{Fish} = mass of fish sample (g). The values are not reported in this work.

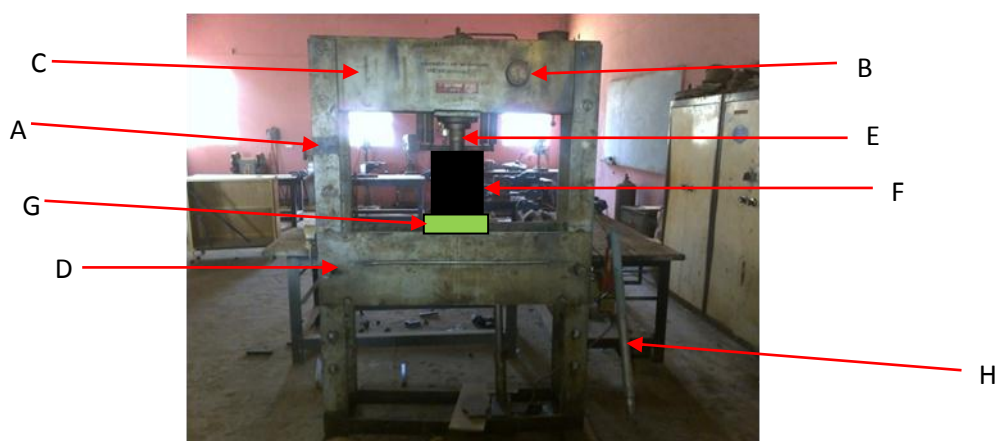


Figure 1: Hydraulic Press used for the oil Expression (Model: CAM Radstock-Avon 076134226)

Main frame (A); Graduated pressure gauge (B); Fixed upper beam (C); Fixed lower beam (D); Moveable spindle (E); Press cage cylinder (F); Adjustable supporting platform (G); Lever arm (H).

2.5 Characterization of Fish Oil

The characterization of the expressed oil was carried out at the National Agency for Food, Drugs and Administration Control (NAFDAC) Laboratory, Maiduguri, to determine the saponification value, acid value and relative density.

2.5.1 Determination of Acid Value (AV)

The method described by ASTM (2002) was used in determining the acid value of the fish oil. Twenty-five ml of diethylether was mixed with 25ml of ethanol in a flask. About 2g of each oil sample was dissolved in the prepared solution and 1ml of 1% phenolphthalein was added and then heated in a water bath for 15min. The hot mixture was titrated with 0.1ml sodium hydroxide (NaOH)

as base, using phenolphthalein end point (pink) as an indicator. The end point was noted when the addition of a single drop produced a slight colour change persisting for at least 15s this was replicated three times. The acid value was determined using the following expression:

$$\text{Acid Value (AV)} = \frac{\text{Titration (ml)} \times 5.61}{\text{weight of sample used}} \quad (2)$$

2.5.2 Determination of Saponification Values

The method used was as described by Shridhar *et al.*, (2010). To determine the saponification value, 2 g of the oil sample was weighed into 250 ml conical flask with accuracy of 1mg. 50ml of 0.5N ethanolic potassium hydroxide solution was added to the cold oil and the reflux condenser attached to the flask. The mixture was heated, and when the ethanol started to boil, the flask was shaken occasionally until the oil was completely dissolved, 1ml of phenolphthalein indicator was added and the hot soap solution obtained was slowly titrated with 0.5N hydrochloric acid (and volume V_a was recorded). A blank determination was carried out upon the same quantity of potassium hydroxide solution at the same time and under the same conditions (and volume V_b was recorded). The saponification value was determined using the following expression:

$$\text{Saponification Value (SV)} = \frac{(V_b - V_a)28.05}{\text{wt of sample}} \text{ (Mg KOH/g)} \quad (3)$$

where: V_a = volume of hydrochloric acid used in the test, ml, V_b = volume of hydrochloric acid used in blank, ml

2.5.3 Determination of Relative Density (Specific gravity)

Density bottle was used to determine the specific gravity of the oil following the procedure described by Shridhar *et al.*, (2010). A clean and dry density bottle of 25ml capacity was weighed (W_0) and then filled with the oil sample, stopper inserted and weighed to give (W_1). The oil was substituted with water after washing and drying the density bottle and weighed to give (W_2). The expression for specific gravity is:

$$\text{Sp. gr.} = \frac{(W_1 - W_0)}{(W_2 - W_0)} = \frac{\text{mass of substance}}{\text{mass of equal vol. of water}} \quad (4)$$

where, W_0 = mass of empty density bottle, g

W_1 = mass of density bottle filled with oil, g

W_2 = mass of density bottle filled with water, g

2.6 Statistical Analysis

Data on oil properties collected were subjected to one-way analysis of variance (ANOVA)

Experimental data generated in the oil expression process were statistically analyzed using Design-Expert 7.0 Software (Stat-Ease Inc. USA).

3. Results and Discussion

Table 1 shows the results of the oil yield and characterization of the extracted oil, while Tables 2, 3, 4 and 5 show the ANOVA of acid value, saponification value, relative density and oil yield respectively. The observed differences in Tables 2, 3 and 4 were not significant; however, the heating temperature significantly affected the oil yield as observed in Table 5.

Table 1: Characterization of Fish oil

| Parameters | A | B | C | D |
|----------------------|---------|---------|--------|--------|
| Time (minutes) | 60 | 90 | 60 | 90 |
| Temperature (°C) | 70 | 96 | 96 | 70 |
| Acid value mg/KOH | 3.5 | 3.6 | 2.9 | 2.75 |
| Saponification value | 94.42 | 84 | 82.8 | 90 |
| Relative density | 0.04305 | 0.04301 | 0.0433 | 0.0407 |
| Oil Yield % | 1.37 | 3.40 | 2.58 | 2.04 |

Table 2. ANOVA Table of Acid Value

| Source | Sum of Squares | df | Mean Square | F Value | p-value Prob > F | |
|------------------------|----------------|----|-------------|---------|------------------|-----------------|
| Model | 0.28 | 2 | 0.141 | 2.778 | 0.3906 | not significant |
| A-Heating Time | 0.01 | 1 | 0.006 | 0.111 | 0.7952 | |
| B- Heating Temperature | 0.28 | 1 | 0.276 | 5.444 | 0.2578 | |
| Residual | 0.05 | 1 | 0.051 | | | |
| Cor Total | 0.33 | 3 | | | | |

R-Squared = 0.85

Table 3. ANOVA Table of Saponification Value

| Source | Sum of Squares | df | Mean Square | F Value | p-value Prob > F | |
|------------------------|----------------|----|-------------|---------|------------------|-----------------|
| Model | 85.51 | 2 | 42.756 | 16.495 | 0.1715 | not significant |
| A-Heating Time | 77.62 | 1 | 77.616 | 29.943 | 0.1151 | |
| B- Heating Temperature | 7.90 | 1 | 7.896 | 3.046 | 0.3312 | |
| Residual | 2.59 | 1 | 2.592 | | | |
| Cor Total | 88.10 | 3 | | | | |

R-Squared = 0.97

Table 4. ANOVA Table of Relative Density

| Source | Sum of Squares | df | Mean Square | F Value | p-value Prob > F | |
|------------------------|----------------|----|-------------|---------|------------------|-----------------|
| Model | 3.31E-08 | 2 | 1.65E-08 | 0.9067 | 0.5962 | not significant |
| A-Heating Time | 9.03E-09 | 1 | 9.03E-09 | 0.4952 | 0.6096 | |
| B- Heating Temperature | 2.4E-08 | 1 | 2.4E-08 | 1.3182 | 0.4562 | |
| Residual | 1.82E-08 | 1 | 1.82E-08 | | | |
| Cor Total | 5.13E-08 | 3 | | | | |

R-Squared = 0.64

Values of "Prob > F" less than 0.0500 indicate model terms are significant. In this case, heating temperature, B are significant model terms.

Table 5. ANOVA Oil Yield (%)

| Source | Sum of Squares | df | Mean Square | F-Value | p-value Prob > F | |
|------------------------|----------------|----|-------------|----------|------------------|-----------------|
| Model | 2.2063 | 2 | 1.1031 | 196.1111 | 0.0504 | not significant |
| A-Heating Time | 0.5550 | 1 | 0.5550 | 98.6711 | 0.0639 | |
| B- Heating Temperature | 1.6512 | 1 | 1.6512 | 293.5511 | 0.0371 | |
| Residual | 0.0056 | 1 | 0.0056 | | | |
| Cor Total | 2.2119 | 3 | | | | |
| R-Squared = 0.9975 | | | | | | |

The response surface contours, which are the graphical results of the interactive effects of heating temperature and heating time (A x B) on acid value, saponification value, relative density and oil yield are shown in Figures 1, 2, 3 and 4 respectively.

Design-Expert® Software

Acid Value

● Design points above predicted value

○ Design points below predicted value



X1 = A: TIME
X2 = B: TEMPERATURE

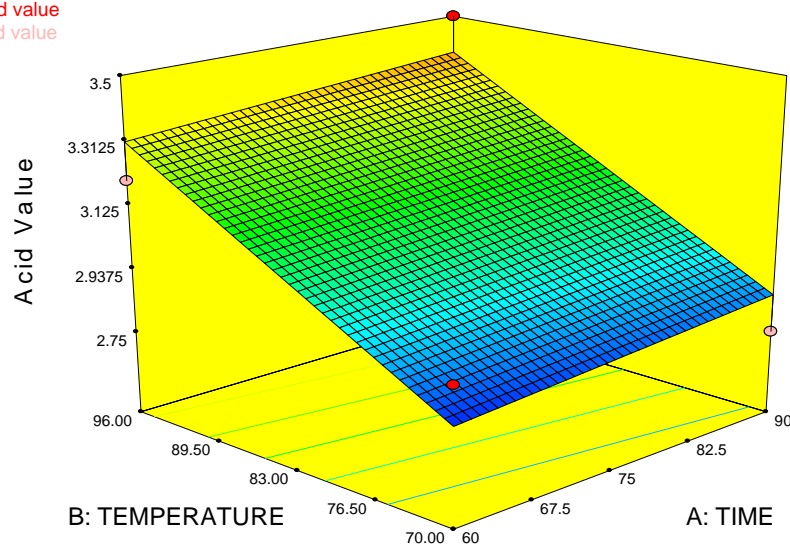


Figure 1: Relationship between in acid value, heating time and heating temperature

Design-Expert® Software

Saponification Value

● Design points above predicted value

○ Design points below predicted value

94.42

82.8

X1 = A: TIME

X2 = B: TEMPERATURE

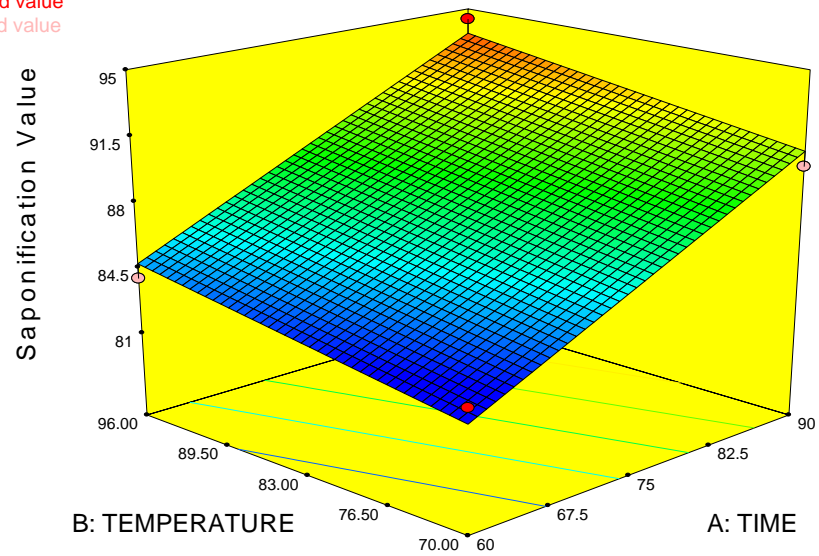


Figure 2: Relationship between saponification value, heating time and heating temperature

Design-Expert® Software

Relative Density

● Design points above predicted value

○ Design points below predicted value

0.0433

0.04301

X1 = A: TIME

X2 = B: TEMPERATURE

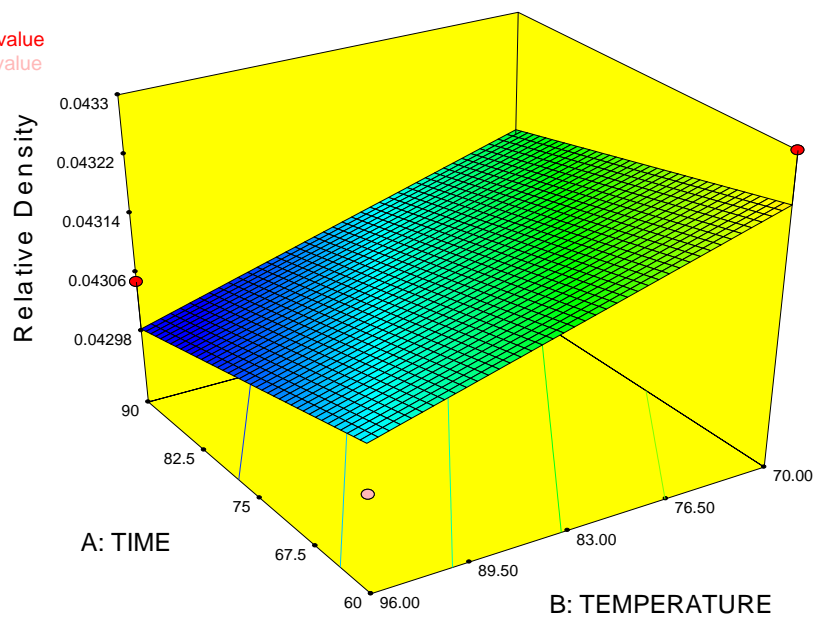


Figure 3: Relationship between saponification value, heating time and heating temperature

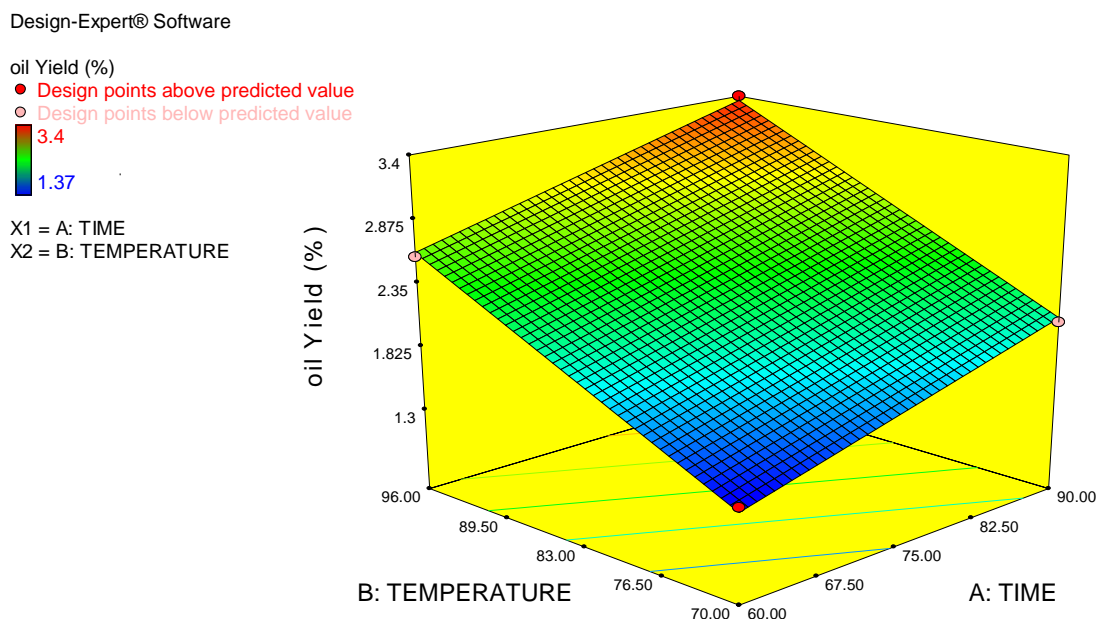


Figure 4: Relationship between Oil Yield value, heating time and heating temperature

Figures 1 and 2 show that as the heating temperature and heating time increase the acid and saponification values increased. Figure 3 shows that the relative density decreased with increase in heating temperature and heating time. This agrees with the findings of Oluwole *et al.*, (2015).

Table 1 showed that Sample A had acid value 3.5mg/KOH, B, 3.6mg/KOH, C, 2.9mg/KOH and D 2.75mg/KOH. These values agreed with the results on mackerel oil in which it was found that the crude oil had an acid value of 2.5 mg/KOH, whereas the refined oil had 2.27mg/KOH. The saponification value sample A was 82.8mg/KOH/g, B, 94.42mg/KOH/g, C, 82.8mg/KOH/g and D, 70mg/KOH/g. These values were slightly lower than the standard value for fish oil (165 -195 mg/KOH/g). This may be as a result of the fish pre-treatment methods. The relative density was found to be 0.04305 for sample A, 0.04301 for B, C had 0.0433 and D, 0.04307.

Tables 2, 3 and 4 show that the observed acid values, saponification values and relative density of the oils were not significantly different. Table 5, however, shows that the observed differences in oil yield were significant. This agrees with the finding of Oluwole *et al.* (2015).

4. Conclusion

The investigation of the properties of oil expressed from fish subjected to some pre-treatment conditions using hydraulic press revealed the following:

- i. The quality of fish oil by measurement of acid value, saponification value and relative density was within the recommended values.
- ii. *Brycinus nurse* oil are suitable for application in pharmaceutical and food industries.
- iii. Heating temperature and heating time affected the oil yield positively.

References

- Ackman, RG., Barlow, SM. and Stansby, ME. 1982. Fatty Acid Composition of Fish Oils: Nutritional evaluation of long-chain fatty acids in fish oil. Academic Press, New York, p.25-80.
- ASTM. 2002. Petroleum Products, Lubricants and Fossil Fuels. Annual Book of ASTM Standards Section Five. 05. 01. D2597-D4927. ASTM International, West Conshohocken, PA. USA.
- Cleland, L., James, M. and Proudman, S. 2003. The role of fish oils in the treatment of rheumatoid arthritis. *Drugs*, 63: 845-853.
- Eyo, AA. 2001. Fish processing Technology in the Tropics. University of Ilorin Press. Ilorin, pp. 112-129.
- FAO 1986. The production of fish meal and oil. Food and Agricultural Organization of the United Nations, Fishery Industries Division, Technical Paper, FAO, Rome, pp142.
- Hall, GM. 1992. Fish Process Technology. Food Engineering and Biotechnology Group, University of Technology, Loughborough, pp. 4-7 and 172-181.
- Kris-Ethertan, PM., Harris, WS. and Appel, LJ. 2002. Fish consumption, fish oil, Omega-3 fatty acids and cardiovascular disease. *Circulation*, 106: 747-757.
- Lam, M. 2002. Omega-3 fatty acids, Academy of Anti-ageing Research, New York, USA, pp. 24.
- Oluwole, FA., Aviara, NA. Umar, B. and Muhammad, AB. 2015. Influence of variety and pre-treatment on oil properties of mechanically expressed castor oil. *Global Advanced Research Journal of Engineering, Technology and Innovation*, 4(1): 001-009.
- Osuji, FNC. 1976. The influence of traditional handling methods on the quality of processed fish in Nigeria. In: Proceedings of the conference on the Handling, processing and marketing of Tropical fish. 13th to 19th April, 1976 organized by the Tropical Product Institute, London, pp201.
- Peet, M. and Stokea, C. 2005. Omega-3 Fatty Acids in the treatment of psychiatric disorders drugs. *Polyunsaturated Fatty acids in Acylylglycerols from Marine oils*, 65 (8): 1051-1059.
- Paul, W. and Southgate, L. 1978. Fish and Meat Consumption in the Tropics. MacMillan, London, pp 287.
- Ruxton, CHS., Reed, SC., Simpson, MJA., and Millington, KJ. 2004. The health benefits of omega-3 Polyunsaturated fatty acids: a review of the evidence. *Journal Human Nutrition and Dietetics*, 17: 449-459.
- Shridhar, BS., Beena, KV., Anita, MV. and Paramjeet, KB. 2010. Optimization and characterization of castor seed oil. *Leonardo Journal of Sciences*, 17: 59-70.
- Sidhu, KS. 2003. Health benefits and potential risks related to consumption of fish or fish oil. *Regulations on Toxicological Pharmacology*, 38:336-344.