Project Report
On
"Assessment of Oil Extracted from *Pterygota alata* Seeds"

Submitted by

Koushik Mittra
ID: 163-34-574
Department of Nutrition & Food Engineering
Daffodil International University

Supervised by

Nasima Akter Mukta
Lecturer (Senior Scale)
Department of Nutrition & Food Engineering
Daffodil International University

Prof. Dr. Md. Abdur Rahim
Dean
Faculty of Agriculture
Bangladesh Agricultural University

Date of Submission: 20th June, 2021

Table of Contents

Certificate of Approval

This is to certify that the research work presented in the report titled "Assessment of Oil Extracted from *Pterygota alata* Seeds" is being submitted by Koushik Mittra, bearing ID: 163-34-574 is an authentic work which is carried out in the laboratory of the Department of Nutrition and Food Engineering, Daffodil International University, Dhaka. The project report is approved for the partial fulfillment of the Bachelor degree of Science in Nutrition and Food Engineering.

…… **Dr. Sheikh Mahatabuddin** Head Department of Nutrition & Food Engineering Daffodil International University.	 …… **Nasima Akter Mukta** Lecturer (Senior Scale) Department of Nutrition & Food Engineering Daffodil International University	 …… **Prof. Dr. Md. Abdur Rahim** Dean Faculty of Agriculture Bangladesh Agricultural University

Letter of Transmittal

Date: 20th June, 2021
Dr. Sheikh Mahatabuddin
Head
Department of Nutrition & Food Engineering
Daffodil International University.

Subject: Submission of Project Report

Dear Sir,

This is my pleasure to submit the project report titled "Assessment of Oil Extracted from *Pterygota alata* Seeds" as partial fulfillment for the requirement of the BSc. in Nutrition & Food Engineering (NFE) program. I would like to thank you for your support and guidance.

I made my best effort in collecting and studying necessary data to make the report as analytical as possible. The practical and analytical knowledge gathered during the thesis work will be fruitful for my career.

I therefore would like to submit this report to you for your kind suggestion and consideration.

Sincerely yours,

Koushik Mittra

……..

Koushik Mittra

ID: 163-34-574

Dept. of Nutrition and Food Engineering

Daffodil International University.

Declaration

This project report entitled “Assessment of Oil Extracted from *Pterygota alata* Seeds” is being submitted to the Department of Nutrition and Food Engineering, Faculty of Allied Health Sciences, Daffodil International University, Dhaka, Bangladesh as a part of partial fulfillment of the requirements for the degree of Bachelor of Science in Nutrition & Food Engineering. This project report is unique and carried out by Koushik Mittra’s authentic work.

Submitted by

Koushik Mittra
ID: 163-34-574
Dept. Of Nutrition and Food Engineering
Daffodil International University

Supervised by

……

Nasima Akter Mukta
Project Supervisor
Lecturer (Senior Scale)
Department of Nutrition & Food Engineering
Daffodil International University

……

Prof. Dr. Md. Abdur Rahim
Project Co-Supervisor
Dean
Faculty of Agriculture
Bangladesh Agricultural University

Acknowledgement

At first, I would like to express my gratitude to my Project Supervisor **Nasima Akter Mukta,** Lecturer (Senior Scale)**,** Department of Nutrition & Food Engineering for her continuous supervision, support and advice without which it would be impossible for me to complete the thesis work. I am also grateful to my Project Co-supervisor **Prof. Dr. Md. Abdur Rahim**, Dean, Faculty of Agriculture, Bangladesh Agricultural University for his important advice and support during the entire thesis work.

My sincere thanks go to **Dr. Sheikh Mahatabuddin**, Head, Department of Nutrition & Food Engineering, Daffodil International University and to **Professor Dr. Ahmad Ismail Mustafa,** Dean, Faculty of Allied Health Sciences for their suggestion and support to conduct this thesis successfully.

I would definitely like to show my gratitude to the officials of the entire **Department of Nutrition and Food Engineering** for the provided support and facilities during the thesis work.

At last, my gratitude goes to the Almighty to give me strength and patience to complete the work and report successfully.

Abstract

The chemical properties of oil extracted from the seeds of *Pterygota alata* were investigated by established methods. The seed oil was analyzed for acid value, saponification value and iodine value. Oil was extracted from the seeds by solvent extraction process using Petroleum Ether (40-60ºC) as solvent. Amount of oil extracted from the seeds was 42.27% (±0.02) which indicates a very high yield percentage. The properties of the oil extracted from *Pterygota alata* seeds were observed as acid value 0.39 (±0.03) mg KOH/g, iodine value of 0.955 (± 0.036), and saponification value of 207.84 (±1.05) mg KOH/g. The oil may not be suitable for consumption as the iodine value is low which reflects a high degree of saturation. However, the high saponification value and low iodine value indicated that the oil could be a potential raw material for soap processing. The low acid value is a good indication to stability towards hydrolysis during storage and processing of the oil.

Chapter 1:

Introduction

1.1 Chemistry of Fats and Oils

Fats and oils are made of water insoluble non-volatile organic substances called triglycerides (or triacylglycerols) which are esters composed of three fatty acids linked to glycerol. The other minor components of most of the fats comprise phosphatides, fat soluble vitamins such as vitamin A & D, sterols, tocopherols, fatty alcohols, carotenoids & chlorophyll etc.[1] The terms 'oils' and 'fats' generally refer to the same group of compounds and can be used interchangeably[1]. Usually, Fats are solid and Oils are liquid in room temperature. They are commonly found from vegetable source (e.g. palm oil, sunflower oil, soybean oil, olive oil, cocoa butter, etc.) or animal source (e.g., pork lard, fish oil, beef tallow, animal milk fats etc.).

Fatty acids are mainly saturated and unsaturated. Unsaturated fatty acids are grouped by monounsaturated, polyunsaturated and trans. Saturated fatty acids contain single bonds between carbon atoms. Monounsaturated fatty acids contain one and polyunsaturated fatty acids contains two or more double bonds between their C-C bonds. In case of trans fatty acids some or all of the double bonds are in the trans configuration.

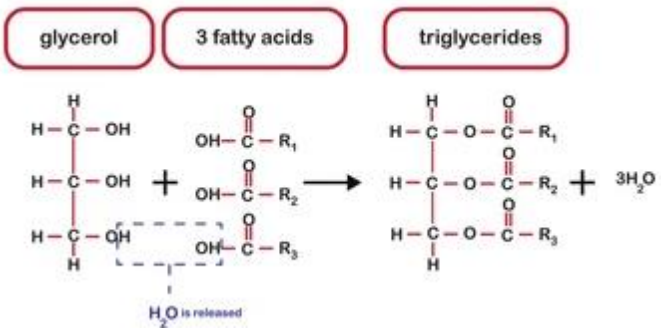

Figure 1.1: Basic Structure of Triglyceride

Fats which contain the largest proportion of saturated fatty acids are solid in room temperature. These fatty acids are less chemically reactive and usually more stable than unsaturated fatty acids. Most of the animal fats such as meat, cream, butter, cheese and vegetable oils like coconut oil, palm oil etc. contain high proportion of saturated fat. Limitation of consuming these fats are necessary.

Trans Fats are most commonly produced as a result of hydrogenation of fats and in used frying oils. These are not acceptable in a healthy diet plan.

On the other hand, cis-unsaturated fatty acids are beneficial for human health as they increase the beneficial HDL cholesterol and decrease the detrimental LDL cholesterol. Olive oil, sunflower oil, rapeseed oil, soybean oil etc. are rich in unsaturated fatty acids.

1.2 Importance of Fat in the Diet

Along with carbohydrates and proteins edible fats and oils are the major components of the human diet. Compared to proteins and carbohydrates (4 Cal/g), Fats are the most energy-rich molecules in the diet containing 9 calories per gram [2]. Recent recommendation for total fat intake is between 20 and 35% of total calories [3,4]. Fats and oils are the source of essential fatty acids such as linoleic, linolenic acid to human. These Essential fatty acids must be consumed in the diet as our human body cannot synthesize them on its own [1]. Linoleic acid (Omega-6/ N-6) and α-linolenic acid (Omega-3/ N-3), both are essential for proper functioning of human body if they are present in balanced proportion in the body. Omega -6: Omega-3 ratio should be 2:1 by the recommendation of some specialists [5].

According to WHO and FAO fats are important in diet for mainly 5 reasons [6] :

1. As energy source
2. For cell structure and membrane function
3. As a vehicle for fat soluble vitamins
4. As source of essential fatty acids for cell structure and prostaglandin synthesis
5. For control of blood lipids

Besides most of the dietary fats contain significant amounts of Vitamin A,D. Almost all the vegetable oils contain Vitamin E [6]. For healthy eating, instead of animal fats plant-based cooking oils should be selected because most of the plant-based oils contain higher amount of polyunsaturated and monounsaturated fats which are beneficial for our heart health and contain much lower proportion of harmful saturated fat which is bad for our hearts.

1.3 Adverse Health Effects of Fats and Oils

All the fats and oils are not good for our health. Some of the fats are responsible for creating negative effect to our body:

- Limiting the consume of saturated fat is recommended as researchers have found that consuming higher amount of saturated fat can increase the cholesterol level causing cardiovascular diseases[7,8].

- Trans fatty acids were found the most harmful of all the fatty acids as they increase the levels of harmful LDL cholesterol and decrease the levels of beneficial HDL cholesterol [9]

- As a small amount of fat can provide high energy, excessive intaking of fat can cause obesity as well as other health problem.

1.4 Application of Fats and Oils

From annually produced Fats and Oils around 80% is used for human food, around 6% for animal feed, and around 14% for the oleochemical industry [10].

- In food applications they are consumed as butter, salad oils, shortening, margarine, cooking oils, mayonnaise etc. Fats and Oils are used to make a lot of processed foods like chocolates, ice-cream and bakery products like cake, biscuits, cookies etc.

- Oleochemicals (chemical compounds derived from natural fats and oils) are used to make soaps, detergents, biodiesel, paints, lubricants, greases, pharmaceutical and cosmetic products etc.

1.5 Overview of "*Pterygota alata*"

Pterygota alata, commonly named as Buddho narikel or Buddha coconut (family: Malvaceae) is an evergreen or semi-deciduous large tree which can grow up to 30-35m. The Trunk of the tree is erect and cylindrical, up to more than 1m diameter with grayish bark based on tabular roots. Leaves are oval and heart-shaped, 10-30 cm long, 8-18 cm wide [11]. The most interesting thing is that it's called Pagla Gach (mad tree) because every leaf of the plant is differently shaped. This tree is found mainly in the hilly area of southern China, India, Bhutan, Bangladesh, Myanmar, Thailand, Vietnam, Malaysia, Philippines [11]. In Bangladesh it is also planted in parks and botanical gardens as an ornamental tree. Fruit is big, round, hard, dehiscent, and brown in color containing numerous winged seeds. Raw or roasted seeds are edible. Some narcotic properties were found in the seeds. It was claimed that oil extracted from the seeds is nutritious [12]. However, a very little scientific research have conducted based on the seeds. Recently antioxidant properties have found from the leaves and bark of the plant [13,14].

Figure 1.2: Fruits of "*Pterygota alata*"

1.6 Origin of the study

Thesis or project report is a requirement for completing the Bachelor in Nutrition and Food Engineering (NFE) at Daffodil International University. Department of NFE provides thesis opportunity for students in the university laboratory. This report was made based on the experiments conducted and by studying related data.

1.7 Purpose of the study

1.7.1 General Objective:

No oil from a particular source has been found to be suitable for all purposes because oils from different sources have different characteristics. So, interest is growing in producing oils from new sources. In this regard, plant seeds are known to be a good source of oils of nutritional, industrial, and pharmaceutical importance. There are many rare and unfamiliar plants whose seeds can be a good source of oils having unique characteristics. ***Pterygota alata*** is such a rare plant. Characterization of ***Pterygota alata*** seed oil will help to know and study the different characteristics of the oil and to determine which purpose it should be used as it can be a valuable ingredient for the food and nonfood industries.

1.7.2 Specific Objectives:

- to investigate the characteristics of oil extracted from ***Pterygota alata*** seed;
- to know the storage stability of the oil;
- to the learn methodologies of finding different characteristic of the oil;
- to find out the applications of the oil; and
- to fulfill the requirement of Bachelor in Nutrition and Food Engineering.

1.8 Limitation of the Study

- Due to the pandemic situation created for COVID-19 some important analysis could not be done.

- Lacking of some chemicals and specific instruments is another reason to not able to conduct some of the important tests.

Chapter 2:

Determination of Chemical Properties of *Pterygota alata* Seed Oil

2.1 Method and Materials

All the experiments were carried out in triplicate by following standard methods and the found data were expressed as the mean ± (standard deviation).

2.1.1 Collection and Processing of Seeds:

Fully matured fresh fruits were harvested from Germplasm Center (GPC), Bangladesh Agricultural University, Mymensingh, Bangladesh under the supervision of Prof. Dr. Md. Abdur Rahim.
Major processing steps are described below:

- Seeds were separated from the hard pod of the fruit by the help of knives.
- Seeds were then separated from the shells.
- Dirt and foreign materials were cleaned.
- Seeds were then dried in the oven at 60ºC for about 3 hours until the moisture content is <8% as drying sample up to 8% moisture content was recommended for better yield [15].
- After drying, homogenous powder was produced by grinding the seeds with a food grinder.
- Powdered sample were packaged with airtight polyethylene bag and kept at -20ºc.

Figure 2.1: a) Seeds of *Pterygota alata*; *b*) Powdered seeds of *Pterygota alata*

2.1.2 Extraction of Oil:

Extraction of oil was done by solvent extraction method with soxhlet apparatus. Oil was extracted for 8h with Petroleum Ether (BP 40-60ºC). Solvent-Sample Ratio was kept as 10:1. Solvent was evaporated with a rotary evaporator under low pressure at 40-45ºC.

Major steps of extraction process are mentioned below:

- At first, weight of empty round bottom flask (RBF) was taken.
- Then the weight of powdered seed sample (~ 25g) was taken and placed in thimble and then thimble was inserted into the Soxhlet apparatus.
- 250 ml of solvent was taken into the round bottom flask and the flask was connected with the Soxhlet apparatus.
- The flask was then fitted with the heating mantel and it was heated at 55ºC for 8 hours.
- After 8 hours the flask was removed from the apparatus and connected with vacuum rotary evaporator and solvent was evaporated at low pressure.
- After finishing the evaporation weight of flask with remaining oil was noted and collected oil was kept in airtight glass bottle and stored at -20ºC until further analysis.

Calculation:

$$\% \text{ of Oil} = \frac{\text{Weight of RBF after evaporation} - \text{weight of empty RBF}}{\text{weight of powdered seed sample}} \times 100$$

Figure 2.2: a) Soxhlet Extraction Process; b) Extracted Oil Sample

2.1.3 Determination of Acid Value

Acid value (AV) is the milligrams of potassium hydroxide required to neutralize the free fatty acids in one gram of fat or oil. As rancidity is usually accompanied by free fatty acid formation, determination of acid value is used as a general indication of the condition of oils. AV is determined by titrating a solution of the oil in solvent mixture with alcoholic potassium hydroxide by following standard AOCS official method (AOCS cd 3d-63)[16].

Reagents:

- 0.1N KOH
- 1% phenolphthalein indicator
- Solvent mixture – Toluene: Isopropyl Alcohol = 1:1

Procedure:

- 125ml solvent mixture was taken in an Erlenmeyer flask and 2ml indicator was mixed.
- Neutralization was done by titrating the solution with 0.1N KOH until faint permanent pink color appeared.
- Then about 20g of oil was taken in another flask (as expected AV was 0-1) and neutralized solvent mixture was added.
- The oil was mixed by shaking.
- Then titration is done by vigorous shaking against 0.1N KOH until permanent pink color persisted 30s.

Calculation:

Formula used to calculate AV is shown below:

$$\text{Acid Value, mg KOH/g of sample} = \frac{(A-B) \times N \times 56.1}{W}$$

Where,
A= ml of KOH used in titration
B= ml of KOH used in titration of Blank
N= Normality of KOH
W= Gram of sample

Figure 2.3: (Acid Value Determination)
a) Blank Solution after Neutralization; b) Sample Solution before titration;
c) Sample solution after titration

2.1.4 Determination of Iodine Value:

Iodine value (IV) is the measure of the degree of unsaturation of an oil. It measures total number of double bonds present in fats and oils. Number of g of iodine which reacts with the double bonds of 100 grams of fat or oil is the IV of that particular fat or oil. The iodine value was determined according to AOCS official method (AOCS Cd 1d-92) by dissolving oil sample in a solvent mixture and then adding iodine monochloride (Wijs solution)[16]. After completion of the reaction, the excess iodine monochloride was decomposed to iodine by the addition of aqueous potassium iodide solution, which was then titrated with standard sodium thiosulfate solution.

Reagents:

- Solvent Mixture- Glacial Acetic Acid: Cyclohexane = 1:1
- 10% Potassium Iodide (KI) solution
- 1% Soluble Starch Solution
- 0.1N Sodium Thiosulfate ($Na_2S_2O_3$) Solution

Procedure:

- Melted oil was filtered at 80-85ºC
- Then ~3g oil sample was taken into Erlenmeyer flask (as expected IV was <5)
- 15 ml solvent mixture was added and oil was dissolved by swirling.
- 25 ml of Wijs solution was dispensed and mixed by swirling and kept the mixture in dark for 1 hour with keeping the glass stopper on.
- After 1 hour 20 ml KI solution and 100 ml dist. water was added.
- Titration was done with adding starch solution until dark blue color has disappeared.
- Titration of blank solution was also conducted with the same manner.

Calculation:

$$\text{Iodine value} = \frac{(B-S) \times N \times 12.69}{\text{Weight of the sample (g)}}$$

Where,
B = Volume of $Na_2S_2O_3$used in titration of Blank
S= Volume of $Na_2S_2O_3$ used in titration of Sample
N= Normality of $Na_2S_2O_3$

Figure 2.4: (Determination of Iodine value)

a) Sample solution before titration
b) Sample solution after adding indicator
c) Sample solution after titration

2.1.5 Determination of saponification value

The saponification value is the milligrams of potassium hydroxide required to neutralize the fatty acids (including free fatty acids) resulting from the complete hydrolysis of 1g of fat. Lower saponification value indicates higher mean molecular weight of fatty acids and vice-versa. Saponification value of the oil was determined by following AOAC Official Method (AOAC 920.160). The oil sample is saponified by refluxing with excess of ethanolic KOH. Triglyceride reacts with Potassium hydroxide and glycerol is produced along with soap (Potassium palmitate) (Fig: 2.5).

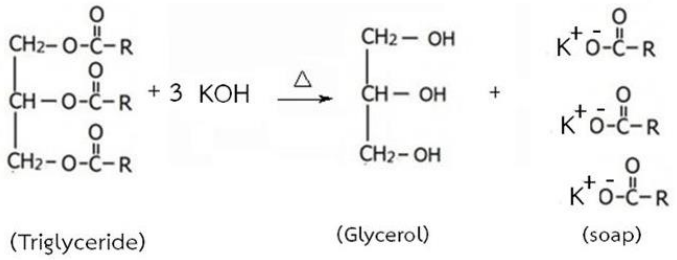

Figure 2.5: Production of Soap during saponification

Apparatus and Machineries:

- Reflux Condenser
- Round Bottom Flask
- Pipette

Reagents:

- Alcoholic Potassium Hydroxide Solution
- 1% Phenolphthalein Indicator
- Hydrochloric acid, 0.5 N

Procedure:

1. About 5g of oil sample was take in Round Bottom flask (RBF)
2. 50 ml of Alcoholic KOH was added and the flask was connected with reflux indicator.
3. RBF was placed in the heating mantle and Boiling was done for about 30 minutes
4. After cooling, 2 drops of indicator was added.
5. Titration was done against 0.5N KOH until purple color disappeared.
6. Blank titration was also done.

Calculation:

Formula used for determination saponification value:
mg of KOH =28.05（B-S) /g of oil

Where,
B= ml of HCL required by blank
S= ml of HCL required for sample

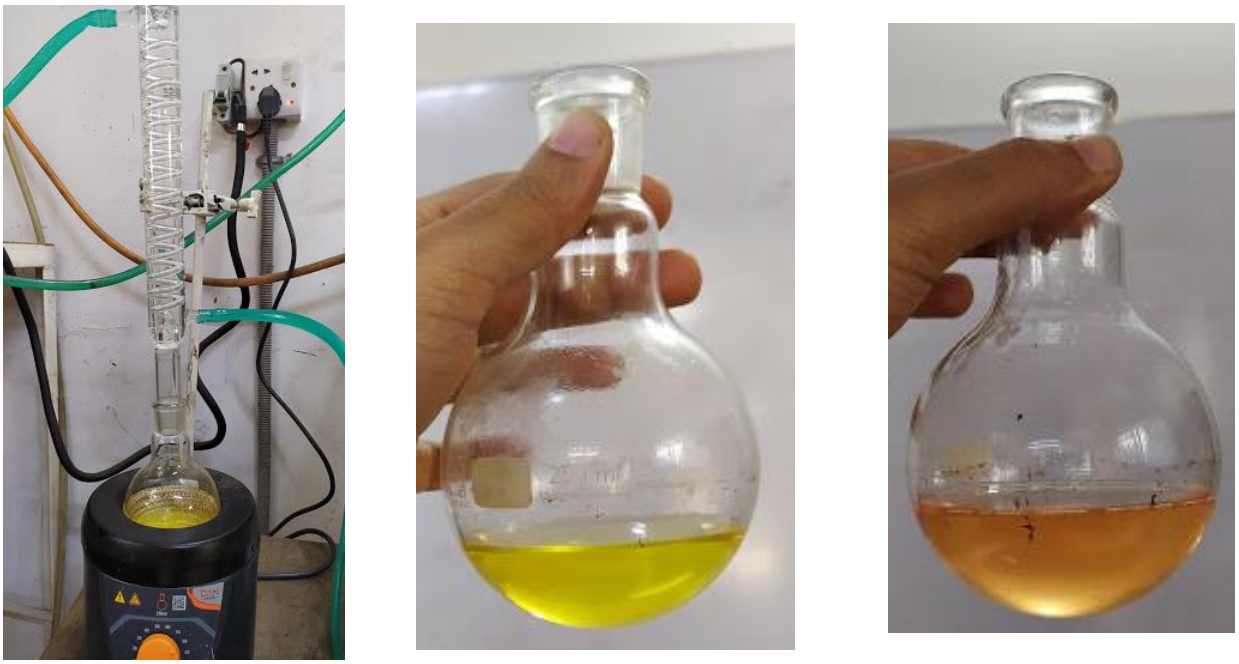

Figure 2.6 a) Refluxion during saponification; b) Sample Solution before saponification;

c) Sample Solution after saponification

Chapter 3:

Result and Discussion

Oil Extraction:

Total yield of oil from the seed sample of *Pterygota alata* was 42.27% (±0.02%), which is higher than reported of *Hibiscus esculentus* (29.31%, ± 0.83), *Hibiscus sabdariffa* (27.22, % ± 0.17) and lower than reported of *Pterygota macrocarpa* (63.97%, ± 0.23) which all are the members of Malvaceae family [17–19]. The significant percentages of oil yield make the seeds suitable for the application in oil industry.

Acid Value:

The acid value of the seeds of *Pterygota alata* obtained in this study was 0.39 (±0.03) mg KOH/g oil which is a permissible value according to Codex Standard for Some Known Vegetable Oils[20]. The lower acid value indicates that the triacylglycerols of the oil have not been hydrolyzed during the processing and storage time (about 3 weeks), which indicates that the oil is stable enough. The oil could be a source towards industrial interest like soap manufacturing for its good stability [21].

Iodine Value:

The iodine value represents the degree of unsaturation of the fatty acids of an oil or fat and that can be used to quantify the amount of double bonds present in the oil which reflects the susceptibility of oil to oxidation. The Iodine Value of *Pterygota alata* seed oil was found 0.955 (± 0.036), which reflects that the oil contains a very high amount of saturated fatty acids. As the iodine value is lower than 100, this oil could be classified as a non-drying oil [22]. The oil may not be the healthy choice for eating as it has high amount of saturated fatty acids which are unhealthy for human body. However, the oil can be used industrially for the production of hard soaps, as non-drying oils don't harden when they are exposed to air [23].

Saponification Value:

Saponification Value found from the oil sample was 207.84 (±1.05) mg KOH/g. The high saponification value indicates that the oil can be a good raw material for soap making [24]. High proportion of saponification value indicates high proportion of shorter chain fatty acids since saponification value is inversely proportional to the average molecular weight or chain length of the fatty acids[25].

Table 1: Chemical Analysis of the oil of *Pterygota alata* Seed:

Parameters	Value
Acid Value (mg KOH/g)	0.39 (±0.03)
Iodine Value	0.955 (± 0.036)
Saponification Value (mg KOH/g)	207.84 (±1.05)

Chapter 4: Conclusion

Conclusion:

This study has been carried out to determine the yield percentage oil extracted from the seeds of *Pterygota alata* and to assess the physicochemical properties of the oil. Total yield of the oil was determined as 42.27% (±0.02%). Due to some inadequacy only acid value, iodine value and saponification value were investigated and the result was obtained as 0.39 (±0.03) mg KOH/g, 0.955 (±0.036), 207.84 (±1.05) mg KOH/g respectively. The oil was primarily found essential for the raw material of soap production as it has high saponification value and low iodine value. The low acid value is appreciated as it is an indication of the stability of the oil. The high yield of oil from the seeds could draw the attention to use of the seeds in oil industry as the natural raw material that are decreasing day by day. The oil may not be a good choice for consuming as an edible oil for its low iodine vale which is a reflection of high saturated fatty acids content. However, more study and research are needed to identify the other physicochemical properties of the oil to understand and ensure its application range as well.

Reference:

(1) Strayer D, Belcher M, Dawson T, Delaney B, Fine J, Flickinger B, Friedman P, H. C.; Hughes J, Kincs F, Linsen L, McBrayer T, McCaskill D, McNeill G, Nugent M, P. E.; Rosegrant P, Tiffany T, Wainwright B, W. J. *Food Fats and Oils.*, 9th ed.; Institute of Shortening and Edible Oils.: Washington, DC, 2006.

(2) Weber, N. C. E. Stauffer: Fats & Oils. Eagan Press Handbook Series. 149 Pages, 50 Illustrations and 24 Tables. Eagan Press, St. Paul, Minnesota, USA. Price: 59.- US $. *Food / Nahrung* **1996**. https://doi.org/10.1002/food.19960400514.

(3) Fats and Fatty Acids in Human Nutrition. Report of an Expert Consultation. *FAO Food Nutr. Pap.* **2010**.

(4) Trumbo, P.; Schlicker, S.; Yates, A. A.; Poos, M. Dietary Reference Intakes for Energy, Carbohydrate, Fiber, Fat, Fatty Acids, Cholesterol, Protein and Amino Acids. *J. Am. Diet. Assoc.* **2002**. https://doi.org/10.1016/S0002-8223(02)90346-9.

(5) Simopoulos, A. P.; Leaf, A.; Salem, N. Workshop on the Essentiality of and Recommended Dietary Intakes for Omega-6 and Omega-3 Fatty Acids. *J. Am. Coll. Nutr.* **1999**. https://doi.org/10.1080/07315724.1999.10718888.

(6) FAO & WHO. *Fats and Oils in Human Nutrition*; Rome, 1977.

(7) Choe, E. Effects and Mechanisms of Minor Compounds in Oil on Lipid Oxidation. In *Food Lipids: Chemistry, Nutrition, and Biotechnology, Fourth Edition*; 2017. https://doi.org/10.1201/9781315151854.

(8) Bester, D.; Esterhuyse, A. J.; Truter, E. J.; Van Rooyen, J. Cardiovascular Effects of Edible Oils: A Comparison between Four Popular Edible Oils. *Nutr. Res. Rev.* **2010**. https://doi.org/10.1017/S0954422410000223.

(9) Hu, F. B.; Willett, W. C. Optimal Diets for Prevention of Coronary Heart Disease. *J. Am. Med. Assoc.* **2002**. https://doi.org/10.1001/jama.288.20.2569.

(10) McKeon, T.; Sharma, B.; Lin, J.; Erhan, S.; Alander, J.; Gunstone, F. Nonfood Uses of Oils and Fats. *Lipid Handb. with CD-ROM, Third Ed.* **2007**, No. 2001, 591–635. https://doi.org/10.1201/9781420009675.ch9.

(11) Puccio Pietro. Pterygota alata - Monaco Nature Encyclopedia https://www.monaconatureencyclopedia.com/pterygota-alata-2/?lang=en (accessed May 24, 2021).

(12) Chancerel, L. *Flore Forestière Du Globe / Par Lucien Chancerel.*; 2011. https://doi.org/10.5962/bhl.title.45099.

(13) Jahan, N.; Parvin, M. S.; Das, N.; Islam, M. S.; Islam, M. E. Studies on the Antioxidant Activity of Ethanol Extract and Its Fractions from Pterygota Alata Leaves. *J. Acute Med.* **2014**, *4* (3), 103–108. https://doi.org/10.1016/j.jacme.2014.05.001.

(14) Jahan, N.; Parvin, M. S.; Khan, A.; Das, N.; Islam, M. S.; Islam, M. E. Evaluation of Free Radical Scavenging and Polyphenolic Contents of Bark of *Pterygota Alata* Roxb. *J. Sci. Res.* **2014**, *6* (3), 543–552. https://doi.org/10.3329/jsr.v6i3.17586.

(15) Dekker, M. Crude Fat Methods – Considerations. *Lab. Methods Serv. Comm.* **2014**.

(16) AOCS. AOCS Official Method Cd 3d-63. *American Oil Chemists' Society*. 2009.

(17) Nzikou, J. M.; Mvoula-Tsieri, M.; Matouba, E.; Ouamba, J. M.; Kapseu, C.; Parmentier, M.; Desobry, S. A Study on Gumbo Seed Grown in Congo Brazzaville for Its Food and Industrial Applications. *African J. Biotechnol.* **2006**, *5* (24), 2469–2475. https://doi.org/10.4314/ajb.v5i24.56046.

(18) Hainida, K. I. E.; Amin, I.; Normah, H.; Mohd.-Esa, N. Nutritional and Amino Acid Contents of Differently Treated Roselle (Hibiscus Sabdariffa L.) Seeds. *Food Chem.* **2008**, *111* (4), 906–911. https://doi.org/10.1016/j.foodchem.2008.04.070.

(19) Amoo, I. A.; Agunbiade, F. O. Some Nutrients and Anti-Nutrients Components of Pterygota Macrocarpa Seed Flour. *Electron. J. Environ. Agric. Food Chem.* **2010**, *9* (2), 293–300.

(20) Standard, C.; Oils, N. V. SECTION 2 . Codex Standards for Fats and Oils from Vegetable Sources. **2015**, 1–19.

(21) Aremu, M. O.; Ibrahim, H.; Bamidele, T. O. Physicochemical Characteristics of the Oils Extracted from Some Nigerian Plant Foods – A Review. *Chem. Process Eng. Res.* **2015**.

(22) Pearson, D. The Chemical Analysis of Foods.; Churchill Livingstone: Edinburgh, New York, 1981; pp 504–530.

(23) Ebong, U.; Christopher. A Comparison of Iodine Values of Some Co. 2019, pp 1–38.

(24) Zauro, S. A.; Abdullahi, M. T.; Aliyu, A.; Muhammad, A.; Abubakar, I.; Sani, Y. M. Production and Analysis of Soap Using Locally Available Raw-Materials. *Appl. Chem.* **2016**, *96* (7), 41479–41483.

(25) Jain, J. L.; Jain, N.; Jain, S. Fundamentals of Biochemistry. S. Chand. & Co. Ltd; S. Chand. & Co. Ltd.: New Delhi, India, 1995; p 115.