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Investigation on Fatty Acid Composition of Oil Extracted from *Carica papaya* L. Seed

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ABSTRACT: This study was aimed to extract oil from *Carica papaya* L. seeds by solvent extraction with petroleum ether. The extraction process was carried out for 36 hours and reddish yellow coloured oil (24.01%) was obtained. Esterification and saponification of extracted seed oil can be carried out by using different catalyst. But in this study the extracted oil was esterified with BF₃-methanolic solution and the percentages of fatty acids were estimated as the methyl esters of fatty acids by Gas Chromatography. The fatty acid compositions of *Carica papaya* L. seed oil were oleic (78.88%), palmitic (15.96%), stearic (4.71%) and arachidic (0.44%) acids. It was implied that the unsaturated fatty acid of the seed oil was more than the saturated one. On the basis of this result, the papaya seed oil can be considered as potential source of oleic acid and alternative source of vegetable oils.

KEYWORDS: Solvent extraction, Esterification, Saponification, Fatty Acid, Carica papaya L.

I. INTRODUCTION

Natural product chemistry is that branch of chemistry which deals with the isolation, identification, structure elucidation, and study of the chemical characteristics of chemical substances produced by living organisms [1]. A natural product is a chemical compound or substance produced by a living organism that is, found in nature [2]. In the broadest sense, natural products include any substance produced by life. Bennett defined natural products as being plant derived, occurring naturally, wild harvested or cultivated or cultivated in suit, sustainably harvested and pro-poor. These natural products could include an array of medicinal and aromatic plant species, oil seeds, flowers, teas, fruits and nuts and the availability of these plant species varies from country to country [3].

Fatty acid is a carboxylic acid with a long aliphatic tail, which is either saturated or unsaturated. Fatty acids are the major component of lipids, and the physical, chemical, and physiological properties of a lipid depend primarily on its fatty acid composition. Fats contain higher portion of saturated fatty acids component and oils contain higher portion of unsaturated fatty acids component [4-5]. Most of naturally occurring fatty acids have a chain of an even number of carbon atoms from 4 to 28. Extraction plant oils from seeds can be done in several ways like hot extraction, cold extraction, steam distillation, maceration, effleurage, cold pressing, solvent extraction, carbon dioxide extraction, pressure method etc. Hot extraction is used for some seeds having low percentage of oil. *Carica papaya* L., belonging to family Caricaceae, is commonly known as papaya in English, Papita in Hindi and Erandakarkati in Sanskrit. The plant is native to tropical America and was introduced to India in 16th century. The plant is recognised by its weak and usually unbranched soft stem yielding copious white latex and crowded by a terminal cluster of large and long stalked leaves, is rapidly growing and can grow up to 20m tall [4-5].



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Figure 1. Carica papaya L. fruit (left), seed (middle) and dry seed (right)

Both leaf and fruit of the Carica papaya L. possess carotenoids namely β-carotene, lycopene, anthraquinones glycoside, as compared to matured leaves and hence possess medicinal properties like anti-inflammatory hypoglycaemic, antifertility, abortifacient, hepatoprotective, wound healing, recently its antihypertensive and antitumor activities have also been established [4-5]. The papaya seeds are small, black, round and covered with gelatinous aril (Figure 1). There are many health benefits of the seeds. Beside this it has been proven to be a good source of oil (25.6%) that may be useful for medicinal, biofuel, and industrial purposes [6]. The physicochemical properties of oils determine their quality and whether they are suitable for consumption [7-8]. The papaya seed oil possessed a reddish yellow colour. Oleic, palmitic, linoleic and stearic acids are the most abundant fatty acids found in the papaya seed oil [9-10]. The papaya seed oil is used in biodiesel production. Biodiesel is a nontoxic, biodegradable and renewable fuel consisting of mono-alkyl esters of long fatty acid chains. Recently, the utilization of biodiesel as a promising substitute to petroleum diesel has grown considerably. Many researchers have tried a variety of plant oils to develop new possible feedstock sources [11]. Fatty acid composition of seed oil is used to calculate the biodiesel properties. Papaya is important for its fruit and it is cultivated mainly for eating purpose. The seeds of papaya fruits are generally discarded. Hence our main focus on the use of papaya seeds. This research was aimed to extract oil from Carica papaya L. and to investigate the fatty acid composition in extracted oil. Beside this, additional aim was to assess the activity of BF₃-methanolic solution as catalyst. However, in order to make a more efficient use of papaya, it is worth investigating the use of the seeds as a source of oil.

II. LITERATURE REVIEW

The extraction and use of vegetable oils has for centuries played an important role in the manufacture of a large number of industrial products and food items. *Puangsri et al.* (2005) studied the physico-chemical properties of oil from *Carica papaya* seed. The oil was extracted with petroleum ether and aqueous-enzymatic methods. The results showed that the major fatty acids in the extracted oil were oleic (76.9%), followed by palmitic (13.4%), stearic (4.6%) and linoleic (3.2%) acids with no significant difference between enzyme- and solvent-extracted oils. The oil was found to contain trace quantities of lauric, myristic, palmitoleic, margaric, linolenic, arachidic and eicosenoic acids [10]. *Singh* (1990) reported that the oil from the seeds of other papaya varieties contained lauric (0.4%), myristic (0.04%), palmitic (16.2%), stearic (5.0%), arachidic (0.9%), behenic (1.6%), palmitoleic (0.08%), oleic (74.3%) and linoleic (0.4%) acids [12].

Li et al. (2015) studied the suitability of Ultrasound-Microwave synergistic Extraction (UMAE) for the recovery of papaya seed oil as compared to Ultrasound-Assisted Extraction (UAE). The efficiency of these two methods was assessed by comparing the physicochemical properties and oxidative stability of papaya seed oil. Analysis of fatty acid composition revealed that the predominant fatty acids in papaya seed oil were oleic (18:1, 72.60%), palmitic (16:0, 18.00%), linoleic acid (18:2, 5.80%) and stearic (18:0, 3.60%) [13]. *Foluso and Tolulope* (2014) studied methanolysis



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of *Carica papaya* L. seed oil for the production of Biodiesel. The oil was transesterified by two-stage catalysis with oil to methanol mole ratio of 1:9. The biodiesel produced was subjected to standard fuel tests. The mean percentage oil yield was $31.18 \pm 0.1\%$. The result of the oil content of the C. papaya indicates that the seed has relatively high oil content and is considerable for commercial or industrial applications [14].

Badami and Daulatabad (2006) reported the fatty acid composition of *Carica papaya* L. seed oil to be lauric acid (0.4%); myristic acid (0.4%); palmitic acid (16.2%); stearic acid (5.0%); arachidic acid (0.9%), behenic acid (1.6%); hexadecenoic acid (0.8%); oleic acid (74.3%); linoleic acid (0.4%) [15]. *Samaram et al.* (2013) studied to evaluate the suitability of ultrasound-assisted extraction (UAE) for the recovery of oil from papaya seed as compared to conventional extraction techniques (Soxhlet extraction and solvent extraction) [16].

Robert et al. (2011) investigated the physicochemical characteristics, fatty acid, tocopherol, and carotenoid composition of a crude oil extracted from papaya (*Carica papaya* L.) seeds. The oil yield from the seeds was 29.16%. The data obtained for the analytical indexes were in agreement with those of other edible oils. The oil obtained had high oxidation resistance (77.97 hours). The major fatty acids in total lipid were oleic (71.30%), palmitic (16.16%), linoleic (6.06%), and stearic (4.73%) acid. The α and δ -tocopherol were the predominant tocopherols with 51.85 and 18.89 mg kg⁻¹, respectively. The β -cryptoxanthin (4.29 mg kg⁻¹) and β -carotene (2.76 mg kg⁻¹) were the carotenoids quantified, and the content of total phenolic compounds was 957.60 mg kg⁻¹ [17].

III.MATERIALS AND METHODS

The sample *Carica papaya* L. seeds were collected from Dhaka, Bangladesh. Then the seeds were washed with distilled water then dried in sunlight for 7 days. The dried seeds were grinded by mortar to make powder and it used for the extraction of oil. Analytical grade chemicals and reagents namely Acetone (Fischer Scientific), Petroleum ether (Merck, Germany), n-hexane (Merck, Germany), Methanol (Merck, Germany), Hydrochloric acid (Sigma Chemical Co. USA), Boron trifluoride 12% (1.5 M) in Methanol (Acros Organics, USA), NaOH pellets (Loba Chemie Pvt Ltd., India) and Anhydrous sodium sulphate (Loba, Chemie Pvt Ltd., India)were used for all the analyses.

Extraction of oil from seed of Carica papaya L.

The oil was extracted from *Carica papaya* L. by soxhlet extractor. In this method the dried sample was placed in a packet of markin cloth, a porous cellulose thimble. The thimble was placed in an extraction chamber, suspended above a round bottom flask containing the solvent (petroleum ether) and below a condenser. The flask was heated by heating mental and the solvent evaporates and moves up into the condenser where it was converted into a liquid that trickles into the extraction chamber containing the sample. The extraction chamber was designed so that when the solvent surrounding the sample exceeds a certain level it overflows and trickles back down into the boiling flask. This process was continued for 36 hours. At the end of the extraction process, the round bottom flask containing the solvent and oil is removed and the mixture containing petroleum ether and oil was taken in a dry pear shaped flask. The mixture of the seed oils and petroleum ether was separated using rotary evaporator. Then the oil was stored at 0 to 4 °C. The content of the oil in the seed was calculated to evaluate the percentage of oil in the seed by following equation.

 $Percentage of oil content = \frac{Weight of total oil \times 100}{Weight of total seeds taken}$

Determination of fatty acid composition of Carica Papaya L. seed oil

The oil extracted from *Carica papaya* L. seeds were esterified first and then analysed for the composition of fatty acids by the gas chromatography with flame ionization detector. Approximately 400 mg of seed oil extracted from seed sample was taken in a pear shaped flask and 3.0 mL of n-hexane was added to the oil. Then the mixture was evaporated with a rotary evaporator to dryness. 10.0 ml of 0.5 M methanolic NaOH was added to the pear shaped flask. The



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mixture was ultrasonicated for 5 minutes and then refluxed on boiling water at about 96°C for an hour. The mixture was evaporated with a rotary evaporator to dryness and 4.0 ml of water was added to it. The pH of the solution was adjusted to 4.5 (just acidic) with 0.2M HCl in which the blue litmus paper was turned to red. The mixture was shaken vigorously and then extracted with n-hexane. The organic layer was collected. The hexane part was made free from water by adding anhydrous sodium sulphate. The solution was filtered, evaporated to dryness and 10 ml of boron trifluoride-methanol (BF₃-MeOH) complex was added. The mixture was refluxed on a boiling water bath for an hour. The mixture was then evaporated with a rotary evaporator to nearly dryness and finally 2.0 ml of hexane was added and filtered through a pasture pipette containing cotton filter and anhydrous sodium sulphate on the cotton filter. The filtrate was concentrated to 1 mL and was taken in a GC vial. The vial was wrapped with aluminium foil and parafilm for protection of solution from air and moisture and then labelled the vial. The meniscus of the solutions was marked with permanent ink. These solutions were stored in the freezer of a refrigerator and sent for analysing by GC-FID to find out the fatty acid composition of seed oil at department of Chemistry, Dhaka University, Dhaka, Bangladesh.

Analysis of seed oils for Fatty acids by GC-FID

A mixture of methyl esters of twelve fatty acids (Capric acid, Caprilic acid, Lauric acid, Myristic acid, Palmotelic acid, Palmitic acid, Linolic acid, Oleic acid, Stearic acid, Arachidic acid, Behenic acid and Lignoceric acid) standard was used as the reference. The identification of fatty acids were done by comparing retention times of the samples with that of the corresponding fatty acid standards in the chromatograms. Quantification was carried out by the accounting the areas of individual fatty acids and the results were expressed in terms of the relative percentages. The amount of individual fatty acids present in the seed oil was calculated by using the following equation and the retention time, name of fatty acid and area under the peak were taken from sample chromatogram.

 $Amount of individual fatty acid (\%) = \frac{Area of individual fatty acid \times 100}{Total area}$

IV. RESULTS AND DISCUSSION

The oil was extracted by hot extraction method by using petroleum ether as solvent and soxhlet as extractor. The yield of the oil from Carica papaya L. seed was 24.01%. The result of the oil content of the Carica papaya L. indicates that the seed has relatively low oil content and is considerable for commercial or industrial applications. According to GC analysis the fatty acid composition in the extracted seed oil were oleic (78.88%), palmitic (15.96%), stearic (4.71%) and arachidic (0.44%) acids (**Table 1**).

Serial No.	Retention time	Name of fatty acid	Area of fatty acid	Relative %of fatty acid
1	13.990	Palmitic acid	3160880	15.96
2	16.809	Oleic acid	15625472	78.88
3	16.938	Stearic acid	933948	4.71
4	19.412	Arachidic acid	87301	0.44
		Total area	19808658	

Table 1: Determination of relative percentage of fatty acid in Carica Papaya L. seed oil

The oil from papaya seeds could be considered similar to other vegetable oils containing large amounts of unsaturated fatty acids with 18 carbon atoms. Analysis of fatty acid composition revealed that the predominant fatty acids in papaya seed oil were oleic (18:1, 70.5%-74.7%), palmitic (16:0, 14.9%-17.9%), stearic (18:0, 4.50%-5.25%), and linoleic acid (18:2, 3.63%-4.6%) [10,18]. The high-oleic acid content of papaya seed oil (78.88%) was also comparable with commercially available high-oleic vegetable oils such as sunflower (>80%), canola (75%) and safflower (77%). On the



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basis of these results, papaya seed oil can be considered as potential oleic oil and, hence, may be viewed as a healthy alternative to many other vegetable oils [10,18]. Fatty acid methyl esters (FAME) from *Carica papaya* L. (papaya) oil were investigated as a potential biodiesel fuel. The fatty acid composition can also be used to evaluate the stability and the nutritional quality of fats and oils.



Figure 2. Sample Gas Chromatogram of the fatty acid methyl ester mixture

V. CONCLUSIONS

This study revealed that papaya seed contains lower content of oil. BF_3 -Methanol catalysed esterification of papaya seed oil was a potential source of unsaturated fatty acid like oleic acid and the percentages of yield was almost similar to other methods. The high proportion of unsaturated fatty acids would make the oil an acceptable substitute for other highly unsaturated oils. However, before being considered adequate for food applications, toxicological studies need to be carried out to ascertain whether or not this oil is safe. Information provided by the present study is of great importance for further chemical investigations of papaya seed oil and industrial utilization of the papaya seeds as a raw material to foods and cosmetics.



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