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Characterisation of the Oil Extracted from the Seed of Pepper Fruit (*Dennettia Tripetala)*

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Abstract

The oil from the dehulled seed of *Dennettia tripetala* (pepper fruit) was extracted and analyzed for its physicochemical properties (refractive index, specific gravity, saponification value, peroxide value, iodine value, acid value, and free fatty acid) using standard analytical procedures. The extracted oil appeared as a brown liquid at room temperature, with a percentage yield of 8.31 %. It exhibited a specific gravity of 0.914 g/cm³, 1.418 for refractive index, 48.96% for acid value, and a free fatty acid content of 21.9%. Additional parameters included saponification value of 188.10 mg KOH/g, an iodine value of 30.28 g I_2/100 g, and a peroxide value of 24.40 meq/kg. Based on its iodine value (below 115 g I_2/100 g), it is classified as a non-drying oil. The peroxide value (less than 30 meq/kg) suggests that it is relatively stable and less susceptible to rancidity. The high saponification value indicates potential for use in soap and shampoo production. The data from the fatty acid profile revealed that the oil is primarily composed of saturated fatty acids (SFAs), with a percentage composition of 64.6 %, followed by monounsaturated fatty acids (MUFAs) at 17.94% and polyunsaturated fatty acids (PUFAs) at 14.29%. The dominant SFAs were stearic acid (24.39%), margaric acid (20.76%), and palmitoleic acid (4.06%). These results suggest that *Dennettia tripetala* seed oil (PFSO) holds promise as a valuable source of oil for both domestic and industrial applications, provided it is effectively harnessed.

Keywords: *Dennettia tripetala*; Fatty Acid Methyl Esters (FAMEs); Pepper Fruit Seed Oil (PFSO); Saponification Value; Stearic Acid

Introduction

The role of vegetable oils cannot be overemphasised in the human diet as sources of energy, fat-soluble vitamins, and essential fatty acids [1]. These fatty acids have numerous importance and could perform functions as maintaining healthy bones, supporting liver function, strengthening the immune system, and ensuring the structural integrity of cells. The oils are extracted from seeds, kernels, and nuts of various plants primarily through mechanical pressing or using solvent extraction. Vegetable oils are largely composed of triacylglycerols [2].

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Oils derived from plants serve both edible and industrial purposes, including use as lubricants, raw materials for soap and cosmetics, insulating agents, and in the production of biodiesel [3-5].

Dennettia tripetala, commonly known as pepper fruit, belongs to the *Annonaceae* family and is native to the tropical rainforests and parts of the savannah, particularly in western Cameroon, Ivory Coast, and southern Nigeria [6]. The fruit is known for its pungent flavor and is locally referred to as 'mmịmị' (Igbo), 'nkarika' (Ibibio and Efik), 'imako' (Urhobo), and 'ata igbere' (Yoruba) [7]. Its ripe fruits are reddish, while the unripe ones are green, both having a spicy, peppery taste and a distinct aromatic fragrance due to essential oils or oleoresins [8].

The fruit is consumed either ripe or unripe and is valued for its unique flavor, often described as citrusy, fruity, and mildly spicy, similar to tomatoes or peaches in complexity [9]. It is mildly used as a stimulant [6] and often used as a seasoning or condiment [10].

Lipids are essential nutrients, consisting primarily of fatty acids (FAs), which are characterized by the presence of single and double bonds. These include saturated fatty acids (SFAs, characterized by the presence of single bonds), mono-unsaturated fatty acids (MUFAs, having one double bond), and polyunsaturated fatty acids (PUFAs, having more than two double bonds). PUFAs are further classified as cis or trans based on the special configuration of the bonds, and as n-3 or n-6 depending on the position of the first double bond from the methyl end. The human body cannot synthesize PUFAs because it lacks the enzymes responsible for their synthesis.

The fatty acid composition of vegetable oils comprises a mixture of saturated and unsaturated fatty acids, the latter being further divided into MUFAs and PUFAs based on the number of double bonds.

However, the fatty acid composition of vegetable oils varies by plant source, allowing their health effects to be evaluated based on the distinct impacts of individual fatty acids on human health and the risk of serious diseases. Despite the ample nutritional and medicinal importance of the *Dennettia tripetala*, not much is known about the physicochemical properties of the plant. Therefore, this study focused on examining the physicochemical properties and fatty acid profile of oil extracted from *Dennettia tripetala* seeds. This research will help bridge the gap in the existing literature on the physicochemical properties and fatty acid composition of the oil from *Dennettia tripetala* seed, and also expand the scope of knowledge on the utilization and nutritional qualities of *Dennettia tripetala* oil.

Materials and Methods

Collection and Preparation of Plant Material

The fruits of *Dennettia tripetala* used in this study were bought from Sabo Market in Ikorodu North Local Government Area, Lagos State, Nigeria. Then, the samples were identified by a taxonomist at the herbarium, of the Department of Botany, University of Lagos; and assigned an herbarium number LUH7001A. The Samples were

transported to the laboratory, Department of Chemical Sciences, Lagos State University of Science and Technology, Ikorodu, where they were handpicked to removed spoilt and other unwanted materials, washed, and the fruits were peeled and the seeds dehulled using a knife. They were then air-dried for seven days, after which they were pulverized with the aid of a mortar and pestle before being stored in an air-tight polythene bags before use.

A B



Figure 1: A sectional view of the ripped pepper fruit (A) and dehulled pepper fruit seeds (B) of *Dennettia tripetala*

Extraction of the Oil

Pepper fruit seed oil (PFSO) was extracted from pulverized seed samples using the Soxhlet extraction method with *n*-hexane (boiling range: 60–80 °C) as the extracting solvent, following standard procedures [11]. A 100 g of the pulverised seed sample was enclosed in a Whatman filter paper and placed in the Soxhlet apparatus, with *n*-hexane serving as the extraction solvent. The extraction was carried out continuously for four hours after which the solvent was recovered through simple distillation. The resulting crude oil was oven-dried at 60 °C for 30 minutes and then cooled in a desiccator and weighed. This drying, cooling, and weighing process was repeated until a constant weight was achieved. The oil yield (%) was calculated using Equation (1).

$$\% \text{ Oil Yield} = \frac{\text{weight of extracted oil}}{\text{Weight of sample}} \times 100 \quad (1)$$

Physicochemical Characteristics of the Oil

The Physicochemical properties of the oils were determined using standard methods [11]. It was employed for the determination of refractive index, specific gravity, saponification value, peroxide value, iodine value, acid value, and free fatty acid of the oil.

Fatty Acids Profile of Oil Extract

Quantitative determinations of Fatty Acid Methyl Esters (FAMEs) were conducted using a Shimadzu GC-2010 gas chromatograph

(Shimadzu Corporation, Tokyo, Japan) with a flame ionization detector (FID) and capillary column HP-88 Agilent Technologies (100 m × 0.25 mm) with a stationary phase (88% cyanopropyl, aryl-polysiloxane) with the thickness of 0.2 μm. The injection volume was 1.0 μL, the temperature of the injection port was 250 ºC with split ratio of 1:100, and nitrogen was used as carrier gas. The temperature program consisted of 80 ºC for 5 min, 200 °C for 30 min, and 250 ºC for 15 min. Identification of FAMEs was performed by comparing their retention times with those of reference standards (mixture FAME Mix, SUPELCO, which included 37 FAMEs). For quantification of FAMEs, methyl-undecanoate (Sigma Aldrich Chemical Co., St. Louis, MO, USA) was used as the internal standard. The results of FAs were expressed as percentages of total FAMEs [12-13].

Statistical Analysis

Three determinations were carried out for each analysis. The mean values and standard deviation were calculated using SPSS statistical software.

Results and Discussion

The quality of oils and their suitability for consumption are determined by their physicochemical properties [14]. Table 1 reveals the physical characteristics of Pepper Fruit Seed Oil (PFSO). The PFSO is a brown liquid at room temperature (25 ℃) with a pleasant and refreshing smell. The percentage oil yield for PFSO was 8.31±0.0.3 %. The results of the oil yields indicate that the extracted seed oil exhibited a very low oil yield, which makes it unsuitable for consideration in biodiesel production. The oil yield of PFSO is lower than that obtained in papaya seeds 30.7 % [15] and 31.2 % [16], soya bean 14.51 %, groundnut oil 10.54 %, and maize 6.63 % as reported by [17].

The oil yield range of 40-60 % for African pear seed was reported by [18]. The specific gravity and refractive index of 0.912 and 1.418 for the PFSO obtained are similar to most edible oils [19]. The refractive index of PFSO is in the range as that obtained for Soya bean oil (1.4662), groundnut (1.4622), and maize (1.4632) [17].

Table 1: Physical characteristics of Pepper Fruit Seeds Oil (PFSO)

Parameter	**Values**
Oil yield (%)	8.31 ± 0.0.3
Colour	Brown
Odour	Pleasant and refreshing
Specific gravity	0.914 ± 0.03 g/cm^3
Refractive index	1.418 ± 0.20

Table 2.0 shows the results of the chemical parameters of the PFSO investigated.

Acid value is a direct measure of the percentage of free fatty acid in a given amount of oil. It is a

measure of the extent to which triglycerides in the oil have been decomposed by lipase action into free fatty acids. An acid value depends on the degree of rancidity, which is used as an index of freshness [20-21]. The acid value obtained from PFSO was 48.96 mg/KOH/g, which was lower than 57.74±0.47 mg KOH/g as reported in earlier studies [22], and higher than that of soya bean and groundnut (19.21 mg KOH/g, 4.63 mg KOH/g) and lower than maize oil 65.50 mg KOH/g [17]. The acidity of the oil makes it unsuitable for human use, due to the value of the acid being higher than the maximum acceptable value of 4 mg KOH/g as recommended by the Codex Alimentarius Commission 1999 [23-24]. The oil's free fatty acid (FFA) content was 21.9% which is higher than that of soya bean oil and groundnut oil (6.85 mg NaOH/g, 1.65 mg NaOH/g) except that of maize (23.35 mg NaOH/g), which is slightly higher, indicating a significant hydrolytic activity likely due to the presence of moisture [17]. Since the FFA level surpasses 5%, the oil is also considered inappropriate for use as a biodiesel feedstock [25].

Table 2: Chemical characteristics of pepper fruit seeds oil (PFSO)

Parameter	**Unit**	**Value obtained**
Acid value	mg KOH/g	48.96±1.56
Free fatty acid	%	21.9 ± 2.20
Saponification value	mg KOH/g	188.10± 11.80
Iodine value	gI_2/100g	30.28±0.84
Peroxide value	meq/kg	28.40±3.12

The data are the mean ±standard deviation (SD) of the replicated determinations

A saponification value of 188.10 ± 11.80 mg KOH/g was recorded for *Dennettia tripetala* seed oil (PFSO) in this study, which is slightly lower than the 191.36 ± 10.17 mg KOH/g reported by [22] for pepper fruit oil and also lower than that of soya bean oil (228.19), groundnut (227.49), and maize (211.37 mg KOH/g) as reported by [17]. According to [26], a saponification value around 200 mg KOH/g suggests a high content of low-molecular-weight fatty acids. Such a value indicates that the oil may have potential applications in soap production, cosmetics, industrial uses, and thermal stabilization of polyvinyl chloride (PVC). It also reflects that the presence of essential fatty acids is beneficial to human health [27]. The saponification value also offers insight into the structural composition of fatty acids [28].

However, the relatively low saponification value observed in this study suggests that PFSO has limited potential for both local and industrial-scale production, particularly in the

manufacturing of soap. Instead, it may be more suitable as a food ingredient, owing to its medicinal, cleansing, and flavor-enhancing properties.

The iodine value, which indicates the degree of unsaturation and thus affects the oil's shelf life, was measured at 30.28 g I_2/100 g, which is less than 73.02 I_2/100 g, 38.65 I_2/100 g, and 47.25 I_2/100 g for soybean, groundnut, and maize oils. This value suggests that PFSO is a drying oil and may be suitable for certain industrial applications [17]. Iodine value also influences the oil's susceptibility to rancidity, as peroxidation typically begins at sites of unsaturation [29]. The iodine value of PFSO is lower than that of *Carica papaya* seed oil (72.78 g/100g) reported by [30] and lower than 59.6 g/100g found in African pear seeds [18].

Compared to other oils, PFSO's saponification value is also lower than the 193.5 mg KOH/g reported for *Carica papaya* seed oil [31] and 213.54 mg KOH/g for African pear seed oil [32]. The peroxide value of PFSO was found to be 28.40 ± 3.12 Meq/kg. This value indicates the level of primary oxidation, which is influenced by the degree of unsaturation in the oil. Oils rich in double bonds are more prone to autoxidation. Fresh oils typically have peroxide values below 10 Meq/kg, while values between 30 and 40 Meq/kg can result in a noticeable rancid flavor. Thus, the peroxide value of PFSO suggests that the oil is nearing the threshold where rancidity becomes perceptible and may not be considered fresh.

Table 3: Fatty acid profile of pepper fruit seed oil (PFSO)

Fatty acid	**Class of fatty acid**	**Structure**	**Retention time (min)**	**Concentration %**
Caprylic acid	Saturated	C8:0	11.76	2.54
Capric acid	Saturated	C10:0	13.73	2.08
Undecenoic acid	Saturated	C11:0	15.54	0.25
Tridecanoic acid	Saturated	C13:0	18.67	2.39
Myristic acid	Saturated	C14:0	18.81	1.51
Myristoleic acid	Monounsaturated	C14:1	20.07	4.76
Pentadecanoic acid	Saturated	C15:0	20.23	4.37
Palmitic acid	Saturated	C16:0	21.34	6.24
Palmitoleic acid	Monounsaturated	C16:1	21.61	4.06
Heptadecanoic acid	Saturated	C17:0	22.76	20.76

Stearic acid	Saturated	C18:0	23.08	24.39
Elaidic acid	Monounsaturated	C18:1Trans	24.08	4.73
Linolelaidic acid	Polyunsaturated	C18:2(n-6)	24.19	8.77
Oleic aid	Monounsaturated	C18:1 Cis	24.45	4.39
Linoleic acid	Polyunsaturated	C18::2	27.79	1.35
Arachidic acid	Polyunsaturated	C20;0	28.39	3.40
Linolenic acid	Polyunsaturated	C18:3	31.18	0.27
Behenic acid	Saturated	C22:0	34.88	0.07

Mean values of duplicate determinations

Total saturated fatty acid (SFA) = 64.6%, Total Monounsaturated fatty acid (MUFA) = 17.94%
Total Polyunsaturated fatty acid (PUFA) =14.29%

The fatty acid composition of vegetable oils is a main feature of their description and identification. It is also used as an indication of the purity and quality of the oil because the type and quality of each fatty acid vary from one vegetable oil to another. Table 3.0 shows the fatty acid profile of PFSO extract. The major fatty acids detected in higher levels in PFSO were Stearic acid (24.39 %), Heptadecanoic or Margaric acid (20.76 %), Linolelaidic acid (8.77 %), Palmitic acid 6.24 %). Myristoleic acid (4.76%), Elaidic acid (4.73 %), Oleic acid (4.39, Pentadecanoic acid (4.37 %) Palmitoleic acid (4.06 %) Arachidic acid (3.39 %), Caprylic acid (2.54 %), Tridecanoic acid (2.39 %), Myristic acid (1.51 %) and Linoleic acid (1.31 %) respectively. Other fatty acids detected in trace amount in PFSO include; Undecanoic acid (0.25 %), Linolenic acid (0.26 %) and Behenic acid (0.069 %) respectively.

Table 3.0 also depicts fatty acids distribution according to saturation and unsaturation of components (%). The total unsaturated fatty acid was higher than the saturated ones. It has been established that relative to carbohydrates, the saturated fatty acids elevate serum cholesterol while polyunsaturated fatty acids (PUFA) lower serum cholesterol [33]. Margaric acid is classified as an odd-chain fatty acid, which occurs as a trace component of the fat and milk fat of ruminants. Margaric acid (Heptadecanoic acid, C17) may be used as a reference during the analysis of medium-chain fatty acids from biomaterials such as seed oils from plant and also, animal oils.

Oleic acid is an 18-carbon MUFA, essential in human nutrition, and helps to reduce triglycerides, LDL cholesterol, total cholesterol, and glycemic index [34]. The increase in stability over oxidation of vegetable oil is attributed to oleic acid [35]. Stearic acid, a long 18-carbon straight-chain SFA, has been found to bind and

plasticize composites [36], human serum albumin [37], and α-helical sites in biomolecules [38], indicating the usefulness of pepper fruit seed oil.

Fats and oils rich in stearic acid are more abundant in animal fat (up to 30%) than in vegetable fat (typically <5%). The important exceptions are foods like cocoa butter (4 %) and shea butter, where the stearic acid content (as a triglyceride) is 28–45 % [39]. Stearic acid is primarily used industrially in the production of detergents, soaps, and cosmetics such as shampoo and shaving cream products. Soaps are not made directly from stearic acid, but indirectly by saponification of triglycerides consisting of stearic acid esters. Figure 2.0 shows the chromatogram of identified Saturated, Monounsaturated, and Polyunsaturated fatty acids analyzed by GC with FID detector.

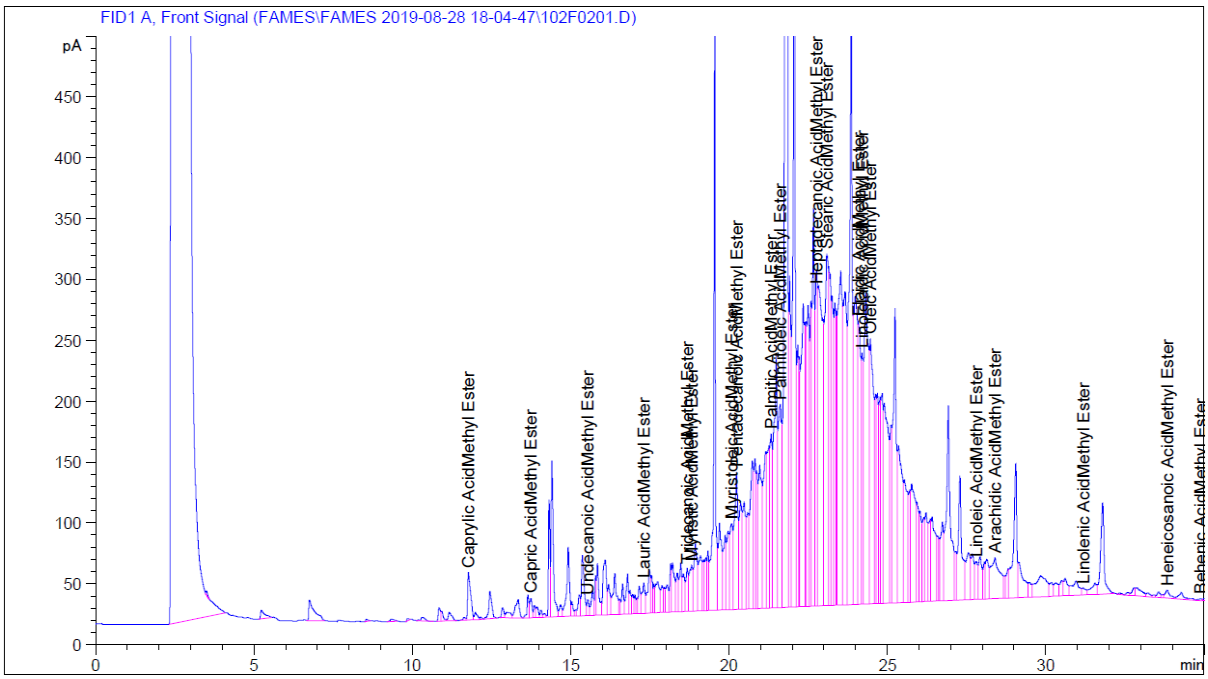


Fig. 2: An overlaid GC-MS chromatogram of PFSO in mg/kg

However, the total compositions of monounsaturated fatty acids (MUFA), saturated fatty acids (SFA), and polyunsaturated fatty acids (PUFA) in PPSO were 17.94 %, 64.6 %, and 14.29% respectively. Therefore, the predominance of SFA over MUFA and PUFA reveals that PFSO could have industrial applications such as soap production, lubricants, and ointments.

Conclusion

In this study, the extraction and characterization of oil from *Dennettia tripetala* have been determined. The results revealed that PFSO could exhibit both domestic and industrial applications based on the oil parameters, if properly harnessed.

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