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Physico-chemical and Fatty Acid Composition of Oil Extracted by Acetone Solvent from Alligator Pepper (*Aframomum Melegueta*)

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Abstract To achieve a suitable concentration of the active ingredients contained in the plant or seed samples and that their action can be more effective, it is necessary to perform several procedures through which the active ingredients are extracted with the appropriate solvents, These are selected according to the solubility and stability of the beneficial substances. The acetone (a polar solvent) extracting method by soaking where adopted for the extraction of oil from alligator pepper seeds *(Aframomum Melegueta*) for adequate extraction. Since the water content easily soluble in acetone after decantation, a freeze drying method at -20 ℃ and vacuum pressure of 1.034 mBar was used to remove both the acetone and the water content. This prevented the effect of heat during drying on the essential oil present in the oil extracted from the sample. This method leaved the oil in its natural state for its Physico-chemical and fatty acid composition. The values obtained for the chemical composition were. Acid value (mg/KOH/g) 34.22±0.2, Iodine Value (wijs) 29.44±0.2, Saponification Value (Mg/KoH/g) 145.86±0.3 and Peroxide Value (mg/kg) 30.90±0.2 respectively, 1.4605±0.1 and 0.9034±0.1 were the values obtained for the physical parameters (Refractive Index and Specific gravity)respectively. The fatty acid Composition obtained were Palmitic Acid(16:0) 17.269, Palmitoleic Acid(16:1)0.156, Margaric Acid(17:0)0.083, Stearic Acid(18:0)2.791, Oleic Acid(18:1)64.944, Linoleic Acid(18:2)13.885, Linolenic Acid(18:3)0.320, Arachidic Acid(20:0)0.351 and Behenic Acid(22:0)0.200 respectively. This result indicated that the values obtained were within the range of convectional oils consumed and good industrially because the level of the unsaturated fatty acids in the oil sample.

Keywords Alligator pepper, fatty acid, Essential oil, freeze drying

Introduction

Seeds of plants are a good source of food for animals, including humans, because they contain nutrients necessary for plant's initial growth, including many healthy fats, such as omega fats. In fact, the majority of foods consumed by human beings are seed-based foods. Edible seeds include cereals, legumes and nuts [1]. Oil seeds are often pressed to produce rich oils – sunflower, flaxseed, rapeseed, sesame. Seeds are typically high in unsaturated fats and, in moderation, are considered a healthy food, although not all seeds are edible [2]. Vegetables fats and oil are lipid materials derived from plants. Physically, oils are liquids while fats are solids at room temperature [1].

Chemically, both fats and oils are composed of triglycerides, as contrasted fatty acids while oils are up to mostly unsaturated fatty acids. Although many different parts may yield oil, in commercial practice, oil is extracted primarily from seeds of plants which grow in many different parts of the world. The chief importance of vegetables oils lies in their food value. Oils and fats are vital ingredients of a balanced diet because they supply body warmth

and build physical energy, and in recent years, vegetable sources have accounted for about three-fifths of the world's consumption of fats and oils, the rest coming from animal and marine fats.

These edible oils are consumed in various ways; the liquid form are used for cooking in warmer climate, in the Western world, they are eaten in spreadable form, they are applicable in food industry in the preparation of fats ands oil, the seeds of several of these plants have high protein content, the residue (cake) after the oil has been extracted in many cases provides animal fodder [3].

There are wide varieties of seeds and nuts that can produce oils for food nutraceuticals, skincare products, aromatherapies, fuels and industrial lubricant. Also, some of these plant derived oils can be used to make soap, body and hair oils, detergents and paint. A wide range of oilseeds and other oil producing plants are high quality feedstock for biodiesel. Oilseed processing expands the use of crops and also brings value to waste products. Vegetables oils derived from plant seeds have playing vital roles to provide comfort in human lives in various aspects. Outside the realm of food manufacture, vegetable oils feature in a variety of industrial uses ranging from the manufacture of soap to the production of paints, varnishes, lubricants and plastics. For instance, they have been used for illumination and lubricating purpose, production of detergents and cosmetics and for coatings and paint for many centuries before an abundant and cheap supply of mineral oil became available [4].

In the last few decades, there been growing concerns over vegetable oils as source of material in preference to petroleum or mineral oil. The main factor for this concern is due to environmental issues that regard mineral oil as major contributor of volatile organic compounds (VOCs) which themselves are responsible for most of our present recalcitrant pollution.

Aframomum is propagated by seed or by rhizome division, and cultivated as a mono crop. Division of rhizome is the preferred method. The plant could be successfully cultivated under cocoa plantations in warm humid areas and under partial shade. Pruning of the old dry leaves is crucial for better production of the plant. Seeds normally germinate between 7 to 12 days after nursing. Seedlings can be transplanted to the field after six to eight weeks. The first crop can be taken off nine to eleven months after planting. Under good management profitable yield can be obtained up to the tenth year of cultivation. Aframomum is harvested between February and June, when the pod changes colour from green to red [5].

No serious diseases or pest are known for Alligator pepper (*Aframomum melegueta*). The rust puccinia aframomi has been observed on Aframomum leaves in Ethiopia. Fruits are mostly harvested from the wild. The seeds are best harvested when the fruits are red and mature.The yield can be as much as 500kg of dried fruits per ha (without fertilizers) [5].

The essential oil from Alligator pepper consists of two sesquiterpenes, humelene and caryphyllene and the oxides of these. It has an exotic tropical scent and flavor and is used for the production of beer, wine and spirits, and the flavouring of vinegar. It is used in the Surinam cuisine to flavor dishes such as vegetables (Okra and Tomatoes recipes), soup (Lenti and Chicken) and fish recipes. The rhizome of the plant is used medicinally and is also an important part of the diet of western low land gorillas in Africa. Melegueta pepper is a spice native to tropical West Africa. In the 13th century, trades from West Africa carried the spice across the desert to sell in the high value of the product, the secrecy of the country of its origin. Europe acquired a taste for spice as a substitute for real pepper [6]. Therefore the aim of this work is to evaluate physico-chemical (acid value, iodine value ,peroxide value and the saponification value like refractive index and specific gravity), and fatty acid composition of oil with its essential oil extracted from alligator pepper using team distillation method for industrial application and its suitability for home consumption. The results obtained also were correlated with the results obtained by any other methods and some convectional oil whether they are good for human consumption and industrial application.

Materials and Methods

Collections of the sample: Mature alligator pepper was purchased from Obashoto market, Owo in Owo local government area of Ondo state, Nigeria. Proper sampling was done to remove the immature pod from the matured ones right in the market before the purchase.

Preparation of the sample: The matured alligator pepper was sundried for 3-4hrs for two days with the pod. After the complete dryness, the pod was removed and the seeds separated manually. The particles were blown off in order to have a clean seeds. The seeds were milled mechanically by small milling machine in the market and stored in a clean bottle for extraction process.

Figure 1: a) Whole Alligator pepper, b) Alligator pepper seed, c)Alligator pepper oil

Extraction Procedure: The extraction was done by acetone soaking method. Acetone is a polar solvent and had boiling point of 56 ℃ and melting point of -95 ℃. About 100 g of the grounded fresh sample was soaked in 1dm^3 acetone for 24 hrs inside a tightly covered sample jar with occasional shaking. This process was done on four different portions for a required quantity of the oil. The mixture of the oil, acetone and the water content were filtered using a muslin cloth, allowed the suspended particles to precipitate inside a tight jar bottle, a clear portion was decanted inside 250 ml beaker for freeze drying. The mixture was frozen at -20 ℃, the oil and the water froze remaining acetone in liquid form which can be easily decanted and the water sublimed at 1.034mbar pressure, living the oil in a solid state. The oil was carefully collected and stored below room temperature until it is required for analysis.

Characterization of the Extracted Oil

In evaluating the quality of the extracted oil with its essential oil for the refractive index, Specfic gravity, saponification values, acid value, iodine value, free fatty acid value and peroxide value of the oil were determined using AOAC [7].

Fatty Acid Methyl Ester Analysis (Gas Chromatographic Method): 50 mg of the extracted fat content of the sample was saponified (esterified) for 5 min at 95 °C with 3.4 ml of the 0.5m KOH in dry methanol the mixture was neutralized by using 0.7 M HCl 3 ml of the 14 % Boron triflouride in methanol was added, the mixture was heated for 5 min at the temperature of 90 °C to achieve complete methylation process. The fatty acid methyl esters were thrice extracted from the mixture with redistilled n-hexane. The content was concentrated to 1ml for gas chromatography analysis and 1µ was injected into the injection port of GC.

Results and Discussion

Table 1: The physicochemical properties of alligator pepper

Properties	Alligator pepper
Acid value (mg/KOH/g)	34.22±0.2
Iodine Value (wijs)	29.44±0.2
Saponification Value (Mg/KOH/g)	145.86±0.3
Peroxide Value (mg/kg)	30.90±0.2
Refractive Index	1.4605±0.1

Specific gravity	0.9034±0.1

Table 2: Fatty acid composition of alligator pepper Oil

Parameter (%)	Alligator pepper Oil
Palmitic Acid(16:0)	17.269
Palmitoleic Acid(16:1)	0.156
Margaric Acid(17:0)	0.083
Stearic Acid(18:0)	2.791
Oleic Acid(18:1)	64.944
Linoleic Acid(18:2)	13.885
Linolenic Acid(18:3)	0.320
Arachidic Acid(20:0)	0.351
Behenic Acid(22:0)	0.200

Discussion: The results for the physic-chemical analysis were as shown in table 1. The acid value of 34.22±0.2 mg KOH/g was obtained and was relatively high compared to that reported for tropical almond (7.6 mg KOH/g) and that of fluted pumpkin (3.5 mg/KOH/g) [8]. The high acid value of the oil indicates that it is not a good source as edible oil except in a little proportion for curative purposes [9]. The iodine value of alligator pepper seed oil, 29.44mg is extremely lower to those of unsaturated fatty acid-rich oils such as peanut (86.0 – 107.0), cottonseed (100.0 – 123.0), sesame (104.0 – 120.0), sunflower (118.0 – 141.0) but higher than that of soybean oil (24.0 – 29.0) [11].

Alligator pepper oil has a peroxide value of 30.9±0.2mg/kg. This value is higher than the value recorded for *Bauchinia racemora* seed oil (4.9) [11]. Peroxide value depends on a number of factors such as the state of oxidation (quality of oxygen consumed), the method of extraction used and the type of fatty acids present in the oil. The high peroxide value recorded for alligator pepper oil in this study may be due to too much exposure of the seeds to sun during drying, causing lipid oxidation process or the moisture that accompanied the oil during extraction process. Heat favours oxidation of fatty acids increasing the formation of peroxides [12]. Thirdly, the oil contains mostly polyunsaturated fatty acids which easily undergo oxidation, raising peroxide value of the oil.

The saponification value of alligator pepper was 145.86±0.3mg/KOH/g which disagrees with values obtained for some vegetable oil ranging from 188 – 196mgKOH/g [13]. However, there were some vegetable oils with higher saponification values such as coconut oil (253.0 mg/KOH/g), palm kernel oil (247.0mg/KOH/g) and butter fat (225.0mg/KOH/g) [10]. As reported by Pearson 1976 [13], oil with higher saponification values contain high proportion of lower fatty acids. Therefore, the values obtained for egusi melon oil in this study show that it contains high amounts of higher fatty acids. The refractive index, 1.46±0.2 is in same agreement with the value of 1.46 obtained for *B. sapida* [14] oil and higher than value obtained for the same sample but different processing method by Aladekoyi and Shakpo, 2014 [9]. This shows that the oil is less thick compared with most drying oils whose refractive indices were between 1.48 and 1.49 [15]. This may as a result of the essential oil embedded in the oil sample. The value of the specific gravity (0.9034) has no significant different to values obtained by Aladekoyi and Shakpo, 2014 [9].

Table 2 showed the fatty acid profile of alligator pepper oil. The results showed that saturated palmitic acid (17.269) and stearic acid (2.792). High percentage of monounsaturated Oleic acid (64.944) and linoleic (13.885) are the main fatty acid present in alligator pepper oil. The linoleic acid level in alligator pepper oil obtained in this study disagreed to that of alligator pepper seeds oil from Niger (30.0–74.0 %) [16]. This may as a result of different extraction method used. The lower value in lenolenic acid reduced the off-flavour and oxidation of some harmful product as reported by Gupta and Warner, 2003 [17]. These results show that alligator pepper oil is better than animal fats in their content of linoleic acid, while animal fats contain mostly oleic acid (29.0 – 0.48%) [18]. Present results are also showed no significant difference to that of previous study on *Cucurbita pepo* seed oil which was found to contain palmitic, stearic, oleic and linoleic with oleic acid as the most abundant [19].

Conclusion

The physico-chemical analysis carried out on the oil from alligator pepper seed indicated that the values obtained were within the range of convectional oils consumed except the higher level of the acid value obtained and good industrially because the level of the unsaturated fatty acids in the oil sample like palmitoleic acid(16;1)(0.156), Oleic Acid(18:1) (64.944) and Linoleic Acid(18:2) (13.885). The oil is prone to oxidative rancidity due to the level of the acid and peroxide value present for a long period of time.

Recommendation

The results obtained in this work for the physico-chemical and fatty acid composition indicated that the oil had good industrial and pharmaceutical application due to the level of the unsaturated fatty acid present. It is recommended that further analysis should be carried out to know its biofuel potency.

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