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Evaluation of antioxidant capacity and physicochemical properties of Sudanese baobab *(Adansonia digitata)* seed- oil

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Abstract

The oil quality parameters of the seed oil of Baobab *(Adansonia digitata)* were evaluated using standard methods of analysis. The Iodine value, Peroxide value, Saponification value were 86 g/100g, 4.08 mEq/Kg, 188 mg/g, respectively, for seed oil. The oil content of the kernel was higher 23% compared to the hulls that contain 5.4% oil. The kernel oil contains substantial quantities of calcium, potassium, and magnesium, which were found to be 4116, 2339 and 1629 mg/Kg, respectively. The fatty acid profile showed that oleic and linoleic were the major unsaturated fatty acids, whereas palmitic was the major saturated acid. The oil also, showed considerable amount of total phenolic content (TPC) and worthy antioxidant activity. Baobab oil has great nutritional and industrial potentials. It is therefore recommended that more and advanced research should be undertaken for this abundant source of natural nutritious oil.

Introduction

Seed oils have been utilized by various communities as food, medicine, for cosmetic applications and as fuel. The multimillion dollar seed oil and natural products industry has grown enormously with an annual growth rate of 15 – 20% because of the increasing demand for seeds as a source of oils (Vermaak *et al*., 2011). Oil are used in many ways for food texturing, frying, manufacture of soap, cosmetics, detergent and oils paint (Birnin-Yauri and Garba, 2011). Researchers have controlled their efforts at discovering new ingredients in cosmetic and can be used as sources of food, Baobab seed oil is a marvelous source of unsaturated fatty acids including oleic, linoleic, and linolenic. Seeds oil are particularly essential sources of vitamins D and E (McKevith, 2005), which were founded in Baobab oil, including vitamins A, D, E and K (Nkafamiya *et al.*, 2007) Vitamins A and K are polyunsaturated fatty acids and these acids are directly responsible of the renewal of cell membranes also, rich in vitamin E, which affords oxidative stability and a long shelf life (Koroch *et al*., 2007) Also, the existing of Vitamin E, Linoleic oils which are useful for the protection of the skin production and moisturization, It can help with decreasing inflammation and promote the reform of the cells and tissue generation (Vermaak *et al*., 2011). Linoleic acid (found in baobab seed oil) is the most accustomed fatty acid in cosmetic products. The oil is used in wound care treatment and bath oil preparations, moisturizer and massages oil, and is used for hot oil hair bathtubs. On the other hand, the high proportion of linoleic acid in the seeds of baobab oil, food reflects the importance of the seed and the potential of baobab seed oil (Osman, 2004). The oil from the baobab seed contains about 1 - 2 mg/g linoleic acid, it is necessary for the body for growth and development of essential fatty acids. Studies showed the potential Baobab seed oil for food pharmaceutical and cosmetics applications. The baobab seed oil is an important source of minerals. Oil seeds are also used in animal feed because of their high protein content. Their seeds contain energy for the sprouting embryo mainly as oil, compared with cereals, which contains the energy in the form of starch. The aim of the present study was to investigate the information of antioxidant properties and physio-chemical characterization of the oil and its mineral and fatty acid composition, therefore, search on the antioxidant activity of the baobab seed oil that expected to exhibit good antioxidant properties.

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Materials and methods

Materials Collection and Preparation

Baobab fruit was harvested from the area closed to the city Kosti in Sudan. Fruits were cracked and grind and the seed kernels were manually removed from the seed shell using filter. Sohxlet extractors were obtained from the laboratory of engineering Department of biotechnology, IIUM.

Solvent extraction (Soxhlet)

The seed were ground in a hammer mill, the powdered seed kernel and seed hull, 10 grams, was used for the extraction process using a Sohxlet extractor. Organic solvents, 200 mL was measured into separate 250 mL round bottom and the Sohxlet with a thimble containing the seed powder and seed hull, a condenser were assembled. The solvent refluxed for 8 hours. Then, it was concentrated using rotary evaporator to obtain light yellowish oil. The oil was stored in a labeled sample bottle.

Physiochemical characteristics of the oil

The acid value, saponification value, iodine value were determined using the procedures described by AOCS.

Determination of iodine value (IV)

Wiji's method was adopted in determining the iodine value. 0.1 M iodine mono-chloride in acetic acid was added to 0.2 g of the oil dissolved in cyclohexane. The mixture was allowed to stand for 10 min. The mixture was allowed to stand for 10 min, to allow for halogenation. 0.1 M of potassium iodide solution was added to reduce excess iodine mono-chloride to free iodine. The liberated iodine was titrated with a standardized solution of 0.1M sodium thiosulphate using starch indicator (Wijs, 1929). The iodine value was calculated from the following equation [1]:

$$IV=((S-B)x\ M\ x\ 12.69)/(Sample\ weight\ (g)) \quad [1]$$

Where B = blank titre value,
S = sample titre value,
M = molarity of $Na_2S_2O_3$ and 12.69 = conversion factor from Meq. N

Determination of saponification value (SV)

Two grams of the oil sample was added to excess alcoholic KOH. The solution was heated for two minutes to saponify the oil. The unreacted KOH was back-titrated with standardized 0.1 M HCl using phenolphthalein indicator. The saponification value was calculated from the following equation [2]:

$$SV=((S-B)x\ M\ x\ 56.1)/(Sample\ weight\ (g)) \quad [2]$$

Where S = Sample titre value,
B = blank titre value,
M = molarity of the HCl and 56.1 = the molecular weight of KOH

Determination of peroxide value

Exactly 1.0 g of potassium iodide and 20 mL of solvent mixture (glacial acetic acid: chloroform, 2:1 v/v) were added to 1.0 g of the oil sample and the mixture was boiled for one minute. The hot solution was poured into a flask containing 20 mL of 5% KIO_3 solution. Few drops of starch solution were added to the mixture and the latter was titrated with 0.025 M sodium thiosulphate $Na_2S_2O_3$ solution (Nkafamiya *et al*., 2007).

Elemental analysis

Mineral analysis was carried out after 2 g of the seed sample was ashed and 10 mL of Conc. HNO_3 was added to it and digested until a clear solution was obtained. The digest was allowed to cool and then transferred into a 100 mL standard flask and made up to mark with de-ionized water. The mineral elements were analyzed with atomic absorption spectrophotometer (GBC Avanta Ver. 2.02 Model, Australia) equipped with air-acetylene flame.

Fatty acid composition

Fatty acid composition was analyzed by gas–liquid chromatography after derivatization to fatty acid methyl esters (FAMEs) with 2 M KOH in methanol at room temperature according to the IUPAC standard method (Dieffenbacher and Pocklington, 1992).

Antioxidant assay

DPPH radical-scavenging activity was determined by the suggested method (Kedare and Singh, 2011) with slight modification. Briefly, 0.025 ml of sample extract of various concentrations, 0.075 ml of distilled water and 0.1 ml methanol were put into a 96-well microplate. Then, 0.025 ml of 1mM DPPH in methanol solution was added. The sample mixture was incubated at 37°C for 30 min. The absorbance was measured at 517 nm. A DPPH radical solution without sample extract was used as a control. All analyses were run in triplicates. The scavenging activity was expressed as percentage of inhibition, which was calculated according to the

below equation [3]:

Scavenging ability (%)=(ΔA517 of control-ΔA517 of sample)/(ΔA517 of control) x10 [3]

Results and discussion

Baobab seed is the one of the major additional sources of oil, due to the appreciate amount of oil extracted from baobab seed kernel.

Oil in Kernel = 23 %

Oil in Hull = 5.4 %.

In general, baobab seeds are rich in oils; their content varies due to differences in species and environmental factors. (Birnin-Yauri and Garba, 2011) reported that the oil content of baobab seed from Kebbi State, Nigeria has an appreciate amount of oil 22.5%, this result is closed to our result, while (Chindo *et al*., 2010) found that the seed from Bauchi state, Nigeria has oil content 35% a higher percentage of oil than our result.

The presence of important natural antioxidants in plant food is attracting further interest because of their clear benefits as anti-carcinogenic agents and as inhibitors of biologically harmful oxidation reactions in the body. The analytical methods used to determine the oxidation and the oxidation condition are the mean factors that must be careful when evaluate the antioxidant (Frankel, 1993; Barros *et al.*, 2009). In our knowledge our study is the first study dealing with the antioxidant activity of phenolic compounds of baobab seed oil. The oxidation of unsaturated fatty acids is one of the major causes of the development of off-flavor compounds and in the reduction in the nutritional value of food products. The oxidative stability is an important quality and safety parameter of oils for their potential commercial applications and utilizations in food and other commercial products (Parker *et al*., 2003). 2,2-Diphenyl-1-picrylhydrazyl (DPPH) is a stable free radical and has been commonly used to screen phenolic compounds containing high free radical scavenging ability (Lee *et al*., 2007). Our data shows that extracts of baobab oils have far greater antioxidant capacities; the antioxidant of the oil is found to be 1430 mg/100g of Gallic acid. Indeed, many seeds oil are source of vitamin E (an antioxidant) the level of phenol in seed oils is an important factor while assessing the quality of oil because these compounds have been correlated with color and the shelf-life of oil, and particularly its resistance to oxidation. Baobab oil had been shown to be rich in the content of polyphenols.

Table 1. Physio-chemical properties of baobab seed oil*

Solvent	SV (mgKOH/g)	IV (g/100g)	PV (mEq/kg)
Hexane	188 ± 13	86 ± 9	4.08 ± 0.7

*Results are mean of three replicates ± Standard deviation

Table 1 represents some physico-chemical characteristics of the oil from the seeds of baobab. The saponification number of the oil was high, value was 188 mg KOH/g oil, and this value is within the range of some edible oils as peanut oil (165) and vegetable oil (186), whereas this indicates that the oil could also be used in soap making. While the saponification number of the oil was high iodine value is low which indicate the high stability of oil. The iodine value measures the unsaturation of the oils, which reflects the susceptibility of oil to oxidation. It is one of the parameters used to measure the oil quality.

Determination of peroxide value can give an idea about the early stages of oil oxidation. The low value of peroxide indicates the high quality and stability of the oil. Rancidity reaction that occurred during storage is extent of Peroxide value, it is a result of oxidation of unsaturated fatty acids and it depends on the degree of oil unsaturation. Whereas, baobab oil is rich of polyunsaturated fatty acid, were expected to be faster oxidation which is not seen because the Baobab seed oil has high resistance to rancidity due to presence of high antioxidant content.

Baobab seed oil contained significant amounts of important mineral element (Table 2). The mineral analysis of baobab seed oil shows that baobab seed oil is an excellent source of minerals such as Ca, Mg, and K. Potassium was the most abundant element in the baobab seed oil, followed by calcium, Sodium and Magnesium. Seed oil provides appreciable amounts of microelement iron. The high calcium contents of seed oil make the baobab seed oil attractive as a natural source of calcium supplementation. The levels of these minerals suggest that the consumption of baobab seed oil could contribute to the daily requirements of the elements studied; the seed oils are consumed in relatively small amounts, the average weekly intake of seeds oil, including non-consumers, was 17 g for men and 12 g for women (Whitton *et al.*, 2011) that could contribute partially to the overall daily intake of these elements (Chadare *et al.*, 2008).

Fatty acid composition of baobab seed oil (Table 3) showed that total unsaturated fatty acids account more than 67.15% of the total fatty acids. Saturated fatty acids (SAFA) accounted for 31.49% of total fatty acids, among them the main saturated acids were palmitic C16:0 and Stearic acid C18:0, with

Table 2. Mineral composition of Baobab seed oil*

Mineral content of seed oil (mg/kg)	
Parameter	**Results**
Calcium (ca)	411.6 ± 27
Iron (Fe)	031.8 ± 2.4
Potassium (K)	2339 ± 88
Sodium (Na)	235.9 ± 32
Magnesium (Mg)	162.9 ± 21

*Results are mean of three replicates ± Standard deviation

Table 3. Fatty acid profile of Baobab seed oil*

Type of fatty acid	Name	symbol	%
Saturated	Myristic	C14:0	0.168 ± 0.01
	Palmatic	C16:0	21.76 ± 1.45
	Stearic	C18:0	08.85 ± 0.53
	Arachidic	C20:0	00.17 ± 0.00
	Behenic	C22:0	00.33 ± 0.02
	Lignoceric	C24:0	00.21 ± 0.01
Monounsaturated	Margaroleic	C17:1	00.29 ± 0.02
	Oleic	C18:1	36.40 ± 2.41
	Gadoleic	C20:1	00.30 ± 0.02
Polyunsaturated	Linoleic	C18:2	25.50 ± 1.68
	Linolenic α	C18:3n3	02.60 ± 0.22
	Linolenic γ	C18:3n6	02.06 ± 0.13
	SFA		31.49
	MUFA		36.99
	PUFA		30.16

*Results are mean of two replicates ± Standard deviation
Abbreviations: SFA Saturated fatty acids, MUFA monounsaturated fatty acids, PUFA polyunsaturated fatty acids

minute amounts of arachidic, lignoceric, myristic and margaric. As well as linoleic C18:2 and oleic acid C18:1, were observed in baobab seed oil, While the percentage of Linolenic γ and Linolenic α were relatively low. The ratio of high polyunsaturated fatty acids (PUFA) to SAFA was 2: 1 is due to a high level of PUFA. Whereas, the biological activities and potentials in disease prevention have led to recent recommendations for an increase in daily intake of n-3 unsaturated fatty acid from 0.1–0.2 g to 0.6 g will provide potential health benefits in preventing cancers, heart diseases, hypertension, and autoimmune disorders (Parker *et al*., 2003). Significant amounts of the, an n-3 fatty acid, and linoleic acid, an n-6 fatty acid. From these two fatty acids, the body can make all the fatty acids it needs (McKevith, 2005). Generally, high blood cholesterol levels normally correlated with high intakes of saturated fatty acids and trans fatty acid, one of the risk factors associated with CVD. In comparison, MUFA decrease the 'bad cholesterol'. The major MUFA present in all foods was oleic acid (C18:1) which was particularly high in baobab seed oil. Linoleic acid (C18:2) was the most abundant polyunsaturated fatty acid (PUFA), presence of high content of linoleic acid reflect the nutritive significance and potential of baobab seed oil as healthy food oil.

This result was similar to those reported the fatty acid for baobab seed oil showed that the oils of baobab seeds contained saturated (33%), monounsaturated (36%) and polyunsaturated (31%) fatty acids. The comparison of data with those of the variety from Kano, Nigeria (Eteshola and Oraedu, 1996) showed that there were significant differences in the case of saturated fatty acid, the most abundant saturated fatty acids were myristic, whereas the main unsaturated fatty acids present were oleic and linoleic in agreement with our results

Conclusion

In this study, the proximate analysis of *Adansonia digitata* seeds oil has been determined. Also, physicochemical characterization and anti-oxidant capacity has been analyzed. The results showed that the oil has a very good yield with a high degree of unsaturation. This can make it serve as potential dietary, and rich in oleic acid (18:1n-). Baobab containing no added synthetic antioxidants may exhibit very good oxidative stability and be suitable for food preparations and useful in various industries and for human consumption and makes it unique and desirable oil.

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