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PHYSICO-CHEMICAL CHARACTERIZATION AND EXTRACTION OF OIL FROM BALANITES AEGYPTIACA PLANT (SEED)

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ABSTRACT

Balanites Aegyptiaca is deep rooted arid zone tree has a very wide natural range. The tree is value for its fruits and seeds. The Balanite Aegyptiaca seeds are good source of oil, which can be used as a diesel substitute. In this paper, Balanite Aegyptiaca oil was extracted from the seeds using n-hexane (60 ^{o}C) by using sohxlet extraction method. The oil yield was 87% and the Physico-chemical properties of the oil investigated were Acid value (1.25), saponification value (28), iodine value (62.2), peroxide value (6), specific gravity (0.9), and viscosity (26.5). By using Gas Chromatography (GC) three fatty acids in the extracted oil were identified as; Linoliec acid, Palmitic acid, and Stearic acid with 75.87%, 14.70%, and 9.40% respectively. The oil exhibited high oil content and good Physico-chemical properties which resemble within ASTM 6751 standard specifications.

KEYWORDS: Physic-chemical properties, Balanites, Oil, Gas Chromatography, n-hexane

INTRODUTION

In Ethiopia oil scarcity a major problem. Most of the time in this country oil manufactured from coconut, Maringa, olive, castor, etc. But there are other potential sources of oil in which the country still didn't utilize them. Balanites Aegyptiaca plants fruit is one of the source of oil rich fruit product that must be used in order to increase the oil yield to fulfill the demand of the people, and to upgrade the oil quality in order protect people from health risk.

Balanites Aegyptiaca is a species of tree, classified either as a member of the zygophyllaceae or Balanitaceae. It is multi branched, ever green, and the flowers are small.[1] The plant grows in tropical and desert areas. It can be found in many kinds of habitats, tolerating a wide

variety of soil types from sand to heavy clay and climatic moisture.[2,3] It is believed indigenous to all dry lands of Ethiopia, extending southward to Arbaminch. It is allowing land species, growing up to 1000m altitude.[3]

Balanites Aegyptiaca is perennial plant used in food preparations, especially in Africa and developing countries. It has multiplicity of uses and almost every part of the plant is useful including, leaves thorns, back of root and fruit. The fruit is used to treat liver disease and as a purgative and sucked by schools children as a confectionary in some countries. It is Edible fruit and its seed have 40-87%t of edible oil; leaf and fruits are eaten by goats, sheep and camels. Balanites Aegyptiaca seed kernel is considered as an extremely useful edible product. And it is used for extraction and the oil is used for human consumption and cosmetics. The Balanites Aegyptiaca seed oil has been used in many countries as ingredient and substituent to ground nut oil in the preparation of local food.[4,5]

The seed kernel obtained after cracking the nut is an oil source. Local methods for harvesting and processing of balanites products were examined as a step towards promoting their wide use and development of improved processing method.[7]

Oil is composed of molecules called triglyceride which chemically contain glycerol molecules with each other and fatty acid. Oil can be hydrolyzed by water or alkali with the production of long chain fatty acid and glycerol.[8]

In the presence of Alkali hydroxide catalyst, the chemical reaction between oil (triglyceride) and alcohol undergo Transesterification (alcoholysis) reaction to produce monoesters and to reduce viscosity of the seed oil.

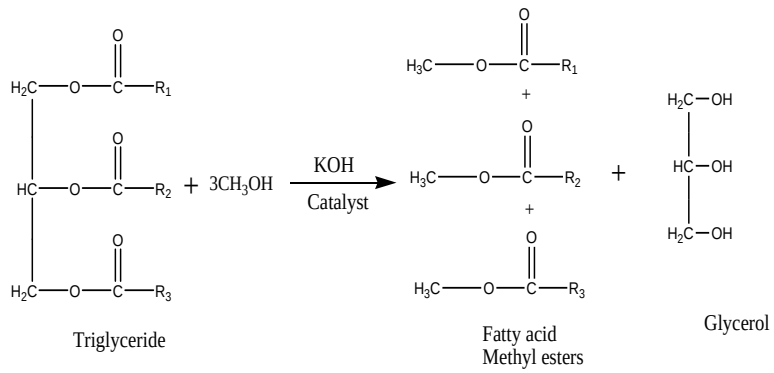

Figure 1: transesterification reaction of oil with methanol in the presence of sodium hydroxide as catalyst.

Oil shows chemical properties of Iodine value (direct representation of the degree of unsaturation), Saponification value (a measure of the amount of triglycerides), Acid value (a measure of the amount of free fatty acids) and Peroxide value (a measure of oxidation or rancidity).[9,10]

Palm oil and Peanut oil are the most common cooking ingredients of food in most countries.[10,11,12] Biodiesel is a diesel replacement fuel for use in Compression ignition engines. It is manufactured from plant oils (soybean oil, cottonseed oil, canola oil), recycled cooking greases or oils (e.g., yellow grease), or animal fats (beef tallow, pork lard) etc. Because plants produce oils from sunlight and air, and can do so year after year on crop lands, these oils are renewable.

There are many standardized procedures for production of biodiesel. The commonly used methods are: Blending, Micro Emulsification, Thermal Cracking and Transesterification. Among these, transesterification of vegetable oils appears to be more suitable because the byproduct (glycerol) has commercial value. Transesterification (alcoholysis) is the chemical reaction between triglycerides and alcohol in the presence of catalyst to produce monoesters.[13]

Solvent extraction is an industrial process in which highly purified, volatile hydrocarbons are used to dissolve aroma compounds from plant material, followed by the removal of the solvent by distillation.[14] The objective of this research was to investigate the production of oil from Balanites Aegyptiaca plant seed and to characterized physico-chemical properties of its extracted oil.

MATERIALS AND METHODS

Study area

Arba Minch is a city of Gamo Gofa zone in southern nations nationalists and peoples region at about 500 km south of Addis Ababa (capital city of the country) while 275km south of Hawasa (capital city SNNP). Arba Minch lays at an altitude of between 1200 and 1275 meters above mean sea level, its geographical position is $6^{o}2$”N 37^{o} 33’’ E. Its geographical landscape is low land and comfortable for the growth of Balanites Aegyptiaca plants.

Sample collection and Preparation

The seeds were collected from Arba Minch University main campus, Arba Minch, Ethiopia. To avoid the bias on data, samples were collected by dividing sub areas. It was screened to remove the defective ones and discarded while those in good condition were soaked in large bowl of clean water (overnight), to remove the glycoside pulp from seed coats. The washed seeds plus its heavy coats were further dried in the oven at 60oC for 4hrs and then crushing by a metal hammer crusher and pestle to obtain its kernel.

OPERATION OF SOHXLET EXTRACTOR

300 ml of normal hexane was poured into round bottom flask. 10 g of the Sample was placed in the thimble and was inserted in the center of the extractor. The sohxlet was heated at 60^{o}C. When the solvent was boiling, the vapor raised through the vertical tube into the condenser at the top .The liquid condensate drips into filter paper thimble in the center, which contains the solid sample to be extracted.

The extract seeps through the pores of the thimble and fills the siphon tube , where it flows back down into the round bottom flask. This was allowed to continue for 30 minutes. It was then removed from the tube, dried in the oven, cooled in desiccators and weighed again to determine the amount of oil extracted. Further extraction was carried out at 30 minutes interval until the sample weight at further extraction and previous weight becomes equal. The experiment was repeated by placing 5g of the sample into the thimble again. The weight of oil extracted was determined for each 30 minute interval. At the end of the extraction, the resulting moisture (miscella) containing the oil was heated to recover solvent from the oil.

Determination of Moisture Content of the Seeds

Plate weight was noted primarily and then Sample weight (of the ground Sample with plate 5-7g) was taken and dried in an oven at 60 ^{o}C for four hours and the final weight was taken. The percentage moisture in the seed was calculated using equation.

$$\text{Moisture content} = \frac{W_1 - W_2}{W_1 - W_P} \times 100\% \quad (1)$$

Where W_1 = original weight of the sample before drying, W_2 = weight of the Sample (with plate) after drying and Wp = plate weight.

Extraction and Determination of Seed Oil Content

Oil extraction was conducted at Arbaminch University Chemistry department laboratory. Sohxlet extraction method (horoitz, 1984) with hexane as extracting solvent was used each extraction was proceed for four hours. It was then removed from apparatus, cooled in the discator and solvent removed using rotavapour. The experiment was repeated in triplicate. The weight of the oil Extracted was determined for each replicate, and the mean value was recorded and the percentage of oil extracted was determined using (equation 2). Sample weight was taken dry base, based on the moisture content determined.

$$(2)\ \text{Seed Oil content} = \frac{W_0}{W_s} \times 100\%$$

Where W_o = weight of oil extracted and W_s = weight of sample (dry base)

Analysis of Physico- Chemical Properties of the Extracted Oil

DETERMINATION OF SAPONIFICATION VALUE

First, 5g of the sample was weighted in to a flask and 50 ml of alcoholic KOH was added from burette by allowing it to drain for a definite period of time and a blank was also prepared by taking only 50 ml of alcoholic KOH allowing it to drain at the same duration of time. The flask was connected to the air condenser and it was boiled gently for about one hour. Then the flask and condenser was cooled, the condenser was rinsed with a little distilled water and then the condenser was removed. Finally, 1 ml of indicator was added and it was titrated against 0.5N of HCl until the pink color was disappeared.

DETERMINATION OF ACID VALUE

2 grams of the given oil was weighted in 250ml conical flask and it was dissolved in 25ml of alcohol. Then two drops of phenolphthalein indicator was added. The contents were titrated with alcoholic KOH. Blank titration was performed on 100 mL of the titration solvent and 0.5 mL of the indicator solution, adding 0.1 ml increments of the 0.1 M KOH solution. The KOH solution was standardized frequently to detect morality change of 0.0005.

The volume of 0.1M KOH (VA), for the sample titration, and volume for the blank (VB) were noted. Then, The total acidity (Acid Value) - **AV (**mg KOH/g**)** was calculated using (Equation 3):

$$(3)\ \text{Acid Value} = \frac{(V_A - V_B) \times N \times 56.1}{W_0}$$

Where, W_O = sample weight, V_A = Volume of KOH used for the sample (ml), V_B = Volume of KOH used for the blank (ml), and N= Concentration of KOH used (Molar),

DETERMINATION OF PEROXIDE VALUE

One gram of the Balanites aegyptiaca oil was weighed in to a clean dry boiling tube and 1 g of powdered KI and 30ml of solvent (2 volumes of chloroform and 3 volumes of glacial acetic acid) mixture was added. After this, the tube was placed in boiling water so that the liquid boils with in 30 seconds and allowed to boil vigorously for not more than 30 seconds. The contents was transferred quickly to a conical flask containing 20 ml of 5% KI solution and the tube was washed twice with 25ml water each time and collected in to the conical flask. Then, the solution was titrated against 0.01N $Na_2S_2O_3$ solution until yellow color disappeared and 0.5ml of starch was added with vigorous shaking and titrated carefully till the blue color just disappeared.

DETERMINATION OF IODINE VALUE (IV)

The weighed amount (0.25g) of substance (W) was dissolved in 15mL carbon tetrachloride in a conical flask. 25.0 mL of 0.2N Hanus reagent (prepared by dissolving 9g of iodine trichloride in a mixture of 700mL glacial acetic acid (purity at least 99%) and 300mL carbon tetrachloride) was added from a burette. The flask was closed, mixed, and allowed to stand in the dark at about 20^{o}C for 1hour. After standing, 20 mL potassium iodide solution and approximately 150mL water were added. The iodine liberated by the process was titrated with sodium thiosulphate solution while shaking and starch indicator was added towards the end of titration (and volume Va was recorded). Blank determination was made with the same quantities of reagents at the same time and under the same conditions (and volume Vb was recorded). Finally the iodine value (IV) was calculated using (Equation 4).

$$\text{Iodine Value} = \frac{12.69 \times (V_A - V_B) \times N}{W} \quad (4)$$

Where, W = weight (g) of sample taken, Va =Volume (mL) of thiosulphate solution used in test, V_b =Volume (mL) of thiosulphate solution used in blank and N =Normality of thiosulphate solution.

Fatty acid compositions

The composition of oil sample was analyzed using a Gas Chromatography (GC) of (model GC-2014, Shimadzu, JAPAN). The GC oven was kept at 65ºC for 3.0 min, heated at 10

C/min up to 280 ºC, where it was kept for 5.0 min, and a total analytical time was 26 min. The carrier gas was helium. The analysis of a sample by GC was carried out by injecting 1 µl of the sample solution into the GC. The identification of the fatty acids was achieved by retention times when compared with authentic standards analyzed under the same conditions and relative percentages of each fatty acids was determined based on peak area measurements.

RESULTS AND DISCUSSIONS

The oil yield from Balanites Aegyptiaca seeds was 87%. The Balanites aegyptiaca oil was bright yellow in color, Liquid at room temperature with palatable flavor.

Table 1: Physico-chemical Properties of Balanites Aegyptiaca seed oil

Properties	Volume of analyte	Volume of blank	Obtained value
Acid value	0.55ml	0.1ml	1.25 mg/g
Saponification value	25ml	0.5ml	28 mg/g
Iodine value	50ml	0.4ml	62.2 mg/g
Peroxide value	0.6ml	0ml	6 mill eq/kg
Oil content	-	-	87%
Specific gravity	-	-	0.9
Viscosity	-	-	26.5
Oil color	-	-	Bright Yellow
Moisture content	-	-	17.65

Table 2: The Relative percent composition of fatty acid in Balanites Aegyptiaca oil

No.	Name	Retention Time (Min)	Area %	Height%	Concentration %
1	Palmitic Acid	15.57	14.635	27.5651	14.700
2	Linoliec Acid	17.45	75.899	51.7429	75.864
3	Stearic Acid	17.69	9.5051	20.6920	9.436

The result shows that, the oil content of the extracted oil of Balanites Aegyptiaca was high, it was found to be 87% (Table 1) which shows that the processing of the oil for industrial or edible would be economical feasible. The value was greater than as research works presented in.[13,16] this show that Balanites seed is productive. Moisture content of the seed is high (17.65%), that indicates the oil has high content and this indicates also that the seeds and oil can have long shelf live.

The viscosity of the oil at room temperature was 25.6 (table 1). From the result presented, it can be seen that the viscosity is on the high side when compare to the ASTM standard value expected for an engine to work properly. The viscosity of Balanite aegyptiaca oil seeds must

be reduced for oil application since the viscosity of this oil was very low compared to vegetable oils. The result shows that the oil had low friction; this was due to the intramolecular interaction (force) in oil was low and the oil had high unsaturation that leads the oil poorly close pucks between the molecule, since the oil easily flow.

Acid value was determined to quantify the fatty acid found in the oil. The acid value was very low (1.25 mg/g). It was lower than the research work presented in.[16] this shows that this oil was stable.

Saponification value was determined to quantify the amount of triglyceride. Saponification value of the oil was greater than acid value this was due to inversely related proportional, but this value was lower than saponification value of.[13,16]This shows that the oil was less useful in the production of soap.

Iodine value was determined to quantify the degree of unsaturation in oil and it was an identity characteristic of native oil. It indicates the degree of unsaturation in the fatty acids of triglyacylglycerol. This value could be used to quantify the amount of double bonds present in the oil which reflects the susceptibility of oil to oxidation. The iodine value obtained was high which suggest the presence of unsaturated fatty acid and this places the oil in the drying groups. This oil may find application as raw material in industries for the manufacture of vegetable oil based ice cream. The result obtained was agreed with.[13] but lower than.[16] This enables the oil to resist rancidity (autoxidation) Peroxide value was determined to quantify the rancidity of the oil. The value was (6 meq/kg) lower than the expected of rancid which ranges from 10-20 meq/kg. This value overcomes due to careful preservation of the oil. The oil exhibited better peroxide value than others. The result indicated that the oil had no rancidity or it had good flavor quality and considered as stable.

Table 2 shows the presence of three major fatty acids in the Balanite aegyptiaca oil, the main unsaturated fatty acid is linoliec acid (C18:2) (75.86%), while the saturated fatty acids are palmitic acid (C16:0) (14.73%) and stearic acid (C18:0) (9.40%). The overall oil sample contained saturated and unsaturated acids of 24.13% and 75.86% respectively. According to[17], the properties of the triglyceride and oil fuel are determined by the amounts of each fatty acid that are present in the molecules.

CONCLUSSIONS

Generally, high yield oil was investigated from Balanites Aegyptiaca seed. The concept of extracting and physicochemical analysis (acid value, saponification value, iodine value, peroxide value, viscosity, and specific gravity) of oil produced from Balanites Aegyptiaca was a novel to the country and indispensable to solve oil scarcity problems and to upgrade its quality. The oil was characterized to determine the presence of free fatty acids, the amount of unsaturated bond, fluidity of oil and flavor quality of the oil. This study showed that Balanites Aegyptiaca seed was good source rich oil. Balanites Aegyptiaca seeds oil have high contents of unsaturated fatty acid, the free fatty acids and peroxide value were low indicating oils could quality as good edible oils because of satisfying properties that could be compared with ASTM 6751

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