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Physicochemical Properties of *Balanites aegyptiaca*
(Laloub) Seed Oil

الخصائص الفيزيوكيميائية لزيت بذرة شجرة الهجليج (الللوب)

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الخواص الفيزيوكيميائية لزيت بذرة شجرة الهجليج (اللاوب)

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Dedication

TO MY HUSBAND

TO MY FATHER

TO MY MOTHER

TO MY TEACHERS,

TO MY FRIENDS

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First of all, I would like to thank almighty God who gave me his blessings, good health and support to accomplish this study.

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List of Contents

Title	Page No.
Dedication	I
Acknowledgements	II
List of Contents	III
List of Tables	VI
List of Figure	VII
Abstract.....	VIII
.....	IX
CHAPTER ONE.....	1
Introduction	1
CHAPTER TWO.....	3
LITERATURE REVIEW.....	3
2.1 Description of plant.....	3
2.2 History of the plant.....	4
2.3 Distribution	4
2.4 A tree adapted for arid and semiarid regions	6
2.5 Nutritional value.....	6
2.6 Utilization.....	8
2.6.1 Food utilization	8
2.6.2 Medicinal utilization	9
2.6.3 Folkloric utilization.....	10
2.6.4 Fodder utilization	10
2.6.5 Gum or resin utilization	10
2.6.6 Toxicity of <i>Balanites aegyptiaca</i> fruit	10
2.6.7 Hedges and forestation.....	11
2.7 Processing of oils	11
2.7.1 Back ground	11
2.7.2 Vegetable oil processing	11
2.7.3 Technology of oils processing	12

2.7.4 Refinement level	12
2.7.5 Extraction processes.....	13
2.7.6 Chemical and solvent extraction	14
2.7.7 Mechanical extraction	14
2.8 <i>Balanites aegyptica</i> seed oil.....	16
2.8.1 Chemical composition of <i>B.eagyiatica</i> seed	18
2.8.1.1 Fats of <i>B.eagyiatica</i> seed.....	18
2.8.1.2 Crude Proteins of <i>B.eagyiatica</i> seed	19
2.8.1.3 Minerals of <i>B. eagyiatica</i> seed.....	19
2.8.2 Physico-chemical characteristics of <i>Balanites. eagyiatica</i> seeds oil	20
2.8.2.1 Colour.....	21
2.8.2.2 Refractive index	21
2.8.2.3 Viscosity	21
2.8.2.4 Density	22
2.8.2.5 Free fatty acid.....	22
2.8.2.6 Peroxide value.....	22
2.8.2.7 Acid value	23
2.8.2.8 Saponification value.....	23
2.8.2.9 Iodine number	23
2.8.2.10 Fatty acid composition	24
CHAPTER THREE.....	25
MATERIALS AND MATHODS	25
3.1 Material	25
3.2 Methods.....	25
3.2.1 Chemical composition of Seeds.....	25
3.2.1.1 Moisture content.....	25
3.2.1.2 Ash content.....	26
3.2.1.3 Fat content.....	26
3.2.1.4 Crude protein content.....	27
3.2.1.5 Crude fibre content.....	28
3.2.1.6 Minerals content.....	28
3.2.1.7 Total and available carbohydrates.....	29

3.3 Physical characteristics of <i>B.aegyptiaca</i> oil.....	29
3.3.1 sample preparation	29
3.3.1.1 Colour of oil	29
3.3.1.2 Refractive index of oil.....	30
3.3.1.3 Viscosity of oil	30
3.4 Chemical characteristics of <i>B. aegyptiaca</i> oil.....	31
3.4.1 Acid Value of oil.....	31
3.4.2 Peroxide value of oil	31
3.4.3 Saponification number of oil.....	32
3.4.4 Fatty acid compositions of oil.....	33
3.4.5 Iodine value of oil	33
3.4.6 Free fatty acids of oil	34
3.3 Statistical analysis	35
CHAPTER FOUR	36
RESULTS AND DISCUSSION	36
4.1 Chemical composition of <i>B. aegyptiaca</i> seed.....	36
4.2 Minerals content of <i>B.aegyptiaca</i> seed:	36
4.3 Physical properties of <i>B.aegyptiaca</i> oil	39
4.4 Chemical properties of <i>B.aegyptiaca</i> oil:.....	41
4.5 Fatty acid composition of <i>B.aegyptiaca</i> oil	41
CHAPTER FIVE.....	44
CONCLUSION AND RECOMMENDATIONS.....	44
5.1 Conclusion.....	44
5.2 Recommendations:.....	44
References	45

List of Tables

Title	Page No.
Table 4. 1: Chemical composition of <i>B. egyptiaca</i> seed.....	37
Table 4. 2: Minerals content of <i>B.egyptiaca</i> seed:.....	38
Table 4. 3: Physical propertis of B.egyptiaca oil:	40
Table 4. 4: Chemical properties of <i>B. egyptiaca</i> oil:	42
Table 4. 5: Fatty acid composition of <i>B. aegyptiaca</i> oil	43

List of Figure

Title	Page No.
Fig 1 :Atypical Oil Extraction Process (Fawad, 1993)	15

Abstract

The main goal of this research was to study the physical and chemical properties of *Balanites. aegyptiaca* (laloub) oil.

Two kilograms of *Balanites aegyptiaca* fruit were purchased from local market in Khartoum, Sudan. Fruits were then crushed using a steel hummer and seeds were then obtained. One kilogram of seeds was collected and was then ground using grinding machine. Processed ground seeds were then eventually ready for further analysis.

The results revealed that *B. aegyptiaca* seed contain considerable percentage of moisture (3.10%) , oil (42.95%) ,protein (31.08%), fiber (12.64%), ash (3.19%) and carbohydrate (3.05%) on dry matter basis .Also the seeds contained different minerals e.g calcium (0.41mg), sodium (0.09mg), magnesium (0.13mg) phosphorus (0.30mg) and potassium (1.09mg) per 100g dry matter (DM).

Also, the results of the physicochemical evaluation of *B. aegyptiaca* oil showed that it contains viscosity 19.63 cp, density 0.9109 , refractive index 1.483, colour 7.633 (R.y.b), free fatty acid 3.17%, peroxide value 1.18 (meq /kg) , saponification value 224.63 (mgKOH/g), iodine value 122.42 and acid value 1.53 a (mgKOH/g). The fatty acid profile of *B. aegyptiaca* seed oil showed that it contains linoleic34.36%, palmitic 13.37%, stearic acid 15.03% , oleic acid 28.57%.

From the previous physicochemical tests of *B,aegyptiaca* oil could be used for edible purpose.

ملخص الأطروحة

كان الهدف الأساسي لهذا البحث هو دراسة الخصائص الفيزيائية والكيميائية لزيت بذرة اللالوب. تم الحصول على 2 كجم من عينات ثمرة اللالوب من السوق المحلي بمنطقة الخرطوم. تم تكسير ثمار اللالوب بواسطة مطرقة حديدية للحصول على البذور. تم جمع 1 كجم من البذور وتم طحنها بواسطة ماكينة الطحن ثم أخذت لإجراء التحاليل الأخرى.

أوضحت نتائج الدراسة أن بذور اللالوب تحتوي على نسب مقدره من الرطوبة (3,10%) الدهون (42,95%)، البروتين (31,08%)، الألياف (12,64%)، الرماد (3,19%) والكربوهيدرات (3,05%) على أساس الوزن الجاف. كما احتوت البذور أيضا على نسب عالية من المعادن المختلفة مثل الكالسيوم (0,41)، الصوديوم (0,09)، الفسفور (0,30)، البوتاسيوم (1,09) والماغنسيوم (0,13) ملجم لكل 100 جرام من المادة الجافة.

كما أوضحت نتائج التقييم الفيزيوكيميائي لزيت اللالوب احتوائه على لزوجة (19,63 cp)، الكثافة النسبية (0,919)، معامل الانكسار (1,483)، اللون، (7,633R.y.b)، الأحماض الدهنية الحرة (3,17%)، رقم البيروكسيد (1,18)، رقم التصبن (224,63). الرقم اليودي (122,42) رقم الحموضة (1,53).

أما اختبارات الأحماض الدهنية أوضحت الدراسة احتواء زيت بذور اللالوب على الأحماض الدهنية الآتية اللينولك (34.36%)، (بالميتك 13.37%)، (الاستيرك 15.03)، (الاوليك 28.47%).

من خلال التحاليل الفيزيوكيميائية السابقة يمكن استخدام زيت اللالوب كزيت طعام للاستهلاك الآدمي.

CHAPTER ONE

INTRODUCTION

Balanites aegyptiaca (Laloub) tree is indigenous to all dry lands south of Sahara and extending southwards (Sands, 2001; Hall and Walker, 1991; Shanks, 1991; Sidiyene, 1996). It is also found in Arabian Peninsula (Arboneir, 2004), India, Iran and Pakistan (Amalraj and Shankarnarayan, 1986). In Sudan it is more likely the species with widest natural range, occur in all zones, except in very high altitudinal areas or when the rainfall exceeds 1100 mm/annum (Badi *et al*, 1989). It makes up to one third of the total tree population in central region of the Sudan (NRC, 2008). *B. aegyptiaca* had been used over thousands of years (Von Maydell, 1986). The fleshy pulp of the fruit is eaten fresh or dried. It contains 64– 72% carbohydrates, plus crude protein, steroidal saponins, vitamin C, ethanol and other minerals (Abu Al-Futuh, 1983). All parts of the tree has a medicinal uses including fruits, seeds, barks and roots. The most important is a steroidal saponins, which yield diosgenin, a source of steroidal drugs, such as corticosteroids, contraceptives and sex hormones (Farid *et al.*, 2002; and FAO, 1985). *Balanites* seed is considered as an extremely useful edible product. It contains good quality oil and high protein content (Mohamed *et al*, 2002; Abu AlFutuh, 1983). The debittered seed is used as snacks (nuts) by humans.

The extracted oil is used for many uses and it is used in Western Sudan remaining cake is used as animal feed. Both fruits and seed were widely used in many countries during the dry season and drought periods including Nigeria (Lockett *et al.*, 2000), Ethiopia, (Guinand and Lemessa, 2001) and Sudan (Grosskinsky and Gullick, 2001).

The need for vegetable oil is rising worldwide so it has to be to look for good sources for the production of high-quality oil can be exploited for industrial purposes This study was conducted on the physical and chemical properties of *Balanites aegyptiaca* (laloub)seed oil as a good source for the production of oil.

The objectives of the study

- To determine the chemical composition of *Balanites aegyptiaca* seed.
- To determine the physicochemical properties of *Balanites aegyptiaca* seed oil.
- To estimate the nutritional value of *Balanites aegyptiaca* seed oil.

CHAPTER TWO

LITERATURE REVIEW

2.1 Description of plant

Balanites aegyptiaca (laloub) is a semi-evergreen, usually spiny, extremely variable shrub or small tree of the Zygophyllaceae family that grows up to 12 m high. The bole is usually straight with a 60 cm diameter, often fluted, the branches are generally spread irregularly or pendulous, and sometimes form a round crown. The tree produces yellow date-like fruit. The trees bear heavy yields as many as 10,000 fruits annually on a mature tree in good condition each fruit, weighing 58 g, consists of an epicarp (5-9%), a mesocarp or pulp (28-33%), an endocarp (49-54%), and a kernel (8-12%). The oil content of *B. aegyptiaca* seed approaches 50% (Chapagain and Wiesman, 2005).

Vernacular name:

Ayurvedic: Ingudi, Angaar Vrksa, Taapasadrum.

Taapasa vrksa, Dirghkantaka.

Unani: Hingan, Hanguul.

Siddha: Nanjunda.

Folk: Hingol, Hingota, Hingothaa

English: Desert date, Soapberry tree, Thorn tree.

Egyptian: balsam

Arabic: Heglig

French: Dattier du desert, Hagueleg, Balanite

Spanish: corona di Jesus

2.2 History of the plant

The Balanites tree has a long history of use as a resource, especially in the African continent where it is the most wide-spread woody plant. According to Sprague (1913), *B. aegyptiaca* has been planted in Egypt for over four thousand years. Stones of the fruits were placed as votive offerings in the tombs of the Twelfth Dynasty. The tree also has biblical connections. It is believed that Balanites was the source of one of the ingredients of the perfume 'spikenard' as used by the Egyptian royalty (Hall and Walker, 1991). Today this plant plays a diverse cultural and traditional role in different societies. The Huasa community of West Africa uses the fruit of *B. aegyptiaca* to ensure immunity and protection against defeat in boxing (Irvine, 1961). Chips of the wood placed in elephant dung are used to prevent elephants from attacking people or property in East Africa. The mistletoe grown in *B. aegyptiaca* is taken as concoctions to enhance scholastic ability. The fruit is used to hang around the neck of a potential victim toward off blood-sucking sorcerers in Saharan Morocco (Burkhill, 1985). If, as is generally believed, humanity began in Africa, then the bittersweet Balanites fruit is likely among the oldest of all foods. Certainly this resilient evergreen tree has been helping mankind for thousands of years. Its fruits have been found in Pharaohs' tombs dating back to at least the 12th dynasty in ancient Egypt; thus even royalty has appreciated it for 4,000 years (Ladipo, 1989).

2.3 Distribution

Balanites aegyptiaca is perhaps one of the most wide-spread woody plants of the African continent. It is distributed through much of Africa from costal Mauritania

and Senegal to Somalia and Egypt, southwards to Zambia and Zimbabwe, as well as in the Middle East from Yemen to Jordan and Israel (Sands, 2001). Benin, Burkina Faso, Cameroon, Chad, Djibouti, Ethiopia, Gambia, Ghana, Guinea, Bissau, Guinea, Ivory Coast, Kenya, Mali, Mauritania, Nigeria, Niger, Senegal, Sudan, Somalia, Tanzania, Togo, Uganda, Zaire, and Zambia are the primary African countries where Balanites are grown (Booth and Wickens, 1988). Algeria, Angola, Burundi, Central African Republic, Libya, Morocco, and Rwanda are the other African countries where Balanites are found (Hall and Walker, 1991).

Balanites are not only grown throughout the African continent but also in the Middle East, the Arabian Peninsula, and Southern Asia. Jordan, Saudi Arabia, North and South Yemen, India, and Myanmar are the countries beyond Africa where Balanites naturally grow. Balanites are found in the Arava Valley (near the Jordanian border), Eilat (near Red Sea coast), Ein-Gedi oasis (near the Dead Sea), (Chapagain and Wiesman, 2005). The growth range of Balanites was found to extend across more than 50° of latitude: from 35° N to about 19° S (Budi district, Zimbabwe) (Zohary, 1973). However, it is mainly distributed in semiarid and arid zones in tropical Africa. The tree is called as Lalob in Arabic, Aduwa in Hausa, Hingota in Hindi, and Zaquum mitzri in Hebrew. In the Indian subcontinent, so far *B. aegyptiaca* and its subspecies are reported to be found only in India and neighboring Burma (Union of Myanmar) In India, Balanites are widely grown in Rajasthan and neighboring states, whereas in Burma this plant is recorded so far only from the Irrawady Valley and one of two adjacent areas between Yeu in the North and Prome in the South. The Burmese species is better known as *B. triflora*. The Indian species of Balanites is known *Balanites roxburghii*). Although some taxonomists have indicated the differences between these two species of Balanities, until they are precisely examined, the relationships between triflora and roxburghii remain uncertain (Sands, 2001).

2.4 A tree adapted for arid and semiarid regions

Balanites aegyptiaca has developed an armory of weapons against the conditions it must endure in its native semiarid and arid regions. In the Sahel and sub-Saharan Africa, the tree rarely exceeds 10 m in height, but the thick and tough leaves with a glossy coating provide protection from the dry air (Hall and Walker, 1991). The double root system runs vertically and horizontally, finding water up to 7 m below the surface and within a radius of up to 20 m from the trunk. This root system also helps the tree endure sandstorms which are common in this region and which uproot and severely damage other trees. Under such harsh conditions, the coating of sand around each root provides an insulating layer of air, which helps moderate temperature fluctuations and reduce evaporation. The long green spines and branches continue to photosynthesize even long after the leaves have fallen off, and thus ensure the trees' survival. Thus, from the crown of the tree down to its roots, *B. aegyptiaca* is well adapted to survive under the extreme conditions of the desert.

2.5 Nutritional value

Booth and Wickens (1988) summarized the chemical composition, minerals and vitamins content of *Balanites aegyptiaca* fruits pulp in Table (2.1) and (2.2).

Cook, *et. al.* (1998) indicated that *Balanites aegyptiaca* fruit pulp contains high amounts of sugar, protein, lipid, minerals and vitamins. As reported by NRC (2008), the total sugar of Laloub fruits pulp ranges from 40-70 % and it contains about 5 % proteins and 0.1 % fat. The mineral contents of *Balanites aegyptiaca* fruits pulp were investigated by Abdulrazak, *et. al* (2010). The concentrations of calcium, magnesium, phosphorus, sodium, sulphur and iron were found to be 24.4, 6.33, 1.58, 0.542, 1.81 and 1.23 as g/kg on dry basis (DM), respectively. While,

the concentrations of manganese, copper, zinc and selenium as trace elements were 22.5, 33.7, 32.5 and 48.0 as mg/kg on dry basis (DM), respectively.

Table (1.1): Chemical composition of *Balanites aegyptiaca* fruits pulp

	Dougal, <i>et al.</i> , (1964)	Abu_al futuh <i>et.al.</i> , (1983)	Becker <i>et . al.</i> , (1983)	Nour, <i>et . al.</i> , (1985)
	% On weight dry	% On fresh weight basis		
Dry matter or moisture Content	-	75.4 - 82.1	78.9	88.7- 89.5
Protein	8.5	3.2 - 6.6	4.9	1.2 - 1.5
Fat	6.6	01 - 0.7	0.1	0.1 - 0.4
Carbohvdrates	39.0	64 – 74	69.9	
Fiber	40.8	0.9 - 4.4	3.5	
Total sugars	-	56.7	-	34.9-37.1
Reducing sugars	-	56.1	-	28.5-33.8
Non reducing sugars	-	0.6	-	
Ash	5.1	4.9 - 6.9	-	2.4 - 2 9

Source: Booth and Wickens (1988)

Table (1.2): Minerals and vitamins content of *Balanites aegyptiaca* fruits pulp

	Dougal, <i>et. al</i> (1964)	Abu-Al Futuh (1983) <i>et.al</i>	Becker <i>et. al</i> (1983)	Nour, <i>et. al</i> (1985)	
	On dry weight basis (mg/100g)	On fresh weight basis (mg/100g)0			71.3
Phosphorus(P)	91	-	58	143-	
Calcium (Ca)	220	-	147	86.9	-
Iron (Fe)	-		4	14.5	-
(Vitamin (BI)	-	-	0.27		
Vitamin (B2)	-	-	0.07		-
Vitamin (B6)		-	1.74		-
Vitamin (C)	-	12 -27	46		-

Source: Booth and Wickens (1988)

2.6 Utilization

2.6.1 Food utilization

According to the literature, the fruit of *Balanites aegyptiaca* can be eaten either fresh or dry or it can be processed to produce different traditional products.. Sometimes, the fruits pulp is fermented to make an alcoholic drink or may be macerated in water to make a refreshing beverage (NRC, 1983 and Von- Maydell, 1986).

In Kordofan, the fleshy mesocarp is extracted and mixed with gum Arabic to make sort of sweet called "Sernev". Also, the seed of the fruit is boiled with continuous change of water to remove the bitterness of the fruit and then eaten with sorghum, this kind of food is called "Kornaka"(Abdoun, 2005 and Wildpedia, 2010).

Also, in Kordofan and Darfur the green leaves, young sprouted leaves and green thorns are eaten fresh as vegetable salad or may be cooked (NRC, 1983 and Von-Maydell, 1986 and Abdoun, 2005). Besides.. the *Balanites aegyptiaca* yellowish edible oil is released by extended boiling of the fruit seed (El Amin, 1979).

2.6.2 Medicinal utilization

Von-Maydell (1986) and EI-Ghazali, *et.al* (1994) reported that, *Balanites aegyptiaca* leaves and branches clean the malignant wounds and enhance wound healing, while, the bark extract of the tree is used as a treatment for toothaches, stomach complaints, heart burn sore throat and as a remedy for mental diseases, sterility, epilepsy, yellow fever and syphilis.

Balanites aegyptiaca fruits contain steroidal saponins which can be used as starting material for the synthesis of certain steroidal drugs for the treatment of stomach complaints, sterility, mental diseases and as sex hormones. In India, it is also used in curing of some skin diseases like Leucodermia. Moreover, the fruits and leaves are used for the treatment of syphilis, yellow fever, bilharzia and as laxative material (Karlyn and Deboraha, 1993 and Abdoun, 2005) In North Kordofan, people usually use *Balanites aegyptiaca* fruits as a drink against constipation and as anti-diabetic (E1 Ghazali *et. al.*, 1994).

The *Balanites aegyptiaca* fruits are very rich in saturated fatty acids that can be used as cooking oil. Also, it contains steroids (saponins, saponins, diosgenins) that are normally used as raw materials for industrial production of contraceptive pills, corticoids, anabolisants and other sexual hormones (Ndoye, *et. al.* 2004).

The mace-rated *Balanites aegyptiaca* fruit pulp is also mixed with millet to make porridge which is Usually given to women after child birth or during the lactation period to give them energy and to increase the milk production. Also, the

B.aegyptiaca seed was mentioned in the treatment of headache, influenza and rheumatism (Abdoun, 2005).

2.6.3 Folkloric utilization

Balanites aegyptiaca tree wood is a very good fire wood and produces good quality charcoal. The wood is resistance to the insects so it is used for making tool handles, gun stocks and furniture (NRC, 1983 and Von-Maydell, 1986). In Sudan, people use the seeds for making Sibha or for playing Siga, while, the wood of the tree is used to make wooden spoons, stools and combs (Abdoun, 2005).

2.6.4 Fodder utilization

The fresh and dried leaves, fruits and sprouts are all eaten by live stock. In Burkina Faso, *Balanites aegyptiaca*- contributed up to 38% of the dry-matter intake of goats in the dry season. Also, seed meal (the residue remaining after oil extraction) is widely used in Senegal, Sudan and Uganda as a stock feed (NRC, 1983 and Orwa, *et. al.*, 2009).

2.6.5 Gum or resin utilization

A greenish-yellow to orange-red resin is produced from the tree stem. The stem is soaked and chewed when fresh, after that, it is used as glue for sticking feathers on to arrow shafts and spearheads and in the preparation of handle cracks and arrows (Orwa, *et. al.*, 2009).

2.6.6 Toxicity of *Balanites aegyptiaca* fruit

According to the literature, the fruit and bark emulsion is lethal to the fresh water snails which are the host of miracidia and cercaria stages of bilharzias as well as to a water flea that acts as a host to the guinea worm. Also, a fish poison can be obtained from the fruit, root and bark. The saponin was found to be the active

agent of the poison, toxic only to fish without any effects on mammals and rapidly becomes inert, so that fish retrieved are edible (Orwa, *et . al.* 2009).

2.6.7 Hedges and forestation

The thorny branches of the tree are massed together to form lives hedging and boundaries. Also, the cut branches are used to make live stock enclosures (NRC, 1983 and Orwa *et. al.*, 2009).

2.7 Processing of oils

2.7.1 Back ground

(Fawad, 1993) reported that edible oils fall into two categories:

- 1- Vegetable oils which are obtained by processing: soybeans, olives, coconuts, corn, peanuts, sunflower seeds, cotton seeds, sesame seeds, flax seeds and safflower seeds.
- 2- Animal oils and fats which are rendered from the trimmings of freshly slaughtered animals.

2.7.2 Vegetable oil processing

Vegetable oils are recovered by grinding, cooking, expelling and pressing, or by solvent extraction of the raw materials.

The oils are filtered and put in:

- (A) Crude oils storage- with foots: (Foots are the solid fragments of crushed seeds foots in suspension and ensure a uniform feed to down-stream equipment. These tanks are often built with heating pipes. Agitation below these pipes will be reduced, resulting in solids settling and the need for periodic cleaning.

The next process step is called refining. In:

(B) Batch refining

The oil is treated with 20% caustic solution to react with fatty acids. This results in the formation of soft soaps which are decanted off. Too much agitation will emulsify the mix resulting in long separation times. . Use gear drive portable mixers.

Some refining is done in continuous "packaged" systems built by suppliers of continuous separations equipment (Fawad, 1993).

The oil now goes to

- (C) Refined oil Storage: These tanks provide surge capacity in the system.
Lower power levels are used (Fawad, 1993)

2.7.3 Technology of oils processing

Oil is extracted from a number of fruits, nuts and seeds for use in cooking and soap making or as an ingredient in other foods such as baked or fried goods. Oil is a valuable product with universal demand, and the possible income from oil extraction is therefore often enough to justify the relatively high cost of setting up and running a small scale oil milling business. There are two things when considering the methods by which oil is extracted from a plant: The refinement level and the physical process used to extract the oil (Fawad, 1993).

2.7.4 Refinement level

Oils are generally grouped into two groups: unrefined and refined. Unrefined oils taste more like the substances, while refined oils are blander (Fawad, 1993).

(1) Unrefined oils

These oils are used as salad oils (warm salad dressing, and pasta sauces) or light cooking oils (light sautés and low heat baking). As a general rule, they should not be cooked at high temperatures. Unrefined oils are processed by cold-pressed and expeller-pressed methods, unrefined oils carry with them the true bouquet of olives, corn, sesame seeds, peanut, soybeans, sunflower, or whatever plant was the oils original home. The strong flavors of unrefined oils can dominate whatever dish or baked good is made with them. Strong flavor is not always a drawback; and in some cases unrefined oils are used as flavoring agents (Fawad, 1993).

(2) Refined oils

These oils are used as medium cooking oils , high cooking oils and deep-frying oils (greater than 232°C). If the oil is bland and pale, it is certain that it has been fully refined, bleached and deodorized. In essence, refined oils have negligible flavor and aroma, which can be useful in delicately, flavored dishes (Fawad, 1993).

2.7.5 Extraction processes

All oil extraction processes involve heating the oil in some way. However, temperature over 70 °C destroy the proteins and natural vitamin E in oils. Lower temperatures (12-11°C to 70 °C) do not damage the oil significantly, but do reduce the yield, making good oils a little more expensive. It is essential to retain vitamin E in the oil as it prevents the oil from oxidation with little vitamin E tend to go rancid quickly unless treated with antioxidant chemicals (Fawad, 1993).

1. Expeller-pressed

These oils obtained by squeezing the seed, grain, or fruits at pressure up to 15 pound per square inch. The higher the pressure, the more heat is generated. At extremely high pressure, the temperature can exceed 100°C (Fawad, 1993).

Cold-pressed

The term cold pressed theoretically means that the oil is expeller-pressed at low temperature .Olive oil, sesame oil, and peanut oil are really the only kinds that can be truly cold-pressed on any sort of large commercial scale. True cold-pressed oils are prized. They contain minerals, phosphatides, vitamin E and high in trace nutrient (Fawad, 1993).

Oil extraction

Extracted oils are invariably subjected to some sort of applied heat during processing (Fawad, 1993).

2.7.6 Chemical and solvent extraction

The cheaper brands of oil (most regular commercial brands) generally use chemical solvents to extract the oil. The oil is separated from its food source with hexane or other petroleum solvents and then boiled to drive off the toxic solvents. The oil is next refined, bleached, and deodorized, which involves heating it to over 200°C. The oil extracted this way still contains some undesirable solvent residues, while the amounts of many key nutrients (especially vitamin E) are significantly reduced (Fawad, 1993).

Antioxidants or preservatives such as BHA (butylated hydroxyanisole) or BHT (butylated hydroxytoluene) are then frequently added. The resulting product lacks flavor, aroma, pigments, and nutrients. All that can be said for such oil is that it has an extended shelf life, a clear, uniform color and an oily texture (Fawad, 1993).

2.7.7 Mechanical extraction

In order to get high quality edible oils, various processing techniques are used. The process of obtaining oil from seeds involves the separation of oil from oil-bearing materials by mechanical means, chemical means (Fawad, 1993), etc. A typical oil extraction process is shown in Fig .1.

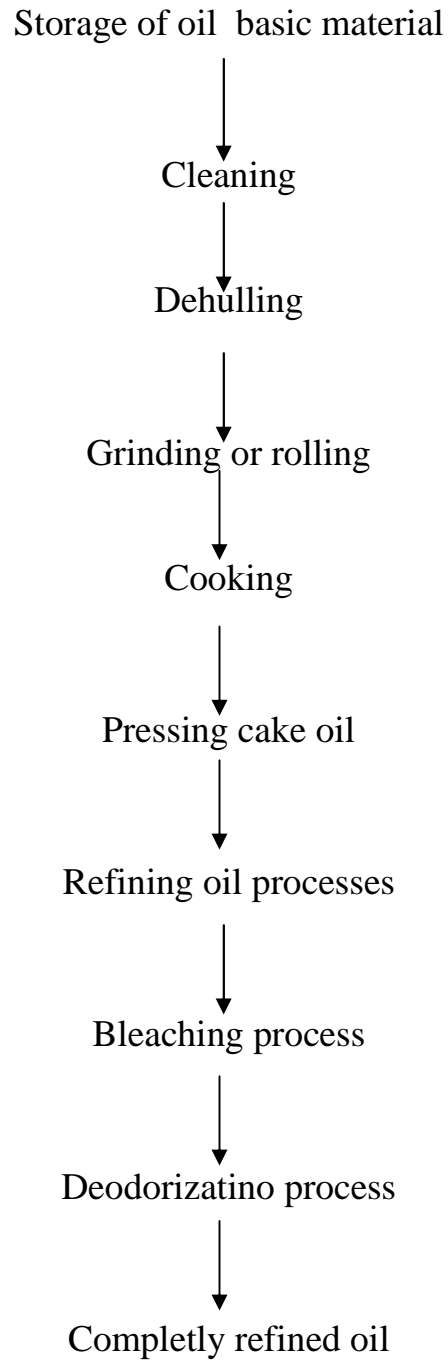


Fig 1 :Atypical Oil Extraction Process (Fawad, 1993)

2.8 *Balanites aegyptica* seed oil

Edible oils are major dietary component and plays important nutritional role as concentrated source of energy and carrier of fat –soluble vitamins .They also impart flavour and taste to foods , provide essential fatty acids and fats are required for normal functions of the body (Frezzotte *et al.*, 1956).

The term oil is used in generic sense to describe all substances that are greasy or oily fluids at room temperature. They are non-volatile and are insoluble in water but are soluble in organic solvents. Oils from seeds or kernels or nuts along with proteins and carbohydrates, constitute the majority of foodstuffs. They are also found in wide industrial applications, like formulation of soap toiletries, paints, varnishes, bio-diesels and lubricant. The criteria for the selection of oil for industrial use are: presence of natural characteristic aroma, clarity, good natural colour, very low moisture content, freedom from solid particles and freedom from flat and rancid (unpleasant) odour (Okoye, 1999).

The seed oil of *Balanites aegyptiaca* is reported to be rich in saturated fatty acids and is used as cooking oil (Hall and Walker, 1991; NRC, 2008). It also contains steroids (saponins, sapogenins, diosgenins) used as raw material for industrial production of contraceptive pills, corticoids, anabolisants and other sexual hormones (UNIDO, 1983). Reports on studies of *B. aegyptiaca* seed oil (Hussain *et al.*, 1949 ., Cook *et al.*, 1998 and Mohamed *et al.*, 2002) indicate that the seed oil consists of four major fatty acids; linolein, olein, stearic and palmitic acid but in varying proportions across study sites. Some studies (WIPO, 2006 ., Deshmukh and Bhuyar, 2009 and Chapagain *et al.*, 2009) have demonstrated and recommended use of *Balanites* oil for biodiesel production. There is therefore growing interest in understanding the development potential of *B. aegyptiaca* as a resource for improving livelihoods of dryland communities. Natural vegetable oil

and fats are increasingly becoming important worldwide in nutrition and commerce because they are sources of dietary energy, antioxidants, biofuels and raw material for the manufacture of industrial products. They are widely used in food, cosmetic, pharmaceutical and chemical industries. According to FAO (2007), vegetable oils account for 80% of the world's natural oils and fat supply. Nutritional information on Balanites oil will prove useful to nutritionists, policy makers, development agencies and the general public in Uganda and elsewhere where nutrition and health benefits would be most beneficial.

Most of the reported biological activities included using Balanites seed extract as anticancer and fruit mesocarp extract as fasciolicidal related to polar constituents (1991 and Koko *et al.*, 2000). The plant contains high amount of nutritive oil (38.2–54.5%, wt/wt) extracted from Balanites seeds with petroleum ether using soxhlet extraction; the majority of studies focused on that oil and oil from seed of fruits prepared by oil pressing as edible oils (Eromosele *et al.*, 1994 and Mohamed *et al.*, 2002). Unsaturated fatty acids reported to have anticancer and antimutagenic activity (O'Hagan and Menzel, 2003); in addition, fatty acids had antimicrobial activity (Abdelrahman *et al.*, 2003).

In Nigeria, the seed oil obtained from *B. aegyptiaca* has been used especially in the Northpart, as substitute to groundnut oil which is usually relatively expensive. The oil is used for frying food and adding flavor to the food. It is also used to add flavor to tea. This is in addition to medicinal uses such as treatment of skin disease and rheumatism. Despite such wide spread use, there is limited literature on the possible effects of long term consumption of the oil. (Abdel Rahim *et al.*, 1986).

Balanites seed is considered as an extremely useful edible product. It contains good quality oil and high protein content (Mohamed *et al*, 2002 and Abu Al Futuh, 1983). The debittered seed is used as snacks (nuts) by humans., The extracted oil

used for many uses and the remaining cake is used as animal feed (Nour *et al.*, 1985). Both fruits and seed were widely used in many countries during the dry season and drought periods including Nigeria (Lockett *et al.*, 2000), Ethiopia, (Guinand and Lemessa, 2001) and Sudan (Grosskinsky and Gullick, 2001). Schmidt and Joker (2000), Hall and Walker, (1991) and Sayda, (2002) recorted that balanites appears to be highly variable in growth and seed chemical contents. The wide variation under the range in which the tree is found suggests genetic differences between and within locations. Determining this genetic variation is very important for improvement and domestication of this species based on seed parameters.

Dietary exposure of crude *Balanites aegyptiaca* seed oil to rats did not show any toxicological concern but should be used with caution having indicated subtle hepatotoxic effects in the 5% treated group (Wilson *et al.* , 2009).

2.8.1 Chemical composition of *B.aegyptiaca* seed

The seed of *Balanites aegyptiaca* is rich in oil, protein, minerals and edible as snacks after boiling.

2.8.1.1 Fats of *B.aegyptiaca* seed

Importance of lipids in human nutrition and health has been long known. Fats are a major source of energy for the body and aid in vitamin absorption and tissue development. They also play an important role as antioxidants (Anhwange *et al.*, 2004; NAS, 2005). In order for a body to meet its daily nutritional needs while minimising risk of chronic diseases, NAS (2005) recommended that adults should obtain 20 – 35% of their calories/energy from fat. Lohlum (2012) and Elfeel (2010) found *Balanites aegyptiaca* seed oil content 40 and 50 % respectively.

2.8.1.2 Crude Proteins of *B.aegyptiaca* seed

Play an important role in nutrition and diet since they are the major structural components of all body cells. Their function as enzymes, membrane carriers, hormones and provide energy. According to NAS (2005), the recommended daily allowance (RDA) for proteins is 0.8 g kg⁻¹ of body weight for adults and an increased value of 1.1 g kg⁻¹ of body weight for pregnant and breast feeding women. WHO (2007) recommended a slightly higher protein value of 0.83 g kg⁻¹ of body weight which translates to about 33 - 66 g day⁻¹ for adults and about 16.2 – 59.9 g day⁻¹ for boys and girls aged between 4 -18 years. Christian and Ukhun (2006) noted that protein quality and quantity are major concerns in human diets. Protein deficiency causes growth retardation, muscles wasting, oedema, kwashiorkor and abnormal collection of fluids in the body (Anhwange *et al.*, 2004). According to WHO (2004). Lohlum (2012), Elfeel (2010) reported the *Balanites aegyptiaca* seed protein content 37.7 and 37 % respectively.

Lohlum (2012) and Babeker and Fatmah (2013) found *B.aegyptiaca* seed moisture content 3.40 and 3.58% respectively. Babeker (2013)and Lohlum (2012) found *B.aegyptiaca* seed fiber content 9.4 and10.18% respectively. Babeker (2013) and Lohlum (2012) found *B.aegyptiaca* seed ash content 2.9 and 3.98% respectively. Lohlum (2012) and Babeker (2013) found *B.aegyptiaca* seed carbohydrate content 4.74 and 7.72% respectively.

2.8.1.3 Minerals of *B. egyptiaca* seed

Macronutrients are important in human diet because of their various functions in the body. (Christian and Ukhun, 2006).

Sodium is vital for maintaining fluid volume, osmotic equilibrium and acid-base balance. Its deficiency during hot weather is attributed to heavy work in hot

climate (Christian and Ukhun, 2006). Omer (2002), Elfeel (2010) and Lohlum (2012) reported *Balanites aegyptiaca* seed sodium content 0.02, 0.90 and 0.93 mg/100g respectively. Magnesium Functions as a cofactor of many enzymes involved in energy metabolism, protein synthesis, RNA and DNA synthesis and maintenance of electrical potential of nervous tissues and cell membranes (AlGhamdi *et al.*, 1994). Lohlum (2012) , Omer (2002) and Elfeel(2010) reported *Balanites aegyptiaca* seeds content 0.025 , 0.10 and 0.90 mg/100g respectively.

Potassium is very important in the human body where, along with sodium, it regulates the water balance and the acid-base balance in the blood and tissues. In the nerve cells. Lohlum (2012), Omer (2002) and Elfeel (2010) reported that *Balanites aegyptiaca* seeds content 1.09 , 1.88 and 1.95 mg/100g respectively. Calcium required for proper bone and tooth growth; during adolescence, as the bones develop, calcium is again essential to support the growth. Finally, when we get older, our bones tend to get porous and weak, thereby requiring ample calcium intake .Omer (2002), Lohlum (2012) and Elfeel (2010) found calcium content 0.12 , 0.19 and 0.415 mg/100g respectively. phosphorus is required by the body for bone and teeth formation. Calcium alone can't build strong bones and tissues. New research shows calcium needs phosphorus to maximize its bone-strengthening benefits, and taking a lot of calcium supplements without enough phosphorus could be a waste of money .Elfeel (2010), Lohlum (2012) and Omer (2002) found 0.16 , 0.28 and 0.280 mg/100g respectively.

2.8.2 Physico-chemical characteristics of *Balanites. eagyptiaca* seeds oil

Physico-chemical characteristics of any oil are important for determining its nutritional quality and commercial value (Omuja, 2008; Chapagain *et al.*, 2009).

2.8.2.1 Colour

Colour in Balanites oil to presence of carotene. According to FAO/WHO (1994) and WHO (2004), carotenoids and their derivatives are responsible for the yellow colour of fruits, vegetables, cereals and some crude oils. The presence of carotene makes Balanites oil nutritionally important because carotenoids are highly unsaturated polyisoprene hydrocarbons that are lipid and are precursors for vitamin A (WHO, 2004). The light yellow colour of the oil also makes it visually attractive thus, along with other good attributes; this could make Balanites oil a viable and competitive market commodity. Okia (2013), Babagana *et.al.*, (2012) and Babeker (2013) found *Balanites aegyptiaca* oil colour was light yellow. (7,633R.y.b).

2.8.2.2 Refractive index

Is an important attribute of oil quality (Omujaal, 2008). Ecey and Lawrence (1954) defined refractive index as the ratio of the velocity of light in vacuum to the velocity of light in the medium being measured. Schultz *et al.*, (1962), noticed that, the RI of oils and fats were closely related to oxidation products and development of rancidity. It is useful for identification purpose and for establishing purity, and also for observing the progress of reactions, such as catalytic, hydrogenation and isomerization. Babeker (2013) Okia(2013) and Manji(2013) reported RI of *Balanites aegyptiaca* oil were 1.46, 1.47 and 1.48 at 40°C respectively.

2.8.2.3 Viscosity

Is one of the quality parameters of oil. Eugene *et al* (1991) defined the viscosity as the measure of resistance to flow. Viscosity is the measure of the internal friction in the oil and is the important index of the study of oil and their intermolecular forces and its useful criterion for degradation or depolymerization such as that occur in initial stage of hydrolysis of fat and oil during storage (Joslyn, 1971). Okia

(2013) , Babagana *et.al.*, (2011) and Babeker (2013) found viscosity of *Balanites aegyptiaca* oil were 22.60 ,34 and 37 cp at 40°C respectively.

2.8.2.4 Density

Babagana *et.al.*, (2011), Babeker (2013) and Manji (2013) found density of *Balanites aegyptiaca* oil 0.277, 0.92 and 1.001 respectively

2.8.2.5 Free fatty acid

The low %FFA reduces the tendency of the oil to undergo hydrolytic activities. In most oils, the level of free fatty acid which causes deterioration. Babeker (2013), Babagana *et.al.*, (2011) reported that FFA of *Balanites aegyptiaca* oil were 1.84 , 2.8% respectively.

2.8.2.6 Peroxide value

Hydroperoxides are the primary products of lipid oxidation; therefore, determination of peroxide value can be used as an oxidation index for the early stages of lipid oxidation (Amany *et al.*, 2012) . Mohammed *et al.*,(2013) noticed that peroxides are formed as a result of oxidation ,under normal conditions ,these peroxides can break down in to secondary oxidation products usually containing carbonyl group. Oxidation of lipid to hydroxide , referred to as the peroxide value. The change of PV of oils and fats during storage under controlled condition is an important parameter for detecting their quality .Standards showed that the PV of oil should be not more than 10 milliequivalents of peroxide.Manji (2013), Babeker (2013) found PV of *Balanites aegyptiaca* oil were 6.0 , 8 (mgEq/kg) respectively.

2.8.2.7 Acid value

The (AV) of an oil and fat is defined as the number of milligrams of potassium hydroxide required to neutralize the free fatty acids in one gram of the sample. Standards showed that the acid value should be not more than 0.8 mg KOH/g. The AV is measure of the extent to which the glycerides in the oil have been decomposed by lipase action. Babagana *et .al.*, (2011), Okia (2013) and Babeker (2013) reported AV of *Balanites aegyptiaca* were 0.57 , 1.41 and 2.08% respectively.

2.8.2.8 Saponification value

The high saponification value suggests that *Balanites* oil is suitable for soap making.

Babeker (2013) , Manji (2013), Babagana *et.al.*, (2011) and Okia (2013) found saponification value of *Balanites aegyptiaca* were 168.3 ,168.80 ,174.5 and 182.80 mgKOH g⁻¹. respectively.

2.8.2.9 Iodine number

(I N) is the number of milligrams of iodine absorbed by one-gram fat . It is a measure of proportion of unsaturated constituents present in fat . Hence ,it is the halogen addition double bonds of the unsaturated fatty acids and the quantity of halogen take up expressed in terms of iodine as iodine number (Hartley,1967).The iodine number gives an indication of the number double bonds in any particular oil or fat it, however also indicates the total amount of unsaturation .Babagana *et.al.*, (2011), Manji (2013) and Okia (2013) reported 1N of *Balanities egyptiaca* were 56.4 ,76.8 and 98.28 mg I₂/g respectively.

2.8.2.10 Fatty acid composition

Balanites aegyptiaca seed oil as potentially linoleic/oleic oil having good nutritional properties. Thus, *Balanites* kernel oil could be a good source of essential polyunsaturated fatty acids. The fatty acid profile is important for determining the nutritional value of oils (WHO, 2004; NAS, 2005 and Ajayi *et al.*, 2006). The fatty acid composition of *Balanites* oil revealed linoleic acid as the predominant fatty acid. Four major fatty acids in the order linoleic>oleic>stearic>palmitic were found in oils. linoleic acid derivatives serve as structural components of the plasma membrane and as precursors of some metabolic regulatory compounds (Vles and Gottenbos, 1989). The presence of one of the three essential fatty acids in *Balanites* oil makes it nutritionally valuable and highly recommended for human consumption. Babagana *et.al.*, (2011) , Chapagain (2009) and Okia(2013) reported that the main fatty acid composition of *B.aegyptiaca* oil were palmitic acid ,stearic acid , oleic acid ,linoleic acid oil (14.73,14.98,15.40)% , (9.40,19.01,19.01)% , (25.74 ,26.76)% and (37.78 , 39.85 , 75.85)% respectively.

CHAPTER THREE

MATERIALS AND MATHODS

3.1 Material

Two kilograms of *Balanites aegyptiaca* fruit were purchased from local market in Khartoum, Sudan. Fruits were then crushed using a steel hammer and seeds were then obtained. One kilogram of seeds was collected and was then ground using grinding machine. ground seeds were then eventually ready for further analysis.

3.2 Methods

3.2.1 Chemical composition of Seeds

3.2.1.1 Moisture content

The moisture content was determind according to of the Association of Official Analytical Chemists (AOAC,2008).

Two grams were weighed into a pre-dried and tarred dish. Then, the sample was placed into an oven (No.03-822, FN 400, Turkey) at $105 \text{ }^{\circ}\text{C} \pm 1 \text{ }^{\circ}\text{C}$ until a constant weight was obtained. After drying, the covered sample was transferred to a desiccator and cooled to room temperature before reweighing. Triplicate results were obtained for each sample and the mean value was reported to three decimal points according to the following formula;

Calculation:

$$\text{Moisture content (\%)} = \frac{W_1 - W_2}{W_{t1}} \times 100$$

Where;

W1= Sample weight before drying

W2 =Sample weight after drying

Wt1=Initial sample weight

3.2.1.2 Ash content

The ash content was determined according to the method described by Pearson (1981). Five grams were weighed into a pre-heated, cooled, weighed and tarred porcelain crucible and placed into a Muffle furnace (No.20. 301870, Carbolite, England) at 550 to 600 °C until a white gray ash was obtained. The crucible was transferred to a desiccator then allowed to cool to room temperature and weighed. After that, the ash content was calculated as a percentage based on the initial weight of the sample.

Calculation:

$$\text{Ash \%} = \frac{[(\text{Wt of crucible} + \text{Ash}) - (\text{Wt of empty crucible})]}{\text{Initial weight (Wt)}} \times 100$$

3.2.1.3 Fat content

Fat content was determined according to the official method of AOAC (2008).

Five grams were weighed into an extraction thimble and covered with cotton that previously extracted with hexane (No.9-1.6-24/25-29-51, LOBA Cheme, India). Then, the sample and a pre-dried and weighed extraction flask containing about 100 ml hexane were attached to the extraction unit (Electrothermal, England) and the extraction process was conducted for 16 hr. At the end of the extraction period, the flask was disconnected from the unit and the solvent was redistilled. Later, the flask with the remaining crude hexane extract was put in an oven at 105 °C. for 3

hr, cooled to room temperature in a desiccator, reweighed and the dried extract was registered as fat content according to the following formula;

Calculation:

$$\text{Fat content (\%)} = \frac{(W2 - W1)}{W3} \times 100$$

Where;

W2 =Weight of the flask and ether extract

W1 =Weight of the empty flask

W3=initial weight of the sample

3.2.1.4 Crude protein content

The protein content was determined in all samples by micro-Kjeldahl method using a copper sulphate-sodium sulphate catalyst according to the official method of the AOAC (2008).

Two grame sample was accurately weighed and transferred together with 4g Na₂SO₄ of Kjeldahl catalysts (No. 0665, Scharlau chemie, Spain) and 25ml of concentrated sulphuric acid (No.0548111, HDWIC, India) into a Kjeldahl digestion flask. After that, the flask was placed into a Kjeldahl digestion unit (No.4071477, type KI 26, Gerhardt, Germany) for about 2 hours until a colourless digest was obtained and the flask was left to Cool to room temperature.

The distillation of ammonia was carried out into 25ml boric acid (2%) by using 20ml distilled. water and 70ml sodium hydroxide solution (45%). Finally, the distillate was titrated with standard solution of HCl (0.1N) in the presence of 2-3 drops of bromocresol green and methyl red as an indicator until a brown reddish colour was observed.

Calculation:

$$\text{Nitrogen\%} = \frac{\text{Titre volume} \times \text{HCl (N)} \times \text{Nitrogen equivalent weight} \times 100}{\text{Sample weight} \times 1000}$$

$$\text{Crude protein\%} = \text{Nitrogen\%} \times \text{Protein conversion factor (6.25)}$$

3.2.1.5 Crude fibre content

It was determined according to the official method of the AOAC (2008). Two grams of a defatted sample was placed into a conical flask containing 20ml of H₂SO₄ (0.26 N). The flask was then fitted to a condenser and allowed to boil for 30 minutes. At the end of the digestion period, the flask was removed and the digest was filtered (under vacuum) through a porcelain filter crucible (No.3). After that, the precipitate was repeatedly rinsed with distilled boiled water followed by boiling in 20 ml NaOH (0.23 N) solution for 30 min under reflux condenser and the precipitate was filtered, rinsed with hot distilled water, 20ml ethyl alcohol (96%) and 20 ml diethyl ether.

Finally, the crucible was dried at 105 °C (over night) to a constant weight, cooled (in a desiccator), weighed, ashed in a Muffle furnace (No.20. 301870, Carbolite, England) at 550-600 °C until a constant weight was obtained and the difference in weight was considered as crude fibre.

Calculation:

$$\text{Crude fibre \%} = \frac{[\text{Dry residue crucible Cal}] - [\text{Ignited residue} + \text{crucible (g)}] \times 100}{\text{Sample weight}}$$

3.2.1.6 Minerals content

Five grams were weighed into a pre-heated, cooled, weighed and tared porcelain crucible and placed into a muffle furnace (No.20. 301870, Carbolite, England) at 550 to 600 °C until a white grey ash was obtained. Then, the ash content was

cooled and 10 ml of HCl (2.0N) was added to each crucible and placed in a hot sand bath for about 10-15 min. After that, the ash solution of each sample was filtered with ashless filter paper and the filtrate was made up to volume in a volumetric flask (50 ml) with hot distilled water and the concentrations of calcium, magnesium, iron, sodium and zinc were determined using, Atomic Absorption Spectrophotometer (3110-Perkin Elmer. USA).

3.2.1.7 Total and available carbohydrates

Total and available carbohydrates were calculated by difference according to the following equations:

$$\text{Total carbohydrates} = 100 - (\text{Moisture} + \text{Protein} + \text{Fat} + \text{Ash})$$

$$\text{Available carbohydrates} = \text{Total carbohydrates} - \text{Crude fibre.}$$

3.3 Physical characteristics of *B.aegyptiaca* oil

3.3.1 sample preparation

Seed were expressed using the unrefined oil cold pressed method of extraction in the food research center labs in shambat.

3.3.1.1 Colour of oil

The colour intensity was measured using a Lovibond tintometer, units of red, yellow and blue were recorded according to the AOAC (2008). Samples were filtered through a filter paper immediately before testing. Appropriate cell (2 inches cell) was filled with oil and placed in the tintometer placed near by the window for light. The instrument was switched on and looked upon through the eye piece. Slides were adjusted until colour match was obtained. The readings of the filter, used to make the match (red, yellow, and blue).

3.2.1.2 Refractive index of oil

Refractive index was determined by Abbe-60 refractometer as described by AOAC (200) where a double prism was opened by means of screw head and few drops of sample were placed on the prism. The prism was closed firmly by lighting screw head. The instrument was left to stand for few minutes before reading in order to equilibrate the sample temperature with that of the instrument. The prism was cleaned between readings wiping off oil with soft cloth, then with cotton moistened with petroleum ether and left to dry. The refractive indices of all samples were determined at 35 - 40°C.

3.2.1.3 Viscosity of oil

The viscosity of the oil samples under investigation were recorded using Ostwald-U-tube viscometer according to Cocks and Van Rede (1966). The viscometer was suspended in the constant temperature water bath so that the capillary was vertical.

The instrument was exactly filled to the mark at the top of the lower reservoir with the oil by means of a pipette inserted in the side arm, so that the tube wall above the mark is not wetted. The instrument was then left to stand for few minutes before reading in order to equilibrate the sample temperature with that of the instrument (35°C).

By means of pressure on the respective arm of the tube, the oil moved into the other arm so that the meniscus is (1 cm) above the mark at the top of upper reservoir. The liquid was then allowed to flow freely through the tube and the time required for the meniscus to pass from the mark above the upper reservoir to that at the bottom of the upper reserve was recorded.

Wl= weight of water at 40°C.

3.4 Chemical characteristics of *B. aegyptiaca* oil

3.4.1 Acid Value of oil

The acid value was determined According to B.S.I (1984). The oil or melted fat was mixed thoroughly before weighing. About 5 to 10 gm of cooled oil sample was weighted accurately in a 250 ml conical flask and 50 to 100 ml of freshly neutralized hot ethyl alcohol and one ml of phenolphthalein indicator solution were added. The mixture was boiled for about five minutes and titrated while hot against standard alkali solution, shacked vigorously during the titration. The weight of the oil/fat taken for theestimation and the strength of the alkali used for titration shall be such that the volume of alkali required for the titration does not exceed 10 ml.

Calculation

$$\text{Acid value} = \frac{56.1 \times V \times N}{W}$$

Where:

V = Volume in ml of the standard potassium hydroxide or

Sodium hydroxide used

N = Normality of the potassium hydroxide solution or sodium hydroxide solution

W = Weight in g of the sample

3.4.2 Peroxide value of oil

The peroxide value (PV) of oils was determined according to Wail *et al.* (1995). About one gm of the sample was weighed into 250 ml conical flask, 30 ml of a glacial acetic acid/chloroform solution (3:2) were added, and the flask was' swirled until the sample was dissolved. A 0.5 ml of saturated potassium iodide was added. The solution was again swirled for one minute, 30 ml of distilled water were added

and 0.5 ml of 1% starch solution were also added. The contents of the flask were then titrated with 0.1 N sodium thiosulphate added gradually with constant and vigorous shaking and the titration was continued until the blue colour just disappeared. A blank test was carried out. The number of 0.1 N sodium thiosulphate required was recorded.

Calculation:

$$PV = \frac{(V_a - V_p)N \times 100}{W}$$

Where:

V_a= Volume of sodium thiosulphite solution used in titration

V_b= Volume of sodium thiosulphite solution used in blank test

W= Weight of sample in grams

N= Normality of sodium thiosulphate solution

3.4.3 Saponification number of oil

The determination of saponification number was carried out according to the AOAC (2008) method.

One gram of oil sample was weighed accurately in to 200 ml conical flask. 25 ml of 0.1N alcoholic KOH solution was added, and the contents of the flask were boiled under reflux for one hour with frequent rotation. one ml of phenolphthalein indicator was added, while the solution was still hot, and the excess alkali was titrated with 0.5N HCL. The numbers of ml of HCL required (a) were noted. The same process was repeated without oil and the numbers of ml of HCL (b) were also recorded.

Calculation:

$$\text{Saponification Number} = \frac{(b-a) \times 0.02805 \times 1000}{S}$$

Where:

a = ml of HCL from sample.

b = ml of HCL from blank.

S = weight of oil in gram.

3.4.4 Fatty acid compositions of oil

The composition of oil sample was analyzed using a Gas Chromatography (GC) (model GC-2014, Shimadzu, JAPAN). It is equipped with an FID –Flame ionization detector and capillary column (30m (column length) x 0.25 μm (film thickness) x 0.25 mm (internal diameter)). The GC oven was kept at 70°C for 3.0 min, heated at 10 C/min up to 280 °C, where it was kept for 5.0 min, and a total analytical time was 26 min. The carrier gas was helium (1.45 mL/min, column flow, and 33.5mL/min, total flow). 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 (Emil *et.al* 2009).

3.4.5 Iodine value of oil

The iodine value (I V) of the oils which quantifies their unsaturation level was determined according to the B.S.I (1985). Approximately ,0.2 grammes of oil was accurately weighed and placed in a dry and clean flask specially offered for the test .A 10 ml of chloroform was used for dissolving the oil . A 25 ml of pyridine sulphate dibromide solutions was added and finally 20 ml of KI (0.1 N)

were added to the contents of the flask was then stoppered and the mixture was allowed to stand for 10 minutes in a dark place . The stopper and the side of the flask were rinsed with enough amount of distilled water , the contents of the flask was then stoppered and the mixture was allowed to stand for 10 minutes in a dark place .The stopper and the side of the flask were then shaken and titrated against 0.1N sodium thiosulphate solution using starch liquid as indicator . A blank determination was carried out simultaneously .

Calculation:

$$\text{Iodine value (IV)} = (b-a) \times 0.01269 \times 100 / S$$

Where :

b; Volume (ml) of sodium thiosulphate in blank solution

a; Volume (ml)of sodium thiosulphate in test active solution

S: Weight (gm) of the oil sample

3.4.6 Free fatty acids of oil

Free fatty acids content was carried out according to the B. S. I (1985) . About five grammes of the oil was weighed accurately into 250 ml conical flask. Fifty ml mixture of 95% alcohol and ether solvent (1:1) were added. The solution was neutralized after addition of one ml of phenolphthalein indicator . The contents of the flask were then heated with caution until the oil was completely dissolved .The contents of the flask were then titrated with 0.01N KOH with constant shaking until a pink colour persisted for 15 seconds .The number of ml of 0.1 N KOH recorded as % .

Calculation:

$$\text{Free fatty acid as (\% oleic acid)} = a \text{ (ml of KOH) } \times N \times 56.1 / S$$

Where:

a. Reading with sample (ml)

N : Normality of KOH

S : Original weight of sample.

3.3 Statistical analysis

The results were subjected to Statistical Analysis System (SAS) by using One-Factor Analysis of Variance (ANOVA). The Mean values were also tested and separated by using Duncan's Multiple Range Test (DMRT) as described by Steel, *et. al.*, (1997).

CHAPTER FOUR

RESULTS AND DISCUSSION

4.1 Chemical composition of *B. aegyptiaca* seed

Table (4-1) shows the chemical composition of *Balanites aegyptiaca* seed. Moisture, oil, protein, fiber, ash and total carbohydrate contents were found to be 3.10, 42.95, 31.09, 12.64, 3.19 and 3.05% respectively

moisture content was found to be 3.10% which is similar with that reported by both Babeker (2013) and Lohum (2012). Oil content was noticed to be 42.96% which is similar to that reported by Lohlum (2012) but it was lower than that reported by Elfeel (2010) which was 50%.

Protein content was found to be 31.09% which is lower than the 39% reported by both Elfeel (2010) and Lohlum (2012). Fiber content was 12.64% which is higher than the 9.4% reported by Babeker (2013) but it is lower than the 17.18% reported by Lohlum (2010). Ash content was noticed to be 3.19% which is lower than the 3.98% reported by Lohlum (2010) but it is higher than the 2.98% reported by Babeker (2013). Total carbohydrate content was recorded to be 3.05% which is lower than the 7.72% reported by Babeker (2013).

4.2 Minerals content of *B.aegyptiaca* seed:

Table (4-2) shows that the minerals content of *Balanites aegyptica* seed, the potassium, calcium, phosphorus, magnesium, sodium content were found to be 1.09, 0.41, 0.30, 0.13 and 0.09 mg/ 100g respectively.

Table 4. 1: Chemical composition of *B. egyptiaca* seed

Constituent (%)	Mean	Std. Deviation	Minimum	Maximum
Moisture	3.10	0.0300	3.07	3.13
Oil	42.95	0.4843	42.40	43.30
Proten	31.08	0.10263	31.00	31.20
Fibre	12.64	0.05132	12.60	12.70
Ash	3.19	0.05568	3.14	3.25
Carbohydrate	3,05	0.0301	3.06	3.06

Table 4. 2: Minerals content of *B.egyptiaca* seed:

Minerals	Mean	Std. Deviation	Minimum	Maximum
Calcium mg/100g	0.41	0.032	0.390	0.450
Sodium mg/100g	0.09	0.005	0.090	0.099
Magnesium mg/100g	0.13	0.186	0.025	0.350
Phosphor mg/100g	0.30	0.015	0.280	0.310
Potassium mg/100g	1.09	0.015	1.070	1.100

Potassium content was found to be 1,09 mg/100g which is lower than the 1.95,1.88 mg/100g reported by Elfeel (2010) and Omer(2002) respectively but is similar to that 1.9 mg/100g reported by Lohlum (2012).Calcium content was reported to be 0.41mg/100g which is lower than the 1.95,1.88 mg/100g reported by both Elfeel (2010) and Louhlum (2012) respectively but is higher than the 0.12 mg/100g reported by Omer(2002) which was Phosphore.

Content was reproted to be 0,30 mg/100g which is higher than the 0.28, 0.16 and 0.28 mg/100g reported by Elfeel (2010) Lohlum (2012) and Omer (2002) respectively.

Magansium content was recorded to be 0.13mg/100g which is lower than the 0,14 mg/100g reported by Elfeel(2010) but is higher than the 0.02 and 0.10 mg/100g reported by both Lohlum (2012) and Omer (2002) respectively .Sodium content was suggsted to be 0.09 mg/100g which is lower than the 0.90, 0.93 and 0.20 mg/100g reported by Elfeel(2002), Lohlum (2012) and Omer (2002) respectively.

4.3 Physical properties of *B.aegyptiaca* oil

Table (4-3) shows that the physical properties of *Balanites aegyptiaca* oil. The viscosity, density, refractive index and colour were found to be 19.63 , 0.9109 , 1.483 and (7,633R.y.b) respectively.

Viscosity was recorded to be 19.63 which is lower than the 34 , 22.60 and 37cp. reported by Babagana *et.al* (2010), Okia (2013) and Babeker (2013) respectively. Density was noticed to be 0.9109 which its similer to 0.92 reported by Babeker (2013) but is higher than the 0.277 reported by Babagana *et.al* (2011) but is lower than the 1.001 reported by Manji(2013) .Refractive index was found to be 1,483 which is similar to that reported by both Okia (2013) and Manji (2013) but is higher than the 1,46 reported by Babeker (2013) .

Table 4. 3: Physical properties of *B.eagyptiaca* oil:

		Mean	Std. Deviation	Minimum	Maximum
Viscosity [40°C] (cp)		19.63	0.000	19.63	19.63
Density		0.9109	0.9109	0.9109	0.9109
Refractive index [40°C]		1.483	0.0153	1.47	1.50
Colour (degree of colour mixtures)*	R.y.b	0	0	0	0
		7.633	1.097	6.40	8.50
		0	0	0	0

But its higher than the 0.57 (mgKOH/g) reported by Babagana *et.al* (2011).

4.4 Chemical properties of *B.aegyptiaca* oil:

Table (4-4) shows that the chemical properties of *Balanites aegyptiaca* oil the free fatty acid , peroxide value, saponification, iodine value and acid value were found to be 3.17,1.18, 224.63, 122.42 and 1.53 respectively.

Free fatty acid was found to be 3,17% which was higher than the 2.8 and 1.84% reported by both Babagana *et.al* (2011) and Manj (2013) respectively. Peroxide value was noticed to be 1.18 (mgEq/kg) which lower than the 6.0 and 8.0 (mgEq/kg) reported by Manji(2013) and Babeker (2013) respectively. Saponification was reported to be 224,63 (mgKOH/g) which was higher than the 168.80 , 174.5 , 168.3 and 182.80 (mgKOH/g) reported by Manji (2013) ,Babagana gutti (2011) ,Babeker (2013) and Okia (2013) respectively. Iodine was found to be 122.43 mg I₂/g which lower than the 76.8 , 56.4 and 98.28 mg I₂/g reported by Manji(2013), Babagana gutti (2011) and Okia (2013) respectively. Acid value was noticed to be 1.53(mgKOH/g) which is similar to reported by Okia (2013) but its lower than the 2.08 (mgKOH/g) reported by Babeker and Fatmah (2013).

4.5 Fatty acid composition of *B.aegyptiaca* oil

Table (4-5) shows that the fatty acid composition of *Balanites aegyptiaca* the palmitic acid, linoleic acid , Stearic acid and Oleic acid. were found to be 13.37 , 34.36 , 15.03 and 28.57% respectively.

Palmitic acid was recorded to be 13.37% which was lower than the 15.40 and 14.98% reported by both Okia (2013) and Chapagain (2009) respectively. Linoleic acid was found to be 34.36 which is similar to that reported by Chapagain (2009) but its lower than the 39.85 and 75.85% reported by Okia (2013) and Babagana *et.al* (2011) respectively. Stearic acid was noticed to be 15.03 % which lower than the 19.01, 19.1 and 9.40% reported by Okia (2013), Chapagain (2009) and Babagana *et.al* (2011) respectively .Oleic acid was found to be 28.57% which higher than the 25.0 and 26.76% reported by Okia (2013) and Chapagain (2009) respectively.

Table 4. 4: Chemical properties of *B. egyptiaca* oil:

	Mean	Std. Deviation	Minimum	Maximum
Free.Fatty.Acid (%), as oleic acid	3.17	0.00046	3.17	3.18
Peroxid (mgEq/kg)	1.18	0.0400	1.14	1.22
Saponfication.value (mgKOH/g)	224.63	1.5503	223.50	226.40
Iodine valu (mg I ₂ /g)	122.42	0.2000	122.375	122.475
Acid value (mgKOH/g)	1.53	0.000	1.5359	1.5359

Table 4. 5: Fatty acid composition of *B. aegyptiaca* oil

Saturated acid					Un Saturated acid				
Fatty acid	Mean	Std. Deviation	Minimum	Maximum	Fatty acid	Mean	Std. Deviation	Minimum	Maximum
Palmitic acid (%)	13.37	0.1081	13	13.62	Linoleic acid (%)	34.36	0.0541	34.11	34.57
stearic acid (%)	15.03	0.0009	15	15.06	Oleic acid (%)	28.57	0.0562	28.37	28.83
Other Fatty acid composition	6.67	-	-	-	-	-	-	-	-

CHAPTER FIVE

CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

The results indicate that *Balanites* oil yield is high (42.95%) with good physico-chemical properties and rich in polyunsaturated linoleic acid which is an essential fatty acid. The order of fatty acids is linolenic>oleic>stearic>palmitic. Unsaturated fatty acids constituted 62.93% of the oil making it nutritionally beneficial.

The physicochemical characteristics and fatty acid profile of *Balanites aegyptiaca* oil make it a potential raw material for cosmetics, soap and food processing (as edible vegetable oil).

5.2 Recommendations:

- To train and supply the rural communities with the proper equipment and technology for the *B. aegyptiaca* oil production in order to improve their livelihoods through incomes generating money.
- More studies are needed for the possible effects of long term consumption of *Balanites aegyptiaca* oil.

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