

CHARACTERIZATION OF MELON SEED (*Citrillus lanatus*) OIL

BY

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MATRIC. NO 2006/24095EA

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**BEING A FINAL YEAR PROJECT REPORT SUBMITTED IN PARTIAL
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STATE**

FEBRUARY,2012

DECLARATION

I hereby declare that this project work is a record of a research work that was undertaken and written by me. It has not been presented before for any degree or diploma or certificate at any university or institution. Information derived from personal communication, published works were duly referenced in the text.

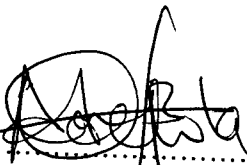
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CERTIFICATION

This is to certify that the project entitled " Characterization of Melon seed (*Citrulus lanatus*) oil" by Bashiru Maude Abdulkadir meets the regulations governing the award of the degree of Bachelor in Engineering (B.ENG) of the Federal University of Technology, Minna and it is approved for its contribution to scientific knowledge and literary presentation.



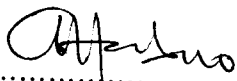
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DEDICATION

This project is dedicated to Almighty Allah

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ABSTRACT

This study was carried out to extract oil from melon seeds using the mechanical and solvent methods of oil extraction. The physical and chemical properties as well as the proximate composition of the extracted oil were determined using the AOAC, (2004) methods. The percentage oil yield was determined for both oils as 42% for solvent and 21.4% for mechanical method of oil extraction. Physicochemical properties determined were as follows: Relative density, Refractive index, specific gravity, colour, Odour, Acid value, Free fatty acid, Peroxide value, Iodine value and Saponification value. The values obtained were 0.9507 and 0.9186, 1.470 and 1.471, 0.965 and 0.9088, Amber for both, Nutty smell for both, 1.48ml/g and 1.70ml/g, 1.04ml/g and 1.64ml/g, 109ml/g and 105ml/g, and 232.53ml/g and 229.45ml/g respectively for solvent and mechanical methods of the extracted oil. The proximate composition determined with the values for the oils were, Moisture, 3.72%, 3.88%, Fats, 42.95%, 42.65%, Crude protein, 27.55%, 28.63%, Crude fibre, 10.90%, 10.65%, Ash, 3.46%, 3.31% and Carbohydrate, 11.43%, 10.88% respectively for solvent and mechanical. The results obtained for both oils were compared and it was discovered that the extraction methods had little or no effect on the physicochemical properties except for the Iodine and saponification values. For the proximate composition determined, the extraction methods also had no effect on it. It was therefore concluded that any extraction methods can be used for the extraction of the oil, except oil that wants to be used for the production of soap, mechanical method of extraction should be used, since it has a higher saponification value.

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CHAPTER ONE

1.0 INTRODUCTION

1.1 Background to the study

Melon (*Citrullus lanatus*) is a cucurbit crop that belongs to the *Cucurbitaceae* family with fibrous and shallow root system. It is a tendril climber or crawling annual crop, mostly grown as a subsidiary crop interplanted with early maize and yam in the savannah belt of Nigeria (Girgis and Said (1968). Melons had their origin from South West India and they belong to the same family as cucumbers, pumpkins, squash, watermelons and bottle gourds. They are differentiated from other melons by their oblong shape and better keeping qualities (Answers.com, 2011).

The *Egusi* melon (*Citrullus lanatus*) is also known as the *Tsamma* melon or wild watermelon and is the biological ancestor of the common watermelon now found worldwide. It is a creeping annual herb with hairy stems and bright yellow flowers. Unlike the common watermelon, whose flesh is sweet and red, the *egusi* melon's juicy flesh is pale yellow or green, and tastes bitter. Its fruits are small and round in the wild, but larger and oval when cultivated. They have smooth pale green skin marked with mottled bands of darker green radiating from the stalk; it is highly adapted to surviving drought and the harsh light of the desert environment (Ogunsua *et al.*; 1989)

Melons are major food crops with several varieties which serve as major food sources (Mabalaha *et al.*; 2007). Their seeds are major soup ingredients and they are used as thickener and flavor component of soups. The seeds are less expensive and widely distributed. They can contribute substantially towards obtaining a balanced diet (Fokou, *et al.*; 2004). Melon seeds are generally a rich source of oil. The oil seeds are generally processed to yield condiments such as 'ogiri'. In West Africa, they are called *egusi* derived from Yoruba language; the seeds are considered a

delicacy. The characteristics and uses of all these seeds are broadly similar. According to Abiodun and Adeleke, (2010) melon seeds are known for their medicinal quality of curing heart diseases. The oil can be extracted using different methods of oil extraction which includes mechanical extraction, solvent extraction, cold press, aqueous, supercritical, steam and high pressure methods of extraction (Microsoft Encarta, 2008).

1.2 Statement of Problem

The derivatives of melon seeds even though numerous still remain unknown to many people in Nigeria. It has been reported by (Abiodun and Adeleke, 2010) that the seed is a medicinal herb and its oil possesses low cholesterol content compared to other oils. The oil with its benefits remains largely unused and untapped, thus despite all its benefits, it has remained largely unreported and underexploited most especially in Nigeria. .

1.3 Objectives of the Study:

The objectives of this project are;

1. To extract oil from melon seeds (*Citrulus Lanatus*) using solvent extraction method
2. To extract oil from melon seeds (*Citrulus Lanatus*) using mechanical extraction method
3. To determine some physical properties of the extracted oils.
4. To determine some chemical properties of the extracted oils.
5. To determine the proximate composition of the oils.
6. To compare the properties of the oil obtained from both methods of extraction

1.4 Justification of Study

Melon seed is very useful because of its numerous and available nutritional properties. The medicinal value of its oil has also been reported. Therefore, research is needed to determine how the extraction methods affect the discussed properties which would of course serve as

available data and an eye opener to many people who may not know the vast benefits of this popular crop even though they consume them.

1.5 Scope of the Study

The study will be limited to the following :Solvent and Mechanical extraction methods; the physical, chemical and proximate compositions to be determined are: odour, color, specific gravity, refractive index relative density, saponification value, acid value, iodine value, peroxide value and free fatty acid, crude fibre, ash, crude protein, carbohydrate and moisture content.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Origin of Melon Seed (*Citrullus Lanatus*)

Human kind has been enjoying melons for more than 4,000 years. Surprisingly, melons have never been found growing in the wild—other than escapees from someone's garden. Melons are believed to have originated in the hot valleys of Southwest Asia, specifically Iran (Persia) and India. Early American settlers grew cultivars of melons back in the 1600s (NGB, 2011). The first documented use of the word "melon" was about 1395. John Ayto's Dictionary of Word Origins suggests that the word is derived from Melos (the Greek Cyclades Islands, best known for the Venus de Milo). In their text, the Mahometans (very early name for the followers of Mohammad) wrote that eating a melon produces a thousand good works (NGB, 2011). All melons are in the same family—*Cucurbitaceae*, the *cucurbit* or gourd family. This large family has more than 100 branches, including cucumbers, gourds, pumpkins, all manner of squash, and even loofahs. "Melons" fall into two genera: *Cucumis* and *Citrullus*. *Cucumis* comprises all melons except for watermelon, which is *Citrullus*—a totally different genus. As a group, all *Cucumis* melons can be called muskmelons or melons (NGB, 2011)

An unusual seed mutant in watermelon (*Citrullus lanatus*) has seeds with a fleshy pericarp, commonly called *egusi* seeds, it is widely cultivated in Nigeria for the high protein and carbohydrate content of the edible seeds. *Egusi* seeds have a thick, fleshy pericarp that appears during the second to third week of fruit development. The *egusi* watermelon is widely cultivated in Nigeria (Anuebunwa 2000; Ezeike and Otten 1989), where the protein and carbohydrate-rich seeds are used as a regular part of the diet. The *egusi* watermelon fruit is not edible because of its bitter, hard, white flesh, and the seeds are often called kernels which are edible (Oyolu, 1977b).

Its seed, which is edible but similarly bitter nutty-flavored, and rich in fat and protein, is eaten whole or used as an oilseed. The oil content of the seeds is 17-19% (w/w), consisting of 67-73% linoleic acid, 10-16% oleic acid, 5-8% stearic acid, and 9-12% palmitic acid. It is estimated that the oil yield is approximately 400 L/hectare. *Agushi*, *agusi* and *egushi* are other names for this fruit. The melon's flesh is bitter and inedible; only the seeds of the fruit are eaten. *Egusi* seeds are high in protein and their flavor is said to be close to that of pumpkin seeds. Each seed is oval shaped and whitish in color with a light tan shell. The shelled seeds are ground for use in Nigerian recipes especially the popular *egusi* soup (Sheri, *et al.*; 2011)

Egusi crops have been overlooked for relatively long time by research and development organizations while they persist in the farming systems of many regions in West Africa Although they looks almost identical to its cousin, the watermelon, they are actually quite different. *Egusi* is filled with very dry, bitter flesh, the seeds are the true delicacy of this melon, composed of nearly 50 percent edible oil and another 30 percent pure protein, these little seeds pack a lot of nutrition into a very small package. In many parts of Africa, where farmers lack access to meat or dairy, the high oil and protein content can make an excellent dietary supplement (Jolaoso *et al.*; 1996).

2.2 Melon Types Case

According to the National Garden Bureau (2011) in the melon fact sheet, there are numerous types of melons available in various regions around the world which includes,

Ananas Melons (a.k.a. Middle Eastern melons) are oval shaped with medium-fine netting over pale green to orange rind. Very sweet, aromatic white flesh. One variety has orange-pink flesh.

Average weight is three to four pounds.

Athena Cantaloupes are Eastern U.S. cantaloupes. They are early maturing, oval-shaped; yellow-orange summer melons with firm, thick, yellow-orange flesh. The skin is slightly sutured with coarse netting. Average weight is 5 to 6 pounds. Left on the vine or harvested, the flesh remains firm.

Canary Melons (a.k.a. Spanish, Juan Canary, Jaune des Canaries, and San Juan canary melons), have bright yellow rinds and an oblong shape. Inside, the pale, cream-colored flesh is juicy, and the flavor is very mild.

Casaba Melons The oval shape with a pointy end, coupled with wrinkled yellow skin sets casabas off from other melons. As does its heft— weighing in at four to seven pounds. The pale, almost white flesh is extremely sweet.

Charentais Melons (a.k.a. French Charentais) are French melons identifiable by their smooth, gray, or gray-blue rinds with sutures and orange flesh.

Christmas Melons (a.k.a. Piel de Sapo and Rochet) have a football shape, weighing upwards of 5 to 8 pounds.

Crenshaw Melons (also seen as cranshaw) are a Casaba cross with a slightly more oblong shape, weighing at least 5 pounds. The slightly wrinkled green rind ripens to yellow. Inside, the flesh is pale peachy orange. It has a strong, spicy aroma.

Galia Melons are Israeli melons that have netted rinds similar to cantaloupes but paler in color.

The sweet pale green to almost white flesh has the consistency of a honeydew with what has been described as a spicy-sweet or banana-like aroma. When ripe, they slip from the vine.

Honeydews (a.k.a. honeydew melon, honey dew melon), second only to "cantaloupes" in popularity, have smooth, white to greenish-white rinds (some may be yellow) and open to reveal refreshingly sweet flesh that may be green, white, or orange. Its texture is similar to a cantaloupe, but the flavor more subtle and sweet.

Muskmelons are the familiar American cantaloupes with orange flesh and netted skin.

Oriental Melons are small (weighing a little more than a pound), elongated yellow melons with white sutures, and sweet, pale peach to white flesh. Because the seeds are so small and the rind is so thin, the entire melon can be eaten.

Persian Melons, bigger than cantaloupes, have a dark green rind with light brown netting. As it ripens, the rind turns to light green. Bright pink-orange flesh has a delicate flavor. Unlike most melons in the Reticulatus group, Persian melons do not slip from the vine when mature.

True Cantaloupe, named for the town of Cantalupo near Rome, Italy has rough-warty (not netted) skin. This is the European cantaloupe, rarely grown in America. Winter Melon- is the catchall name for the long-season, long-keeping (a month or more at room temperature) melons, including crenshaw, casaba, canary and Christmas melons.(NGB, 2011)

2.3 Cultivation

Melons are warm-season fruits, which thrive in temperatures of 70° to 80° F. They prefer slightly acid soil with a pH between 6.0 and 6.5. Melons are thirsty and hungry plants.

2.4 Pests and Diseases of Melon seeds (*Citrullus lanatus*)

Like most other plants, melons are susceptible to a number of pests and diseases, some of which may be more prevalent in one area of the country than another. In the garden, survival of the fittest prevails. If you put a healthy, vigorous melon transplant into rich, well-drained, soil that has been amended with plenty of organic matter, in full sun, with good air circulation, top dress it or fertilize, and provide it with ample water and enough room for the vine to run, the result will be a strong, healthy, well-grown vine, bearing lots of fruit. Take away any of its necessities and the resulting plant will be weaker and/or stressed. A healthy plant is not going to attract pests and diseases; a weak one will. Other diseases that affect melon seeds includes; Bacterial wilt, Anthracnose, Angular leaf spot, powdery mildew and mosaic.(Arthur,1991)

The nutritional and mineral contents of melon seed reported by USDA, (2010) are shown in Table 2.1 and 2.2 respectively;

Table 2.1: Proximate Composition of Melon Seed

COMPOSITION	PERCENTAGE
Protein	33.08
Fat	45.21
Moisture	4.88
Ash	3.75
Crude fiber	2.03
Carbohydrate	10.33

Source: USDA, (2010)

Table 2.2: Mineral composition of Melon seed Oil

COMPOSITION	PERCENTAGE
Calcium (Ca)	2.03
Magnesium (Mg)	8.57
Potassium (K)	5.14
Sodium (Na)	79.24
Manganese (Mn)	66.25
Iron (Fe)	21.43
Copper (Cu)	3.28
Zinc (Zn)	11.66

Source: USDA, (2010)

2.5 Socioeconomic role of *Egusi* crops

Farming in sub-Saharan Africa is characterized by semi-subsistence, low-input, low-productivity farming systems (Govere et al., 2003; Gray, 2005). *Egusi* farming systems reflect similar overview with objectives such as income generation, diversification, household food security, social relationships and seed provision for the next cropping season. However, these objectives are differently prioritized from one socio-cultural region to another. Basically, in most regions the crop is primarily cultivated to provide food for the households. However, in the central region the crop is primarily cultivated to insure cash income. Utilizations of *Egusi* crops are different from one sociolinguistic group to another and could represent a plinth on which a sustainable valuation of *Egusi* crops could be developed. This was recognized and reported by Burkill (1985) who wrote about the traditional uses of *C. lanatus* in Yoruba community: “ A Yoruba food or flavored called “ *oguri* ” is made from the fermented kernels; “ *igbalo* ” is another food made from the seeds Roasted, pounded, wrapped in a leaf and then boiled ”.(Jason. D, 2011)

2.6 Uses of *Egusi* (*citrillus lanatus*)

While the seeds are often shelled and eaten individually as a snack, many processed forms of the seeds have made their way into common cooking practices. After soaking, fermenting, or boiling, the seeds take on different flavors and are frequently added to thicken soups and stews. On their own the seeds can also be roasted and ground into a spread like peanut butter. With further preparation, *egusi*-seed meal can be pressed into patties to be used like a meat substitute, and its oil can be used for cooking. The plant is also easy to grow. It is extremely resilient to pests and diseases and because it blankets the ground as it grows, it can help suppress weeds. Because of this, farmers often intercrop *egusi* with other crops, including sorghum, cassava, coffee, cotton, maize, or bananas. Mature *egusi* melons can also remain in the field for a long

time without rotting, so crop loss and waste is rare. And once the seeds are harvested, they can be a reliable year-round food source because they store well.(Elhussein and ziyada,2008)

The *egusi* can also be an important supplementary baby food, helping to prevent malnutrition. Blending the seeds with water and honey produces a milky liquid that can be used as formula if breast milk is unavailable; making the plant as diverse in its uses as it is easy to grow.

Considerable amounts of oil-rich cucurbit seeds are available in Sudan. They are believed to produce edible oils, but these seeds are not currently exploited as oil sources on a large scale. They are either completely consumed or exported to near-by countries. Recently, several investigations have been carried out on many cucurbit seeds to exploit them as unconventional new sources of oil.

Egusi seed is one of the species which is available in a considerable amount in Sudan (Elhussein, *et al.*; 1994). It is an ancestor type of the cultivated watermelon. It is locally known as “*Gurum*” and is semi-cultivated in the beach of the Nile River in the north of Sudan. The green parts of the plant are used as animal feeds, the seeds are used as a masticatory article and the residue is used as a source of heat energy for cooking (Nadkarni, 1954). Due to the growth of the importance of oilseeds in the national economy of Sudan, the high demand to look for a new source of oil and the sufficient availability of this plant in Sudan has led to its investigation as a new source of vegetable oil (Microsoft Encarta, 2008)

Sociocultural roles played by *Egusi* crops are important in local communities. According to farmers, three main uses are ensured by the household: cash income, household food, the gift to relatives and friends, which indicates that in the Southern and Central regions of Nigeria farmers sell 50 to 70% of their harvest product for cash income. In central region less than 20% of *Egusi* product is allocated for households' consumption (Enoch, *et al.*; 2011).

Few farmers also reported the medicinal role of some *Egusi* species such as *C. lanatus*. The sliced young fruit of this specie is said to heal stomach aches while the seed coat in decoction with Eucalyptus (*Eucalyptus camaldulensis* Dehnh.) roots is a sedative for epilepsy. The roasted seed, ground with salt is taken with warm water or porridge to prevent vomiting. Also used as an alternative to using synthetic anti-diabetic to help control blood glucose (Yibchok-Anun et al., 2006). In pre-modern medicine it was an ingredient in the electuary called *confectio hamech*, or *diacatholicon*, and most other laxative pills; and in such cases as required purging, it was very successful. It is one of the most violent purgative drugs known; insomuch that it excoriates the passages to such a degree as to sometimes draw blood, and induce a so-called "superpurgation". Sometimes, it was taken boiled in water, or beer, in obstruction of the menses, which was considered successful in strong constitutions. Some women used it in the same manner, in the beginning of pregnancy, to cause an abortion, which often occurred due to the violence of its operation (Riddle, 1999).

2.7 Other Beneficial effects of Melon Seeds

The fruit, stems, leaves and roots of melon have all been used in traditional medicine to help treat ailments such as hyperlipidemia, digestive disorders, microbial infections and menstrual problems (Yibchok-Anun *et al.*, 2006;) it has been shown to possess powerful antiviral properties that can stimulate the immune system and activate the body's natural killer cells (Grover and Yadav, 2004) to help fight off viruses such as herpes simplex virus 1 and human immunodeficiency virus 1 (Raman and Lau, 1996)

Studies have also shown that it has anti-carcinogenic properties and can be used as a cytotoxic agent against many types of cancer (Grover and Yadav, 2004). Extract can also be used as a broad-spectrum antibacterial agent to fight off infections caused by *E.coli*, *Salmonella*, *S. aureus*, *Staphylococcus*, *Pseudomonas*, and *Streptobaccilus* (Saeed and Tariq, 2005). In addition,

the plant possesses anti-helminthic properties, which are effective in the treatment of malaria.

Based on the multitude of medical conditions that bitter melon can treat, scientists are more and more interested in studying its bioactive compounds and their actions on the body. However, as many studies report, there has been substantial emphasis on the anti-diabetic compounds and their hypoglycemic properties (Harinantenaina *et al.*,2006).

2.8 Relationship between Melon seed oil and other Oil seeds

Oils and fats are substances of vegetable or animal origin. They are insoluble in water and greasy to touch. The most important characteristic is that they have a caloric content more than twice as high as the other food stuff (9 kcal g⁻¹).¹ Also they act as lubricants during mixing of ingredients and as media for heat transfer carrier for fat soluble vitamins. Also, they are a source of essential fatty acids (Charley, 1982).

The high world demands for oils and fats to meet the multiplex human consumption and the multitudinous industrial needs are the reasons for the increase in the importance of oil seeds and make them play an important role in the national economy of the producing countries.

To ensure the increase of oils and fats, it is necessary to continue not only with the development of new varieties with improved oil yields, but also to search for new sources of oil (Fadala, 1990). Melon seeds like other oil producing seeds are characteristically related in different ways, the relationship can be seen in the different seeds mentioned below which includes; neem seeds and castor seeds.

2.8.1 Neem seed oil (*Azadirachia indica*)

The neem seed is one of the very few trees known in the Indian subcontinent, it is also found in Indonesia in several areas such as Bali, Lombok, West java and Nusa Tenrrara Barat. It grows on most kinds of soils and thrives better than most other plants on dry, stony and shallow soils (Dianne, 2006).

The neem seed is a part of the neem tree which has high concentration of oil (Ikasari and Indraswati, 2008). The seed contains approximately 45% oil which contains Ioeic acid (50-60%), palmitic acid (13-15%), stearic acid (14-19%), linolieic acid (8-16%) and arachidic acid (1-3%). The oil is brownish yellow, non-drying oil with an acrid tasted and unpleasant odor. There are various methods of oil extraction from the neem seed, but a most efficient way has been the solvent extraction method using the soxhlet apparatus for the process. The oil is widely used for insecticides, lubricants, drugs for a variety of diseases. A number of bitter components such as; nimbin (0.12%), nimbinin (0.01%), nimbidin (1.4%) and nimbidiol (0.5%), have been identified in neem oil. There are also pigments, polysaccharides, salts and the proteinaceous material which makes up the cellular matrix of the seed (Johnson and Morgan,1997). The oil is also reported to have several other biological properties such as anti-microbial, anti-fungal and ant-viral effects. An investigation of the active principle of the oil was considered by pongamphia *et al* (2004) as part of a general scheme of research for establishing its industrial uses.

2.8.2 Castor seed oil (*Ricinus communis*)

Castor seed originated in Africa and grows wild in East and North Africa. Castor seeds require warm climate and is killed by frost. It can be grown over a wide altitude range in the tropics and with both low and medium rain fall. The best soil for cultivation is rich well drained sandy or clayey loam. The castor varies greatly in its growth habits, color of foliage, stems, seed size, color and oil content. Castor seed contains between 40% and 60% oil that is rich in triglycerides mainly ricinoleic. The proximate composition of castor ranges as follows; oil 45-51.8%, moisture 3.1%, protein 12-16%, and carbohydrate 3.1-7%, fiber 23.1-27.2%, ash 2-2.2%. Castor oil triglyceride is unique in that its major fatty acid is the unsaturated, hydroxylelated 12-hydroxy 2-octadecanoic acid, also known as ricinoleic acid. The fatty composition of typical castor oil is palmitic acid 2%, stearic acid 1%, oleic acid and linoleic acid 3%, ricinoleic acid 87%.(Microsoft Encater, 2009).

The oil from castor seeds is extracted by variety of processes or combination of processes such as hydraulic presses, continuous screw presses and solvent extraction. The most satisfactory approach is by hot pressing using a hydraulic press, followed by solvent extraction to remove the bulk of oil remaining in the press cake. Hot pressing by hydraulic press extracts between 75-85% of oil contained in the castor seed while the remaining press cake has about 12% oil content (Wikipedia, 2007). The oil extracted from the hydraulic screw process is filtered and collected in a settling tank; the resulting cake contains 8-10% oil. It is crushed into coarse meal and subjected to solvent extraction with hexane or heptane. The soxhlet apparatus is used for solvent extraction of castor oil from the seeds. After extraction, the solvent is removed by distillation and their resulting oil is processed in similar manner as oil from the pressing step. The castor oil can be used for medicines, lubricants, cosmetics, coatings and disinfectants (Wikipedia, 2008).

2.9 Methods of Oil extraction

Many oil-bearing seeds and nuts are broken up by grinding, flaking, or rolling, then subjected to mechanical pressing to liberate the oil. The methods of extraction is likely to affect the composition of oil, since the method used such as pressing (expelling) or solvent extraction are unlikely to remove exactly the same mix of components in the same proportions. The oil yield that can be obtained from melon seeds also varies widely in literature from 45 - 50% (Oyolu, 1977). The oil can be obtained through pressing (grinding) of the seed kernels by cold pressing or through a process incorporating temperature controls. The oil is also extracted by traditional kneading and alternate wetting with hot water until the oil in the dough-material begins to ooze out (Olaifa and Adenuga, 1998).

2.9.1 Mechanical Extraction Method

The NIFOR mechanical screw-press is the latest used by the small-scale oil processing industry in Nigeria. This consists of a perforated tube inside which a transport screw rotates. The pitch of the screw flights gradually decreases towards the discharge end to increase the pressure on the pulp as it is carried through the barrel (Fellows, 1996). The press outlet is more or less closed by a cone that regulates the pressing pressure. The warm transports and gradually compresses the macerated seeds. Released oil drains through the perforations in the tube. The press is mounted directly below a feed conveyor, which is fed by gravity by the horizontal digester. The body of the feed conveyor is perforated to allow oil released in the digester to drain away.

2.9.2 Solvent Extraction Method

The Soxhlet apparatus is used for the solvent extraction of oil. The operation of the Soxhlet apparatus is mainly on the use of appropriate solvent to extract oil from oil producing seeds. This apparatus was designed by Franz von Soxhlet in 1893, firstly with automated extraction apparatus and in later years, several modifications of the apparatus were developed. Solvent extracts are prepared by treating a solid or liquid with a solvent that will dissolve the desired components selectively. For example, the vanilla flavoring is produced by using a solvent to dissolve and separate the compounds that produce the vanilla flavor and aroma from the vanilla seed pods. The process called solvent extraction is widely used in the commercial production of plants and animal by-products. In the Soxhlet apparatus extraction, the solvents used must meet several requirements, they must dissolve the maximum amount of desired components and minimum of undesired materials. The solution must also be easy to separate from the un-dissolved substance. In the extraction of solids, the separation is usually not difficult, but in liquid extraction, a solvent must be chosen that will separate quickly and completely from the liquid being extracted. The final requirement is that the solvent must be easy to separate from the extracted material, usually by distillation without affecting the quality of the product (Microsoft Encarta.2010).

2.9.3 Supercritical extraction Method

This process utilizes Carbon dioxide at critical temperatures and pressures to extract the active ingredients of the melon seeds without the usual high temperature or harmful chemicals. The result is far more concentrated extracts which resembles the herb more closely. Supercritical extracts are superior for many reasons. Beneficial phytochemicals are easily damaged by heat and there is a growing desire for alcohol and solvent free natural extracts.

The supercritical extracts process enables delivery of a broad spectrum of photochemical including both lipophobia (water soluble) and lipophilic (oil soluble) isolates. In addition, this extraction process uses only carbon dioxide as solvent which once the pressure is let off evaporates completely from the extract leaving it totally pure and free of any solvent residues. Then supercritical point is the exact temperature and pressure at which a gas becomes a liquid (Wikipedia, 2007). However, it is only recently that commercial process application of supercritical fluid extraction has been extensively examined (Grandison and Lewis, 1996).

2.9.4 Cold press method

This method of extracting oil is mostly used by the leading manufacturers, though more expensive than the other methods (Wikipedia, 2007).

2.9.5 Aqueous method of Extraction

Aqueous method of extraction is generally reach in water soluble amino acid, pigment, soluble butters and carbohydrates (Wikipedia, 2007).

2.9.6 Steam and High Pressure Method of Extraction:

This method makes use of high pressure to squeeze out oil from seed. Seeds are heated in steam and under high pressure to enabling maximum extraction of oil. This method is not very good as most of the active ingredients and compounds are destroyed by high temperature (Microsoft Encarta, 2008).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1. MATERIALS

3.1.1 Sample Collection and Preparation of samples

The melon seeds that served as the samples for this study were obtained from Bosso market of Bosso Local Government Area of Niger state, Nigeria. The seeds were thoroughly screened so as to remove foreign materials and impurities. Samples of the seeds are shown in Plate 3.1.

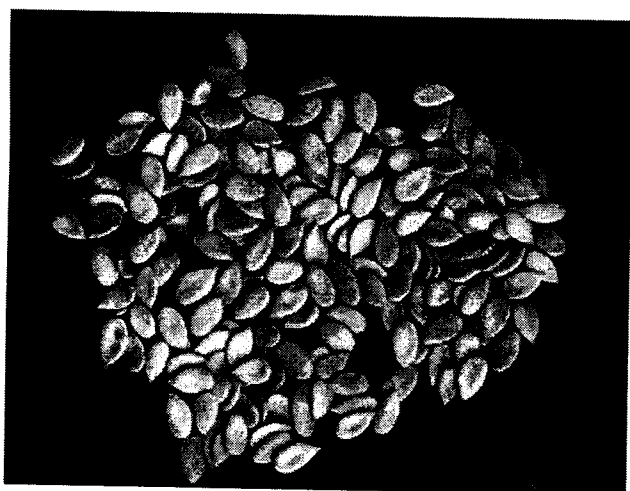


Plate 3.1 Melon seeds

3.1.2 Apparatus used for the Extraction of Melon seed Oil

The following apparatus were used for the study.

Sox let Apparatus (Pyrex England)

Set of muslin cloth

Hydraulic Screw press (Apex 438510 model S15145 by Dor Kent)

Thomas Wiley Milling Machine (Model ED-5)

Test tubes (Pyrex England)

Electrical oven (Gallen Kamp furnace)

Electronic hot plate (Gallen Kamp)

Petri dish

Refractometer

Silica dish

Spatula

Thimbles

Beaker (Pyrex England)

Flame photometer

Reflux condenser

Kjeldahl digestion block (Gerhardt)

Weighing balance (analytical balance machine by Salter, model 250, sensitivity ± 0.005)

Pipette

Desiccators

Water bath (Surgien field instrument (England))

3.1.4 Reagents

The following reagents were used during the determination of the chemical composition of the extracted melon seed oil

Acetic acid

Alcoholic potassium Hydroxide solution

Aqueous Potassium Iodide solution

Carbon tetrachloride

Chloroform solution

Distilled water

Hydrochloric acid

Petroleum ether

Phenolphthalein indicator

Potassium Hydroxide (KOH) solution

Saturated potassium Iodide solution

Sodium-Thiosulphate solution

Sodium Hydroxide (NaOH) solution

Starch indicator

Tetraoxosulphate(VI) acid

3.2 Methods

3.2.1 Solvent Extraction Method

The seeds were cleaned by the use of the Winnowing principle and blowing off the chaff with a tray so as to remove the foreign materials from the seeds. The seeds were then crushed into powder using the Thomas Willey milling machine then sieved (0.5mm sieve) Two liters of normal hexane was added to soak and covered for twenty four (24) hours. Five grams of grinded sample were weighed into a clean thimble and plugged with cotton wool. The Soxhlet apparatus was then assembled and 60ml of petroleum ether was then added to the flask and placed on water bath 40-70⁰c and allowed to reflux for six hours the heat source was then adjusted so that the solvent boils gently and left to siphon over 6 hours, finally the petroleum ether after being siphoned over the barrel, the condenser was detached and the thimble removed the flask containing the oil was then put in the oven and at 100⁰C for 5 minutes so that the petroleum ether can evaporate leaving the oil.

3.2.2 Mechanical Extraction

The seeds were cleaned by Winnowing principle and blowing off the chaff with a tray so as to remove the foreign materials from the seeds. The cleaned seeds were then weighed and oven dried to a moisture content of 3.38%. The oven dried seeds were then put into the Thomas Willey milling machine and grinded. The crushed samples were put inside a muslin bag tied and fed into the hydraulic screw press and pressed to expel the oil from the seeds, the extracted oil from the seed was then collected.

3.3 Determination of the Physicochemical Properties

The determination of the physicochemical properties of the melon seed oil which includes; color, odour, refractive index, specific gravity acid value, free fatty acid, iodine value, peroxide value, saponification value, were carried out using the methods of AOAC (2004). The percentage oil content was determined as described by AOAC (2004). The physicochemical properties of the oil and the percentage yield were determined for oil obtained from the two methods of extraction.

3.3.1 Determination of Refractive Index

The refractive index was determined by setting up the Abbes' refractometer with a light compensator. A drop of the oil sample was applied on the lower prism of the instrument and closed. Light was passed by means of the angled mirror and the reflected light appeared in form of a dark background. The fine adjustment was used to move the telescope tubes until the black shadow appeared central in the cross wire indicator. The control knob was adjusted and readings were recorded and obtained. This is the angle at which a beam of light is bent when passing through a thin film of melted fat or oil. To determine the Abbes' refractometer and sodium vapour lamp, the following vapor temperatures are employed; 20°C for oil, 40°C for solid fat which are fully melted at that temperature, 60°C for hydrogenated fat and 80% for waxes. Equations for correction readings for temperature are given by the International Unit of Pure and Applied Chemistry (IUPAC 1987).

3.3.2 Determination of Specific Gravity

Specific gravity bottle OF 50ml was thoroughly washed with detergent, water and petroleum ether, dried and then weighed. The bottle was filled with water and weighed. The bottle was dried, filled with the oil sample and then weighed. The same procedure was carried out for both oils.

$$\text{Specific gravity} = \frac{\text{Weight of Xml of oil}}{\text{weight of Xml of water}}$$

$$\text{Density} = \frac{\text{weight of oil}}{\text{volume of oil}}$$

3.3.3 Determination of Colour and Odour

The colour and odour of the produced oil was determined through physical observation, that is through smelling and visual observations.

3.3.4 Determination of Acid Value

The acid value is the number of milligram of potassium hydroxide required to neutralize one gram of a sample. The acid value measures the extent to which the glycerides in the oil have been decomposed by lipase action. To determine the acid value of the oil, the following procedure was carried out; 25ml diethyl ether with 25ml alcohol and 1ml phenolphthalein solution were mixed and carefully neutralized with 0.1M sodium hydroxide NaOH. Two grams of the oil was dissolved in the mixed neutral solvent and titrated with aqueous 0.1M NaOH shaking constantly until a pink colour of the solution persisted for 30 seconds. This procedure was carried out for both oils extracted.

$$\text{Acid Value} = \frac{\text{Titration (ml)} \times 5.61}{\text{weight of sample}}$$

$$\text{Acid value} = \text{FFA} \times 2$$

3.3.5 Determination of Saponification Value

The saponification value is the number of milligram of potassium hydroxide required to neutralize the fatty acids resulting from the complete hydrolysis of 1g of the sample. 2g of the sample was weighed into a conical flask, 25ml of alcoholic potassium hydroxide solution was then added. A reflux condenser was attached to the flask containing the mixture which was constantly stirred and was allowed to boil gently for 60mins. Few drops of phenolphthalein indicator was added to the warm solution and then titrated to 0.5M hydrochloric acid to the end point until a pink colour of the indicator just disappeared. The same procedure was used for other samples and blank and was carried out for both oils.

$$\text{Saponification Value} = \frac{(b-a) \times 28.05}{\text{weight of sample}}$$

3.3.6 Determination of Free Fatty Acid (FFA)

Rancidity is usually accompanied by free fatty acid formation; the determination is often used as a general indication of the condition and edibility of oils. The FFA figure is usually calculated as oleic acid 25ml of alcohol and 1ml of phenolphthalein was mixed to neutralize 0.1M NaOH. 2g of the oil was dissolved into the mixed neutral solvent and titrated with aqueous 0.1M NaOH shaking constantly until a pink colour was obtained after twenty seconds. The same procedure was carried out for both oils.

$$(1\text{ml } 0.1\text{M sodium hydroxide} = 0.0282\text{g oleic acid}) = 0.0282\text{g}$$

$$\text{FFA Value} = \frac{\text{Titre value} \times 0.0282 \times 100}{\text{weight of sample}}$$

3.3.7 Determination of Peroxide Value

Fats/oils undergo changes during storage which results in production of an unpleasant taste and odour which is commonly referred to as rancidity. The peroxide value of oil is the measure of its oxygen, this is used to monitor the development of rancidity through the evaluation of the quantity of peroxide in the product. The test is a volumetric one where I_2 formed from potassium iodide in the presence of peroxide, is titrated with thiosulphate. This means $\text{meq}_{\text{peroxide}} = \text{meq}_{\text{thiosulphate}}$ at the equivalence point. 1g of the sample was weighed into a 250ml Erlenmeyer flask, 30ml acetic acid, chloroform solution (3:2) was added under a fume hood and swirled to dissolve the oil. 0.5ml iodide solution was added and swirled for one minute. 1ml of indicator was also added and titrated using starch. The same procedure was performed for blank at the same time. This procedure was carried out for the solvent extracted oil and for the mechanically extracted oil.

$$\text{Peroxide Value} = \frac{S - B \times 0.1 \times 1000}{\text{weight of sample}}$$

3.3.8 Determination of Iodine Value

Iodine measure the degree of unsaturation in oil. Iodine value is the weight of iodine absorbed by 100 parts by weight of the sample. 0.4g of the sample was weighed into a conical flask and 20ml of carbon tetrachloride was added to dissolve the oil. 25ml of wiji's solution was added to the flask using a safety pipette in fume chamber. A stopper was inserted and the content of the flask was vigorously swirled. The flask was placed in the flask for 30 minutes. At the end of this period, 20ml of 10% aqueous potassium iodide and 100ml water were added, using a measuring cylinder and titrated with 0.1M of sodium thiosulphate solution using starch as indicator until the yellow colour almost disappeared. The same procedure was also carried for blank at the same time commencing with 10ml of carbon tetrachloride. This procedure was carried out for both oils.

$$\text{Iodine Value} = \frac{(b-a) \times 1.269}{\text{weight of sample}}$$

3.3.9 Determination of percentage oil yield

The percentage oil yield was gotten by subtracting the final weight of the sample (cake) from the initial weight of the sample. It was calculated as follows;

$$\text{Percentage oil yield} = \frac{\text{initial weight} - \text{final weight}}{\text{initial weight}} \times 100$$

3.3.10 Determination of Proximate Compositions

The proximate composition of the melon seed oil for moisture content, crude protein, crude fibre, carbohydrate and ash were determined using the AOAC (2004) methods.

CHAPTER FOUR

4.0 RESULTS AND DISCUSSION OF RESULTS

4.1 Results

The results obtained from the experimental work are presented on tables 4.1 to 4.4 respectively

4.1.1 Physicochemical properties for Solvent Extracted Melon seed oil

The results gotten for the solvent extracted melon seed oil are shown on Table 4.1 below

Table 4.1: Results for Physicochemical Properties of solvent extracted melon seed oil

Parameters	Values
Relative density	0.9507kg/m ² ± 0
Refractive index	1.470 ± 0
Specific gravity	0.965 ± 0
Color	Amber
Odour	Nutty smell
Acid value	1.48ml/g ± 0.53
Free fatty acid	0.74 ml/g ± 0.0008
Peroxide value	1.04ml/g ± 0.0009
Iodine value	109.33ml/g ± 0.0002
Saponification value	232.53ml/g ± 15.36
Oil yield	42%

4.1.2 Physicochemical properties of Mechanically Extracted Melon seed oil

The results gotten for the mechanically extracted melon seed oil are shown on Table 4.2 below

Table 4.2: Results for Physicochemical Properties of Mechanically Extracted Melon seed oil

Parameters	Values
Relative density	0.9186kg/m ² ± 0
Refractive index	1.471 ± 0
Specific gravity	0.9088 ± 0
Color	Amber
Odour	Nutty smell
Acid value	1.70ml/g ± 0.0008
Free fatty acid	0.85 ml/g ± 0.0016
Peroxide value	1.64ml/g ± 0
Iodine value	105.10ml/g ± 0
Saponification value	229.45ml/g ± 2.59
Oil yield	21.40%

4.1.3 Proximate Composition of Solvent Extracted Melon Seed oil

The result for the proximate composition of the solvent extracted melon seed oil is shown on Table 4.3

Table 4.3: Results for Proximate Composition of Solvent Extracted Melon seed oil

Parameters	Mean Values %
Moisture Content	3.72 ± 0.0004
Fats	42.95 ± 0.065
Crude Protein	27.55 ± 0.36
Crude Fibre	10.90 ± 0.04
Ash	3.46 ± 0.006
Carbohydrate	11.43 ± 0.123

4.1.4 Proximate Composition of Mechanically Extracted Melon Seed oil

The result for the proximate composition of the solvent extracted melon seed oil is shown on Table 4.4

Table 4.4: Results for Proximate Composition of Mechanically Extracted Melon oil

Parameters	Mean Values %
Moisture Content	3.88 ± 0.053
Fats	42.65 ± 0.057
Crude Protein	28.63 ± 0.4225
Crude Fibre	10.65 ± 0.01
Ash	3.31 ± 0.0004
Carbohydrate	10.88 ± 0.01

4.2 DISCUSSION OF RESULTS

4.2.1 Physicochemical Properties of Solvent Extraction Method

The oil had an amber colour and a very characteristic nutty flavour. The refractive index was obtained to be 1.470 at 20°C, this shows the angle through which a beam of light is bent when passing through the oil and is very applicable at different temperatures, this result is similar to the 1.466 for pumpkin seed oil reported by John *et al.*; (1997). The specific gravity of the oil was gotten as 0.965 which is similar to the 0.924 reported by Mirjana, *et al.*; (2005) for melon seed oil. The iodine value was determined to be 109.33ml/g which is in agreement with the 108.8mgI/g gotten for Ammon seeds by Salunke *et al.*; (1992). Peroxide and saponification values were gotten respectively as 1.04mgI/g and 232.53gKOH/g, this results are slightly greater than the 1.01mgI/g and 118Gkoh/g gotten for grape seeds as reported by Basil, *et al.*; (1985), This is the quality of the oil which qualifies its use in soap production.

The acid value as well as the free fatty acid were obtained to be 1.48mgKOH/g and 0.74mgKOH/g, and 1.45mgKOH/g and 0.70mgKOH/g were gotten for pumpkin seeds as reported by John *et al.*; (1997). The moisture content for the melon seed oil was obtained as 3.72%, which is in line with the 3.26% gotten for Chinese kernel oil as reported by Olaofe, *et al.*; (2009). Crude protein was gotten as 27.55%, this value is slightly greater than the 21.75% gotten for Castor seed oil as reported by Okorie, *et al.*; (1985). Crude fibre and the ash contents were obtained as 10.90% and 3.46% respectively, this values are in line with the 9.65% and 4.97% gotten for Chinese kernel as reported by Olaofe, *et al.*; (2009). Fats for the melon seeds were obtained to be 42.95% but for that of castor seeds oils were obtained to be 51% as reported by Devendra, (1988). Finally, the Carbohydrate content for the Melon seed oil was determined to be 11.43% and 10.33% was gotten for melon seed oil by Mabalaha, *et al.*; (2007). The percentage

yield of the oil for the soxhlet extraction was found to be 42%, these and the initial values gotten for Melon seed was 45% by Fokou *et al.*; (2004)

4.2.2 Physico chemical Properties Of Mechanical Extraction Method

The percentage yield of the oil for the mechanical extraction of the oil was found to be 21.40%, this is in line with the 20.1 – 54% range for melon seed oil yield as reported by Mirjana, *et al.*; (2005), Mabalaha *et al.* (2007) also reported oil yields of seeds ranging from 24.8-30.0% in *Citrillus lanatus* and *C. colocynth* species respectively while Madaan and Lai (1984) recorded oil content values of 41.0-56.6% in melon seeds. The refractive index was obtained to be 1.471 at 20°C, this shows the angle through which a beam of light is bent when passing through the oil and is very applicable at different temperatures, this result agrees with the 1.465 gotten for Olive oil as reported by Kiritsakis, (1989). The specific gravity of the oil was gotten as 0.9088, this value is in line with the 0.910 gotten by Bemis, *et al.*; (1968) for melon seed oil. The colour and odour for the oil were gotten as Amber and groundnut smell respectively. The colour and smell as compared to the IUPAC (1976) chattered standard were found to be amber and nutty smell respectively.

The acid value was found to be 1.70mgKOH/g and the free fatty acid was also gotten as 0.85mgKOH/g. this values agrees with the 2.11mgKOH/g and 1.03mgKOH/g respectively gotten for Acid and free fatty acid content for grape seeds as reported by kermel, *et al.*; (1985). The iodine value was gotten to be 105.10ml/g; this is comparable to the values earlier obtained for Arachis oil and Cottonseed oil, 106.12mgI/g and 94.78mgI/g respectively (Pearson, 1981). The saponification value was found to be 229.45mgKOH/ which is in line with the 220.52mgKOH/g for *Leganaria siceraria*, Specie of the melon seed (Budifu, *et al.*; (1991). The peroxide value was found to be 1.64mEq/kg. One should note that a rancid taste begins to be noticeable when the peroxide value is between 20 and 40mEq/kg. this result is in line with 1.70mEq/kg reported

for melon seed by Paris, (2001). The moisture content, 3.88% was close to that of *Leganaria siceraria*, Specie of the melon seed (Budifu, *et al.*; (1991). Crude protein was gotten as 28.63% which is in line with the 39.0% gotten for Chinese Kernel as reported by Olaofe, *et al.*; (2002) Crude fibre 10.65% and Ash, 3.31% were obtained from the study which was compared to the values obtained for *Leganaria siceraria*, Specie of the melon seed to be exactly, 10.65% and 3.31 respectively as reported by Mercy, (2005). The value for the fats obtained was 42.65% which agrees with the 41.9% for *Cucurbita moschata* specie of melon seeds, mostly found in Cameroun (*Felicité*, (2005). Carbohydrate content in the oil was gotten to be 10.88% which is exactly the same value gotten by Mercy, (2005) for *Leganaria siceraria*, Specie of the melon seed.

4.2.3 Comparison of the effects of solvent and mechanical methods of oil extraction of melon seed oil

The Figure 4.1 summarizes the effect of the solvent and mechanical methods of oil extraction from melon seeds.

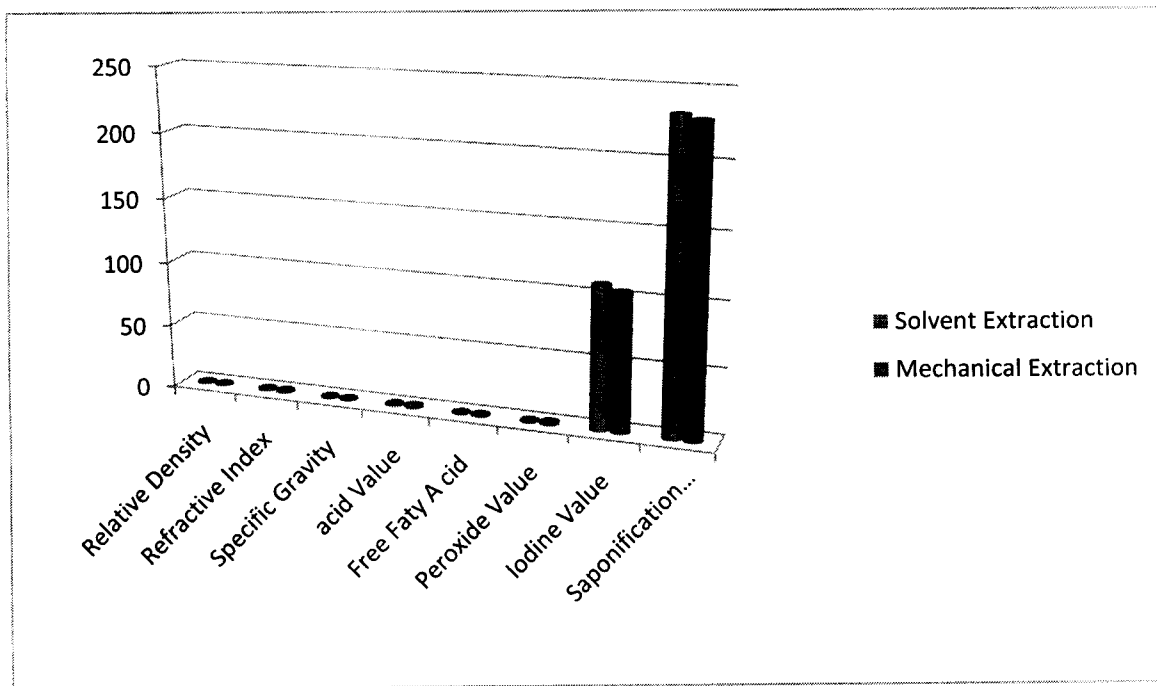


Figure 4.1: Graph of mean values against physicochemical properties parameter for both solvent and Mechanically Extracted Melon seed oil.

From the graph, it was observed that the major difference in the extraction methods was the oil yield, 42% and 21.40% for solvent and mechanical extraction methods respectively. Other differences found there in were that of the Iodine value and the saponification values, it was seen that the iodine value as well as the saponification values for that of the mechanical extraction method were higher than that of the solvent extracted oil, this was seen in their values, it was noted that the difference may have been as a result of the extraction method.

It was also noted that there was little or no effect on the other physicochemical properties of the extracted oil.

Figure 4.2, summarizes the effect of the extraction methods on the extracted oil.

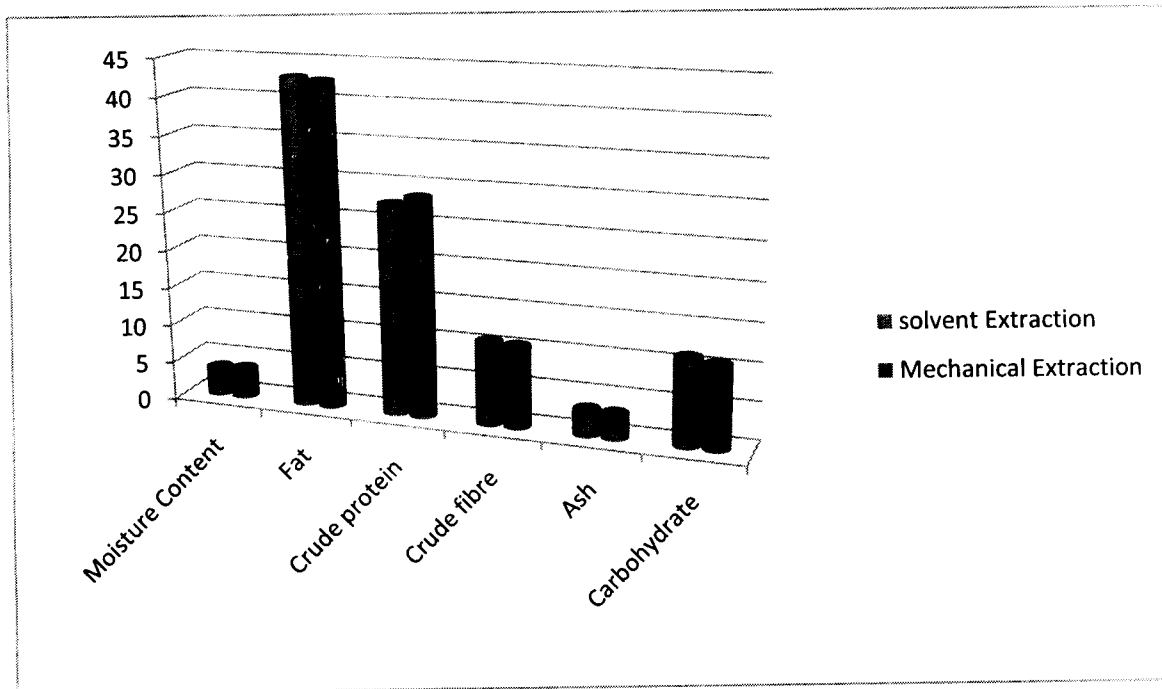


Figure 4.2: Graph of percentages against proximate composition of the solvent and mechanically extracted oil

From the Figure 4.1 above, it can be deduced that the extraction process had little or no effect on the melon seed oil, this is seen in the symmetric pattern of the graph. This results therefore agrees with that which was reported by Mirjana and ksenija, (2005).

CHAPTER FIVE

5.0 CONCLUSIONS AND RECOMMENDATIONS

5.1 Conclusions

This study was carried out to determine the physicochemical properties as well as the proximate compositions of melon seed oil and comparing the results obtained for both methods of extraction for solvent and mechanical extraction respectively,

Peroxide value, 1.04ml/g, 1.64ml/g, Iodine value, 0.1269ml/g, 0.114ml/g, Saponification value, 232.53ml/g, 229.45ml/g, freefatty acid, 0.74ml/g, 0.85ml/g, acid value, 1.48ml/g, 1.70ml/g, Refractive index, 1.470, 1.471, Specific gravity, 0.965, 0.9088, Relative density, 0.9507, 0.9186, color, yellow for both, smell, melon smell for both. The values for the proximate compositions were as well gotten for the two extraction process as shown below respectively; Moisture, 3.72%, 3.88%, Fats, 42.95%, 42.65%, Crude protein, 27.55%, 28.63%, Crude fibre, 10.90%, 10.65%, Ash, 3.46%, 3.31%, Carbohydrate, 11.43%, 10.88%. it was also seen that the oil yield for both solvent and mechanical extraction was gotten as 42% and 21.40% respectively.

The results showed little or no difference in the physicochemical properties of the oil except for the iodine and saponification values which makes the solvent extracted oil less useful in the manufacture of soaps, detergents or industrial uses because of their low values compared to that of the mechanically extracted oil. It is therefore advisable that although the solvent method produced more oil, the mechanical method has more application in the soap industry. Therefore, any method of extraction can be used depending on what the oil wants to be used for.

5.2 RECOMMENDATIONS

- Based on the research carried out, it is recommended that more attention should be given on the other methods of oil extractions such as; supercritical, cold press, aqueous and steam and high pressure method.
- It is also recommended that mineral content of this oil should be determined.
- More research should be carried out to determine the maximum usefulness of this oil Which has low amount of cholesterol which enables body building.

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APPENDICES

Calculations for Parameters of Extraction Methods of melon seed oil

Percentage Oil Yield

Percentage Oil yield for extraction of Melon Oil

$$\text{Percentage oil yield} = \frac{\text{initialweight} - \text{finalweight}}{\text{initialweight}} \times 100$$

Specific Gravity

$$\text{Specific gravity} = \frac{\text{Weight of Xml of oil}}{\text{weight of Xml of water}}$$

$$\text{Mean Iodine Value, } \bar{x}, = (\text{SG}_{S1} + \text{SG}_{S2}) / 2$$

$$\text{Standard Deviation for Acid value} = \sqrt{\sum_{x=1}^n ((x - \bar{x})^2) / 2}$$

Acid Value

$$\text{First Acid value } AV_{S1} = \text{AcidValue} = \frac{\text{Titration (ml)} \times 5.61}{\text{weight of sample}}$$

$$\text{Mean Acid Value, } \bar{x}, = (AV_{S1} + AV_{S2}) / 2$$

$$\text{Standard Deviation for Acid value} = \sqrt{\sum_{x=1}^n \frac{((x - \bar{x})^2)}{3}}$$

Free Fatty Acid

(1ml 0.1M sodium hydroxide = 0.0282g oleic acid) = 0.0282g

$$\text{FFA}_{S1} = \text{FFA Value} = \frac{\text{Titre} \times \text{titre} \times 0.0282 \times 100}{\text{weight of sample}}$$

$$\text{Mean FFA Value, } \bar{x}, = (\text{FFA}_{S1} + \text{FFA}_{S2}) / 2$$

$$\text{Standard Deviation for FFA value} = \sum_{x=1}^n ((x - \bar{x})^2) / 2$$

Saponification Value

$$\text{SV}_{S1}, \text{ Saponification Value} = \frac{(b-a) \times 28.05}{\text{weight of sample}}$$

Where b = Blank reading, a = Sample reading.

$$\text{Mean Saponification Value, } \bar{x}, = (\text{SV}_{S1} + \text{SV}_{S2}) / 2$$

$$\text{Standard Deviation for Acid value} = \sum_{x=1}^n ((x - \bar{x})^2) / 2$$

Peroxide Value

$$\text{PV}_{S1} = \text{Peroxide Value} = \frac{S - B \times 0.1 \times 1000}{\text{weight of sample}}$$

Where , S = Sample, B = Blank

$$\text{Mean Peroxide Value, } \bar{x}, = (\text{PV}_{S1} + \text{PV}_{S2}) / 2$$

$$\text{Standard Deviation for Acid value} = \sum_{x=1}^n ((x - \bar{x})^2) / 2$$

Iodine Value

$$IV_{S1} = \text{Iodine Value} = \frac{(b-a) \times 1.269}{\text{weight of sample}}$$

where, b = blank titre, a = sample titre

$$\text{Mean Iodine Value, } \bar{x}, = (IV_{S1} + IV_{S2}) / 2$$

$$\text{Standard Deviation for Acid value} = \sqrt{\sum_{x=1}^n ((x - \bar{x})^2) / 2}$$