Sudan University of Science and Technology

College of Graduate Studies and Scientific Research

Effect of Heavy Metals on Qualities of Solvent Extracted and Mechanical Extracted Oils

أثر المعادن الثقيلة في خصائص الزيوت المستخلصة بالمذيب والمستخلصة ميكانيكيا

A Thesis Submitted for Partial FullFillment of the Requirements of the Degree of Master of Science in Chemistry

By:

Sara Ismail Safe Aldeen Aldikhairy

Supervisor:

D. Mai Maki Mahmoud

December 2014


بسم الله الرحمن الرحيم

ٹ ڈ

چ ڃ ڄ ڇ

ۆ ۇ ۈ ۋ ۅ ۉ ۊ ۏ ې ۑ ے ۓ ھ ہ ھ ے ے ۓ ك ك ڪ ڪ ۇ ۈ ۆ

چ النور: ٣٥ ی ی ی ی

Dedication

I dedicate this research to mother, father, sisters, brothers and my husband.

Acknowledgements

The authors acknowledge Dr.Mai Maki for her guidance, encouragement, understanding and supervision of this work. Thanks were extended to my friends and all teachers in chemistry lab of Khartoum University everybody that can help me to complete this research work.

Special thanks to all my teachers distinguished….

Abstract

This work was carried out to determine heavy metals contamination levels and comparison between vegetable oils extracted by solvent and mechanical press. Groundnut oil (solvent) contain high level for lead(Pb) 0.9 ppm and compressed oil .02ppm,sesame oil (solvent, compressed) was found in standard limit(0.1ppm).The copper(Cu) content in sesame oil (solvent, compressed) was found in standard (0.1ppm),in groundnut oil (solvent, compressed) 0.064,0.138ppm respectively.Iron(Fe) content in sesame oil(solvent, compressed) 0.2,0.157ppm respectively and standard 0.1ppm,groundnut oil (solvent, compressed) 0.158,0.131ppm respectively. Peroxide value increased in groundnut oil (solvent) 12.6meq/kg, groundnut oil (compressed, manufacture) 4, 9.33meq/kg respectively, the sesame oil (solvent, compressed) 9.66,3.33 meq/kg. Acid value increased in sesame oil (solvent) 1.066 mgKOH/g, groundnut oil (manufacture ,solvent,compressed)1.514,0.505,0.309mgKOH/g respectively, sesame oil (compressed) 0.336mg KOH/g .Iodine value was increased in all sample groundnut oil (solvent, compressed, manufacture) 260.18,213.63,158.65 mg iodine/100g fat respectively, sesame oil (compressed, solvent) had the same value 225.28 mg iodine/100g fat. High value of saponification in groundnut oil (manufacture) 200.0 mg/g, sesame oil (compressed, solvent) 185.97,172.34mg/g, groundnut oil (compressed, solvent)178.42, 163.08 mg/g respectively. The specific gravity of groundnut oil (compressed, solvent) had the same value 0.909, sesame oil (compressed, solvent) 0.911, groundnut oil (manufacture) 0.911.The refractive index in sesame oil (compressed, solvent) 1.4755, groundnut oil (compressed, solvent, manufacture) 1.4735.

الخلاصة

أجريت هذه الدراسة للكشف عن تلوث المعادن الثقيلة فى بعض الزيوت النباتية المستخلصة بواسطة المذيب والعصارة.زيت الفول السودانى (المذيب) إحتوى على نسبة عالية من الرصاص ٩, جزء من المليون وزيت العصارة ٢.,جزء من المليون ، زيت السمسم (المذيب،العصارة)لم يتجاوزا الحد المسموح به (١، جزء من المليون). نسبة النحاس فى زيت السمسم (العصارة،المذيب) فى الحد المسموح به (١, جزء من المليون) ،زيت الفول السودانى (العصارة ، المذيب) ١٣٨,/ ٦٤., جزء من المليون على التوالى. نسبة الحديد فى زيت السمسم (المذيب ،العصارة) ٢, _ ١٥٧, جزء من المليون على التوالى والحد القياسى (١، جزء من المليون)،زيت الفول السودانى (المذيب،العصارة) ١٣١/,١٥٨، جزء من المليون على التوالى.أعلى قيمة للبيروكسيد فى زيت الفول السودانى (المذيب) ٦,١٢ملمكافئ/ كجم ، زيت السمسم (المذيب،العصارة) ٩.٦٦ / ٣٣،٣ ملمكافئ /كجم على التوالى،زيت الفول السودانى (العصارة ، المصانع) ٤/ ٩،٣٣ ملمكافئ/كجم على التوالى.القيمة الحمضية أعلى قيمة فى زيت السمسم (المذيب) ١,.٦٦ مل جرام هيدروكسيد بوتاسيوم / جرام ، زيت الفول السودانى (مصانع ، المذيب ،العصارة) ٥١٤,١/ ٥,٥. / ٣,٩, ملجرام هيدروكسيد بتاسيوم/جرام على التوالى،زيت السمسم (العصارة) ٣٣٦, ملجرام هيدروكسيد بوتاسيوم /جرام .رقم اليود قيمته عالية في كل العينات، زيت الفول السودانى (المذيب ،العصارة ،المصانع) ٢١٣,٦٣/١٨,٢٦٠ /١٥٨,٦٥ مل جرام / ١٠٠جرام دهن على التوالى ، زيت السمسم (العصارة، المذيب) قيمتهما واحدة ٢٨,٢٢٥ملجرام / ١٠٠ جرام دهن .رقم التصبن أعلى قيمة فى زيت الفول السودانى (مصانع) ٢٠٠ ملجرام /جرام ،زيت السمسم (العصارة ، المذيب) ١٨٥،١٧٢،٣٤,٩٧ ملجرام /جرام على التوالى ، زيت الفول السودانى (العصارة ، المذيب) ١٧٨,٤٢، ١٦٣,٠٨ ملجرام/جرام على التوالى .الكثافة النوعية الزيت الفول السودانى (العصارة،المذيب) ٩٠٩, زيت السمسم (العصارة،المذيب) ٩١١, زيت الفول السودانى (المصانع) ٩١١, .معامل الإنكسار لزيت السمسم (العصارة ،المذيب) ٤٧٥٥,١ ، وزيت الفول السودانى (العصارة،المزيب،المصانع) ٤٧٣٥,١ .

List of Contents

List of Tables

List of Figures

Chapter One

Introduction

1 Introduction

1.1 Vegetable oil:-

Vegetable oils rich in polyunsaturated fatty acids (PUFA) are highly digestible as compared to saturated fats. The presence of fats and oils in poultry rations provides a suitable medium for rancidity and nutrients react with oxygen to form free radicals(Sherwin.1978).The products of oxidation include shorter chain fatty acids, fatty acid polymers, aldehydes, ketones, epoxides and hydrocarbons(Wiseman.1986). These highly reactive compounds can damage animal cells and have been implicated in certain negative diseases. These compounds formed of oxidation can be measured by acid, peroxide value (AOAC.1990).

1.2 Sesame seed

Sesame (sesamum indicum,L.) belongs to the family of pedaliaceae. Sesame considered having both nutritional and medicinal value. Sesame grows in long day areas , it flowers about 45days under 10-hours day length. Sesame plant is cultivated in relatively hot and dry regions because the seeds are adaptable and drought -resistance. Sesame crop matures in about 80 days (Beech, D.F.1996).

Sesame seed contain 50-60% oil and 19-25% protein with antioxidants lignans such as sesamolin and sesamin which prevent rancidity and give sesame oil along shelf- life, also it is a good source of minerals, particularly calcium, phosphorus, potassium and iron. Sesame oil is highly resistant to oxidation and displays several medicinal effects (Abou-Gharbia.2000).

Sesame oil: The presence of high polyunsaturated fatty acids content make it possible to use sesame oil for cooking in place of other edible oils and to

help reduce high blood pressure, control hypertension, reduction of cholestrol levels, slowing down certain types of cancer.

1.3 Peanut seed

Peanut (Arachis hypogaea.L.) belongs to genus Arachis the family leguminosae (Nigam et al.1983). Peanut is a major source of edible oil and protein 25-36% ,groundnut have desirable fatty acid profile and are rich in vitamins, minerals and bioactive material (include resveratrol in the seed, flavonoids, phytosterols, they contain monosaturated and polysaturated fatty acids, potassium, magnesium, copper, fiber,α-tocopherol (Sabate.2003).
Peannuts commonly contain 40-50% oil and contain high proportion of unsaturated fatty acids, particular oleic, lunoleic and palmitic. Oxidative stability of groundnut oil is highly correlated with the ratio of oleic acid to linoleic acid.

Groundnut crop is grown during kharif season under rain dependent conditions, grow best in light, sand-loam soil, high temperatures $20C^0$to $28C^0$, it need moderate rainfall from sowing until flowering, growth period of 110 to150 days (5 months).It requires a soil PH 5-6.5,high acidity could induce magnesium or aluminum toxicity, in this type of soil, calcium should be added to maintain the PH above 6 (IBPGR,ICRISAT.1992).

The time of harvesting is very critical as it can significantly affect the economic yield and the quality of seeds. Both pre-mature harvesting and over maturity can be harmful. When the crop is mature, the leaves start yellowing. The mature pods become reticulated and within it, seed is separated from the shell of the pod and the inside of the shell becomes dark in color. The crop should be harvested when 70-75% of the pods are mature .A few representative plants in the field should be uprooted and their pods

should be studied to determine the optimum time for harvesting, (Baldwin.1990).

1.4 Aflatoxin

Aflatoxin are toxic secondary metabolites and a group of mycotoxins(fungi) produced mostly by Aspergillus flavus and A. parasitcus. The most member of aflatoxin group are B_1,B_2,G_1,G_2, they are carcinogenicity and found in groundnut, cereals and their products, dried fruits, herbs).Aflatoxin contamination of groundnuts and groundnuts include poor agricultural practices during planting, harvesting, drying, transportation and storage of product and in animal feeds.(Oliveira et al.2009).Aflatoxin range in sesame oil 0.2-0.8µg/kg and groundnut oil 0.6µg/kg. Degradation of aflatoxins were evaluated in samples subjected to gaseous ozonation under various temperatures (25,50,75) and exposure times (5,10,15min),also use gamma irradiation, microwave heating to decrease the aflatoxins.(Ghanem,Orfi,shamma.2008).

Unrefined oils are contaminated with aflatoxin (B_1), so the refining is an essential process for elimination of aflatoxins from edible oils.(Younis,Y.M.H.Malik,M.K.2003).

1.5 Cancer

The American Institute for Cancer Research (AICR) has suggested that 30-40% of all cancers are linked to the diet,cancer risk may be modified to a certain extent by lifestyle change. Adapting healthful diets and exercise practices at any stage of life can promote health and reduce the risk of cancer. To reduce cancer as well :-

1-Eat a variety of health foods with an emphasis on plant sources.

2-Adopt a physically active lifestyle.

3-Mantain a healthy weight throughout life,(American Cancer Society.2005).

Fatty acid synthesis is over expressed in a number of cancers Researchers intrigued by this observation have tested inhibitors of fatty acid synthesis on mice to see if the inhibitors slow tumor growth. These inhibitors do indeed slow tumor growth, apparently by inducing apoptosis. However, another startling observation was made, mice treated with inhibitors of the condensing enzyme showed remarkable weight loss because they ate less. Thus, fatty acid synthesis inhibitors are exciting candidates both as antitumor and as antiobesity drugs,(Berg,Jeremy Mark.2007).

1.2 Literature review

Fats and oil, a group of naturally occurring organic compound called triglycerides ester, are comprised of three molecules of fatty acids and one molecule of the alcohol glycerol.

CH_3-(CH_2)-CH=CH-$(CH_2)_7$-COOH

Unsaturated fatty acid

HO-CH_2

HO-CH

HO-CH_2

Glycerol

CH_3-$(CH_2)_n$-CH_2-CH_2-$(CH_2)_n$-COOH

Saturated fatty acid

They are oily, greasy or waxy substances that in their pure state, are normally stateless, colorless and odorless. They are lighter than water and are insoluble in it; they are slightly soluble in alcohol and readily dissolved in ether and other organic solvent. Fats are soft and greasy at ordinary temperature (Hui.1996).

1.2.1 Natural oil constituents

1.2.1.1 Fatty acid

Fatty acids are long hydrocarbon chains of various lengths and degree of unsaturation terminated with carboxylic acid groups. The systematic name for a fatty acid is derived from the name of its parent hydrocarbon by the substitution of oic for the final e .For example, the C18 saturated fatty acid is called octadecanoic acid because the parent hydrocarbon is octadecane.A C_{18} fatty acid with one double bond is called octadecenoic acid;with two double bonds, octadecadienoic acid. The notation 18:0 denotes

a C_{18} fatty acid no double bonds, whereas 18:2 signifies that there are two double bonds. Fatty acid carbon atoms are numbered starting at the carbonyl terminus, the methyl carbon atom at the distal end of the chain. The position of a double bond is represented by the symbol Δ followed by a superscript number. For example, cis-Δ^9 means that there is a cis double bond between carbon atoms 9 and 10; trans-Δ^2 means that there is a trans double bond between 2 and 3. Fatty acid in biological systems usually contain an even number of carbon atoms, typically between 14 and 24 .The hydrocarbon chain is almost in variably unbranched in animal fatty acids. The alkyl chain may be saturated or it may contain one or more double bonds. The configuration of the double bonds in most unsaturated fatty acids is cis. The double bonds in polyunsaturated fatty acids are separated by at least one methylene group.

The properties of fatty acid and of lipids dependent on chain length and degree of saturation. Unsaturated fatty acids have lower melting points than saturated fatty acids of the same length.

Fatty acids are elongated and desaturated by enzyme systems in the endoplasmic reticulum membrane.

Fatty acids are important as:-

1-Fuel molecules,

2- Components of phosphlipids and glycolipids.

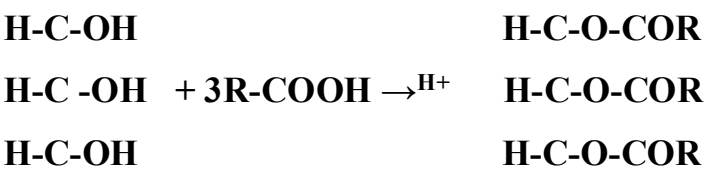
3 -Hydrophobic modifies of proteins.

4- Hormones and intracellular messengers.

They are stored in adipose tissue as triacylglycerols (neutral fat).(Berg,Jeremy Mark.2007).

1.2.1.2 Lipids

Lipids are esters of long chain monocarboxylic fatty acid with alcohol, most trihydric alcohol (glycerol).



Triglyceride formation

Lipids are insoluble in water and can be extracted from the cell and tissues by non-polar solvents, such as chloroform, ether, and benzene.

1.3 Nonsaponifiable Lipids

Lipids are subdivided into main classes**:-**

1.3.1 Steroids:-

These are a third class of lipids, steroids, a long with lipid vitamins and terpenes, are classified as isoprenoides because their structures are related to the five-carbon molecule isoprene. Steroids contain four fused rings (three cyclohexane rings and one cyclopentanes rig fused together), the characteristic ring structure is derived from squalene. (H.Robert Horton.2006).

1.3.2 Terpenoids:-

These are derivatives of the isoprene cell 2-methyl-1,3-butadiene. Example is B-carotene which is the precursor for vitamin A in animals, terpenes mostly produce vitamins.

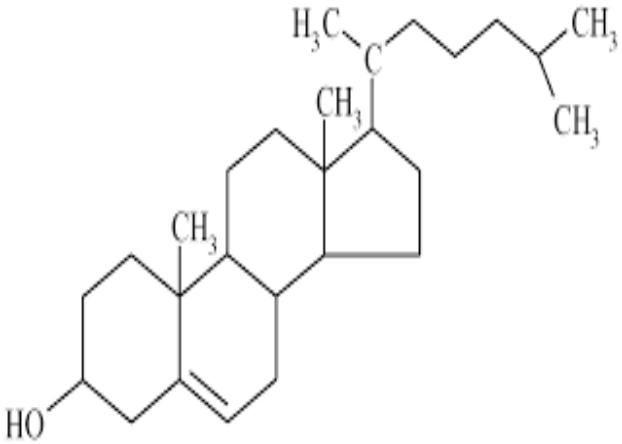
Generally on hydrolysis, the lipids yield two major classes:-

(a)-Portion of the lipids, these contain fatty acids and saponifiables which constituent the major glycerol.

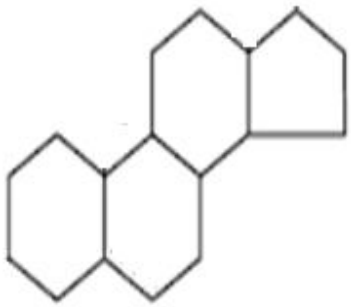
(b)-Unsaponifiables which constitute only very small proportion.
These include sterols, tocopherols aromatic and aliphatic hydrocarbons.

1.3.3 Sterols

Unsaponifiable matter, hydrocarbon compound without fatty acids or glycerol, it contain steroid nucleus (4 fused rings, three with 6 carbons and one with 5 carbons), sterol present in vegetable oils in free form or sterol esters, sterol glycosides.



Cholesterol



Steroid nucleus

Cholesterol is a major sterol in animal tissues, amphipathic molecule (polar-OH head, non polar alkyl side chain), other steroids include sex hermones are synthesized from cholesterol,(H.Robert.Horton.2006).

1.4 Antioxidants

Antioxidants are substances that can prevent the oxidation and have ability to trap the free radicals produced as a result of different metabolic processes and protect lipids, proteins and nucleic acid from the oxidative damage. (Frei et al.1988).

Tocopherols are major natural antioxidants of vegetable and animal fats. The animal body is unable to synthesize these compounds and the small a mounts found in fats of animal origin are derived from the vegetable components of the animal food. These compounds known as alpha, beta, and gamma, tocopherol are isomers or homologs formed by methylated phenol by methylation of a substituted chroman nucleus.(Hashim I.1993).

1.5 Waxes

Waxes are non polar esters of long-chain fatty acids monohydroxylicalcohol, for example, myricylpalmitate, a major component of beewax. Waxes are widely distributed in nature. They provide protective waterproof coatings on the leaves and fruits of certain plants and on animal skin, feathers. Ear wax is a complex mixture made up mostly of long chain fatty acids, cholestrol and ceramides, also contain squalene, triacylglycerols.(H.Robert Horton.2006)

1.6 Reaction of fats and oils

1.6.1 Hydrolysis of fats

Like other ester, glycerides can hydrolyze readily, partial hydrolysis of triglycerides will yield mono- and diglycerides and fatty acids. When the hydrolysis is carried to completion with water in the present of an acid catalyst, the mono-di, and triglycerides will hydrolyze to yield glycerol and fatty acid.

Lypolytic enzymes are present in some edible oil sources.

Any residues of these lypolytic enzyme present in some crude fats and oils are deactivated by elevated temperatures used in oil processing, so enzymatic hydrolysis is unlikely in refined fats and oils (O'Brian,R.D.2004).

1.6.2 Oxidation of fats and oils

1.6.2.1 Autoxidation

In autoxidation oxygen reacts with unsaturated fatty acids. Initially, peroxides are formed which may break down into secondary oxidation products (hydrocarbons, ketones, aldehydes and smaller a mounts of epoxides and alcohols).Metals, such as copper or iron, present at low levels in fats and oils are normally treated with chelating agent such as citric acid to complex, these trace metals (thus inactivating their prooxidant effect).The result of the autoxidation of fats and oils is the development of objectionable flavors and odors characteristic of the condition. Some fats resist this change to a remarkable extent while others are more susceptible depending on the degree of unsaturation, the presence of light, for example, increases the rate of oxidation. It is common practice in the industry to protect fats and oils from oxidation to preserve their acceptable flavor and to maximize shelf-life. (Food Fats and Oils.2006).

1.6.2.2 Oxidation at higher temperatures

Although the rate of oxidation is greatly accelerated at high temperatures, oxidative reactions which occur at higher temperatures may not followed precisely the same routes and mechanism as the reactions at room temperature; thus differences in the stability of fats and oils often become

more apparent when the fats are used for frying or slow baking. The more unsaturated fat or oil, the greater will be its susceptibility to oxidative rancidity. Predominantly unsaturated oils, such as cotton seed, or corn oil, are less stable than predominantly saturated such as coconut oil. Dimethylsilicone is usually added to institutional frying fats and oils to reduce oxidation tendency and foaming at elevated temperatures and partial hydrogenation is also employed in the processing of liquid vegetable oil to increase the stability of oil (Food Fats and Oils.2006).

1.6.3 Polymerization of fats

All commonly used fats and particularly those highly polyunsaturated fatty acids tend to form some larger molecules known broadly as polymers when heated for long time under extreme conditions of temperature and time. Under normal processing and cooking conditions polymers are formed in insignificant quantities.

Although the polymerization is not well understood, but it is believed that polymers in fats and oils arise by formation of either carbon to carbons bond or oxygen bridges between molecules. When an appreciable amount of polymer is present, there is a marked increase in viscosity.(Food Fats and Oils.2006).

1.6.4 Reactions during heating and cooking

Glycerides are subject to chemical reactions (oxidation, polymerization, and hydrolysis) which can occur particularly during deep fat frying. The extent of these reactions, which may be reflected as a decrease in iodine value of the fat and increase in free fatty acids, depends on the frying conditions, principally the temperature, aeration and duration. The composition of frying fat also may be affected by the kind of food being

fried. When frying high fat foods, some fat form will be rendered and blend with the frying fat and some frying fat will be absorbed by the food. In this manner the fatty acid composition of the frying fat will change as frying progresses, since absorption of fat by the fried food continuous generation of smoke from deep fryer is a good indication that the fat is being overheated and could ignite if high heating continuous. If smoke is observed during a frying operation, the heat should be reduced. Furthermore, if a consumer wishes to save that fat or oil after cooking, the hot fat or oil should never be poured back into its original container. Most containers for cooking oils are not designed to with stand the high temperatures reached by the oil during cooking. Pouring hot oil into containers could result in breakage or melting of the container and possible burns to the user. (Food Fats and Oils.2006).

1.7 Hydrogenation

Hydrogenation is the process by which hydrogen is added to point of unsaturation in the fatty acids. Hydrogen was developed as a result of the need to:-

1- Convert liquid oils to the semi-solid form for greater utility in certain food uses.
2- Increase the oxidative and thermal stability of the fat or oil. It is an important process to our food supply, because it provides the desired stability and functionality to many edible oil products.

In process of hydrogenation, hydrogen gases react with oil at elevated temperature and pressure in the presence of catalyst. The catalyst most widely used in nickel supported on an inert carrier which is removed from the fat after the hydrogenation process is completed. Under these conditions, the gaseous hydrogen reacts with double bonds of the unsaturated fatty acid

as illustrated bellow:

$$-CH_2-CH=CH-CH_2- + H_2 \rightarrow^{Ni} -CH_2-CH_2-CH_2-CH_2-$$

The hydrogenation process is easily controlled and can be stopped at any desired point. As hydrogenation progresses there is generally a gradual increase in melting point of fat or oil,(Food Fats and Oils.2006).

1.8 Interesterification

The processes used to modify the physico-chemical characteristic of oils and fats. Interesterification is an acyl-rearrangement reaction on the glycerol molecule. On other hand, hydrogenation involves addition of hydrogen to the double bonds of unsaturated fatty acids. Two types of interesterification, i.e.chemical(random) and enzymatic. Interesterification produces a complete positional randomization of acyl groups in triacylglycerol, by using chemical catalysts. Enzymatic process uses lipases as catalyst (Nor and Noor.2005).The interesterification equation is shown below:-

$$C_3H_5(OOCR)_3 + 3CH_3OH \rightleftarrows 3CH_3OOCR + C_3H_5(OH)_3$$

1.9 Esterification

Fatty acids are usually present in nature in the form of esters and are consumed as such. Triglycerides, the predominant constituents of fats and oils, are examples of ester. When consumed and digested, fats are hydrolyzed initially to diglycerides and mono-glycerides which are also esters. Carried to completion, these esters are hydrolyzed to glycerol and fatty acid. In the reverse process, esterification, an alcohol such as glycerol is reacted with an acid such as fatty acid to form an ester such as mono-,di-,and triglycerides. In an alternative esterification process, called alcoholysis, an alcohol such as glycerol is reacted with fat or oil to produce esters such as

mono- and diglycerides. Using the foregoing esterification processes, edible acids, fats, and oils can be reacted with edible alcohols to produce useful food ingredients that include many of the emulsifiers.(Food Fats and Oils.2006).

$$CH_3{-}COOH + CH_3{-}CH_2{-}OH \rightarrow CH_3{-}COOCH_2{-}CH_3 + H_2O$$

1.10 Rancidity of oils and fats

The edible oils, fats ad their food products on storage show deterioration which is indicated by the development of off-flavor, off-odor and some times change in color and taste in the fatty food products. This change occurs as soon as the oils, fats and fatty food products come into contact with atmospheric oxygen. The enzymes and micro-organisms also react with them and bring about alteration in the structure of oils and fats. This phenomenon of the development of off-flavor, off-odor and change in color and taste is in general, called rancidity. Type of rancidity:-

1.10.1 Oxidative rancidity

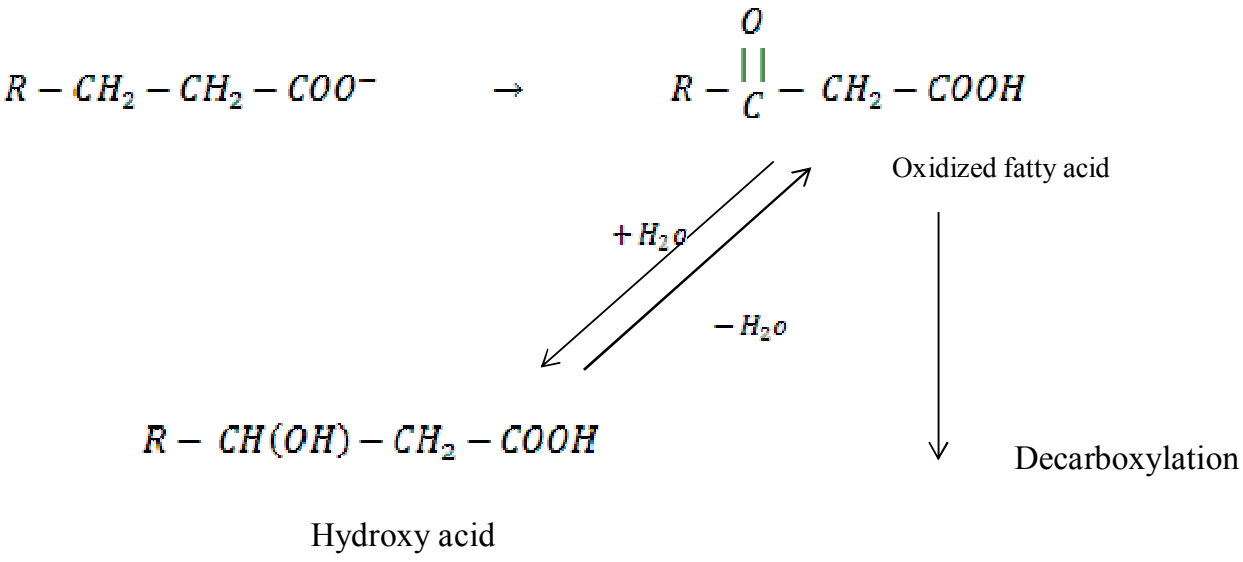
Such a rancidity develops sooner or later on exposure to atmospheric oxygen in all substances with appreciable contents of glycerides of long chain fatty acids which results in off-flavor, off-odor and some times change in color and taste. It is due to the formation of peroxide at the double bonds of fat molecules with subsequent break down of these peroxides to form aldehydes, ketones and acids of lower molecular weight. This rancidity not only causes undesirable flavors and odors but exerts harmful nutritional and physiological effects. Oxidative deterioration leads to the destruction of fat soluble vitamins and essential fatty acids as well as concern of toxicological effects of various types,(Food Fats and Oils.2006).

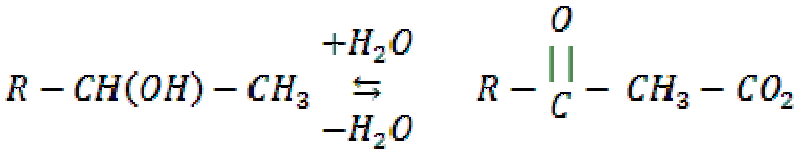
1.10.2 Hydrolytic (lypolytic) rancidity

Refers to the odor that develops when triglycerides are hydrolyzed and free fatty acids are released. This reaction of lipid with water sometimes requires a catalyst, but results in the formation free fatty acids and salts from free fatty acids (soap). In particular, short chain fatty acids, such as common butter fats, are odorous. Various micro-organisms also contain some enzymes that may produce ketones and other oxidation products from the fatty acid esters which become responsible for the hydrolytic rancidity,(Food Fats and Oils.2006).

1.10.3 Microbial rancidity

In which micro-organisms such as bacteria, molds and yeast use their enzymes to break down chemical structures in the oil, producing unwanted odors and flavors. Water needs to be present for microbial growth to occur (Robin Koon 2009). The mechanism of enzymatic oxidation is as follow:-





1.10.4 Flavor reversion rancidity

This type is accomplished with oxidative rancidity which appears stepwise. This refers to the accumulation of hydroperoxides compounds due to the oxidation of lipids gradually in the presence of low concentration of natural antioxidants. This type of oxidation occurs in the oil containing the linoleic acid in the presence of small amounts of oxygen (Ozkan,G,Simesk.2007).

1.11 Extraction and refining of edible oils

1.11.1 Crude oils:

Crude vegetable oils and fats derived from oil bearing seeds and fruits, etc. Generally obtained by either of two methods:

1- Mechanical pressure.

2- Solvent extraction.

The effect of treatment by either method is to separate more or less completely the oil or fat from the solid matter naturally associated with it, the solid matter being generally utilized for agricultural purposes. Now the winning of the oil from the original matter is much more efficient by solvent extraction than by mechanical pressure. The residue contains approximately 4 to 8% and a round 1% or 2% when solvent extraction is employed. The solid residues from mechanical pressure processes appear in the form of the well known oil cake, or uneven lumps.

1.11.2 Extraction of oil by mechanical pressure

Wherever pressure is employed the ground meal has to be tempered by

heating in an open vessel known as kettle consist of a jacketed pan and stirrer and some mean off introducing moisture in to the meal as required in the form of steam.

The meal is heated is such kettles to the desired temperature and fed from here to the presses. Modern methods of pressing are practically confined to two; hydraulic presses and screw presses.

1.11.3 Extraction of oil by solvent

Compared with mechanical pressing methods, solvent extraction is comparatively new process, and although improvements have been made, there is more scope. The general principles of solvent extraction are very simple and comprise the washing out of oil from the ground seed by means of a hot solvent and the subsequent evaporation of the solvent in order to recover the dissolved oil, the solvent being over and over used again. The extracted meal is freed from solvent first by heating and finally steaming.

1.12 Refining of oil

The oil obtained by the solvent extraction or by mechanical pressure, contains trace of impurities in the form of meal, moisture and other non-fatty contamination, which are removed by simple mechanical means like:-

(a)- Sedimentation.

(b)- Filtration.

(c)- Centrifugation.

1.12.1 Sedimentation

The oil is kept warm by storage in tanks which are heated either by means of steam oil in a warmed building. The moisture and foots are drawn off at the bottom and cleared at the top.

1.12.2 Filtration

The most common and most efficient apparatus is well-known filter press consisting of a series of corrugated and perforated plates, so arranged that the oil is driven by pressure through a cloth inserted between the plates. Clear oil flows from outlet at the bottom of each plate and the foot and meal are retained on the surface of the cloth as filter press cake.

Various other types of filter are in use, some of special design and of varying efficiency (Frankel,E,N.1984).

1.12.3 Centrifugation

The various type of centrifugation machines are fully described by their makers and many of them are reasonably efficient for rapidly separating meal and moisture from oil, some of the small machines working at very high speed are of exceptional efficiency, but methods of really continuous centrifugation have yet to be designed.

The clear oil obtained by any of the methods described above, that has been free from non-fatty material, constitutes the crude oil for commercial use, such oil is suitable as raw material for soap or candle manufacture and variety of other purposes (Guston,F.D.1983).

1.15 Neutralization

The acidity of fat is generally removed by treatment with alkali, the most commonly employed being caustic soda, though magnesia, lime, carbonate of soda and other alkalis are occasionally used. The crude oil which should be as clear and free from sediment as possible, having been raised to the desired temperature, is agitated and the caustic soda run in, preferably by means of coarse spray on the surface of the oil, as soon as the amount of alkali necessary has been added, agitation is stopped and the mass

allowed to settle. The free fatty acid approximately an equal amount of neutral oil, after standing for few hours, the supernant neutral oil is drawn off, preferably by means of dipping siphon, and washed free from alkali.

The reaction bellow explains the formation of soap:

$$RCO_2H+NaOH \rightarrow RCO_2Na+H_2O$$

1.16 Bleaching

Substances known as "adsorptive earths" are mixed with the oil or liquid fats, any colors that are in the oils adsorb on the earth and hence are removed from the oil.

1.17 Deodorization

The edible qualities suitable for margarine, baking, confectionery and other special purposes must be quite odorless and tasteless and for this purpose the common edible grade is subjected to live superheated steam in a closed vessel (a deodorizer).

The best form of deodorizer subjects the oil to the action of highly superheated steam injected through the oil contained in the vessel under greatly reduced pressure. The oil is then cooled and run in to barrels or storage-tanks and constitutes the refined deodorized edible oil for commercial use, such oil should be tasteless and odorless, clear and bright, when in liquid condition and free from moisture. These are the basic processes for producing the finished oils and fats. These fats may then be modified either physically (such as by fractionation) or chemically (such as by hydrogenation),(M.K.Gupta.2000).

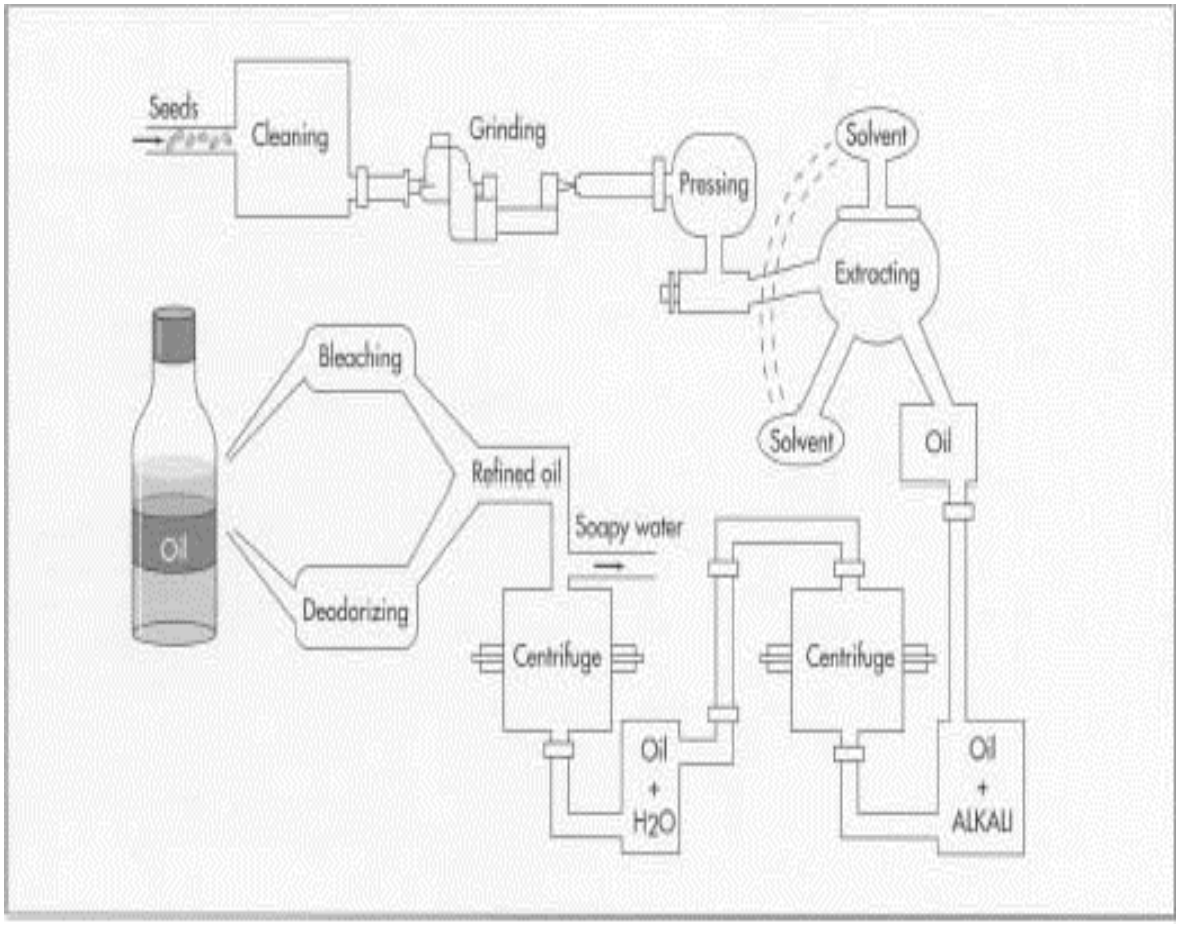


Figure 1.12 Refining of oil

1.18 Modification of edible oil

1.18.1 Fractionation

The objective of fractional crystallization of fats may be to eliminate from a fat or oil a fraction which is the cause of undesirable properties, or it may be produce a new product with a narrower range of triglycerides and hence different properties.

The process has two main steps; crystallization and filtering. The first is achieved by slowly cooling the butter oil in stirred tank. Accurate control of cooling rate and agitation yields “slurry” of high melting “slearine” crystals

in a low melting liquid “olein” which is easy to filter. Fractionation is often carried out after the oil has been deodorized, but sometimes is done before the deodorizing step.

1.19 Chemical modification

The main process used here is hydrogenation, which is used to harden oils, although other processes are also used.

1.19.1 Hydrogenation

This is the process of adding hydrogen to a double bond in the molecule of triglyceride. The reaction was carried out in the presence of a catalyst and result in an increase in the saturation of molecule (i.e. a decrease in the number of multiple bonds) and arise in the melting point. The hydrogenation process can be taken to completion, in which case the product is a mixture of fully saturated glycerides and wholly solid. This is carried out only in the case of coconut oil and with tallow to produce hard fats with special applications. More generally, hydrogenation is not taken to completion but to a pre-determined point where the required chemical and physical properties result. The reaction is controlled by the iodine value which is a measure of the degree of saturation of the fat.

$$-C=C- + H_2 \rightarrow -CH-CH-$$

During hydrogenation the unsaturated glycerides adsorb on the catalyst surface. The hydrogenation process is carried out a large vessels equipped with stirrers and cooling coils, the latter being essential as the reaction is exothermic. The catalysts used in the industry are usually reduced nickel catalysts supported on silica, catalyst preparation is a highly skilled process and there are firms which specialize in the manufacture of these products (Food fats and oils.1988).

1.20 Physical characteristic of edible oils

1.20.1 Color

Oils generally colored, as they contain in true or colloidal solution varying quantities of different lip double pigments. These may be lipo-chromes originated from olciferous tissue, or artifacts caused by degradation, most usually thermal during processing treatments.

Color is important quality factor and in order to maintains a bright color in the final product, chemical treatment or additive are often used in place of bleaching by heat to in activate enzymes.

The color of oil was closely correlated with the increasing amount of carbonyl compound (Luh,B.S,Feinberg.1986).

1.20.2 Viscosity

Viscosity is amount of the internal friction in oil and is an important index in the study of oils and their intermolecular forces.

Viscosity is also useful criterion on desegregation or depolymerization such as that which occurs in the initial stages of hydrolsis of fats and oils during storage. Unsaturation reduced viscosity in fats and oils, e.g. linoleic acid is less viscous than oleic acid and oleic acid is less viscous than stearic.

Hydrogenation of oil increases its viscosity as it decreases its instauration (G.E.LeBlanc.1995).

Viscosity procedures:

Fill the viscometer such the level on the left side is at point C and the level on the right side is at point A (i.e., a certain volume of liquid depending on the volume of the viscometer).Because of the height difference between C and A (h), there is a hydrostatic head or driving pressure to cause the liquid to flow through the capillary or narrow diameter section of the

viscometer. The experiment is to measure the time it takes the liquid to flow from point A to B .In other words the time it takes the volume of liquid between A and B to flow through the capillary.

1.20.3 Refractive index

Refractive index of fats and oils has been reported to indicate an increase in autoxidation, the increase in refractive index with autoxidation is possible attributable to conjugation known to precede hydroperoxide formation in the secondary stage and polymerization of partially oxidized fats in the tertiary stage of autoxidation, since both conjugation and polymerization are reported to result in increased refractive index of oils and fats. Moreover, refractive index of fats and oils decreased in most conditions "controlled or normal conditions" with contaminant increase in the free fatty acids (Mondal GC.and Nadi.1984).

1.21 Chemical characteristic of vegetable oils And fats

1.21.1 Iodine value

The progress of oxidative rancidity can be followed in number of ways, these include measurement of iodine absorption and uptake of oxygen by oil. Hence it has reported that the iodine value probably an excellent indication of oxidation, or at least oxidation plus polymerization as it occurs in herring meal, where the changes in the oil will be mainly oxidation followed by oxidation induced polymerization and degradation. Furthermore, it has been suggested that iodine value is reasonable satisfactory measure of chemical oxidation and subsequent polymerization in a system such as fish meal.

Iodine value of the fat at elevated temperature show no significant change (Eromosele.I.,C.,C.O.1997).

1.21.2 Peroxide value

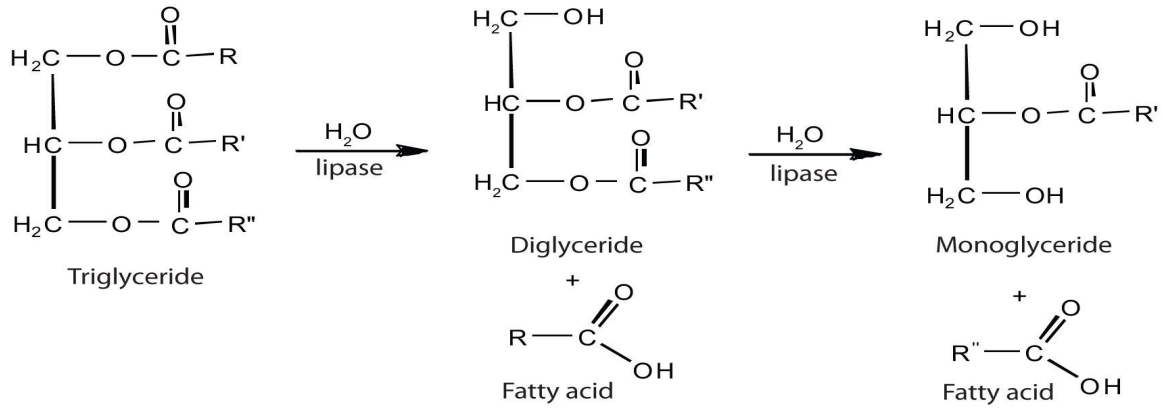
Oxidation of lipids to hyroperoxide, referred to as the peroxide value of fat represent the reactive oxygen contact, and estimated through the liberation of iodine from potassium iodine in glacial acetic acid and expressed as milliequivalents of peroxides\100gm of sample.

Change in peroxide value can often be used to monitor the potential shelf-life and is a good guide to the quality of fat.

Rancidity as defined in its broadest meaning is deterioration of flavors and odors of fat or fatty proteins of foods due to hydrolysis, oxidation or microbial action. The change of peroxide value of oils and fats during storage under normal or controlled condition is an important parameter for detecting their quality. It has been established that since lipid per-oxidation mechanism initiated by autoxidation that can be catalyzed by either metallic portions or enzymes; the reaction once initiated is propagation forming more hyroperoxide and more free radical and or break down products depending on the condition. The products so formed are complex compounds with amino acids, proteins or enzymes (Gan,H.L.,2005).

1.21.3 Free fatty acid

Free fatty acid (FFA) or unesterified fatty acids are ubiquitous of minor components of all living tissues. Free fatty acid and monoacylglycerols are released and absorbed in the small intestine. In intestinal mucosa cells, FFA are re-esterified to triacylglycerols, which are transported via lymphatic vessels to the crticulation as part of chylomicron.FFA are taken up into cell mainly by protein transporters in the plasma membrane and are transported in tracelluarly via fatty acid-binding protein (FABP) activated acyl CoA.(Muller.2001).



1.21.4 Saponification value

In this case of neutral fats and oils which are largely mixed triglycerides containing several varieties of acid, the saponification value usually found between190-200.

In several instance in which longer chained acids make up large proportion of the fatty acids saponification value are markedly lower than those for common fats (ISO 3657 :2002.Animal and vegetable fats and oils.).

1.22 Food and edible oils contamination

It is important to protect food from risk of contamination to prevent food poisoning and the entry of foreign objects. These are four main ways in which food and edible oil can become contaminated: bacterial, physical, chemical and metallic contamination.

1.22.1 Bacterial contamination

If food is assumed to be contaminated by certain, harmful bacteria (pathogenic bacteria) or their toxins; (poisons produced by some of these bacteria), food poison may result.

Food contamination with pathogenic bacteria may appear to taste and smell

perfectly also the micro-organism such as bacteria, molds and yeast can use their enzymes to breakdown chemical structure in the oil and fat, producing unwanted odors and flavors.

1.23 Metallic contamination of edible oils and fats

Edible oils and fats are frequently subjected to processing including refining, bleaching and deodorization, which inevitably makes the oils come in contact with metallic surface area, often at high temperature .This lead to contamination of edible oils and fats by heavy metals.

As certain heavy metals such as lead, copper and mercury have been recognized to be potentially toxic with in specific limiting values, a considerable potential hazard exists for human nutrition. Not all the trace of heavy metal in plants and animals are the results of human activity. Some arise through the absorption process of naturally occurring soil components, theoretically, every 1000kg of "normal" soil contain 200g, 80g nickel, 16g lead 0.5g mercury and 0.2g cadmium.

Some of them, such as copper, nickel, chromium and iron, for example, are essential in very low concentration for the survival of all forms of life. These are described as essential trace elements, only when they are present in greater quantities, these like the heavy metal lead, cadmium and mercury which are already toxic in very low concentrations, cause metabolic anomalies.

1.24 The toxic heavy metals

1.24.1 Lead

The origin of lead in the food stuffs and their surrounding:

Lead has been mine since ancient times and has been processed in many ways e.g. for water pipes, container and as acetate, even for sweetening wine

(lead sugar).

World production amount of millions of tons and is used in the manufacture of accumulators, solders, pigments, cables and anti-rust agent and a considerable amount still into anti-knock petrol.

The main source of lead pollution in the environment is:-

Industrial production processes and their emissions road traffic with leaded petrol, the smoke and dust emissions of coal and gas-fired power station, the laying of lead sheets by roofers as well as the use of paints and anti-rust agents.

1.24.1.1 Toxic Effect

Lead can trigger both acute and chronic symptoms of poisoning. Acute intoxication only occurs through the consumption of relatively large single doses of soluble lead salts. High affinity for proteins, the lead ions consumed bond with the hemoglobin and the plasma protein of the blood. This leads to inhabitation of the synthesis of red blood cells and thus of the vital transport of oxygen, if the bonding capacity here is exceeded, lead passes into the bone-marrow, liver and kidneys, such intoxication leads to:

1- Encephalopathy's in central nervous system (CSN);
2- Disturbance in kidney and liver functions progressing as for necrosis;
3- Damage to the reproductive organs.
4- Anemia and many metabolic deficiency symptoms.

1.24.2 Copper (Cu)

Copper is a very common substance that occurs naturally in the environment and spreads through the environment. Humans widely use copper, for instance it applied in the industries and in agriculture, copper can be released into the environment by both natural sources and human

activities.

1.24.2.1 Health Effects

Long-term exposure to copper can cause irritation of nose, mouth and eyes and it causes headaches, stomachaches, vomiting and diarrhea. Intentionally high up takes of copper may cause liver and kidney damage and even death.

Industrial exposure to copper fumes, dusts or mists may result in metal fume, fever with atrophic changes in nasal mucous membranes.Chronic copper poisoning result in Wilson's disease, characterized by hepatic cirrhosis, brain damage, renal disease and copper deposition in cornea (Fiona.Marshall.2003).

1.24.3 Irons (Fe)

Iron is the most abundant trace mineral in the body and is an essential element in most biological systems and essential for developing aerobic life on earth, iron has ability to donate and accept electrons means that if iron is free with in the cell, it can catalyze the conversion of hydrogen peroxide in to free radicals. About 70% of iron is found in hemoglobin, and about 5% to 10% is found in myoglobin. When bound to normal hemoglobin and myoglobin, iron is in the ferrous (Fe^{2+}) form. Up to 25% of iron in the body is in ferric (Fe^{3+}) form and stored in the liver, spleen and bone marrow.

1.24.3.1 Toxic iron

- Chronic anemia
- Excess iron accumulates in the heart, liver and other vital organs.

Once the body' storage capacity for iron is exceeded, non-transferrin-bound iron is created. This is toxic form of iron that causes oxidative stress,

attacking organ systems at cellular level and causing tissue damage.

Free radical can cause damage to a wide variety of cellular structures, and ultimately kill the cell (Abetz L, Baladi.J.F, Jones, P.2006).

1.24.4 Zinc (Zn)

Zinc is an essential nutrient in humans and animals that is necessary for the function of large number of metalloenzymes.

This enzyme includes alcohol dehydrogenase, alkaline phosphates, carbonic anhydrase, leucine amino peptidase, superoxide dismutase, and deoxyribonucleic acid (DNA) and ribonucleic acid (RNA) polymerase.

Zinc is required for normal nucleic acid, protein and membrane metabolism, as well as cell growth and division. Zinc is also play an essential role in the maintenance of nucleic acid structure of genes.

1.24.4.1 Toxic Effect:-

Symptoms:-

Nausea, vomiting, epigastria pain lethargy.

Zinc deficiency has been associated with dermatitis, anorexia, growth retardation, poor wound healing. Hypogonadism with impaired reproductive capacity, depressed mental function, increased incidence of congenital malformations in infants has also been associated with zinc deficiency in the mothers, and may also have impact on carcinogenesis, though the direction of the influence seems to vary with the agent (American Journal of clinical.2014).

1.25 Objective:

1- Comparative study between sesame oil, groundnut oil (extracted by soxhlet) and sesame oil, groundnut oil (compressed) and which is perfect to the people.

2- Measure the amount of some trace element which contaminated the edible oil.

3- Which is perfect groundnut oil or sesame oil?

Chapter Two

Material and Methods

2 Material and Methods

2.1 Sample preparation:-

Commercial Sudanese peanut oil and sesame oil were extracted by mechanical pressers, bleached and deodorized under industrial processing. The Sudanese sesame oil and peanut oil were extracted by local domestic oil presser, also extracted by petroleum ether (b.p40-60C^0) in a continuous soxhlet apparatus.

Chemical characteristic related to quality (acid value, peroxide value and iodine value) and some physical characteristic (refractive index and density) of oil sample were determined.

Metallic contamination of Sudanese oil samples (sesame, groundnut) were determined by perking Elmer3030 atomic absorption spectrophotometer.

2.2 Materials:-

Petroleum ether, ground samples (groundnut, sesame).

2.3 Methods:-

2.3.1 Oil extraction:-

The ground sample 3.5g was weighed in an empty thimble of known weigh plugged with a piece of cotton wool; then the thimble with the material was placed in soxhlet extractor. A dry and accurately weighed round bottomed flask was fitted to the extractor; then petroleum ether was poured into the flask until it filled approximately two thirds of the flask. The flask, the extractor and the condenser were fitted together, water was allowed to flow through the condenser and heat was continued 4 hours. The apparatus was carefully dismounted and the solvent in the flask was evaporated to dryness in an air.

Calculation:-

Oil% = w_2-$w_{1\backslash}$ s x100

Where = original weight of sample g

W_1 = weight of empty round bottom flask g

W_2 = weight of round bottom flask +oil g

2.4 Physical characteristic of edible oils

2.4.1 specific gravity

2.4.1.1 Pycnometer method:-

The method based upon a comparison of weights of equal volumes of liquid and water at T.C^0.

Procedure:-

A pycnometer was calibrated with distilled water (D.W), the dry pycnometer was filled with dry liquid and the stopper was inserted making sure that the fat column completely filled the capillary in the topper. The specific gravity of the oil was finally calculated.

2.4.2 Refractive Index:-

Refractive index was determined by refractometer. The double prism of the refractometer was opened by means of screw head and few drops of sample were placed on prism. The prism was closed firmly by tightening screw head. The instrument was left to stand for few minutes before reading to equilibrate the sample temperature with that of the instrument. Prisms were cleaned between readings by wiping off oil with soft cloth then with cotton moisture with petroleum ether and left to dry. The refractive index of all sample were determined at 40C^0.

2.5 Chemical Characteristics

2.5.1 Peroxide Value

Peroxide value is used for determine the contents of reactive oxygen of fats and oils in terms of mill equivalents of oxygen per 1000grams of fat and hence for evaluating their keeping qualities.

Reagent:-

Acetic acid glacial–Chloroform – Potassium iodide(KI 15%) -0.01M standardized sodium thiosulphate-starch (indicator).

Procedure:-

3.0g of oil was dissolved in 10ml chloroform and 15ml acetic acid glacial was added together with 1ml of potassium iodide (KI 15%).The flask was quickly stoppered, shaken for 1min and kept away from light for exactly 5minute in dark place.30ml of distilled water was added then added 1ml starch as indicator, blue color was obtain and the liberated iodine was titerated with standarized solution (0.01M) of sodium thiosulphate ($Na_2S_2O_3$) and titrated until the blue color disappears.

Blank determination:-

10ml chloroform+15ml acetic acid glacial+1ml KI +30ml distilled water then added 1ml starch and titrated against $Na_2S_2O_3$.

Peroxide Value = $(V_s-V_b)xNx100/M$

V_s= Volume of thiosulphate (sample)

V_b= Volume of thiosulphate (blank)

N= Normality of $Na_2S_2O_3$

M= Weigh of sample (g).

2.5.2 Acid Value and free fatty acids

The acid value of an oil or fat is defined as the number of mg of potassium hydroxide required to neutralize the free fatty acid in one gram of the sample. The acid is measure of the extent to which the glycosides in the oil have been decomposed by lipase action. The relation between acid value and free fatty acid is 1:0.503%

2.5.2.1 Acid value:-

Reagents:-

Diethyl ether, potassium hydroxide 0.1M-phenolphthalein(ph.ph) indicator

Procedure:-

2.00g of oil was weighted into 250ml conical flask and dissolved in 50ml of (v/v) mixture of ethanol and diethyl ether (ratio 1:1), the solution was titrated against potassium hydroxide 0.1M by using ph.ph until the color was changed to pink color.

Equation:-

$KOH+RCOOH \rightarrow RCOOK+H_2O$

$KOH + HCL \rightarrow KCL + H_2O$

Acid Value = titrant (cm^3)x0.1Mx5.610/wt of sample used, g

2.5.2.2 Free fatty acid:-

Reagent and procedure:-

Method of an acid value

Free fatty acid (as oleic acid) % =a x w x M/10ρ

Where:-

a= number of cm^3potassium hydroxide solution

w= molecular weight adopted oleic acid=282

ρ= weight of sample in gram

M= morality of potassium hydroxide

2.5.3 Iodine Value

Principle:-

Halogens add across the double bonds of unsaturated fatty acids to form addition compounds.

Iodine monochloride (ICL)(wij's solution) is allowed to react with the fat in the dark. The amount of iodine consumed is then determined by titrating the iodine released (after adding KI) with standard sodium thiosulphate and comparing with a blank in which the fat is omitted.

R-CH=CH-R +ICl→ R-CHI-CHCL-R' +ICL (remaining)

ICL+KI→HCL+I_2

I_2+2$Na_2S_2O_3$→2NaI+$Na_2S_4O_6$

The reaction mixture is kept in the dark and the titration carried out as quickly as possible since halogens are oxidized in the light. The quantity of material is calculated.

The iodine number is the number of grammas of iodine taken up by 100g of fat.

Materials:-

1- Fat
2- Iodine monochloride
3- Sodium thiosulphate
4- Starch indicator
5- Stoppard bottle
6- Burette
7- Chloroform

8- potassium iodide 10%

Procedure:-

10ml of fat was pipette and mixed with 10ml chloroform and transferred to a stoppered bottle, added 25ml of ICL solution, stopper the bottle and leaved to stand in the dark for 1hour, after shaking thoroughly. At the same time, set up a blank with all reagents except the fat. Rinse the stoppers and necks of the bottles with 50ml of water, added 10ml of KI solution, and titrated the librated iodine with the standard sodium thiosulphate. when the solution was pale straw color added about 1ml of starch solution and the blue color was appear, continue titrating until the blue color disappeared. The bottle must be shaken thoroughly throughout the titration to ensure that all the iodine was removed.

Iodine value = (v_b-v_s) Nx 126.90/10w of sample g

Where:-

V_b = titeration value of blank

V_s = titeration value of sample

N=Normality of $Na_2S_2O_3$

W= Weight of sample (g)

2.5.4 Saponification value

Principle:-

On refluxing with alkali, glyceryl esters are hydrolysed to give glycerol and the potassium salts of the fatty acids (saops).

The saponification value is the number of milligram's of KOH required to neutralize the fatty acids resulting from the complete hydrolysis of 1.0g of fat. The saponification value gives an indication of the nature of the fatty acids in the fat since the longer the carbon chain the less acid is liberated per

gramme of fat hydrolysed.

Material:-

1- Fat (g)
2- Fat solvent 95% ethanol
3- Alcoholic KOH
4- Reflux condenser
5- Water bath
6- phenolphthalein (ph.ph)
7- Hydrochloric acid (HCL)
8- Burette
9- Conical flask (250ml)

Procedure:-

1.0g of fat was weighted in a beaker and 3ml ethanol was added to dissolve the fat then transferred to conical flask by rinsing the beaker three times with further milliliter of solvent;25ml of 0.5M,alcoholic KOH and attach to a reflux condenser. Set up another reflux condenser as blank with everything present excepted the fat and both flasks was heated on boiling water bath for 30min.Leaved to cool at room temperature and was titrated against HCL by using ph.ph as indicator until get faint pink color.

Saponification value = (b-s) x Mx56.1 / wt of sample (g)

b= volume of blank

s = volume of sample

wt = weight of sample

2.6 Determination of trace element by atomic absorption spectroscopy:

Model: Buck scientific

210 VGP

USA 2005

Atomic absorption spectroscopy is used for qualitative and quantitative determination of elements of atomic methods lei in the part-permillion.

The principle in atomic absorption spectrophotometric analysis based on the estimation of experimental solution in which the atoms are chemically bonded in the reacting flame, the solvent is removed and the chemical bond is broken to form free atoms under the steam of respective hallow cathode lamp lines. These free atoms absorb as definite amount of radiation for specific element, this absorb energy provides a qualitative as well as quantitative determination of a particular element in the matrix. The analysis is based upon calibration curves involving a plot of absorbance versus various concentration of standard solution of the element to be determined.

There are five components of an atomic absorption instrument:

1- The light source that emits the spectrum of the element of interest
2- An "absorption cell" in which atoms of the sample are produced (flame,graphite furnace,MHS cell)
3- Amonochromator for light dispersion
4- Adetector, which measures the light intensity and amplifies the signal
5- A display that shows the reading after it has been processed by the instrument electronics.

There are two basic type of atomic absorption instruments: Single- beam and double-beam.

Reagent:-

Hydrochloric acid (HCL) 20% -Distillid water

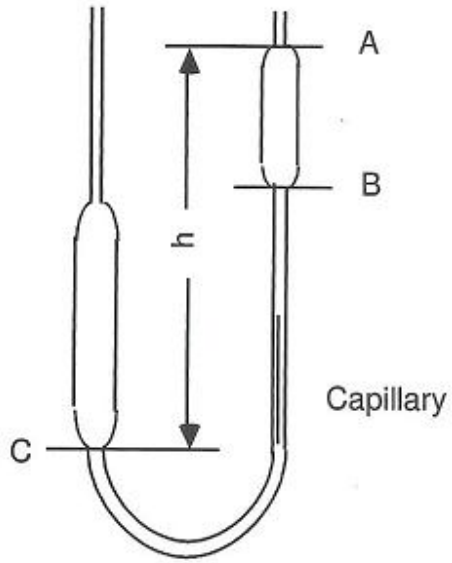
Preparation of the sample:-

2.0g of sample was ashed in muffle furnace at $500C^0$ and after cooled in a discator dissolved in 5ml HCL 20% and then filtrated and complete the volume to 100ml by distilled water.

Analysis:-

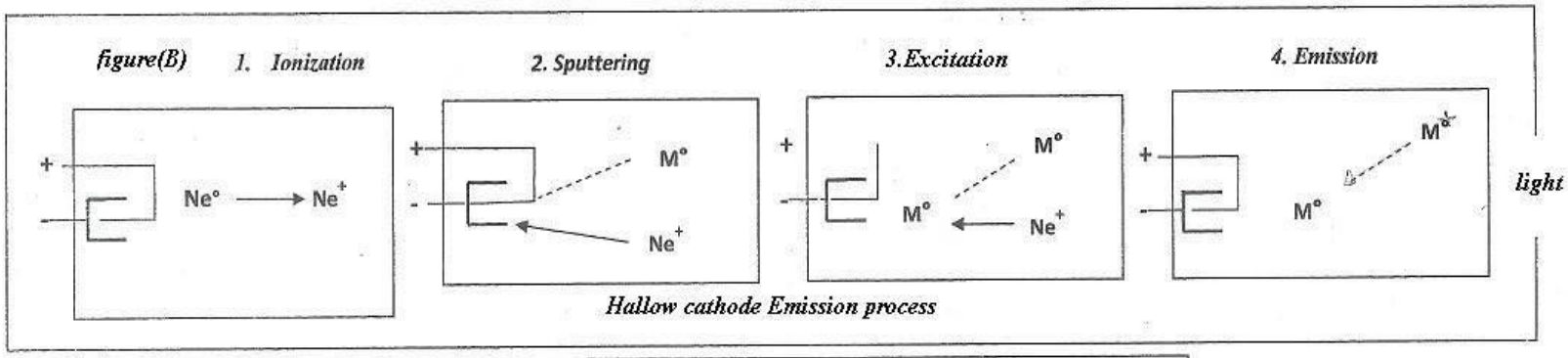
Sample and standard were aspirated in to the flame without dilution or concentration.

Figure 1.20.2



simple viscosity

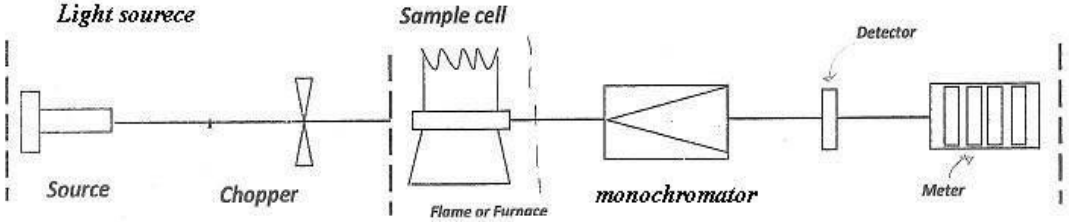
figure2.9 *Atomic Absorption Spectroscopy*



Hallow cathode Emission process

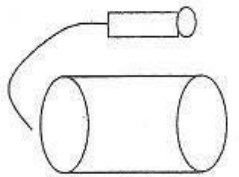
figure(C)

specific light measurement

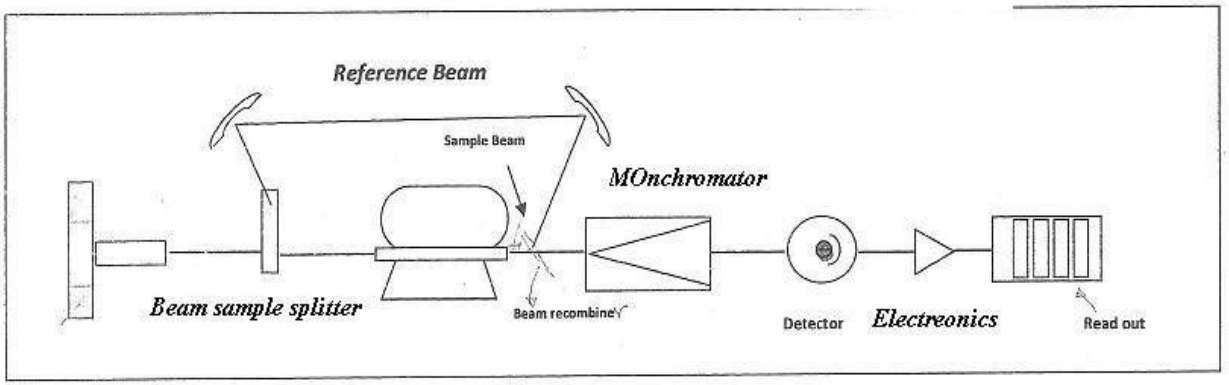


single-Beam Atomic absorption spectrometer

figure(A)



Hallow cathode lamp



Double -Beam Atomic Absorption Spectormeter

Chapter Three

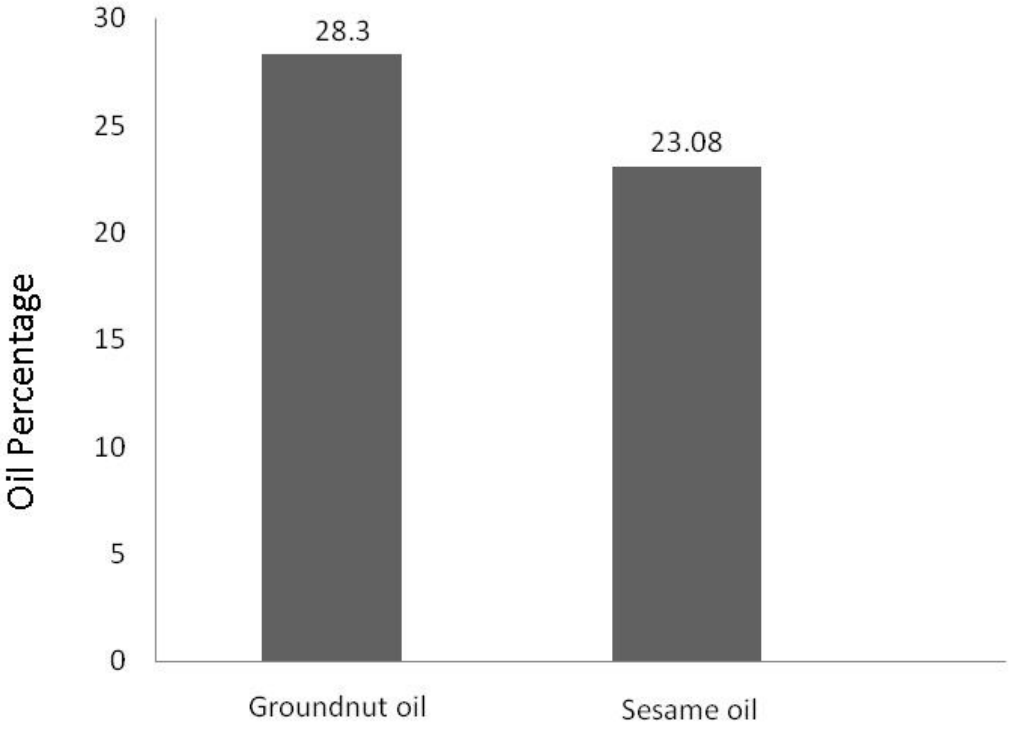
Result and Discussion

Result and Discussion

Determination of chemical and physical properties

Sample	Oil Percentage	Peroxide value	Acid value	Free fatty acid	Iodine Value	Saponification	Specific gravity	Refractive index
Sesame oil (soxhlet)	23.08	9.66	1.07	5.36	225.28	172.34	0.9115	1.47551
Sesame oil (compressed)	-	4.0	0.34	1.69	225.28	185.97	0.9119	1.47551
Groundnut Oil (soxhlet)	28.3	12.6	0.31	1.57	260.18	163.08	0.9094	1.47351
Groundnut Oil (compressed)	-	3.33	0.51	2.24	213.63	178.42	0.90097	1.47351
Groundnut Oil (manu-facture)	-	9.33	1.51	7.61	158.65	200.0	0.9109	1.47351

Figure 3.1 Determination of oil percentage



3.2 Discussion of peroxide value

Peroxide value is increased in sesame oil (soxhlet) 9.66,groundnut oil (soxhlet) 12.6 and groundnut oil (manufacture) 9.33.The oxygen is absorbed at the unsaturated bonds, exposure to light and heat cause significant increase in the peroxide value and indicates a poor resistance of the oil to peroxidation during storage(index of rancidity). Sesame oil (compressed) and groundnut oil (compressed) in the standard range (standard value in sesame oil 7.45, groundnut oil 3.3 (AOAC(1990)).

3.3 discussion of acid value

Acid value increased in sesame oil (soxhlet) 1.07, groundnut oil (compressed, manufacture) 0.34 .1.51respectively indication of rancidity, oxidation and high degree of biological activity and deterioration of non-oily constituents such as carbohydrates and proteins, which may also affect the nutritive value of oils. Sesame oil (compressed) and groundnut oil (soxhlet) in the standard range (standard value of sesame oil 0.38-0.66,groundnut oil 0.42 (Seegeler 1983), and indication of the condition and edibility of oils and the quality of oils.

3.3 Discussion of free fatty acid

Increase of free fatty acid sesame oil (soxhlet, compressed) 5.36, 1.69 respectively, groundnut oil (compressed, manufacture) 2.54, 7.61 respectively, is an indication of the beginning of spoilage and offensive odor and taste in the oil. Decrease of free fatty acid (FFA) in groundnut oil (soxhlet) 1.57 could be result of removal of some fatty materials and free fatty acids during refining process, show that this oil is stable and it makes higher

quality oil. Standard value of sesame oil 0.8% and groundnut 2% (Kirk and Sawyer(1991)).

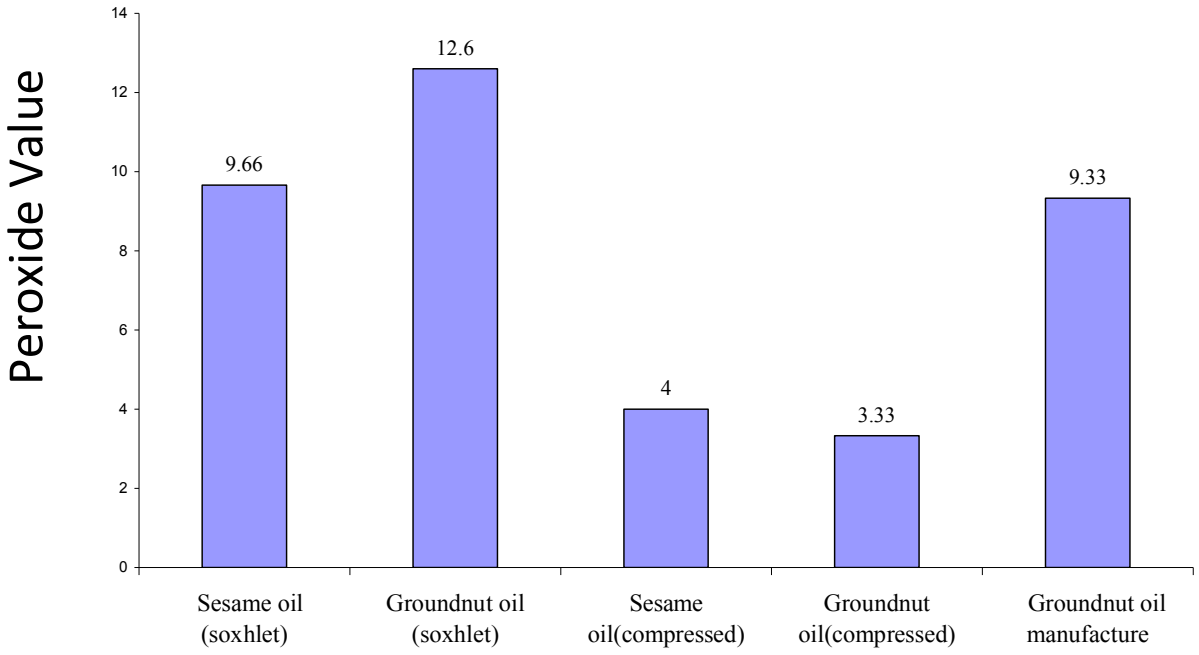
3.4 Discussion of iodine value

Increase in iodine value is indicates of the presence of unsaturated fatty acid and it will increase the rate of oxidation and degree of heat treatment during oil processing (all samples contains high value).Standard value of sesame oil 116mg/100g and groundnut oil 86mg/100g.(Kirk and Sawyer(1991)).

3.5 Discussion of saponification value

Increase of saponification value groundnut(manufacture) 200.0 indicated the presence of greater number of ester bond fatty acids of low molecular weight, this shows that the oil does not use in soap making industry and for thermal stability of poly vinyl chloride(PVC) may be used in liquid soap,shampoos.Decreases in sesame oil (compressed,soxhlet) 185.97 ,172.34 respectively and groundnut oil (compressed,soxhlet) 178.42 ,163.08 respectively may be due to neutralization of fatty acids which result from the hydrolysis of oil. Standard value of sesame oil 188 and groundnut oil 191. (Kirk and Sawyer(1991)).

gure 3.2 Determination of peroxide value



3.6 Discussion of specific value

Increase in specific gravity it determined the purity of edible oils during processing and storage and increase with the degree of unsaturation of the oil. Standard value of sesame oil 0.941 and groundnut oil 0.915 (Kirk and Sawyer (1991)). The increase in specific gravity (Density) might be attributed to oxidation of oil due to the presence of heavy metals Different Refractive index is related to the molecular structure and degree of unsaturation of the oil.

3.7 Discussion of refractive index

The increase in refractive index (sesame oil (soxhlet, compressed) might be attributed conjugation known to precede hydroperoxide formation, polymerization and oxidation. Standard value of sesame oil 1.460 and groundnut oil 1.496.(Kirk and Sawyer (1991)).

Figure 3.3 Determination of acid value

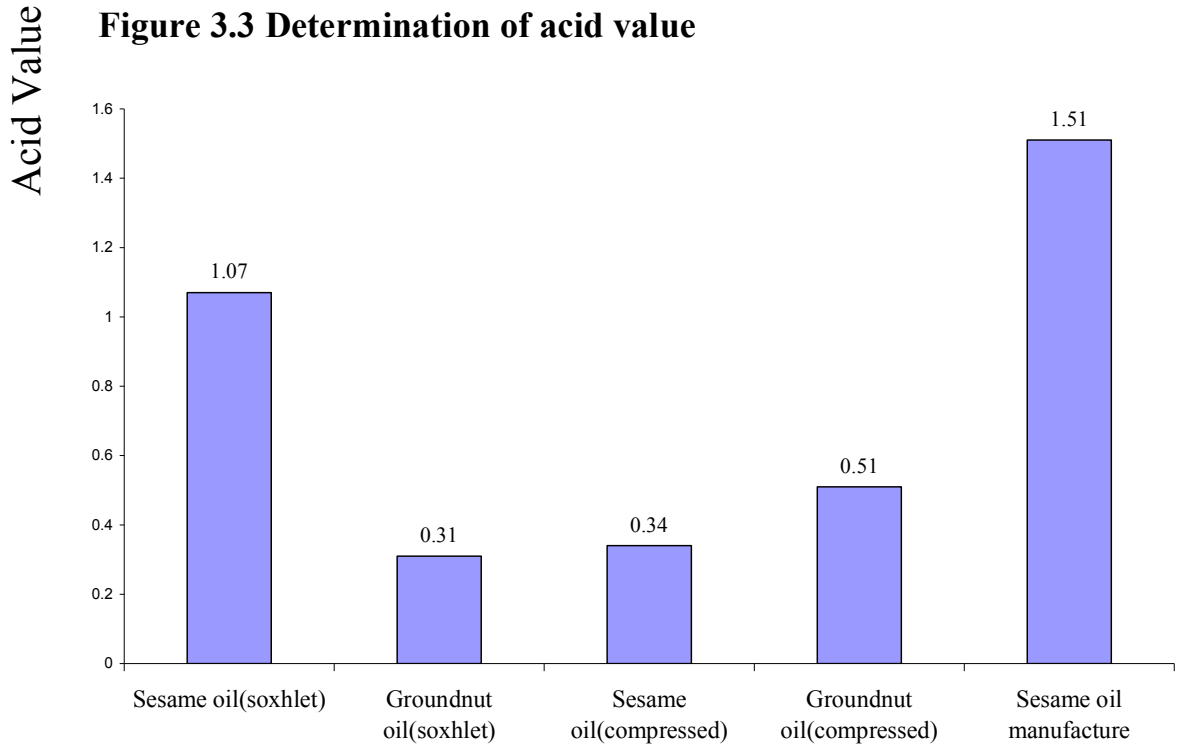


Figure 3.3 Determination of free fatty acid

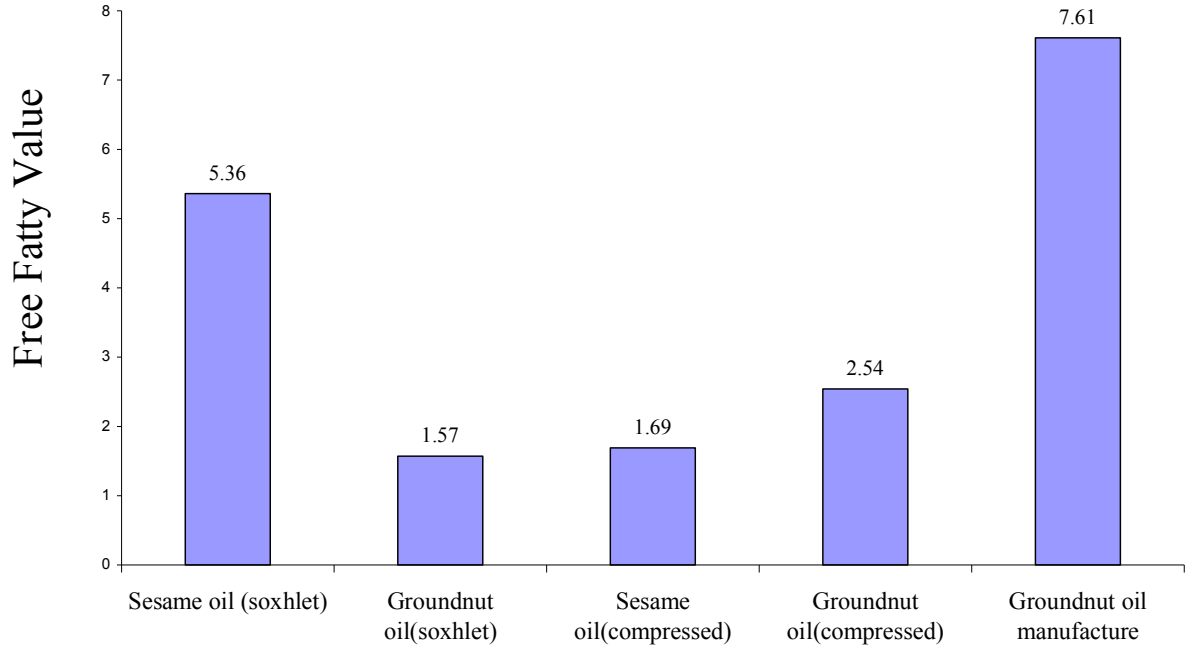


Figure 3.4 Determination of iodine value

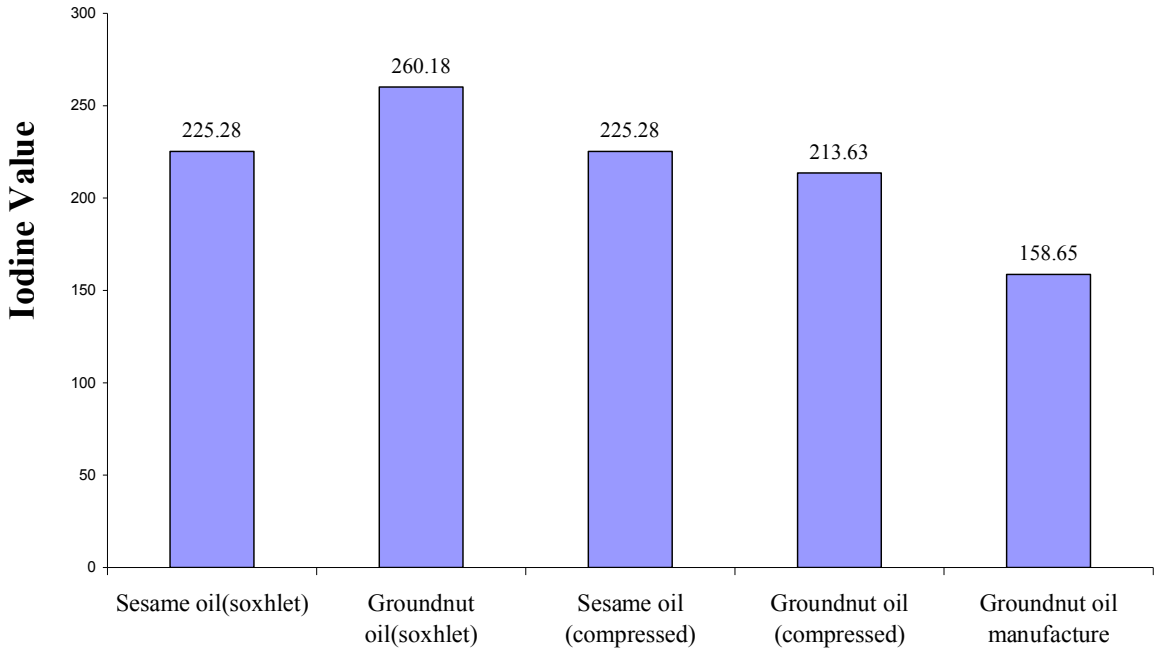


Figure 3.5 Determination of Saponification value

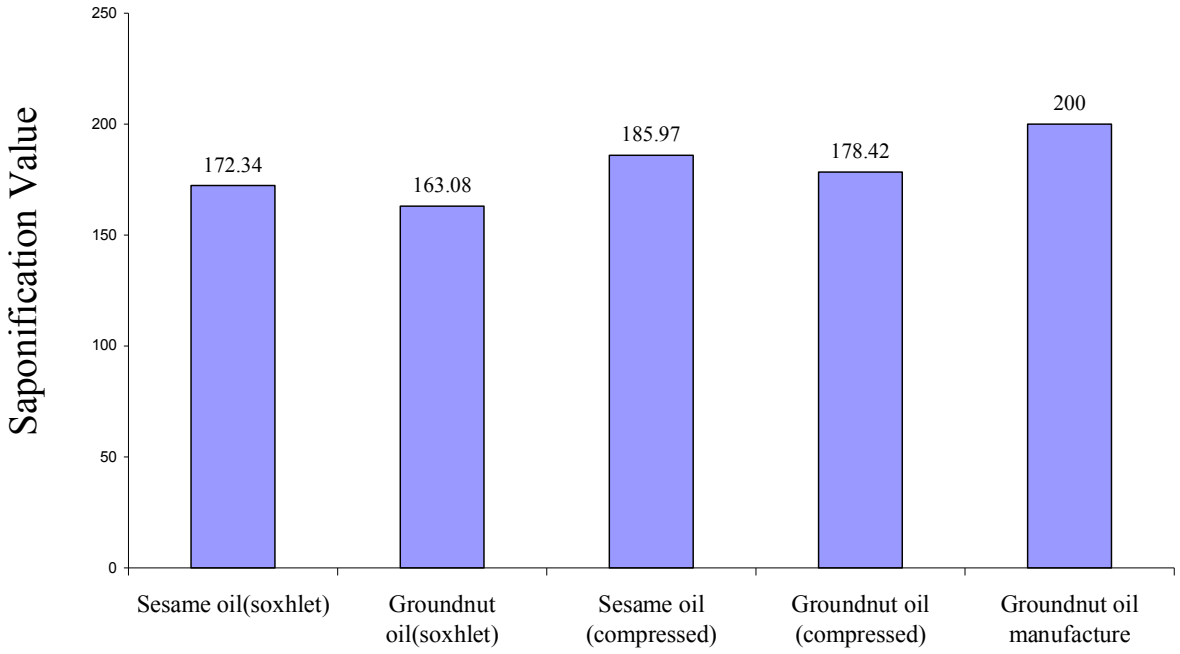


Figure 3.6 Determination of density (specific gravity)

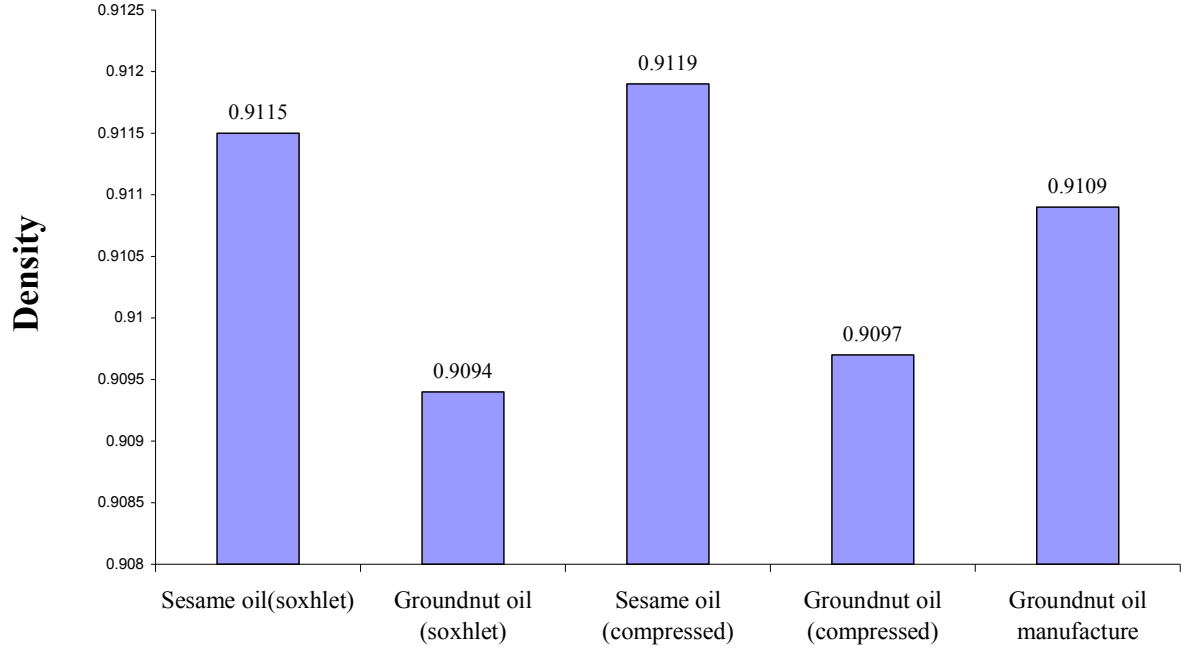
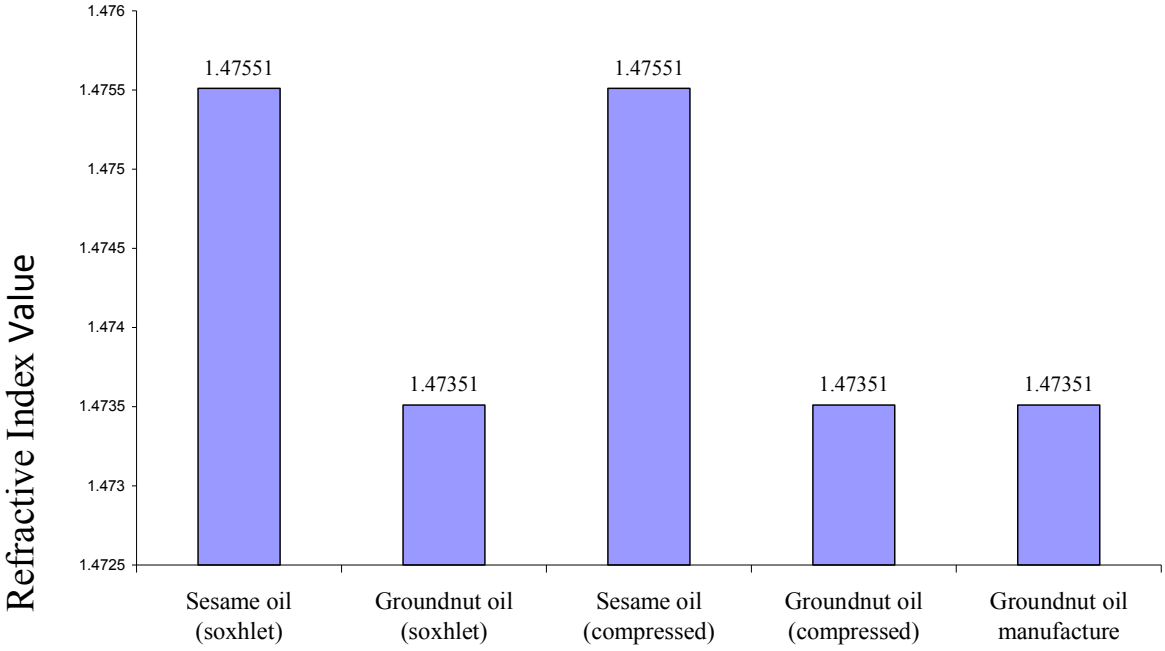


Figure 3.7 Determination of refractive index



3.8 Heavy metal in vegetable oil (contamination)

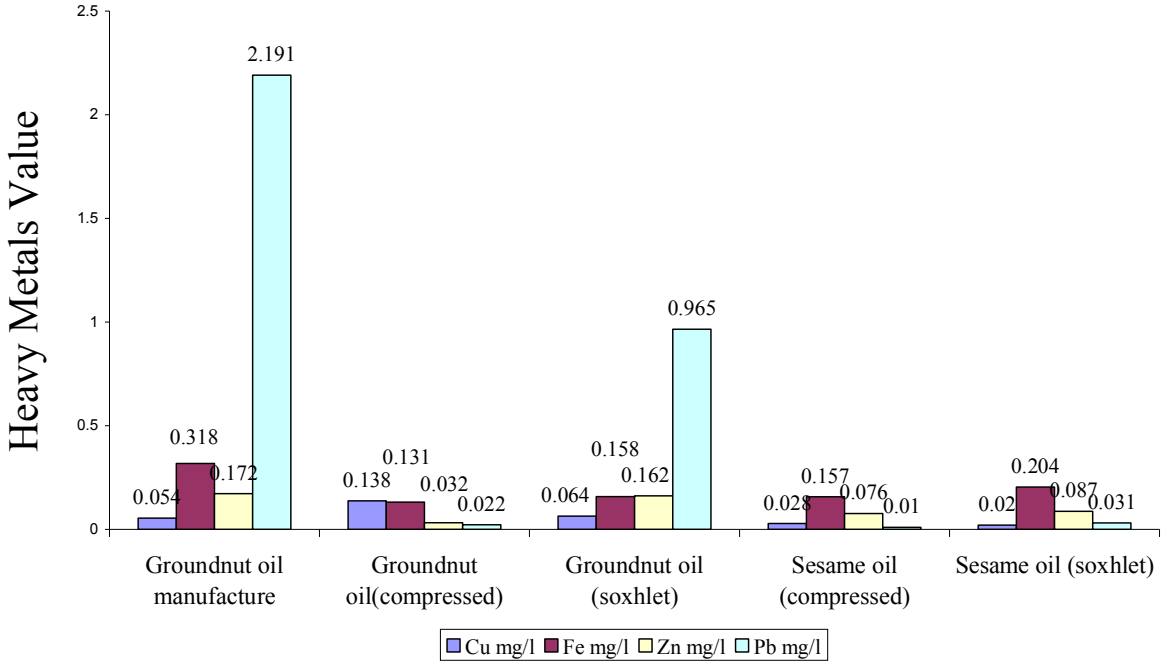
Table 3.8 Change in heavy metals value

Pb mg/l	Zn mg/l	Fe mg/l	Cu mg/l	Sample
2.191	0.172	0.318	0.054	Groundnut oil manufacture
0.022	0.032	0.131	0.138	Groundnut oil(compressed)
0.965	0.162	0.158	0.064	Groundnut oil (soxhlet)
0.010	0.076	0.157	0.028	Sesame oil (compressed)
0.031	0.087	0.204	0.020	Sesame oil (soxhlet)

The range of quality parameters due to Sudanese Standard and Metrology Organization (SSMO).

Heavy metals contaminate vegetable oils from alloys making locally, from the soil, mechanical pressing or during storage. Lead increases in groundnut oil (manufacture) 2.191 mg/l ,groundnut oil (soxhlet) 0,965 mg/l and iron increase in groundnut oil 0.318 mg/l and sesame oil 0.204 mg/l ,during study observed that the groundnut contain high contamination level because it contain aflatoxin (mycotoxin).

Figure 3.8 Determination of heavy metals value



Conclusion

a. The stability of vegetable oils was critically affected by the presence of heavy metals. These harmful effects were investigated by determining the physical and chemical properties of vegetable oils.

b. In presence of heavy metals;(Pb,Cu,Fe,Zn) ,stored vegetable oil (sesame seed, groundnut seed), displayed significant increase in peroxide value in sesame oil and groundnut oil (soxhlet),acid value in sesame oil (soxhlet, manufacture), all samples show increases in iodine value.

c. High level of lead in groundnut oil (soxhlet, manufacture) 2.191 ppm and 0.965 ppm respectively (standard level of each 0.1 ppm maximum.

d. The refining process decreased the impurities (contamination during storage, oil containers or mechanical pressing) which affected the physical and chemical characteristics of oil.

Recommendations

I. Elimination the heavy metal from vegetable oils by reducing their concentration in soil for example use of chelators by adding synthetic chelators such as EDTA (ethylenediamine-tetracetic acid).

II. Improvement the mechanical press.

III. The seed must be storage not more than one year and oil their crystal (bottle) not more than 6 months to avoid the rancidity.

IV. Use the sesame oil because it contain many antioxidant which it has various healthful properties are attributed to the presence of lignans such as sesamin,sesamolin,sesaminol, sesangolin, 2- episalatin,ect..

References

1-AOAC,1990.Official Method of Analysis of the Assocation of official Analytical chemist Inc.Arling.Virginia.USA.

2- Abetz l**.**Baladi J.F,Jones P,Rofail D.The impact of iron overload and its treatment on quality of life:results from a literature review.Health Qual life outcomes.2006;4:73.

3-American Cancer Society.Cancer Fats and Figures.2005.Atlanta:American Cancer Society;2005.

4- Abou-Gharbia.HA,shehata AAY,shahidi,(2000)

Effect of processing on oxidative stabitit and lipid classes at sesame oil.Food.Res Inter 33:331-340.

5-Berg,JeremyMark.Biochemistry,Jeremy M.Berg,John L.Tymoczko,Lubert Stryer. -6th ed.2007 by W.H.Freeman and Company.New york,NY 10010.Houndmills,Basingstoke RG21 6XS,England.

6- Beech,D.F(1996). Sesame SeedsIndustry.In:Australian Grains,ed.B.Coombs Morescope publishing Pty Ltd.PP.414-420.

7-Baldwin John A and Lee R Dewey.1990.Producing high quality seed peannuts.The Coorperative Extension Service Bulletin 1037, The University of Georgia College of Agriclture,Athens,USA.pp 11.

8-Binran Y(1971) A Hand Book of Oil Chemistry,Nippo Yukaaku Kyokai Brito OJ,Nunez N(1982).Evaluation of sesame flour as acomplementry protein source for combination with soy and corn flours.J.Food sci.,47:457-

460.

9- Borchani C.,Besbes S.,Blecker C.H.,and Atti H.(2010).Chemical Characteristic and Oxidation Stability of Sesame Seed,Sesame Paste and Oilve Oil .Journal of Agriclture,Science and Technology.12:585-596.

10- Department Of Agriclture,Forestry and Fisheries Direction Plant production private Bag x144,pretoia,0001-South Africa.

E-mail:DPP@daff.gov.Za.

11-Eromosele.I.C.,C.O.Eromosele,P.Innazo and P.Njerim.1997.Short Communication:Studies on some seeds and seeds oils.Biore sour.Technol.64:245-247.

12- Frankel,E,N,,1984.Lipid oxidation:Mechanisms,Products,Biological significance,J.Am.Oil chem.Soci,61:1908-1916.

13- Fiona-Marshall-2003.Heavy metal contamnation of vegetable oil.Science and Total Enviroment 220:137-145.

14- Food fats and oils.Institute of shortening and Edible oil,Inc Januray 1988.

15- Food and Chemical Toxicology-volume 24,ssues 10-11,october-Novembern 1986,published by Elsevier Ltd.1021-1030.

16- Frei,B.,R.Stocker and N.A.Brue.1988.Antioxidant defenses and lipid peroxidation in human blood plasma.proceding of National a cademy of Science ASU,85:9748-9752.

17- Food Fats And Oils.Inc,Institute of shortening and Edible Oils,1750

New York Avenue,NW,Suite 120 Washington,DC 20006.Ninth Edition-2006.

18- Guston,F.D,Norrs,F.A 1983.Lipid in Foods,chemistry,Biochemistry and Technology.pergamonpress.

19- Ghanem,I.Orfi,M.,shamma,M.(2008) Effect of Gamma Radiation On The Inactivation of Aflatoxin B_1 in Food and Feed Crops.Brazilian Journal of Microboilogy, Vol.39,Oct-Dec,PP.787-791,1517-8382.

20- Gan,H.L.,Y.B.Che Man .C.P.Tan.I.Nor Aini and S.A.H.Nazimah.2005.Characterisation of vegetable oils by surface a coustic wave sensing electronic.nose.Food Chemistry 89:507-518.

21- G.E.LeBlanc and R.A.Secco,High pressure stockes viscometry:A new in-site technique for sphere velocity determination,Rev.Sci.Instrum.,66(10),5015-5018,1995.

22- Hui,Y.H,ed,Bailey's Industrial oil and Fat products.1996.Vol 2,Edible oil and fat product:oils and oil seed,5 Edn,New York,John Wiley and Sons,Inc.p.214 .

23- H.Robert Horton,2006.Principles of Biochemistry,fourth edition.Upper Saddle River,New Jersey 07458.

24- Hashim I, Koehler P.Eitenmller R,Kvien CK.1993.Fatty acid content and tocopherol content of drought streesed florunner peannuts.Peannut Sci 20:21-4 .

25- ISO 3657:2002.Animal and vegetable fats and oils.

26- IBPGR and ICRISAT.1992.Descriptors for groundnut. International Board for Plant Genetic Resources, Rome, Italy: International Crops Research Institute for the Semi-Arid Tropics, Patancheru, India,pp 125.

27- Kochlar,S.P,The british Food Manufacture Industerial Reasrch Association, Scientific and technical survay,1,133(1982).

28-Kirk RS,Sawyer R(1991).Pearson's Composition and Analysis of Foods.9th ED.Longman Sci.Tech.Engl.pp.607-617.

29- Luh,B.S,Feinberg,B,Chung,J.I,and wood root,J.G.1986.Freezing fruit,Ch.7,in commerical treatments(additives are often used in place of bleaching by food processing(EN),P.263,Ari-Publishing.Co,West port.CT.

30- Muller,Kirkus B and Pedersen JI.2001.Serum cholesterol predictive equations withspecial emphasis on trans and saturated fatty acids.An analysis from designed controlled studies.Lipids. 36:783-791.

31- Moure.A.J.M.Crwz,D.Natural antioxidants from residual sources,Food Chemistry 72:145-171.

32- M.K Gupta.2000.Ol Quality improvement through processing,(Introduction to Fat and Oils Technology,2 Edn.Am.Oil Chemists Society Publication, Peoria, Illinis).

33- Mondal,GC, and Nadi,B.1984.Role of fungi on oil quality of stored seed of sesame,J of Food Science 46:1394-1395.

34- Nor,A.I and L,H.M.D.Noor.2005.Interesterification palm products as alternatives to hydrogenation. Asia.Pac.J.Clin.Nutr. 14:396-401.

35- Nes,W.R.Acomparison of methods for the identification of sterols-mass spectroscopy,Enzymology,3,27,Acodemic Press,Orlando,Florida(1985).

36- Ozkan,G.Simesk,B.2007.Antioxidant activity of satureja cilicia essential oil in butter and in vitro-Journal of Food Engineering,79:1391-13996.

37-Oliveira,C.A.F,Goncalves,N.B.,Rosim,R.E,Fernandes,M.(2009) :Determination of aflatoxins in peannuts in the Northeast region of saoppaulo,Brazil.Int.J.Mol.Sci.10:174-183.

38- O'Brian,R.D. Characterization of Fats and Oils, in fats and oils :Formulating and Processing for Applications, second edition.CRC Press,Boca Raton,p8,2004 .

39- Sabate,J. 2003. Nut composition and body weight, American Journal of Clinical Nutrition78:647s-650s.

40- Steen Land,K.,and P.Boffetta.2000.Lead and Cancer in humans.Med.38:295-299.

41- Utilization of natural hybrids in the improvement of groundnut (Arachis hypogea).Expl Agric.,19:355-359.

42- Wiseman , J. 1986. Anti nutritional Factors Associated With Dietary Fats And Oils.Reacet Advances In Animal Nutrition.W.Haresign and D.J.A.Cole,ed.Butterworths,London,U.K.PP:47-75.

43- Younis,Y.M.H.,Malik,M.K.2003.TLC and HPLC,assay of aflatoxin contamination in sudanease peannuts and peannut products.Kuwait J.Sci.Eng,30(1),79-93.