Chapter one
Introduction and Literature Review

1.1 Introduction

Gums are high molecular weight polymeric compounds, composed mainly of polysaccharides capable of possessing colloidal properties in appropriate solvent or agent at low weight (Glicksman, 1973).

Gums are either hydrophobic or hydrophilic. Hydrophobic gums are insoluble in water and include resins, rubber … ect. Whereas hydrophilic gums are soluble in water and can be subdivided to natural semi synthetic and synthetic gums (Glicksman, 1973). Natural gums are those derived from plant and animals. Natural gums of plant origin seem to be associated with plant life process

The Sudan is the world's largest producer of commercial gum it is one of the four important agricultural export commodities, livestock, cotton and sesame. Over the last 20 years, gum Arabic export value amounted to U 40 million annually. UNCIAD, 2013 While there has been government intervention in the marketing of all agricultural exports in the past, (Islam, Phillips, Sljivo, Snowden, and Williams, 1997), gum Arabic is the only one for which government control remain. Small-scale farmers in traditional rain farming areas mostly produce Gum Arabic. They represent 20 percent of Sudan's population and are the poorest.

The impact of the current gum Arabic marketing policy has not been beneficial to this group.(Glicksman and Sand, 1973). This has led to reduced production and consequently exports, declining for the past forty years at an average rate of 2.2 percent per annum. One of the key commitments made by the government of national unity under the joint assessment mission framework was to "abolish the export monopoly "over raw gum Arabic. This commitment has not been implemented. The development of the processing industry over the last three years has resulted in increased domestic competition for raw gum, and in turn better prices paid to farmers as well as more added value captured in Sudan. This positive development comes at a propitious time as increased consumption of soft drinks and confectionary

products, as well as rapid development of health and dietetic products is boosting the world demand for gum Arabic.

Table 1.1 Classification of gums(Ahamed 2012)

Natural gums	**Modified (semi synthetic) gums**	**Synthetic gums**
Plant exudates: Arabic Tragacanth Karaya Ghatti Plant extracts: Pectins Arabinogalactan (larch gum) Plant seed flours Locust Bean Quar Psyllium seed Quince seed Seaweed extracts: Agar Alginates Carrageen an	Cellulose derivatives: Carboxymethcy cellulose Methylcellulose Hydroxypropylmethyl- Cellulose Methylethylcelulose Hydroxypropylcellulose (klucel) Low methoxy pectin Microbial fermentation gums: Dextran Xanthan gum	Vinyl polymers: Polyvinylpyrrolidone (PVP) Polyvinyl alcohol (PVA) carboxyvinyl polymer (carbopol) Ethylene oxide polymers: Polyox

Furcellaran Cereal starches : Seed starches Corn Wheat Rice Waxy maize Sorghum Waxy sorghum Tuber starches Potato Arrowroot	Pregelatinized starches Pregel at inlzed starches Modified starches: Carboxymethyl starch Hydroxethyl starch Hydroxypropyl starch	

1.2. Gum Arabic

Gums Arabic are the most important polysaccharide of commerce, gum Arabic is probably the oldest food hydrocolloid in current use. It was used at least 4000 years ago. Ancient Egyptians used it, largely, in paintings as an adhesive for mineral pigments. Eventually it found its way to Europe through Arabian ports, hence acquired the name gum Arabic. (caius, 1939,wister 1993)

1.3. Definition of gum *Acacia*

Gums are defined as the air hardened exudates flowing naturally from or obtained by incision of the trunk and branches of *Acacia* and other species of *Acacia* of Africa origin(European pharmacopoeia, 1990), FAO/WHO joint Expert committee for food Additives. (JECFA) has developed this definition "gum Arabic as a dried exudation obtained from the stems and branches of

Acacia Senegal (L) willdenow and closely related species. It is a complex polysaccharide of high molecular weight and its calcium, magnesium and potassium salts, which on hydrolysis yield arabinose, galactose, Rhammnose and glucuronic acid. The article of commerce may be further specified as to viscosity (FAO, 1990). These definitions are very broad when it is considered that over 1000 species of Acacia occur worldwide, over100 of which are indigenous to Africa.

1.4. Types of gum:

The genus *Acacia* is the second largest within the Leguminoosae family and contains at least 900 species.(Kull and Rangan, 2008). With their extensive root system, Acacia trees can be found in semiarid areas in Australia, India, and America and mainly in the Sahelian region of Africa. They are multipurpose trees, not only producing gum, but also preventing desert encroachment, restoring soil fertility and providing fuel and fodder. Almost all-commercial gum comes from the so-called gum belt of Africa. A vast area which extends over Mauritania, Senegal, Mali, Burkina Faso, Benin, Niger, Nigeria, Chad, Sudan, Eritrea, Ethiopia, Somalia, Uganda, and Kenya (FAO1995).

Sudan is the world largest producer of gum Arabic, followed by Nigeria, Chad, Mali, and Senegal. Gum from the Sudanese kordofan region is known as the best quality gum. It is used as the standard to judge gums obtained from other areas. (FAO1986).

Commercial gum Arabic is collected from a number of *Acacia* species of which *A senegal, A. seyal*, and *A.polyacantha* are the most widespread in the gum belt. *A. laeta, A. Karoo, and A. Gourmaensis* are some other gum yielding species with a more limited distribution (Islam, *etal.*, 1997). In Sudan, the gums from *A. Senegal* and *A. seyal* are referred to as Hashab and Talha respectively. The former considered of higher quality (Baldwin *et-al.*,1999); most of these species grow scattered in the wild, semi-nomadic people collect Gum from theses untended trees. Cultivation is only practiced for *A. Senegal* particularly in Sudan, wild stands of acacia trees are replaced by monocultures of *A. senegal* in order to facilitate collection and obtain a more consistent quality.

Gum Arabic from other African countries may be variable in quality, because it may contain gums obtained from different species, which occur jointly in the collection area.

1.5The gum belt of Sudan:

"The gums belt, where A. *Senegal* grows naturally, coincides with the area of central Sudan mainly between latitudes 10^0 and 14^0N. The two most conspicuous gum Arabic belt areas outside this limits are the northeast (Faw-Gedaref –Kassala),and in the southeast along the blue Nile /upper Nile state border. the total gum belt area in Sudan amounts to 520,000Km^2, which is equal to one -fifth of the area of the country. A field survey conducted in 1989 indicated that there existed scarcely any *A. Senegal* north of latitude 13^0 45 in kordofan or Darfur (HID& IES, 1990, John, 2002). The gum belt provides a buffer against desertification across the vast region of the Sudan Sahelian one *A. Senegal* provide a variety of valuable economic and ecological functions, such as gum Arabic, fodder for livestock fuel wood and shade.

and massive root system reduces soil erosion and runoff and, a leguminous tree. It fixes nitrogen, which encourages grass and crop growth. The tree is also essential in sand dune fixation. For these reasons, it is the preferred species in bush –fallow rotational and intercropping farming systems in the dry lands of western Sudan (Barbier, 2000).

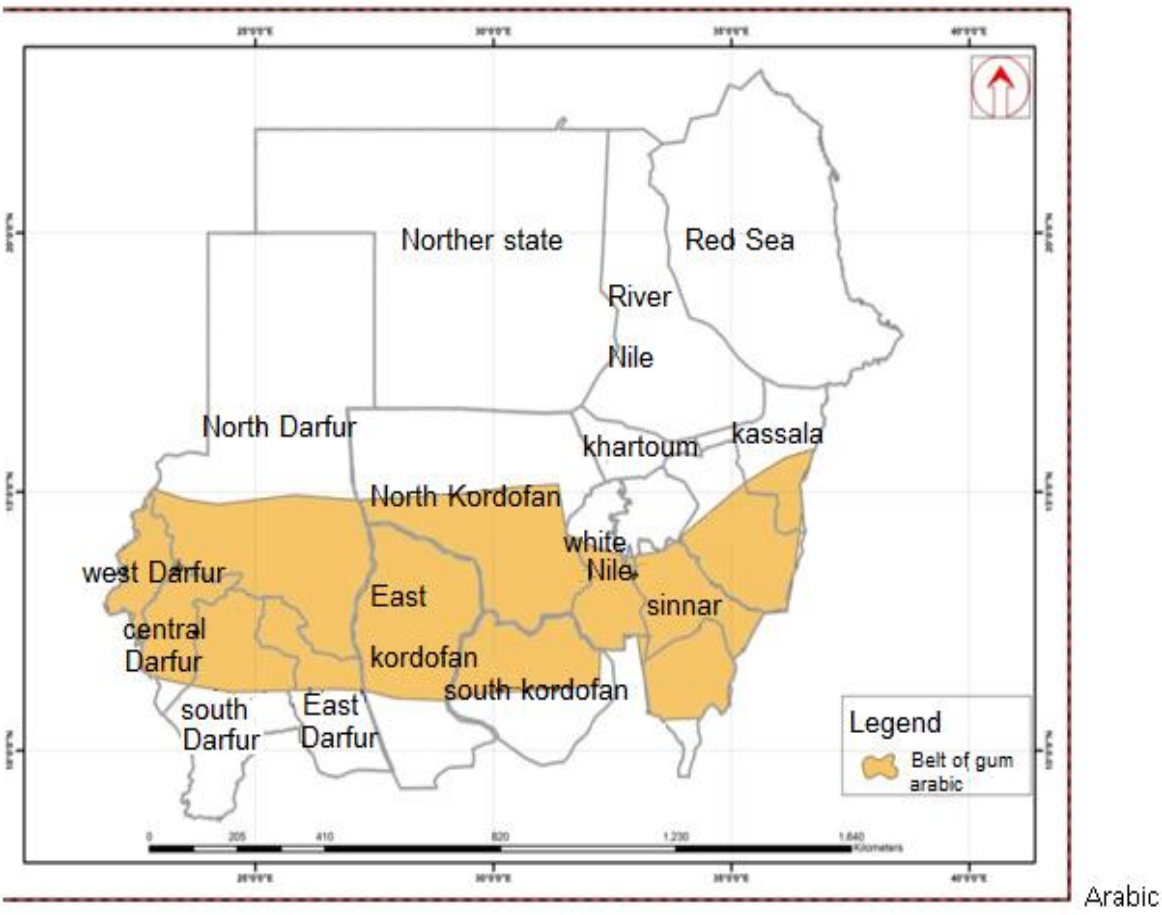


Fig 1.1 gum belt of Sudan

1.6Theories of gum formation.

Gummosis; this word refers to the process by which the tree exudates gum. Gum Arabic is unique in that it produced by tree only when they are in unhealthy condition (Hirst and Jones, 1958, Smith and Montgomery, 1959). So many theories have been proposed to explain gummosis .So some authorities suggested that it might be a product of normal plant life process, most of them believed that it may be a protective mechanism against pathological condition, or it may arise as a result of fungal attack (Smith and Montgomery, 1959)

One theory proposed that it may be a result of starch transformation in to gum, but that is not the fact since Anderson and Dea (1969). found that starch was not present in tissues of excised branches and therefore proposed that the gum has a hemi-cellulose arabinogalactan precursor to which is added rhammnose ,glucuronic acid and 4-0-methyl D-glucuronic acid –terminated

side chain in the final stage of gum formation. However, Miskiel (1990) suggested that gum is formed by enzymatic.

1.7 properties of gums

Gum Arabic readily dissolves in cold and hot water in concentrations up to 50% because of the compact, branched structure and a low viscosity, allowing the use of the high gum concentration in various applications (Dziezak 1991), characterizes therefore small hydrodynamic volume gum Arabic solutions. Solution exhibit Newtonian behavior at concentrations. The pH of the solution is normally around 4.5-5.5, but maximal viscosity is found at PH 6.0.

The other major functional characteristic of gum Arabic is its ability to act as an emulsifier for essential oils and flavours , Gum Arabic has excellent emulsifying properties of it AGP fraction. The hydrophobic polypeptide backbone strongly adsorbs at the oil- water terrace while the attached carbohydrate units stabilize the emulsion by satiric and electrostatic repulsion. Fractionation studies show that ,although emulsifying properties generally improve with increasing molecular weight and protein content, the best results are obtained with mixtures of different fractions (Ray *etal*., 1995) Seemingly the terogeneous nature of the gum makes it an excellent emulsifier .(Buffo *etal.*,2001) found that stability of beverage emulsions is influenced by a number for processing factors, such as pasteurization and demineralization, and by the PH of the emulsion (Buffo *etal.*, 2001). Emulsions can remain stable for long periods of time (several months) prolonged heating of gum Arabic solutions causes the proteinaceous components to precipitate out of solution thus affecting the gums emulsification properties capabilities.

1.8Application of Gums

Emulsifying and stabilizing properties are utilized in the food industry, which is the most important application of gum Arabic. It is also used in medicinal and pharmaceutical fields.(Al-Khalifa, 1996).

1.8.1Gums in food industry

The uses of Gum Arabic in food industry can be summarized as follows (Cossalter, 1991)

1.8.1.1Confectionery

It has been used, widely, in the confectionery industry, where in most cases it has two important functions (a) to retard sugar crystallization and (b) to emulsify the fat and keep it evenly distribution throughout the product. For prevention of sugar crystallization, gum Arabic finds its greats application in confections such as jujubes and pastilles in which sugar content is high and moisture is low. With these products, the technique of incorporating the flavours is extremely important. Usually, gum Arabic is dissolved in water and the solution is filtered, mixed with sugar, and boiled. The flavour is added with a minimum of stirring to prevent formation of bubbles or opaque spots. The second function, as a fat emulsifier, is essential in keeping fat distributed uniformly throughout an oxidizable, greasy film. This property makes gum Arabic extremely useful as an emulsifying agent in caramels and toffees.(Sims,2003).

1.8.1.2 Flavours:

The emulsification properties of gum Arabic are utilized in various liquid flavor emulsions. Many citrus oils and other beverage flavour emulsions utilize the emulsification properties of the gum. When used as a fixative, the superior film forming ability of gum Arabic makes it ideal for protecting the flavour from oxidation evaporation and absorption of moisture from the air. It is equally used as a from stabilizer and agent to promote cohesion of foam to glass (Rao, 2001).

1.8.1.3 Bakery:

Gum Arabic is widely used in the baking industry for its low water absorption properties it is cold water soluble and has impressive adhesive properties for use in glazes and toppings.(Cossalter, 1991).

1.8.1.4 Beverages:

The ability of the gum to stabilize foam is used in the manufacturing of drinks, the gum is also used to clarify wine and fix tanning (Williams, *etal*.,

Gum Arabic suspending and stabilizing properties are employed to suspend insoluble drugs and to prevent the precipitation of heavy metals. Its emulsifying property is used for calomine, magnesia and kaolin suspensions, liquid petrolatum, and cod liver emulsions. Many cough drops and syrups utilize gum Arabic because of its demulcent of soothing characteristics. Gum Arabic is used as an adhesive and binder for pharmaceutical tablets as well as in their coating. Gum Arabic has a long history as additives to ceramic glzes. (Parmalee, *etal*., 200)Also gum Arabic were used for treating kidney disease, the latter is now adopted by the kidney unit in Khartoum, ministry of health (Al-Khalifa, 1996).

1.8.2 Inks and other Industries

Its uses in ink industry have also been reported (Neils *etal.,* 1947). In lithography, gum Arabic is used as a sensitizer for lithographic plates (wood, 1953). also used in electroplating to make the highly active elites on the metal plates allowing more deposition of coatings (Meer, 1980). It has been also used in the preparation of geological specimen slides and in textile industries.

1.9 Sudan gums producing trees

Sudan is endowed with more than 30 Acacia species (Table 1.2), most of which yield gum (EL-amin, 1990), species with greatest distribution that includes, *Acacia Senegal* (Hashab), *Acacia seyal* (Talha), *Acacia polyacantha* (Kakamut), *Acacia laeta* (Shubahi), *Acacia mellifere* (Kitir), *Acacia nilotica* (sunt), *Acacia sieberiana* (kuk), and *Acacia oerfota* (Lao't) (Abdel Nour, 1999).

Table 1.2Sudan gum producing trees

1	*Acacia Senegal var Senegal*	24	*Adansonia*
2	*Acacia Seyal var Seyal*	25	*AfzzeliaAfricana*
3	*Acacia Seyal var fistula*	26	*Albizzlaamara*
4	*Acacia drepanolobium*	27	*Anogeissusschimperi*
5	*Acacia soirocarpa*	28	*Balanitesaegyptiaca*
6	*Acacia Raddiana*	29	*Bauhenia reticulate*
7	*Acacia Stenocarpa*	30	*Bauheniafassoglensis*
8	*Acacia Campylacantha*	31	*Boswelliapapyrifera*
9	*Acacia Sieberiana*	32	*Cassia arereh*
10	*Acacia Arabica*	33	*Ceibapentandra*
11	*Acacia oerfota*	34	*Cordylaafricana*
12	*Acacia usambarensis*	35	*Crataeraadansonii*
13	*Acacia mellifere*	36	*Dannielaoliveri*
14	*Acacia albida*	37	*Isoberliniadoca*
15	*Acacia ethaica*	38	*Khayagrandifoliola*
16	*Acacia ehrenbergiana*	39	*Khayasenegalensis*
17	*Acacia ehyssinica*	40	*Lophiraeleta*
18	*Acacia famesiana*	41	*Moringapterogosperma*
19	*Acacia hecatophlla*	42	*Sterculiacinerea*
20	*Acacia lacta*	43	*Sterculiatomentosa*
21	*Azadirachta indica*	44	*Tomarindusindica*
22	*Combretumsplendens*	45	*Terminaliaspe*
23	*Combretumcollinum*	46	*Terminaliasehimperiana*

1.10 Gum collection in Sudan:

Gum Hashab is collected from *Acacia Senegal* by tapping, whereas all gum *Talha* from *Acacia Seyal* is collected as a result of natural and tap exudation. Tapping begins when the trees are just starting to shed their leaves, around mid of October or the beginning of November. In order to reach this stage, trees have to grow for a period of 3 to 5 years depending on the method of establishment. However, there are two tapping seasons, an earlier one before the onset of the colder weather and a later one in the dry spell after March.

After tapping, exudation occurs, gradually, forming a hard but slightly elastic nodule (N.A.S, 1980; Anonymous, 1980-2008).

1.11Other non-acacia gum

Gum ghatti (Indian gum) is exudates from *Anogessus latifolia*, a tree that is found in India and Sri Lanka. The exudations are natural, but the yield can be increased by making artificial incisions. Gum ghatti occurs naturally as calcium and magnesium salt of a complex polysaccharide acid complex. Acid hydrolysis has shown the gum to consist of L-arabinose, D-galactose, D-mannose, D-xylose and D-glucuronic acid. On dispersion in water, gum ghatti forms viscous solution of viscosity intermediate between gum Arabic and gum karaya. Gum ghatti have emulsification and adhesive properties equivalent to or superior to those described for gum Arabic (Meer *etal.*, 1973).

Gum tragacanth is the dried exudates of several species of the genus *Astaragulus* (family *Leguminoosae*).Astaragulus species is a small perennial shrub with a relatively large tap root, which, along with the branches, is tapped for gum. Gum tragacanth does not dissolve in water, but absorbs a large amount of water and swells greatly to form a soft adhesive gel. The soluble fraction of the gum is a complex mixture of acidic polysaccharides that consists of D-glucuronic acid. On hydrolysis the gum produces D-galactose, 6-deoxy-galactose, and D-xylose. Gum tragacanth powder is white to pale yellow and odorless (Coppen, 1995).

Gum Karaya (Fig.1.2) is the name given to the dried exudation of the

Fig 1.2 Karaya gum sample

Sterculiaurens tree. Almost all Gum karaya comes from India. In the early days of its importation into the United States, many types of gums were introduced which had properties similar to those of Gum karaya (Eljack, 1999) in its monographs of "Drugs, Chemicals and Preparations," "The National Formulary" named the gum Sterculia gum, with the alternate name of Gum karaya. It described Sterculia gum as the dried, gummy exudation from Sterculiaurens (Roxburgh), Sterculiavillosa (Roxhurgh), *Sterculiatra gacantha* (Lindley) or other species of *Sterculia* (*Sterculiaceae* family) or from Cochlospermum gossypinm De Candolle or other species of Cochlospermum.

(*Bixaceal* family).' Howe's established the fact that the gum shipped from India as Gum karaya is obtained from the Sterculiaurens tree.' other gums of the Sterculia type are not collected commercially (Meer, W.1980) .The gum of Cochlospermum gossypinm is very similar to Karaya Gum and is marketed in India (Alamin, 1999).

1.12 *Azadirachta Indica* (Neem)

Neem or Margosa is a botanical cousin of mahogany. It belongs to family Meliaceae. The Latinized of neem- *Azadirachta indica* - is derived from the Persian.

Azad=free

Dirakht= Tree

i-Hind= of India Origin

Which literally means:

'The free Tree of India'

1.12.1 Scientific classification:. (Mukherjee 1955)

Kingdom: pl*antae*

Division*: Magnoli*phyta

Order : *sapindales*

Family: *meliaceae*

Genus: *azdirachta*

Species: *A. indica*

Botanical name(s) *Azadirachta indica* (Puri,1999).

Other names of neem Tree: Miracle tree, Nimba, Arishtha, Margosa.

Synonyms

Antelaea *Azadirachta* (L.) Adelb

Neem tree (*Azadirachta indica*) is one of the very few trees known in India subcontinent (Puri,1999). This tree belonged to Meliceae family, and grows rapidly in the tropic and semi-t ropic climate . It is also observed that this tree could survive in very dry and aird conditions, (Puri,1999). The Neem Tree is an incredible plant that has been declared the Tree of the 21st century by the United Nations (Puri,1999). In India it is variously known as "Divine Tree", Life give tree nature's Drugstore village pharmacy and panacea for all disease . It is one of the major components in Ayurvedic medicine, which has been practiced in India since many centuries .

Extracts from the neem tree (*Azadirachta indica*. A juss) also called Dogonaro in Nigeria are most consistently recommended in ancient medical texts for gastrointestinal upset, diarrhoea and intestinal infections, skin ulears and malaria (Schmutterer, 1995).

All parts of Neem plant such as leave, bark, flower, fruit, seed, root and gum have advantages in medical treatment and industrial products, Its leaves can be used as drug for diabetes, eczema and reduce fever . Barks of neem can be used to make tooth brush ant roots has and ability to had diseases and against insects (Puri, 1999).

The seeds of neem tree has a high concentration of oil .Neem Oil is widely used as insecticides, lubricant, drugs for variety of diseases such as diabetes and tuberculosis (Puri,1999; Ragasa *etal.*,1996).

India encouraged scientific investigation on neem tree as part of program to revitalize India tradition and also increase commercial interest on neem (Stix, 1992). And presently some authors believe that no other plant or tree in the world has been so extensively researched or used in all possible capacities so for In Africa, extracts from neem leaves have provided various medicinal preparations (Ekanem, 1971, Udeinya, 1993). Neem plant (*Azadirachta indica*)

has been of great benefit in human health due to its biochemical, pharmacology, and medicinal properties.(Anna, 2006)

Fig1.3 Neem Tree

1.12.2Uses of neem tree

The neem tree is a tropical evergreen tree native to India sub-continent (Roxburgh, 1874); it has been used in Ayurvedic medicine for more 4000 years due to its medicinal properties. Most of the plant parts such as fruits, seeds, leaves, bark, and roots contain compounds with proven antiseptic, antiviral, antipyretic, anti-inflammatory, antiulcer and antifungal uses .It has great potential in the field of pest management, environment protection and medicine. Neem is natural source of eco-friendly insecticides, pesticides and agrochemicals. The tree has adaptability to a wide range of climatic,

topographic and edaphic factors . It thrives well in dry, stony shallow soils and even on soils having hard clay pan, at a shallow depth. Neem tree requires little water and plenty of sunlight (Sateesh,1998). The tree grows naturally in areas where the rainfall is in the rang 450 to 1200mm. However, it has been introduced successfully even in areas where the rainfall is as low as 150 to 250mm. Neem grows on altitudes up to1500mm (Jattan *etal.*, 1995;Chari, 1996) . It can grow well in wide temperature range of 0 to 490C .It cannot withstand water-logged areas and poorly drained soil. The Ph range for the growth of neem tree lies in between 4 to10. Neem trees have the ability to neutralize acidic soils by a unique property of calcium mining (Hegde,1995).

Neem or Margosa is a botanical cousin of mahogany . It belongs to the family Meliaceae The Latinized name of neem- *Azadirachta indica* – iderived from the Persian : Azad=free, Dirakht=Tree, i –Hind= of India .

Biologically active materials isolated from different parts of the plant include: azadarachtin, melaicin, gedunin, salanin, nimbin, valassin and many other derivatives of these principle, Meliacin forms the bitter components of neem seed oil; the seed also contain tiginc acid (5-methyle-2-butanic acid) responsible for the distinctive odor of the oil (Schmutterer, 1990; Uko and Kamalu,2001; Lale, 2002). These compounds belong to natural product called triterpenoids (Limonodies). The active principles are slightly hydrophilic, but freely lipophilic and highly soluble in organic solvent like, hydrocarbon , alcohols, ketones and esters . (Schmutterer and Singh, 1995).

The neem tree |(*azdirachta indica* A. Juss.) has been known as the wonder tree for centuries in the Indian subcontinent . It has become important in the global context today because it offers answers to the major concerns facing mankind . Neem tree (*Azadirachta indica*) neem is versatile tree, it is considered to be one of the most promising trees of 21 century .It has great potential in the pest 2management, environment protection and medicine .Also it has showing reappraise as potential fertilizer .

The history of the neem tree is inextricably linked to the history of the India way of life. Although the antiquity of neem is shrouded in the mists of time, this evergreen robust looking tree has long been cherish as a symbol of health in the country as a symbol of its origin. It has, for a very long time, been a friend and protector of the villager .

(http://www.neemfoundation.org).

Neem is a fast growing tree that usually reaches a height of 15-20 m, and under very favorable condition up to approximately 30-35 m. As rule it is an evergreen tree, but under extreme circumstances, such as extended dry periods, it may shed most of nearly all of its leaves. The branches spread widely. The fairly dense crown is roundish or oval and may reach diameter 12-20 m in old free standing specimens. (Rajeev, Seenappa, 2009).

The trunk is relatively short, straight and may reach a girth of 1.5-3.5 m.

The bark is hard fissured or scaly and whitish-gray to reddish-brown. The sap wood is grayish-white and the heart wood reddish.

The root system consists of a strong taproot and well developed lateral roots.

The lateral surface root may reach over 18 m. Vesicular-arbuscular mycorrhiza (VAM) is associated with the rootlets categorized neem as a highly VAM dependant plant species .

The leaves are unpaired, pinnate, 20-30 cm long and the medium to dark green leaflets, which number up to 31, are approximately 3-8 cm long. The terminal leaf is often missing. The petioles are short. The shape of mature leaflets is more or less asymmetric.

Natural hybrids between *A. indica* and *A. Siamensis*, found in Thailand on places where both species grow together, have an intermediate position regarding the shape and consistency of the leaflets.

The white gragrant flowers are arranged in auxiliary, normally more or less drooping panicles which are up to 25 cm long. (Puri,1999).

The glabrous fruits are olive-like drupes which vary in shape from elongate ovea to nearly roundish and when ripe are 1.4-2.8×1.0-1.5cm . They are green when young and yellowish-green to yellow, rarely reddish when mature. The fruit skin (exocarp) is thin and the bitter-sweet pulp (mesocarp) is yellowish-white and very fibrous. The mesocarp is 0.3-0.5 cm thick. The white hard ‘shell (endocarp) of the seed encloses one, rarely two and very rarely three elongated seed kernels having brown testa. (Ogbuewu, 2008).

1.12.3Distribution:

Two species of *Azadirachta* have been reported, *Azadirachta indica* A. Juss- native to Indian subcontinent and *Azadirachta excelsa* Kack. Confined to Philippines and Indonesia (Jattan *etal.*,1995; Hgde, 1995). The former grows as a wild tree in India, Bangladesh, Burma, Pakistan, Sir lanka, Malaysia, Thailand Indonesia. Presently neem can seen growing successfully in about 72 Countries worldwide, The neem native of Indian subcontinent; it is widely distributed by introduction, mainly in the drier (arid) tropical and subtropical zones of Asia, Africa, the Americas, Australia and South Pacific island. In India it is widely disrupted in many states. In Myanmar it is very common in the central parts of the country . In the South Pacific Neem occurs in the Fiji Islands. In Australia it was first introduced about 60-70 years ago. In Indonesia, neem exists mainly in low-lying northern and eastern parts of Java and in the friar island to the east (Bali, Lombok, and Sumbawa). In the Philippines it was introduced during the seventies and eighties of the last century . In China, *A.indica* was planted on subtropical island of Hainan and southern china, In Nepal neem trees are found in the southern, low-lying areas (Tarai region). In Sir Lanka it is widespread in the drier northern parts of the island.

In Qatar and Abu Dhabi neem was planted under irrigation using desalted seawater along avenues and parks. A large plantation was established on the Arafat plains near Makah to provide shade for Pilgrims (Ahamed *etal.*, 1995).

1.12.4 Neem in Sudan.

The neem (*Azadirachta Indica* A. Juss) trees have been grow successfully in all parts of Sudan. Neem has became a naturalized species in various parts of the Sudan. (Elteraifi *etal.*, 2001). In Sudan, neem which was introduced in 1921 is frequent in Kassala, in threats in towns and village along the blue and the white Nile irrigated areas of central Sudan and rain fed regions in kordofan and Darfur (Schumutterer, 1995). Neem tree occurs throughout Sudan; it is performance is quite good even in the harshest condition. Most of the original plantation were carried out by the colonial officers along the railway and the Nile banks. Then they spread all over the country . The neem tree is noted for its drought resistance. Normally it thrives in areas with sub-humid condition, with an annual rainfall between 400 and 1200mm. It can grow in region with an annual rainfall below 400mm, but in such cases it depends largely on ground water . neem can

grow in many different types of soil, but it thrives best on well drained deep and sandy soil. It is a typical \ subtropical tree and exists at annual mean temperatures between 21-32^0 C. It can tolerate temperatures below 40^0 C. Temperatures is one of the most important factors affecting seeds. Water up take, gas diffusion, respiration and other metabolic processes all proceed faster at higher temperatures. Germination is dependent on all these processes and thus is, strongly, affected by temperature.

1.12.5 Chemistry of neem

To give a brief background, chemical investigation of Neem were undertaken by Indian pharmaceutical chemist in 1919. Whereby they isolated acidic principle in neem oil, which they named as ‘margosic acid”. However, real chemical research originated in 1942 with isolation of three active constituents, viz, nimbin, nimbidin, and nimbinene. In 1963 an Indian scientist, extensively, examined the chemistry of the active principle of neem following the discovery of neem kernel as locust feeding deterrent. Its chemistry has grown considerably. Several compounds have been isolated and characterized. The main feature is that most of them are chemically similar and biogenetically derivable from a tetracyclicterpenes. These are also called liminoids (azadarachtin, meliantrol, salanin etc).|(Govindachari, T. R., 1992) bitter principles and occur in other botanical species as well (Rutacease and Simaroubaceae). The unraveling of high complex structural features and biogenetic interrelationship represent classic piece of work on natural product chemistry . from the practical side these compounds also exhibit a wide varty of biological activity, for example, pesticides, antifeedants, and cytotoxic properties. Mordu (Luniz) and A. Blackwell (1993).

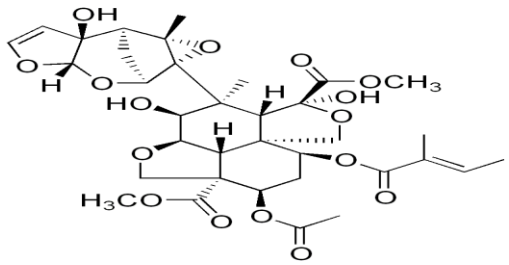


Fig 1.4 Azadarachtin

1.12.6.The chemicals classified are:

- Nimbin: anti-inflammatory, anti-pyretic, anti-histamine, anti-fungal
- Nimbidin: anti-bacterial, anti-ulcer, analgesic, anti-arrhythmic, anti-fungal
- Ninbidol: anti-tubercular, anti-protozoan, anti-pyretic
- Gedunin: vasodilator, anti-malarial, anti-fungal
- Sodium nimbinate: diuretic, spermicide, anti-arthritic
- Quercetin: anti-protozoal
- Salannin: insect repellent

Azadarachtin: insect repellent, anti-feed ant, anti-hormonal

Other chemicals that form its therapeutic value are:

- Limonoids
- Terpenoids and steroids
- Tetranortarpenoids
- Fatty acid derivatives like margosinone and margosinolone-
- Coumarins like scopoletin, dihydrosocoumarins
- Hydrocarbons like docosane, pentacosane, hetacosane, octacosane etc.
- Sulphur compounds
- Phenolics
- Flavonoglycosides
- Tannins

The highest concentrations of the active ingredients are found in the seed and oil, however the active ingredients are also found in lesser amounts in the bark and the leaves.(Anna, 2006)

1.12.7Application of neem

Products made from neem have been used in India for over two millennia for their medicinal properties. They are said to be antifungal, ant diabetic, antibacterial, antiviral, contraceptive and sedative. Neem products are also used in selectively controlling pests in plats. Neem is considered a part of Ayurvedic medicine .

- All parts of neem are used for preparing many different medicines,especially for skin disease.(Zillur, *etal.,* 1996)
- Part of neem tree can be used as a spermicide.
- Neem oil is used for preparing cosmetics (soap and shampoo, as well as lotions and other), and is useful for skincare such as acne treatment. Neem oil has been used effectively as a mosquito repellent.(Sudaravalli *etal.,*1952)
- Neem useful for damaging over 500 types of insects, milts, ticks and nematodes by changing the way they grow and act. Neem does not normally kill pests right away, rather it slows their growth and drives them away. As neem product are cheap and not poisonous to animals and friendly insects, they are good for pest control.

IN The UK, plant protection product that contain azadarachtin, the active ingredient, of neem oil are illegal.

1.12.8 Medical Properties of neem

- For thousands of years the beneficial properties of Neem (*Azadirachta indica* A. Juss) have been recognized in the Indian tradition. Each part of the neem tree has some medicinal properties (Biswas *etal.,* 2002).
- Traditionally Neem was used in Ayurveda for a number of conditions. It is one of the main ingredients in every blood purification formula used in Ayurveda and it appears in most diabetic formulas as well. It is also used for arthritis, rheumatism, the removal of external and internal parasites, including malaria and fevers and as an insect repellent. Neem tree beside it use in

medicine, the neem tree is great importance for its anti-desertification Properties and possibly as good carbon dioxide sink.

- Leaf: Leprosy, skin problems, skin ulcers, intestine worms, anorexia, eye problems, epistaxis, biliousness
- Bark: Analgesic, curative of fever
- Flower: Elimination of intestine worms, phlegm, bile suppression,
- Fruit: Diabetes, eye problem, piles, intestine worms, urinary disorder, wounds, leprosy, epistaxis
- Twig: Asthma, cough, piles, intestine worms, obstinate urinary disorder, phantom tumor, spermatorrhoea
- Gum: Scabies, wounds, ulcer, skin diseases
- Seed: Intestine worms and leprosy
- Oil: Intestine worms, skin diseases and leprosy
- Root: Refrigerant, diuretics

(Stix 2008)

1.12.9 *Azadirachta indica*(Neem)gum

Azadarichta Indica gum, which belong to the family of galactan gums(Anderson, 1971), it is very complex condensate of hetero polysaccharide and protein. The proteins are tightly linked to the polysaccharide, which constitute the major component. Drastic degradation of a smaller gum complex component show that it contains D-glucose, D-glucuronic acid, L-arabinose, L-fucose, mannose and xylose. Investigation of amino acid composition of the gum show a aspartic acid as the most abundant, aspartic acid in *Azadirachta indica* gum were reported by few authors (Anderson DMW, Hendrie A,1971). In addition to that it also found to contain organic fatty acids (Zechmester,1999). *Acacia* gum (gum Arabic) consists of a group of macro molecule characterized by a high proportion of carbohydrates (approximately about 97%), which are predominantly composed of D-galactose and L-arabinose unites and low proportion of proteins (< 3%) .(Islam, *etal.*,1997) (Montenegro, *etal.*,2012).

However, neem gum has unusual structural features in that it contains appreciable amount of D-glucosamine and protein (Ushalakshmi *etal.*,1967)un like other plant gums. *Azadirachta indica* gum (Neem gum) occupies a special position among plant gums in that, it contains about one-third of its weight as protein (Anderson and Henrie, 1971), the highest concentration reported for any plant gum. Thus, neem is an excellent experimental material for the study of the biological activities of proteins in exudates gums. (Anderson *etal.*, 1968).

In the present study, the morphological, compressional, disintegrant and mechanical properties of neem gum obtained from the trunk of *Azadirachta indica* (A. Juss) tree were evaluated in comparison with standard gum binder

Fig 1.5 neem gum(trunk)

Fig 1.6 neem gum

1.12.10. Origin

Many plants exudates viscous, gummy, liquids, which exposed to air and allowed to dry, from clear, glassy masses. The shapes of these masses vary from spherical, tear-drop balls typical of gum producing by *Azadirachta indica* trees. The coloures of these exudates also vary widely from almost clear white to dark brown, depending on the species, climate, soil, and adsorbed impurities.

1.12.11. General Description

Gum is a byproduct obtained as result of certain metabolic mechanism of plants and trees.

Neem gum is a natural exudates from Neem tree by induced or natural injury. The neem bar, due to some internal activity discharges clear, bright and brown- coloured gum material non- bitter in taste and is soluble in cold water. The gum is a multipurpose by product. Natural gums obtained from viscous solution. Neem has been commercially tapped for using its gum which is of use in large number of industries. It is being grown on a large scale basis for using all its parts, no wonder it is called a "Universal Tree" having a cure for almost everything. It has been used, traditionally, as an adhesive for painting. It is used as a bulking agent and for the preparation of special purpose food . (Joneja .S.K., Harcum w.w.,1999).

1.12.12 Chemical characteristics

Neem gum gives on hydrolysis L-arabinose, L-fucose, D-galactose, and D-glucuronic acid. The aldobiuronic acid component of the gum obtained by graded hydrolysis is shown be 4-0-(D-Glico pyranosyl uronic acid) D-galacto pyranose. Information on the structure of the gum has also been obtained by period ate oxidation studies. Neem gum atypical plant gum is the salt complex polysaccharide acid (K.M.NadKarni,1927). The gum acid, obtained from acidified aqueous solution of the gum by precipitation with alcohol, is a white amorphous powder which has an equivalent weight of 1080.(Mukherjee *etal.*,1955) This communication is concern with the composition of the neem gum and with the determination of the structure of an aldobiuronic produced when the gum is hydrolyzed with acid. Complete hydrolysis of the gum followed by partition chromatography and he preparation

of the crystalline derivatives has shown that the gum contains L-arabinose, L-fucose, D-galactose, D-glucuronic acid and traces of D-xylose. The ratio of D-galactose to L-arabinose proved to 3:2. By mild acid hydrolysis, the arabinose and fucose units indicate that they were present in the gum in farinose form.

More drastic hydrolysis of the gum afford D-galactose and an aldobiuronic acid **(**Ramakrishna ,*etal.,* 1981) composed of unit of D-gulocuronic acid and one of D-galactose**,** the structure of the aldobiuronic acid was established as follows, upon methylation of the barium salt of an octa-o methyl –D galactose and 2,-3,4-tri-o-methy –Dglucuronic acid, the former being identified as crystalline ÿ-Lactone and the later as 1,5-Lactone 6-mtheyle ester (J.K.N, Jones and F. Smith1949). These fact prove that the structure assigned to the aldobiuronic acid is correct, a view supported by the further observation that oxidation with period ate result information of 2-3 moles of formic acid and 0.5 mole of formaldehyde per mole of aldobiuronic acid. (Gill., *etal*., 1025). When the gum its self was oxidized with period ate . 2 moles 0f formic acid were produced by equivalent weight of the gum and chromatographic analysis of the oxidation gum, after hydrolysis, shown that certain galactose units had survived period ate oxidation. This evidence demonstrated the, highly, branched chain character of the gum and that the branching in the, molecular, complex is located at those galactose units of the gum which are not affected during period ate oxidation.(Osuna,2000).

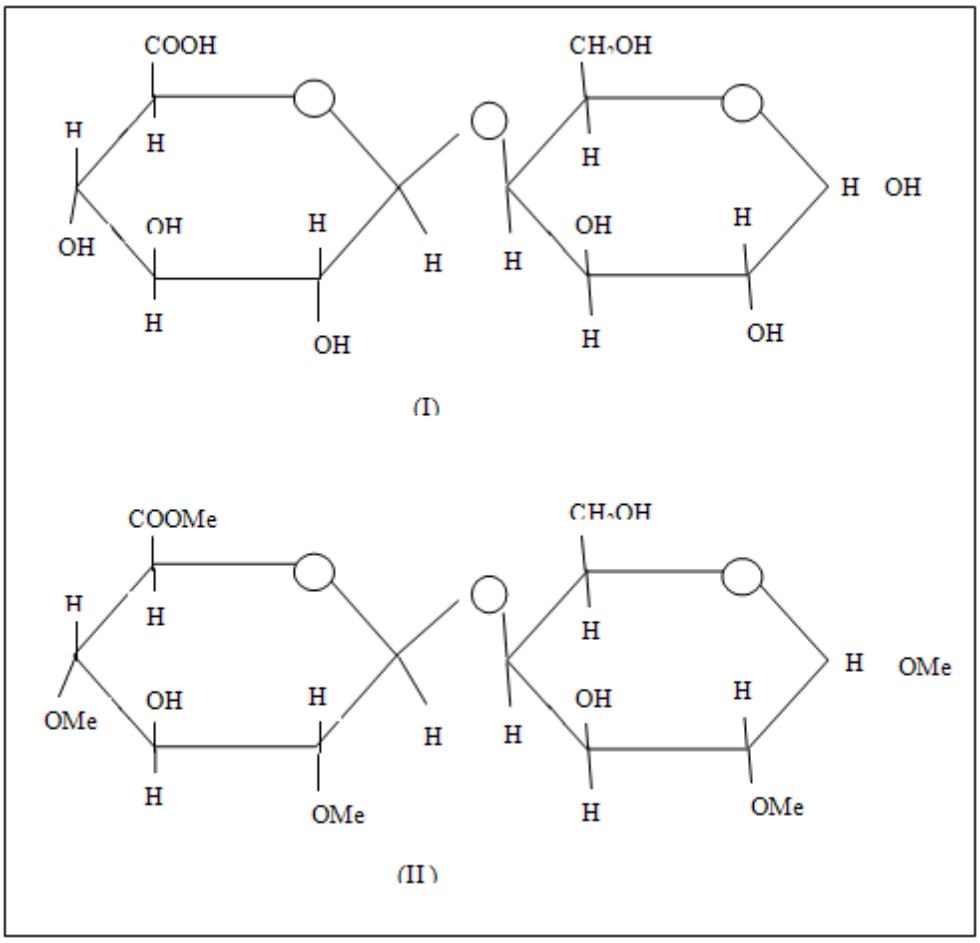


Fig 1.7chemical characteristic of neem gum (Mukherjee 1954)

1.12.13Application of neem gum

Pharmaceutical industry: used in antiseptic creams, tablet binder, and coater.

- Food industry :used as stabilizing agent, and thickening agent.
- Cosmetic industry: used in facial masks, lotions, face powder, protective creams.
- Textile industry : used in dyeing and printing of fabrics.
- Paper industry : used as an adhesive and strengthening the paper.
- Bakery: *Azadirachta indica* gum used in the baking industry for its low water absorption properties, its cold water soluble and has adhesive properties

- Personal Hygiene industry : used in soap, tooth paste, tooth powder.(Satya Narayan, V.; Pattabiraman 1973).

1.13Physicochemical properties of gum

1.13.1Solubility:

Gums can be classified into three categories with regard to their solubilities:

1. Entirely soluble gums: e.g. A. *senegal*, A. seyal. *Azadirachta indica* gum

2. Partially soluble gums: e.g. *Gatti* gum.

3. Insoluble gums: e.g. *Tragacanth* gum (Omer, 2004)

1.13.2Colour

The colours of gums vary from water- white (colourless) through shades of yellow to black. The best grades of gum are almost colourless with slight traces of yellow; some possess pink likes (Siddig, 2003). On the other hand dark or even black gums sometimes occur e.g. mesquite gum. There are also the pale rose pinks, darker pink and yellowish gums. The pink colour is probably due to the presence of different quantities of tannin materials (Omer, 2004).

1.13.3Shape

Natural gums are exuded in a variety of shapes and forms: usually the fragments are irregularly globular or tear globular or tear shaped. The best known being the tear or drop shape of various grades of gum Arabic. Other shapes are flakes or threat like ribbons with gum *tragacanth.* The surface is perfectly smooth when fresh but may become rough or crusty, covered with small cracks (Omer, 2004).

The identification of a particular gum from a series of different gum exudates needs an extensive number of analytical tests to perform. This approach enables “a chemical finger print” of each gum to be determined. The Several parameters can be used in this study but to most important are: Moisture ,Ash content, Nitrogen content, Specific optical rotation, content and Absence of tannins.(Karamalla, 1999).The most fundamental properties of a gum which makes it unique amongst polysaccharide, generally, are its solubility and viscosity. The majority of gums dissolve in water at different

concentrations; gum Arabic readily dissolves in cold and hot water in concentrations up to 50% (Hassan, 2000; Karamalla, 1999).

1.13.4 Moisture content

Moisture content of the gum determines its hardness and hence the variability of densities and the amount of air entrapped during nodule formation. It is determined by measuring the weight loss after evaporation of water. Reducing the moisture content of the natural gum can be readily used as a tenable method of reducing the microbial counts. (Karamalla, 1999). Anderson in (1968) reported the moisture content of the *azdirachta indica* gum (neem gum) to be 11.9% .

1.13.5Ash content

The ash content indicates the presence of inorganic elements existing in salt form. (Karamalla, Siddig and Osman, 1998).and Karamalla (1999) showed that the type of soil (clay or sand) affects the ash content significantly. the ash content of the neem gum 3.5 %reported by Anderson(1968).

1.13.6 pH value

The hydrogen ion concentration plays an important role in the chemistry and industrial use of gums. The change in the concentration of hydrogen ion may determine the solubility of gum and the precipitation of protein, therefore functional properties of a gum may be affected by change in pH for example viscosity and emulsifying power. Crude gum Arabic is slightly acidic because of the presence of few free carboxyl groups of its constituent acidic residues, D-glucuronic acid and its 4-O-methy1 derivatives.

1.13.7 Specific optical rotation

The optical activity of organic molecules (saccharides and carbohydrates) is related to their structure and is a characteristics property of the substance, and thus the specific rotation is considered as the most important criterion of purity and identity of any type of gum. Anderon *etal.*, 1971 the specific optical rotation reported -62^0. also(Mukherjee *etal.*, 1955) reported the optical rotation of neem gum was -71^0.

1.13.8 Nitrogen and protein content

The role of nitrogen and nitrogenous component in the structure, physicochemical properties and functionality of gum Arabic was subjected to intensive investigation (Dickinson *etal.*, 1988 (Randall, Phillips, and Williams, 1989). Dickinson, Galazka, and Anderson (1991), studied the emulsifying behavior of gum Arabic and concluded that there was a strong correlation between the proportion of protein in the gum and emulsifying stability. Protein is a part of gum molecule was established by (Anderson and Henrie, 1971;Anderson *etal.*,1972). Anderson and Henrie, (1971)reported the protein content in neem gum 35%.

The micro Kjeldahl method was used to determine the total nitrogen

1.13.9 Number average molecular weight

An important group of absolute methods allowing the determination of the molecular weight of macromolecules is based on the measurement of colligative properties. Here, the activity of the solvent is measured in a polymer solution via determination of the osmotic pressure (π). The value of π required to determine the number-average molecular weight can be obtained using a membrane Osmometere.

1.13.9.1Osmotic pressure

Osmosis is the phenomenon of penetration of a solvent into a solution through a semi permeable membrane. The tendency of solvent molecules to pass spontaneously into a solution, due to the inequality of chemical potential of pure solvent and solution estimated quantitatively by osmotic pressure, which has the dimension of pressure (atm). The osmotic pressure of a solution is equal to the additional pressure which must be applied to the solution to make the chemical potential of the component in solution equal to the chemical potential of the pure solvent (Billmeyer, 1971; Krigbaum and Flory, 1953).The Van't Hoff equation $\pi = C\ RT$ does not apply to polymer solutions, even though they are very dilute. The concentration dependence of osmotic pressure is expressed by a more complex equation which results if the concentration C is replaced by power series (Flory, 1953):

$$\frac{\pi}{C} = RT(\frac{1}{M_n} + A_2C + A_3C^2 + \cdots)$$

Π = osmotic pressure, A1 A2 = first and second virial coefficients, R = Gas constant

T is Temperature = Concentration

A_2 and $A_3C^2$is very small then:

$$Theintercep = \frac{\pi}{C_{\rightarrow 0}} = \frac{RT}{M_n}$$

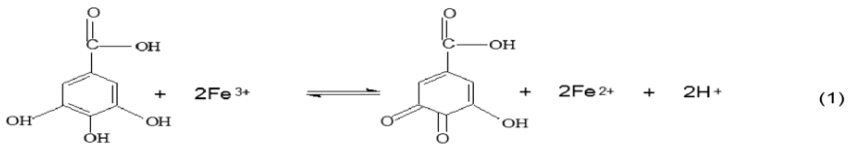
1.13.10 Equivalent weight and total uronic acid

Uronic acids are widely distributed in animal and plant tissues. They constitute a major component of many natural polysaccharides. Different methods have being developed for the determination of uronic acid in Arabic *gums*. These include colorimetric, decarboxylation and acid-base titrimetric methods (Ibrahim, 2006). The acid equivalent, or the titrable acidity, is determined as the mls of 0.02N sodium hydroxide that neutralizes 10 cm^3 of 3% w/v *Acacia gum* solution. Gums differ widely in their equivalent weight and uronic acid content. Anderson (1976) reported that the equivalent weight of *Azadirachta indica* gum as 620 and Mukherjee *etal.,* 1955 reported 1080.

1.13.11Tannin content

One of the most important tests that can be used to identify *Azadirachta indica* gum and distinguish it from some *Acacia* gums which are characterized by absence of tannins. Tannin content conducted by UV/Visible spectroscopy (Fig. 1.8).

A study had been done by Zahir 1998, cited by Karamalla, 1999) on raw gums from different *gums* species of Sudan-for their taxonomic classification, showed that these *Acacia* species could be divided into two main groups. Out of the thirteen gums tested, all but one fell into one group. The species falling in the large group showed presence of tannins in their gums. The tannin content ranged between 0.03 to 1.63%. The only gum that did not show presence of tannin was the gum from *A. senegal,* thus distinguishing itself distinctly and distantly from other *Acacia* gums. This finding was of significant importance when considering gums as food additives. It was established that tannins are anti-nutritional (Karamalla, 1999).



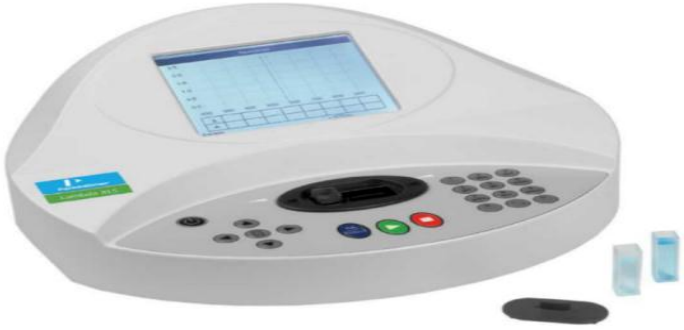


Fig 1.8 Perkin Elmer Lambda 40 UV/Vis spectroscopy

1.13.12 Viscosity

The viscosity of a liquid is its resistance to shearing, to stirring or to flow through a capillary tube. Viscosity was considered as one of the most important analytical and commercial parameters, since it is a factor involving the size and the shape of the macro – molecule (Anderson *etal*., 1969). Viscosity can be presented in many terms such as relative viscosity, specific viscosity, reduced viscosity, inherent viscosity and intrinsic viscosity.

The intrinsic viscosity has great practical value in molecular weight determinations of high polymers. This concept is based on the Mark-Houwink relationship suggesting that the intrinsic viscosity of a dilute polymer solution is proportional to the average molecular weight of the solute raised to a power in the range of 0.5 to 0.9. Values of the proportionality constant and the exponent are well known for many polymer-solvent combinations. Solutions viscosities are useful in understanding the behavior of some polymers.

Relative viscosity (η_{rel}) is the ratio of the dynamic viscosity of the solution to that of the pure solvent ($\eta_{rel} = \eta/\eta_0$ where η is the dynamic viscosity of the solution and η_0 is the dynamic viscosity of the solvent). As it is a ratio, it is dimensionless having no units. It is related to the intrinsic viscosity $[\eta]$ by the Huggins-Kramer equation: $in(\eta_{rel})/c = [\eta] + k_2[\eta]^2c$, where c is the concentration.

Reduced viscosity(η_{red}) is the specific viscosity divided by the concentration. It has units of reciprocal concentration, for example, mL g^{-1}. It is related to the intrinsic viscosity [η] by the Huggins-Kramer equation: $\eta_{red} = [\eta] + k_1[\eta]^2c$, where c is the concentration.

Specific viscosity (η_{sp}) is one less than the relative viscosity ($\eta_{sp} = \eta_{rel} - 1$; $\eta_{sp} = (\eta - \eta_0)/\eta_0$ where η is the dynamic viscosity of the solution and η_0 is the dynamic viscosity of the solvent). As with the relative viscosity, it has no units.

1.13.13 Calorific Value

The calorimeter C1 (Fig. 1.9) is used to determine the calorific value of solid and liquid materials according to national and international standards (DIN 51900, BS 1016 T5, ISO1928, ASTM 5468, ASTM 5865 and ASTM 480

Fig. 1.9 Calorimeter IKA C1System and the accessories.

1.13.14 The Rheology of *Azadirachta indica* gum

1.13.14.1Introduction

Rheology is the science of flow and deformation of matter and describes the interrelation between force, deformation and time. Rheology is most sensitive method for material characterization, because flow behavior is responsive to properties such as molecular weight and molecular weight distribution (Wang, 2007).

Rheology is an applied science, and its aim is twofold: Firstly, rheologists try to understand the relation between structure and flow properties. This important for the intelligent design and/or formulation of materials for certain applications. Secondly, by studying the material behavior using simple deformations, fundamental relations will be derived between deformation and force. Thus we need Rheology to measure fluid properties, understand structure-property relations, model behavior and to predict flow behavior of complex liquids under processing conditions(Gandhi, *etal.*,1988).

In principle, this definition includes everything that deals with flow, such as fluid dynamics, hydraulics, aeronautics and even solid state mechanics. However, in rheology we tend to focus on materials that have a deformation behavior in between that of liquids and solids, The term comes from The Greek philosopher Heraclitus described rheology as everything flows. Translated into rheological terms by *Marcus Reiner* this means everything will flow if you just wait long enough.

Fluid rheology is used to describe the consistency of different products, normally by the two components viscosity and elasticity. By viscosity is usually meant resistance to flow or thickness and by elasticity usually stickiness or structure. Rheology is applicable to all materials, from gases to solids. It was founded by two scientists meeting in the late '20s and finding out having the same need for describing fluid flow properties. The scientists were Professor *Marcus Reiner* and Professor *Eugene Bingham*. The science of rheology started in the 1920's when polymers started to be produced, leading to novel polymeric substances and new colloidal fluids (e.g. paints). Hence the Newtonian fluid and elastic solid outside the scope of rheology and material behavior intermediate to these classical extremes will be studied here. The term

"viscoelastic" is used to describe this behavior. Some fluids are however essentially inelastic, but have a viscosity which changes with the deformation state, they are called Non-Newtonian fluids(Braun *etal*., 2000).

The quantities measured in Rheology in principle this is very simple - there are only three basic ideas. First: stress, the amount of force applied to a given area of the sample. Second: strain, the degree to which the material deforms. And third: the ratio of stress to strain, which defines the elastic modulus for a solid, and the ratio of stress to rate of strain (or flow rate), which defines the viscosity for a liquid. The big complication is that most materials, and especially all biological materials, have both liquid and solid aspects. Consequently, material properties like elastic modulus and viscosities are not constants but functions of time, force, the direction in which the force is applied, and so on (Mofrad MRK, 2006).

Classification of materials Fluids are normally divided into three different groups according to their flow behavior: Newtonian fluids, Non-Newtonian fluids, time independent, and Non-Newtonian fluids, time dependent (Fig. 1.10).

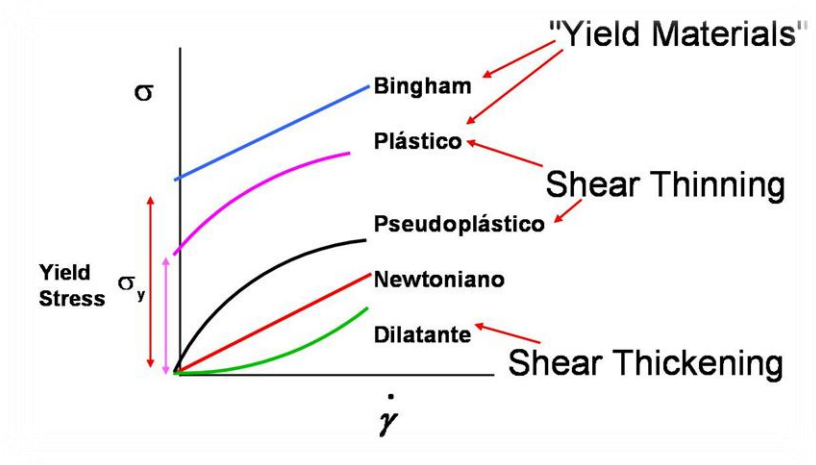


Fig. 1.10 Flow curves are normally use for the graphical description of flow behavior.

1.13.14.2 Stress and strain of the gum

Strain is what millions of people feel in their backs, or their relationships, and the cause of it is stress. Stress causes strain, but the amount of strain depends in some sense on how tough one is. Stress in rheology is, as one might expect, related to force. More precisely stress is the ratio of force to the area over which

that force is exerted; it has units of force/distance^2. Bioengineering literature, stress is often given in cogs units of dyne/cm^2; in most other studies, stress is given in SI units of N/m^2, which is the same as a Pascal (1 Pa = 1 N/m^2 = 10 dyne/cm^2). Neither unit is particularly well scaled to cell biology, but 1 Pa = 1 pN/μm^2. There are different kinds of stress depending on the direction in which the force is exerted. A shear stress is parallel to the surface. For example, endothelial cells feel a shear stress on their apical surface due to blood flow in an artery, much as a river's edge is subjected to shear stress that depends on how fast a river flows. Strain in rheology is somewhat different from its everyday meaning: it's a purely geometrical quantity, a way to quantify the amount of deformation in a given material, and has no units. Much of the complexity of rheology relates to defining or measuring strain. Measurable quantities such as the distance by which some point moves in response to stress requires sometimes complicated formulas to convert them to strains, depending on the shape of the material and the place where the force is applied. The elastic modulus is the quantity that allows the prediction of how much a material will deform elastically when a certain amount of stress is applied, and viscosity is the analogous quantity that tells how fast the material will flow. The more a tissue is stressed, cell or protein network, the more it is strained. But the rate and degree to which it strains is, usually, a complex function of the magnitude of the stress and how long it is applied (Discher, *etal.,* 2005).

1.13.14.3 Viscosity and elasticity of the gum

The ratio of stress to strain is all what is needed to know the properties of the material. The difference between elastic and viscous is, basically, whether the strain reaches a limit or continuously increases in response to a constant stress. An ideal elastic material such as a spring deforms to a given strain in response to a given stress and then sits there forever unless the stress is removed, at which time the material returns to its initial shape. The elastic modulus, the ratio of stress to strain, is a constant in this case. All the work done by the initial stress (work = force × distance) was stored in the material (hence the term storage modulus, see below) and elastically recovered when the stress is removed. Elasticity in this context does not refer to whether a material is `stretchable' or not, but whether it returns to its initial shape when you stop pulling or pushing on it. There are different kinds of elastic modulus depending on the kind of stress. If it's a shear stress, then the ratio of stress to strain is the shear modulus.

If it's elongational or compressional stress, then the ratio of stress to strain is generally called the *Young's* modulus, named for *Thomas Young* who also identified the cause of astigmatism. If a simple material conserves volume, then shear and Young's modulus are related by a factor of three. An ideal viscous material changes strain in proportion to the time that the stress is applied. In this case, the ratio of stress to the rate of strain defines the viscosity (Mothe` , C. G., & Rao, M. A.,1999).

1.13.14.4 The Visco elasticity

As noted above, most materials are both viscous and elastic. The shear modulus and viscosities that define them depend on strain rates as well as strain magnitudes. Differentiating elastic from viscous effects requires measurements at different time scales and is usually done by performing oscillatory deformations at different frequencies. As a result, data are often reported in terms of storage (elastic) or loss (viscous) modulus. The time dependence is important.

1.13.14.5 Kinematic and dynamic viscosity

Kinematic viscosity is measured with kinematic instruments, normally different types of cups which means that the knowledge and control of shear rates is limited or non-existent. Therefore kinematic viscosity values are of little or no use for design of equipment for non-Newtonian fluids. Dynamic viscosity takes into account the effect of shear rate and time and is therefore the only type of viscosity relevant for non-Newtonian design purposes. Dynamic viscosity is measured with dynamic instruments, either rotating (shearing) or oscillating (Heilbrunn L, *etal.*, 1956). An instrument only capable of measuring shearing viscosities is called a viscometer and the oscillating type is called a rheometer.

Basic constitutive equations Various models for approximation of rheological data have been presented. One of the most widely spread models is the so-called power law for approximation of viscosity data. The main reason for the power law being so popular is that the shearing rheological behavior of a fluid is represented simply by a straight line in a log-log shear rate/shear stress graph. Another reason is that the shearing behavior of most fluids lends itself to a good approximation applying the power law (Table 1.3).

Table 1.3Mathematical models for flow behavior

Newtonian	$\tau = \mathrm{K}\gamma$
Shear thinning	$\tau = \mathrm{K}\gamma^{n}$ (n<1)
Shear thickening	$\tau = \mathrm{K}\gamma^{n}$(n>1)
Bingham	$\tau = \tau_0 + \eta_p\gamma$
Herschel-Bulkley	$\tau = \tau_0 + \mathrm{K}\gamma^{n}$
Casson	$\tau^{1/2} = {\tau_0}^{1/2} + {\eta_\infty}^{1/2}\gamma^{1/2}$
Sisko	$\eta = \eta_\infty + \mathrm{K}\gamma^{n-1}$
Ellis	$\gamma = \mathrm{K}_1\tau + \mathrm{K}_2\tau^{\mathrm{n}}$
Carreau	$\frac{\eta - \eta_\infty}{\eta_0 - \eta_\infty} = [1 + (\lambda\gamma)^2]^{(n-1)/2}$

***τ shear stressγ shear rate$\tau_0$yield stress η_∞limiting viscosity.**

1.13.14.6 Viscous and elastic modulus

Rheological measurements are normally performed in kinematic instruments in order to get quantitative results useful for design and development of products and process equipment. For design of products, e.g. in the food, cosmetic or paint industry, rheometric measurements are often performed to establish the elastic properties, such as gel strength and yield value, both important parameters affecting e.g. particle carrying ability and spread ability. For design of process equipment the properties during shearing of the product is of prime interest. Those properties are established in a normal viscosity measurement.

A rheometric measurement normally consists of a strain (deformation) or a stress analysis at a constant frequency (normally 1 Hz) combined with a frequency analysis, e.g. between 0.1 and 100 Hz. The strain sweep gives information of the elastic modulus G', the viscous modulus G" and the phase angle d. A large value of G' in comparison of G" indicates pronounced (Fig. 1.11).

elastic (gel) properties of the product being analyzed. For such a product the phase angle is also small, e.g. 20º (a phase angle of 0º means a perfectly elastic material and a phase angle of 90º means a perfectly viscous material). The

frequency sweep gives information about the gel strength where a large slope of the G' curve indicates low strength and a small slope indicates high strength.

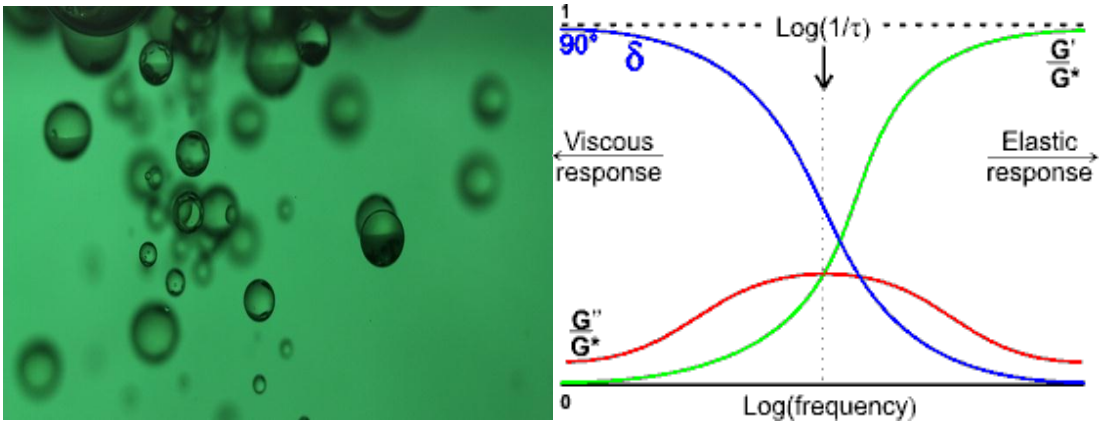


Fig. 1.11The elastic modulus G', and the viscous modulus G''

A viscometric measurement normally consists of a shear rate analysis. The shear rate sweep should preferably cover the range applied in the intended equipment. For liquid foods a shear rate range from around 1 to 1,000 s-1 covers the needs for a low-viscous product.

1.13.14.7Structural effects of the gum molecule

Linear and substantially linear polymers behave in a qualitatively predictable way with respect to the relationship of their viscosity to their structure and conformation. In dilute solutions this relationship depends effectively on the volume "swept out" (that is, the hydrodynamic volume) by the molecules as they tumble in the solution. At these low concentrations, where there is effectively no interaction between molecules and they are at their most extended, the viscosity may be little different from that of water; this small difference depending on the total spherical volume (itself dependent on concentration and radius of gyration of the solute) taken up by the freely rotating molecules. The relationship between viscosities with concentration is generally linear up to viscosity values of about twice that of water (Barnes, *etal.*, 2001). This dependency means that more extended molecules increase the viscosity to greater extents at low concentrations than more compact molecules of similar molecular weight. Generally less-flexible links between sequential monomers in the

polymeric chains give rise to more extended structures but the linkage spacing, direction and charge density are all important factors. Where residues are negatively charged, the repulsion between similar charges increases molecular extension but this can be reduced at higher ionic strength or below the pK_a's of the anionic groups and this reduction is particularly noticeable for polymers with high molecular mass. The lack of much change in viscosity of such molecules with ionic strength is indicative of an inflexible rod-type conformation. It should be noted that although attaching short sugar units as branch-points to linear polysaccharides does increase their rigidity into an extended structure, this is at the cost of greatly increased molecular mass. The extended nature of the molecules has an extreme effect on the molecular mass dependency of the viscosity as the hydrodynamic volumes (and hence viscosities) of compact (highly flexible but poorly hydrated) molecules increase approximately as the cube root of their molecular mass whereas those of more-extended well hydrated molecules (such as alginate and xanthan gum) increase approximately linearly with molecular mass. The relationship between the intrinsic viscosity $[\eta]$ and the relative molecular mass (M_w) is given by $[\eta] = K M_w^a$, the Mark-Houwink equation where K and a are constants. Amylose, carboxymethylcellulose, arabinoxylans and guar all have exponents (a) of about 0.7. Knowledge of these constants allows the viscosity-average molecular mass to be calculated from viscosity data.

The viscosity increases with concentration until the shape of the volume occupied by these molecules becomes elongated under stress causing some overlap between molecules and a consequent reduction in the overall molecular volume with the resultant effect of reducing the amount that viscosity increases with concentration (under stress).

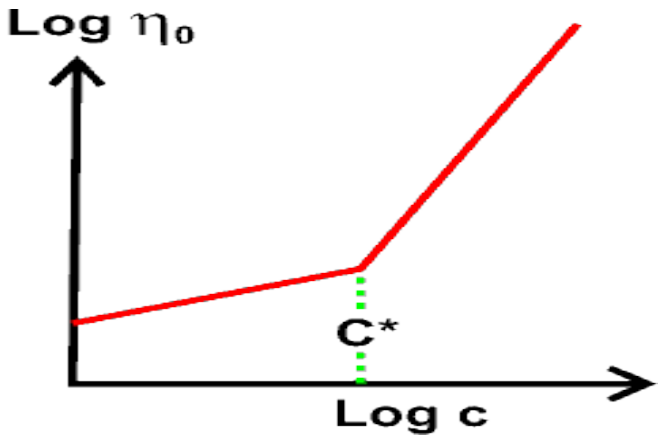


Fig. 1.12A plot of log η_0 vs. log C

At higher concentrations (above a critical concentration C*) all the polymer molecules in the solution effectively overlap (Fig. 1.12), interpenetrate and become entangled (that is, their total hydrodynamic volume appears greater than the solution volume) even without being stressed, so changing the solution behavior from mainly viscous to mainly elastic with the viscosity (η_0 at zero stress) being mainly governed by the mobility of the polymer molecules. C* will depend on the shear strain rate as, at high shear strain rate, the molecules take up a less voluminous shape. At higher concentrations the viscosity increases up to about the fifth power of the concentration and this can cause apparently synergic behavior of hydrocolloid mixtures, particularly if they cause phase separation with its inherent concentration increases.

At high shear strain rate (and sufficient concentration) molecules may become more ordered and elastic. Shear flow (and its related stress) causes molecules to become stretched and compressed (at right angle to stretch) resulting in isotropic solutions becoming anisotropic. After release from such conditions, the molecules relax back with time (the relaxation time). At low concentrations below the critical value (C*), the shear modulus of hydrocolloid solutions is mainly determined by the loss modulus at low frequencies (that is, G" is relatively high for viscous materials). As G" depends on the frequency but G' depends on the square of the frequency, G' becomes more important at higher frequencies. At higher concentrations in viscous solutions G' is generally greater than G" throughout a wide frequency range. This difference is very large for strong gels when the frequency has almost negligible effect (that is, G' is high for elastic materials). Such gels often form above another critical concentration

specific to the hydrocolloid, where junction zones occur so stabilizing intermolecular associations (Li, X., Fang, (2009).

Dilatancy (**shear thickening**) shows an increase in viscosity with shear stress and strain due to structural enhancement. An example is uncooked corn starch paste where shear stress squeezes the water from between the starch granules allowing them to grind against each other. This property is often used in sauces where, for example, tomato sauce flow is prevented under small shear stress but then catastrophically fails, producing too great a flow, under greater stress (shaking). Another (and the strictly correct usage for the term) meaning for Dilatancy concerns the increase in volume of suspensions of irregular particles with shear due to the creation of small but empty cavities between the particles as they scrape past each other.

Eutectic point is the lowest possible melting point (equilibrium freezing point) that a mixture of solutes may have. No other composition of the same materials will have a lower melting point. Thin films of fluid may remain below the eutectic point in microcrystalline ice due to surface effects.

Fluidity is the reciprocal of the viscosity (= $1/\eta$).

Stoke (St) is a unit of kinematic viscosity ($cm^2\ s^{-1}$). The SI unit of kinematic viscosity is $m^2\ s^{-1}$ (= 10000 stoke).

Thixo tropic liquids exhibit a time-dependent response to shear strain rate over a longer period than that associated with changes in the shear strain rate. They may liquefy on being shaken and then solidify (or not) when this has stopped.

At moderate concentrations above a critical value hydrocolloid solutions exhibit non-Newtonian behavior where their viscosity depends on the shear strain rate, typically as opposite, where γ is the shear strain rate, η_0 and η_∞ are the viscosities at zero and infinite shear strain rate respectively and τ is a shear-dependent time constant that represents the reciprocal of the shear strain rate required to halve the viscosity.

Tan(δ) = G''/G' where tan(δ) quantifies the balance between energy loss and storage. As tan(45°) =1, a value for tan(δ) greater than unity indicates more "liquid" properties, whereas one lower than unity means more "solid" properties,

regardless of the viscosity. The shear modulus (resulting from changing strain) is the ratio of the shear stress to the shear strain.

1.13.14.8The goal of rheology

•To understand the kinds of flow and deformation effects exhibited by complex systems.

•To apply qualitative rheological knowledge to diagnostic, design, or optimization problems.

•To in diagnostic, design, or optimization problems, use or devise quantitative analytical too is that correctly capture rheological effects.

Table 1.4Process shear, shear rate, and applications from Barnes (1989)

Process shear	**rate [s^{-1}]**	**Applications**
Sedimentation	10^{-6} -10^{4}	Medicines, Paints,…
Leveling	10^{-3}-10^{-1}	Paints, inks
Draining under gravity	10^{-1}	Emptying tanks
Extrusion	10^{1}-10^{2}	Polymer melts, Dough
Chewing, Swallowing	10^{1} -1^{02}	Food
Dip Coating	10^{1} -10^{2}	Paints, confectionery
Mixing and stirring	10^{1}-10^{3}	Manufacturing liquids
Pipe flow	10^{2}-10^{3}	Pumping, blood flow
Spraying, brushing	10^{3} -10^{4}	atomization, painting
Rubbing	10^{4} -10^{5}	Skin cream and Lotion
Injection molding	10^{2} -10^{5}	Polymer melts
MILLING	10^{3}-10^{5}	inks, coatings
coating flows	10^{5} -10^{6}	coating flows
Lubrication	**10^{3}-10^{7}**	**Engines**

1.13.15 Molecular weight distribution of *Azadirachta indica* gum

The molecular weight and molecular weight distribution of *azdirachta indica* gum was studded using Gel Permeable Chromatography (GPC) with different

detectors such as Light scattering (LS), Refractive index (RI) and Ultra violet (UV).

Gel Permeable chromatography (GPC) is a type of size exclusion chromatography (SEC) that separates analysts on the basis of size. The technique is often used for the analysis of polymers. As a technique, SEC was first developed in 1955 by Lathe and Ruthven Lathe. The term gel permeation chromatography can be traced back to J.C. Moore of the Dow Chemical Company who investigated the technique in 1964 and the proprietary column technology was licensed to Waters Corporation, who subsequently commercialized this technology in 1964.GPC systems and consumables are now also available from a number of manufacturers. It is often necessary to separate polymers, both to analyze them as well as to purify the desired product (Moore, J.C., 1964).

When characterizing polymers, it is important to consider the Polydispersity index (PDI) as well the molecular weight. Polymers can be characterized by a variety of definitions for molecular weight including the number average molecular weight (M_n), the weight average molecular weight, the size average molecular weight (M_z), or the viscosity average molecular weight (M_v). GPC allows for the determination of PDI as well as M_v and based on other data, the M_n, M_w, and M_z can be determined.

Radius of gyration (R_g) is defined as the distance (r_i) between all pairs of entities (n). Where (n) is the number of entities in the chain and (r_i) is the radius of each from the center of mass(Fig. 1.13).

GPC separates based on the size or hydrodynamic volume (radius of gyration) of the analytes. This differs from other separation techniques which depend upon chemical or physical interactions to separate analysts (Skoog, D.A., 2006). Separation occurs via the use of porous beads packed in a column.

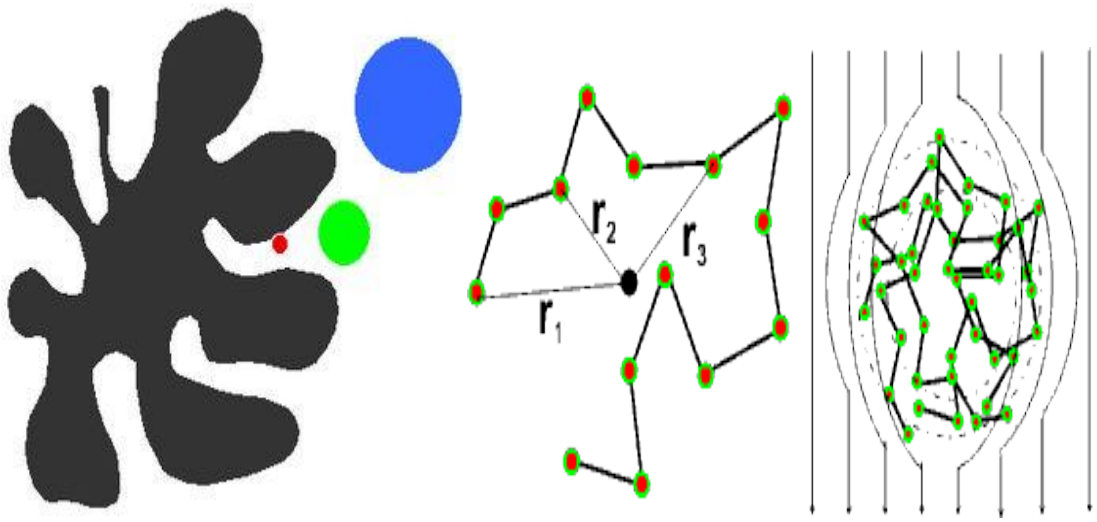


Fig.1.13 Schematic of pore vs. analytes size and the radius of gyration

The smaller analysts can enter the pores more easily and therefore spend more time in these pores, increasing their retention time. These smaller molecules spend more time in the column and therefore will elute last. Conversely, larger analysts spend little if any time in the pores and are eluted quickly. All columns have a range of molecular weights that can be separated.

If an analyte is either too large or too small, it will be either not retained or completely retained, respectively. Analytes that are not retained are eluted with the free volume outside of the particles (Vo), while analytes that are completely retained are eluted with volume of solvent held in the pores (Vi). The total volume can be considered by the following equation, where Vg is the volume of the polymer gel and Vt is the total volume.

$$V_t = V_g + V_i + V_o$$

As can be inferred, there is a limited range of molecular weights that can be separated by each column and therefore the size of the pores for the packing should be chosen according to the range of molecular weight of analytes to be separated. For polymer separations the pore sizes should be on the order of the polymers being analyzed. If a sample has a broad molecular weight range it may be necessary to use several GPC columns in tandem with one another to fully resolve the sample.

Typically for a linear polysaccharide (M_w 10^6) the Radius of gyration would be approximately 6 nm if spherically packed, 940 nm if as an extended stiff rod, and 17 nm if as a random coil. Its relationship to the effective radius the tumbling molecule represents to a flowing liquid (hydrodynamic radius, R_h) depends upon the flexibility and density of the structure; R_g/R_h generally being about 1.6 but lower for branched and globular structures and gels.

Light scattering If a beam passes through of suspended particles the light is scattered; this phenomenon is called Rayleigh scattering. From the theory of scattering we can calculate the molar mass of the particle and obtain some idea about its shape. First, we consider the scattering by a particle that is small compared to the wavelength of the light. The monochromatic light beam passed into cell and the intensity of scattered light measured as a function of the scattered angle and as a function of the wave length. The filter selects the wavelength to be used; the detector can be moved through an angle centered on the 90 degree position so that any dissymmetry can be measured. The scattering for large particles is not symmetric about the 0 degree angle (G.H.; Ruthven, C.R.J., 1956).

Whenever an electrical charge is accelerated, it radiate energy. If the oscillating electric field of a light beam act on a charge, the charge oscillates, is accelerated, and radiates a light beam of the same frequency. This oscillating charge is equivalent to an oscillating dipole moment, a charge q displaced through a distance x.

1.13.15.1 Rayleigh scattering

Scattering is a good approximation to the manner in which light scattering occurs within various media for which scattering particles have a small size parameter. In a typical Rayleigh scattering experiment, a well collimated, single frequency polarized light beam (e.g. from a laser) is used to illuminate a solution containing a suspension of the macromolecules of interest. The electric field of the polarized light beam is generally produced perpendicular to the plane in which the intensity and angular dependence of the subsequently scattered light is to be measured. The intensity carries information about the molar mass, while the angular dependence carries information about the size of the macromolecule. Light scattering is a non-invasive technique for characterizing macromolecules and a wide range of particles in solution. In contrast to most

methods for characterization, it does not require outside calibration standards. In this sense it is an absolute technique. There are two different types of light scattering measurements for absolute molecular characterization.

Absolute molecular weights can be determined also via mass spectrometry, membrane osmometry, and sedimentation equilibrium (analytical centrifugation), only light scattering covers so broad a range of macromolecules including their oligomeric states. Most importantly, light scattering permits measurement of the solution properties of macromolecules. While a sedimentation equilibrium run may require 72 hours, a size exclusion chromatography/light scattering study may be completed in well under an hour and a batch mode analysis in a few minutes (Billmeyer,. (1971).

1.13.15.2Scattered Light and Molar Mass

The intensity of the radiated light depends on the magnitude of the dipole induced in the macromolecule. The more polarizable the macromolecule, the larger the induced dipole, and hence, the greater the intensity of the scattered light. Therefore, in order to characterize the scattering from a solution of such macromolecules, it is first necessary to know their polarizability. This may be determined from a measurement of the change, *dn*, of the solution's refractive index *n* with the molecular concentration change, *dc*, by measuring the *dn/dc*. When there are many macromolecules in solution, each macromolecule scatters light via the aforementioned induced dipole mechanism.

During the time of passage of the incident light wave front over the macromolecule, each element scatters in phase with the scattering of adjacent elements. Thus the scattered waves will add destructively or constructively producing constructive or destructive interference in certain directions. If the angular dependence of the scattered light is measured, it is possible to determine the size of the molecule. Here, the size measurement is known as the root mean square (rms) radius, or sometimes the "radius of gyration". The latter term is a misnomer since it describes a kinematic measure of a molecule rotating about a particular axis in space. The rms radius, on the other hand is a measure of its size weighted by the mass distribution about its center of mass. Once the molecule's conformation is determined, (e.g., random coil, sphere, or rod), the rms radius can be related to its geometrical dimensions. For a sample containing a broad distribution of molecular masses, following separation by chromatographic

means, the measured rms radius may be plotted against the correspondingly measured molar mass to determine the sample's conformation (Billmeyer,. (1971).

1.13.15.3 GPC instrumentation

GPC is often used to determine the relative molecular weight of polymer samples as well as the distribution of molecular weights. What GPC truly measures is the molecular volume and shape function as defined by the intrinsic viscosity. If comparable standards are used, this relative data can be used to determine molecular weights within ± 5% accuracy.

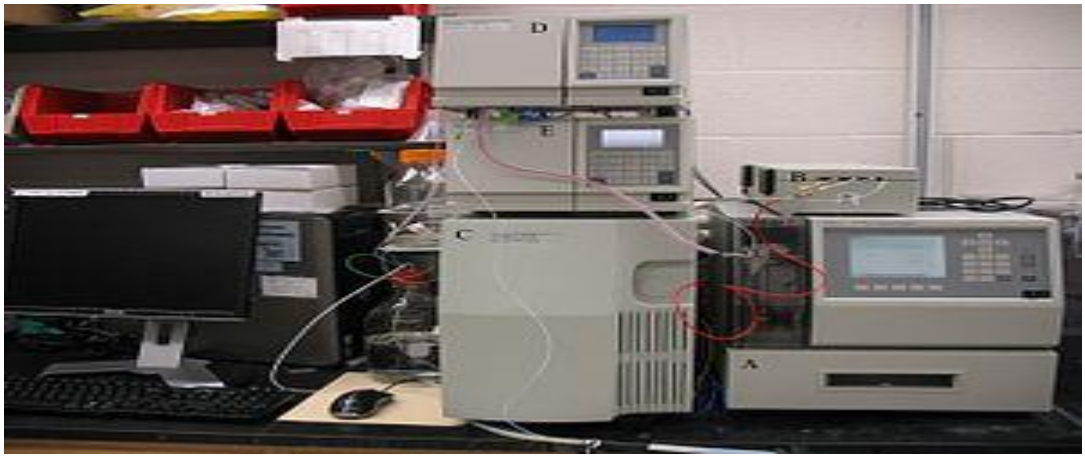
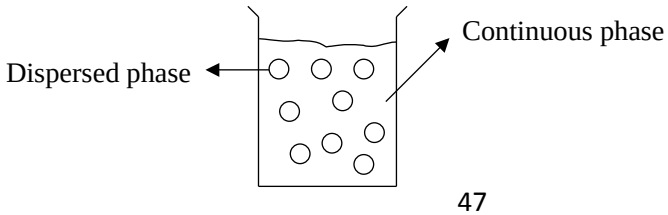


Fig. 1.14 Atypical GPC instrument including: Auto sampler, Column, Pump, RI detector, and UV- vs. detector

1.14. Emulsification Properties of *Azadirachta indica* Gum

1.14.1 Definition of emulsion

An emulsion is liquid preparation containing two immiscible liquids, one of which is dispersed as globules name as dispersed phase or internal phase in the other liquid name as continuous phase or external phase (Dickenson, 2003).



To stabilize these droplets, emulsifying agent should be added

1.14.2 British Pharmacopoeia (BP) definition of oral emulsions

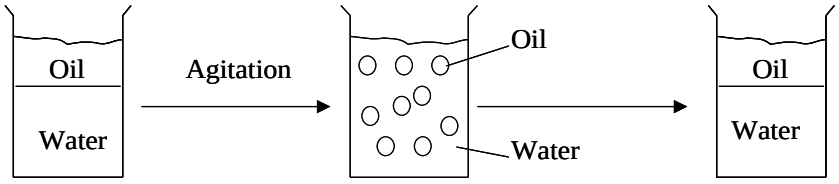
Oral emulsions are oral liquids containing one or more active ingredients. They are stabilized oil-in-water dispersions, either or both phases of which may contain dissolved solids. Solids may also be suspended in oral emulsions. When issued for use, oral emulsions should be supplied in wide-mouthed bottles. Micro emulsion has droplets size range 0.01 to 0.1 microns, but Macro emulsion has droplets size range approximately 5 microns (Dickenson, 2003).

1.14.3 The primary and secondary emulsion

Primary emulsion containing one internal phase, for example, oil-in-water emulsion (o/w) and water-in-oil emulsion (w/o).Secondary emulsion or multi pleemulsion it contains two internal phase, for instance, o/w/o or w/o/w. It can be used to delay release or to increase the stability of the active compounds.

1.14.4Theories of Emulsification

In case of two immiscible liquids as figure below the oil was separated from the water.



Separate rapidly into two clear defined layers

An explanation of this phenomenon is because of cohesive force between the molecules of each separate liquid exceeds adhesive force between two liquids. This is manifested as interfacial energy or tension at boundary between the liquids.

The producing of more small droplet in emulsion lead to increasing the surface area, increasing interfacial tension and the system thermodynamically

unstable (high energy); and the system tend to separate in two layer to reduce the surface area. Therefore, to prevent the coalescence and separation, emulsifying agents have been used (McClements, 2007).

1.14.5 The emulsifying agents

The emulsifying agents defined as surface active agent adsorbed at oil/water interface to form monomolecular film to reduce the interfacial tension. Also emulsifying agents defined as hydrophilic colloids forming a Multi molecular film around the dispersed droplet. Other definition of emulsifying agents is finely divided solid particles adsorbed at the interface between two immiscible liquid phases to form particulate film (Dickenson, 1992).

Table 1.5 The emulsifying agents

Emulsifying Agents	Examples
Carbohydrate Materials	Acacia, Tragacanth, Agar, Pectin.
Protein Substances	Gelatin, Egg yolk, Caesin
High Molecular Weight Alcohols	Stearyl Alcohol, Cetyl Alcohol, Glyceryl Mono stearate emulsion, cholesterol
Wetting Agents	Anionic, Cationic, Nonionic
Finely divided solids	Bentonite, Magnesium Hydroxide, Aluminum Hydroxide

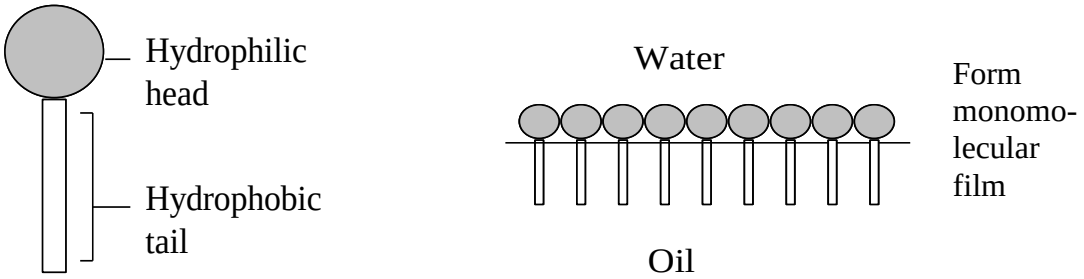
1.14.6 Monomolecular adsorption

In emulsion, the surface area is high to maintain the dispersion of the droplets. Thus, based on the equation, surface free energy becomes high consequently. The only way to keep it low is to reduce the interfacial tension (Dickinson, 1992).

$$W = \gamma_{o/\omega} \cdot \triangle A$$

Surface free energy — Interfacial tension — Surface area

Surface active agent (SAA) is molecule which have two parts, one is hydrophilic and the other is hydrophobic.



The functions of emulsifying agents is to provide stability to dispersed droplets are as following: Reduction of the interfacial tension by Forming coherent monolayer to prevent the coalescence of two droplet when they approach each other, or provide surface charge which cause repulsion between adjust particles(McClements, 2005).

Table 1.6 The Multimolecular adsorption examples

	Polysaccharides	Amphoterics	Synthetic or semi-synthetic polymers
colloids	Acacia Agar Alginic acid Carrageenan Guar gum Karraya gum Tragacanth	Gelatin	Carbomer resins Cellulose ethers Carboxymethyl chitin PEG-n (ethylene oxide polymer)

Other main function as emulsion stabilizers is by making coherent multi-molecular film. This film is strong and resists the coalescence. They have, also, an auxiliary effect by increasing the viscosity of dispersion medium. Most of the hydrophilic colloids form oil-in-water emulsions. Some of them can provide electrostatic repulsion like *acacia*, which contains Arabic acid and proteins (COOH and NH_3).

1.14.7 Solid particle adsorption

Finely divided solid particles are adsorbed at the surface of emulsion droplet to stabilize them. Those particles are wetted by both oil and water (but not dissolved) and the concentration of these particles form a particulate film that prevent the coalescence. Particles that are wetted preferentially by water from o/w emulsion, whereas those wetted more by oil form w/o emulsion. Note that they are very rare to use and can affect rheology of the final product size of the particle is very important, larger particles can lead to coalescence.

1.14.8 The factors affecting the choice of emulsion type

The choice of emulsion depends on

(1)properties and uses of final products

(2) the other material required to be present.

- Oil-soluble drug is prepared in o/w emulsion due its solubility and its taste can be masked by adding flavoring agents
- For intravenous injection, o/w emulsion is the only type could be used.
- For intramuscular injection, both o/w and w/o types of emulsion could be used. Water-soluble drug can be prepared in w/o emulsion to get prolonged action (depot therapy)

Table 1.7 The differentiation between oil in water and water in oil emulsion

Oil in water emulsion	**Water in oil emulsion**
• For insoluble drug • For local effect • Easily to wash from skin • Doesn't have greasy texture of oily preparation • Acceptable by consumer	• For water soluble drug • Can be used to hydrate the upper layer of stratum corneum (moisturizing cream) • Can increase the absorption of drug from these formulation • Can be used to clean skin from dirt • Not acceptable by consumer

1.14..9 Emulsion Preparation Methods

1.14.9.1Continental or Dry Gum Method

4 parts (volumes) of oil and 2 parts of water and one part of *Azadirachta indica* gum. or other o/w emulsifier is triturated with oil in a perfectly dry Wedgwood or porcelain mortar until thoroughly mixed. Glass mortar has too smooth a surface to produce the proper size reduction of the internal phase (Do not use glass mortar). After the oil and gum have been mixed, the two parts of water are then added all at once and the mixture is triturated immediately (Hunter, 1986).

1.14.9.2 English or wet Gum Method

Same proportion of oil, water and gum are used as in the continental or dry gum method but the order of mixing is different. Mucilage of the gum is prepared by triturating *neem gum*(or other emulsifier) with water. The oil is then added slowly in portions, and the mixture is triturated to emulsify the oil. Should the mixture become too thick during the process, additional water may be blended into the mixture before another successive portion of oil is added.

1.14.9.3Bottle or Forbes Bottle Method

Useful for extemporaneous preparation of emulsion from volatile oils or oleaginous substance of low viscosity. Put powdered *neem gum* in a dry bottle add 2 parts of oil, then thoroughly shake the mixture in the capped bottle. A volume of water approximately equal to the oil is then added in portions, the mixture being thoroughly shaken after each addition. This method is not suitable for viscous oils (i.e. high viscosity oil) (McClements, 2005).

1.14 .9. 4 Control emulsion type during formulation

Volume of internal and external phases controls the type of emulsion. The smaller volume will be for the internal phase and the larger volume will be for external phase. In some cases, internal phases can be more than 50% of the total volume. The other control is Dominance of polar and non-polar characteristic of

emulsifying agents (relative solubility of emulsifying agent in water and oil). Dominance of polar part results in formation of o/w emulsion and dominance of non-polar part results in formation of w/o emulsion. Note that polar groups are better barriers than non-polar; therefore, o/w emulsion can be prepared with more than 50 % of oil phase "internal phase".

1.14.10 Instability mechanisms of Emulsion

An emulsion is considered to be physically unstable if the internal phase tends to form aggregates of globules, or large globules or aggregates of globules rise to the top or fall to the bottom of the emulsion to form a concentrated layer of the internal phase. If all or part of the liquid of the internal phase becomes un emulsified on the top or bottom of the emulsion. Separation of the internal phase from the external phase is called breaking of the emulsion(Whitehurst, 2004).

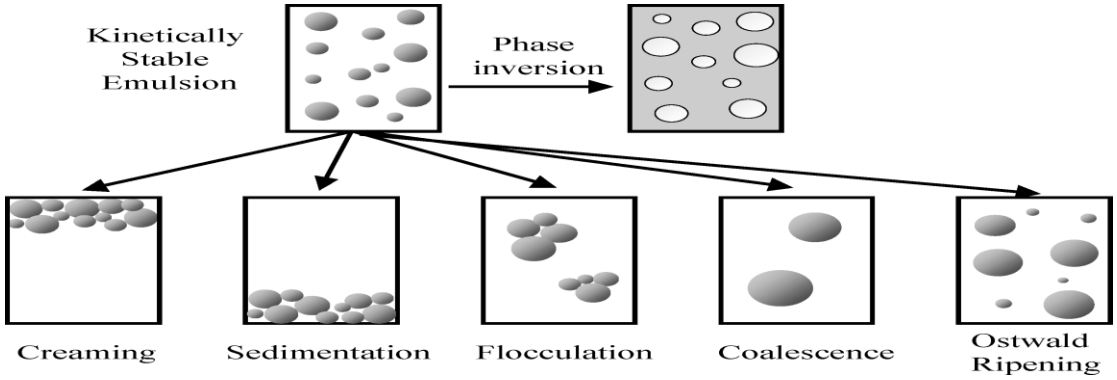


Fig 1.15 Schematic diagram of most common instability mechanisms

1.14.11 Phase Inversion

The relative volume of internal and external phases of an emulsion is important. Increase internal concentration increase viscosity up to a certain point. Viscosity will decrease after that point. At this point the emulsion has undergone inversion i.e. it has changed from an o/w to a w/o, or vice versa. In practice, emulsions may be prepared without inversion with as much as about 75% of the volume of the product being internal phase.

1.14.12 Applications of emulsions

The emulsions can be applied in many aspects such as food industry, mask the taste in many applications, convenient means of orally administration of water-insoluble liquids, facilitates the absorption of water-insoluble compounds, drugs, cosmetic and therapeutic uses(Dalgleish, 2001).An emulsifier is a surface active substance that adsorbs to the surface of emulsion droplets to form a protective coating that prevents the droplets from aggregating with one another, such as certain proteins, polysaccharides, phospholipids and small molecule surfactants (Whitehurst, 2004). An emulsifier also reduces the interfacial tension and therefore facilitates the disruption of emulsion droplets during homogenization, which aids in the formation of emulsions containing smaller droplets (Dalgleish, 2001).

1.15 Objectives:

1. To Characterize *Azadirachta Indica*(Neem) gums by studying the physicochemical properties.
2. To study the rheological behavior of *Azadirachta indica* gum.
3.To study the emulsification properties of *Azadirachta indica* gum.

Chapter two
Materials and Methods

2.1Materials

Forty five samples of authentic *Azadirachta indica* (Neem trees*)* gum collected from three different states from Sudan were collected during in season (2014-2016) from three different areas .from Alkadrow-Sooba-EL mogran regions Khartoum state, other sample from Kosti –Al jabaleen –Tandalty region White Nile state and other sample From Um rowaba –Elobied- Bara regions Northern Kordofan state

Details of Neem gum(*Azadirachta indica*) gums are given in tables (2.1, 2.2 and 2.3) for Seasons 2014, 2015 and 2016 respectively.

Table 2:1 content sample code, locations, date of collection, type soil and rain fall season 2014 (*azadirachta indica gum*)

Code	Location		Date of collec-tion	Type of soil	Rain fall
	State	area			
1A1	Khartoum	Al kadrow	Feb-14	Clay	126-150mm
1A2	Khartoum	Elmogran	Feb-14	Clay	126-150mm
1A3	Khartoum	Soba	Feb-14	Clay/Sand	126-150mm
Comp 1A 20014	is prepared by mixing equal amounts of (1A1+1A2+1A3)				
1B1	White Nile	Kosti	Dec-14	Clay	75-150
1B2	White Nile	Elgabaleen	Dec-14	Clay	75-150
1B3	White Nile	Tandalty	Dec-14	gardood	75-150
Comp 1B 2014	is prepared by mixing equal amounts of (1B1+1B2+1B3)				
1C1	Northern Kor-dofan	Um Rawaba	Apr-14	sand	< 400mm
1C2	Northern Kor-dofan	El obied	Apr-14	sand	< 400mm
1C3	Northern Kor-dofan	Bara	Apr-14	sand	< 400mm
Comp 2C 2014	is prepared by mixing equal amounts of ($1C_1$+$1C_2$+$1C_3$)				

Table 2:2 content sample code, locations, date of collection, type soil and rain fall season 2015 (*Azadirachta indica gum*)

Code	Location		Date of collection	Type of soil	Rain fall
	State	area			
2A1	Khartoum	Al kadrow	Mar-15	Clay	126-150mm
2A2	Khartoum	Elmogran	Mar-15	Clay	126-150mm
2A3	Khartoum	Sooba	Mar-15	Clay/Sand	126-150mm
Comp 2A 2015	is prepared by mixing equal amounts of ($2A_1$+$2A_2$+$2A_3$)				
2B1	White Nile	Kosti	Feb-15	Clay	75-150
2B2	White Nile	Elgabaleen	Feb-15	Clay	75-150
2B3	White Nile	Tandalty	Feb5	gardood	75-150
Comp 2B 2015	is prepared by mixing equal amounts of ($2B_1$+$2B_2$+$2B_3$)				
2C1	Northern Kordofan	Um Rawaba	Apr-14	Sand	< 400mm
2C2	Northern Kordofan	El obied	Apr-14	Sand	< 400mm
2C3	Northern Kordofan	Bara	Apr-14	Sand	< 400mm
Comp 2C 2015	is prepared by mixing equal amounts of ($2C_1$+$2C_2$+$2C_3$)				

Table 2:3 content sample code, locations, date of collection, type of soil and rain fall season 2016(*azadirachta indica gum*)

Code	Location		Date of collection	Type of soil	Rain fall
	State	Specific area			
3A1	Khartoum	Al kadrow	Feb-16	Clay	126-150mm
3A2	Khartoum	Elmogran	Feb-16	Clay	126-150mm
3A3	Khartoum	Sooba	Feb-16	Clay/Sand	126-150mm
Comp 3A 20016	is prepared by mixing equal amounts of (3A1+3A2+3A3)				
3B1	White Nile	Kosti	Dec-16	Clay	75-150
3B2	White Nile	Eljabaleen	Dec-16	Clay	75-150
3B3	White Nile	Tandalty	Dec-16	gardood	75-150
Comp 3B 2016	is prepared by mixing equal amounts of ($3B_1$+$3B_2$+$3B_3$)				
3C1	Northern Kordofan	Um Rawaba	Apr-16	sand	< 400mm
3C2	Northern Kordofan	El obied	Apr-16	Sand	< 400mm
3C3	Northern Kordofan	Bara	Apr-16	Sand	< 400mm
Comp 3C 2016	is prepared by mixing equal amounts of ($3C_1$+$3C_2$+$3C_3$)				

- Fifteen samples from season 2014 labeled as (comp1A2014, comp1B2014, comp1C2014).
- From season 2015 also Fifteen samples were used, labeled as (comp2A2015, comp2B2015 and comp2C2015).
- Fifteen samples were used from season 2016 labeled as (comp3A2016, comp3B2016 and comp3C2016)
- Three composite samples were made for each season . 2014, 2015 and 2016
- The whole composite samples were prepared by mixing the three season's 1A+2A+3A Khartoum locations composite samples, 1B+2B+3B White Nile locations composite samples and 1C+2C+3C Northern Kordofan locations samples.

2.1Preparations of samples:

Gum nodules were dried at room temperature, then hand cleaned by to insure free from sand, dust and bark impurities, then ground using a mortar and pestle, kept in labeled container for analysis.

Fig. 2.1 Neem gum Sample

2.3Analytical Methods

2.3.1Determination of Moisture content

The moisture content was calculated according to (AOAC, 1990) method. Accurately weighed one gram of the ground gum was heated in an oven (Heraeus) at 105°C to a constant weight. Then, the moisture content was determined as percentage of the lost weight to the total weight as follows;

$$\text{Moisture content (\%)} = \frac{W_1 - W_2}{W_1} \times 100 \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (2.9)$$

W_1 = Original weight of sample (g).

W_2 = Weight of sample after drying (g).

2.3.2Ash content

Ash content was determined according to(AOAC, 1990) method. Ash content was determined by weighing the sample (1g) in a pre-weighed ashing dish after heating at 550 **C** for 5 h. The dish was cooled to room temperature in a desiccators and was weighed again. The weight loss was calculated as a percentage of the initial weight taken as follows;

$$\text{Ash (\%)} = \frac{W_3 - W_1}{W_2 - W_1} \times 100 \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (2.10)$$

Where:

W_1 = Weight of the empty crucible (g).

W_2 = Weight of the crucible + the sample (g).

W_3 = Weight of the crucible + ash (g).

2.3.3 pH Measurement

The pH values were measured directly in a homogenate prepared with 1% (w/v) gum powder (on dry basis) in distilled water, using a JENWAY 3510 pH meter at room temperature.

2.3.4Specific optical rotation $[\alpha]_D^T$

The specific rotation was measured for 1% (w/v)solution (on a dry basis) using an Optical Activity Bellingham and Stanley Ltd. polarimeter fitted with a sodium lamp and with a cell of path length of 20 cm at 25**C**. The specific optical rotation was calculated according to the relationship:

$$\text{Specific optical rotation}[\alpha]_D^T = \frac{\alpha \, x \, 100}{L \, x \, C} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (2.13)$$

Where:

α = Observed angle of rotation.

L = the length of sample holder in decimeters (dm).

C = concentration in gm/100ml

T = Temperature.

2.3.5 Viscosity measurements

The viscosity was determined on gum solutions using U-tube viscometer immersed in a constant temperature water bath set at 25 C°. Gum solutions (0.5%, 1%, 2%, 3%, and 4%) was prepared in 0.2 M NaCl, then filtered and transferred into the viscometer. The intrinsic viscosity was calculated by:

Relative viscosity [η_{rel}] = $\eta/\eta_{\circ}$ = $t/t_{\circ}$

Specific viscosity [η_{sp}] = $\eta_{rel} - 1$

Reduced viscosity [η_{red}] = η_{sp}/C

Intrinsic viscosity [η] = $\lim_{c \to 0} \eta_{sp}/C$]

Intrinsic viscosity [η] is determined from the intercept in the plot of $\eta_{reduced}$ as a function of a sample concentration at zero concentration (infinite dilution). The inherent viscosity is determined from the intercept in the plot of η_{rel}/c as a function of a sample concentration at zero concentration (infinite dilution) .

2.3.6 Nitrogen and protein Content

The total nitrogen in gum samples was determine by used The Kjeldahl method according to AOAC (1990). The method consists of three basic steps: step one digestion of the samples in sulfuric acid with a catalyst, which results in conversion of nitrogen to ammonia, step two distillation of the ammonia into a trapping solution, and step three quantification of the ammonia by titration with a standard solution. The reactions involved in these steps can be shown as follows:

- Sample + H_2SO_4 (conc.) +Heat → $(NH_4)_2SO_4$
- $(NH_4)_2SO_4 + 2\ NaOH \rightarrow 2NH_3 + Na_2SO_4 + 2H_2O$
- $NH_3 + H_3BO_3 \rightarrow NH_4^+ + H_2BO_3^-$
- $H_2BO_3^- + HCl \rightarrow H_3BO_3 + Cl^-$

0.5 gram of each sample (in duplicate) was weighed and transferred to Kjeldahl digestion flasks and Kjeldahl tablet (copper sulphate-potassium sulphate catalyst) was added to each. 10 cm^3 concentrated, nitrogen free, sulphuric acid was added. The tube was then mounted in the digestion heating system which was previously set to 240^oC and capped with an aerated manifold. The solution was then heated at the above temperature until a clear pale yellowish-green color was observed which indicates the completion of the digestion. The tubes were then allowed to attain room temperature. Their contents were quantitatively transferred to Kjeldahl distillation apparatus followed by addition of distilled water and 30% (w/v) sodium hydroxide. Steam distillation was then started and the released ammonia was absorbed in 25 cm^3 of 2% boric acid. Back titration of the generated borate was then carried out versus, 0.02M, hydrochloric acid using methyl red as an indicator. Blank titration was carried in the same way.

$$N\% = \frac{14.01\ x\ M\ x\ (\ volume\ of\ titrant - volume\ of\ blank) x\ 100}{weight\ of\ sample\ (grams)}$$

Where: M is the molarity of hydrochloric acid. Protein content was calculated using nitrogen conversion factor resulting from amino acid analysis as follows:

protein% = N% x 6.51

2.3.7 Acid Equivalent weight

A glass column was packed with an Amberlite Resin IR ($120H^+$). HCl 0.2M was passed through the column until the resin was thoroughly washed with the acid. Then this was followed by distilled water until the column was chloride free (20 cm^3 eluent equalize to one drop 0.1N NaOH). 50 cm^3 of 3.0% gum solution was passed through the column, followed by the distilled water until a volume of 250 ml of the eluent and washing were collected. This 250 cm^3 of the eluent titrated

against 0.1N NaOH. The apparent equivalent weight of the acid was calculated by:

$$Equivalent\ weight = \frac{weight\ of\ the\ sample\ \times\ 1000}{Volume\ of\ titer\ \times\ molarity\ of\ alkali}$$

2.3.8 Total Uronic acid

Uronic acid% was determined by multiplying the molecular weight of uronic acid (194) by 100 and dividing by the apparent equivalent weight of gum sample as follows:

$$Uronic\ acid\% \quad = \frac{194\ \times\ 100}{acid.Eq.wt.}$$

2.3.9 Determination of sugar composition

Industries (Marcrae, 1985). HPLC is widely considered to be a technique mainly for biotechnological, biomedical, biochemical research, and for the pharmaceutical industry, is as well widely used in a lot of fields such as cosmetics, energy, environmental, and food

2.3.9.1 Sample preparation

The samples were hydrolyzed to liberate the sugar residues. Sample was weighed out (100 mg, taking into account the moisture content) and added to 10 cm_3 of 4% H_2SO_4 and incubated at 100 ^0C for 6 hours. Following this, 1g of $BaCO_3$ was added to the solution and left overnight (minimum of 12 hours) to neutralise the solution. After $BaCO_3$ treatment, universal indicator strips were

used to ensure that the sample was neutral before proceeding to the next stage. The solution was then centrifuged at 2500 rpm for 10 minutes to allow the Barium Sulphate (formed from neutralizing the H_2SO_4) to settle. The supernatant was removed and filtered through a 0.45 μm Whatman nylon filter and then diluted 1:1 with 70/30 Acetonitrile/buffer. This constituted the final solution of which 1ml was put in a vial (filtered via 0.45 μm filter) prior to injection into HPLC column.

2.3.9.2 Method

The purpose of analyzing the gum samples by HPLC was to determine the relative concentration of each sugar residue present in the sample, namely rhammnose (Rha), arabinose (Ara), galactose (Gal) and glucuronic acid (GlcA).

Before analysis of the gum samples, calibration curves of these sugars were prepared. Stock concentrations of 5 mg cm-3 for each sugar were made up by hydrating in 70/30 acetonitrile/buffer for 2 hours. Dilutions of the stock solution achieved six different concentrations for each sugar over a range of 2.5–0.5 mg cm-3. This allowed six levels for the calibration curve and an average of 3 replicates for each level was used to ensure accuracy. This calibration allowed the determination of the unknown sugar content for the gum samples. The concentration of each sugar was calculated by peak height and expressed as a % of the total sugar content.

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2.3.10 Determination of Total Polyphenol (Tannin %)

The tannin content determined according to the Prussian blue assay originally devised by price and Butler and subsequently modified (Graham, 1992). Tannin content is taken her to represents the "total phenols" and more accurately the "Gallic acid equivalents" as Gallic acid – 99% in purity purchased from sigma Aldrich – was used as analytical standard for determining the hydrolysable tannins. 500 μg/g Gallic acid was prepared in distilled water. This was then serial diluted to obtain concentration of 400, 300, 200, 100, and 50 μg/g as standards. 0.10*ml* of each sample or standard was dispensed in a 30*ml* universal. 3ml of distilled water was added fallowing by vortex mixing for 30sec. Next 1.00ml of 0.016*M* (0.526*g*/100*ml*,w/v) Potassium hexacyano ferrate(III) [$K_3[Fe(CN)_6]$,Fallowed by 1.00ml of 0.02(0.324$gFeCl_3$/100*ml*d.w+ 0.83*mlHCl*) Ferric Chloride ($FeCl_3$) were added and immediately mixed by vortex mixer 30sec. Exactly 15 min after adding the reagent to the sample 5.00ml of stabilizer

was added and vortex mixed 30sec.The stabilizer was prepared by mixing 10.00ml of 85% phosphoric acid, (H_3PO_3), 1.00ml of 1% gum Arabic , and 30 ml of distilled water, then exactly after15 min The absorbance was read at 700 nm in triplicate for standard solutions, using (Perkin Elmer Lambda XLS+, UV/Vis spectroscopy). Solvent only blank were also prepared by adding all reagents and 0.1ml of solvent instead *Azadirachta indica* gum or Gallic acid standards. The absorbance was read at 700 nm in triplicate for all using Perkin Elmer Lambda 40 UV/Vis spectroscopy. The error in measuring the tannins content was below 10% for all samples and the average was taken.

2.3.11Calorific Value

The IKA C1 calorimeter system was calibrated by standard IKA C723 Benzoic acid tabs about 1g (2 Tabs), with cross cal.val. 26461J/g, RSD 0.03%, and LOT SZBD2180V.The temperature was 19 C, the gas pressure (Oxygen) was 30 bar, and the Pump flow 2700 rpm. Then weighted about 0.5g of A*zadirachta indica* gum and placed into a small plastic bag which have cross value 46463j/g, the bag was covered by rolling and placed into a decomposition vessel which is surrounded by a water jacket. The sample was combusted in an oxygen atmosphere, and the calorific value of the sample was calculated.

2.3.12Cationic composition

Atomic absorption spectrometry is a technique which can be applied effectively to determine about 70 elements. It is based on the absorption of radiations by the atoms of a particular element in the ground state, raising them to exited states. Excitation is produced by radiation energy at a wavelength equivalent to the energy needed to lift an atom from its ground state to higher level, the energizing radiation is thus absorbed and the amount of absorption is directly dependent on the population of the ground state atoms in the flame. The sample solution is aspirated in the gaseous state by vaporization and dissociation of molecules. A hollow cathode lamp, which consists of a cathode of the element of interest or coated with it and anode at a low pressure of neon or argon, is used as a source of radiation. A monochromator is used in conjunction with the hollow cathode lamp to isolate the desired spectrum. The radiation that finally reaches the detector system is amplified.

Na, K and Ca were determined by flame photometer and Ca determined vbc'\ by spectrophotometer according to vanadate / molybdate method Duke, (1981).

2.3.13Amino acid analysis

Wet dry 4An accurately weighed quantity of sample (~5mg) is placed in a sample tube and hydrochloric acid (Pierce, constant boiling (6N), 40 ml) containing phenol (saturated solution in water, 0.6 ml) and dodecanethiol (Sigma, 3 ml) are added. The tube is sealed and placed in a heating block at 160 0C for 1 hour. After being allowed to cool the tube is opened and placed in a dessicator under vacuum over sodium hydroxide for 60 minutes. Sodium citrate loading buffer (pH 2.2) is then added to dissolve the residue, and the resulting solution is filtered under centrifugation through a 0.2 µm filter, prior to chromatographic analysis.

2.3.13.1 Chromatographic analysis

An aliquot of the filtrate is injected into an amino acid analyser (Biochrom 30 instrument) and separation performed using a cation exchange sulphonic acid high performance sodium column by elution with a series of buffers over the pH range 3.2 to 6.45 (buffer flow rate 35 mL/hour). Peak detection is achieved by post-column derivatisation using ninhydrin (flow rate 25 mL/hour) at 1350C and measuring the absorbance (at 570 and 440 nm). Quantization is performed using Chrome eon software and calibration curves for each amino acid of interest.

2.3.14 Determination of number average molecular weight by Osmotic pressure

150 micro meter of different concentration(0.25%, 0.5%, 1%, 1.5%, 2%, 2.5%, and 3%) of *Azadirachta indica* gum samples was ejected into Osmometere 150 and the osmotic pressure was digital determined at 25 C^0. The number average molecular weight determined using equation:

$$Theintercept = \frac{\pi}{C_{\rightarrow 0}} = \frac{RT}{M_n}$$

Π = osmotic pressure, R = Gas constant, T is Temperature = Concentration, M_n = Number average molecular weight.

2.3.15Molecular weight and molecular weight distribution

2.3.15. 1Sample preparation

2.0 mg/ml fallowed by one dilution (1.0mg/ml) gum samples was prepared (based on dry weight) in 1mM phosphate buffer at pH 7 containing 0.2M NaCl, and hydrated by roller (SRT9. Stuart Scientific, UK) mixing the solution overnight to ensure that the sample fully dissolved and hydrated. The solutions were then centrifuged for 10 minutes at speed of 3000 rpm using Megafuge 1.0R (Heraeus SEPATECH, Germany) centrifuge. The gum solutions were then filtered using 0.45-µm nylon filter (Whatman, 13 mm) prior to injection into the GPC-MALLS system.

2.3.15.2 GPC-MALLS

The gum exudates from *azdirachta indica* gum tree were subjected to average molecule weight M_w and molecular weight distribution study using different techniques such as Osmometere, gel permeation chromatography (GPC-MALLS). The system utilises Waters (Division of Millipore, USA) Solvent Delivery System Model 6000A connected to a column containing Superose 6 (Amersharm Biosciences) (10 x 300mm), manual Rheodyne Model 7125 syringe. The column eluent was monitored by three detectors, refractive index (RI) Wyatt Optilab DSP inter ferometricre fractomter operated at 633 nm (Wyatt Technology Corporation, USA), multi-angle laser light scattering photometer DAWN EOS using He–Ne laser at 690 nm (Wyatt Technology Corporation, USA), and an Agilent 1100 series G1314A UV detector (214 nm, Agilent Technologies). RI provides an accurate concentration profile, MALLS enables absolute molecular mass and radius of gyration (Rg), and the UV detects the proteinaceous components of the gum. (Katayamaa, *et al*., 2006).The data was processed by the Astra for Windows software (version 4.90.07, Wyatt Technology Corporation).

2.4 Rheological measurement

The 50% w/w (based on dry weight) of gum *Azadirachta indica* solution were prepared in distill water, then the solution agitated on a tube roller mixer (SRT9. Stuart Scientific, UK) overnight to ensure that the sample fully dissolves and hydrated. The solutions were then centrifuged for 10 minutes at speed of 3000 rpm using (Megafuge 1.0R, Heraeus SEPATECH, Germany) centrifuge. One dilution was prepared from stock solution (25% w/w) and recent rifuged as previous procedure to investigate the rheological behavior. Rheological measurements were carried out using Malvern Series KINEXUS pro+ rheometer (Malvern Instruments Ltd., Malvern, Worcester, UK), fitted with cone and plate geometry with a cone diameter of 40 mm and an angle of 20. Steady shear viscosity curves were measured for gum solutions ranging from 10 to 50 % w/v both upon shear rate ramp-up (from 0.01 to 10000 s-1) and subsequent shear rate ramp-down (from 10000 back to 0.01 s-1). Dynamic rheological measurements, to determine the elastic modulus (G′), viscous modulus (G″) and dynamic viscosity, were performed in the frequency range of 0.1–10 Hz. The linear viscoelastic region was assessed, at 1 Hz. The temperature of the samples were controlled within 0.1 C using a Peltier element. The rheometer control and data processing was done by computer software.

Fig. 2.2 Homogenizer

2.5 Emulsification properties of the gum

2.5..1 Emulsion preparation

Distilled water was added to 8 g of the gum sample (based on dry weight) in a glass bottle to give 40 g in total with a concentration of 20 % (w/w) gum

solution. The sample was agitated on a roller mixer overnight until the sample is completely dissolved. Exact calculated grams for each samples (in the range from about 19.97 to about 20 g) of the prepared gum solution was filtered using 100 µm mesh then mixed with 0.52 ml of 10 % (W/V) sodium benzoate solution as a preservative, and 0.48 ml of 10 % (W/V) citric acid solution to adjust the pH to 4, 15.71ml , and 15.73ml of distilled water was added, then, 4.2 g of ODO oil (10%) was added to the gum solution to give a total of 40 g and final concentration of 10%.The mixed solution was homogenized for 3 minutes using a POLY TRON (PT 2100, KINEMA TICA AC) homogenizer at 22000 rpm (Fig. 2.3). Impeller (PTDA21 9 mm tip diameter) was used as dispersing tool. To achieve small particle size < 1 micron, the pre-emulsified mixture was homogenized using a high-pressure NanoVater (NV30-FA, MITSUBISHI GOT1000.). In order to achieve effective disaggregation of the gum it was passed twice at 75 MPa. The final emulsion kept in closed glass universals, and emulsion particle size was measured. then putted at 60^{o}C in the Vacuum Oven (GALLENKAMP. OVA031.XX1.5). Droplet size was remeasured after 3 and 7 days.

Fig. 2.3POLY TRON (PT 2100, KINEMA TICA AC) homogenizer

2.5..2 Droplet size analysis

The droplet size distribution of the emulsions was measured, using Mastersizer 3000 (Fig. 2.4), a laser diffraction particle size analyzer (Malvern Instruments). Distilled water was used as dispersant and a value of 1.45 was used for the refractive index for oil phase (ODO). Emulsification stability of samples kept at 60C was evaluated by particle size change after accelerated stability test

for 3 and 7 days. The particle size of the emulsions was described by the volume median diameter (VMD).

Fig. 2.4 Mastersizer 3000 Instrument

2.5..3 Emulsion stability index of *Azadirachta indica* gum

Emulsification stability was evaluated by the change in the particle size of emulsion after acceleration test. Emulsion stability index (ESI) was calculated according to PHRC grading system using the equation:

ESI= d0.5as prepared + (d0.53 days@60C - d0.5as prepared) + (d0.57days@60C - d0.5as prepared).

Chapter Three

Result and Discussion

A number of physicochemical and chemical methods were used to characterize *Azadirachta indica* gum. The characterization of gums is very important when we need to use the gum in the industrial applications. The study of chemical and physical properties of gum is used to ensure the purity and to a void mixed samples and to report the specification of the samples under study. Tables 3.1, 3.2 and 3.3 show analytical data of *Azadirachta indica* gum samples seasons 2014- 2016 and 2016from Khartoum ,White Nile and Northern Kordofan States respectively.

Tables (3-1)(3-2)(3-3) shows analytical data of the Samples under the study .Analysis of samples was carried out in triplicate and the averaged . and Standard Division.

3.1 Moisture content

The moisture content of the gum is usually affected by the season of collection, the prevailing climate conditions and the storage condition. The moisture content of *azdirachta indica* gum samples collected from Khartoum ranged between 11.2-13.1% with an average value of 12.4% as shown in Table (3.1).*azdirachta indica* gum samples from White Nile had moisture contents in the range of 11.3-11.2% with an average value of 11.2% as shown in Table (3.2). Table (3.3) shows the moisture content of *Azadirachta indica* gum samples collected from Northern Kordofan which were found in the range of 12.0-11.7% with an average value of 11.7%. The moisture contents result were relatively in the range mentioned by Anderson (1968) who claimed that moisture content of *Azadirachta indica* gum range between 11.9-13.0.

Table (3-1) Physiochemical Properties—Khartoum State

Sample	Mois-ture	Ash %	PH	Nitrogen	Pro-tein%	O.R	Intrinsic viscosity	Acid Eq.wt	Uronic acid	Mn×10^5
A_1	11.2	3.2	4.8	5.1	31.9	-67	36.5	1714	11.3	12.3×10^5
A_2	12.9	3.4	4.6	5.5	31.6	-67	35.7	1711	11.3	11.9×10^5
A_3	13.1	3.0	4.6	4.9	30.6	-67	35.9	1700	11.4	11.8×10^5
Mean	12.4	3.2	4.7	5.2	31.9	-67	36.6	1708	11.3	12.0×10^5
S.D										

Table 3.2Physiochemical Properties—White Nile State

Sample	Moisture	Ash%	PH	Nitrogen	Pro-tein%	O.R	Intrinsic viscosity	Acid Eq.wt	Uronic acid	Mn×10^5
B_1	**11.3**	**3.4**	**4.8**	**4.8**	**30.0**	**-65.0**	**26.3**	**1631**	**12.00**	**8.6×10^5**
B_2	**10.9**	**3.0**	**4.8**	**4.8**	**30.0**	**-65.1**	**24.9**	**1601**	**12.11**	**9.9×10^5**
B_3	**11.5**	**3.5**	**5.0**	**5.0**	**31.3**	**66.1**	**26.9**	**1653**	**11.70**	**11.8×10^5**
Mean	**11.2**	**3.3**	**4.9**	**4.9**	**30.4**	**65.5**	**26.3**	**1628**	**11.9**	**10.8×10^5**
S.D										

Table 3.3 Physiochemical Properties— Northern Kordofan State

Sample	Moisture	Ash%	PH	Nitrogen	Protein%	O.R	Intrinsic viscosity	Acid Eq.wt	Uronic acid	Mn×10^5
C1	**11.2**	**3.2**	**4.8**	**5.1**	**31.9**	**-67**	**23.6**	**1714**	**11.3**	**5.8×10^5**
C2	**12.9**	**3.4**	**4.6**	**5.5**	**31.3**	**-67**	**24.9**	**1711**	**11.3**	**6.1×10^5**
C3	**13.1**	**3.0**	**4.6**	**4.9**	**30.9**	**-67**	**24.2**	**1700**	**11.4**	**7.3×10^5**
Mean	**12.4**	**3.2**	**4.7**	**5.2**	**31.3**	**-67**	**24.2**	**1708**	**11.3**	**6.4×10^5**
S.D										

3.2 Ash content

Tables (3.1, 3.2 and 3.3) show the ash content of *Azadirachta indica* gum from Khartoum state, White Nile state and Northern Kordofan state 2010 respectively. The mean values was found to be 3.2%, 3.3% and 3.0% which is almost similar to those results obtained by Anderson (1968) which fell in the range of 3.3- 3.0%. but far less than those obtained by Andreson, *etal*., (1966)

3.3 pH value

Tables 3.1, 3.2 and 3.3 show the PH value that there is no significant variation in three locations 4.8-4.6 for Khartoum location , 4.8-5.0 for White Nile location, and Northern Kordofan 4.3-4.4 . the average value is about 4.7, 4.9, and4.2. respectively. Accordingly the gum Azadirachta indica gum has low acidity nature. This results is less than the result reported by Anderson (1968) which was 5.5.

3.4 Nitrogen --Protein Content%

Tables 3.1, 3.2 and 3.3 show the nitrogen % of Khartoum samples fall in the range 5.1– 4.9 with average value5.2, that of White Nile samples fall in the range 4.8– 5.0 with average 4.9. and that of Northern Kordofan samples fall in the range 4.6-4.9 with average value4.8. The protein % of Khartoum samples fall in the range 31.9 – 30.6 with average 31.9 , that of White Nile samples fall between30.0–31.0 with average 30.4.of Northern Kordofan samples fall in the range 29.0–31.0 with average value30.2. The percentage of the nitrogen and hence the protein from the three locations are almost the same. These values are less values than Anderson(1986) results(35.0-37.5).

3.5 Specific Optical Rotation

Tables 3.1, 3.2, and 3.3 show Specific rotation of Khartoum samples fall between-67 ^{o}to-67 ^{o} with average -67^{0}whereas that of White Nile samples fall between-65^{o}to-66.1^{o} with average 65.5^{o}.and the Northern Kordofan samples fall between -66^{0}to -65 ^{o} with average value -65.5 almost identical. These values are all same from white Nile and northern kordofan locations and this result is greater than Anderson(1968) -58 – -62^{0}the result of optical rotation of *azdirachta indica* gum in this study was small than result add by Mukherjee 1954) -72 .

3.6 The intrinsic viscosity

The viscosity of *Azadirachta indica* gum (neem gum)was measured using U-tube Ostwald viscometer. They are no significant different between three locations. The results is found to be in the range of Khartoum 36cm^{3}g^{-1}–,White Nile 26cm^{3}g^{-1} and Northern Kordofan 24cm^{3}g^{-1} as shown in Fig. 3.1.These results is shows Khartoum samples is greater than the results obtained by Anderson, *etal.*,(1966) 25cm^{3}g^{-1}.but samples from White Nile and Northern Kordofan which are almost similar to those results obtained by Anderson (1968) which fell in the range.

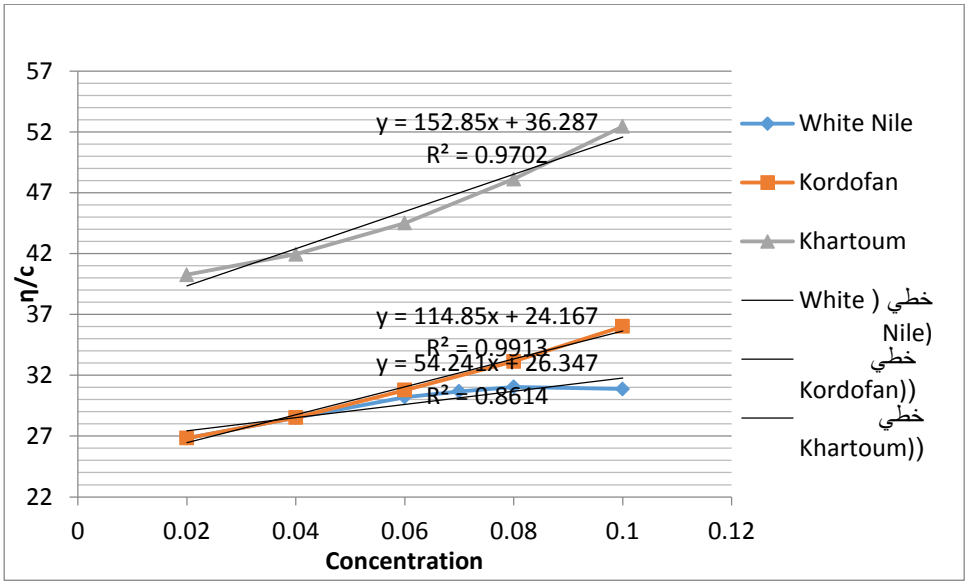


Fig.3. 1 Intrinsic viscosity: (η/c) Variation with concentration

3.7The equivalent weights and Uronic acid

Tables 3.1, 3.2 and 3.3 obtained values of the equivalent weights for Khartoum samples fall between 1714 and 1700 with average 1708 , those White Nile samples fall between 1631-1653 with average 1628. And samples from Northern Kordofan fall between 1514-1554 with average 1543 The values are slightly different. equivalent weights of acid was greater in the samples than what was found by Anderson(1968) who illustrated arrange of 957 to 990.

The Uronic acid% of Khartoum samples falls between 11.3 - 11.4 with average 11.3, that of White Nile samples falls between 12.0-11.10 with average 11.9. and samples from Northern Kordofan samples falls between 12.8-12.4 .

with average 12.5.The Uronic acid is greatly lesser than what was found by Anderson *etal.*,1968 (28.3).

3.8 Cation composition

Tables 3.4show the Cationic composition of *azdirachta indica* gum (neem gum) from three locations (Khartoum, White Nile and Northern Kordofan) .The most valuable cation were Magnesium followed by sodium, Calcium, potassium, Copper, Zinc, Phosphor, Arsenic, Cadmium, but the Lead, Iron, Nickel, and Chrome not found in the samples.

Table 3.4: **Cationic composition of Neem gum from three locations.**

Cation	Khartoum State	White Nile State	Kordofan State	Unit
Ca	**12.600**	**9.6700**	**10.200**	**mg/g**
Mg	**57.100**	**52.900**	**38.900**	**mg/g**
Zn	**0.0710**	**0.0780**	**0.5800**	**mg/g**
Pb	**0.0000**	**0.0000**	**0.0000**	**mg/g**
P	**0.0200**	**0.0400**	**0.0400**	**mg/g**
Na	**13.939**	**24.545**	**18.409**	**mg/g**
K	**6.6870**	**11.932**	**6.2050**	**mg/g**
Cu	**0.1852**	**0.1367**	**0.1136**	**mg/g**
As	**31.4**	**55.7**	**32.2**	**Ppm**
Cd	**25.1**	**14.4**	**31.6**	**Ppm**

3.9 Amino acids composition

Table 3.5 shows The amino acid content of *Azadirachta Indica* (neem tree) gum)were determined the results showed the presence of seventeen amino acids expressed in mg/g .namely Asparagine, Threnine, Serine, Glutamine, Proline, Glysine, Cysteine, Alanine, Valine, Methionine, Isoleucine, Leucine, Tyrosine, Phenylalanine, Histidine, Lysine, and Araginine were the principal amino acids in *Azadirachta Indica* gum.

Table 3.5 Amino Acid of Neem gum from three locations

No.	Name	White Nile (mg/g)	Kordofan (mg/g)	Khartoum (mg/g)
1	ASP	12.55	6.32	21.23
2	THR	4.39	2.04	10.49
3	SER	1.75	0.55	6.58
4	GLU	4.31	1.93	14.54
5	PRO	11.06	10.90	14.96
6	GLY	3.03	1.92	5.90
7	ALA	9.73	8.72	8.73
8	CYS	2.45	3.22	0.31
9	VAL	13.64	10.46	19.04
10	MET	0.19	0.14	0.71
11	ILE	8.69	6.75	13.32
12	LEU	12.39	9.20	20.01
13	TYR	3.60	1.73	5.19
14	PHE	10.67	5.64	16.19
15	HIS	4.32	3.04	8.20
16	LYS	3.88	2.77	7.66
17	ARG	7.73	8.05	22.43

3.10 Sugar composition

Table 3.6 showed the sugar contents of *azdirachta indica* gum were measured using HPLC technique and were found to be Khartoum samples22% arabinose, 24% galactose ,4% xylose and 4% rhammnose, While in White Nile samples 14% Arabinose 17% galactose ,3% xylose and 3% rhammnose and Northern Kordofan samples. And arabinose content18%, galactose 20%, xylose3% and rhammnose 2%. classification (Anderson.1974), in which arabinose had a higher percentage than galactose, and the lowest percentage of rhammnose. The results greater than that reported by Nyak *etal.*,(1982) reported sugar composition of 15.5% arabinose ,12.4% galactose, 5.1% xylose.

An earlier communication reported the characterization of a high-molecular-weight glycoprotein from neem gum (Ramakrishna, 1981).This glycoprotein had a protein: carbohydrate ratio 4.24:1 and a large molecular size. The carbohydrate potion of the glycoprotein consisted of glucosamine mannose, arabinose, galactose, fucose. Xylose and glucose in molar ratio of 3:4:3:2:2:1:1. Furthermore, the carbohydrate-peptide link in this glycoprotein was fond to be an acylgucosa- minyl- asparaginyl bond. These observation confirmed the isolated protease to be the same high-molecular-weight glycoprotein characterized previously.((Ramakrishna, 1981).

Table 3.6 Sugar Composition of Neem gum from three location

Region / Sugar%	Khartoum	White Nile	Northern Kordofan
Arabinose	22	14	18
Galactose	24	17	20
Xylose	4	3	3
Rhammnose	4	3	2

3.11 Colour Gardner and Tannin content

The colour Gardner of gum *Neem* is the range of (0.1 – 0.9).Table 3.7 shows that the values are increasing with time (24 hours and48 hours) due to oxidation of Polyphenol. Tannin content is calculated according to Kjeldahl method, according to table xxthe three samples have tannin content of about 413.3,915.6, and 1014.9 ppm for Kordofan, White Nile, and Khartoum respectively.

Table 3.7 Colour Gardena and tannin content

Sample	After 3 hours				Tannin (ppm)
	L*	a*	b*	Color Gardner	
Kordofan	98.1	-0.19	1.86	0.2	413.3
White Nile	97.18	-0.59	4.86	0.7	915.6
Khartoum	97.72	-0.36	3.09	0.4	1014.9
Sample	After 24 hours				Tannin (ppm)
	L*	a*	b*	Color Gardner	
Kordofan	98.63	-0.16	1.57	0.2	413.3
White Nile	96.53	-0.5	6,96	0.9	915.6
Khartoum	98.63	-0.35	2.77	0.4	1014.9
Sample	After 48 hours				Tannin (ppm)
	L*	a*	b*	Color Gardner	
Kordofan	96.99	-0.13	2.92	0.4	413.3
White Nile	95.58	-0.48	7.9	1.2	915.6
Khartoum	98.35	-0.4	3.23	0.4	1014.9

3.12 Calorific value

Table 3.8 showed The calorific value of *azdirachta indica* gum was about 4.26 Kcal/g This Calorific value is very low. That's mean the gum very suitable to used as food additives.

Table 3.8 Calorific Value of Neem gum from three locations composite

sample name	sample three loca-tions	sample weight	bag Cal. value J/g	Gross cal. Value	Net Cal. value j/g	net.Cal.value. cal/g	Cal.Value Kcal/g
A. *Indic*a-gum	Composite	0.5121	46463	16037	17829.2	4259.5	4.26

3.13The number average molecular weight

The Fig 3.2 Shown the number average molecular weight determined by osmometry (Mn) of *Azadirachta indica* gum for Khartoum samples fall between 12.3×10^5–11.8×10^5g/mol .with average 12.0×10^5g/mol, those for White Nile samples fall between 8.6×10^5-11.8×10^5g/mol with average 10.8×10^5g/mol. And Northern Kordofan samples fall between 5.8×10^5–7.3×10^5g/mol with average 6.4×10^5g/mol .These values of Khartoum and White Nile samples are wide range than those reported for Anderson (1968).but samples from Northern Kordofan which is almost similar to those results obtained by Anderson (1968) which fell in the range Mn = 5.2×10^5– 7.1×10^5g/mol.

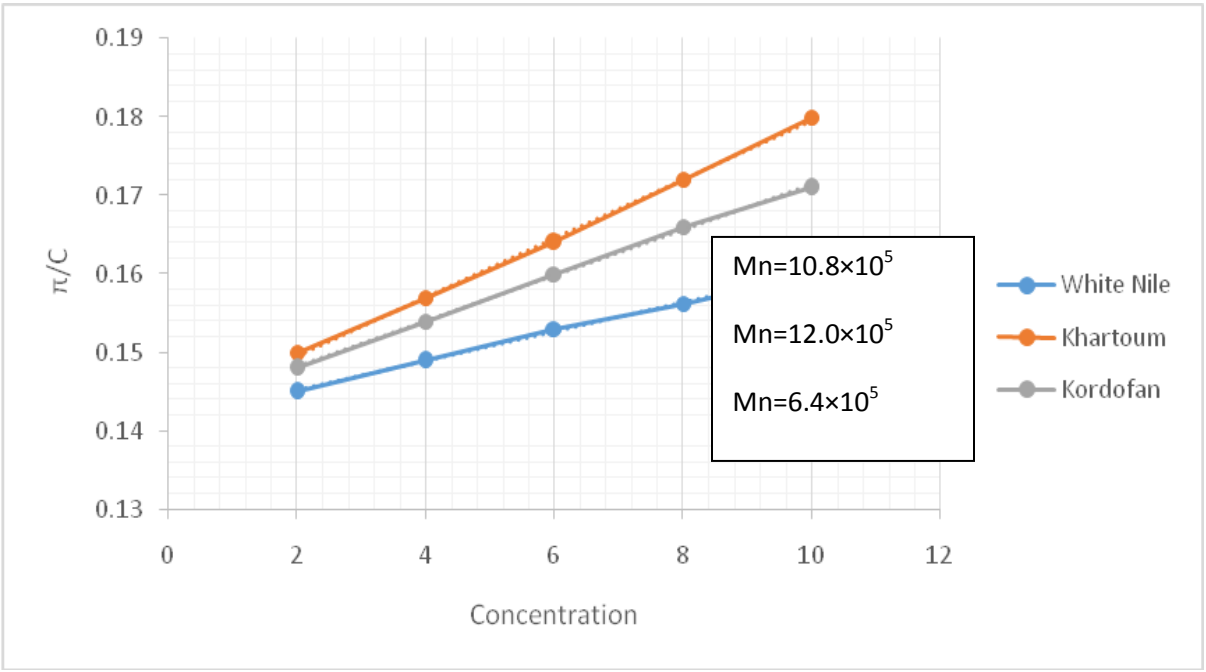


Fig 3.2 Number average molecular weight

3.14 Molecular Weight and Molecular Weight Distribution

GPC MALLAS show three fraction of the *Azadirachta indica* gum; Arabino-galactan protein (AGP), Arabinogalactan (AG) and Glycoprotein (GP) molecular weight of whole *Azadirachta indica* gum value form the three locations is $4.004x10^5$, $3.6x10^5$ and $3.7x10^5 g.mol^{-1}$respectively, with radius of gyration 25.6, 36.6 and 66.

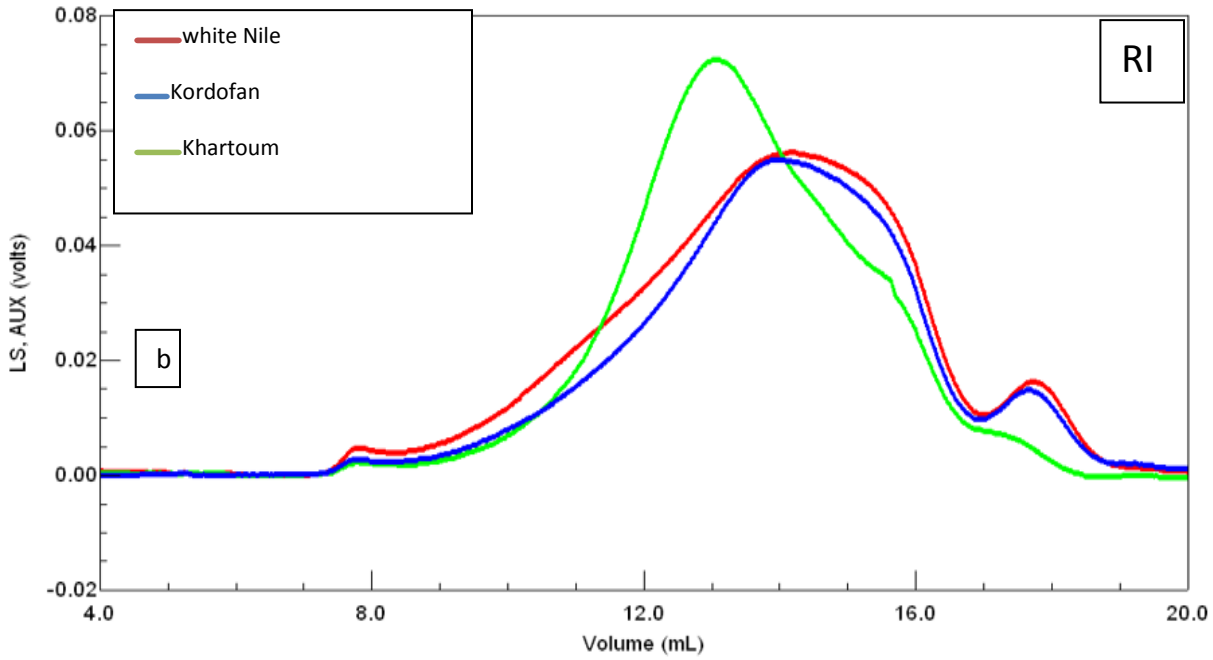
The light scattering (LS) detector on GPC-MALLS response for the three locations show two peaks. The first peaks has a high response corresponds molecular weight (AGP) content of $3.924x10^5$, $3.916x10^5$ and $5.086x10^5$ $g.mol^{-1}$, with radius of gyration 43.2, 52.2 and 63.6. The second peak is broader with lower response and it is due to see of AG and GP fraction $3.346x10^5$, $3.292x10^5$ and $3.139x10^{5.}$

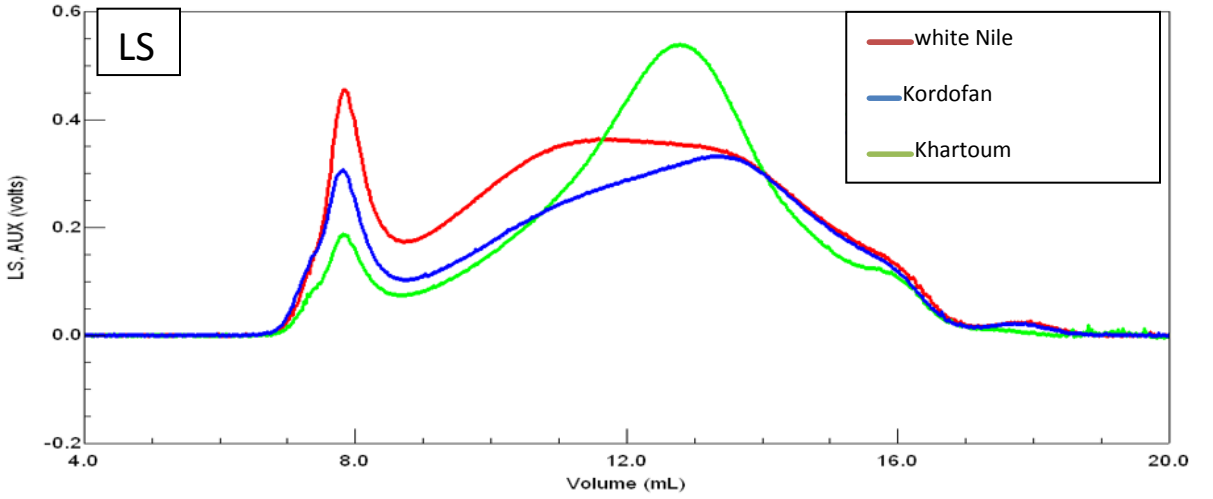
Figs 3.3 shows a typical elution profile for *Azadirachta indica* gum monitored using refractive index, response shows three peaks. The first beak AGP is very small mass 1.77, 0.87 and 1.34 for the three samples respectively. The second beak with very high mass is due to AG+GP fraction percent Masses 98.23, 99.13 and 98.66.

UV, GPC profiles for the *Azadirachta indica* gum clearly, indicate the significant high AGP content.

Table 3.10 Molecular weight parameters determined by GPC-MALLS

Samples	Mw whole gum ($x10^5$)	% Mass recovery	Rg (whole gum)/nm	Mw AGP	% mass (AGP)	Rg-AGP	Mw (AG+GP) ($x10^5$)	% mass (AG+GP)
White Nile	3.994	120.57	25.6	3.924	1.77	43.2	3.346	98.23
Kordofan	3.606	115.115	36.6	3.916	0.87	52.2	3.292	99.13
Khartoum	3.785	106.675	66	5.086	1.34	63.7	3.139	98.66


white Nile
Kordofan
Khartoum
RI
b
LS, AUX (volts)
0.08
0.06
0.04
0.02
0.00
-0.02
4.0
8.0
12.0
16.0
20.0
Volume (mL)


LS
white Nile
Kordofan
Khartoum
LS, AUX (volts)
0.6
0.4
0.2
0.0
-0.2
4.0
8.0
12.0
16.0
20.0
Volume (mL)

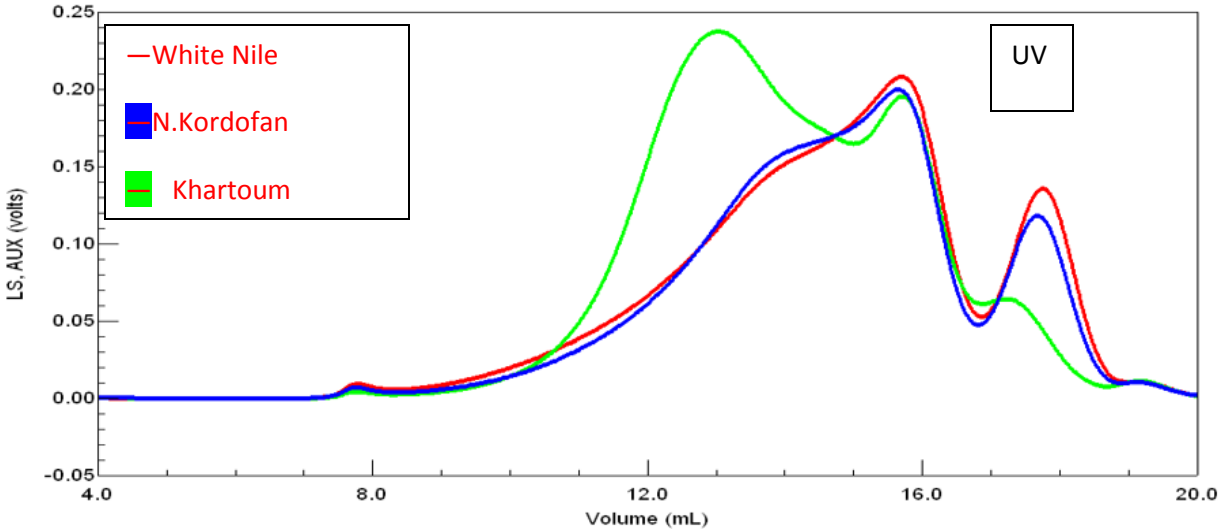


Fig.3.3 GPC-MALLS elution profile of *Azadirachta indica* gum monitored by (a) refractive index, (b) light scattering and (c) UV

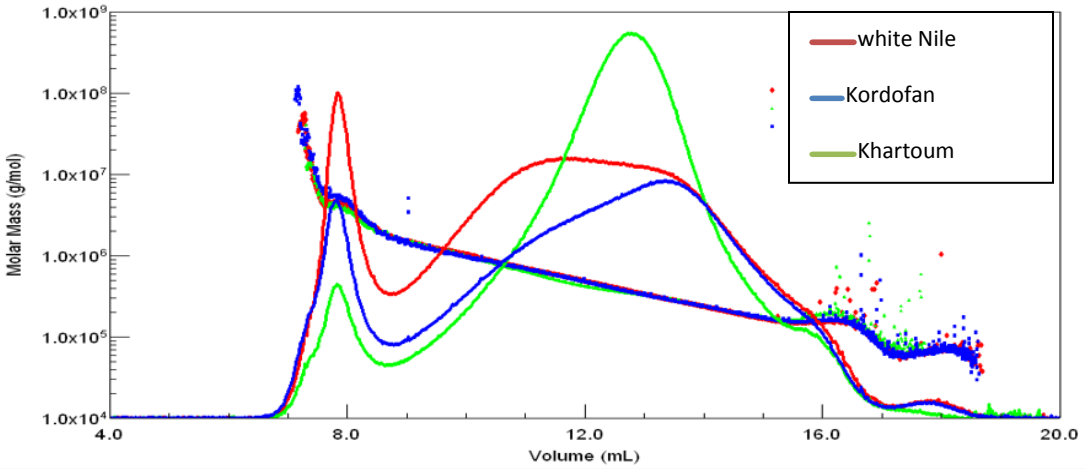


Fig. 3.4 Molar mass of gum neem

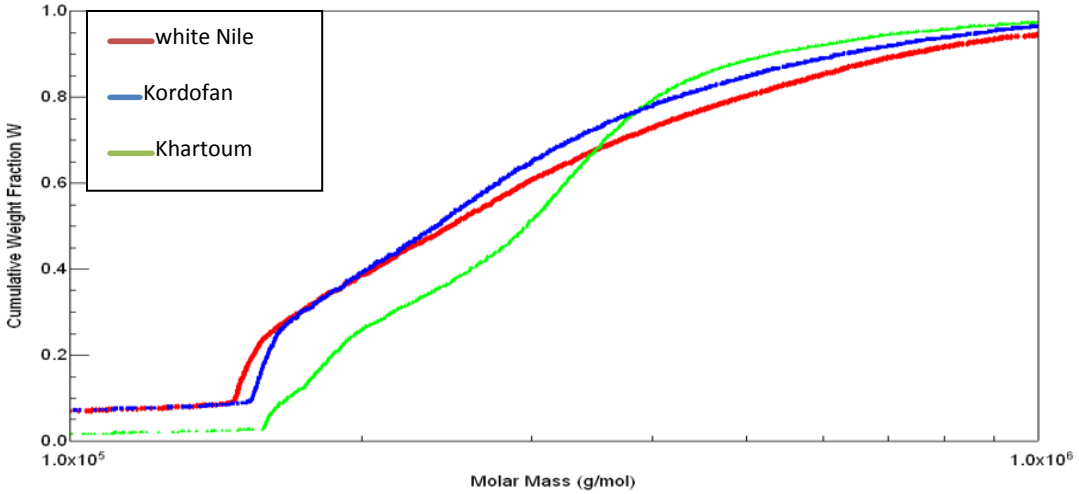


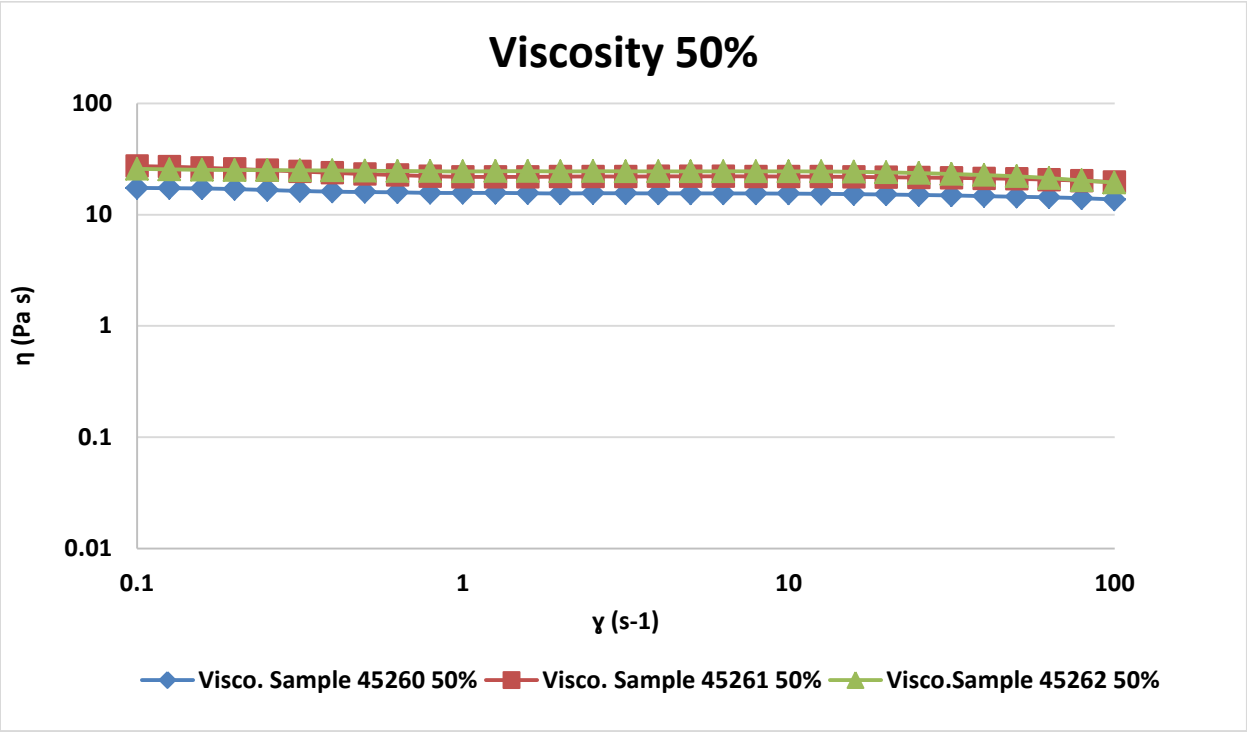
Fig. 3.5 CWF of *Azadirachta indica* gum

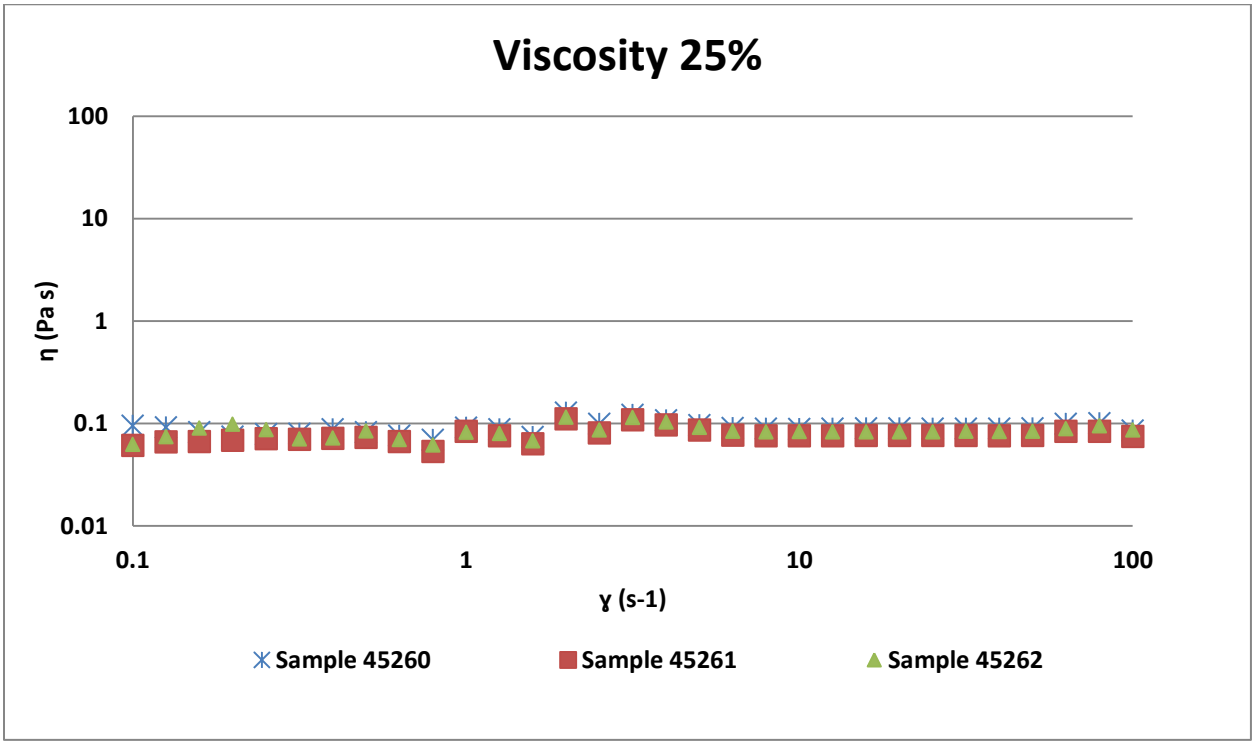
3.15 Dynamic Rheology

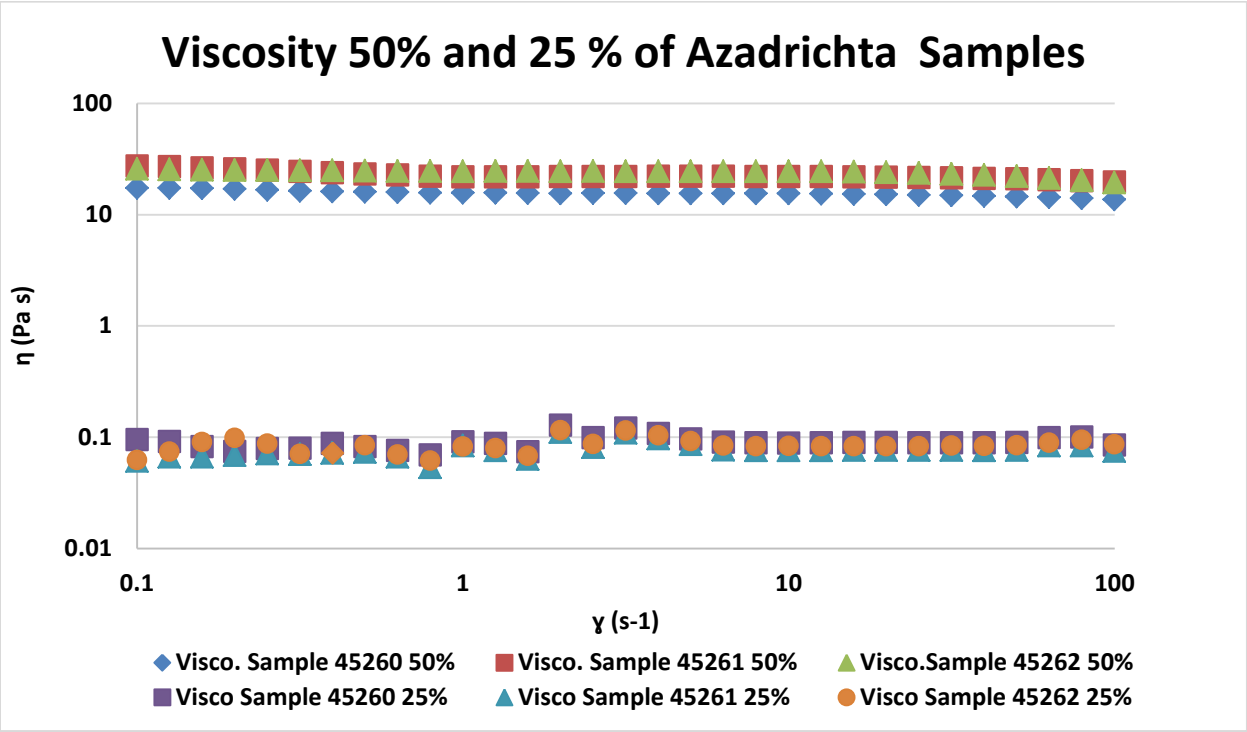
Fig. 3.6 shows that the loss modulus (G″) of Neem gumwas higher than the storage modulus (G′) and they do not cross, accordingly, gum Neem is a viscous not, elastic or viscoelastic.

3.16.1Shear flow viscosity

The effect of shear rate on viscosity, at 25Co, of *Azadirachta indica* gum solutions at different concentrations are shown in Figures (3.6). The results show Newtonian flow behavior in the shear rate range from 0.1 s-1 to 100 s-1, for *Azadirachta indica* gum at (50%)concentration. At low concentration in the shear rate range from 0.1 s-1to 8 s-1*Azadirachta indica* gum exhibit quite different of Newtonian flow behavior then from 8 s-1 to 100 s-1*Azadirachta indica* gum show Newtonian flow behavior. The presence of a large number of high molecular weight molecules increases the resistance to flow which, in turn, increases the apparent viscosity of gum solutions (Tanaka *etal.*, 2006).


Viscosity 50%
100
10
1
0.1
0.01
η (Pa s)
0.1
1
10
100
γ (s-1)
Visco. Sample 45260 50%
Visco. Sample 45261 50%
Visco.Sample 45262 50%


Viscosity 25%
100
10
1
0.1
0.01
η (Pa s)
0.1
1
10
100
γ (s-1)
Sample 45260
Sample 45261
Sample 45262



Figs 3.6 dynamic rheology

KEY

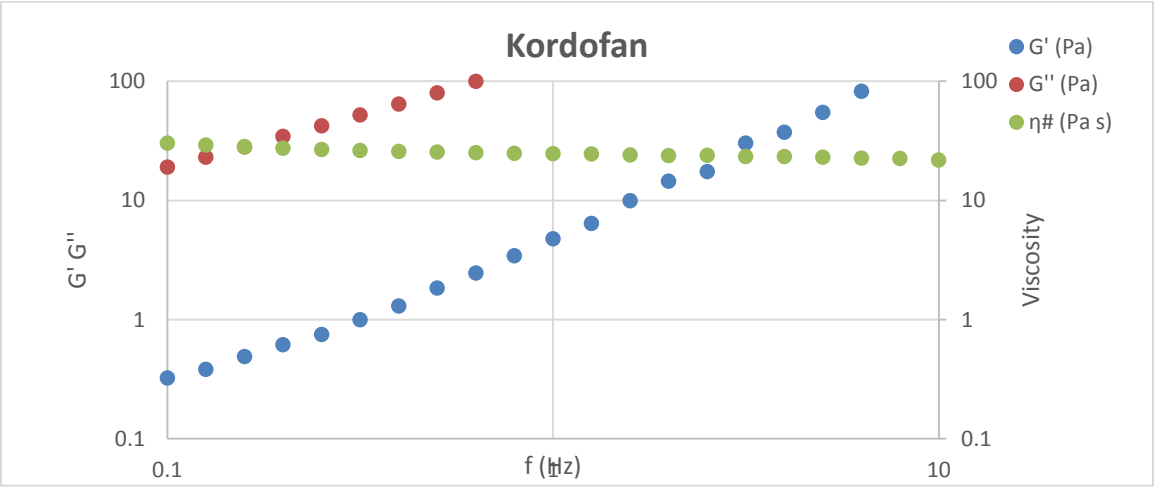
Sample 45260 White Nile

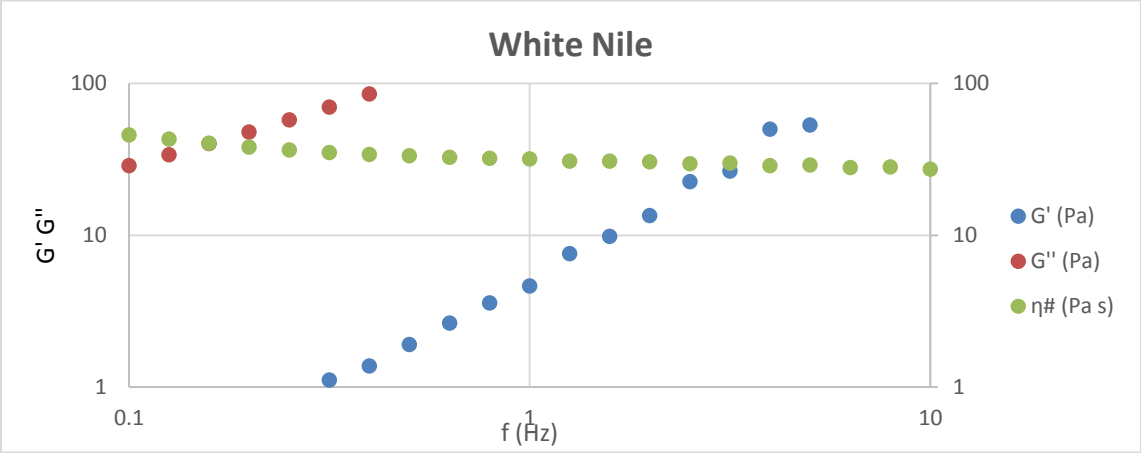
Sample 45261 Kordofan

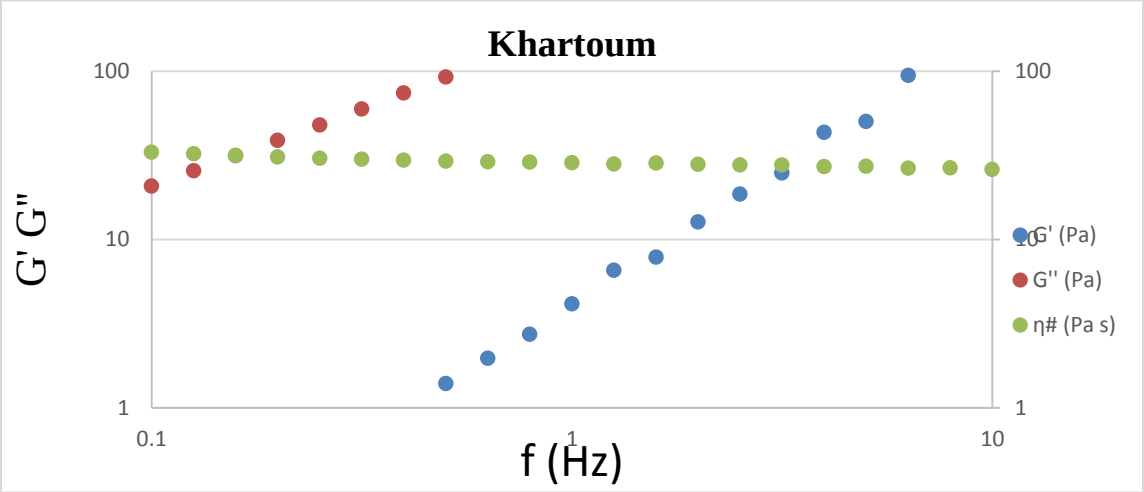
Sample 45262 Khartoum

3.15.2Dynamic rheological behavior

Viscoelastic properties of Azad *Azadirachta indica* gum., solutions at 50% concentrations were determined using oscillatory testing. Mechanical spectra of solutions revealed a typical liquid-like behavior Figures (3.6). The loss modulus (G″) of all samples of *Azadirachta indica* gum, was higher than the storage modulus (G′), the energy used to deform the material is dissipated viscously and the samples exhibits liquid-like behavior. The modules showed less frequency dependence at lower frequency range and relatively higher frequency dependence at higher frequency range.





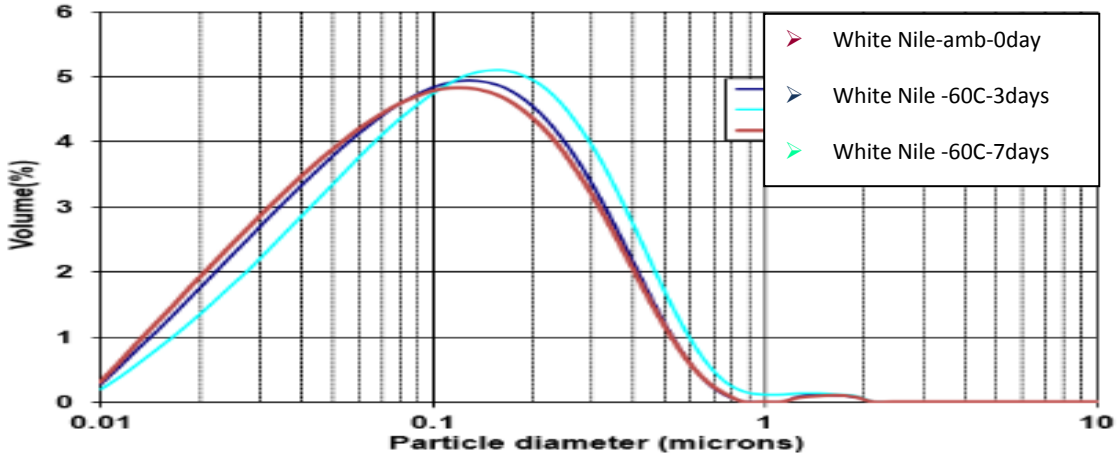


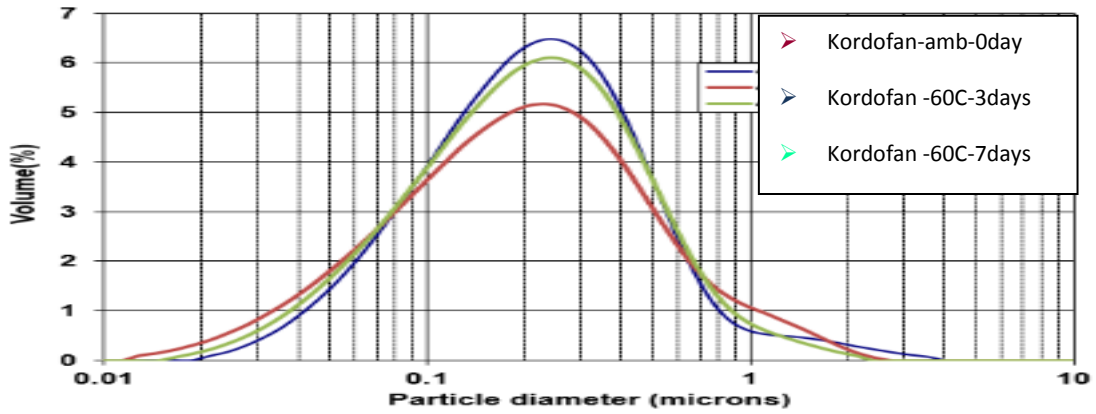
Figs 3.7 The effects of frequency on G' and G''

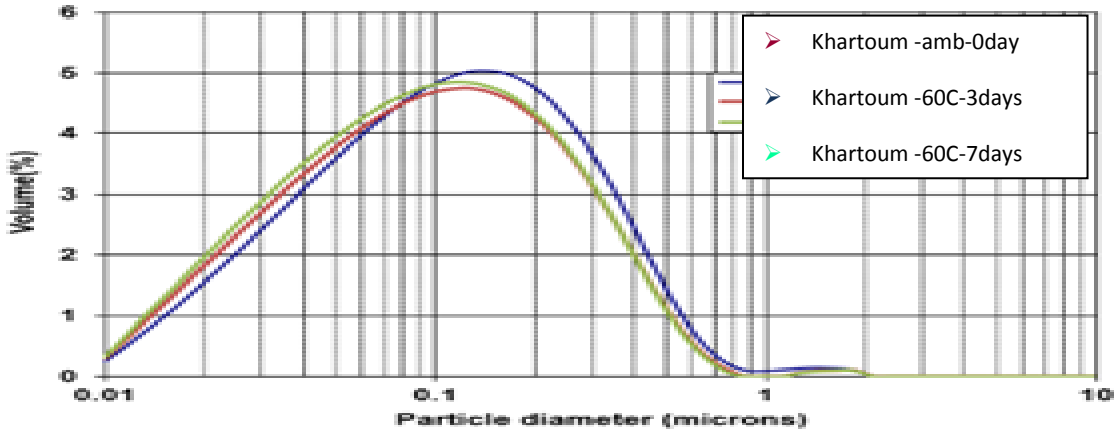
3.16 Emulsification properties

Figure3.8 shows that all of the *Azadirachta indica* gum samples produces very broad peaks with droplet diameters extended from 0.01 to 1 microns. As can be seen from the Figure, the curves of the zero day and the accelerated conditions at 60C for three and seven days were coincided which indicates almost identical stabilities of the emulsions. One possible reason for this could be due to *Azadirachta indica* gum has higher molecular weight in general and a higher molecular weight of AGP and great amount of nitrogen content .All these parameters leads to better emulsification characteristics.

The short term stability on emulsion may be due to the AGP fraction decrease in the first peak Fig 5.1 (c), it established that there is no direct relationship between the total proportion of the gum and emulsification stability. And the stability of the emulsion is direct relationship to AGP.







Figs. 3.8 Emulsion particle size of *Azadirachta indica* gum

3.16.1Span%

Figs3.8shows (span %) for the fresh emulsion and after incubation for 3 and 7 days at 60^{o}C respectively. The study shows that neem gum is a first- grade emulsion.

Table 3.10 Emulsification characters of neem gum Khartoum sample

Sample Name	As Prepared	After 3 days	After 7 days
D [3,2]	0.0665	0.0766	0.0638
D [4,3]	0.154	0.181	0.151
Span	2.869	2.721	2.9736
DX(10)	0.028	0.0324	0.0414
DX(20)	0.0436	0.05177	0.104
DX(50)	0.108	0.128	0.104
DX(80)	0.241	0.278	0.236
DX(90)	0.338	0.232	0.335
Grad	1	1	1

Table 3.11 Emulsification characters of neem gum White Nile sample

Sample Name	As Prepared	After 3 days	After 7 days
D [3,2]	0.163	0.142	0.151
D [4,3]	0.32	11.4	0.674
Span	2.239	4.782	2.421
DX(10)	0.0781	0.0618	0.0699
DX(20)	0.114	0.0981	0.105
DX(50)	0.23	0.232	0.224
DX(80)	0.432	0.552	0.438
DX(90)	0.592	1.7	0.612
Grad	1	1	1

Table 3.12 Emulsification characters of neem gum Kordofan sample

Sample Name	As Prepared	After 3 days	After 7 days
D [3,2]	0.0119	0.0664	0.0029
D [4,3]	0.17	3.13	0.148
Span	2.53	3.228	2.962
DX(10)	0.0304	0.0275	0.0263
DX(20)	0.0479	0.0429	0.0407
DX(50)	0.118	0.108	0.101
DX(80)	0.26	0.254	0.23
DX(90)	0.365	0.378	0.357
Grad	1	1	1

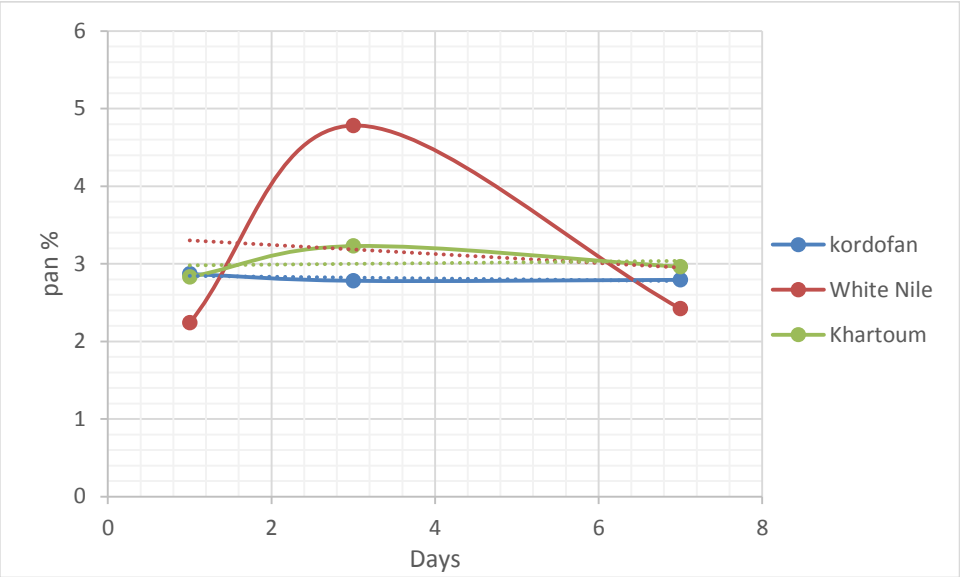


Fig. 3.9 The span% of *Neem* gum from three locations

Fig. 3.9 Show specific surface area (m^2/g) of cumulative droplet distributions of (Dx10, Dx50, and Dx.90). The results clearly showed that extreme changing was found in emulsions during the incubation for 3 and 7 days at 60^oC. Also the instability of the emulsion enhanced by decreasing of Dx 50 area to less than 6%, and increasing the droplet particles size with the diameter of more than one microns to more than 80%.

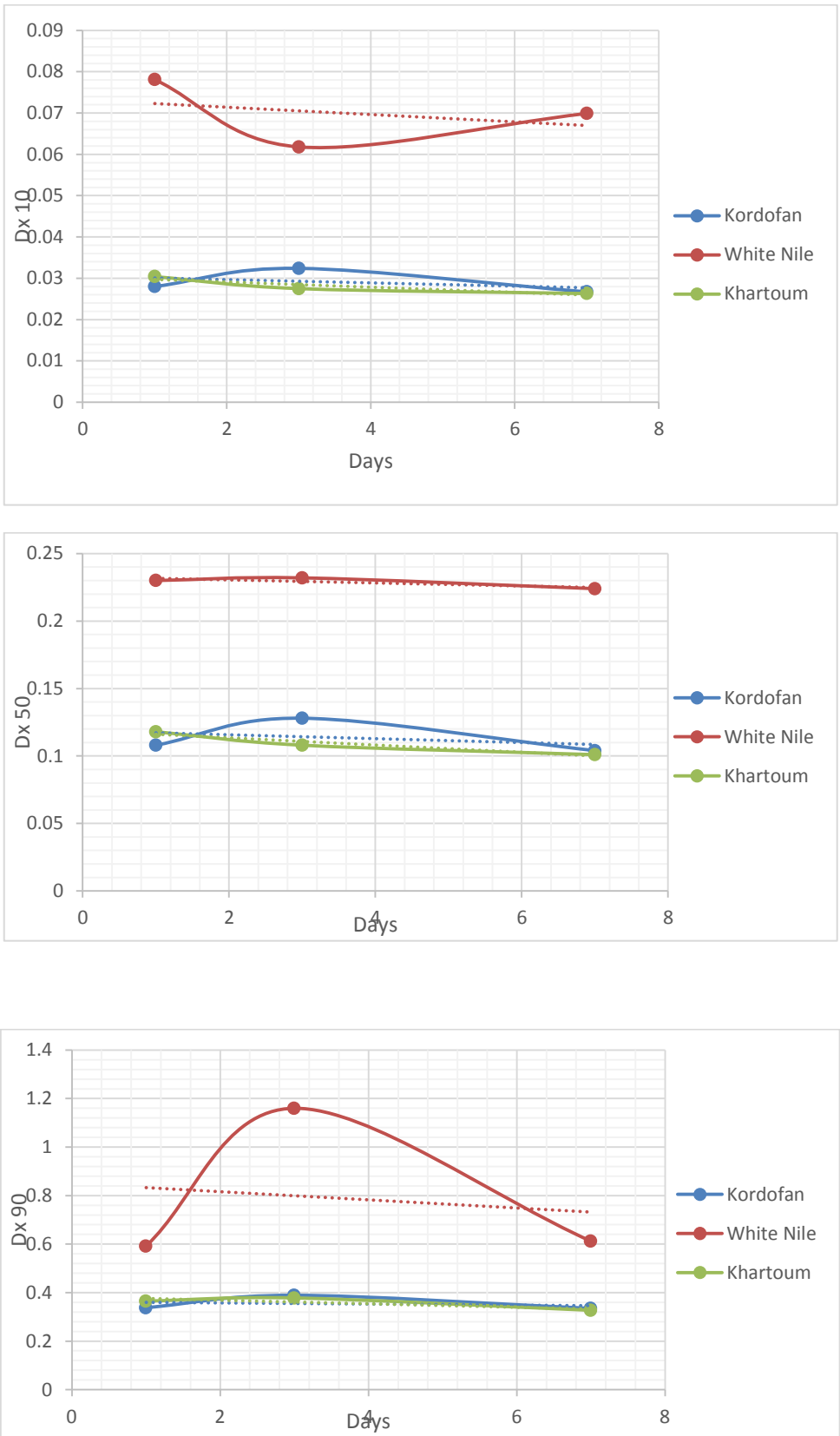


Fig. 3.10The Volume Weighted Mean Diameter of A Neem gum Emulsion for three location

Conclusion

- *Azadirachta indica* Gumshows physicochemical properties different from*Gummiferae* and *Volgaria.*
- *Azadirachta indica* gum contains the biggest amount of protein than all entire gums studied tell now. The molecular weight of *Azadirachta indica* Gum ranges between ($3.606x10^5$ and $4.00x10^5$ g/mole) with the radius of gyration (Rg). (25.6 - 66)The *Azadirachta indica* gum contains three fractions AGP, AG. and GP.
- The rheology of *Azadirachta indica* gum solution shows that the gum is simply a Newtonian fluid.
- The loss of modulus of *Azadirachta indica* Gumis higher than the storage modulus.
- *Azadirachta indica* Gumforms first grade highly stable emulsions.

Recommendation and further work

- Toxicology behavior of the gum needs to be investigated.
- Fractionation of the gum and investigation of the characteristics of the fractions are required.

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