Microflora Contamination of Gum Arabic

(*Acacia senegal* Gum) from Tree to Store

By

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DEDICATION

TO THE THREE CANDLES THAT
SHED LIGHT UPON MY WAY
WHEREVER I AM
PARENTS,
BROTHERS AND
SISTERS.

WITH ALL MY LOVE
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Before all, my special praise and thanks should be to Allah, the Almighty, most gracious and most merciful who gave the means, strength, and patience to conduct this research.

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ABSTRACT

This study comprises microbiological and physicochemical examination of 72 *Acacia senegal* gum samples from the seasons 2002/03 and 2003/04. The samples were randomly collected from North and West Kordofan areas, from fields, villages, markets and stores.

All samples were randomly tested for viable count of bacteria, moulds and yeasts and thermophilic spore-formers, as well as some pathogens such as *Staphylococcus aureus*, *Salmonella*, and contamination indicators such as coliforms and faecal coliforms (*E.coli*) for 36 samples.

Viable count of bacteria of gum samples after exudation ranged from $1.2 \times 10^3$ to $4.0 \times 10^4$ colony-forming units/gram (cfu/g) while those taken after collection in the field showed a range from $2.8 \times 10^3$ to $2.2 \times 10^4$ cfu/g, and those taken at village market ranged from $2.0 \times 10^3$ to $9.0 \times 10^9$ cfu/g, and those taken at crops market showed a range from $5.0 \times 10^2$ to $8.0 \times 10^3$ cfu/g. With samples of recent storage (three months) the count ranged from $3.1 \times 10^3$ to $8.0 \times 10^3$ cfu/g, and with samples of long storage (one year) the count ranged from $3.1 \times 10^3$ to $4.5 \times 10^3$ cfu/g.

Viable count of moulds of gum arabic samples (after exudation, collection, village market, crops market, recent storage and long storage) showed a range from $8.0 \times 10^2$ to $1.7 \times 10^4$, $1.0 \times 10^2$ to $2.0 \times 10^2$, $2.0 \times 10^2$ to $8.0 \times 10^2$, $2.0 \times 10^2$ to $2.0 \times 10^4$, $1.0 \times 10^2$ to $2.0 \times 10^3$ and $3.0 \times 10^2$ to $6.0 \times 10^2$ cfu/g, respectively.

Yeast were not detected in any sample of gum arabic.

Viable count of thermophilic spore-formers of gum arabic samples after exudation showed viable count ranging from $4.7 \times 10^2$ to $7.0 \times 10^3$ cfu/g while those taken after collection showed a range from
1.2×10^3 to 3.5×10^3 cfu/g. The samples at village market gave a range from 1.0×10^3 to 5.0×10^3 cfu/g but the samples at crops market showed a range from 7.0×10^2 to 6.5×10^3 cfu/g. The sample of recent storage gave a range from 2.3×10^3 to 5.0×10^4 cfu/g, while samples of long storage gave a range from 3.5×10^3 to 3.1×10^4 cfu/g.

Coliforms were not detected except in eight samples; two samples drawn after collection in the field showed a low value of 3.6 /g and a high value of 2.4×10^3 /g; one sample drawn at the village market showed a low value of 3.6 /g; four samples drawn at crops market showed a value of 9.3×10^2 /g, 4.3×10^2 /g, 2.0×10^3 /g and 3.6 /g; and one sample drawn at recent storage showed a high value of 2.1×10^2 /g. The ten isolates obtained were identified as *Klebsiella sp* (8 isolates) and *Moraxella sp* (2 isolates), which confirmed the absence of *E. coli*.

Staphylococci were not detected except in three samples (at village market, crops market and long storage). The suspected thirteen isolates obtained were identified as *Staphylococcus aureus* (8 isolates), *Streptococcus faecium* (4 isolates) and *Aerococcus viridians* (1 isolate).

The presence of *Salmonella* was suspected in all samples except in samples after exudation. The fifteen suspected isolates were identified as *Salmonella sp* (9 isolates), *Serratia sp* (3 isolates), *Pseudomonas sp* (2 isolates) and *Cedecea sp* (1 isolate).

**On the other hand twelve isolates of gram-positive, rod-shaped bacteria were isolated and identified as* Bacillus sp*, except that one isolate was found to be *Corynebacterium sp*.

Physico-chemical characterization of the gum arabic samples including moisture, ash and crude protein
CONTENT, PH, VISCOSITY, OPTICAL ROTATION AND MINERAL CONTENT WAS CARRIED OUT. THE SAMPLES AFTER EXUDATION, COLLECTION, AT VILLAGE MARKET, CROPS MARKET, RECENT STORAGE AND LONG STORAGE SHOWED THAT THE MEAN VALUE OF MOISTURE CONTENT WAS FOUND TO BE 23.6%, 22.97%, 16.7%, 15.5%, 13.1%, AND 11.6%; THE ASH CONTENT WAS 3.03%, 3.29%, 3.6%, 3.3%, 3.5% AND 3.6%; THE PH WAS 4.33, 4.33, 4.33, 4.37, 4.34 AND 4.35; THE PROTEIN CONTENT WAS 2.1%, 1.95%, 1.8%, 1.8%, 1.71% AND 1.74%; THE VISCOSITY WAS 52.6, 53.87, 50.6, 53.9, 50.6 AND 54.6 CENTIPOISES (CPS)/SEC; AND THE OPTICAL ROTATION WAS –25.8°, -25.0°, -27.2°, -28.0°, -29.2° AND -26.5°, RESPECTIVELY.

ALSO THE MEAN VALUE OF THE MINERAL COMPOSITION FOR THE GUM ARABIC SAMPLES WAS 0.49%, 1.4%, 0.24% AND 0.04% FOR CA⁺⁺, K⁺, Mg⁺⁺ AND Na⁺, RESPECTIVELY, IN THE EXUDATION SAMPLES; 0.85%, 1.39%, 0.31% AND 0.04%, RESPECTIVELY, IN THE COLLECTION IN FIELD SAMPLES; 0.78%, 1.46%, 0.24% AND 0.04%, RESPECTIVELY, IN THE VILLAGE MARKET SAMPLES; 0.65%, 1.59%, 0.29% AND 0.02%, RESPECTIVELY, IN THE CROPS MARKET SAMPLES; 0.63%, 1.28%, 0.27% AND 0.09%, RESPECTIVELY, IN THE RECENT STORAGE SAMPLES; AND 0.81%, 1.33%, 0.28% AND 0.05%, RESPECTIVELY, IN THE LONG STORAGE SAMPLES.
ملخص الأطروفة


تم تقدير العد الحي للبكتيريا والفطريات والخمائر والعدد الحي للبكتيريا المتجرثة المحبة درجة الحرارة العالية، بالإضافة إلى بعض الممرضات مثل بكتيريا المكورات العنقودية والسالمونيلا وبعض دلال الشمس مثل بكتيريا القولون والغانطية ( أشيرشا كولاي ) لـ36 عينة.

تراوح العد الحي للبكتيريا لعينات الصمغ بعد النضج مباشرة بين 1.2×10³ و 4.0×10⁴ وحدة مكونة للمستعمرة /جرام (م/جم). في حين أن العينات التي أخذت بعد الجمع في الحقل أعطت مدى تراوح بين 2.8×10³ و 2.2×10⁴ م/جم، والعينات من سوق القرية أعطت مدى تراوح بين 2.0×10³ و 9.0×10⁴ م/جم وعينات سوق المحصورات أعطت مدى 5.0×10² و 8.0×10³ م/جم، بينما أعطت العينات المخزنة حديثا تمتد (مدة ثلاثة أشهر) مدى بين 3.1×10³ و 8.0×10³ م/جم، والعينات المخزنة لمدة عام أعطت مدى بين 3.1×10³ و 4.5×10³ م/جم.

أعداد الفطريات لعينات الصمغ العربي بعد النضج مباشرة وبعد الجمع في الحقل، وسوق القرية، وسوق المحصورات، والمخازن الحديث والمخازن لمدة عام أعطت مدى تراوح بين 2.0×10³ و 8.0×10³ م/جم، و 1.0×10⁴ و 2.0×10⁴ م/جم، ومدى تراوح بين 2.0×10² و 8.0×10² م/جم، و 2.0×10³ و 2.0×10⁴ م/جم، ومدى 3.0×10³ و 6.0×10³ م/جم، و 3.0×10³ و 6.0×10³ م/جم، على التوالي.

لم يتم تبادل الخمائر في جميع العينات المفحوسة.

تراوح العد الحي للبكتيريا المتجرثة المحبة للحرارة العالية لعينات الصمغ بعد النضج مباشرة بين 4.7×10² و 7.0×10³ م/جم، وللعينات التي أخذت بعد الجمع في الحقل بين 1.2×10³ و 3.5×10³ م/جم. أما عينات سوق القرية فاعطت ما بين 5.0×10³ و 1.0×10⁴ م/جم، بينما عينات سوق المحصورات أعطت مدى بين 1.0×10³ و 1.0×10⁴ م/جم.
لم يتم تبين بكتيريا القولون إلا في ثماني عينات، اثنتان منها أثناء الجمع في الحقل، وعُدّت قيمة 3.6/جم، و 2.4×10³/جم، وواحدة في سوق القرية وعُدّت قيمة 9.3×10⁴/جم، وربعة في سوق المحصولات وعُدّت القيم الآتية 2.0×10⁴/جم، 2.3×10³/جم، 3.6/جم، و 3.1/جم، وواحدة في التخزين الحديث وعُدّت قيمة 2.1×10²/جم، و اثنتان. Klebsiella sp كانت واربعة منها كانت Staphylococcus aureus، وواحدة فقط كانت Moraxella sp.

وتبين عدم وجود بكتيريا المكورات العنقودية المرضية في كل من عينات النضج، وبعد الجمع والتخزين الحديث، بينما وجدت في عينات سوق القرية وسوق المحصولات والتخزين لمدة عام، حيث تم تعين ثلاث عشرة عزلة، ثماني منها Streptococcus faecium واربعة منها كانت Salmonella sp وواحدة فقط كانت Cedecea sp، واثنتان منها Pseudomonas sp، واثنتان منها Serratia sp.

وفي جانب آخر من خلال عدد اثنتي عشرة عزلة من البكتيريا العصبية الموجبة لصبغة جرام وتبين أن اغلبها كان Bacillus sp وواحدة فقط كانت Corynebacterium sp.

تم إجراء عدة اختبارات كيميائية لعينات الصمغ العربي تضمنت نسبة الرطوبة، الرماد، البروتينات، الأس الهيدروجيني، الزوجة، الدوران النوعي، والعناصر المعدنية للرماد.

وقد أوضحت نتائج التحليل للعينات بعد المضح مباشرة، وبعد الجمع في الحقل، وفي سوق القرية، وسوق المحصولات، والتخزين الحديث، والتخزين لمدة عام أن متوسط الرطوبة يبلغ 23.6%, 22.97%, 16.7%, 15.5%, 13.1% و 11.6% و 11.6%.
مهام الرطوبة: 3.03%, 3.29%, 3.6%, 3.3%, 3.5% و 3.6%; وينبغي متوسط
الأكس الهيدروجيني: 4.33%, 4.33%, 4.37%, 4.35%, 4.30% و 4.35%; ومتوسط البروتين الخام
2.1%, 1.95%, 1.8%, 1.71% و 1.74%; ومتوسط اللزوجة: 52.6, 53.87, 50.6, 53.9, 50.6 و 54.6 (cps، centipoises/sec)
الذبائح: 10.85%، 10.82%، 10.81%، 10.80% و 10.85%.

التوصيات المعملية أثبّت أن متوسط محتوى العناصر المعدنية للرماد لعينات
الصمغ العربي بعد النضج مباشرة كان 0.49%, 1.4%, 0.24% و 0.04% لكل من
الكالسيوم، البوتاسيوم، المغنيسيوم والصوديوم. على التوالي: 1.39%,
0.31% و 0.04% على التوالي لعينات بعد الجمع في الحقل; و 0.78%, 1.46%,
0.24% و 0.04% على التوالي لعينات سوق القرية; و 0.65%, 0.59%, 0.29% و
0.02% على التوالي لعينات سوق المحصولات.; و 0.63%, 0.28%, 0.27% و
0.09% على التوالي لعينات التخزين الحديث.; و 0.81%, 1.33%, 0.28% و
0.05% على التوالي لعينات التخزين لمدة عام.
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CHAPTER ONE

2. INTRODUCTION

Sudan is a large country, extending over many climatic regions and different environments. It is considered the first country in gum arabic production, as it produces annually more than 80% of the total world production (Taha, 1999).

Gum arabic is defined as the dried exudate, obtained from the stems and branches of *Acacia senegal* (L.) Willd (called in Sudan, hashab) or related species of *Acacia* (family: leguminoseae), which is extensively found in the so-called gum arabic belt. As one of the main export crops from the Sudan, gum arabic plays an important role in the international market.

The process of gum arabic production in Sudan is traditional and classical. The gum production channels from the stages of tapping and collection through marketing up to storage provide good chances for contamination with microorganisms that are found mainly in the surrounding environment. To insure unique identity of gum arabic and to protect it from competition from other substitutes, standardization measures are critically needed. One of the major problems facing gum arabic production in the Sudan is the contamination with microorganisms. The gum arabic contains basically carbohydrates (complex polysaccharides of high molecular weight), acids, salts and many other elements (Leung, 1980; and Evans, 1989).

Immediately after gum picking, different microorganisms can decompose the carbohydrates found as part of the gum. This process can lead to changes in the chemical structure of gum (Yousif et al., 1987).
THE PROCESSING OF RAW GUM ARABIC INTO POWDER IMPLIES THAT GUM SHOULD FIRST BE DISSOLVED IN WATER BEFORE DRYING. FERMENTATION OF THE CARBOHYDRATES WILL TAKE PLACE DURING THE DISSOLUTION PERIOD UNLESS THE PROCESSES ARE DONE FAST ENOUGH. THIS INDICATES THAT GUM ARABIC IS HIGHLY SUSCEPTIBLE TO MICROBIAL CONTAMINATION AND SPOILAGE (ALAMIN AND HASSAN, 1995).

Abdala (1988) revealed contamination with great numbers of Penicillium, Aspergillus and Rhizopus species in gum arabic produced from trees of different ages and the branches of the hashab trees were contaminated with aflatoxin which is caused by Aspergillus flavus. Gum arabic trade has advanced dramatically during the past 30 years with the advent of processed gums. Cleaning and sorting are expensive processes, and result in decreasing the earnings of gum arabic for the Sudan. Hence, the spray-drying technique has been developed in Sudan Industrial Institute since 1969 (Elmubarak et al., 1970).

In order to give improved functional properties of gum and to meet specifications required by gum manufacturers, by ensuring a constant product quality, spray-drying technique of gum arabic was applied in Babanousa dry milk factory in 1969. Unfortunately, this trial stopped partially because of unacceptable microbiological quality and complaints from international customers. For example, Agroproducts Company in Switzerland showed that total microbial count was 16000/g whereas the acceptable number was only 1000/g (Dirar, 1976b).

In this respect, gum arabic has not received any special technical standardization research work regarding microbial contamination (Alamin and Hassan, 1995).
The physico-chemical and biological properties of gum arabic are very closely correlated with the effect of microorganism. Generally, microorganisms reduce the quality and structure of gum arabic. Therefore, microbiological standardization is internationally important for production of good quality gum arabic and acceptability by customers in the international market.

Justification of the study

**GUM ARABIC IS VERY IMPORTANT FOR HUMAN BEINGS. IT IS INVOLVED IN DIFFERENT BASIC USES, E.G., IN FOOD AND PHARMACEUTICAL INDUSTRIES. GUM ARABIC HAS NEITHER A SUITABLE SUBSTITUTE NOR CLEAR STANDARDS AS FAR AS THE MICROBIAL CONTAMINATION IS CONCERNED. THIS STUDY WILL FOCUS ON THE COUNTS AND THE NATURE OF MICROORGANISMS ASSOCIATED WITH GUM ARABIC IN ORDER TO HELP BRIDGE THE GAP OF OUR KNOWLEDGE IN THIS AREA.**

Objectives of the study

The main objectives of this study were: -

1- To count, isolate and identify different species of microorganisms that contaminate gum arabic immediately after exudation of gum arabic nodules, after collection in the fields, after storage in the villages, after transportation to the crops markets and at stores (recent storage and storage for one year).

2- To study the physico-chemical properties of gum arabic.
CHAPTER TWO

3. LITERATURE REVIEW

2.1 The *Acacia senegal* tree

The name *Acacia* was first given to the genus by Miller (1754). A large number of species of *Acacia* (family leguminoseae) yield gum in commercial quantities (Ghosh and Purkayastha, 1962). In the Sudan, the majority of the gum produced is from *Acacia senegal* (L.) Willd tree (Abdel Gadir, 2002) (Plate 2.1).
Plate 2.1: *Acacia senegal* tree with gum nodules

A- Classification

*Acacia senegal* (L.) Willd is classified in family Leguminoseae, subfamily Mimosoideae, genus *Acacia*, subgenus *Aculeiferum* (Vassal), series vulgares (Bentham), species *senegal* ( Sahni, 1968).

Scientific name is *Acacia senegal* (L.) Willd. English name is gum arabic tree ( Vogt, 1995). Sudanese name is hashab tree ( Abdel Gadir, 2002).

*Acacia senegal* tree is a deciduous small tree or shrub 2.8 m in height and 0.25 m in diameter of the stem, with umbrella-shaped crown, very branchy with many upright twigs and wider spreading in the upright parts. The bark is light gray to light brown, smooth on young but very scaly on older trees. The tree is grown on sandy and clay plains in short grass savanna forming a continuous belt from east to west central Sudan.

The tree is more common in the western sand plains of Darfur and Kordofan as pure stands or associated with *Acacia mellifera* ( Maydell, 1990).

The flowering period of the *Acacia senegal* tree is usually from November to February and the fruiting period is from January to April.
GUM TAPPING USUALLY STARTS AT THE END OF THE RAINFALL SEASON IN NOVEMBER AND GUM COLLECTION CONTINUES UP TO MAY OR JUNE.

B- SIGNIFICANCE

*ACACIA SENEGAL* IS A MULTIPURPOSE TREE; BESIDE THE ECONOMIC IMPORTANCE OF IT IN GUM PRODUCTION, IT HAS MANY AND VARIED USES; ITS WHITE HARD WOOD MAY BE USED AS A LOCAL BUILDING MATERIAL, AS FUEL WOOD AND AS CHARCOAL. THE WOOD ALSO YIELDS POTASH, WHICH IS USED SOMETIMES AS A SUBSTITUTE FOR SALT.


2.2 Gum arabic belt
Gum arabic is produced in the so-called gum arabic belt of sub-Saharan Africa (FAO, 1995). The gum arabic belt occurs as a broad band from Mauritania, Senegal and Mali in the West, through Burkina Faso, Benin, Niger and Northern parts of Nigeria, Cameroon and Chad, northern Central Africa Republic to Sudan, Eritrea, Ethiopia and Somalia in the horn of Africa. It extends through East Africa to Southern Africa covering southern Angola, Namibia, Botswana, Zimbabwe, South Africa and parts of Mozambique (Taha, 1999).

In the Sudan the term gum belt is applied to that part of the country in which various types of gum are produced, the most important one being the gum produced by *Acacia senegal* “hashab”. The gum belt is the Sahelian zone south of the desert stretching from Chad in the west to the Ethiopian border in the east, approximately between latitudes 11 and 15° north. More precisely the location of the gum belt corresponds to the natural distribution of *Acacia senegal* which primarily depends on climatic conditions. In administrative terms in the Sudan the gum belt covers parts of North Kordofan, South Kordofan, West Darfur, Kassala, Gadaref, Sinnar, Blue Nile and Upper Nile states (Karamalla, 1999).

The most important region for gum arabic production in the Sudan is Kordofan, but the gum-yielding *Acacia* trees grow along a 3000 Km-wide band following the southern frontier of the Sudan desert from West Africa to the Middle East, and in India (FAO, 1995) (Fig. 2.1).
**A-Climate**

There is large variation in climate in the Sudan, from tropical forest in the south with an average annual rainfall of 800 mm and an extensive savannah in the north. Rainfall is most important climatic factor in the gum belt. Combined with soil condition, average annual rainfall determines the distribution of *Acacia senegal* and thus the location of the gum belt. In the west annual rainfall ranges from 250 to 600 mm and in the east the annual rainfall ranges from 450 to 1000 mm. The rainy season extends from June to September. The total annual rains are very variable and unreliable. The yearly variation has different effects on various locations in the gum belt. However, the gum belt is prone to drought that affects gum production (MNP, 1980).

**B-Soil**
Basically, the gum belt comprises two kinds of soil, sandy soil in the west (goz in North and South Kordofan and Southern Darfur and patches of White Nile states), and clay soil in the east (in Kassala and Blue Nile states). However in the west, clay soils may be found either pure or mixed with sand (gardude in Southern Darfur and South Kordofan) (Karamalla, 1999).

2.3 Definition of gum arabic

Gum arabic is defined as the polysaccharide obtained as exudate of the *Acacia* tree (Whistler, 1973). It consists of six carbohydrate residues i.e. D-glucose, L-arabinopyranose, L-arabinofuranose, L-rhamnose, D-glucuronic acid and 4-0-methyl glucuronic acid, and as their Ca, K, Mg and Na salts (Ismail, 2000).

*It can also be defined in terms of its functionality; as a natural, nontoxic, multimolecular, glucoproteinaceous, negatively optically active, anionic hydrocolloid with very unique rheological properties (Idris, 1989).*

The Joint FAO/WHO Expert Committee on Food Additives (JECFA, 1990) defined gum arabic as a “dried exudate obtained from the branches and stems of *Acacia Senegal* (L.) or closely related species”. It consists mainly of high molecular weight polysaccharides and their calcium, magnesium and potassium salts which on hydrolysis yield, arabinose, galactose, rhamnose and glucuronic acid (Ismail, 2000).

The gums collected from the same species of tree are chemically identical with respect to some components in
THEM WHILE GUMS FROM DIFFERENT SPECIES DIFFER CHEMICALLY AND PHYSICALLY (Mantell, 1965).

Gum arabic can better be defined as an arabinogalactan protein complex (instead of polysaccharide) based on the findings of Akiyama et al. (1984), and Lamport et al. (1991) who demonstrated that the nitrogen in Acacia senegal gum is structurally significant and that Acacia senegal gum is a kind of glycoprotein with the carbohydrate bound covalently to the protein part of the gum via the amino acids hydroxyproline and serine (Ismail, 2000).

2.4 Theories of gum formation and function

It is still a matter for debate whether gum exudates are formed at the site of a wound or whether they are generated elsewhere in the tree and then transported to the site of exudation.

Many theories concerning the origin and formation of plant gums have been advanced (Mantell, 1965; GAC, 1991). These include that gum:

I- Is a product of normal plant metabolism.

II- Arises from a pathological condition of the tree.

III- Arises from some infection or invasion by microorganisms, which may be fungal or bacterial in nature (Smith and Montgomery, 1959).

However, some trees produce gum while in apparently healthy condition and since the composition, properties and structure of Acacia
*senegal* gum appear to be independent of the nature of the wound or stimulus inducing gum production, it does not seem necessary to propose that bacteria or other microorganisms are obligatorily involved in gum formation. It seems likely that the gum is the product of a normal metabolic process which in influenced by environmental conditions such as water stress, temperature and infectious agents (Smith and Montgomery, 1959; Whistler, 1973).

2.5 Gum arabic tapping, collection and handling

Natural exudation of gum arabic commonly occurs due to damage caused by animal, insects, or by sand particles carried by wind (Hall, 1961). Tapping by man, is traditionally done by a tapping tool (axes or “soonky”) (Ballal, 1991).

Tapping generally starts when the trees are 3 (coppice origin) to 6 or 7 (seed origin) years old with a height of about 1.2 to 3.7 m and a stem diameter of 5 cm. However, trees from seedlings origin are tapped at the age of 5 years. The trees can be tapped annually up to the age of 18 years (Dale and Greenway, 1961; Weber and Stoney, 1986). (Plate 2.2).

After tapping the tree, the first harvest of gum nodules is picked after 3 –6 weeks as stated by Awouda (1973) or 30–45 days as stated by Obied and Seif Eldin (1970).

Gum continues to exude from the same injured spot, and is collected every 10–15 days, and the same tree is picked 4–6 times during the season and a tree can be tapped five times during its life time (Ballal, 2002). (Plate 2.3).
PLATE 2.2: TAPPING OF *ACACIA SENEGAL* TREE USING “SOONKY”
PLATE 2.3: COLLECTION OF GUM ARABIC (ACACIA SENEGAL GUM)
2.6 Production of gum arabic

Sudan is the major producer of gum arabic and supplies about 85% of the annual world requirements of gum arabic. This accounts for 11% of the exports value of the Sudan (GAC, 1991).

Kordofan and Darfur states in the Sudan are the biggest producers and exporters of the best qualities of gum arabic which is also known as Kordofan gum (Awouda, 1974b).

NAS (1979) and GRD (1965-1986) reported that the average yield of gum arabic is 250g per tree per year with variability that ranges from 0 to 6.6 Kg.

Total production of gum arabic in Sudan from season 1969/70 to 2002/03, was found to be 804 thousand tons, about 87.19% of which was produced from Acacia senegal (hashab). Hashab gum has shown a uniformity of high quality and the presence of the trees in pure stands results in ease of collection. Acacia seyal (talha) represents about 12.81% of total production. Therefore Acacia senegal is dominating gum production in Sudan (GAC, 2004).

To follow the trend of gum production (Fig 2.2), about 35 thousand tons were produced in season 1969/70. The quantity declined to about 23 thousand tons due to drought in season 1973/74. Then the production fluctuated till season 1984/85, in which production fell to 11 thousand tons as a result of severe drought (GAC, 2004).

In 1990/91 and 1991/92 production fell to about 11 and 7 thousand tons, respectively. This was due to environmental and biological factors such as lack of rainfall and invasion by locusts. Then
production increased to about 39 thousand tons in 1994/95. This was attributed to high prices in the local market (3500SD/Kontar).
Fig 2.1 Gum arabic production in Sudan (1969-2003)
Unfortunately this was followed by a reduction to about 3 thousand tons in 1999/00 in response to low prices (GAC, 2004).

However, the production substantially increased during the period 2001–2003 as the consequence of the rapid escalation of prices. Therefore, gum production is greatly affected by environmental and biological problems as well as by pricing policy (GAC, 2004).

2.7 Gum arabic

2.7.1 Specifications of gum arabic


The European Economic Community (EEC) conforms to the definition of gum arabic as dried gummy exudates from stems and branches of *Acacia senegal* (L.) Willd or related species of *Acacia* (EEC, 1978).

Commercial specifications also included viscosity, emulsifying property and the moisture content (not more than 15%), ash content (not more than 6%), acid-insoluble ash (not more than 0.5%) and acid-insoluble matter (not more than 1%). Also two microbiological parameters were included: *Salmonella* and *Escherichia coli* should be absent/1g (FAO, 1986).

In 1990 the above specifications were revised again (FAO, 1990) adding three aspects to the superseded, which are:
(i) Insertion of the words “closely related species to Acacia” for further distinction between the gum exudates from Acacia senegal and other commercially inferior Acacia gum.

(ii) Specific rotation was specified by the lower limit –34° and the upper limit -26°.

(iii) Nitrogen content was also specified by lower and upper limits: 0.27% and 0.39%, respectively.

Specifications were revised again in 1995 excluding the most important distinguishing parameters such as specific rotation and nitrogen content (JECFA, 1995).

In 1997 specifications were revised once more with some amendments to the superseded one (JECFA, 1997):

(i) The word “closely” was replaced by Acacia seyal closely related species.

(ii) Specific rotation was re-included and expressed as laevorotatory, and dextrorotatory for Acacia senegal and Acacia seyal, respectively.

Very recently, specifications were revised and the word “closely” was excluded and replaced by the expression “or Acacia seyal”. Gums from other Acacia species are not included in these specifications (JECFA, 1999).

2.7.2 Description of gum arabic

Gum arabic from Acacia senegal is a pale white to orange-brown solid, which breaks with a glassy fracture. The best grades are in the form of whole, spheroidal tears of varying sizes with a matt surface texture. When ground, the pieces are paler and have a glass appearance. Gum arabic is also available commercially in the form of white or yellowish-white flakes, granules, powder, roller dried or spray-dried
material. An aqueous solution of 1g in 2ml water flows readily, is acid to litmus and is insoluble in ethanol (JECFA, 1999).

2.8 Gum arabic grading, processing and marketing

2.8.1 Gum arabic grading

Currently gum derived from *Acacia senegal* is sold in different grades. Kordofan gum from *Acacia senegal* is considered the best gum type.

The commercial grades of gum arabic are as follows:

A. Handpicked selected (HPS)

It is considered as the best grade comprising the cleanest and brightest gum with the largest particle size with smooth surface texture.

B. Cleaned and sifted

A grade just below handpicked selected with relatively small particle size.

C. Cleaned

Smaller particle size with some impurities.

D. Siftings

Small fragments with some impurities.

E. Gum dust

It is the residue of the operations of gum sorting and is considered as the poorest with a high percentage of dust and impurities. It is used locally and not exported (Karamalla, 1999).

2.8.2 Gum arabic processing
AFTER THE INITIAL CLEANING FROM IMPURITIES, GUM ARABIC MAY BE FURTHER PROCESSED INTO THE FORMS NEEDED FOR INDUSTRIAL USE. THE PROCESSING INCLUDES:

(a) **Kibbling**

*Making uniform pebble size pieces, in which mechanically or manually cleaned gum is simply crushed through kibbler or hammer mill and then sifted into a certain size required by customers. Size of kibbled gum ranged from 0.5 to 8 mm.*

(b) **Powdering**

*Milled gum arabic in which the oversize and undersize particles of gum arabic are reduced to powder with 100 –150μ particle size. It is called milled product.*

(c) **Spray-drying**

Spray-drying entails creating a concentrated gum solution, filtering out the impurities and forcing the heated solution through a fine nozzle into hot air, creating fine, highly uniform, very pure and clean spherical particles of gum (Karamalla, 1999).

2.8.3 *Gum arabic marketing*

**The production and trade of gum arabic in modern times have been dominated by Sudan. Levels of export from Sudan are therefore good indicators of consumption.**

Towards the end of the 1960s total gum arabic production (hashab and talha) was in excess of 60 thousand tons/year. Considering supplies of gum arabic from other countries, total world consumption was around 70 thousand tons/year.
Drought in 1970s, and 1980s, led to fluctuations in both the supply and price of gum arabic and as a consequence to changes in demand. The severe Sahelian drought of 1973/74 resulted in a world shortage of gum arabic and high prices which in turn, accelerated the replacement of gum arabic by substitutes such as modified starches.

A low point of approximately 20 thousand tons of Sudanese exports was reached in 1975s, which recovered to around 40 thousand tons during 1979.

A further drought, 1982 –1984, combined with political and civil-unrest, caused a fall of exports to below 20 thousands in mid/late 1980s and early/late 1990s.

Demand for gum arabic has therefore been constrained at times by supply and under these circumstances end users who switch to alternatives do not always revert to gum arabic when supply problems are solved.

It is unlikely, therefore that world market for gum arabic will reach the heights that they once had, although the superior properties of gum arabic (especially high quality material from *Acacia senegal*) will ensure that it retains substantial markets if availability is assured and prices are favorable (FAO, 2002).

2.9 Gum arabic in human diet

Because gum arabic is a complex proteinaceous, water-soluble polysaccharide, resistant to human gut enzymes, it can be described as dietary fiber. Gum arabic has been awarded by the European Economic Community (EEC) the designation E414 as a safe food additive. Gum arabic does not cause any health trouble as it is degraded extensively in the human colon and it is completely decomposed to volatile fatty acid
by the natural anaerobic micro-flora in the large bowel. Ross et al. (1983), Titgemeyer et al. (1991) and Wyatt et al. (1986) reported that, gum arabic fermenters, species of Bacteroides and Bifidobacterium, are able to utilize gum arabic as a source of carbohydrate. Gum arabic has a digestible energy value of 3.5 Kcal/gram i.e. 84% of the value for starch. Independent studies have confirmed this relatively high value (Anderson, 1991).

Although gum arabic has been considered as GRAS (Generally Recognized As Safe) since 1961, recent extensive toxicological studies have again confirmed its safety (FDA, 1973; Anderson et al. 1982; Ross et al. 1983).

In 1982 the American National Institute of Health (ANIH, 1982) published extensive data resulting from 103 weeks bioassay studies of gum arabic feeding experiments on rats and mice. The conclusion was that the ingestion of gum arabic over the normal life span of rats and mice, of either sex (male-female), did not result in carcinogenesis or any other compound-related clinical signs or effects on survival.

Bliss et al. (1996) reported that an increase in nitrogen metabolites concentration in the plasma caused by chronic renal failure can be reduced by using gum arabic as a dietary supplement aimed at increasing the excretion of nitrogen waste via feces. The bacteria that inhabit the human digestive tract grow by fermenting dietary fiber. They absorb nitrogen and degrade urea to ammonia then excrete it in the feces taking some of the body’s nitrogen waste with it. As a result, patients who find themselves rapidly losing weight on such a diet might be able to eat slightly more protein if they consume gum arabic fiber.

2.10 Uses and applications of gum arabic
THE USE OF GUM ARABIC DATES BACK TO ABOUT 5000 YEARS TO THE TIME OF THE ANCIENT EGYPTIANS, AND IT IS THE OLDEST AND BEST KNOWN OF ALL NATURAL GUMS. GUM ARABIC HAS A WIDE RANGE OF USES AS FOOD ADDITIVE, IN PHARMACEUTICALS, MEDICINAL USES AND COSMETICS. IT ALSO HAS MANY INDUSTRIAL APPLICATIONS SUCH AS IN TEXTILE, LITHOGRAPHY, POLISHES, PAINT, INKS AND PIGMENTS, CARBONLESS PAPER, CRAYONS AND CERAMICS (DUKE, 1983; FENNEMA, 1996).

2.10.1 Food industries

Gum arabic is used mainly as food additive, because of its desirable properties such as being colorless, odorless, tasteless and completely water-soluble and does not affect the flavor, odor or color of the food to which it is added. Gum arabic acts as emulsifying agent, flavor, fragrance, encapsulation and thickening agent, in addition to being nontoxic and a natural product.

It is widely used as a preferred emulsifier for citrus-containing flavor emulsions. The ability of gum to stabilize foam is still used in the manufacture of beers and soft drinks. It is also used to clarify wine and fix tannins.

Gum arabic is used extensively in the production of spray-dried flavors and fragrances. It is used to stabilize emulsions and when subsequently spray-dried, the gum prevents evaporation and oxidative deterioration of flavor, fragrance oil or absorption of water from the air.

Perhaps second only to its uses in flavor and beverage industries, gum arabic has found a widespread use in confectionery industry. It is used primarily to retard sugar crystallization and in emulsifying fat, and
it is used in a wide variety of confections. In high sugar pieces, the gum has been used in concentration from 10 to 45%.

As an emulsifier, gum arabic functions in keeping fat distributed uniformly throughout the piece and preventing the fat from migrating to the surface. This property makes gum useful in caramel and toffee confections.

Gum arabic is also used as a glaze and as a component in various chewing gums as a thickener and it contributes to softer chew. Also the gum provides a protective initial coat and acts as a plasticizer for subsequent sugar coating. Also caloric content for gum arabic has not been firmly established, it is an integral ingredient in reduced-sugar or non-sugar coated hard candy and gumdrops.

Gum arabic is widely used in the baking industry for its low water absorption properties. Gums are cold water-soluble and have impressive adhesive properties for use in glazes and toppings.

Gum is used in processed meat industry, where it provides good water retention and cohesive properties. It is used in sauces and dressings where it contributes a smooth, short texture, while adding a high degree of emulsion stability.

Gum arabic had widespread applications in diet or meal; replacing drinks, because of its low viscosity, bland flavor and high fiber content.

2.10.2 Pharmaceuticals industry

Gum arabic is listed in U.S “Pharmacopoeia” as an effective suspending agent and has been employed to suspend soluble drugs and to prevent the precipitation of metals from solutions through the formation of colloidal suspensions. Gum arabic is commonly employed as a demulcent in preparations designed to treat diarrhea, dysentery, coughs,
throat irritations and fevers. It serves as an emulsifying agent and gives viscosity to powder drugs materials; also it is used as a binding agent in making pills and tablets and particularly cough drops and lozenges. Also gum arabic is used as a film-forming agent in peel-off masks.

2.10.3 Medicinal uses

GUM ARABIC IS USED FOR THE TREATMENT OF LOW BLOOD PRESSURE CAUSED BY HEMORRHAGE OR SURGICAL SHOCK. IT IS ALSO USED INTERNALLY IN INFLAMMATION OF INTESTINAL MUCOSA AND EXTERNALLY TO COVER INFLAMED SURFACES, AS BURNS, SOME NIPPLE AND NODULAR LEPROSY. ALSO IT WAS SAID TO BE USED FOR CATARRH, Colds, Coughs and Dysentery (Duke and Wain, 1981).

GUM ARABIC IS USUALLY DISSOLVED IN WATER TO MAKE MUCILAGE, WHICH IS SWEETENED AND HAS BEEN USED TO TREAT THE EARLY STAGES OF TYPHOID FEVER AND MAKES A GOOD VEHICLE FOR OTHER MEDICINES.

RECENT SCIENTIFIC FINDINGS SHOWED THAT SUPPLEMENTATION OF THE DIET OF PATIENTS SUFFERING FROM CHRONIC RENAL FAILURE WITH GUM ARABIC FIBERS, RESULTED IN A SIGNIFICANT REDUCTION IN URINARY NITROGEN EXCRETION AND IN AN INCREASE OF FAECAL NITROGEN EXCRETION (Bliss et al. 1996).

SUDANESE PHYSICIANS CARRIED OUT A TRIAL ON TREATING SOME PATIENTS, WHO SUFFERED FROM CHRONIC RENAL FAILURE, WITH GUM ARABIC. ENCOURAGING RESULTS WERE OBTAINED SHOWING A RELATIVE REDUCTION IN SERUM CREATININE, SERUM PHOSPHOROUS
AND URIC ACID LEVEL WHILE SERUM CALCIUM WAS INCREASED (SULIEMAN ET AL. 1998).

2.10.4 COSMETICS INDUSTRY

GUM ARABIC IS USED TO INCREASE VISCOSITY, IMPART SPREADING PROPERTIES AND GIVE A PROTECTIVE COATING AND SMOOTH FEEL IN LOTIONS. IT IS ALSO USED AS A SUSPENDING AGENT AND VICTORIES IN HAND CREAMS.

2.10.5 INDUSTRIAL APPLICATION:

GUM ARABIC IS USED AS AN INGREDIENT IN MICRO-ENCAPSULATION, AND IN THE PRODUCTION OF CARBONLESS PAPER. IN TEXTILE, IT IS FOUND IN WATER PROOF EMULSIONS AND SIZING. GUM WORKS IS USED AS A BINDER FOR COLOR PIGMENT IN CRAYONS. THIS LIST OF USES GOES ON TO INK AND PIGMENT MANUFACTURE, CERAMICS, LIQUID INSECTICIDES, POLISHES AND PAINTS... ETC.

GUM ARABIC IS ALSO USED IN THE PREPARATION OF ETCHING AND PLATING SOLUTIONS IN LITHOGRAPHY INDUSTRY. THE GUM FUNCTIONS AS A SENSITIZER FOR PLATES, AS AN INGREDIENT IN FUNCTION SOLUTIONS, PLATE WASHING AND PROTECTIVE COATINGS FOR THE PLATES IN STORAGE.

2.11 SUBSTITUTE OF GUM ARABIC

AS MENTIONED ABOVE, GUM ARABIC HAS BEEN USED WIDELY INTO THREE MAIN AREAS, FOOD, PHARMACEUTICAL AND TECHNICAL,
which makes it difficult to substitute completely. The gum arabic is highly soluble in water and forms solutions over a wide range of concentrations without becoming highly viscous. The combination of high solubility in water and low viscosity confers on gum arabic its highly valued emulsifying, stabilizing, thickening and suspending properties. Moreover, gum arabic is a natural product and generally recognized as safe (GRAS) by American Food and Drug Administration (FDA, 1974).

Recently, factors, such as the droughts of 1971/1972 and 1984/1985, unstable prices of gum, lack of quality control and instability of supply, enhanced the appearance of gum arabic substitutes (Awouda, 1974b).

Now there is a growing competition from new generations of permitted modified starches as substitutes for gum arabic (Smith and Bill, 1986).

There are substitutes which have somewhat similar properties as gum arabic but they are cheaper. Recently it has been claimed that certain extra-cellular microbial products from bacteria, yeasts and algae, have an emulsification ability that is as good as, or even better than gum arabic (Shepherd et al. 1995).

Turbian (1995) reported that sodium starch octenyl succinates are unique specialty food starches characterized by excellent emulsion stabilizing properties. Starches and derivatives are the most important gum substitutes in confectionery industry, but the characteristic of being non-
CALORIFIC GIVES GUM ARABIC AN ADVANTAGE OVER OTHER CARBOHYDRATES.

Also gelatin obtained from a variety of animal sources and skins finds application in the food and pharmaceutical industries. However, gelatin is much more expensive than gum arabic. The inferior performance of many substitutes such as gum trangacanth, gum karaya, starches, arabinogalactans and some synthetic gums prevented them from retaining any major share of the traditional markets of gum arabic (Ali, 1996).

To maintain markets of gum arabic, a system of total quality management must be applied from field up to end user, so as to ensure homogeneity of quality and systematic supply (Ajmi and Arbish, 2000).

2.12 Microflora of gum arabic

Gum arabic has been an article of commerce since ancient times; being used as early as 3000 BC by the Egyptians for thickening paints. Today, gum arabic is one of the most profitable cash crops and has an increased importance for Sudan’s economy and because of its desirable properties, it has been used widely. Chemical, physical and agronomic studies on gum arabic started long ago, but very few researches have been done concerning the microbiological aspects, although gum arabic is involved in very highly sensitive industries such as food additive, pharmaceuticals
AND COSMETICS, WHICH REQUIRE STANDARD LEVELS OF MICROBIOLOGICAL CRITERIA.

SOME RESEARCHERS HAVE CLAIMED THAT GUM FORMATION (GUMMOSIS) IS A PATHOLOGICAL PROCESS RESULTING FROM A BACTERIAL OR FUNGAL INFECTION OF THE INJURED TREE. BLUNT (1926) AND MALLOY (1972) CLAIMED THAT GUMMOSIS IS ACCELERATED BY MICROBIAL INFECTION. THEY ALSO INDICATED THAT THEY HAD ISOLATED MICROORGANISMS, WHICH PLAYED CERTAIN VITAL ROLES IN GUM EXUDATION AND/OR FORMATION.

VOLLARD (1972) PROVIDED ANALYTICAL DATA, ON DIFFERENT SAMPLES OF GUM ARABIC (BOTH NODULES AND POWDERED FORMS). THE RESULTS OBTAINED SHOWED THAT GUMS ARE IN GENERAL, OF ACCEPTABLE MICROBIOLOGICAL QUALITY.

2.12.1 BACTERIAL LOAD OF GUM ARABIC

Khalid (1984) stated that the total viable count per 1g of gum ranged from $4.8 \times 10^5$ to $9.9 \times 10^6$cfu/g.

OSMAN (1998) AND KARAMALLA (1999) REPORTED THAT THE BACTERIAL LOAD FOR GUM ARABIC SAMPLES PICKED FROM TREES, LOCAL MARKET AND IN THE STORE RANGED BETWEEN $5.9 \times 10^3$ AND $1.3 \times 10^5$CFU/G, $2.9 \times 10^4$ AND $7.7 \times 10^5$CFU/G AND $1.7 \times 10^4$ AND $3.1 \times 10^6$CFU/G, RESPECTIVELY, IN ELOBIED AREA.

ISMAIL (2000) STATED THE TOTAL Viable COUNT OF CRUDE GUM ARABIC RANGED FROM $4.5 \times 10^4$ TO $8.4 \times 10^4$CFU/G.

Mohamed (2000) stated that bacterial load of gum arabic samples did not exceed $8.2 \times 10^3$cfu/g.
Recent study done by Obied (2002) reported that the total viable count of raw gum arabic samples ranged from $2.1 \times 10^2$ to $7.0 \times 10^2$ cfu/g.

2.12.2 mould and yeast counts

Khalid (1984) stated that total viable counts of fungi/g of gum ranged from $2.0 \times 10^3$ to $2.0 \times 10^5$ cfu/g. They included *Cladosporium sp*, *Aspergillus sp* and a species of the filamentous bacteria Actinomycetes (*Streptomyces*). Was also detected.

Osman (1998) and Karamalla (1999) reported that mould counts of gum arabic samples picked from trees, local market and in the stores ranged between $2.3 \times 10^3$ and $7.0 \times 10^3$ cfu/g, $3.1 \times 10^4$ and $4.2 \times 10^5$ cfu/g and $2.6 \times 10^4$ and $3.8 \times 10^5$ cfu/g, respectively, in Elobied area.

Ismail (2000) stated that the total viable moulds of crude gum arabic ranged from $3.0 \times 10$ to $1.5 \times 10^2$ cfu/g.

Mohamed (2000) reported that the fungal load was lower than bacterial load. No yeasts could be detected.

The North East Wales Institute (NEWI, 1987) reported that gum arabic (Kordofan gum), contained different species of moulds such as *Aspergillus*, *Rhizopus*, *Penicillium* and other unidentified species of moulds.

Idris (1989) isolated from gum arabic samples five genera of moulds, namely *Aspergillus*, *Penicillium*, *Rhizopus*, *Gilocladium* and *Cladosporium*.

Bokhary et al. (1983) found that natural gum arabic, in different growth media, carried appreciable numbers of microorganisms such as the fungi *Aspergillus sp*, *Alternaria sp*, *Curvolaria sp* and *Helminthosporium sp*. 
Anderson and Mc Dougall (1987) reported that natural gum arabic carried a wide range of yeasts and moulds such as *Aspergillus sp.*

Obied (2002) stated that the total viable moulds of raw gum arabic ranged from Nil to $6.5 \times 10^{cfu/g}$, and no yeasts could be detected.

### 2.12.3 Spore-former count

**Anderson and Mc Dougall (1987) reported that natural gum arabic carried a wide range of thermophilic spore-forming bacilli of remarkable tolerance to heat treatment (*Bacillus stearothermophilus*).**

This result confirmed the conclusion reported by British Pharmacopoeia (BP, 1986) that thermal or other sterilization processes are necessary to ensure that *Acacia senegal* gum conforms to established microbiological criteria for pharmaceutical use.

Idris (1989) isolated from gum arabic samples some bacterial genera such as gram-positive spore-forming bacilli, gram-positive non-spore-forming bacilli, gram-negative bacilli and gram-positive cocci.

Recent study done by Obied (2002) stated that total viable count of spore-forming bacteria of raw gum arabic samples ranged from $1.5 \times 10$ to $1.4 \times 10^{2} cfu/g$.

### 2.12.4 Some health indicators

Bokhary *et al.* (1983) found that natural gum arabic (*Acacia senegal*), contained appreciable numbers of microorganisms such as the bacteria *Bacillus sp*, *Serratia sp* and *Micrococcus varians*. 
Osman (1998) stated that no coliform bacteria were isolated from raw gum arabic samples.

Obied (2002) reported that coliform bacteria (Enterobacter cloacae, Cedecea lipagei and Klebsiella pneumoniae) in raw gum arabic samples did not exceed $1.5 \times 10$ cfu/g and that *Escherichia coli* was not detected. On the other hand, isolates of gram-negative rod bacteria were identified as *Salmonella* sp, *Providencia*, *Proteus*, *Serratia* sp, *Klebsiella* sp, *Pseudomonas* sp and *Moraxella* sp.

2.13 Physico-chemical characterization of gum arabic

2.13.1 Moisture content

Anderson and Stoddart (1966) reported that Kordofan gum arabic contained (11±1)% moisture content. Also later it was found that moisture content of commercial gum arabic ranged from 11.0 to 12.7% Awad Elkarim (1994).

Osman (1998) stated that the moisture content of commercial *Acacia senegal* gum ranged from 10.35 to 11.31%.

The FAO/WHO Joint Expert Committee of Food Additives (JECFA, 1990) specified that in good quality crude commercial gum arabic moisture content should not exceed 15%.

2.13.2 Ash content and minerals

Anderson (1976) reported that *Acacia senegal* gum contained 3.93% ash. Also later it was found that the average ash content of *Acacia senegal* specimens and commercial gum arabic samples was 4.1% (Anderson *et al.* 1983).
The ash content of gum arabic subjected to degradation using auto-hydrolysis, mild acid hydrolysis increased slightly from 3.6 to 3.8% (Anderson and Mc Dougall, 1987).

Snowden et al. (1987) found that gum arabic contained 0.89%, 0.98% and 0.02% of Ca\(^{++}\), K\(^+\) and Na\(^+\) ions, respectively.

2.13.3 pH value

Gum arabic is slightly acidic, with a pH of about 4.5–5.5 (Glicksman, 1969).

Anderson et al. (1990) stated that the mean value of the pH of Sudanese gum arabic was found to be 4.40. Later Awad Elkarim (1994) reported a range of pH from 4.2–4.8 for crude commercial gum arabic samples.

2.13.4 Brookfield viscosity

The viscosity of a liquid is its resistance to shearing, stirring or to flow through a capillary tube. It can be presented in many terms e.g. relative viscosity, intrinsic viscosity…etc.

Viscosity is considered one of the most important analytical and commercial parameters of gums, since it is a factor involving the size and shape of macromolecules (Anderson and Dea, 1969).

Recently, Karamalla et al. (1998) reported intrinsic viscosity value of 82.67 ± 3.3 cps (centipoises/sec) at 20% gum solution.

The gum arabic forms very viscous solutions at low concentration (1–5%). Gum arabic is extremely soluble and can form solutions over a wide range of concentrations, up to 37% at 25\(^\circ\)C (Osborne and Lee, 1951; Mason, 1943).
Anderson et al. (1990) reported that the Brookfield viscosity of Sudanese gum was 78 cps.

### 2.13.5 Specific optical rotation

Specific rotation is considered the most important criterion of purity and identity of gum arabic. It represents the fingerprint in physico-chemical differentiation between *Acacia senegal* gum and other botanically related *Acacia* gums (Obied, 2002).

In the revised specifications of gum arabic by FAO (1990), the specific rotation of *Acacia senegal* falls in the range between \(-26^\circ\) to \(-24^\circ\). On the other hand Fenyo (1982) reported that specific rotation to be ranging between \(-29^\circ\) to \(-34.4^\circ\). Also Stephen (1987) reported that specific rotation of gum arabic was \(-32^\circ\).

Studies by Jurasek et al. (1993) showed that specific rotation ranged between \(-20^\circ\) to \(-30^\circ\), while Osman et al. (1993) reported that specific rotation of commercial gum arabic samples ranged from \(-29^\circ\) to \(-30^\circ\).

Awad Elkarim (1994) and Omar et al. (1995) reported that specific rotation for the commercial gum arabic samples was \(-22^\circ\) to \(-28^\circ\) and \(-32^\circ\) to \(-33.3^\circ\), respectively.

### 2.13.6 Nitrogen (protein) content

Anderson and Stoddart (1966) and Anderson et al. (1983) reported that *Acacia senegal* gum contained 0.33% nitrogen (2.1% protein).

Anderson et al. (1968) investigated the nitrogen content of *Acacia senegal* gum from different locations. On heavy soil (eastern Sudan) the gum contained 0.35% nitrogen (2.2% protein), while samples from
sandy soil (Kordofan Province, western Sudan) contained 0.39% nitrogen (2.44% protein).
CHAPTER THREE

4. MATERIALS AND METHODS

3.1 Materials

3.1.1 Collection of gum arabic samples

The study was conducted in Kordofan states, for the two seasons (2002/03 and 2003/04). The samples of gum arabic for this study were collected from West Kordofan State (Elnuhode, Eyal Bakheet and Wad Banda) and North Kordofan State (Shekan, Um Rwaba, Nawa, El Rahad and Damukya), as well as from Elobied crops market and Gum Arabic Company stores. One hundred and twenty samples of gum arabic were used in this study. A random sampling technique was adopted in sample collection from the fields, markets and stores at different phases of gum production as follows:

1- immediately after the exudation of gum nodules.
2- after collection in fields.
3- after store in villages.
4- after transportation to Elobied crops market.
5- at store (recent storage) at Gum Arabic Company stores at Elobied.
6- after one-year storage at Gum Arabic Company stores at Elobied.

3.1.2 Sterilization of glassware

Before sterilization, glassware were washed thoroughly and left to dry, then they were sterilized in a hot oven at 160°C for at least 3 hours (Harrigan and McCance, 1976).

Instruments such as loops, needles, forceps, spoons and knives were sterilized by flaming directly after dipping in spirit.
3.1.3 Culture media and chemicals

The following media and chemicals, supplied by Oxoid Unipath Ltd. England, were used to enumerate and detect different types of Microorganisms, according to Harrigan, (1998).

3.1.3.1 Solid media

3.1.3.1.1 Plate count agar

This was a non-selective medium for general viable counts of bacteria in foods (Harrigan, 1998). It was obtained in a dehydrated form. The medium was composed of yeast extract, tryptone, D-glucose and granulated agar. It was prepared according to the manufacturer’s instructions by using 17.5g in one liter distilled water. The medium was allowed to boil in water bath until it was completely dissolved. The pH was adjusted to pH 7.0 and then the medium was sterilized in an autoclave at 121°C for 15 minutes.

3.1.3.1.2 Malt extract agar

This medium was used for the isolation, counting and cultivation of yeasts and moulds. It was obtained in a dehydrated form. The medium was composed of malt extract, yeast extract, peptone, glucose and agar. It was prepared according to the manufacturer’s instructions by suspending 50g in one liter distilled water. The medium was allowed to boil in water bath until it was completely dissolved. The pH of the medium was adjusted to pH 5.6 and then the medium was sterilized in an autoclave at 115°C for 10 minutes (Harrigan, 1998).
3.1.3.1.3 Nutrient agar

This was a general-purpose culture medium for sub-culturing of bacteria. It was obtained in a dehydrated form. The constituents of the medium were lab-lemco powder, yeast extract, peptone, sodium chloride and agar. It was prepared according to the manufacturer’s instructions by suspending 40g in one liter distilled water. The medium was allowed to boil until it was completely dissolved. The pH of medium was adjusted to pH 7.0 and then the medium was sterilized in an autoclave at 121°C for 20 minutes (Harrigan, 1998).

3.1.3.1.4 Staphylococcus medium No.110

This selective medium was used for the detection of *Staphylococcus aureus*. The medium consisted of yeast extract, tryptone, lactose, mannitol, sodium chloride, dipotassium hydrogen phosphate, gelatin (Oxoid L18) and agar No.3. It was prepared according to the manufacturer’s instructions by suspending 150g in one liter distilled water. The medium was allowed to boil in water bath until it was completely dissolved. The pH of the medium was adjusted to pH 7.0 and then the medium was sterilized in an autoclave at 121°C for 20 minutes.

3.1.3.1.5 Bismuth sulphite agar

This was used for *Salmonella* detection. It was obtained in a dehydrated form. The medium was composed of Bacto-beef extract, peptone, Bacto-dextrose, disodium phosphate, ferrous sulphate, bismuth sulphate indicator, Bacto-agar and brilliant green. It was prepared according to the manufacturer’s instructions by suspending 52g in one
liter distilled water. The medium was allowed to boil in a water bath until it was completely dissolved. The pH of the medium was adjusted to pH 7.0 and then the medium was sterilized in an autoclaved at 121°C for 20 minutes.

3.1.3.1.6 Eosin methylene blue agar (EMB)

This medium was used for the detection of coliform bacteria and/or *Escherichia coli*. It was obtained in a dehydrated form. The constituents of the medium were peptone, lactose, dipotassium hydrogen phosphate, eosin Y, methylene blue and agar. It was prepared according to the directions of the manufacturer’s by suspending 37.5g in one liter distilled water and bringing to the boil to dissolve completely. The pH of the medium was adjusted to pH 6.8 and then the medium was sterilized at 150°C for 20 minutes (Cowan and Steel, 1974).

3.1.3.1.7 Triple sugar agar

**THE MEDIUM WAS USED FOR DIFFERENTIATION OF THE ENTEROBACTERIACEAE (SALMONELLA).** IT CONSISTED OF PEPTONE, LACTOSE, SUCROSE, D-GLUCOSE, SODIUM CHLORIDE, SODIUM THIOSULPHATE PENTAHYDRATE, DI-AMMONIUM ION (II) SULPHATE HEXAHYDRATE, PHENOL RED AND AGAR. IT WAS PREPARED ACCORDING TO MANUFACTURER’S INSTRUCTIONS BY SUSPENDING 65G IN ONE LITER DISTILLED WATER. THE MEDIUM WAS BOILED TO DISSOLVE AND THEN DISPENSE IN 7 ML AMOUNTS IN TEST TUBES. IT WAS STERILIZED IN AN AUTOCLAVE AT 121°C FOR 15 MINUTES. IT WAS THEN ALLOWED TO SET AS SLOPE WITH A 3 CM BUTT (HARRIGAN, 1998).
3.1.3.1.8 Simmon’s citrate agar

The medium was used for the determination of citrate utilization, particularly for differentiation of enterobacteria. The medium was composed of sodium chloride, magnesium sulphate hydrated, ammonium dihydrogen phosphate, dipotassium hydrogen phosphate, sodium citrate, bromothymol blue and agar. Suspending 34.28 prepared it in one liter distilled water. Add all the ingredients except the indicator solution to the water and dissolve by steaming. Adjust to pH 7.0 and the bromothymol blue. Mix and dispense in test tubes and the medium then sterilized in an autoclave at 121°C for 15 minutes. Set as short agar slopes (Harrigan, 1998).

3.1.3.1.8 Christensen’s urea agar

The medium was used for the determination of urease utilization. The medium was composed of peptone, sodium chloride, potassium dihydrogen phosphate, D-glucose, phenol red and agar. It was prepared according to manufacturer’s instructions by suspending 29g in one liter distilled water. Dissolve the ingredients in the water by steaming. Adjust to pH 7.0 and distribute in bottles or test tubes and sterilize by autoclaving at 121°C for 15 minutes. Set it in the sloped position (Harrigan and McCance, 1976).

3.1.3.1.9 SIM medium

The medium was used for the determination of motility and indole tests. The medium was composed of tryptone, meat extract, disodium thiosulphate, cysteine hydrochloride, sodium chloride and agar. It was prepared according to manufacturer’s instructions by suspending 43.7g in one liter distilled water. Dissolve the ingredients in the water by
heating. Dispense the medium into bottles or test tubes and sterilize by autoclaving at 121°C for 15 minutes. (Barrow and Gelthan, 1993).

3.1.3.2 Semi-solid media

3.1.3.2.1 Hugh and leifson’s medium

This was used for differentiating oxidative and fermentative metabolism of carbohydrates (Harrigan, 1998). The medium consisted of tryptone, yeast-extract, D-glucose, bromocresol purple and agar. The ingredients were added to one liter distilled water and dissolved by steaming. The pH was adjusted to pH 7.0 and then the medium was sterilized by autoclaving and sterile glucose (sterilized by tendallization) was aseptically added to the previously sterilized basal medium to give a final concentration of 1%. The medium was steamed for 10-15 minutes before use to expel the excess oxygen.

3.1.3.3 Liquid media

3.1.3.3.1 Nutrient broth (NB)

This medium was used for the detection of Salmonella. The medium consisted of peptone, sodium chloride and lab-lemco extract (Oxoid). The ingredients were dissolved in distilled water by heating at 100°C. Then the pH was adjusted to pH 7.6 and the medium was sterilized in an autoclave at 121°C for 15 minutes (Harrigan, 1998).

3.1.3.3.2 MacConkey broth

This medium was used for the detection and enumeration of lactose-fermenting enterobacteria (coliorm bacteria) by the multiple tube technique (FAO, 1992). It was obtained in a dehydrated form. The medium consisted of peptone, lactose, bile salt, sodium chloride and
bromocresol purple. It was prepared according to the manufacturer’s
instruction by suspending 40g in one liter distilled water. The medium
was distributed in test tubes with inverted Durham tubes. The pH was
adjusted to pH 7.0 and the medium was sterilized in an autoclave at
121°C for 20 minutes.

3.1.3.3.3 Selenite cystine broth

This was used as an enrichment medium for *Salmonella* detection.
The medium was composed of peptone, lactose sodium hydrogen
selenite, disodium hydrogen phosphate, and cystine. The ingredients
were dissolved in one liter distilled water, distributed in suitable amounts
and sterilized in a boiling water bath for 10 minutes (Harrigan, 1998).

3.1.3.3.4 Brilliant green bile lactose broth

This is a selective medium for isolation and counting of *coliform*
and *Escherichia coli* by the multiple tube technique. It was obtained in a
dehydrated form. The medium was composed of peptone, lactose,
purified bile and brilliant green. It was prepared according to the
manufacturer’s instructions by suspending 40g in one liter distilled
water. The pH was adjusted to pH 7.4, distributed in test tubes with
inverted Durham tubes and the medium then sterilized in an autoclave at
121°C for 15 minutes (Harrigan, 1998).

3.1.3.3.5 EC medium broth

This was used for the detection of coliform bacteria and/or
*Escherichia coli* by the multiple tube technique. It was obtained in a
dehydrated form. The medium was composed of tryptose, lactose, bile
salts, dipotassium hydrogen phosphate, potassium dihydrogen phosphate
and sodium chloride. It was prepared according to the manufacturer’s
instructions by suspending 37g in one liter distilled water. The pH of the medium was adjusted to pH 7.0, distributed in test tubes with inverted Durham tubes and the medium then sterilized in an autoclave at 121°C for 15 minutes (Harrigan, 1998).

3.1.3.3.6 Glucose phosphate broth

This medium was used for the methyl red and Voges-Proskauer test. The medium consisted of peptone, D-glucose and dipotassium hydrogen phosphate. It was prepared according to the manufacturer’s instruction by suspending 15g in one liter distilled water and then the pH was adjusted to pH 7.5. And distribute it in test tubes. The medium was sterilized in an autoclave at 121°C for 20 minutes (Harrigan and McCance, 1976).

3.2 Methods

3.2.1 Microbiological methods

3.2.1.1 Sampling methodology

The sampling procedure was done according to Kiss (1984). To avoid any microbial contamination the following procedure in gum arabic sampling at different phases of gum production (after exudation, after collection, at village market, at crops market, at recent and long storage) was fully employed.

1- Wearing sterile hand-gloves after disinfection of hands.
2- Alcohol-sterilized forceps were employed in picking gum samples.
3- Samples were kept in labeled sterilized polythene bags.
4- Samples were kept at 4-10°C until used.
PLATE (3.1): MATERIALS AND TOOLS USED IN GUM SAMPLING PROCEDURE

(1): Polythene bags           (2): Spirit lamp
(3): Aluminum foil            (4): Forceps
(5): Hand-gloves              (6): Alcohol
Plate (3.2): Collection of gum arabic samples after gum exudation

Plate (3.3): Collection of gum arabic samples after gum collection in field
Plate (3.4): Gum arabic at village market

Plate (3.5): Gum arabic at crops market
Plate (3.6): Gum arabic at stores

3.2.1.2 Preparation of serial dilutions

The gum nodules were dissolved in sterile peptone water. Ten grams of each sample were weighed and added to a conical flask containing 90 ml sterile 0.1% peptone water and were shaken for 45 minutes using electric shaker at speed of 150 RPM to release any microorganisms trapped in the nodules. This dilution was referred to as mother (dilution $10^{-1}$). One ml of mother solution were pipetted aseptically, with sterilized pipette into 9 ml sterile peptone water (dilution $10^{-2}$) and serial decimal dilutions up to $10^{-6}$ were prepared as described by Harrigan (1998).

3.2.1.3 Microbial parameters studies

3.2.1.3.1 Determination of microbial load of gum arabic

Total viable count was carried out using the pour-plate method as described by Harrigan, (1998). One ml of each dilution was transferred aseptically into sterile Petri dishes. To each dilution 10-15 ml of melted and cooled (42°C) plate count agar were added. The inoculum was mixed with medium and allowed to solidify. The plates were then incubated aerobically in an incubator at 37°C for 48 hours. A colony counter was used to count the viable bacterial colonies. The count was expressed as colony-forming units (cfu) per gram (Dirar, 1976).
3.2.1.3.2 Mould and yeast enumeration

From suitable dilutions of sample 0.1 ml was aseptically transferred onto solidified malt extract agar containing 0.1 g chloramphenicol per one liter of medium to inhibit bacterial growth. The sample was spread all over the plates using sterile bent glass rod. Plates were then incubated at 25-28°C for 48 hours as described by Harrigan and McCance (1976). Colonies were counted using a colony counter and the results were expressed as cfu/g.

3.2.1.3.3 Spore-formers enumeration

From each dilution were heated in a water-bath at 80°C for 15 minutes and cooled. 1 ml of each dilution was pour-plated on nutrient agar medium and then incubated at 37°C for 24 hours. Counting was expressed as cfu/g.

3.2.1.3.4 Staphylococcus aureus enumeration

From suitable dilution, 0.1 ml was spread on solidified Staphylococcus medium 110 plates and the plates were incubated at 37°C for 24 hours. Counting was expressed as cfu/g.

3.2.1.3.5 Determination of Salmonella

Ten grams of gum samples were weighed aseptically and mixed well with 90 ml sterile nutrient broth and incubated at 37°C for 24 hours. A loopful of incubated NB were transferred aseptically into sterilized selenite broth and incubated at 37°C for 24 hours. A loopful of 24 hours inoculum was streak-plated on bismuth sulphate agar surface and incubated at 37°C for 24 hours. Salmonella colonies appear as gray
colonies with black centers (black metallic sheen). Taking gray colony and sub-culturing it in triple sugar iron agar tubes carried out a confirmatory test. Production of a black color at the bottom of the tube confirmed the presence of *Salmonella.*
3.2.1.3.6 Determination of coliform bacteria

3.2.1.3.6.1 Presumptive coliform test

One ml of each of the three first dilutions (10^{-1}, 10^{-2} and 10^{-3}) was inoculated aseptically in triplicates of 9 ml sterilized MacConkey broth using the three-tube technique with Durham tubes. The tubes were incubated at 37\(^\circ\)C for 48 hours. Positive tubes gave gas in the Durham tubes.

3.2.1.3.6.2 Confirmed coliform test

All tubes of the dilution showing gas fermentation in 24 hours, were submitted to the confirmed test using brilliant green bile lactose broth fermentation tubes with Durham tubes, and then incubated at 37\(^\circ\)C for 48 hours. The most probable number (MPN) was recorded. The most probable number (MPN) tables were used according to FAO (1992) to record the coliforms number (FAO, 1992).

3.2.1.3.6.3 Faecal coliform test

At least 3 loopfuls of each confirmed positive tube were sub-cultured into EC broth medium and then incubated at 44.5\(^\circ\)C for 24 hours. Tubes showing any amount of gas production were considered positive. The most probable number (MPN) was recorded.

3.2.1.3.6.4 Differentiation of faecal coliform

For further confirmation of faecal coliforms, tubes giving positive reaction at 44.5\(^\circ\)C for 24 hours were streaked
ON EMB AGAR. COLONIES WITH GREEN METALLIC SHEEN INDICATED A POSITIVE TEST.
3.2.1.4 Purification and identification of isolates

Predominant microorganisms from morphologically different colony types were selected from plate count agar. Sub-culturing purified these isolates; typical colony was streaked onto sterile nutrient agar plates. The plates were incubated at 37°C for 24 hours. The representative colonies of various microorganisms were sub-cultured in their respective media (on slopes) and then the cultures were kept in the refrigerator at 4°C until used for further test. The identification of purified isolates was carried out according to Cowan and Steel (1974).

3.2.1.5 Biochemical tests

Below are some important biochemical tests:

3.2.1.5.1 Gram stain test

A discrete colony was picked carefully with sterile wire loop. The colony was emulsified in a drop of physiological saline (0.85%NaCl), placed on a clean slide and spread evenly to make a thin film. The slide was allowed to dry. The smear was fixed by using a flame. Then the smear was stained as described by Harrigan and McCance (1976).

3.2.1.5.2 Oxidase test

A piece of filter paper was impregnated with oxidase test solution (Difco laboratories, Detroit Michigan USA). Then a loopful from a 24 hours culture was streaked onto the filter paper. A positive reaction was indicated by purple color after 10-15 seconds, any later reaction being recorded as negative (Harrigan, 1998).
3.2.1.5.3 Catalase test

This demonstrates the presence of catalase, an enzyme that catalyses the release of oxygen from hydrogen peroxide.

One drop of 3% hydrogen peroxide solution was placed on a clean slide. A loopful from 24 hours culture was added. The release of bubbles of oxygen indicated the presence of catalase in the culture under test (Harrigan, 1998).

3.2.1.5.4 Motility test

A TUBE OF MOTILITY MEDIUM (SIM MEDIUM WITH CONCENTRATION OF 0.4% OF NUTRIENT AGAR) WAS INOCULATED WITH A 24-48 HOURS CULTURE. THIS WAS DONE ASEPTICALLY USING A STRAIGHT WIRE TO HALF DEPTH OF THE TUBE. DURING GROWTH, MOTILE BACTERIA WILL MIGRATE FROM THE LINE OF INOCULATION TO FORM TURBIDITY IN THE SURROUNDING MEDIUM. NON-MOTILE BACTERIA WILL GROW ONLY ALONG THE LINE OF INOCULATION.

3.2.1.5.5 Oxidation-fermentation (O/F) test

Fresh culture (18-24 hours) was tested for O/F test by stab inoculation onto pairs of freshly steamed Hugh and Leifson's medium (Hugh and Leifson's, 1953), contained in test tubes. One of the tubes was sealed with sterile paraffin oil and the other left unsealed. Inoculation was carried out at 37°C for 2-7 days. Acid production is shown by change in the color of the medium from blue to yellow but fermentative organism produce acid in both tubes, and oxidative organisms produce acid in the open tube only.
3.2.1.5.6 Glucose (acid) test

After prepared the medium of glucose broth methyl red was added as indicator and the pH adjusted to pH 7.4, then the medium distributed in test tubes with inverted Durham tubes. Some bacteria ferment certain sugars with the production of acid and gas; others produce neither acid nor gas. The positive result is the change in color (acid) and production gas in the Durham tubes (Hobbs, 1978).

3.2.1.5.7 Methyl red (MR) test

The medium used was glucose phosphate broth to which five drops of the methyl red were added as indicator and then inoculated and incubated at the optimum growth temperature for 2-7 days. A red color was described as positive. A yellow coloration was negative (Harrigan, 1998).

3.2.1.5.8 Voges-Proskauer (VP) test

This test was used to assist in the differentiation of enterobacteria that ferment glucose with the production of acetyl-methyl carbinol (acetoin), which could be detected by an oxidation. The medium used was glucose phosphate broth, as in the methyl red test, and then add 0.05 ml creatine and 1 ml of KOH (40%) were added and the tube shaken well. The development of a pink color in the medium, usually within 30 minutes, indicated a positive reaction (Harrigan, 1998).
3.2.1.5.9 Urease test

Testing for urease enzyme activity is important in differentiating enterobacteria and excluding non-lactose fermenters producing urease. The medium used is basal medium (Christensen’s urea agar) which was sterilized by autoclaving. Then sterile 20% urea solution was added and the test culture streaked and incubated. The ammonia produced from the decomposition of urea changes the color of the indicator from yellow to pink or red (Kiss, 1984).

3.2.1.5.10 Production of hydrogen sulphide

Hydrogen sulphide may be formed from organic sulphur compounds (e.g. cysteine and cystine) or from the reduction of inorganic sulphur compounds (e.g. sulphite). Sim media was prepared and stab culture made. Filter paper is soaked in saturated acetate solution; this was used as indicator paper and incubated at 37°C for 2 days. Production of hydrogen sulphite causes blackening of the indicator paper (Kiss, 1984).

3.2.1.5.11 Production of indole

This test is important for the identification of enterobacteria. Prepare the SIM media was prepared to make slope culture and put filter paper soaked in Kovac’s indole reagent or oxalic acid and incubate at the optimum growth temperature for 2 days. Only a red or pink color should be recorded as an indole-positive result (Harrigan, 1998).
3.2.1.5.12 Utilization of citrate as the sole source of carbon

Simmon’s citrate agar medium was prepared as a slope as described by Harrigan (1998). The slope was inoculated with the test organism and incubated. The change of color from green to bright blue is positive results.

3.2.1.5.13 Decarboxylation of lysine

The test was based on the ability of some bacteria to Decarboxylate an amino acid to the corresponding amine with the liberation of carbon dioxide. The production of these carboxylases is induced by low pH and, as a result of their action; the pH rises to neutrality or above.

A small amount of bacteria was taken by straight wire, and then the butt of the medium for carrying out the lysine decarboxylase test was inoculated by stabbing technique and the slant by zig sag technique. The test was incubated at 37°C for 18-24 hours. Positive lysine was indicated by the appearance of violet color throughout the medium. A negative result was indicated by keeping the yellow color as it was (Colle et al. 1996).

3.2.1.5.14 API test

API manual system which was done to confirm the biochemical tests, which were used above.

3.2.2 Physico-chemical methods
3.2.2.1 Moisture content
An empty crucible was dried in Imperial IV Laboratory oven at 105°C for 30 minutes, cooled in a desiccator and weighed. About one gram of air-dried ground gum samples was placed in the crucible and weighed. Then the crucible and content were heated for three hours at 105°C, cooled in a desiccator and weighed. This was repeated till constant weight was obtained (Allen, 1989).

Calculation:

\[
\text{Moisture} \,(\%) = \left( \frac{\text{loss in weight on drying} \,(\text{g}) \times 100\%}{\text{Initial sample weight} \,(\text{g})} \right)
\]

3.2.2.2 Ash content

An empty crucible was heated at 550°C, cooled in a desiccator and weighed (W1). About one gram of gum sample was transferred to the crucible, dried at 105°C for two hours and weighed (W2), after that it was ignited at 550°C in Fisher Scientific Isotemp Muffle Furnace for two hours until free from carbon, then cooled in a desiccator and weighed (W3) (Allen, 1989).

Calculation:

\[
\text{Ash} \,(\%) = \left( \frac{W_3 - W_1 \times 100\%}{W_2 - W_1} \right)
\]

3.2.2.3 pH value

The pH was determined in 10% aqueous solution, using Jenway 3320 pH meter. The temperature was kept at 30°C.

3.2.2.4 Brookfield viscosity
Viscosity was measured using Brookfield model DV-II+ viscometer, using 25% gum arabic solution. The viscosity of gum arabic solution was read directly in centipoises/second (cps). The readings were taken three times and averaged out.

3.2.2.5 Specific optical rotation

The specific optical rotation was determined for 0.1% solution on dry weight basis using an Optical Activity Ltd. Automatic Polarimeter fitted with a sodium lamp and with a cell path length of 20 cm. The solution was passed through a No.42 filter paper before carrying out the measurement at room temperature (30°C). Readings were taken three times and averaged. The specific optical rotation for the gum arabic solution was calculated as follows:

\[
\text{Specific optical rotation} = \frac{\alpha \times 100}{C \times L}
\]

Where:

\( \alpha = \) observed optical rotation.

\( C = \) concentration of the solution.

\( L = \) length of the Polarimeter tube.

3.2.2.6 Nitrogen and crude protein
The protein was determined according to AOAC (1984). The nitrogen content was estimated by micro Kjeldahl system, and then the nitrogen content was multiplied by the factor 6.25 to obtain the percentage of crude protein as follows:

\[
\text{Protein (N} \times 6.25\text{) \%} = \frac{T \times N \times 14 \times 100\%}{S \times 1000}
\]

Where:

\(T = \text{titration figure of HCl (0.02N).}\)
\(N = \text{HCl normality (0.02N).}\)
\(S = \text{weight of sample (0.5g).}\)
\(14 = \text{nitrogen molecular weight.}\)

3.2.2.7 Analysis of inorganic matter

Two hundred mg of dry gum were ignited in a crucible in a Muffle Furnace at 550°C for two hours. When cooled, 5 ml HCl (50%) were added to the crucible and heated on a sand bath for 15 minutes. One ml HNO₃ (98%) was added and the material evaporated to dryness, then heated again for one hour. One ml mixer of 1ml H₂O +1ml HCl was added and swirled to dissolve the residue, and then the solution was diluted to 10 ml with distilled water and warmed to complete the dissolution. The mixture was filtered and distilled water was added to complete the volume to 50 ml. Na⁺ and K⁺ were read on the Atomic Absorption Spectrophotometer. Calibration standards were prepared for Na⁺ and K⁺. The sample readings were recorded and the percentages of
minerals were calculated. Ca$^{++}$ and Mg$^{++}$ determinations were carried out according to Allen (1989).
5. RESULTS AND DISCUSSION

Gum arabic samples under study were taken from six different phases of gum production (immediately after exudation of gum nodules, after collection in field, at village market, at crops market, at recent storage and at long storage) for the two seasons 2002/03 and 2003/04. The samples were collected from North and West Kordofan from different fields and villages, namely, Eyal Bakheet and El Rahad, and from markets (Elobied crops market) and stores (Gum Arabic Company, Elobied stores).

4.1 Microbiological quality

4.1.1 Total viable count of bacteria

**Total viable count (TVC) of bacteria, which is regarded as a microbiological indicator of contamination, was determined for the 36 samples collected from different locations and different phases of gum arabic production.**

**Total viable count of bacteria of 36 samples taken during the production of gum arabic after exudation of gum nodules, after collection, at village market, at crops market, at recent and long storage was obtained with the mean value of 1.5×10^4, 1.4×10^4, 2.1×10^4, 4.7×10^3, 4.5×10^3 and 3.7×10^3 CFU/G, respectively (Table 4.1).**

**The load in samples picked from trees immediately after exudation of gum nodules was in the range between 1.2×10^3 and 4.0×10^4 CFU/G, the load in samples after collection**
IN FIELDS WAS IN THE RANGE BETWEEN $2.8 \times 10^3$ AND $2.2 \times 10^4$ CFU/G AND THE LOAD IN SAMPLES AT VILLAGE MARKET WAS IN THE RANGE BETWEEN $2.0 \times 10^3$ AND $9.0 \times 10^4$ CFU/G. BUT THE LOAD

**Table (4.1). Total viable count of bacteria of samples drawn during different phases of gum arabic production**

<table>
<thead>
<tr>
<th>Phases</th>
<th>No. of samples</th>
<th>Minimum</th>
<th>Maximum</th>
<th>Mean</th>
<th>Std. Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>After exudation</td>
<td>6</td>
<td>$1.2 \times 10^3$</td>
<td>$4.0 \times 10^4$</td>
<td>$1.5 \times 10^4$</td>
<td>0.59</td>
</tr>
<tr>
<td>After collection</td>
<td>6</td>
<td>$2.8 \times 10^3$</td>
<td>$2.2 \times 10^4$</td>
<td>$1.4 \times 10^4$</td>
<td>0.38</td>
</tr>
<tr>
<td>At village market</td>
<td>6</td>
<td>$2.0 \times 10^3$</td>
<td>$9.0 \times 10^4$</td>
<td>$2.1 \times 10^4$</td>
<td>0.55</td>
</tr>
<tr>
<td>At crops market</td>
<td>6</td>
<td>$5.0 \times 10^2$</td>
<td>$8.0 \times 10^3$</td>
<td>$4.7 \times 10^3$</td>
<td>0.45</td>
</tr>
<tr>
<td>At recent storage</td>
<td>6</td>
<td>$3.1 \times 10^3$</td>
<td>$8.0 \times 10^3$</td>
<td>$4.5 \times 10^3$</td>
<td>0.15</td>
</tr>
<tr>
<td>At long storage</td>
<td>6</td>
<td>$3.1 \times 10^3$</td>
<td>$4.5 \times 10^4$</td>
<td>$3.7 \times 10^3$</td>
<td>0.06</td>
</tr>
</tbody>
</table>

IN SAMPLES AT CROPS MARKET WAS IN THE RANGE BETWEEN $5.0 \times 10^2$ AND $8.0 \times 10^3$ CFU/G. THE LOAD IN SAMPLES AT RECENT STORAGE WAS IN THE RANGE BETWEEN $3.1 \times 10^3$ AND $8.0 \times 10^3$ CFU/G AND THE LOAD IN SAMPLES AT LONG STORAGE WAS IN THE RANGE BETWEEN $3.1 \times 10^3$ AND $4.5 \times 10^3$ CFU/G.

*The bacterial load during the present study was generally not high, but there was increase in the bacterial load of samples taken after collection in field from after exudation. However, there was some increase in the bacterial load during transport from field to the village market. This could lead to accumulation of high moisture content and*
hence to microbial growth. The microbial load decreased at the crops market and on storage phases Fig (4.1). This decrease could be due to the low moisture content and to the death of the bacteria by the sun.

The present results of TVC of bacteria of all gum arabic samples show much lower values than the loads reported by Khalid (1984) who found a bacterial load in raw gum arabic in the range of $4.8 \times 10^5$ and $9.9 \times 10^6$ CFU/G. The present counts are also much lower than the results obtained by Osman (1998) and Karamalla (1999) who stated that the

![Graph showing variation in total viable count of bacterial for different phases of gum production](image)

Fig. 4.1. Variation in total viable count of bacterial for different phases of gum production
TVC of bacteria of gum arabic samples picked from trees, local market and in the stores were found to be in range from $5.9 \times 10^3$ to $1.3 \times 10^5$; $2.9 \times 10^4$ to $7.7 \times 10^5$ and $1.7 \times 10^4$ to $3.1 \times 10^6$ CFU/g, respectively, in Elobied area.

The present TVC of bacteria is higher than that obtained by Mohamed (2000) who reported that the TVC of bacteria of raw gum was expected not to exceed $8.2 \times 10^3$ CFU/g and Obied (2002) who stated that TVC of bacteria of raw gum arabic samples ranged from $2.1 \times 10^2$ to $7.0 \times 10^2$ CFU/g.

The present TVC of bacteria is in agreement with the loads reported by Idris (1989) which was in the range between $3.0 \times 10^2$ and $9.0 \times 10^4$ CFU/g and Ismail (2000) who reported that TVC of bacteria of raw gum was found to range from $4.5 \times 10^4$ and $8.5 \times 10^4$ CFU/g.

4.1.2 Total viable count of moulds

Generally, no signs of definite spoilage by moulds were detected since TVC of moulds of value $<1.0 \times 10^2$ is considered to be negligible in food (Jay, 1986).

The mean value of TVC of moulds through 36 sampling points of gum arabic production after exudation, after collection, at village market, at crops market, at recent and long storage was found to be $6.2 \times 10^3$ CFU/g (range from $8.0 \times 10^2$ to $1.7 \times 10^4$); $5.9 \times 10^3$ CFU/g (range from $1.0 \times 10^2$ to $2.0 \times 10^4$); $4.5 \times 10^2$ CFU/g (range from $2.0 \times 10^2$ to $8.0 \times 10^3$); $4.2 \times 10^3$ CFU/g (range from $2.0 \times 10^2$ to $2.0 \times 10^4$); $2.7 \times 10^2$ CFU/g (range from $1.0 \times 10^2$ to $2.0 \times 10^3$) and $4.3 \times 10^2$ CFU/g (range from $3.0 \times 10^2$ to $6.0 \times 10^2$), respectively (Table 4.2).
In general, contamination with moulds of samples taken during the exudation of gum nodules and after collection was relatively high compared to the other phases of gum production. This could be attributed to the presence of millet and sorghum fields in the same area; contamination could originate from these millet and sorghum fields. Also there was a decrease in the TVC of moulds at the village market. This decrease could be due to the drying effect and low moisture content. But there was a little increase of moulds load at crops market which could be due to the handling of gum arabic. There was a decrease in count at the storage phase which could be due to the low moisture content and drying effect that lead to a stop of mould growth and to the death of some sensitive species (Fig 4.2).

The present results of TVC of moulds of all gum arabic samples was found to be lower than the results obtained by Khalid (1984) who reported that TVC of moulds of different *Acacia senegal* gum samples ranged from $2.0 \times 10^3$ to $2.0 \times 10^5$ cfu/g. But the values we obtained are

**Table (4.2). Total viable moulds of samples drawn during different phases of gum arabic production**

<table>
<thead>
<tr>
<th>Phases</th>
<th>No. of Samples</th>
<th>Minimum</th>
<th>Maximum</th>
<th>Mean</th>
<th>Std. Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>AFTER EXUDATION</td>
<td>6</td>
<td>$8.0 \times 10^2$</td>
<td>$1.7 \times 10^4$</td>
<td>$6.2 \times 10^3$</td>
<td>0.61</td>
</tr>
<tr>
<td>AFTER COLLECTION</td>
<td>6</td>
<td>$1.0 \times 10^2$</td>
<td>$2.0 \times 10^4$</td>
<td>$5.9 \times 10^3$</td>
<td>0.87</td>
</tr>
<tr>
<td>AT VILLAGE MARKET</td>
<td>6</td>
<td>$2.0 \times 10^2$</td>
<td>$8.0 \times 10^2$</td>
<td>$4.5 \times 10^2$</td>
<td>0.21</td>
</tr>
<tr>
<td>AT CROPS MARKET</td>
<td>6</td>
<td>$2.0 \times 10^2$</td>
<td>$2.0 \times 10^4$</td>
<td>$4.2 \times 10^3$</td>
<td>0.69</td>
</tr>
<tr>
<td>AT RECENT</td>
<td>6</td>
<td>$1.0 \times 10^2$</td>
<td>$2.0 \times 10^3$</td>
<td>$2.7 \times 10^2$</td>
<td>0.50</td>
</tr>
</tbody>
</table>
Fig. 4.2. Variation in total viable count of mould for different phases of gum production
a little higher than the results obtained by Idris (1989) who stated that TVC of moulds of gum arabic ranged from $0.5 \times 10^5$ to $0.9 \times 10^3$ cfu/g, and Ismail (2000) who reported that TVC of moulds of crude gum arabic ranged from $3.0 \times 10^2$ to $1.5 \times 10^2$ cfu/g and much higher than counts reported by Obied (2002) who stated that TVC of moulds of raw gum arabic ranged from nil to $6.5 \times 10^2$ cfu/g.

The TVC of moulds of gum arabic samples obtained in this study was found to be in close agreement with the results obtained by Osman (1998) and Karamalla (1999) who reported that mould counts of gum arabic samples picked from trees, local market and in the stores ranged between $2.3 \times 10^3$ and $7.0 \times 10^3$ cfu/g, $3.1 \times 10^4$ and $4.2 \times 10^5$ cfu/g, and $2.6 \times 10^4$ and $3.8 \times 10^5$ cfu/g, respectively, in Elobied area.

No yeasts were detected in any of the 36 samples of gum arabic. This finding is in agreement with the results obtained by Idris (1989), Osman (1998), Mohamed (2000) and Obied (2002). And disagrees with the results reported by Anderson and Mc Dougall (1987) who stated that natural gum carried a wide range of yeasts, and with the results by Ismail (2000) who reported that TVC of yeast ranged from $0.8 \times 10^2$ to $2.3 \times 10^2$ cfu/g for crude samples of gum arabic.

### 4.1.3 Total viable count of spore-forming bacteria

The spore-forming bacteria were detected in all the 36 samples of gum arabic after exudation, after collection, at village market, at crops market, at recent and long storage with mean value of $2.7 \times 10^3$ cfu/g (range $4.7 \times 10^2$ to $7.0 \times 10^3$); $2.3 \times 10^3$ cfu/g (range $1.2 \times 10^3$ to $3.5 \times 10^3$); $3.5 \times 10^3$ cfu/g (range $1.0 \times 10^3$ to $5.0 \times 10^3$); $3.2 \times 10^3$ cfu/g (range $7.0 \times 10^2$ to $6.5 \times 10^3$); $1.7 \times 10^4$ cfu/g (range $2.3 \times 10^3$ to $5.0 \times 10^4$) and $1.7 \times 10^4$ cfu/g (range $3.5 \times 10^3$ to $3.1 \times 10^4$), respectively (Table 4.3).
It was noted that spore-formers showed high differences between the phases of gum production.

The results generally show a steady increase in the spore-formers load from exudation up to the stage of storage (Fig 4.3).

**Table (4.3). Total viable count of spore-formers bacteria of samples drawn during different phases of gum arabic production**

<table>
<thead>
<tr>
<th>Phases</th>
<th>No. of samples</th>
<th>Minimum</th>
<th>Maximum</th>
<th>Mean</th>
<th>Std. Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>After exudation</td>
<td>6</td>
<td>$4.7 \times 10^2$</td>
<td>$7.0 \times 10^3$</td>
<td>$2.7 \times 10^3$</td>
<td>0.47</td>
</tr>
<tr>
<td>After collection</td>
<td>6</td>
<td>$1.2 \times 10^3$</td>
<td>$3.5 \times 10^3$</td>
<td>$2.3 \times 10^3$</td>
<td>0.20</td>
</tr>
<tr>
<td>At village market</td>
<td>6</td>
<td>$1.0 \times 10^3$</td>
<td>$5.0 \times 10^3$</td>
<td>$3.5 \times 10^3$</td>
<td>0.26</td>
</tr>
<tr>
<td>At crops market</td>
<td>6</td>
<td>$7.0 \times 10^2$</td>
<td>$6.5 \times 10^3$</td>
<td>$3.2 \times 10^3$</td>
<td>0.38</td>
</tr>
<tr>
<td>At recent storage</td>
<td>6</td>
<td>$2.3 \times 10^3$</td>
<td>$5.0 \times 10^4$</td>
<td>$1.7 \times 10^4$</td>
<td>0.51</td>
</tr>
<tr>
<td>At long storage</td>
<td>6</td>
<td>$3.5 \times 10^3$</td>
<td>$3.1 \times 10^4$</td>
<td>$1.7 \times 10^4$</td>
<td>0.47</td>
</tr>
</tbody>
</table>
Fig. 4.3. Variation in total viable count of spore-forming bacteria for different phases of gum production
There is a drop, lower, in endospore count at the crops market. This increase could be due to lower moisture content, drying effect by the action of the sun. These effects may change the vegetative cells to the spore-form.

The present results show in some cases that TVC of spore-formers is higher than the TVC of bacteria. This increase could be due to the fact that some spore-formers do not grow without being activated first by heat treatment.

The present values of TVC of spore-formers at different phases of gum arabic production are higher than those obtained by Obied (2002) who stated that TVC of spore-forming bacteria of raw gum arabic samples ranged from $1.5 \times 10$ to $1.4 \times 10^2$ cfu/g.

This finding was in close agreement with the results obtained by Anderson and Mc Dougall (1987) who stated that natural gum arabic carried a wide range of thermophilic spore-forming bacilli of remarkable tolerance to heat treatment (*Bacillus stearothermophilus*). This result also confirms the finding of British Pharmacopoeia (BP, 1986) which reported that thermal or other sterilization processes are necessary to ensure that *Acacia senegal* gum conforms with established microbiological criteria for pharmaceutical use. Also the presence of spore-formers was in agreement with results reported by Idris (1989) who stated that gram positive spore-forming bacilli were found to be most abundant among the isolated bacteria obtained from *Acacia senegal* samples, the most dominant of which were *Bacillus licheniformis* and *Bacillus subtilis*. 
4.1.4 Detection of some health indicators

Detection of some health indicators was carried out to evaluate the hygienic conditions applied during the different phases of gum arabic production (Table 4.4).

No *Staphylococcus aureus*, *Salmonella sp*, *Coliforms* and *E. coli* were detected in all samples taken after exudation of gum nodules.

Table (4.4). Detection of some health indicators in gum arabic

<table>
<thead>
<tr>
<th>Gum Arabic samples</th>
<th>Sample point</th>
<th><em>Staphylococcus aureus</em></th>
<th><em>Salmonella sp</em></th>
<th>Coliforms</th>
<th><em>E. coli</em></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>AFTER EXUDATION</strong></td>
<td>2.3.7.8.9.10</td>
<td>-ve</td>
<td>-ve</td>
<td>-ve</td>
<td>-ve</td>
</tr>
<tr>
<td><strong>AFTER COLLECTION</strong></td>
<td>1</td>
<td>-ve</td>
<td>+ve</td>
<td>3.6/g</td>
<td>-ve</td>
</tr>
<tr>
<td>3.6.10</td>
<td>-ve</td>
<td>+ve</td>
<td>-ve</td>
<td>-ve</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>-ve</td>
<td>-ve</td>
<td>-ve</td>
<td>-ve</td>
<td></td>
</tr>
<tr>
<td>11</td>
<td>-ve</td>
<td>-ve</td>
<td>2.4×10³/g</td>
<td>-ve</td>
<td></td>
</tr>
<tr>
<td><strong>AT VILLAGE MARKET</strong></td>
<td>1.9</td>
<td>-ve</td>
<td>-ve</td>
<td>-ve</td>
<td>-ve</td>
</tr>
<tr>
<td>4.7</td>
<td>+ve</td>
<td>-ve</td>
<td>-ve</td>
<td>-ve</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>+ve</td>
<td>+ve</td>
<td>-ve</td>
<td>-ve</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>+ve</td>
<td>-ve</td>
<td>3.6/g</td>
<td>-ve</td>
<td></td>
</tr>
<tr>
<td><strong>AT CROPS MARKET</strong></td>
<td>2.9</td>
<td>-ve</td>
<td>+ve</td>
<td>-ve</td>
<td>-ve</td>
</tr>
<tr>
<td>5</td>
<td>-ve</td>
<td>+ve</td>
<td>9.3×10³/g</td>
<td>-ve</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>+ve</td>
<td>+ve</td>
<td>4.3×10³/g</td>
<td>-ve</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>+ve</td>
<td>+ve</td>
<td>2.0×10³/g</td>
<td>-ve</td>
<td></td>
</tr>
<tr>
<td>14</td>
<td>-ve</td>
<td>-ve</td>
<td>3.6/g</td>
<td>-ve</td>
<td></td>
</tr>
<tr>
<td><strong>AT RECENT STORAGE</strong></td>
<td>2.6.7.8</td>
<td>-ve</td>
<td>-ve</td>
<td>-ve</td>
<td>-ve</td>
</tr>
<tr>
<td>4</td>
<td>-ve</td>
<td>+ve</td>
<td>-ve</td>
<td>-ve</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>-ve</td>
<td>-ve</td>
<td>2.1×10³/g</td>
<td>-ve</td>
<td></td>
</tr>
<tr>
<td><strong>At long storage</strong></td>
<td>4.13</td>
<td>+ve</td>
<td>+ve</td>
<td>-ve</td>
<td>-ve</td>
</tr>
<tr>
<td>5.9</td>
<td>-ve</td>
<td>+ve</td>
<td>-ve</td>
<td>-ve</td>
<td></td>
</tr>
<tr>
<td>6.10</td>
<td>-ve</td>
<td>-ve</td>
<td>-ve</td>
<td>-ve</td>
<td></td>
</tr>
</tbody>
</table>

-ve = negative  
+ve = positive

This finding indicates good hygienic conditions in this phase of gum production.
No *Staphylococcus aureus* or *E. coli* were detected in the phase after collection in the field, but *Salmonella* and coliform bacteria were detected. This finding indicates contamination through handling of gum nodules.

*Staphylococcus aureus, Salmonella* and coliform bacteria were detected at village market and at crops market but no *E. coli* was detected at these two points. No *Staphylococcus aureus* or *E. coli* were detected at recent storage but *Salmonella* and coliform bacteria were detected. At long storage both *Staphylococcus aureus* and *Salmonella* were detected but no Coliforms or *E. coli*.

The present results were found to be in close agreement with the results obtained by Bokhary *et al.* (1983) who found that natural gum arabic (*Acacia senegal* gum), contained appreciable numbers of microorganisms such as the bacteria *Bacillus sp, Serratia sp* and *Micrococcus sp*. Also the presence results were found to be in agreement with the results obtained by Obied (2002) who stated that raw gum arabic samples did not exceed 1.5×10 cfu/g of coliform bacteria and no *E. coli* was detected and isolates of gram negative rod bacteria were identified as *Salmonella sp, Serratia sp, Klebsiella sp, Pseudomonas sp, Cedecea sp* and *Moraxella sp*.

Also these results agree with the results obtained by Idris (1989) in presence of coliforms and absence of *E. coli* in *Acacia senegal* gum samples, and disagree in absence of *Salmonella* reported by Khalid (1988), Osman (1998) and Ismail (2000) who reported freeness of raw or processed gum arabic from pathogenic bacteria. Also this finding was in disagreement with the results obtained by Osman (1998) who reported that no coliform bacteria were isolated from raw gum arabic samples.
4.1.5 Biochemical identification of bacterial isolates

4.1.5.1 Biochemical identification of bacterial isolates suspected for *Staphylococcus aureus*

Testing for the presence of *Staphylococcus aureus* was carried out on the 36 samples. Thirteen isolates were obtained. Identification and further comprehensive confirmation tests were conducted and the isolates were found to be as in Table 4.5. Out of 13 isolates, eight proved to be *Staphylococcus aureus*.

Table (4.5): Biochemical identification of bacterial isolates suspected for *Staphylococcus aureus*.

<table>
<thead>
<tr>
<th>Isolates</th>
<th>C4</th>
<th>C7</th>
<th>C10</th>
<th>Fb4</th>
<th>Fb13</th>
<th>E9</th>
<th>Aa7</th>
<th>Fb41</th>
<th>Aa9</th>
<th>E10</th>
<th>E7</th>
<th>C5</th>
<th>Fa6</th>
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</thead>
<tbody>
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<td>+ve</td>
<td>+ve</td>
<td>+ve</td>
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<td>Growth anaerobically</td>
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<td>Glucose (acid)</td>
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<td>Acid</td>
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</tbody>
</table>

C4: *Staphylococcus aureus.*  
C7: *Staphylococcus aureus.*  
C10: *Staphylococcus aureus.*  
Fb4: *Staphylococcus aureus.*  
Fb13: *Staphylococcus aureus.*  
E9: *Streptococcus faecium.*  
Aa7: *Streptococcus faecium.*  
Fb41: *Streptococcus faecium.*  
Aa9: *Aerococcus viridans.*  
E10: *Staphylococcus aureus.*  
E7: *Staphylococcus aureus.*  
C5: *Staphylococcus aureus.*  
Fa6: *Streptococcus faecium.*
4.1.5.2 Biochemical identification of bacterial isolates suspected for Bacillus

Checking the presence of Bacillus sp was carried out on the 36 samples and twelve isolates were obtained. Identification and further comprehensive confirmation tests were conducted and the isolates were found to be as shown in Table 4.6. Eleven out of 12 isolates turned out to be Bacillus.

4.1.5.3 Biochemical identification of bacterial isolates suspected for coliform bacteria

Ten isolates were obtained. Identification and further comprehensive confirmation tests were conducted and the isolates were found to be as shown in Table 4.7. Eight out of the ten isolates proved to belong to the genus Klebsiella while the remaining two were Moraxella.

4.1.5.4 Biochemical identification of bacterial isolates suspected for Salmonella

Testing for the presence of Salmonella was carried out on the 36 samples. Fifteen isolates were obtained. Identification and further comprehensive confirmation tests were conducted and the isolates were found to be as shown in Tables 4.8. Nine out of fifteen isolates were found to be Salmonella.
Table (4.6): Biochemical identification of bacterial isolates suspected for *Bacillus*.

<table>
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<tr>
<th>Isolates</th>
<th>Biochemical tests</th>
<th>Ba1</th>
<th>Fb6</th>
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<th>Fa2</th>
<th>E9</th>
<th>Aa2</th>
<th>Aa8</th>
<th>C1</th>
<th>E10</th>
<th>Fa8</th>
<th>Ba3</th>
<th>Fb10</th>
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</tr>
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<tr>
<td>Growth in air</td>
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<tr>
<td>Growth anaerobically</td>
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<td>Glucose (acid)</td>
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<td>Carbohydrates (O/F) test</td>
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</tbody>
</table>

Ba1: *Corynebacterium sp.*  
Fb6: *Bacillus coagulans.*  
Fa10: *Bacillus plymyxa.*  
Fa2: *Bacillus plymyxa.*  
E9: *Bacillus plymyxa.*  
Aa2: *Bacillus plymyxa.*  
Aa8: *Bacillus plymyxa.*  
C1: *Bacillus plymyxa.*  
E10: *Bacillus plymyxa.*  
Fa8: *Bacillus plymyxa.*  
Ba3: *Bacillus plymyxa.*  
Fb10: *Bacillus plymyxa.*
Table (4.7): Biochemical identification of bacterial isolates suspected for coliform bacteria.

| Isolates | Biochemical tests | \( \text{Bal}_1 \) | \( \text{Bal}_{12} \) | \( \text{C}_{10} \) | \( \text{E}_{14} \) | \( \text{Fa}_{102} \) | \( \text{Bal}_{11} \) | \( \text{E}_{5} \) | \( \text{E}_{7} \) | \( \text{E}_{10} \) | \( \text{F}_{9} \) |
|----------|------------------|----------------|---------------|-------------|-------------|-------------|---------------|-------------|-------------|-------------|-------------|-------------|
| Gram stain | -ve | -ve | -ve | -ve | -ve | -ve | -ve | -ve | -ve | -ve | -ve | -ve |
| Shape | R | R | R | R | R | R | R | R | R | R | R | R |
| Spore | - | - | - | - | - | - | - | - | - | - | - | - |
| Catalase test | + | + | + | + | + | + | + | + | + | + | + | + |
| Oxidase test | - | - | - | + | - | - | + | - | - | - | - | - |
| Motility test | - | - | + | - | - | - | - | - | - | - | - | - |
| Growth in air | + | + | + | + | + | + | + | + | + | + | + | + |
| Growth anaerobically | + | + | + | - | + | + | - | + | + | + | + | + |
| Glucose (acid) |
| Acid /gas | Acid /gas | Acid /gas | Acid /gas | Acid /gas | Acid /gas | Acid /gas | Acid /gas | Acid /gas | Acid /gas | Acid /gas | Acid /gas |
| Carbohydrates (O/F) test | F | F | F | F | F | F | F | F | F | F | F | F |
| Urease test | - | + | - | - | + | - | - | - | + | + | + | + |
| MR test | + | - | + | + | - | + | + | + | - | - | - | - |
| VP test | - | + | - | - | + | - | - | - | + | + | + | + |
| Lysine test | + | + | - | + | + | + | + | + | + | + | + | + |
| Citrate test | + | + | - | + | + | + | + | + | + | + | + | + |
| Indole test | - | - | - | - | - | - | - | - | - | - | - | - |
| H₂S test | - | - | - | - | - | - | - | - | - | - | - | - |
| Acid form: |
| Fructose | + | + | + | + | + | + | + | + | + | + | + | + |
| Sucrose | + | + | + | + | + | + | + | + | + | + | + | + |
| Lactose | + | + | + | + | + | + | + | + | + | + | + | + |
| Dulcitol | - | - | - | - | - | - | - | - | - | - | - | - |
| Inositol | - | - | - | - | - | - | - | - | - | - | - | - |
| Sorbitol | + | + | + | - | + | + | - | + | + | + | + | + |

\( \text{Bal}_1 \): *Klebsiella ozaenae.*

\( \text{Bal}_{12} \): *Klebsiella aerogenes.*

\( \text{C}_{10} \): *Klebsiella rhinoscleromatis.*

\( \text{E}_{14} \): *Moraxella sp.*

\( \text{Fa}_{102} \): *Klebsiella aerogenes.*

\( \text{E}_{5} \): *Moraxella sp.*

\( \text{E}_{7} \): *Klebsiella ozaenae.*

\( \text{E}_{10} \): *Klebsiella aerogenes.*

\( \text{F}_{9} \): *Klebsiella aerogenes.*
Table (4.8): Biochemical identification of bacterial isolates suspected for *Salmonella*.

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<th>E2</th>
<th>E9</th>
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<th>Fa4</th>
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</table>

Fb4: *Salmonella* sp.  
E2: *Serratia* sp.  
E9: *Salmonella* sp.  
Ba1: *Serratia* sp.  
Fa4: *Serratia* sp.  
Ba6: *Salmonella* sp.  
Fb13: *Salmonella* sp.  
Ba10: *Salmonella* sp.
Table 4.8 (cont.): Biochemical identification of bacterial isolates suspected for *Salmonella*.

<table>
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<tr>
<th>Isolates</th>
<th>Ba3</th>
<th>E5</th>
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</tbody>
</table>

Ba3: *Salmonella sp.*  
E5: *Salmonella sp.*  
E7: *Cedecea sp.*  
E10: *Pseudomonas sp.*  
Fb5: *Pseudomonas sp.*  
Fb9: *Salmonella sp.*  
C5: *Salmonella sp.*
4.1.6 Identification of moulds and yeasts

All gum arabic samples at different phases of gum production showed that moulds were detected and isolates obtained were identified as *Aspergillus niger*, *Aspergillus flavus*, *Penicillium citreonigrum* and *Curvolaria sp.* (Table 4.9).

The present results of moulds identification were found to be in close agreement with the results obtained by Khalid (1984), North East Wales Institute (NEWI, 1987), Bokhary *et al.* (1983), Anderson and Mc Dougall (1987) and Idris (1989).

Yeasts were not detected in any sample of gum arabic at different phases of gum production (after exudation, after collection, at village market, at crops market, at recent and long storage), Table (4.10). This finding is in agreement with the results obtained by Idris (1989), Osman (1998), Mohamed (2000) and Obied (2002), and disagrees with the results reported by Anderson and Mc Dougall (1987) who stated that natural gum carried a wide range of yeasts, and Ismail (2000) who reported that total viable count of yeast ranged from $0.8 \times 10^2$ to $2.3 \times 10^2$ for crude gum arabic samples.

<table>
<thead>
<tr>
<th>Phases</th>
<th>No. of samples</th>
<th>Yeasts isolates</th>
<th>Moulds isolate</th>
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</thead>
<tbody>
<tr>
<td>AFTER EXUDATION</td>
<td>6</td>
<td>Nil</td>
<td><em>Aspergillus niger</em>, <em>Aspergillus flavus</em> and <em>Penicillium citreonigrum</em>.</td>
</tr>
<tr>
<td>AFTER COLLECTION</td>
<td>6</td>
<td>Nil</td>
<td><em>Aspergillus niger</em>, <em>Aspergillus flavus</em> and <em>Curvolaria sp.</em></td>
</tr>
<tr>
<td>AT VILLAGE MARKET</td>
<td>6</td>
<td>Nil</td>
<td><em>Aspergillus niger</em> and <em>Aspergillus flavus</em>.</td>
</tr>
<tr>
<td>AT CROPS MARKET</td>
<td>6</td>
<td>Nil</td>
<td><em>Aspergillus niger</em> and <em>Aspergillus flavus</em>.</td>
</tr>
<tr>
<td>AT RECENT STORAGE</td>
<td>6</td>
<td>Nil</td>
<td><em>Aspergillus niger</em> and <em>Aspergillus flavus</em>.</td>
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</tbody>
</table>
4.2 Physico-chemical properties

**Physico-chemical properties of 36 samples of gum arabic collected from different locations and different phases of gum arabic production (Tables 4.10, 4.11, 4.12, 4.13, 4.14, 4.15 and 4.16), showed no important change in the mean value of each physico-chemical property at different phases of gum arabic production.**

4.2.1 Moisture content

As shown in Table 4.10 the mean value of moisture content of different phases of *Acacia senegal* (L.) Willd gum samples, after exudation, after collection, at village market, at crops market, at recent storage and long storage, was 23.6%, 22.97%, 16.7%, 15.5%, 13.1% and 11.6%, respectively.

Gum arabic samples after exudation and after collection had high moisture content which probably aids in contamination of gum nodules with sand grains, dusts particles and microorganisms which can easily stick to the gum.

The comparatively high moisture content of gum samples after exudation and after collection in the fields may had also encouraged microbial growth and resulted in the little high microbial count.

Other phases of gum arabic production, i.e., at village market, at crops market, at recent and long storage, showed mean values of 16.7%, 15.5%, 13.1% and 11.6%, respectively.

These values are a little higher than those obtained by Anderson and Stoddard (1966), Awad Elkarim (1994), Osman (1998) and Obied
(2002), but are similar to values specified by JECFA of FAO/WHO (1990).
Table (4.10). Mean values of moisture content (%) of gum arabic samples collected from different phases of gum production.

<table>
<thead>
<tr>
<th>Phases</th>
<th>No. of samples</th>
<th>Minimum</th>
<th>Maximum</th>
<th>Mean</th>
<th>Std. Deviation</th>
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<tr>
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<td>20.46</td>
<td>25.28</td>
<td>22.97</td>
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<tr>
<td>At village market</td>
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<td>15.4</td>
<td>17.8</td>
<td>16.7</td>
<td>1.07</td>
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<tr>
<td>At crops market</td>
<td>6</td>
<td>15.2</td>
<td>15.9</td>
<td>15.5</td>
<td>0.29</td>
</tr>
<tr>
<td>At recent storage</td>
<td>6</td>
<td>12.2</td>
<td>13.7</td>
<td>13.1</td>
<td>0.71</td>
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<tr>
<td>At long storage</td>
<td>6</td>
<td>10.3</td>
<td>12.7</td>
<td>11.6</td>
<td>1.06</td>
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</table>

4.2.2 Ash content

It is clear from Table 4.11 that the mean value of ash content of different phases of *Acacia senegal* (L.) Willd gum samples, after exudation, after collection, at village market, at crops market, at recent and long storage was 3.03%, 3.29%, 3.60%, 3.30%, 3.50% and 3.60%, respectively.

It is noticed that this value is in close agreement with Anderson (1976), Osman *et al.* (1993), Ali (1996) and Osman (1998) who reported that the values of ash content range from 3.01% to 3.9% with mean value of 3.37%, for the commercial samples obtained from Gum Arabic Processing Company.

**Table (4.11). Mean values of ash content (%) of gum arabic samples collected from different phases of gum production.**

<table>
<thead>
<tr>
<th>Phases</th>
<th>No. of samples</th>
<th>Minimum</th>
<th>Maximum</th>
<th>Mean</th>
<th>Std. Deviation</th>
</tr>
</thead>
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<tr>
<td>After exudation</td>
<td>6</td>
<td>2.80</td>
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<td>After collection</td>
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<td>2.89</td>
<td>3.47</td>
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<td>At village market</td>
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<td>3.40</td>
<td>3.80</td>
<td>3.60</td>
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<td>6</td>
<td>2.80</td>
<td>3.60</td>
<td>3.30</td>
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<tr>
<td>At recent storage</td>
<td>6</td>
<td>2.60</td>
<td>3.90</td>
<td>3.50</td>
<td>0.61</td>
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</table>
4.2.3 pH value

The crude gum arabic from *Acacia senegal* is slightly acidic because of the presence of few free carboxyl groups, viz., D-glucuronic acid and its 4-0-methyl derivative.

Table 4.12 gives the mean values of pH for 36 gum arabic samples at different phases of gum production, after exudation, after collection, at village market, at crops market, at recent and long storage which were 4.33, 4.33, 4.33, 4.37, 4.34 and 4.35, respectively. These values are similar to those obtained by Anderson *et al.* (1990), Awad Elkarim (1994), Osman (1998) and Obied (2002).

Table (4.12). Mean values of pH value of gum arabic samples collected from different phases of gum production.

<table>
<thead>
<tr>
<th>Phases</th>
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<th>Maximum</th>
<th>Mean</th>
<th>Std. Deviation</th>
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<tr>
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<td>4.31</td>
<td>4.37</td>
<td>4.33</td>
<td>0.02</td>
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<tr>
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<td>4.31</td>
<td>4.37</td>
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<td>0.03</td>
</tr>
<tr>
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<td>4.37</td>
<td>4.35</td>
<td>0.03</td>
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</table>

4.2.4 Brookfield viscosity

The viscosity is described as an important commercial property of plant gum.

Table 4.13 gives the mean values of Brookfield viscosity for 36 gum arabic samples at different phases of gum production, after exudation, after collection, at village market, at crops market, at recent
and long storage, which were 52.6, 53.87, 50.6, 53.9, 50.6 and 54.6 (cps), respectively.

These values are similar to values obtained by Obied (2002), but are lower than the values obtained by Anderson et al. (1990) who reported that the mean value of Brookfield viscosity of 13 samples of Sudanese gum drawn from season 1904 to 1989 was found to be 78 cps (range from 60 to 100 cps).

Table (4.13). Mean values of Brookfield viscosity (cps) of gum arabic samples collected from different phases of gum production.

<table>
<thead>
<tr>
<th>Phases</th>
<th>No.of samples</th>
<th>Minimum</th>
<th>Maximum</th>
<th>Mean</th>
<th>Std. Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>After exudation</td>
<td>6</td>
<td>43.4</td>
<td>61.5</td>
<td>52.6</td>
<td>5.59</td>
</tr>
<tr>
<td>After collection</td>
<td>6</td>
<td>43.8</td>
<td>62.3</td>
<td>53.87</td>
<td>8.43</td>
</tr>
<tr>
<td>At village market</td>
<td>6</td>
<td>43.8</td>
<td>57.3</td>
<td>50.6</td>
<td>6.06</td>
</tr>
<tr>
<td>At crops market</td>
<td>6</td>
<td>42.0</td>
<td>67.1</td>
<td>53.9</td>
<td>11.45</td>
</tr>
<tr>
<td>At recent storage</td>
<td>6</td>
<td>42.8</td>
<td>55.4</td>
<td>50.6</td>
<td>5.37</td>
</tr>
<tr>
<td>At long storage</td>
<td>6</td>
<td>48.6</td>
<td>61.2</td>
<td>54.6</td>
<td>5.73</td>
</tr>
</tbody>
</table>

### 4.2.5 Specific optical rotation

The aqueous solution of 1% concentration of gum arabic (Acacia senegal) samples were found to be optically active and rotate the plane of polarized light to the left.

As seen in the Table 4.14, the mean values of the specific optical rotation of 36 gum arabic samples at different phases of gum production, after exudation, after collection, at village market, at crops market, at recent and long storage, were -25.8°, -25°, -27.2°, -28°, -29.2° and –26.5°, respectively.
These results were found to be a little higher than the range obtained by Fenyo (1982) and similar to the results reported by Awad Elkarim (1994), Omar et al. (1995), Osman (1998) and Obied (2002).

**Table (4.14). Mean values of optical rotation of gum arabic samples collected from different phases of gum production.**

<table>
<thead>
<tr>
<th>Phases</th>
<th>No.of samples</th>
<th>Minimum</th>
<th>Maximum</th>
<th>Mean</th>
<th>Std. Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>After exudation</td>
<td>6</td>
<td>-22°</td>
<td>-29°</td>
<td>-25.8°</td>
<td>2.32</td>
</tr>
<tr>
<td>After collection</td>
<td>6</td>
<td>-22°</td>
<td>-30°</td>
<td>-25°</td>
<td>2.68</td>
</tr>
<tr>
<td>At village market</td>
<td>6</td>
<td>-22°</td>
<td>-31°</td>
<td>-27.2°</td>
<td>2.43</td>
</tr>
<tr>
<td>At crops market</td>
<td>6</td>
<td>-25°</td>
<td>-29°</td>
<td>-28°</td>
<td>1.5</td>
</tr>
<tr>
<td>At recent storage</td>
<td>6</td>
<td>-25°</td>
<td>-31°</td>
<td>-29.2°</td>
<td>2.14</td>
</tr>
<tr>
<td>At long storage</td>
<td>6</td>
<td>-22°</td>
<td>-29°</td>
<td>-26.5°</td>
<td>3.15</td>
</tr>
</tbody>
</table>

### 4.2.6 Protein content

**Table 4.15 shows the mean values of protein content for 36 gum arabic samples at different phases of gum production, after exudation, after collection, at village market, at crops market, at recent and long storage, were 2.1%, 1.95%, 1.8%, 1.8%, 1.71% and 1.74%, respectively.**

This result is in agreement with the values reported by Anderson (1966), Anderson et al. (1968) and Osman (1998).

**Table (4.15). Mean values of crude protein (%) of gum arabic samples collected from different phases of gum production.**

<table>
<thead>
<tr>
<th>Phases</th>
<th>No.of samples</th>
<th>Minimum</th>
<th>Maximum</th>
<th>Mean</th>
<th>Std. Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>After exudation</td>
<td>6</td>
<td>1.72</td>
<td>2.41</td>
<td>2.1</td>
<td>0.25</td>
</tr>
<tr>
<td>After collection</td>
<td>6</td>
<td>1.69</td>
<td>2.16</td>
<td>1.95</td>
<td>0.17</td>
</tr>
<tr>
<td>Store Location</td>
<td>Mart Number</td>
<td>Price Factor</td>
<td>Total Price</td>
<td>Storage Cost</td>
<td></td>
</tr>
<tr>
<td>-------------------</td>
<td>-------------</td>
<td>--------------</td>
<td>-------------</td>
<td>--------------</td>
<td></td>
</tr>
<tr>
<td>At village market</td>
<td>6</td>
<td>1.7</td>
<td>1.9</td>
<td>1.8</td>
<td>0.08</td>
</tr>
<tr>
<td>At crops market</td>
<td>6</td>
<td>1.7</td>
<td>1.9</td>
<td>1.8</td>
<td>0.09</td>
</tr>
<tr>
<td>At recent storage</td>
<td>6</td>
<td>1.63</td>
<td>1.75</td>
<td>1.71</td>
<td>0.05</td>
</tr>
<tr>
<td>At long storage</td>
<td>6</td>
<td>1.63</td>
<td>1.85</td>
<td>1.74</td>
<td>0.09</td>
</tr>
</tbody>
</table>
4.2.7 Mineral composition

As can be seen from Table 4.16, the mean values of elemental composition from 36 gum arabic samples at different phases of gum production, after exudation, after collection, at village market, at crops market, at recent and long storage, were 0.49%, 0.85%, 0.78%, 0.65%, 0.63% and 0.81%, respectively for Ca$^{++}$; 1.40%, 1.39%, 1.46%, 1.59%, 1.28% and 1.33%, respectively for K$^{+}$; 0.24%, 0.31%, 0.24%, 0.29%, 0.275 and 0.285, respectively for Mg$^{++}$ and 0.04%, 0.04%, 0.04%, 0.02%, 0.09% and 0.05%, respectively, for Na$^{+}$.

This indicates that gum arabic is a good source of Ca$^{++}$ and K$^{+}$. The results are in good agreement with those obtained by Snowden et al. (1987), Idris (1989), Awad Elkarim (1994) and Osman (1998).

**TABLE (4.16). MEAN VALUES OF MINERAL COMPOSITION (%) OF GUM ARABIC SAMPLES COLLECTED FROM DIFFERENT PHASES OF GUM PRODUCTION.**

<table>
<thead>
<tr>
<th>Phases</th>
<th>Parameters</th>
<th>No. of samples</th>
<th>Minimum</th>
<th>Maximum</th>
<th>Mean</th>
<th>Std. Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>After exudation</td>
<td>Ca$^{++}$</td>
<td>6</td>
<td>0.40</td>
<td>0.60</td>
<td>0.49</td>
<td>0.07</td>
</tr>
<tr>
<td></td>
<td>K$^{+}$</td>
<td>6</td>
<td>1.27</td>
<td>1.50</td>
<td>1.40</td>
<td>0.09</td>
</tr>
<tr>
<td></td>
<td>Mg$^{++}$</td>
<td>6</td>
<td>0.12</td>
<td>0.27</td>
<td>0.24</td>
<td>0.06</td>
</tr>
<tr>
<td></td>
<td>Na$^{+}$</td>
<td>6</td>
<td>0.01</td>
<td>0.08</td>
<td>0.04</td>
<td>0.03</td>
</tr>
<tr>
<td>After collection</td>
<td>Ca$^{++}$</td>
<td>6</td>
<td>0.8</td>
<td>0.9</td>
<td>0.85</td>
<td>0.03</td>
</tr>
<tr>
<td></td>
<td>K$^{+}$</td>
<td>6</td>
<td>1.25</td>
<td>1.50</td>
<td>1.39</td>
<td>0.12</td>
</tr>
<tr>
<td></td>
<td>Mg$^{++}$</td>
<td>6</td>
<td>0.27</td>
<td>0.35</td>
<td>0.31</td>
<td>0.03</td>
</tr>
<tr>
<td></td>
<td>Na$^{+}$</td>
<td>6</td>
<td>0.01</td>
<td>0.06</td>
<td>0.04</td>
<td>0.02</td>
</tr>
<tr>
<td>At village market</td>
<td>Ca$^{++}$</td>
<td>6</td>
<td>0.55</td>
<td>0.95</td>
<td>0.78</td>
<td>0.17</td>
</tr>
<tr>
<td></td>
<td>K$^{+}$</td>
<td>6</td>
<td>1.39</td>
<td>1.51</td>
<td>1.46</td>
<td>0.05</td>
</tr>
<tr>
<td></td>
<td>Mg$^{++}$</td>
<td>6</td>
<td>0.15</td>
<td>0.31</td>
<td>0.24</td>
<td>0.05</td>
</tr>
<tr>
<td></td>
<td>Na$^{+}$</td>
<td>6</td>
<td>0.01</td>
<td>0.07</td>
<td>0.04</td>
<td>0.03</td>
</tr>
<tr>
<td>At crops market</td>
<td>Ca$^{++}$</td>
<td>6</td>
<td>0.50</td>
<td>0.75</td>
<td>0.65</td>
<td>0.08</td>
</tr>
<tr>
<td></td>
<td>K$^{+}$</td>
<td>6</td>
<td>1.51</td>
<td>1.61</td>
<td>1.59</td>
<td>0.04</td>
</tr>
<tr>
<td></td>
<td>Mg$^{++}$</td>
<td>6</td>
<td>0.27</td>
<td>0.30</td>
<td>0.29</td>
<td>0.01</td>
</tr>
<tr>
<td></td>
<td>Na$^{+}$</td>
<td>6</td>
<td>0.02</td>
<td>0.02</td>
<td>0.02</td>
<td>0.002</td>
</tr>
<tr>
<td>At recent storage</td>
<td>Ca$^{++}$</td>
<td>6</td>
<td>0.55</td>
<td>0.71</td>
<td>0.63</td>
<td>0.06</td>
</tr>
<tr>
<td></td>
<td>K$^{+}$</td>
<td>6</td>
<td>1.08</td>
<td>1.50</td>
<td>1.28</td>
<td>0.21</td>
</tr>
<tr>
<td></td>
<td>Mg$^{++}$</td>
<td>6</td>
<td>0.26</td>
<td>0.27</td>
<td>0.27</td>
<td>0.006</td>
</tr>
<tr>
<td></td>
<td>Na$^{+}$</td>
<td>6</td>
<td>0.08</td>
<td>0.09</td>
<td>0.09</td>
<td>0.006</td>
</tr>
</tbody>
</table>
CHAPTER FIVE

6. CONCLUSIONS AND RECOMMENDATIONS

All gum arabic samples tested in this study showed no signs of definite microbial spoilage. The maximum microbial load of the samples (after exudation, after collection, at village market, at crops market, at recent and long storage) were found to be about 4.0×10⁴; 2.2×10⁴; 9.0×10³; 8.0×10³ and 4.5×10³ colony forming unit/gram (cfu/g), respectively, for bacterial load, and about 1.7×10⁴; 2.0×10⁴; 8.0×10²; 2.0×10⁴; 2.0×10³ and 6.0×10² cfu/g, respectively, for moulds, and about 7.0×10³; 3.5×10³; 5.0×10³; 6.5×10³; 5.0×10⁴ and 3.1×10⁴ cfu/g, respectively, for spore-formers bacteria. It can be stated that these loads represent safe limits for raw gum arabic.

*Staphylococcus aureus* and *Salmonella*, were detected in samples after collection and other phases of gum production. Coliform bacteria showed a maximum value of 2.4×10³ in samples after collection. This may be due to faecal contamination from soil or human handlers or due to improper hygienic conditions of handling during collection of gum nodules.

*E. coli* and yeast were not detected in any sample tested.

All gum arabic samples at different phases of gum production tested had physico-chemical properties that generally agree with values reported in the literature, except for the gum samples after exudation of gum nodules and after collection in field, which had about 23.6% and

<table>
<thead>
<tr>
<th></th>
<th>Ca²⁺</th>
<th>K⁺</th>
<th>Mg²⁺</th>
<th>Na⁺</th>
<th>6</th>
<th>0.75</th>
<th>0.85</th>
<th>0.81</th>
<th>0.03</th>
</tr>
</thead>
<tbody>
<tr>
<td>At long storage</td>
<td>Ca²⁺</td>
<td>0.75</td>
<td>0.85</td>
<td>0.81</td>
<td>0.03</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ca²⁺</td>
<td>6</td>
<td>0.75</td>
<td>0.85</td>
<td>0.81</td>
<td>0.03</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>K⁺</td>
<td>6</td>
<td>1.25</td>
<td>1.40</td>
<td>1.33</td>
<td>0.06</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mg²⁺</td>
<td>6</td>
<td>0.27</td>
<td>0.30</td>
<td>0.28</td>
<td>0.01</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Na⁺</td>
<td>6</td>
<td>0.05</td>
<td>0.05</td>
<td>0.05</td>
<td>0.0008</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
22.97%, respectively, for moisture content, which values are rather higher than those reported before.

Gum arabic is considered one of the most valuable cash crops. To enhance and improve the quality of this commodity and maintain its markets against gum substitutes and other competing producing countries, a high quality must be ensured. This can be achieved by adopting an effective quality assurance system throughout all phases of gum production from field up to end users by implementation of Hazard Analysis of Critical Control Point System (HACCP) during tapping, collection, transportation, local market, stores, processing plants and export ports. This would help avoid any microbiological or chemical hazard.

Gum arabic as a natural product which toxicologically approved to be used in food and pharmaceutical industries, deserves further studies extension service to improve its hygienic standards especially during tapping, collection and grading.
REFERENCES


Colle, J.G; Duguid, J.p; Fraser, A.G; Marmion, B.P. (1996). Practical Medical Microbiology, 14 the edit.vol2. London.


