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Emulsion-Stabilizing Effect of Gum from *Acacia senegal* (L) Willd. The Role of Quality and Grade of Gum, Oil Type, Temperature, Stirring Time and Concentration

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Abstract: Gum Arabic (*Acacia senegal*) from Kordofan (Central Sudan) and Damazin (Blue Nile, Western Sudan) were used in this study. Physicochemical properties of gum samples were studied (moisture, ash, nitrogen, total soluble fiber, specific rotation, relative viscosity, refractive index and pH). Results show significant differences in moisture content, protein content and relative viscosity between Kordofan and Damazin gums. Damazin gum contained higher protein (3%) and characterized by higher viscosity (24.81) compared to Kordofan gum. Stability of Acacia gum emulsion in regard to type of refined oil (sesame, groundnut, cotton seed, sunflower and corn), temperature, stirring time, concentration and gum grade was also investigated. Results revealed that emulsion stability is significantly affected by the type of oil used. Cotton seed oil gave the most stable emulsion while groundnut resulted in the lowest stable emulsion. Increase in the length of the stirring time is significantly increased stability of the emulsion. Also emulsion stability was affected by gum grades. Other factors of concentration and temperature did not significantly influence emulsion stability.

Key words: *Acacia senegal* gum, quality, emulsion stability, emulsification factors

Introduction

Gum Arabic refers to dried exudates obtained from the stems and branches of *Acacia senegal* (L) Willdenow or Acacia seyal. However, the quantity and quality produced varies between and among species and this determines the type and extent of use by man. A study shows that there are about 17 species producing commercial Acacia gum in Sub-Saharan Africa. Most of these gums are edible and some are used in the food and pharmaceutical industries (FAO, 1996).

The gum from *Acacia senegal* is a water soluble polysaccharide of the hydrocolloid group and comprised mostly of arabinogalactan and protein moiety, in addition to some mineral elements (Williams and Phillips, 2000). It has considerable variation in physicochemical, functional and toxicological properties according to different locations, type of soil and age of the tree (Anderson *et al.*, 1968). Gum Arabic is known by the worldwide food, beverage and pharmaceutical industry as a versatile additive with polyvalent functions: Protective colloid, film-building and coating agent, encapsulating agent, oxidation inhibitor, stabilizer, emulsifier, texturant, clouding and clarifying agent, food adhesive. More recently, western countries discovered that acacia gum is also a dietary fiber with very interesting nutritional properties (NGARA, 2005).

Many food products in the markets are in the emulsion state such as cheese, milk, salad dressings, sauces, beverages and coconut milk (Gonzalez, 1991; McClements, 1999). An emulsion is a dispersed system that consists of two immiscible liquids (usually oil and water), with one of the liquids dispersed as small droplets in the other called continuous phase (McClements, 1999). The emulsions are thermodynamically unstable systems and have a tendency to break down over time (Dickinson, 1992; Friberg and Larsson, 1997; McClements, 1999). The breakdown of an emulsion may manifest itself through different physicochemical mechanisms such as gravitational separation, coalescence, flocculation, Ostwald ripening and phase inversion (Friberg and Larsson, 1997; McClements, 2000). Therefore, the production of high quality food emulsions that can remain kinetically stable for a certain period of time is necessary. In general, emulsifiers are needed for stabilizing emulsions because they decrease the interfacial tension between the oil and water phases and form a protective coating around the droplets which prevents them from coalescing with each other (McClements, 1999).

Dicknison and Sainsby (1988) distinguished between emulsifier and stabilizer in food system. Emulsifier is defined as a single chemical or mixture of components having the capacity for promotion emulsion formation and short term stabilization by interfacial action while stabilizer is defined as a single chemical or mixture of components which can offer long term stability on emulsion, possibly by mechanism involving adsorption but not necessarily so. Certain polysaccharides such as gum arabic (Silber and Mizrahi, 1975), Xanthan gum (Prud and Long, 1983) and Tragancanth gum (Dea and Madden, 1986; Bergenstahl and Steinius, 1986) have been noted to display specific surface activities and stabilize dispersed particles of oil droplets in aqueous system. The gum from *Acacia senegal* has a functional ability to act as emulsifier that stabilizes oil-in-water emulsion (Yokoyama *et al.*, 1988; Randall *et al.*, 1988). It is now known that the protein-rich high molecular mass component adsorbs preferentially onto the surface of the oil droplets. It is envisaged that the hydrophobic polypeptide chains adsorb and anchor the molecules to the surface while the carbohydrate blocks inhibit flocculation and coalescence through electrostatic and steric repulsions (NGARA, 2005). Therefore, the protein moiety had effects on emulsifying behavior of gum Arabic and the best emulsion capacity and stability is found in gums with highest nitrogen content (Randall *et al.*, 1988; Dickinson, 1992). The emulsifying properties of gum Arabic, however, are directly influenced by the botanical type, the nature of the growing soils and the climate (NGARA, 2005). In addition to the emulsifying properties of Acacia gum, other factors related to the emulsification process influence the properties of an emulsion (Brosel and Schubert, 1999).

In Sudan, the gum belt lies within the arid/semi-arid zone of mainly 520,000 km^2 in area that extends across Central Sudan between latitudes 10^o and 14 N, accounting for one fifth of the country's total area (IIED and IES 1990). This belt covers parts of Kordofan, Darfur, Eastern Sudan and Blue Nile and Upper Nile (Hamza, 1990). The vast production area of gum Arabic in Sudan with its variable climatic and edaphic conditions may produce gums with different physical and chemical properties that reflected in their functional behavior. In line with this concept, the present study is designed to compare effects of factors, such as type of oil, temperature, stirring time and concentration on stability of emulsions prepared from gum Arabic samples collected from two environmentally different locations in Sudan.

Materials and Methods

Sample preparation: Dried gum samples from *Acacia senegal* were obtained from the farms of "Acacia Agricultural Company" in Kordofan (Central Sudan) and Damazin (Blue Nile, Eastern Sudan). Gum samples were sorted out into hand picked select intact nodules with purity of about 98% and cleaned fractured nodules (CFG) with purity less than 98%. Then the samples were ground into fine powder to pass 0.4 mm mesh screen. The prepared samples were kept in tight containers and stored at room temperature until analysis. Hand picked select gum (HPSG) was employed in this study.

Moisture, ash, protein and total soluble fiber: Ash and nitrogen (micro-Kjeldahl) were determined according to AOAC (1990). Moisture content of gum samples was determined by drying samples at 105°C overnight (AOAC, 1990). Total soluble fiber was obtained by subtraction of contents of moisture, ash and protein from 100.

pH: pH of 1% aqueous solution of gum (w/v) was measured using a glass electrode pH-meter (HANNA-pH 210).

Specific rotation: Specific rotation of gum samples was measured in a filtered 1% aqueous solution using a polarimeter (Bellingham and Stanley) equipped with a sodium lamp and a cell of 20 cm path length (Abu Baker, 1996).

Relative viscosity: Relative viscosity of gum Arabic samples was measured in filtered 1% aqueous solution using U-shaped viscometer (AOAC, 1990). A flow time (seconds) of distilled water was measured by filling the viscometer tube (held at 30°C in water bath) with water and then drawn by suction to the upper mark of the viscometer. The distilled water was allowed to fall down passing the lower mark of the viscometer. Initial and final times were recorded using stop watch while the water passing the upper and the lower marks of the U-shaped tube. The flow time of a carbon dioxide free aqueous gum solution (2%) was measured as before.

Calculations:

$$\text{Relative viscosity } (30^oC) = [T - T_o]/ [T_o]$$

Where: T, Flow time of gum Arabic solution (1%) in seconds; T_o, Flow time of distilled water in seconds.

Refractive index: Refractive index of gum samples was measured in a filtered 1% aqueous solution using an Abbe refractometer as described by Karamalla *et al.* (1998).

Measurement of stability of emulsions prepared from gum solution and different types of oils: Five types of refined oil (sesame, groundnut, cotton, sunflower and corn oil) and 20% aqueous gum (HPSG) solution were used to prepare stock emulsions. Emulsions were prepared by blending a measured amount of the gum solution (20%) and the oil (2:1 v/v) for one minute at 1800 rpm using kitchen blender (triplicate preparations were made for each study). Aliquot (1 mL) of the stock emulsion was diluted in distilled water to give final dilution of 1/1000. The absorbance was then read at 520 nm in spectrophotometer (Analgue, S104). Another reading of absorbance was recorded after an hour following the same procedure as before (Karamalla *et al.*, 1998). Emulsion stability was calculated as:

$$\text{Emulsion stability} = \frac{\text{First reading of absorbance}}{\text{Reading of absorbance after an hour}}$$

Tests for stability under influence of emulsification factors: These tests were done with the objective of studying effect of emulsification factors of stirring time, temperature, concentration, gum grade and quality, on emulsion stability. The tests for stability were performed in emulsions prepared by mixing the cotton seed oil (Selected as giving the highest emulsion stability) with 20% aqueous gum solution, as mentioned previously (Karamalla *et al.*, 1998).

Stirring time: This test was done to study effect of the length of stirring on emulsion stability. One mL of the stock emulsion was diluted with distilled water to a concentration of 1/1000 and then stirred for different times (1, 2, 3 and 4 minutes) using magnetic stirrer. Emulsion stability was determined following the previous procedure.

Concentration: Distilled water diluted concentrations of stock emulsion solution of 1, 2, 3 and 4/1000 were prepared and examined for emulsion stability under a fixed stirring time of one minute at room temperature. Emulsion stability was measured as before.

Temperature regime: Emulsions of 1/1000 concentration were subjected to four temperature regimes; 50, 100, 150 and 200°C; for 30 minutes and then emulsion stability was measured as before.

Gum grade: Emulsions were prepared by blending 20% aqueous gum (HPSG and CFG) solutions and cotton seed oil in a ratio of 2:1. Determination of emulsion stability was done following the method described by Karamalla *et al.* (1998) as previously stated.

Statistical analysis: Data are means of three samples. An appropriate statistical analysis of variance (ANOVA) was done (Snedecor and Chochran, 1987).

Results and Discussion

Moisture, protein, ash and total soluble fiber: Table 1 shows the contents of moisture, ash, protein and total soluble fiber of *Acacia senegal* gum (HPSG) collected from Kordofan and Damazin. Results indicated that the moisture content of Damazin gum is significantly ($p \leq 0.05$) lower than that of Kordofan. Moisture in both samples is lower than the value reported by FAO (1998). Ash content of both Kordofan and Damazin gums is similar, which falls within the range reported by FAO (1998). Protein contents of Damazin and Kordofan gums were 3% and 1.7%, respectively. Results revealed that the content of protein in samples studied is significantly ($p \leq 0.05$) different, but both values coincided with those found by Karamalla *et al.* (1998). On the other hand, total soluble fiber of Kordofan gum is significantly ($p \leq 0.05$) higher than that of the Damazin gum. The lower content of protein in Kordofan gum may be responsible for the increase in amount of soluble fiber compared to that in Damazin gum. Considering that total soluble fiber was obtained by subtraction.

Table 1: Physicochemical properties of *Acacia senegal* gum collected from Kordofan and Damazin

	Property*	
Acacia gum	Kordofan	Damazin
Moisture (%)	$10.80^a \pm 0.10$	$10.40^b \pm 0.20$
Protein (%)	$1.70^b \pm 0.20$	$3.00^a \pm 0.10$
Ash (%)	$3.40^a \pm 0.1.00$	$3.40^a \pm 0.10$
Total soluble fiber (%)	$84.10^a \pm 1.00$	$82.10^b \pm 0.90$
Specific rotation (°)	$-29.00^a \pm 2.00$	$-29.00^a \pm 1.00$
Relative viscosity	$14.29^b \pm 0.03$	$24.81^a \pm 0.02$
Refractive index	$1.34^a \pm 0.001$	$1.35^a \pm 0.001$
pH	$3.90^a \pm 0.10$	$3.90^a \pm 0.10$

*Means of triplicate samples ± SD. Means having different superscripts within the row are significantly different ($P \leq 0.05$).

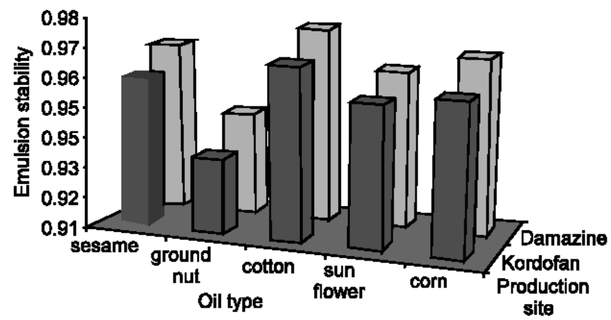

Fig.1: Stability of Kordofan and Damazin gum emulsions as affected by different oil types.

Specific rotation, relative viscosity, refractive index and pH: Results in Table 1 indicated that Kordofan and Damazin gums had similar specific rotation (-29°). This value found within the range reported previously (Anderson *et al.*, 1968; FAO, 1998). Moreover, results illustrated that the relative viscosity (30°C) of Damazin and Kordofan gums were 24.81 and 14.29, respectively (Table 1). These values are significantly ($p \leq 0.05$) different; indicating the more viscous of Damazin gum compared to Kordofan gum and hence reflected the influence of the environmental conditions on some physicochemical properties of the gum produced. Refractive indices and pH of Kordofan and Damazin gums are almost similar, which agreed with those obtained by Karamalla *et al.* (1998).

Effect of emulsification factors on stability of emulsion: Effect of type of oil, temperature, stirring time, concentration and gum grade on emulsion stability is presented in Fig. 1 to 5.

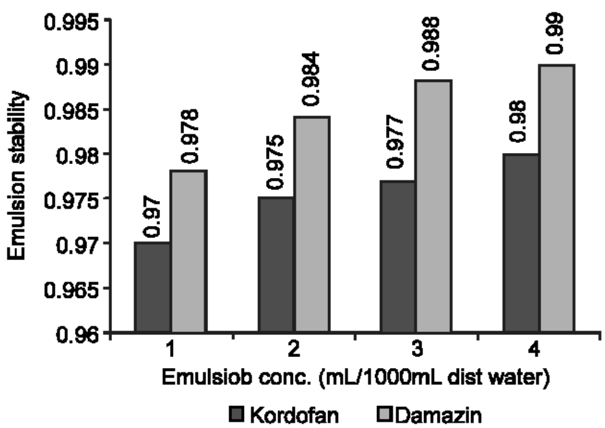

Fig. 2: Effect of varied concentrations on measurement of stability of Kordofan and Damazin gum emulsions

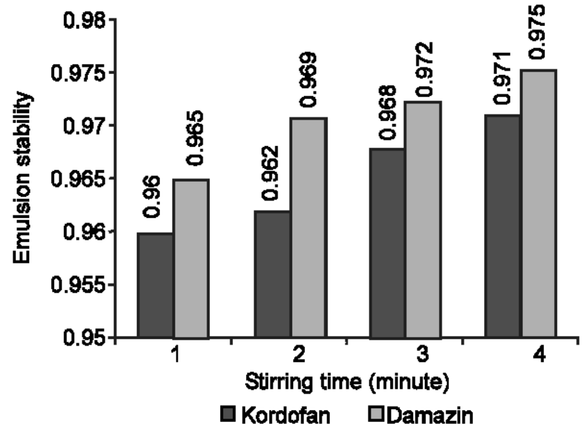

Fig. 4: Stability of Kordofan and Damazin gum emulsions under different stirring times

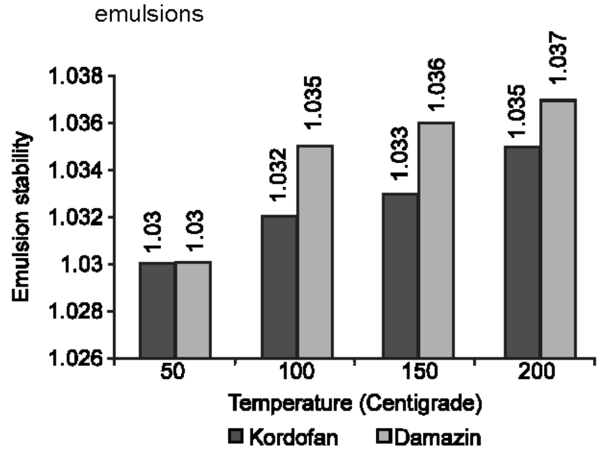

Fig. 3: Stability of Kordofan and Damazin gum emulsion under different heating temperature

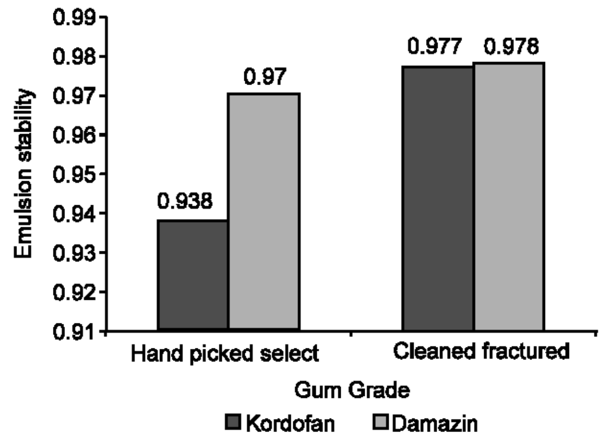

Fig. 5: Effect of gum grade on stability of Kordofan and Damazin gum emulsions.

Oil type: Results show that emulsions prepared by mixing pure oils (sesame, groundnut, cotton seed, sunflower and corn), with aqueous solutions of gums had stabilities varied between 0.935 to 0.967 and 0.944 to 0.974, for Kordofan and Damazin samples, respectively (Fig. 1). However, results indicated that using different types of oils resulted in significant ($p \leq 0.05$) differences in emulsion stability (ES) for both samples of gum. ES when using groundnut oil was the lowest for Kordofan and Damazin gums (0.935 and 0.944, respectively), while that of the cotton seed oil was the highest (0.967 and 0.974, respectively). Damazin gum gave the highest ES compared to Kordofan gum. Differences in ES may be ascribed to difference in protein content (Table 1). Protein is the fraction that provides the functionality of the gum Arabic as emulsion stabilizer. Therefore, the best emulsion capacity and emulsion stability in regard to coalescence and flocculation were recorded in gums with highest nitrogen content (Dickinson *et al.*, 1991). Variability in ES with varied oil type was reported (Pearce and Kinsella, 1978).

Concentration: Fig. 2, illustrates stability of emulsions with concentrations of 1, 2, 3 and 4/1000 for Acacia gum samples. Data revealed an increase, but with insignificant magnitude, in ES with increasing the concentration of the stock emulsion for Kordofan and Damazin gums.

Temperature regime: Emulsion stability at a temperature regime of 50, 100, 150 and 200°C, for both Kordofan and Damazin gums increased, but insignificantly ($p \geq 0.05$), with increasing the heating temperature (Fig. 3). These findings agreed with those reported by Dickinson (1988).

Stirring time: Fig. 4, illustrates that stability of emulsions of Acacia gum, for Kordofan and Damazin, is significantly ($p \leq 0.05$) increased with the increase in stirring time from 1 to 4 minutes. Mechanical blending and homogenization affects droplet-size distribution and hence influences the properties of an emulsion (Brosel and Schubert, 1999). Droplets can be made smaller by applying more intense emulsification giving more stability (Walstra, 1996).

Hand picked selected gum versus cleaned fractured gum: Stability of oil-in-water emulsion of cleaned fractured gums of Kordofan and Damazin is significantly ($p \leq 0.05$) higher than ES from hand picked selected gum samples studied (Fig. 5).

In conclusion, results of this study indicate that location influence the quality of gum produced, which in turn reflected in emulsification properties of the gum. Factors such as oil type, stirring time and gum grade influence emulsion stability.

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