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Moulecular wright distribution and emulsification properties of *Azadirachta indica* (Neem Gum)

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Abstract
Three composite sample collected from different location (White Nile, kordofan, and Khartoum) in the Sudan were subjected to GPC- MALLAS, molecular weight distribution. and emulsification analysis the result show that. The samples have Molar masses of $4.00x10^5$ g.mol^{-1}, – $3.7×10^5$ g.mol^{-1}- $3.8x10^5$ with a radius of gyration (Rg) in the range of (25.6 – 36.6 - 66). respectively. The gum is fractioned into three fractions with different concentrations and different molar masses and different radius of gyration (Rg). The First fraction is an Arabinogalactan protein with molecular weight $3.924x10^5$g.mol^{-1} and an Rg 52.2. The second and third fractions correspond to an Arabinogalactan and a glycoprotein with a total molecular weight of $3.292x10^5$ g.mol^{-1} and mass recovery of 98.66%. Emulsification properties of *Azadirachta indica* gum were examined using droplet size technique as prepared and after accelerated stability test for 3 and 7 days at 60^0C. The results show emulsion droplet size, range 0.01 - 1 microns. Indicating high emulsion stability.

Keywords: gum, neem, molecular weight distribution, emulsion, droplet size

1. Introduction
Two species, *Azadirachta indica* A. Juss. Neem, a member of the family Meliaceae, and in tropical regions. *Philippines and Indonesia.* (Jattan *et al*., 1995) [7] Family Meliaceae found to be distribution (K. Girish *et al*., 2008) [9], Neem tree is a tall tree it has very bright leaves. It has a hard brown grayish bark and it blossoms during spring season with small white colored flowers. Neem can be very easily cultivated in dry and stony soils. All parts of the tree have been used medically for centuries it is used to cure many disease. Neem oil is beneficial to cure most of skin problems. (Sateesh, 1998) [15]. It is also used treating psoriasis and cancer. Traditionally many Indian farmers used neem cake as fertilizers in their fields. (Hegde, 1995) [5] Thus neem tree indeed is a wonderful tree that has many benefits and without any side effect, (Betan, 1972) [3]. (Jhariya *et al*., 2013) [8].
Azadirachta indica gum named as neem gum is natural exudates family is glactan (Aspinal, *et al*., 1996) [1] taking shape bright and amber- colored, tasteless, soluble in cold water, (Srivastava and Rai, 1963) [16] and uses of the *Azadirachta indica* gum has been commercially tapped for using its gum which is of use in large number of industries. (Markhade *et al*., 2006) [12]. It used facial masks, lotions soaps, tooth paste, tooth powder. Antiseptic creams, tablet binders and coaters. It is used as an adhesive for strengthening paper. It is also utilized in dyeing and prating of fabrics. Sometimes it is also used as a stabilizing and thickening agent. The gum cons it of complex polysaccharides and a major component protein. Tightly linked to the polysaccharides, (Mukherjee, *et al*., 1970) [13]. Degradation of the gum complex shows that it contains D- glucose D- glucuronic acid, L– arabinose, L-fucose, mannose and xylose. Investigation of the amino acid composition of the gum shows aspartic acid as most abundant. Amino acid (Anderson, *et al*., 1969) [4].
Several food hydrocolloids exhibit interfacial properties, leading to emulsifying and stable applications. Hydrocolloids stabilize these emulsions through viscosity effects, steric hindrance and electrostatic interactions. High solubility in cold water, low viscosity, high emulsifying capacity and no thickening and/ or gelling effects with aging. (Whitehurst, 2004). The protein which is covalently linked with highly branched polysaccharide structures arabinogalactan protein (AGP) fraction that is critical in establishing the emulsifying properties of the gum (Islam, *et al*., 1997) [6].

The AGP-rich fraction is adsorbed on the oil–water interface; it has been shown to be responsible for the emulsifying properties the gum (Randall, *et al*., 1988) [14]. As a matter of fact, only the AGP molecules adsorb at the surface in an emulsion, whereas the other components have poor affinity for the interface and remain in aqueous phase (Snowden, *et al*., 1987) [17].

2. Material and Methods

2.1 Material

The gum from *Azadirachta indica* fig 1 were collected for Kordofan state, White Nile state, and Khartoum state in Sudan. Season 2016.

2.1.1 Sample preparation

Gum samples were dried at room temperature, hand cleaned to insure purity from sand, dust and bark, ground using a mortar and pestle, and kept in Sealed plastic bags.

Fig 1: *Azadirachta indica* gum

2.2 Methods

2.2.1 GPC-MALLS Analysis

The GPC system comprising a high precision HPLC pump (Waters, USA), an injector (Rheodyne 7125, Rheodyne, UK), a GPC column (Sepharose 6, Pharmacia, Sweden) attached to amulti-angle laser light scattering system (DAWN DSP, Wyatt Technology USA), and UV detector (Pye Unicam, UK), and a software (Astra 4.5 for windows, Wyatt Technology, USA). The system is switched on for two hour to equilibrate. Accurately weighted Gum samples 0.02g in 5ml of 0.2 M were prepared. The samples were kept on a roller shaker for two hours. 100-microlitres of solution were injected into the GPC system (fitted with 100-microlitre loop) via a 0.45µm filter (Water Millipore) the injector was fitted with 100-microlitre loop. Elution buffer, 0.2 M NaCl, was passed at a flow rate of 0.5cm^3/min at ambient temperature. The gum was fraction ated while passing through the Sepharose 6 GPC column and fractions were detected via MALLS, RI and UV detectors. The data was analyzed by Astra software V 4.5 (Wyatt Technology. USA).

2.2.2 Emulsion preparation

Distilled water was added to about sample (based on dry weight) a in glass bottle to give 40 g in concentration 20% (w/w) dry weight was prepared agitated on a tube roller mixer overnight until the sample is, completely dissolved. 20 g of the prepared gum solution were filtered using 100 µm mesh, mixed with 0.52 ml of 10 % (W/V) sodium benzoate solution as a preservative, and 0.48 ml of 10 % (W/V) citric acid solution to adjust the pH to 4. and 15.73ml of distilled water and 4.2 g of ODO oil were added to the gum solution to give a total of 40 g and final concentration of 10%.

The mixed solution was homogenized for 3 minutes using a POLY TRON (PT 2100, KINEMA TICA AC) homogenizer at 22000 rpm. Impeller (PTDA21 9 mm tip diameter) was used as dispersing tool. To achieve small. The pre-twice at 75MPa using a high-pressure Nonvoter (NV30-FA, MITSUBISHI GOT1000.). The emulsion droplet size was measured (Masler size 3000) Malvern instrwrents) as prepared after 3ard 7 days at 60^{0}C in the Vacuum Oven. (GALLENKAMP. OVA031.XX1.5).

3. Results and Discussions

3.1 Molecular Weight and Molecular Weight Distribution

Table 4.1 show the Molecular weight distribution GPC MALLAS show three fraction of the *Azadirachta indica* gum; Arabinogalactan protein (AGP), Arabinogalactan (AG) and Glycoprotein (GP) molecular weight of whole *Azadirachta indica* gum value form the three locations is 4.004x10^5, 3.6x10^5 and 3.7x10^{5}g.mol^{-1} respectively, with radius of gyration 25.6, 36.6 and 66.

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

Figs 2 shows a typical elution profile for *Azadirachta indica* gum monitored using refractive index, response shows three peaks. The first beak AGP is very small mass 1.77, 0.87 and 1.34 for the three samples respectively. The second beak with very high mass is due to AG+GP fraction percent Masses 98.23, 99.13 and 98.66.

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

Table 1: Molecular weight parameters determined by GPC-MALLS

Samples	Mw whole gum (x 10^5)	% Mass recovery	Rg (whole gum)/nm	Mw AGP	% mass (AGP)	Rg-AGP	Mw (AG+GP) (x10^5)	% mass (AG+GP)
White Nile	4.00	120.57	25.6	3.924	1.77	43.2	3.346	98.23
Kordofan	3.6	115.115	36.6	3.916	0.87	52.2	3.292	99.13
Khartoum	3.8	106.675	66	5.086	1.34	63.7	3.139	98.66

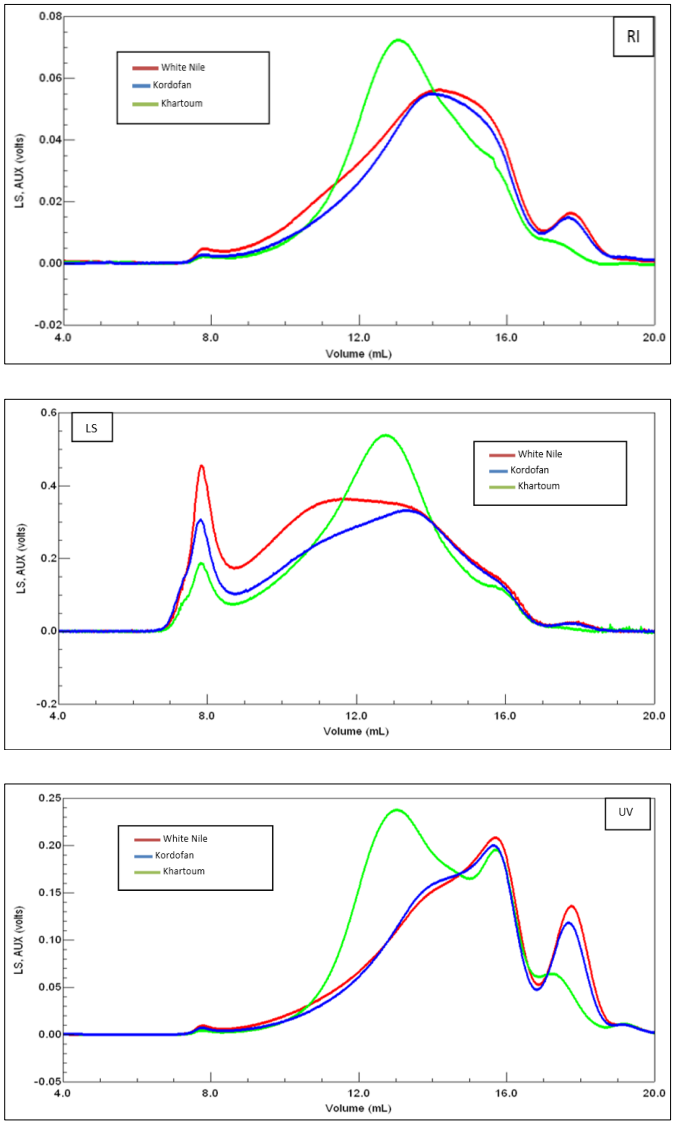

Fig 2: GPC-MALLS elution profile of *Azadirachta indica* gum monitored by (a) refractive index, (b) light scattering and (c) UV at 214 nm

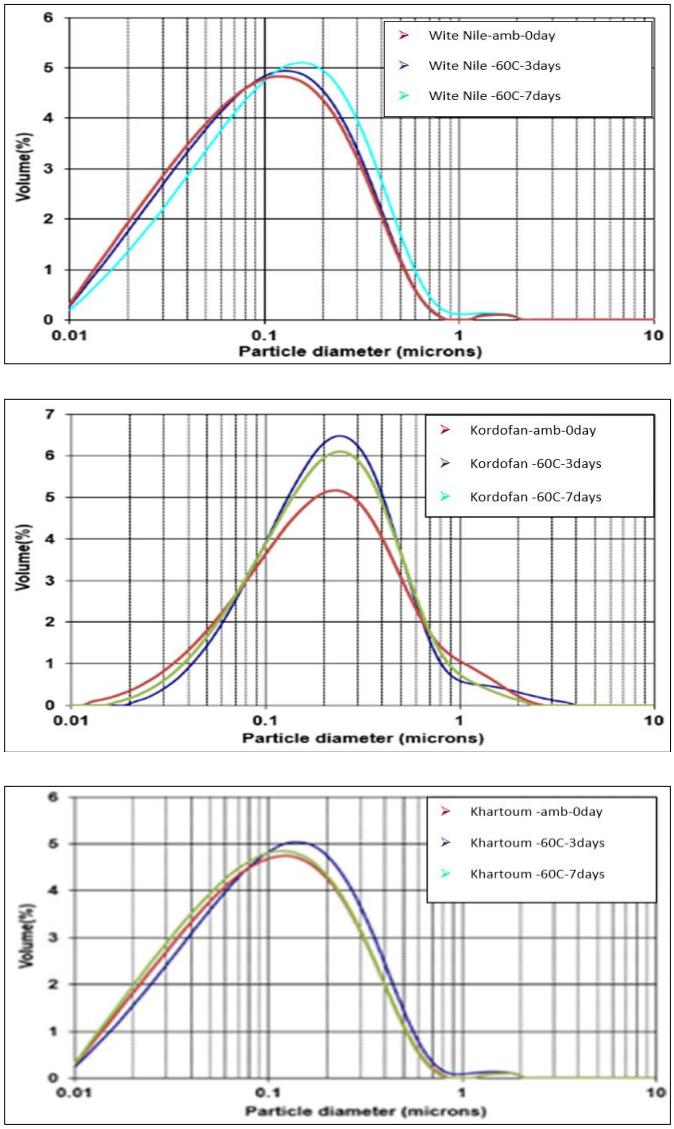

Fig 3: Emulsion particle size distribution from the three states of *Azadirachta indica* gum

3.2 Emulsification properties

Figures 3 show that all of the *Azadirachta indica* gum samples produces very brood peaks with particle diameter size of (0.01 - 1) microns, accordingly, more particle size with different diameter appeared in the emulsions. The more different in particle size lead to coalsence of small particle size and pig particle size together. These ideas can be explain the poor stability of emulsions and the separation of emulsion content is in the experiments.

Figure 3 show particle size distribution for *Azadirachta indica* gum samples for the three location, the practical size diameter lines between 0.01—0.1 micron indicating very high emulsion stability…..the test condition the 7th a similar profited for the gum samples for White Nile and Khartoum states. The high emulsion stability is probably due to high porter composition.

Azadirachta indica gum has higher molecular weight in general and a higher molecular weight of AGP and grate amount of nitrogen content. The short term stability on emulsion may be due to the AGP fraction decrease in the first peak Fig 2(c), it established that there is no direct relationship between the total proportion of the gum and emulsification stability. And the stability of the emulsion is direct relationship to AGP.

4. Conclusion

The molecular weight of the whole *Azadirachta indica* gum is found to be in the range of $3.606x10^5$ - $3.994x10^5$ g.mol^{-1}, with the radius of gyration (Rg) in the range of (25.6 - 66). *Azadirachta indica* gum has a high of protein composition. *Azadirachta indica* gum forms very stable Emulsions.

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6. References

1. Aspinal GO. Advances in carbohydrate chemistry and biochemistry. Wolform M.L., and Tipson R.S Eds., Academic press, New York. 1996; 24:333.
2. Biswas K, Challophyay I, Banerjee RK, Bandyopadyay U. Biological activates and medicine properties of Neem (*azdirachta indica)* curr Sci. 2002; 82:1336-1345.
3. Betan Court A. Growth of *Azadirachta indica* in cuba. Bar-acoa, 1972; 2:17-23.
4. Anderson DMWA. Hendrie, and Munro. J Chromatogr. 1968; 44:178.
5. Hegde NG. Neem and small farmers constraints at grass root level. Indian for. 1995; 121:1040-1048.

6. Islam AM, Phillips GO, Sljivo A, Snowden MJ, Williams PA. A review of recent developments on the regulatory, structural and functional aspects of gum Arabic. Food Hydrocolloids, J Jpn. 1997; 211(3)11:493-505.
7. Jattan SS, Shashikumar, Pujar G, *et al*. Prerspectives in Intensive management of neem plantations. Indian for, 1995; 121:981-988.
8. Jhariya MK, Raj A, Sahu KP, Paikra PR. Neem. A Tree for Solving Global Problem. Indian Journal of Applied Research. 2013; 3(10):1-3.
9. Girlsh K, Bhat S Shankara. Neem-A Green Treasure. Electronic Journal of Biology. 2008; 4(3):102-111.
10. Katayama T, Sasaki Y, Hirose Y, Ogasawara T, Nakamura M, Sakata M, *et al*. Foods Food Ingredients, 2006.
11. Kohli RK, Batish D, Singh HP. Allelopathy and its implications in agro ecosystems. J. Crop prod. 1998; 1:169-202.
12. Markhade DM, Fulzele SV, Satturwar PM, Joshi S. Gum Copal and Gum dammar: novel matrix forming materials for sustained drug delivery Indian J pharma SCI [Internet] [cited 2009 Sep 11]. 2006; 68:53. Available from: *http*:// I Jpsonline. Com.
13. Mukherjee S, Srivastava VN. I solution of pure aldobiuronic acid and aldotriuroic acid from plant gum and mucilage. Indian journal of chemistry. 1970; 8:48-50.
14. Randall RC, Phillips GO, Williams PA. The role of the proteinaceous component on the emulsifying properties of gum Arabic. Food Hydrocolloids, 1988; 2:131-140.
15. Sateesh MK, Sharma G, Hawazhagan, Selvamurthy W. Immunomodulatory effects of neem oil. J. Ethnopharm. 1998; 55:133-139.
16. Srivastava UK, Rai. Psycho-chemical studies on neem gum Dhawa (*Anogessus latifolia*) collid polym. Sci., 1963; 190:140-143.
17. Snowden MJ, Phillips GO, Williams PA. Functional characteristics of gum Arabic. Food Hydrocolloids, 1987; 1:291-300.