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RHEOLOGICAL AND EMULSIFYING PROPERTIES OF SOME NATURALLY OCCURRING POLYSACCHARIDES II: POLYSACCHARIDE IN IRVINGIA GABONENSIS, IRVINGIACEAE

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INTRODUCTION

The work reported in this paper is a part of the screening for useful naturally occurring emulsifying and thickening agents.

Irvingia gabonensis, Irvingiaceae var. excelsia is an evergreen tree found in the rain forests of Nigeria. It extends from Senegal to the Sudan and Southwards to Angola (Keay, Onochie and Stanfield, 1964). The cotyledons are used primarily as thickner in food. This gum or polymerous substance has been shown to contain a reasonably high amount of protein (Irvine, 1961). The polymerous substance seems to aid the dispersion of its high concentration of fixed oils in water. The oil from the seed has been described as being superior to cocoa butter and is often used as adulterant in confectionery (Dalziel, 1948). Already, there is indication that this oil might be a good suppository base superior to cocoa butter (Udeala and Ebiennang, 1977).

In the present investigation, the suitability of the polysaccharide in the edible variety as an emulsifier was investigated using acacia as basis for comparison.

Experimental

Defatting of Seeds:

The decorticated pulverised seeds were soxhlet extracted for 24 hours using n-hexane (Merck) with a boiling range of 60-80°C. The defatted seed flour was air-dried.

Removal of Protein:

Papain (Hopkins and Williams) in fine powder, cetyltrimethyl ammonium bromide or CTAB (Merck), ethanol (Merck), sodium chloride (Fisher), Titriplex a brand of disodium ethylenediamine tetraacetate (Merck), cysteine hydrochloride (B.D.H.) and serum albumin (Merck) were used as supplied by the manufacturers.

The digesting aqueous medium contained 0.001 M Titriplex, 0.005 M cysteine hydrochloride and 0.5%w/w to 1% w/w papain by weight of material to be digested. This medium is similar to that suggested by Spiro (1966).

A portion of the defatted seed flour was dispersed in distilled water in which Titriplex has been dissolved. The pH was adjusted to 6.5 and the dispersion was warmed to 70°C. To activate the papain, its dispersion in distilled water containing cysteine hydrochloride was adjusted to pH 6.5 and warmed at 70°C for 30 minutes. The choice of temperature of digestion was determined by an investigation carried out by Osuoji (1968) who found that papain is most effective at this temperature. Digestion was carried out for 48 hours.

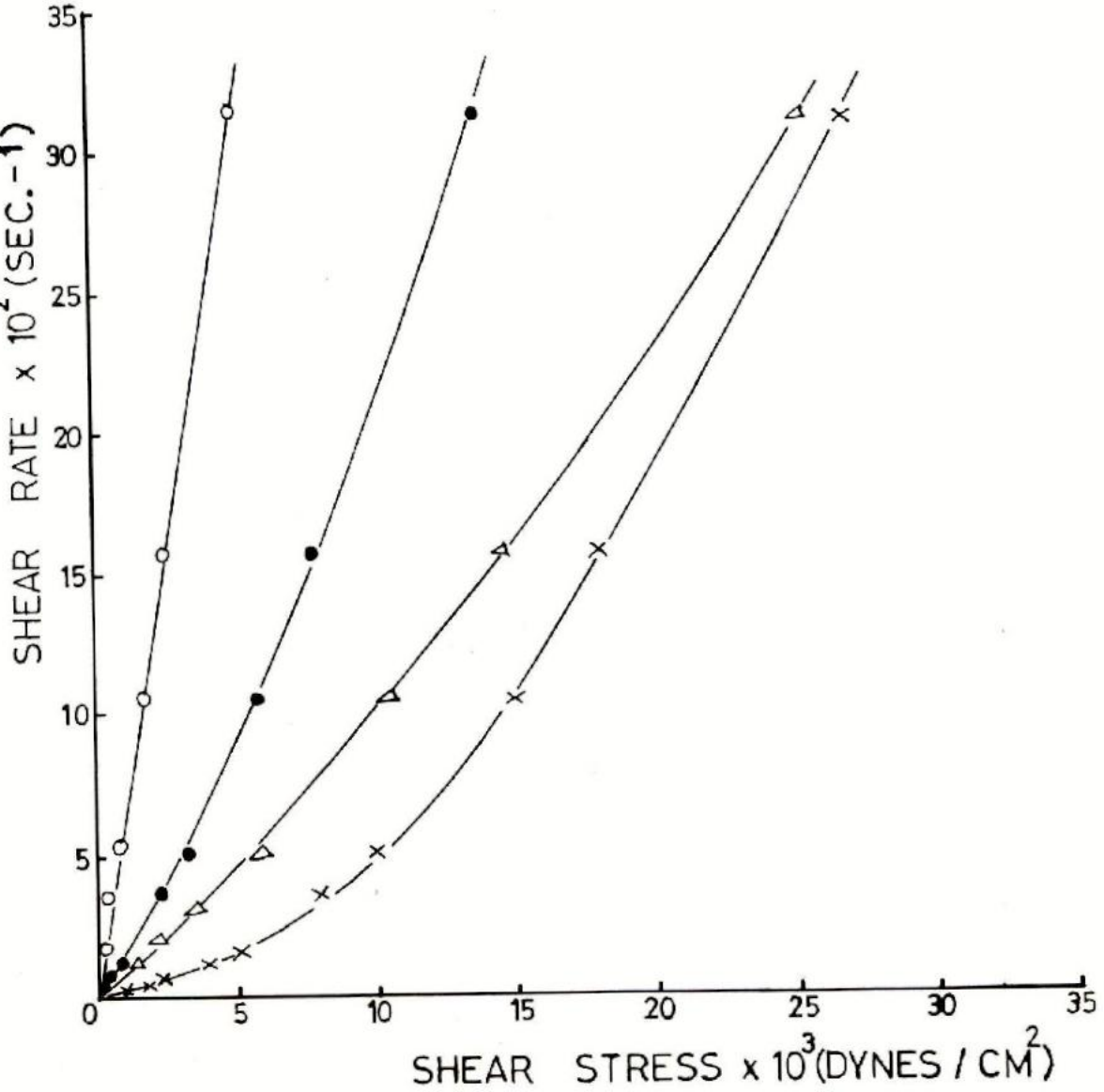
Having removed the undigested fibrous matter by centrifugation at 3000 r.p.m., the polysaccharide in a portion of the supernatant was precipitated out in 95% w/v ethanol. The precipitate was then washed with alcohol. The polysaccharide in the other portion of the supernatant was complexed with CTAB (Scott, 1965), while the protein remained in solution. The flocculent polysaccharide-CTAB complex precipitate, was removed by centrifugation at 3000 r.p.m. The CTAB in the complex was exchanged with sodium ion by dissolving the complex in 2 M sodium chloride. The sodium salt of the polysaccharide was precipitated in 95% w/v ethanol. The resulting precipitate was repeatedly dissolved in sodium chloride and re—precipitated in ethanol. In this manner, the polysaccharide sodium salt was freed from the detergent. The polysaccharide product was dissolved in distilled water and dialysed against tap water for 24 hours. This ensured the removal of excess sodium chloride.

From the dialysed aqueous dispersion, the polysaccharide was precipitated in alcohol. The solid recovery by filtration was dried in a vacuum dessicator containing calcium chloride as dessicant. Using serum albumin as standard, the protein content of the product was determined by the Lowry method for protein estimation.

Rheological Measurement:

Preparation of mucilages: The defatted seed flour was first passed through a 200 mesh sieve. Mucilages were prepared using either the seed flour gum precipitated in alcohol after the removal of fibrous matter by centrifugation of deproteinised and precipitated gum. In each case, the weighed quantity of material was placed in one-half the volume of the required quantity of distilled water, allowed to hydrate for 24 hours and then made up to volume. The acacia mucilage was prepared in the conventional manner. Water used in all preparations contained 0.5% w/v benzoic acid as preservative. Mucilage of acacia contained 12.5% w/v of the gum while concentration of Irvingia gum in the mucilages was varied from 0.02% w/v to 4% w/v.

Fig. 1


SHEAR RATE x 10^2 (SEC.$^{-1}$)
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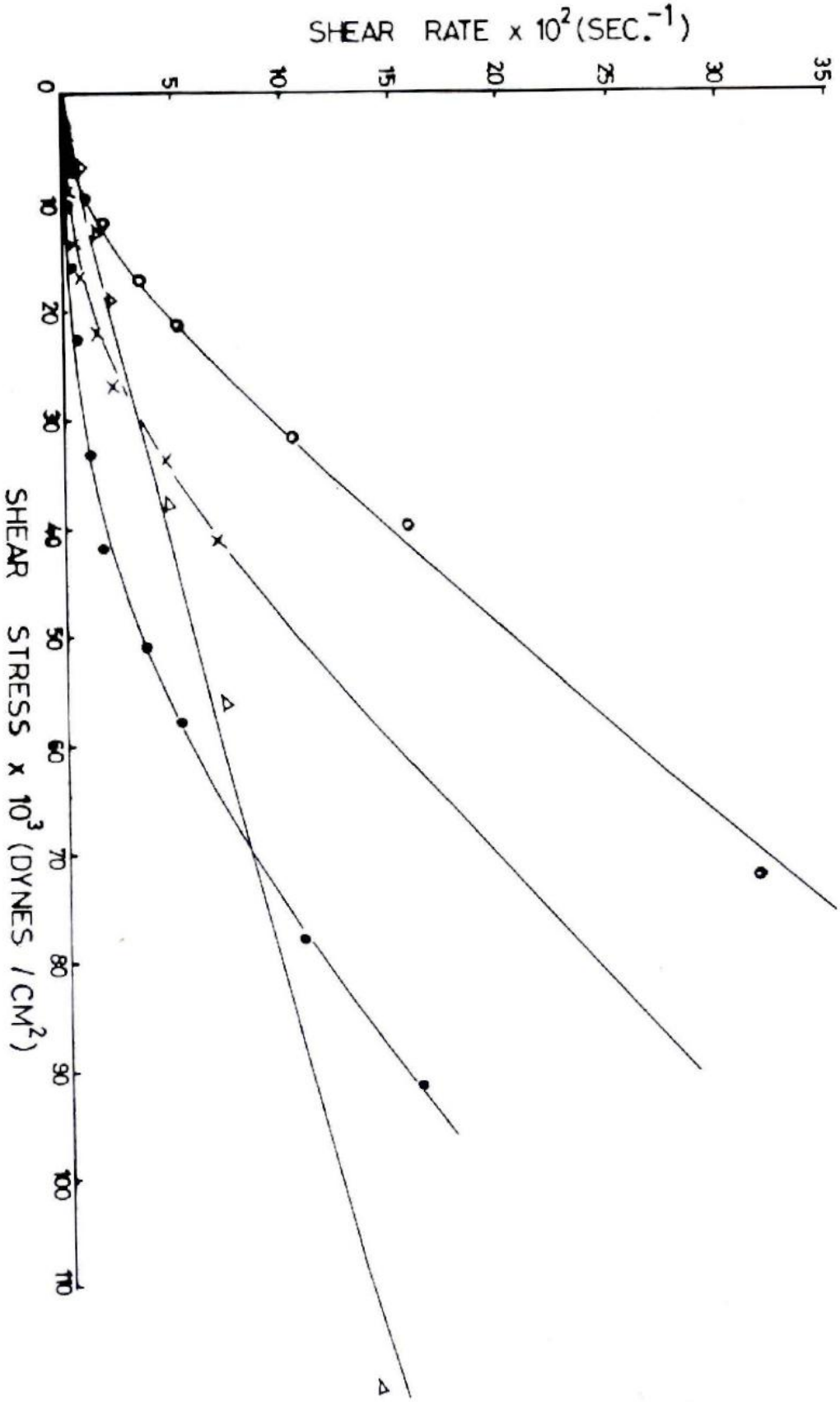
SHEAR STRESS x 10^3 (DYNES / CM^2)

Fig. 2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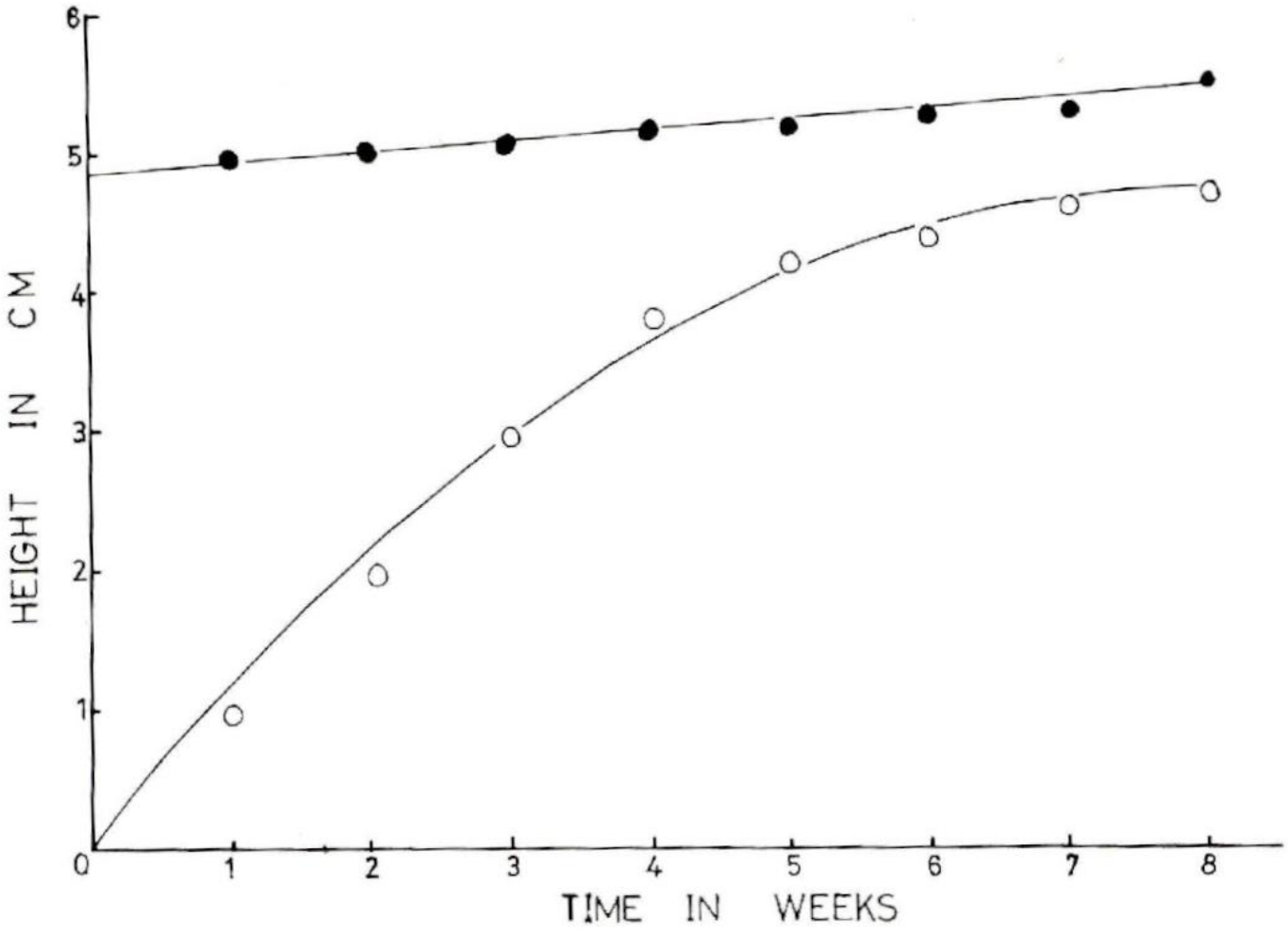

Fig. 3 The degree of phase separation in Emulsions prepared with heat treated and untreated Irvingia seed flour

Preparation of Emulsions:

A method similar to that used in preparing emulsions with Raphia gum (Chiori and Udeala, 1977) was adopted. This involved initial blending of oil, water and gum with Silverson mixer and subsequent passage through a hand homogeniser. In an attempt to obtain enhanced stability, a mucilage was first prepared using the seed flour after autoclaving for 20 minutes at 121°c. The Oil was then blended in and the emulsion made up to the required volume.

Emulsions were formulated using liquid paraffin and water in 4:6 oil—water ratio. The deproteinised and precipitated gum, the ordinary precipitated gum and the seed flour were used in trial formulations. Concentration of the gums were varied from 0.1% w/v to 5% w/v. Liquid paraffin emulsion was also prepared using 12.5% w/v acacia as amulgent. Distilled water used in all preparation contained 0.5% w/v benzoic acid as a preservative. The extent of cracking or creaming of these emulsions were observed over a period of 10 weeks.

Apparatus and Method:

For rheological measurements, the Searle sensor system in a concentric-cylinder viscometer by Haake Instruments were used. This and the manner for obtaining data have been fully described in previous work (Chiori and Udeala, 1977).

RESULTS AND DISCUSSION

Preliminary Studies:

Kjedahl nitrogen analysis showed that the defatted seed flour contained 4.27% w/w nitrogen (27% w/w crude protein). Treatement of the seed flour with liquified phenol only reduced the protein content to 18% w/w. Enzyme digestion with papain proved a more effective method of deproteinization. The protein content was reduced to 4.5% w/w. The presence of this level of protein in the emulgent was not considered unsatisfactory since proteins can add to the value of cosmetic preparations such as lotions (De Navarre, 1962). From these results, it can be inferred that the suspected emulgent is firmly bound to the protein moiety since enzymic digestion was the more effective method of protein removal.

Initially, varying concentrations of the emulgent were tried in emulsion formulation. For the heat treated and untreated gum, concentration of 1% w/v to 4% w/v were found to yield creamy white emulsions. Consistency varied with concentration of emulgent. 1% w/v of the deproteinised and precipitated emulgent yielded the best emulsion while the other preparations of the emulgent yielded relatively good emulsions at higher concentrations.

Rheology and Stability of Preparations:

Mucilages containing 0.5% w/v to 4% w/v of each of the three types of gums were rheologically examined. These preparations in general exhibited pseudoplastic behaviour. Since mucilages prepared with the highly purified gum gave high instrument readings, lower concentrations were examined. Pseudoplastic behaviour disappeared at concentrations lower than 0.05% w/v. The graph of shear stress against shear rate is presented in Fig. 1. It can be seen that the highly purified gum assumes Newtonian behaviour at a concentration of 0.02% w/v. This has been confirmed by capillary viscometry. Reproducible flow patterns were obtained at this concentration. With the same concentration of the gum to which large amount of protein was still attached, the flow pattern was erratic (Udeala and Attwood, 1977). Higher concentrations of the highly purified gum and the other forms are pseudoplastic. The rheogram for 12.5% w/v acacia mucilage has been included in Fig. 1 for comparison. Mucilage prepared with the highly purified gum proved the more stable than the others, but less stable than acacia mucilage.

The emulsions prepared with different preparations of the emulgent are pseudoplastic. The most extreme is that prepared with 1% w/v of the highly purified gum (Fig. 2). The least pseudoplastic was emulsion prepared with 2% w/v of the seed flour. Indeed higher concentration of the seed flour yielded plastic emulsion. This is probably due to high concentration of suspended fibrous matter. The rheograms of emulsions prepared with 12.5% w/v acacia and 1% w/v precipitated gum respectively have been included in Fig. 2 for comparison.

The emulsion prepared with different forms of the Irvingia gum were basically less stable than acacia emulsion. In Fig. 3, it can be seen that a greater stability was achieved in those emulsions prepared with heat treated emulgent. Emulsions prepared with untreated emulgent attained maximal separation of phases in one week. In emulsions prepared with treated seed flour, maximal separation appeared in 8 weeks. Heat treatment is often instituted to inactivate enzymes present in seed gums and thus enhance stability of emulsions (Chudzikowski, 1971). Increased stability might have been achieved in the Irvingia emulgent by denaturation of the proteins which might be enzymic in nature. Emulsion prepared with 1% w/v concentration of the deproteinised gum was most stable. After 8 weeks, cracking was preceeded by a gradual creaming process. The stability achieved in this indicates that the protein is one major contributor to the instability of this gum.

Even with reduction of the protein content from 27% to 4.5%, stability of the Irvingia gum emulsion could not equal the stability of acacia emulsion. This is indicative that the highly purified Irvingia gum is not as an efficient emulsifier as acacia gum. In all preparations using this new gum, colour and odour developed with time.

SUMMARY

The macromolecular substance contained in **Irvingia gabonensis** seeds has been investigated for its emulsifying potential along the same line as Raphia gum (Chiori and Udeala, 1977). It was found that removal of fibrous matter and reduction of the protein content from 27% to 4.5% accorded some stability to the suspected emulgent. Emulsions prepared with it were found to be much less stable than acacia emulsion. The polymerous substance may not be an effective emulsifier but its remarkable ability to increase viscosity warrants further investigation for possible use as a blood extender.

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LEGEND FOR FIGURES

Fig 1: Rheogram of dispersions containing • 1% w/v seed flour: × 1% w/v gum co-precipitated with protein; O 0.02% w/v deproteinised and precipitated gum and Δ 12.5% w/v acacia.

Fig. 2: Rheogram of emulsions prepared with • 1% w/v deproteinised and precipitated gum • 2% w/v seed flour. × 1% w/v gum co-precipitated with protein and Δ12.5% w/v acacia.

Fig. 3: The degree of phase separation in emulsion prepared with 2% w/v • heat treated and • untreated gum.