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Composition and Physicochemical Properties of Linseed (*Linum usitatissimum* L.) Mucilage

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Linseed (*Linum usitatissimum* L.) mucilage, consisting mainly of water-soluble polysaccharides, was isolated from the seeds and a partially defatted meal by different extraction regimes. The mucilage yield (3.6–9.4%) and level of contaminating proteins varied substantially with the temperature of extraction and nature of the raw material; lower yields of relatively pure polysaccharide extracts were obtained from the seeds at 4 °C. Although the relative monosaccharide composition varied with the extraction conditions, galacturonic acid, galactose, xylose, and rhamnose were the major monosaccharides; fucose, arabinose, and glucose were minor constituents. Purified mucilage was further fractionated into mainly neutral and acidic polysaccharides by selective precipitation with cetyltrimethylammonium bromide (it complexes preferentially with polysaccharides of high charge density), as revealed by galacturonic acid analysis and ^{13}C NMR. The neutral polysaccharide fraction, having a higher intrinsic viscosity ([η] = 6.6 vs 4.6 dL/g), exhibited more pronounced shear thinning and viscoelastic responses in solution than its acidic counterpart. All mucilage preparations exhibited stable viscosity in the pH range 5.0–9.0; however, large reductions in viscosity were seen with the addition of electrolytes. The water binding capacity (1600–3000 g of H_2O/100 g of solids) and rheological properties of mucilage resembled those of guar gum.

INTRODUCTION

Linseed mucilage is a gumlike material associated with the hull of linseed and comprises about 8% of the seed weight (Bhatty and Cherdkiatgumchai, 1990; BeMiller, 1973). It is primarily a mixture of polysaccharides which, on acid-catalyzed hydrolysis, yield rhamnose, fucose, arabinose, xylose, galactose, galacturonic acid, and glucose (Erskine and Jones, 1957). Bailey (1935) was the first to separate linseed mucilage into acidic and neutral polysaccharide fractions, while structural analysis of the mucilage was carried out by Erskine and Jones (1957), Hunt and Jones (1962), and Muralikrishna et al. (1987).

Mason and Hall (1948) have reported on the use of linseed mucilage as an emulsifying agent for chocolate milk. BeMiller (1973) also concluded that, functionally, linseed gum resembles gum arabic more closely than any of the other common gums. In this reference the effects of concentration, pH, and temperature on the rheological properties of linseed mucilage solutions were discussed.

There has been increased interest recently in the physical and functional properties of linseed mucilage (Susheelamma, 1987, 1989; Mazza and Biliaderis, 1989; Wannerberger et al., 1991). However, there is limited information on the influence of extraction and purification methods on yield, composition, and properties of this gum material. Moreover, while the composition and structural analysis of its acidic and neutral polymeric constituents have been investigated, rheological and other physical properties of these polysaccharides were not studied in detail. The aim of the present work was to examine the effect of different extraction/purification methods on yield, purity, and monosaccharide composition of linseed mucilage and its polymeric constituents as well as to evaluate the physicochemical properties of these polysaccharides in solution. Such studies are useful in exploring the full potential of linseed mucilage as a hydrocolloid preparation for food and nonfood applications.

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MATERIALS AND METHODS

The linseed used was of the cultivar Linott obtained from the 1989 harvest at Agriculture Canada Research Station, Morden, MB. Guar gum and Celite were obtained from Sigma Chemical Co. (St. Louis, MO). Xanthan gum was an agglomerated product of Zumbro, Inc. (Hayfield, MN), while gum arabic was obtained from Fisher Scientific (Winnipeg, MB). Partially defatted linseed meal was obtained from United Grain Growers (Winnipeg, MB). Vega clay was a product of Pembina Mountain Clays (Winnipeg, MB). All other chemicals were of analytical reagent grade.

Extraction. Water-soluble polysaccharides were extracted from linseed by mixing the seed with water (1:20 w/v), stirring the seed–water mixture for 2 and 24 h at 4, 25, 100–25 (100 °C at start of extraction, allowing to cool to 25 °C), and 80 °C, and separating the mucilage extract from the seed by filtration through a 40-mesh screen. Water-insoluble material was removed by centrifugation on a model RC2-B Sorvall refrigerated centrifuge (10000*g*, 30 min, 4 °C). The water-soluble extract was dialyzed (12 000–14 000 molecular weight cutoff (MWCO), 72 h, 4 °C) and then concentrated on a rotary evaporator at 60 °C and lyophilized or precipitated with 3 volumes of ethanol. Subsequent extractions of linseed were carried out with 0.5% (w/v) ammonium oxalate (80 °C, 1 h) or 0.1 M Na_2CO_3 (4 °C, 18 h). Water-soluble polysaccharides were extracted from linseed meal in a similar manner, with the following modifications: linseed meal–water slurries (1:40 w/v) were stirred for 30 min at 25–80 °C. Following hot-water extraction of mucilage from seed and removal of water-insoluble material, deproteinization was accomplished by treatment of selected extracts with Vega clay, an acid-activated class adsorbent (Figure 1).

Fractionation. The lyophilized material obtained by extraction from linseed (2 h, 80 °C) was fractionated according to the procedure of Hunt and Jones (1962) with the following modifications: a 0.5% (w/w) mucilage solution (2000 mL) was prepared in distilled water, and 100 mL of aqueous 10% (w/v) cetyltrimethylammonium bromide (CTAB) was added slowly dropwise under continuous stirring at 30 °C. The solution was then allowed to stand for 18 h at room temperature, and the precipitate was removed by centrifugation (10000*g*, 30 min, 4 °C). The supernatant was dialyzed (12 000–14 000 MWCO, 72 h, 4 °C) until no further carbohydrates were detected in the dialysate. Some material that precipitated during dialysis was removed by centrifugation, lyophilized, and washed with ethanol to remove excess CTAB; this material is designated CTAB

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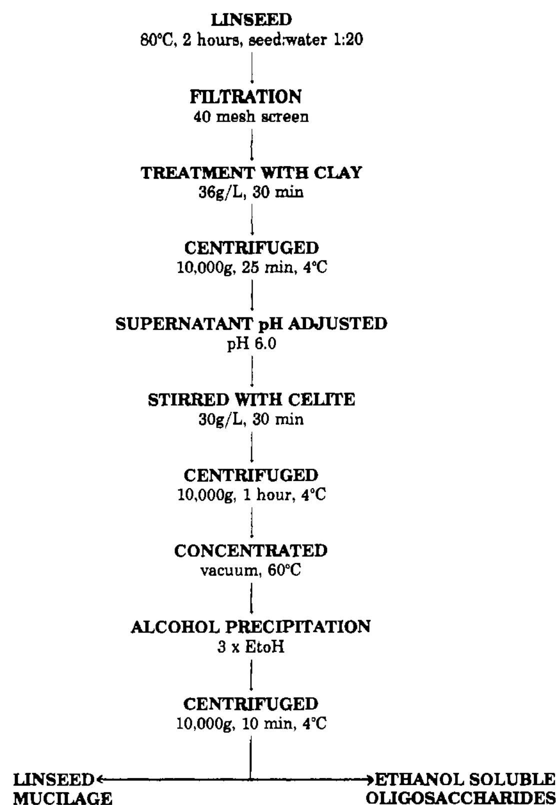

Figure 1. General procedure for extraction and purification of linseed mucilages from linseed.

nonprecipitated insoluble fraction. The CTAB nonprecipitated soluble material was lyophilized and washed with ethanol. The CTAB precipitated material was washed with distilled water and dissolved in 10% (w/v) NaCl solution by overnight stirring to dissociate the complex of CTAB–acidic polysaccharides. The solubilized material (CTAB precipitated soluble) was then separated from the insoluble portion (CTAB precipitated insoluble) by centrifugation. Both of these fractions were treated with 3 volumes of ethanol, redissolved in distilled water, dialyzed, and lyophilized.

Cold-water-extracted mucilage from seed (24 h, 4 °C) was fractionated in a manner similar to the above, with the following modifications: a 0.5% (w/w) solution of mucilage was prepared in 5 L of 0.02 M Na_2SO_4 solution, and 300 mL of 10% (w/v) CTAB was added. The pH was adjusted to 6.0, and the mixture was incubated at 37 °C overnight. The CTAB precipitated material (CP) was removed by centrifugation and further treated as above. Celite (30 g/L) was added to the CTAB nonprecipitated material (CS) and stirred for 30 min. The water-insoluble material was removed by centrifugation, and the supernatant was dialyzed and lyophilized. The lyophilized material was dissolved in 10% (w/v) NaCl to a final concentration of 1% (w/w) and then precipitated with 4 volumes of ethanol and allowed to stand overnight. The precipitate was removed by centrifugation, dissolved in distilled water, dialyzed, and lyophilized.

Chromatography. Gel permeation chromatography of linseed mucilage and its fractions was performed on Sepharose CL-2B (2.5 × 95 cm) by elution with degassed 0.1 M sodium acetate buffer, pH 4.0, or 0.1 M sodium acetate buffer, pH 4.0, in 6.0 M urea at a flow rate of 35 mL/h at room temperature. Samples (10 mg in 5 mL of buffer) were applied to the column, and fractions of 6 mL were collected. The fractions were analyzed for total carbohydrates by the phenol–sulfuric method (Dubois et al., 1956), proteins by the Lowry et al. (1951) procedure, and uronic acids by the Blumenkrantz and Asboe-Hansen (1973) procedure. The void and total volumes were determined with elution of blue dextran and xylose, respectively.

Chemical Analysis. Galacturonic acid concentration in linseed mucilage and its fractions was determined according to the method of Blumenkrantz and Asboe-Hansen (1973). Protein content of the polysaccharide preparations was measured according to the method of Lowry et al. (1951). Methyl ester content of the acidic polymers was assessed by the method of Klavons and Bennet (1986) and total ash according to standard procedures (AOAC, 1975). Monosaccharide composition was determined by gas–liquid chromatography following the procedure of Englyst et al. (1982) with some modifications. The alditol acetate derivatives of sugars in the acid hydrolysates were separated on a SP-2330 glass capillary column, 30 m × 0.75 mm i.d., in a Model 3700 Varian gas chromatograph equipped with a flame ionization detector (injector temperature 240 °C, column temperature 150 °C, detector temperature 240 °C); the chromatographic run involved isothermal heating at 150 °C (1 min) followed by heating to 220 °C at 10 °C/min. Peak area integration was obtained with a Model 3390A Hewlett-Packard integrator. The values of fucose, arabinose, xylose, and galactose were calculated after hydrolysis for 2 h in 1.0 M H_2SO_4. The value of rhamnose was calculated after hydrolysis for 6 h. The value of glucose was determined after hydrolysis for 2 h in 1.0 M H_2SO_4, following solubilization/prehydrolysis in 12.0 M H_2SO_4 at 35 °C for 1 h.

^{13}C NMR. Natural abundance ^{13}C NMR spectra (75.9 MHz) were recorded at 85 °C on a Bruker AM 300 FT spectrometer. CS and CP fractions (2% w/w) were dissolved in D_2O. Number of acquisitions was approximately 30 000. Pulse repetition time was 1.245 s and the radio frequency pulse angle 90.0°; the frequency for proton decoupling was 5000 Hz. Chemical shifts (δ) are expressed in parts per million downfield from external Me_4Si but were actually measured by reference to internal 1,4-dioxane (δ = 67.4 ppm) (Izydorczyk and Biliaderis, 1992).

Rheological Measurements. Limiting viscosity measurements of CS (0.03–0.1% w/v) and CP (0.04–0.12% w/v) polysaccharide fractions of linseed were performed using Ubbelohde viscometers (International Research Glassware, Kenilworth, NJ) at 25 ± 0.05 °C. The reduced viscosities (using 0.2 M NaCl as solvent) were plotted against polymer concentration and extrapolated to zero concentration to obtain the intrinsic viscosities (Huggins, 1942).

A Bohlin VOR rheometer (Bohlin Reologi, Edison, NJ) was used to determine steady shear viscosity as a function of shear rate. Measurements were taken using a concentric cylindrical geometry; the radii of the inner rotor and outer container were 25.0 and 27.4 mm, respectively. The samples were subjected to shear sweeps between 2 and 735 s^{-1}. The reported results are averages of three replicates; the coefficient of variation was less than 5% of the mean values in all cases. Viscosity measurements were carried out using aqueous solutions of 0.1–2.0% (w/w) mucilage and its fractions (pH 5.5). Changes in viscosity of 1.0% (w/w) mucilage and fractions due to pH variation, within 2–12 (adjusted with HCl or NaOH), electrolyte concentration (NaCl or $CaCl_2$), 0.005–0.5 M, and temperature, 15–65 °C, were also examined.

The dynamic rheological properties of concentrated solutions were also assessed with the Bohlin VOR rheometer operated in the oscillatory mode. All measurements were conducted at 25.0 ± 0.1 °C using either a 19.3 or 93.2 g·cm torsion element. Small-amplitude oscillatory measurements were performed at 0.05–20.0 Hz and less than 4.0% strain. Values for the storage (G') and loss (G'') moduli, as well as dynamic viscosity (η'), were obtained using the software analysis programs of the rheometer.

Water Binding Capacity. For water binding capacity measurements, guar gum and xanthan gum were used for comparative purposes. Freeze-dried mucilage samples were first dissolved in distilled water and precipitated with 3 volumes of ethanol. The precipitate was dried, ground to pass through a 415-μm screen, and then dried for 18 h at 60 °C and 740-mm vacuum to correct samples to dry weight. Spontaneous water uptake was measured with the Baumann capillary apparatus at 25 °C using distilled water (Wallingford and Labuza, 1983).

RESULTS AND DISCUSSION

Extraction, Purification, and Characterization. Extraction of linseed with water at 4 °C yielded only 3.6% mucilage, while extractions at higher temperatures yielded larger amounts of mucilage (Table 1). The yields obtained are comparable to those obtained by Mazza and Biliaderis (1989) and Wannerberger et al. (1991). Subsequent use

Table 1. Effect of Extraction and Purification Procedures on Yield and Purity of Linseed Mucilage

mucilage,[a] condition of extraction	yield[b] (%)	protein[f] (%)	ash[f] (%)
Linott 25 °C, 2 h, EtOH ppt	4.0 ± 0.4[c]	ND[e]	ND
Linott 100–25 °C, 2 h, EtOH ppt	5.1 ± 0.4	ND	ND
Linott 80 °C, 2 h, EtOH ppt	8.4 ± 0.4	22.8 ± 0.6	6.3 ± 0.1
Linott 4 °C, 24 h, lyophilized[d]	3.6 ± 0.1	<1	7.7 ± 0.1
Linott 80 °C, 2 h, lyophilized[d]	6.0 ± 0.1	19.7 ± 0.6	4.4 ± 0.1
Vega clay treated	4.7 ± 0.3	4.3 ± 0.1	8.7 ± 0.1
meal 55 °C, 0.5 h, EtOH ppt	9.1 ± 0.2	29.2 ± 0.2	5.7 ± 0.1
meal 80 °C, 0.5 h, EtOH ppt	9.4 ± 0.1	28.6 ± 0.4	4.2 ± 0.1
meal 55 °C, 0.5 h, lyophilized[d]	9.5 ± 0.4	28.3 ± 0.2	3.4 ± 0.3

[a] Source, temperature and time of extraction, and method of isolation. [b] % basis of linseed or meal. [c] Means ± SD (n = 3). [d] Mucilages utilized for further study. [e] Not determined. [f] % on a dry mucilage basis.

of a calcium sequestrant [0.5% (w/v) ammonium oxalate] on linseed previously extracted with hot water (80 °C) yielded less than 0.5% additional carbohydrate material, indicating that pectic polysaccharides held in the cell wall matrices via Ca^{2+} bridges were not present. Alternately, the use of 0.1 M Na_2CO_3 also failed to extract any additional amounts of carbohydrates from the seed.

The protein content of mucilage samples increased with the temperature of the extraction medium (Table 1). Also, the protein content of mucilages extracted from the meal exceeded that of mucilage extracted directly from linseed. This can be attributed to the comminuted nature of the meal; proteins in the matrix would be exposed to the extraction media, thereby facilitating their solubilization and extraction. Ash contents varied from 3 to 8% for the nonpurified polymers. The values are substantially lower than those reported by Mazza and Biliaderis (1989) and may reflect differences in seed composition.

A purification procedure was developed to obtain mucilage of high purity and cause no degradation or other chemical modifications in the constituent polysaccharides. Following hot-water extraction of linseed (80 °C), a substantial decrease in soluble proteins was accomplished by treatment of the extracts with Vega clay. This clay adsorbent has been reported to selectively adsorb water-soluble proteins, and as such it has been employed to purify plant cell wall polysaccharides (Izydorczyk et al., 1990). Up to 80% of the proteins were indeed removed from hot-water-extracted mucilage using this clay treatment. However, the yield of treated mucilage was substantially lowered, suggesting that partial removal of polysaccharides also occurred during the clay treatment. Also, the ash content of clay-treated extracts increased.

For further characterization of linseed mucilage, three materials were chosen to represent diversified extraction conditions (4 vs 80 °C) or raw material (seed vs meal). As a result, the relative monosaccharide composition of the three mucilages varied substantially (Table 2). The glucose content of mucilage extracted from meal was much higher than that extracted from linseed. During the hydrolysis procedure for all linseed mucilage preparations, insignificant differences in glucose content were seen between 2 and 6 h of H_2SO_4 hydrolysis. This implies that cellulose was not present in the material, as a longer hydrolysis period would release larger amounts of glucose from cellulose, increasing the apparent glucose content of the mucilage. Pretreatment of mucilage samples with concentrated H_2SO_4 (35 °C, 1 h) to solubilize cellulosic materials, if present, also did not increase the glucose yields. The galacturonic acid content of Linott 4 °C was almost twice that found for the other two mucilages and was higher than levels reported by Wannerberger et al. (1991) for mucilages extracted from several varieties of linseed (21–36%). The difference may be attributed to the use of different linseed cultivars and different extraction procedures. An increase in galacturonic acid implies an increase of the acidic polysaccharides, which seem to be preferentially extracted at lower temperatures (Table 2). Arabinose, xylose, and galactose levels ranged from 5 to 9%, from 19 to 27%, and from 16 to 19% respectively, similar to the values obtained by other researchers (Wannerberger et al., 1991; Muralikrishna et al., 1987). Mannose was not detected in the mucilage, which disagreed with the studies of Wannerberger et al. (1991), where small amounts of mannose (1–2%) were found. The rhamnose and fucose levels varied within 14–16% and 3–4%, respectively.

Fractionation of Linott 80 °C resulted in four different fractions (Table 3). Both water-soluble and water-insoluble materials were found in the CTAB precipitated and CTAB nonprecipitated fractions. The total yields of the CTAB precipitated and CTAB nonprecipitated polysaccharides were approximately 41 and 27%, respectively. The galacturonic acid content of the CTAB precipitated fractions was approximately 40%, which is comparable to the value reported by Hunt and Jones (1962) (ca. 43%) but much higher than that obtained by Muralikrishna et al. (1987) (ca. 28%). Fractionation of Linott 4 °C resulted in two fractions which were water-soluble, CS and CP fractions. The CP fraction had higher amounts of galacturonic acid and xylose but a lower amount of galactose than the corresponding fraction obtained from

Table 2. Relative Monosaccharide Composition of Linseed Mucilages

material	Rha	Fuc	Ara	Xyl	Gal	Glu	Gal A
Linott 4 °C	16.4 ± 0.3[a]	4.0 ± 0.6	4.9 ± 0.5	18.8 ± 1.3	15.8 ± 0.6	trace	40.1 ± 2.3
Linott 80 °C	18.4 ± 0.2	4.0 ± 0.6	8.8 ± 1.4	26.8 ± 2.8	18.7 ± 0.3	1.4 ± 0.1	22.0 ± 1.1
meal 55 °C	14.5 ± 0.1	3.3 ± 0.3	9.3 ± 1.5	24.4 ± 0.2	16.2 ± 0.1	8.5 ± 0.1	23.8 ± 1.1

[a] Means ± SD (n = 3).

Table 3. Yield and Relative Monosaccharide Composition of Linseed Mucilage Fractions

material	yield	Rha	Fuc	Ara	Xyl	Gal	Glu	Gal A
Linott 80 °C								
CTAB ppt insoluble	5.1 ± 1.3[a]	19.0 ± 0.3[b]	3.3 ± 0.3	8.1 ± 1.0	18.7 ± 0.3	15.9 ± 0.2	nd[c]	35.0 ± 1.1
CTAB ppt soluble	36.0 ± 1.4	22.9 ± 1.8	3.3 ± 0.2	2.4 ± 1.3	3.3 ± 0.7	23.0 ± 0.4	nd	41.9 ± 0.7
CTAB non-ppt insoluble	6.1 ± 1.3	11.4 ± 0.9	3.3 ± 0.6	13.3 ± 1.2	41.4 ± 3.0	14.4 ± 0.4	1.5 ± 0.2	14.7 ± 1.0
CTAB non-ppt soluble	20.6 ± 1.7	4.1 ± 0.2	0.9 ± 0.2	18.8 ± 0.2	58.7 ± 3.7	9.7 ± 0.2	1.5 ± 0.2	6.3 ± 0.6
Linott 4 °C								
CTAB ppt (CP)	49.5 ± 3.5	19.4 ± 0.2	4.3 ± 0.3	2.0 ± 0.3	7.0 ± 0.6	13.6 ± 0.4	nd	53.6 ± 0.8
CTAB non-ppt (CS)	16.9 ± 2.6	9.6 ± 0.3	1.7 ± 0.4	13.2 ± 0.2	50.0 ± 0.7	9.5 ± 0.5	1.1 ± 0.2	14.7 ± 1.1

[a] % on a dry mucilage basis. [b] Means ± SD (n = 3). [c] Not detected.

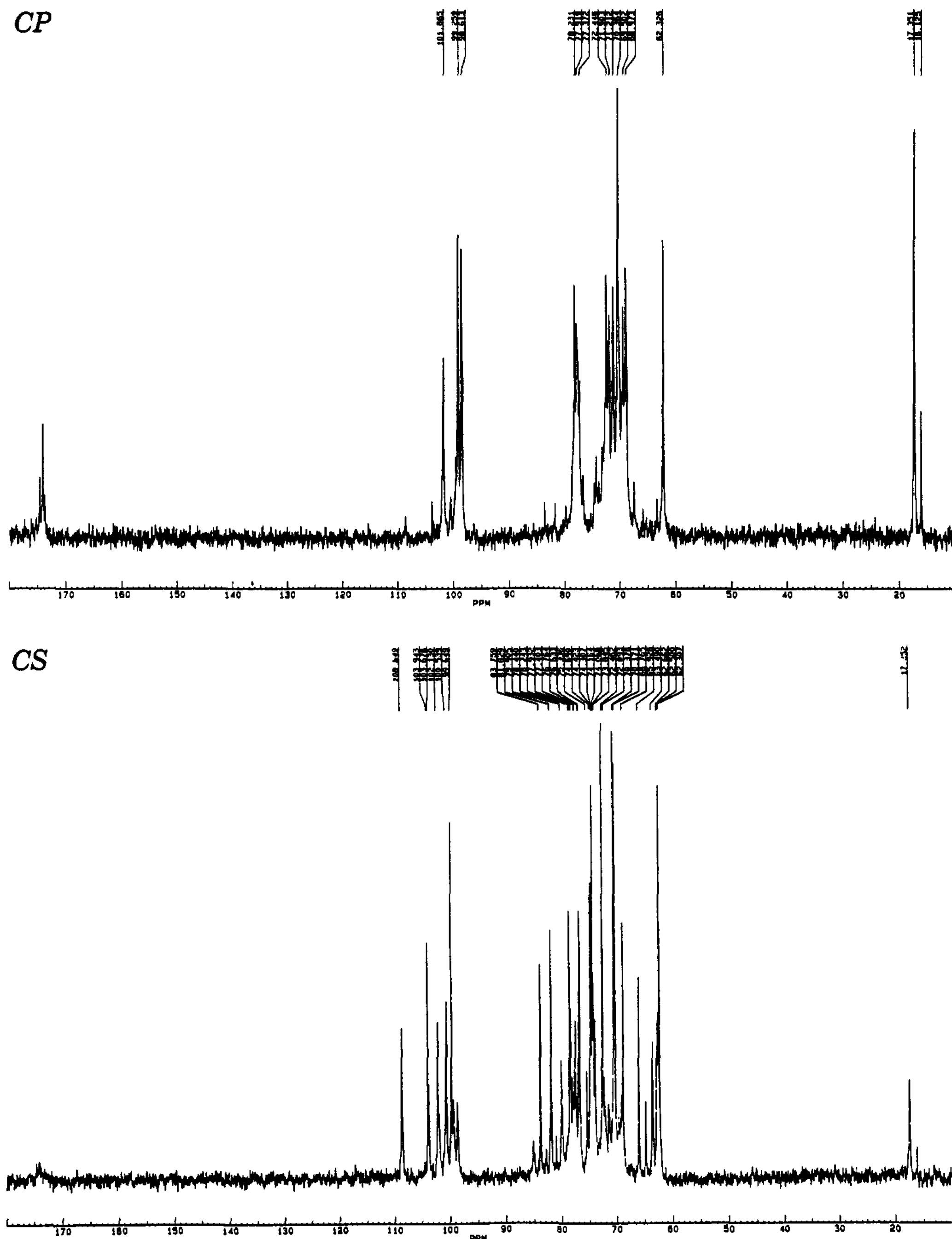

Figure 2. ^{13}C NMR spectra of CP and CS fractions of linseed mucilage (10% D_2O). The chemical shifts were assigned relative to 1,4-dioxane.

Linott 80 °C (CTAB precipitated, soluble). The CP fraction had lower amounts of galactose, fucose, and rhamnose than a similar acidic polysaccharide fraction examined by Muralikrishna et al. (1987) (14 vs 22%, 4 vs 12%, and 19 vs 37%, respectively). In contrast, the galacturonic acid content was much greater for the CP fraction of the present study (ca. 54%) than the value reported by Muralikrishna et al. (1987) (21%). Methyl esters and covalently bound phenolic residues were not detected in any mucilage preparations of the present study. The observed differences in composition among the various mucilage extracts or their fractions clearly indicate that, besides varietal differences as reported by Wannerberger et al. (1991), the extraction conditions greatly affect the nature of the solubilized polysaccharides from the seed.

The ^{13}C NMR spectra of the fractions obtained from Linott 4 °C were fairly complex (indicative of both linkage and residue multiplicity), making difficult the assignment of individual resonances (Figure 2). The CP fraction displayed a peak at 174.1 ppm which is attributed to 1 → 4-linked galacturonic acid (Keenan et al., 1985; Pressey and Himmelsbach, 1984). The presence of a peak at 17.35 ppm confirms the presence of (1 → 2) linked L-rhamnose in the polymer (Keenan et al., 1985), while the resonances at 16.2 ppm can be attributed to fucose residues. The CS fraction lacked the 174.1 ppm peak and exhibited a weaker 17.35 ppm peak than CP. The anomeric regions of the two spectra also differ. The CP had resonances at 101.9, 99.3, and 98.6 ppm, corresponding to C-1 of galactose, rhamnose, and galacturonic acid residues, respectively. In the CS fraction, the signal at 108.6 ppm indicates the presence of α-arabinofuranose in the arabinoxylan structure (Bradbury and Jenkins, 1984) and contradicts the methylation data of Muralikrishna et al. (1987), who reported the presence mainly of arabinopyranose residues. The resonances between 99 and 104 ppm for the CS fraction

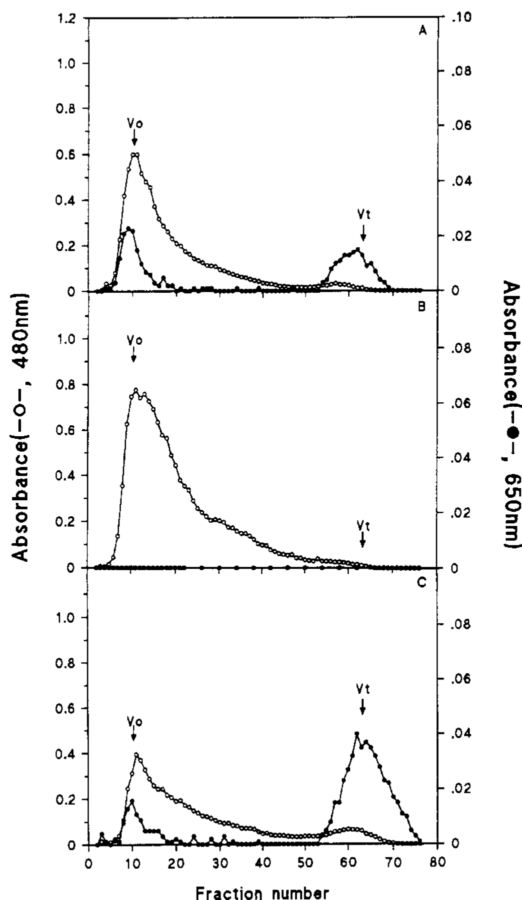

Figure 3. Gel filtration chromatography on Sepharose CL-2B (2.5 × 94 cm, 0.1 M sodium acetate buffer, pH 4.0, flow rate 35 mL/h, 25 °C) of mucilages: (A) Linott 80 °C; (B) Linott 4 °C; (C) meal 55 °C. Absorbances at 480 (○) and 650 nm (●) correspond to eluting carbohydrates and proteins, respectively.

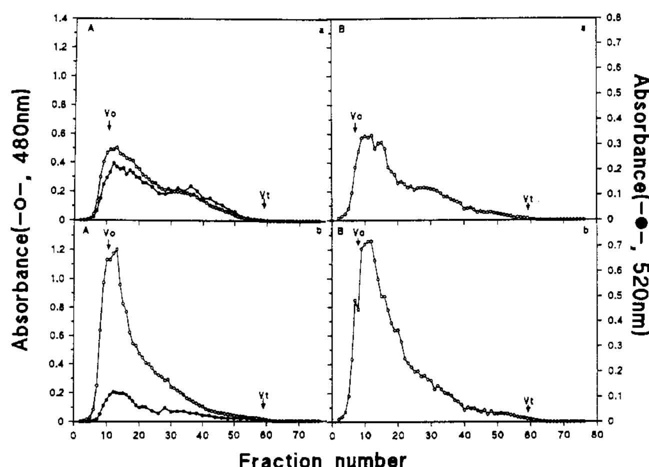

Figure 4. Gel filtration chromatography on Sepharose CL-2B [2.5 × 94 cm, (A) 0.1 M sodium acetate buffer, pH 4.0; (B) 0.1 M sodium acetate buffer, pH 4.0 in 6.0 M urea, flow rate 35 mL/h, 25 °C] of fractions obtained from Linott 4 °C: (A) CP and (B) CS fractions. Absorbances at 480 (○) and 520 nm (●) correspond to eluting carbohydrates and galacturonic acid, respectively.

arise from xylose residues (Bock et al., 1984) The lack of resonances at 21 and 53 ppm, corresponding to acetyl and methyl groups, respectively, indicates the absence of such residues in the native polymers.

The gel chromatographic profiles (Figures 3 and 4) revealed that carbohydrates eluted predominantly at the void volume, while proteins eluted in the void and total volumes. The 6.0 M urea buffer was used to minimize

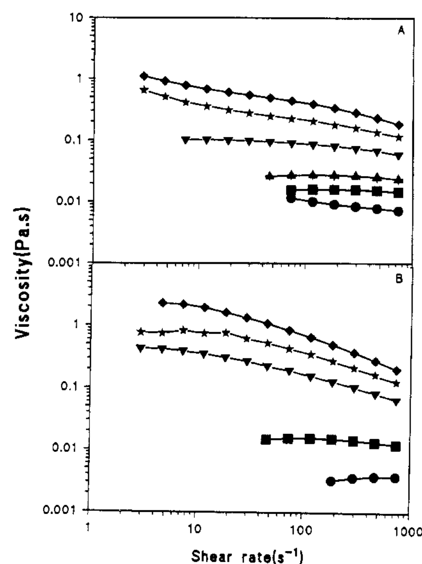

Figure 5. Flow curves of linseed mucilage fractions of varying concentration (pH 5.5, 25 °C): (A) CP fraction; (B) CS fraction. Polymer concentration (w/w): 0.1% (○); 0.3% (■); 0.5% (▲); 1.0% (▼); 1.5% (★); 2.0% (◆). Standard errors of means for triplicate measurements were less than 10% of the absolute values.

interchain associations via hydrogen bonding (Applegarth and Dutton, 1964). However, the chromatographic profiles of the fractions did not differ substantially in the presence or absence of urea, indicating that chain association and aggregation processes were not a determinant factor in the elution behavior of the mucilage preparations. Nevertheless, all samples eluted over a range of size, indicating that linseed polysaccharides are highly polydisperse.

Rheological Properties. The calculated limiting viscosity value of CS (in 0.2 M NaCl) was 6.6 dL/g and that of CP 4.6 dL/g. Pectic substances isolated from flax fibers (Hourdet and Muller, 1987) had much lower intrinsic viscosities (0.04–0.652 dL/g). The shear rate dependent flow behavior of xanthan gum, guar gum, Linott 80 °C, Linott 4 °C, meal 55 °C, CS, and CP aqueous solutions/dispersions was also examined. Shear thinning behavior was observed for all polymers at high concentrations. The degree of shear thinning decreased with decreasing gum concentration as shown for CP and CS solutions in Figure 5. Differences in shear thinning behavior were quantified using the power law model $\eta = m\dot{\gamma}^{n-1}$ where η is the apparent viscosity, m is the consistency coefficient, n is the flow behavior index, and $\dot{\gamma}$ is the shear rate (s^{-1}) (Launay et al., 1986) (Table 4). Guar gum exhibited the highest m and the lowest n values. At equivalent concentration, the CS fraction exhibited higher m and lower n values than the CP fraction, implying that solutions of the former are more viscoelastic and shear thinning than the latter. This is in agreement with the fact that polysaccharide preparations of high intrinsic viscosity also exhibit greater shear thinning properties (Launay et al., 1986). Such differences are generally attributed to differences in structure and molecular size since these intrinsic molecular parameters are the main determinants of the hydrodynamic volume of polysaccharides in solution. The meal 55 °C extract had low values of m and high values of n, which could be attributed to the higher protein contamination of this preparation. Polysaccharides, because of their high intrinsic viscosities, are primarily responsible for the viscous nature of solutions/dispersions (Mitchell, 1979).

The viscosity of aqueous solutions of mucilage decreased with increasing temperature. The decrease in apparent

Table 4. Flow Behavior Index (n) and Consistency Coefficient (m) of Commercial Gum and Mucilage Solutions (11.64–734.3 s^{-1}, 25 °C, pH 5.5) according to the Power Law Model $\eta = m\dot{\gamma}^{n-1}$

polymer	n^a	m
xanthan 2.0%	0.29	6.16
xanthan 1.5%	0.35	3.14
guar 2.0%	0.22	41.85
guar 1.5%	0.27	18.07
Linott 80 °C 2.0%	0.63	1.68
Linott 80 °C 1.5%	0.71	0.60
Linott 4 °C 2.0%	0.61	2.63
Linott 4 °C 1.5%	0.73	0.72
meal 55 °C 2.0%	0.75	0.38
meal 55 °C 1.5%	0.77	0.19
CP fraction 2.0%	0.69	1.53
CP fraction 1.5%	0.73	0.68
CS fraction 2.0%	0.46	8.37
CS fraction 1.5%	0.53	3.19

[a] $n = 3$, linear regression of $\log \eta = \log m + (n-1)\dot{\gamma}$ ($r^2 > 0.98$, $p < 0.01$).

Table 5. Activation Energy (E_a) of Mucilage and Fraction Solutions (1.0% w/w) (116 s^{-1}, 15–65 °C, pH 5.5)

material	$E_a{}^a$ (kJ/mol)	material	$E_a{}^a$ (kJ/mol)
Linott 4 °C	23.5	CP fraction	22.8
Linott 80 °C	22.8	CS fraction	30.8

[a] $n = 3$, linear regression of $\log \eta = \log A + 2.303(E_a/RT)$ ($r^2 > 0.98$, $p < 0.01$).

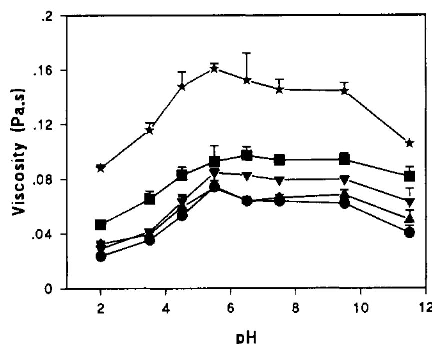

Figure 6. Effect of pH on the apparent viscosity of 1.0% (w/w) mucilage and fraction solutions (116 s^{-1}, 25 °C) [exception: meal 55 °C, 1.5% (w/w)]: Linott 80 °C (●); Linott 4 °C (■); meal 55 °C (▲); CP fraction (▼); CS fraction (★).

viscosity (within 15–65 °C) followed an Arrhenius temperature dependence, and the computed energy of activation values are shown in Table 5. Data are not given for meal 55 °C as reproducible results for viscosity could not be obtained at high temperatures (very thin dispersions) due to the sensitivity limits of the rheometer. Linott 80 °C and Linott 4 °C had similar E_a's. The E_a's for 1% aqueous solutions of guar gum and (carboxymethyl)-cellulose, as reported by Launay et al. (1986), were 11.7 and 23 kJ/mol, respectively. The CS fraction had a substantially higher E_a than other commercial gum solutions, which would imply that a higher energy is required to initiate viscous flow in its solutions (Launay et al., 1986). The CP fraction had a lower E_a value than CS, suggesting that weaker intermolecular associations occur in solutions of the former. This could be related to the structure and conformation of the constituent polymers of CP.

The influence of pH on the apparent viscosity ($\dot{\gamma}$ = 116 s^{-1}) of mucilage solutions and its fractions is shown in Figure 6. In contrast to a study by BeMiller (1973), in which maximum viscosity of mucilage was obtained at pH 8.0, the results of Figure 6 indicate that high viscosity is

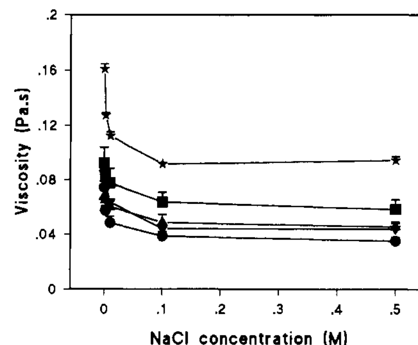

Figure 7. Effect of NaCl concentration on the apparent viscosity of 1.0% (w/w) mucilage and fraction solutions (116 s^{-1}, 25 °C) [exception: meal 55 °C, 1.5% (w/w)]: Linott 80 °C (O), Linott 4 °C (■), meal 55 °C (▲); CP fraction (▼); CS fraction (★).

maintained over a broad range of pH (5.0–9.0). These observations are in agreement with the data of Mazza and Biliaderis (1989). At low pH's, charge suppression would result in a smaller conformation of the polymers, leading to reductions in viscosity. At high pH's, the decrease in viscosity may be attributed to alkaline depolymerization reactions (BeMiller, 1986). The CS and CP fractions showed similar profiles, in spite of having different content of acidic polysaccharides.

The responses in apparent viscosity of linseed mucilage and its fractions to the addition of NaCl are shown in Figure 7. Initial addition of NaCl resulted in reduction of viscosity for the solutions up to 0.1 M. A similar response was seen with the addition of $CaCl_2$. The reduction in viscosity is attributed to the progressive suppression of intramolecular charge–charge repulsion and consequent contraction of the polysaccharide molecules (Mazza and Biliaderis, 1989). The similar rheological responses obtained for NaCl and $CaCl_2$ indicate that Ca^{2+}-mediated cross-linking, as commonly observed for pectins, does not occur for the CP fraction.

The dynamic moduli (G', G'') and viscosity (η') as a function of frequency are shown for 2% (w/w) solutions of guar gum, xanthan gum, Linott 80 °C, Linott 4 °C, CP fraction, and CS fraction in Figure 8. Data on the dynamic rheological properties of meal 55 °C are not shown because the viscosity range of the sample was below the sensitivity limits of the rheometer operated in the oscillatory mode. Xanthan gum exhibited behavior typical of a weak gel, with G' exceeding G'' over the entire frequency range investigated, and both rheological parameters were relatively independent of frequency, agreeing with Morris (1990). Guar gum exhibited typical viscoelastic behavior as is evident by G'' exceeding G' at low frequencies, while the reverse is observed at high frequencies. The mechanical spectra of CS resembled those of guar gum. Solutions of Linott 80 °C, Linott 4 °C and even more of CP showed dynamic responses characteristic of dilute polymer solutions, where $G'' > G'$ and both moduli are highly dependent on frequency. The rheological data of Figure 8 indicate that the neutral polysaccharide fraction of mucilage has a greater tendency for structure formation in an aqueous environment (elastic component) than the acidic polysaccharide fraction. This supports the methylation data of Muralikrishna et al. (1987), according to which the neutral polymers are mainly linear while the acidic polysaccharides of linseed mucilage are highly branched.

Xanthan gum had the highest water binding capacity (WBC) (Table 6). This value was twice as high as that reported by Wallingford and Labuza (1983). This may be attributed in part to the fact that the xanthan gum used in the current experiments was an agglomerated product;

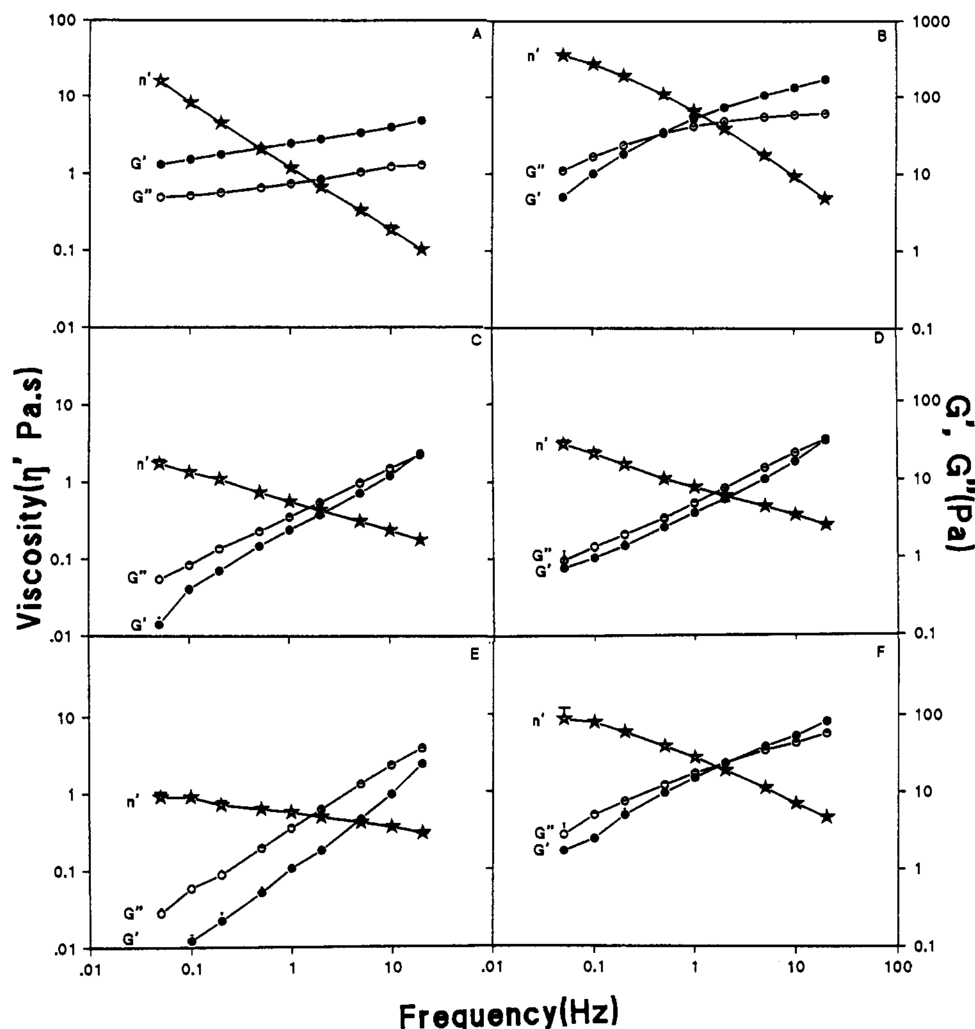

Figure 8. Frequency dependence of storage (G') and loss (G'') moduli and dynamic viscosity (η') of 2.0% (w/w) commercial food gum, mucilage, and fraction solutions (pH 5.5, 25 °C): (A) xanthan gum; (B) guar gum; (C) Linott 80 °C; (D) Linott 4 °C; (E) CP fraction; (F) CS fraction.

Table 6. Water Binding Capacity of Commercial Food Gums and Linseed Mucilages As Determined by the Baumann Capillary Apparatus

material	WBC[a] (g of H_2O/100 g of solids)	material	WBC[a] (g of H_2O/100 g of solids)
xanthan gum	32300 ± 1100	Linott 4 °C	3000 ± 300
guar gum	2200 ± 400	Linott 80 °C	2500 ± 100
		meal 55 °C	1600 ± 100

[a] Means ± SD (n = 3).

more pores would be present in such a product, allowing more water to be held within the gellike structure (Chen et al., 1984). The water binding capacity of the three mucilages decreased in order from Linott 4 °C to Linott 80 °C to meal 55 °C. This order corresponds to the total carbohydrate content of the mucilage samples. Since the water binding capacity of most food gums ranges between 300 and 3200 g of H_2O/100 g of solids (Wallingford and Labuza, 1983), the values obtained for mucilage indicate that this material has a relatively high WBC.

Conclusions. Linseed mucilage is composed primarily of polymeric carbohydrates; the yield and purity of this material vary with the extraction conditions. Maximum yield was obtained when hot water was used as extractant, but the preparation had high levels of contaminating proteins. Cold-water extraction yielded lower amounts of mucilage of reduced protein content. Purification of mucilage can be further accomplished by a clay adsorbent to remove contaminating water-soluble proteins.

Fractionation of mainly acidic and neutral polysaccharide constituents was achieved by CTAB selective precipitation of the former, though complete separation was not possible. These preparations were polydisperse and had constituents of large hydrodynamic volumes in aqueous solutions. The neutral polysaccharide fraction had higher limiting viscosity and exhibited a greater degree of shear thinning and viscoelastic behavior than the acidic fraction. Both salt and pH affected the viscosity of mucilage and its fractions; maximum solution viscosity was obtained in the neutral pH zone and zero salt concentration. The linseed mucilage also had a large water binding capacity.

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