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Prickly pear nopals pectin from *Opuntia ficus-indica* physico-chemical study in dilute and semi-dilute solutions

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Abstract

The water soluble fraction of peeled prickly pear nopals called native sample (NS) has been characterised mainly by SEC/MALLS analysis. Two main components have been identified: one with high average molar mass (M_w of 13×10^6 g mol^{-1}) called the high weight sample (HWS), the other being a low M_w fraction (LWS). After extensive ultra filtration of NS, isolated HWS and LWS are obtained. From sugar composition analysis, HWS has been found to be a pure polysaccharide, without protein of the pectin family. Moreover, HWS contains a low amount of charged sugar. The conformation of HWS has been discussed using molar masses, gyration radii and viscometry results and LWS has been evidenced as a protein. Rheological behaviour is reported to give an initial understanding of the system's behaviour. The effect of the degree and purification and added monovalent and divalent salts were investigated, The low charge density of the polymer backbone resulted in interesting viscosity stability even in the presence of salts.

Keywords: *Opuntia ficus-indica*; Polysaccharide; Pectin; SEC/MALLS; Rheology; Salt effect; Calcium complexes

1. Introduction

Pectins have a number of applications in the pharmaceutical, cosmetic and food industries. These numerous applications justify the search for new sources of pectins. Industrial pectins are extracted from fruits (apple, citrus…) and fibrous plants (flax). *Opuntia ficus-indica* (Prickly pear) belonging to the cactaceae group growing in desert and subdesert areas is a potential source of pectins. This has been evidenced through the works of Trachtenberg and Mayer (1981) on the mucilage. This plant is widely cultivated for its sweet fruit; its nopals are used as cattle food and because of its strength it is used for fences in fields. Its abundance in arid area makes it a good candidate for industrial utilisation.

The extraction and use of the natural pigments contained in blood-red prickly pulp have been investigated by Forni, Polesello, Montefiori and Maestrelli (1992) while Sawaya, Khalil and Al Mhammad (1983) pointed out the nutritional character of the prickly pear pulp. The isolation and characterisation of reserve proteins from the seeds of prickly pear have been studied by Uschoa, Souza, Zarate, Gomes-Filho and Campos (1998). Their main conclusion is that such proteins (molar masses of ~6500 g mol^{-1}) have an amino acid composition similar to the 2S albumin storage protein family.

Forni, Penci and Polesello (1994) focused their search on the determination of the polysaccharide prickly pear peel composition. They have found that the polysaccharide is characterised by a very high neutral sugar content, mainly consisting of rhamnose and galactose and enough galacturonic acid content to be useful as a thickening additive. They suggest applications in sauce production and pastry as well as in cosmetic and pharmaceutical preparations.

However, no data exist concerning the macromolecular properties of polysaccharides extracted from the mucilage of prickly pear nopals. The general aim of this work is to understand the structure–property relationship of this polysaccharide and its potential for use as a commercial pectin.

In the present paper, extraction and purification of polysaccharides are reported. Our interest has been focused on the physico-chemical characterisation using several techniques such as: viscosity in dilute solutions, low angle laser light scattering (LALLS) and steric exclusion chromatography equipped with double detection refractometric and multiangle laser light scattering (SEC/MALLS).

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We also describe the thickening properties of this polysaccharide issued from *O. ficus-indica* nopals from rheological measurements in dilute and semi-dilute aqueous solutions.

2. Extraction and purification

The nopals of prickly pear aged between six and twelve months were collected in the area of Monastir (Tunisian Sahel) at the end of spring (May 1998). This season corresponds to the flowering period of the plant. They ripen at the end of summer.

2.1. Extraction

Samples (1 000 g) of peeled nopals were shredded and blended using a mixing machine. The degreasing process of the mixture, was realised by adding 1 l of petroleum ether under mechanical stirring for 24 h at room temperature. After removal of ether elimination, the blend was first macerated in deionised water (Milli Q) for one day at room temperature, then filtered in vacuum through a Go sintred glass funnel. The resulting filtrate was centrifuged (7000 rpm, 1 h), then finally filtered through a Millex HA 0.45 μm filter (Millipore).

2.2. Purification

The filtrate was subdivided into three fractions. The first was obtained by direct freeze-drying to get the native sample (NS). A second and a third fraction were isolated by ultrafiltration using a Minitan cell equipped with a series of membranes with molecular weight cut off 100,000 g mol^{-1} in order to eliminate salts and compounds of low molecular weight. The ultrafiltration was conducted against deionised water (Milli Q Process) for about 1 week until the conductivity equalled that of the deionised water. The extensively ultrafiltrated sample retained by the membrane, was freeze-dried to obtain the high weight sample (HWS). The filtrate obtained during this ultrafiltration, was concentrated and freeze-dried to get the low weight sample (LWS).

Two degrees of purification were used for the rheological characterisation: HWS obtained from extensive ultrafiltration and mixed sample (MS) obtained after a short ultrafiltration period of 24 h.

All samples (NS, HWS, MS and LWS) were freeze-dried before utilisation.

3. Preparation of solutions

The samples (NS, LWS, HWS and MS) were dissolved in water deionised by the Milli Q Process containing 0.5 M aqueous NaCl or 0.1 M aqueous $LiNO_3$ using magnetic stirring. Each solution was filtered through a 0.45 μm filter (Millex) before analysis.

The semi-dilute solutions were obtained by dissolving the sample in deionised water containing the desired salt level (NaCl, $LiNO_3$ or $CaCl_2$) under gentle mechanical stirring at room temperature. A bactericide (sodium triazide) was added to the resulting solutions, after which the mixtures were kept at 4°C before analysis.

4. Material

4.1. Acid equivalent weight determination

About 0.24 g of the freeze-dried sample was dissolved in pure water. About 1 ml of 1 M NaOH was added to ensure the total ionisation of carboxylic acid groups. The equivalent weight of the polysaccharide sample was determined from indirect conductimetric titration of such a solution with 0.1 M HCl (Eyler, Klug & Siephuis, 1947).

4.2. Sugar content determination

The sugar content was determined by methanolyse and silylation followed by gas chromatography according to the method described elsewhere (Girault, Bert, Rihouay, Jauneau, Morvan & Jarvis, 1997; Goubet, Bourlard, Girault, Alexandre, Vandevelde & Morvan, 1995).

4.3. Protein presence determination

The quantification of protein in aqueous solutions was performed by spectrophotometry at 562 nm with bicinchoninic acid (BCA) after calibration with bovine serum albumin (BSA) (Smith, Krohn, Hermanson, Mallia & Gartner, 1985).

Freeze-dried pronase from *Streptomyces griseus* was purchased from Boehringer Mannheim and added to the HWS and LWS solutions as indicated by Connolly, Fenyo and Vandevelde (1988) for Acacia Senegal Gum.

4.4. Viscosity

Viscosimetric measurements were performed at a low shear rate using a Contraves LS30 viscometer at 25°C. In dilute solution ($C < C^{**}$ and $C/C^{*} < 1$), the polymer chains are separated from each other and the interactions are weak, so that a series expansion from an expression such as Huggins' equation is valid. The slope of the line $\eta sp/C$ versus C is proportional to $k'[\eta]^2$ where k' (Huggins coefficient) indicates the strength of the polymer–polymer interactions and the zero y-axis intercept gives the intrinsic viscosity ($[\eta]$) which is related to the hydrodynamic volume.

With concentration increase, the chains begin to overlap at concentration C^{*}. Between C^{*} and C^{**} (overlapping zone) polymer chains overlap and are entangled with each other. C^{**} is the concentration corresponding to the start of the concentrated regime where the chains are totally entangled.

The Utraki–Simha representation log η_{sp} versus log $C[\eta]$ (Utraki & Simha, 1963) permits the determination of C^*.

4.5. Rheology

Flow and dynamic rheological experiments in the semi-dilute region were performed using a Carri-Med CSL-100 controlled stress rheometer with a double gap concentric cylinder geometry. The rheometer was equipped with a solvent trap to prevent any dehydration during measurement, and the temperature controlled by circulation from of an external water bath. Measurement started after a 15 min delay to ensure the system reached equilibrium.

4.6. Light scattering

The weight average molecular weight M_w and the second virial coefficient A_2 were computed from scattered light intensities using a plot of the excess Rayleigh factor R_θ as a function of polymer concentration C. A LALLS photometer Chromatix KMX-6 was used ($\lambda = 633$ nm).

At a very low angle ($\theta = 4.88°$), R_θ is related to C and M_w by the following equation:

$$KC/\Delta R_\theta = 1/M_w + 2A_2C$$

where the optical constant K takes into account the refractive index (RI) increment dn/dC measured with an optilab refractometer (Wyatt tech.) at 633 nm.

4.7. On-line SEC/MALLS

For Size Exclusion Chromatography, the following experimental device was used: a mobile phase (0.1 M aqueous $LiNO_3$ solution) is pumped at 0.6 ml $^{-1}$ (Kontron HPLC pump 420) through two OHPAK SB 804 and 806 HQ columns (Shodex) in series. These conditions are typically sufficient for screening polyelectrolyte and to protect the chromatography line. The polymer solution, injected through a 100 μl loop, was eluted and separated with respect to the SEC laws and finally detected using two detectors in series: a MALLS (Dawn F, Wyatt Technology) followed by a Refractometric Index Detector (DIR ERC-7515A, Erma CR Inc). Both detections allow the number, weight and z-average molecular weights and size distributions to be calculated. Well-adapted representations of experimental data such as gyration radius versus molecular weight for example, give information about polymer conformation in solution (Capron, Grisel & Muller, 1995; Picton, Merle, Muller & Int, 1996). The HWS was analysed using 15 angles (between 30 and 160°) and fitted the Zimm first-order extrapolation with good precision.

5. Results and discussion

The NS has been characterised by SEC/MALLS analysis using 2 g l^{-1} solutions in $LiNO_3$ 0.1 M. These conditions will be discussed later with respect to the viscometric study. Fig. 1 shows, the evolution of the RI signal and light scattering (LS) response at 90° as a function of the elution volume. The RI signal is proportional to the concentration, whereas the LS response depends on both concentration and molar mass. The first population (called A) of NS was eluted at 12 ml. Fraction A shows a low RI intensity due to its low concentration in the sample (about

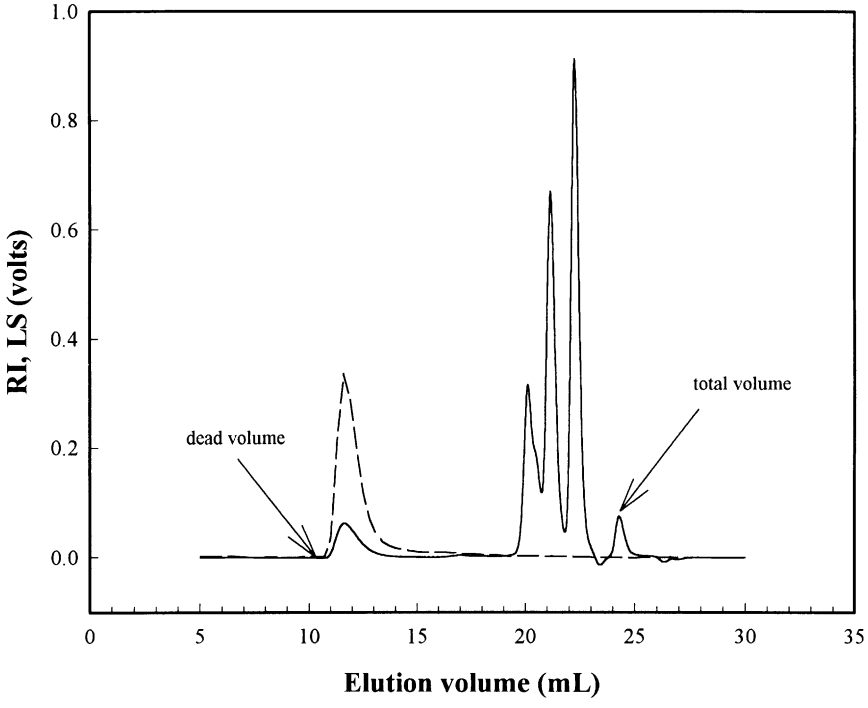

Fig. 1. Refractive index (solid line) and light scattering response at 90° (dotted line) from SEC/MALLS analysis of NS.

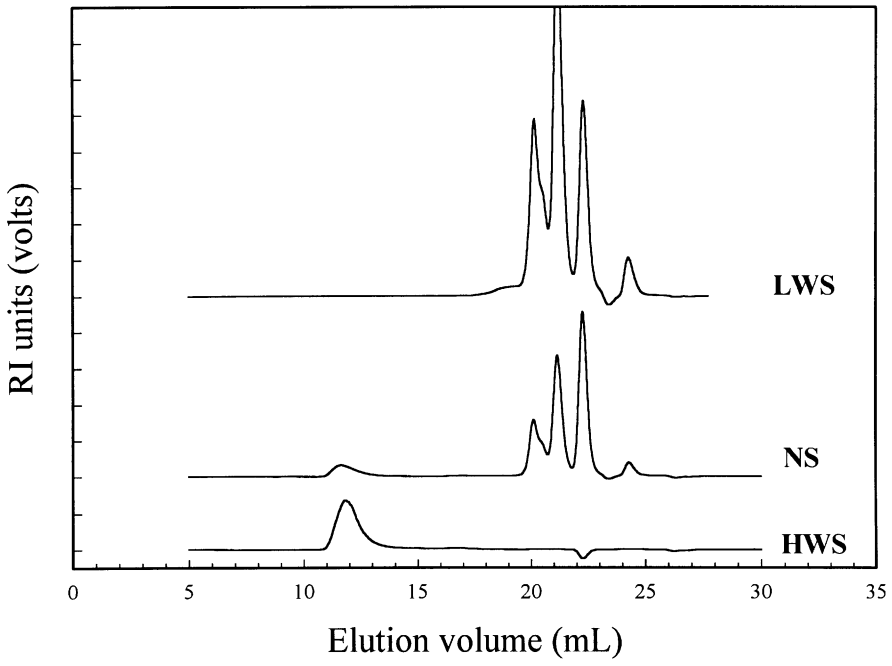

Fig. 2. SEC/MALLS elution profiles of NS, HWS and LWS in 0.1 M $LiNO_3$.

7% obtained by the ratio of peaks area) whereas the light scattering signal is large. This indicates a high molar mass polymer. Other species are eluted at volumes around 20 ml. The three peaks, called B, consist of the major part of the NS as evidenced by the large intensity of the RI response. Nevertheless, the negligible scattered light intensity indicates that B is of very low molar masses.

From the extensive ultrafiltration described above HWS and LWS were obtained, giving the ultra-purified fraction and the residue of filtration (below 100,000 g mol^{-1}). Both products have been analysed by on-line SEC/MALLS in the conditions used for NS measurements. d*n*/d*c* have been measured and are 0.139 and 0.176 ml g^{-1} for HWS and LWS, respectively. The elution profiles (RI response) of both HWS and LWS are compared to NS in Fig. 2. It clearly appears that HWS is equivalent to the pure fraction A without any trace of B species, whereas LWS shows the same profile as fraction B without any trace of A. These results obviously evidence a good efficiency of the purification process.

The macromolecular data could be monitored from the above LALLS and SEC/MALLS experiments. The average weight molar masses ($\overline{M_w}$), radii of gyration ($\overline{R_g}$) and second virial coefficient (A_2) of both fractions A and B from NS as well as HWS and LWS are reported in Table 1.

In agreement with the previous observations, fraction A has a very high $\overline{M_w}$ (about $13 \times 10^{+6}$ g mol^{-1} and a gyration radius ($\overline{R_g}$) of about 200 nm. The HWS chromatographic profile is similar to that of fraction A, and obviously both present the same macromolecular characteristics. More information about the conformation can be obtained from the slope (x) of M_w vs R_g log–log plot (Fig. 3). A value of x near 0.6 is obtained for the HWS and fraction A of NS, suggesting the same random coil flexible conformation. Moreover the second virial coefficient A_2 is about $10^{-4} \times$ mol ml g^{-2} corresponding to a good solvatation of the polymer chain in the solvent used (i.e. 0.1 M $LiNO_3$). The sugar composition of HWS is reported in Table 2 and the data are compared with those of Forni et al. (1994) on the prickly pear peel. It appears that HWS is a polysaccharide from the pectin family. Nevertheless, its composition greatly differs from that of peel pectin. This

Table 1
Light scattering measurements in 0.1 M $LiNO_3$

		M_w (g mol^{-1})	R_g (nm)	x from R_g vs M_w	A_2(mol ml g^{-2})
NS	Fraction A	13.3×10^{6} [a]	188	0.57	
HWS		12.8×10^{6} [a]	190	0.57	1×10^{-4} [b]
		14.2×10^{6} [b]			
LWS		3900 [b]	–		-8×10^{-4} [b]

[a] From SEC/MALLS.
[b] From LALLS.

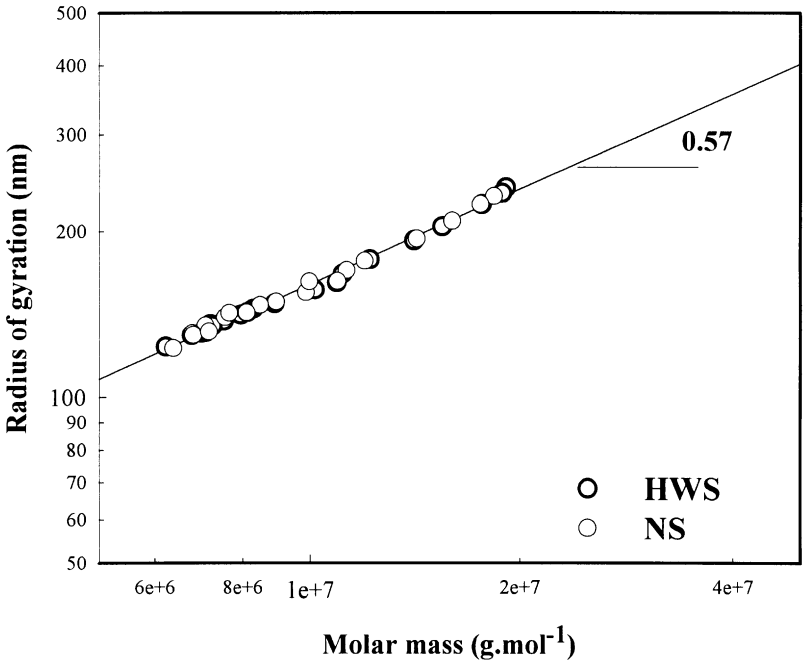

Fig. 3. Radii of gyration as a function of molar masses. High weight fraction of NS and HWS from ultrafiltration in 0.1 M $LiNO_3$.

is particularly clear concerning the charged sugar content that is only about 20% of the whole sugar in nopals composition whereas about 65% of galacturonic acid has been found in peel pectin. Conductimetric titration of HWS gave an acid equivalent weight (AEW) of 840 g per mole of equivalent, which corresponds to about 1 acid group per 4.5 sugar repeat units. This result seems consistent with the charged sugar composition found. As a comparison the parameter AEW is 1722 for gum arabic (Jefferies, Pass & Phillips, 1977), 765 and 250 for flax pectins with the degree of esterification being 38 and 10%, respectively (Hourdet, 1989). This evidence for a charged pectin type structure suggests a potential ability for this type of polysaccharides to interact with divalent cations (Ca^{2+} or Mg^{2+} for example) as largely described in the literature (Thibault & Rinaudo, 1985; Axelos, Mest & François, 1994). This point will be studied later in this paper.

On the other hand the elution profile of LWS is quite similar to that of fraction B of the NS (Fig. 2). This sample consists of very low molar mass species as shown by the average M_w of about 4000 g mol^{-1}, though this result is of a poor precision due to low LS intensities. Moreover, the second virial coefficient is quite insignificant, evidencing poor water solubility. Those results argue in favour of protein based composition in LWS or fraction B of NS as reported by Uschoa et al. (1998) for the 2S albumin storage family from the seeds of *O. ficus-indica.*

In order to determine the presence of proteins, spectrophotometric detection by a BCA reagent has been conducted on HWS and LWS fractions (Fig. 4). The absorption calibration at 562 nm was obtained with BSA. As a result, no protein has been found in the HWS, whereas it represents the major part of LWS (~80%). Moreover, nitrogen content analyses of 0% for HWS and 2.2% for LWS confirms the previous result. In order to confirm the above result, the NS was submitted to a pronase treatment (37°C, 24 h). NS before and after treatment has been studied by SEC/MALLS analysis and compared to pronase alone (Fig. 5): as visible on the profiles, no modification occurs for the pure polysaccharide (fraction A). In summary, it appears that proteins are essentially in the fraction LWS and that HWS is a pure polysaccharide making it quite different from the well-known arabino–galactan (gum arabic) that evidences a protein–arabino–galactan complex (Connolly et al., 1988).

The viscometric study of 0.5 M NaCl has been carried out in dilute and semi-dilute solutions. Intrinsic viscosity ($[\eta]$), Huggin's constant (k') and the critical overlapping concentration (C^*) are reported in Table 3. The Huggin's constant reported for HWS is typical of a highly solvated water-soluble polymer. $[\eta]$ (~850 ml g^{-1}) for the very high $\overline{M_w}$

Table 2
Sugar composition of peel prickly pear pectins (% in weight)

A. Gal.	A. Glu.	Glu.	Xyl.	Gal.	Rha.	Ara.	Reference
64.0	–	2.6	2.1	22.0		6.0	Forni et al. (1994) and nopals
9.6	9.8	1.0	18.7	20.3	6.9	33.1	This work

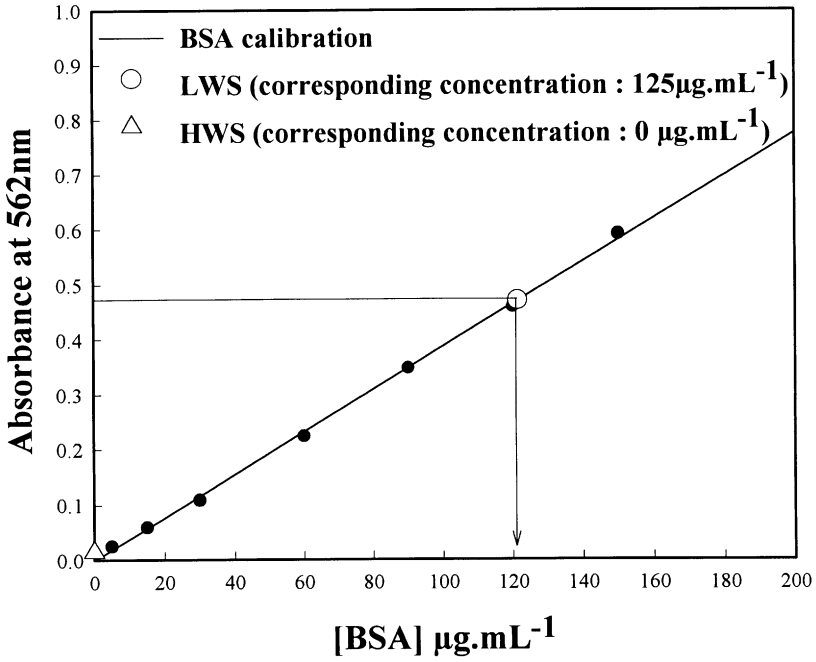

Fig. 4. Spectrophotometric detection of protein in HWS and LWS by BCA protein assay reagent calibration line obtained with bovine serum albumin (BSA).

of the polymer (13 10^6 g mol^{-1}), is consistent with a very flexible random coil conformation and/or a possible high degree of branching due to the presence of rhamnose units. By comparison, amylose with the same molar mass (i. e. 13×10^6 g mol^{-1}) gives an intrinsic viscosity of 2500 ml g^{-1} under the same conditions (Banks & Greenwood, 1969). The slopes of bilogarithmic plots, as determined from experimental data reported, are of 1.4 and 2.6, respectively, before and after the overlap concentration C^*, which indicates dilute/semi-dilute regimes. They are typical of polysaccharide solutions and characteristic of an entanglement process. The polysaccharide starts overlapping in 0.5 M NaCl for a concentration of 1.55 g l^{-1}. This result agrees well with the expected C^* (1.75 g l^{-1}) calculated using the relation given by Morris, Rees, Robinson and Young (1980): $C^* = 1.5/[\eta]$, indicating that the high weight polymer extracted from prickly pear nopals of *ficus-indica* follows the typical viscometric behaviour of a polysaccharide.

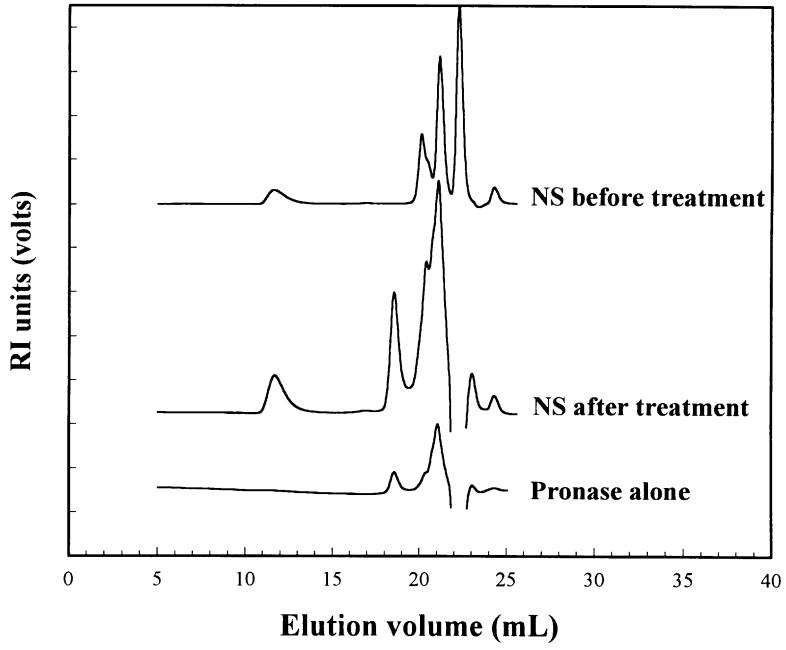

Fig. 5. SEC/MALLS elution profiles of NS before and after a pronase treatment (37°C, 24 h) in 0.1 M $LiNO_3$.

Table 3
Viscometric characteristics of NS in 0.5 M NaCl

	$[\eta]$ (ml g^{-1})	k'	C^* (g L^{-1})	Slope below C^*	Slope above C^*
HWS	840	0.86	1.55[a] 1.75[b]	1.4	2.6

[a] Experimental results.
[b] Results from the approximation $C^* = 1.5/[\eta]$ (Morris et al., 1980).

The other main aim of this work was to explore the semi-dilute rheological properties of the polysaccharide extracted from *O. ficus-indica*. As described above, the NS consists of less than 10% polysaccharide with high molecular weight (about 13×10^6 g mol^{-1}) and about 90% of low molecular weight material.

Regarding the results obtained from SEC/MALLS analysis of the water-soluble portion of the polysaccharide, we have chosen to characterise the rheological behaviour of the two different samples HWS and MS. This part aims to investigate the effect of purification degree on the whole rheological properties of the polysaccharide extracted from *O. ficus-indica*.

5.1. Rheological properties of purified sample HWS

The chemical structure of the polysaccharide as determined by sugar analysis shows the presence of negative charges along the polymer chain (carboxylic groups corresponding mainly to galacturonic and glucuronic acid residues) makes the chain conformation dependent on the salinity. The measurement of the salt dependence of the solution viscosity indicates a slight polyelectrolyte effect due to the presence of charges on the polymer backbone (Fig. 6) confirming the conductimetric data. When examining the viscosity loss of the solution before and after screening the charge effects, it is interesting to note that the loss does not exceed 10%, indicating a fairly limited extension of the polysaccharide chains in solution. This can be attributed to a low proportion of charged groups along the polymer chain and also probably to a slightly branched

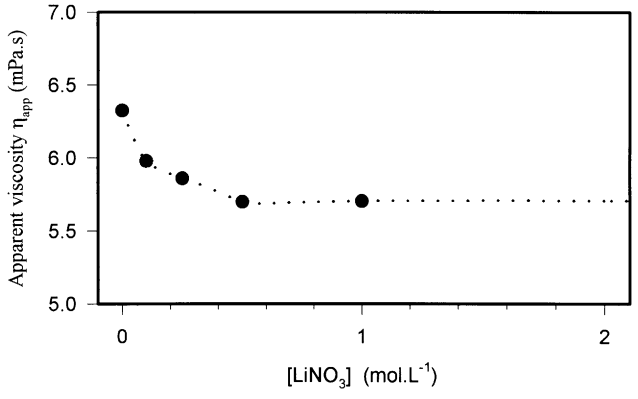

Fig. 6. Influence of ionic strength on the viscities of HWS aqueous solutions (2.5 g/l — salt used: $LiNO_3$ — 25°C).

structure. Total screening conditions have been fixed at 0.5 mol l^{-1} in NaCl but an ionic strength of 0.1 mol l^{-1} has been used for SEC–MALLS experiments for the reason already explained. In fact, Fig. 6 shows that there is no significant difference between viscosities at the two ionic strengths.

For each experiment the polymer solution behaviour is typical of viscous solutions. The zero shear viscosity value becomes higher as the polymer concentration rises, and at the same time the Newtonian plateau limit is shifted to the low shear region. As the system does not show thixotropy, it should be said that the viscometric properties are only related to physical entanglements between the polysaccharide chains in the solution as described by Ross-Murphy (1984).

Fig. 7 allows a comparison between the flow behaviour of HWS solutions (30 g l^{-1}) prepared in pure water and in the presence of 1 mol l^{-1} $LiNO_3$ or $CaCl_2$. When compared to the behaviour of the solution prepared in pure water, the loss of viscosity observed in the presence of monovalent cation (Li^+) is less marked than the one corresponding to the same solution in the presence of divalent cations (Ca^{2+}). This phenomenon can not only be attributed to screening effects of the added salt, but it can be interpreted as the result of the complex formation between the calcium ions and the carboxylic groups present along the polysaccharide backbone. This has been clearly established in the literature for carboxylated polysaccharides like pectins or alginates in the presence of divalent cations (e.g. Durand et al., 1990; Klein, Stock & Vorlop, 1983; Morris, Rees, Thom & Boyd, 1978; Morris, Powell, Gidley & Rees, 1982). Such properties can then be utilised in many industrial fields including removal of polluting metal for ions (Harel, de la Queriere, Mignot & Junter, 2000; Parker, Schram, Plude & Moore, 1996). In most of the cases described elsewhere the complexation induces a large increase of the viscosity or the establishment of a physical gel when sufficient intermolecular links can be formed (semi-dilute regime) (Dronnet, Renard, Axelos & Thibault, 1996; MacDougall, Needs, Rigby & Ring, 1996; Thibault & Rinaudo, 1985, 1986). However, the loss of viscosity observed here results from the high flexibility of the polysaccharide chains and from the low extent of charges, so that the complex formation mainly acts through intramolecular bonds inducing a more compact chain conformation.

Due to the intrinsic flexibility of the polysaccharide chains, the solution viscosity remains fairly low in spite of a large increase in the polymer concentration. For the most

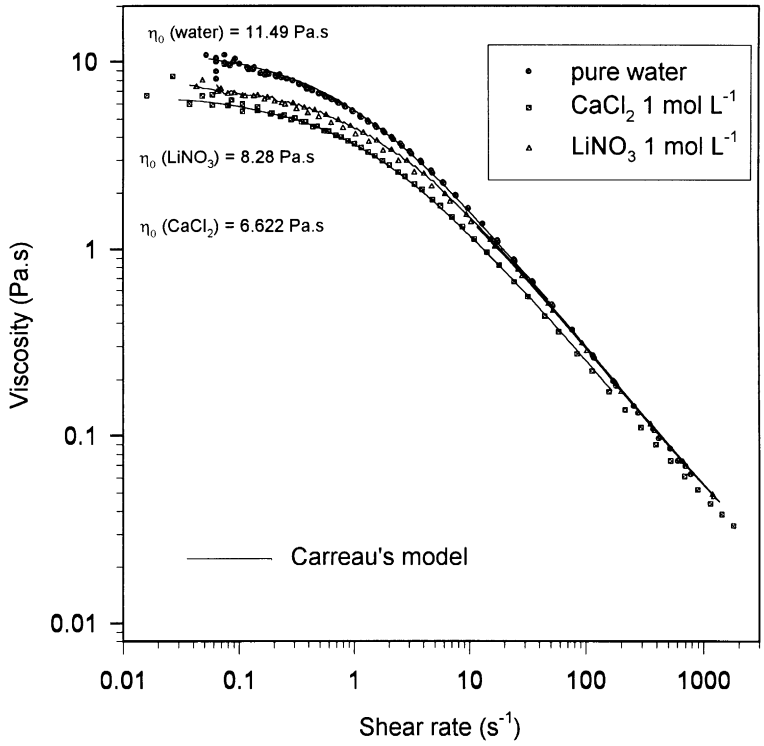

Fig. 7. Salt effect on the viscosity of HWS (30 g l^{-1} — 25°C).

concentrated solution the dynamic spectra (not shown here) are typical of a viscoelastic fluid, the elastic and the loss modulus being of the same order of magnitude in the frequency range studied, with a crossover point around 1 Hz.

Even in the entangled semi-dilute regime, the high flexibility of the chains results in a relatively low viscosity. Getting high viscosity systems would imply a considerable increase of the whole polysaccharide content in the media to over 10% by weight, thus this polysaccharide is of little interest as a viscosity agent.

The establishment of intramolecular complexes in the presence of $CaCl_2$ confirms the high flexibility of the polysaccharide chains, as already observed by Grisel (1996) for disordered schizophyllan polysaccharide in the presence of borate ions as a complexing agent. Due to the non-extended conformation of such a polymer in solution, the exclusive establishment of such "inactive" loops prevents the formation of intermolecular junctions, leading to a tri-dimensional network in the media corresponding to a physical gel. A non-negligible intrinsic rigidity of the polysaccharide then appears as a pre-requisite factor to get such properties. At the same time the loss of the intrinsic viscosity for HWS in the presence of the divalent salt ($CaCl_2$) is also observed as indicated in Table 4 confirming the previous results. Finally, for such a system, the type of interactions depends on the solvent conditions that influence the polymer configuration and the chain–chain interactions (Thibault & Rinaudo, 1985, 1986), and the loss of viscosity can also be interpreted as the result of salting out effects.

Table 4
Effect of adding divalent salt on the intrinsic viscosity of HWS as measured under low shear conditions (25°C)

Solvent conditions	$[\eta]$ (ml g^{-1})	k'
NaCl 0.5 M	820	0.9
$CaCl_2$ 0.5 M	740	0.6

Comparing the initial polymer composition (NS) (roughly 10% high molecular weight polysaccharide (about 13×10^6 g/mol) and 90% low molecular weight proteins) to the totally purified sample (HWS), it appears interesting to look at the effect of purification degree on the rheological behaviour of the system. For this reason, a partially purified sample MS was obtained using a short ultrafiltration period (24 h), and the rheological properties of this partially purified sample (MS) were compared to the ones of the totally protein free sample (HWS).

5.2. Influence of purification on the rheological properties of the polysaccharide

Fig. 8 illustrates the difference between HWS and MS as determined using SEC/MALLS experiments. As discussed earlier in this paper no proteins were present in HWS (no elution peak at high elution volume) while the MS sample still contains roughly 75% of protein-like species (presence of elution peaks at higher volumes). The corresponding sample is named as a Mix Sample (MS) for that reason. This observation concerning MS, the composition of

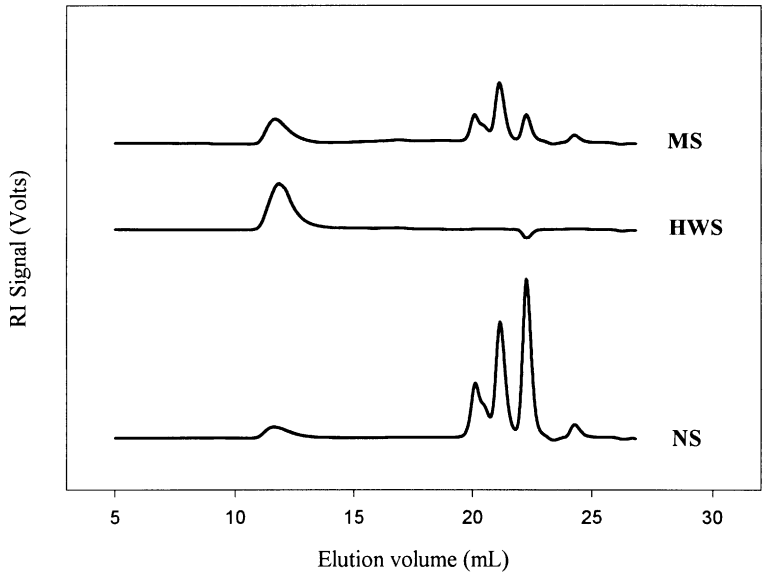

Fig. 8. SEC/MALLS elution profiles of MS and HWS in 0.1 M $LiNO_3$.

which is quite similar to NS, indicates that extensive ultrafiltration (during several days) has to be used to eliminate the rest of the protein species in order to get a highly pure polysaccharide containing sample (HWS).

Fig. 9 allows a comparison between HWS and MS samples dissolved in pure water at a concentration of 30 g l^{-1}. Both solutions show a remarkable shear thinning behaviour, but the HWS solution presents a much higher initial viscosity when compared to the MS. This can be easily explained by the large difference in polysaccharide amount between the two samples, this component being mainly responsible for the thickening behaviour of the solution. Both the solution flow curves can be modelled using Carreau's model Eq. (1)

$$\eta_r = \eta_{r0}/[1 + (\tau\dot{\gamma})^2]^m \quad (1)$$

to describe entangled polymer solution behaviour (Carreau, 1972). In this equation, η_r, η_{r0}, τ and m are, respectively, the relative viscosities of the solution, the relative

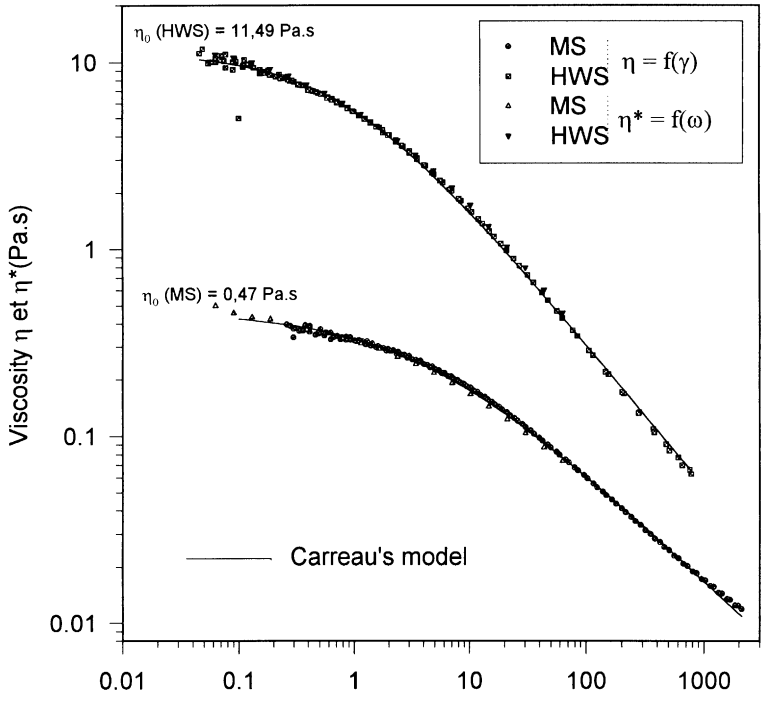

Fig. 9. Comparison of the flow properties of MS and HWS in water (30 g L^{-1} — 25°C).

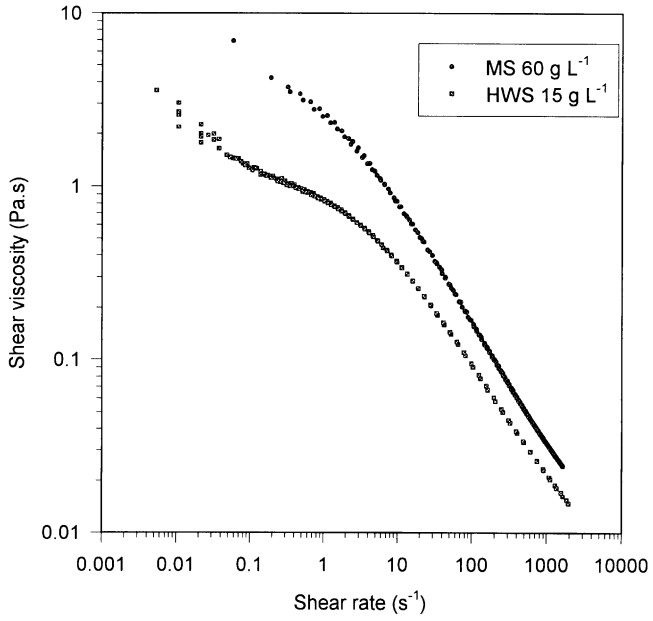

Fig. 10. Proteins influence on the viscometric behaviour of the polysaccharide in water (15 g l^{-1} polysaccharide content — 25°C).

Newtonian plateau viscosity and the relaxation time and the slope of the curve in the shear thinning regime. We also note a perfect superposition between the flow curves and the complex viscosity curves (obtained using oscillatory measurements), as predicted by the rule of Cox and Merz (1958). Furthermore, as mentioned earlier in this paper concerning HWS, MS also shows viscoelastic properties typical of entangled solutions.

As about 75% of the MS sample consists of low molecular weight proteins, it is of prime interest to compare the viscosity of MS and HWS solutions containing equal amount of polysaccharide species. From this point of view a 15 g l^{-1} HWS solution is equivalent to a 60 g l^{-1} MS one, both being prepared in water, but the flow behaviour observed reveals a large difference between the two solutions as indicated in Fig. 10. Proteins actually appear to greatly influence the viscosity of the system, probably because they interact with the polysaccharide backbone through physical associations, so that increases the whole solution viscosity by establishment of intermolecular physical links between the polymer chains. For this reason, it appears that the NS or the MS are probably much more interesting from an economic point of view and there is no point in carrying out the purification step.

6. Conclusion

In summary, the water-soluble function has been extracted from nopals of prickly pear of *ficus-indica*. It represented about 1% of the whole material. Extensive purification gives two distinct water-soluble components. The major component consists of an albumin-like family of proteins. In addition, a high molar mass and very flexible polysaccharide was isolated which was experimentally determined to be pectin. This potential interesting thickening agent needs extensive purification before practical use as it represents less than 10% of the water-soluble material.

The polysaccharide extracted from prickly pear nopals of *O. ficus-indica* revealed a good stability of viscosity of the solutions to added salts. This property can be viewed as a consequence of the low charge density of the polymer backbone, so that it is only slightly sensitive to electrostatic effects in spite of its high flexibility and can also be interpreted as the result of salting out effects. However, despite a high molecular weight this polysaccharide does not give high viscosities and therefore has little potential as a thickening additive in pharmaceutical, cosmetic or food applications. In addition, the purification of this product is lengthy, so at first sight it is of little economic interest.

An interesting approach that is now being considered due to the presence of proteins is to look at the potential emulsifying and/or stabilising properties as a result of the presence of proteins in the non-purified or partly purified material. The economic interest may then become much more evident as the polysaccharide may find applications in many fields of everyday life.

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