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Carbohydrate Polymers 63 (2006) 299–309

Carbohydrate Polymers

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Structural characteristics of gels formed by mixtures of carrageenan and mucilage gum from *Opuntia ficus indica*

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Received 17 December 2004; revised 6 April 2005; accepted 17 April 2005

Abstract

Sol–gel and gel–sol transitions of mixtures formed by mucilage gum from *Opuntia ficus indica* and either κ-carrageenan or ι-carrageenan were studied using differential scanning calorimetry (DSC) and scanning electron microscopy (SEM). A total polymer concentration of 2% (w/w), a mucilage concentration in the interval between 0 and 80% (w/w), and KCl or $CaCl_2$ concentrations of 12 or 60 mM, were used. Results from DSC showed that gelation was independent of the presence of mucilage gum in the mixtures. SEM results showed the non-gelling characteristic of mucilage gum from *O. ficus indica*. However, when mixed with κ-carrageenan a synergistic effect was observed only at the 80/20 κ-carrageenan/mucilage gum ratio. The DSC and SEM results indicated that the mucilage gum gets adsorbed onto the *i*-carrageenan structure in an interaction controlled by electrostatic charges. Regarding the *i*-carradeenan/mucilage mixtures no clear interaction at structural level was observed.

Keywords: Carrageenan; *Opuntia ficus indica*; Gelation; DSC; SEM; Structure

1. Introduction

The carrageenans are linear sulfated polysacchrides, which molecules may include up to 1000 residues providing many possibilities for structural variations (Stanley, 1990). ι-and ι-carrageenan form the thermo-reversible gels, whereas λ-carrageenan does not. Gelation in the κ- and ι-carrageenan solutions are the result of a coil-to-helix molecular transition followed by aggregation that occur upon cooling. However, the exact gelation mechanism has not been established (Stanley, 1990). What is well known is that gel strength and gelation temperature both depend on the ionic strength, i.e. κ-carrageenan is sensitive to K^+ and ι-carrageenan to Ca^{++}. On the other hand, λ-carrageenan is not able to go through the coil-to-helix transition and remains as a random coil in aqueous solutions without forming gels (De-Ruiter and Rudolph, 1997).

From the studies of Picullel (1998) it is now accepted that monovalent counter-ions induce a coil to helix transition leading to a double-helix conformation before gelation takes place. On the other hand Kara et al. (2003) showed that the increase in the KCl/carrageenan ratio raised both transition temperatures and the enthalpy value for the sol-to-gel transition. These authors also reported the formation of stronger gels in the presence of KCl. In the same way MacArtain et al. (2003), examined the effect of divalent counter-ions on the initial coil to helix transition of κ-carrageenan gels. Their results showed that extensive neutralization of the carrageenan by excess of divalent calcium ions produce aggregation of the polymer strands in a precipitation-like process. At lower counter-ion to polymer ratios, the onset of gelation might prevent such phase separation.

More particular information has shown that ι-carrageenan, with additional sulfated groups in the anhydrous galactose residues, displays greater hydrophilic

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0144-8617/$ - see front matter © 2005 Published by Elsevier Ltd.
doi:10.1016/j.carbpol.2005.04.022

properties than κ-carrageenan (Stanley, 1990). This chemical characteristic enhances its ability to inhibit syneresis, although its gels are less rigid than those of κ-carrageenan owing to its lower aggregation capacity. Because of its higher degree of substitution by sulphate groups ι-carrageenan shows a lower degree of ion specificity. Nevertheless, some reports have mentioned that its ion specificity is the result of κ-carrageenan impurities, a common feature of commercial samples (Hugerth and Sundelöf, 2001).

Nowadays, commercial mixtures of bio-polymers with functional properties specifically designed to meet industrial needs are available in the market. However, the lack of scientific grounds to explain their functional properties is a common feature of commercial technical literature. On the side of the carrageenan most of the available scientific reports are for the κ-carrageenan/Locust bean gum (LBG). This system has been studied because the properties of the κ-carrageenan gels, usually brittle, turbid and prone to syneresis, can be improved by the presence of galactomannans like LBG, phenomenon known-as synergism (Morris, 1990). Thus, κ-carrageenan alone does not gel in the presence of 100 mM NaCl, but in a 0.9/0.1 (% w/w) κ-carrageenan/LBG mixture a weak gel forms (Gonçalves et al., 1997). Increasing NaCl concentration up to 200 mM a strong gel was produced and the gel strength increased on adding small proportions of LBG. Lundin and Hermansson (1997), from rheological measurements confirmed the synergistic effect of the κ-carrageenan/LBG system. In contrast, Dunstan, Chen, Liao, Salvatore, Boger and Prica (2001) did not observe a maximum on the Young's modulus for κ-carrageenan/LBG blends, as expected if synergism is present. It seems that variations in the molecular weights and purity of both κ-carrageenan and LBG, explain the synergism observed by many other researchers. More experimental evidence is needed for a better understanding of the synergy phenomenon.

Recently, our group reported the rheological properties of the mucilage gum isolated from *Opuntia ficus indica*, a member of the Cactaceae family (Medina-Torres, et al., 2000). The results showed that at low mucilage concentration (<3%) the behavior is typical of a dilute solution. However, with increasing concentration, the solution shows a weak gel behavior. Viscous properties of mucilage gum are dependent on ionic strength. Thus, an increase in salt concentration results in a decrease in the shear-dependent viscosity, as expected for poly-electrolytes. Viscosity reduction is more dependent on Ca^{++} and Mg^{++} than on Na^{+} or K^{+}. Important elastic properties were also found for this mucilage gum. In a different work Medina-Torres et al. (2003) investigated the mechanical properties of gels formed by mixtures of mucilage gum from *O. ficus indica* and κ- or ι-carrageenan. For the mixed gels of ι-carrageenan and mucilage gum, no enhancement of the mechanical properties with respect to the pure ι-carrageenan was observed. In contrast, for the ι-carrageenan/mucilage gum system, at an 80/20 (w/w) ratio and in the presence of 12 mM KCl, a synergistic interaction was observed in gel elasticity.

Following with our work the objective of this work is to investigate the gelation and synergistic mechanism of mixtures of κ- or ι-carrageenan with mucilage gum from *O. ficus indica*. A better understanding of the mucilage gum/carrageenans interactions would help to describe the general synergistic effect and the particular rheological properties of this and others hydrocolloid mixture systems. Thermal and micro-structure results would allow a better evaluation and interpretation on the role each polysaccharide is playing in the functional and rheological properties of the final gels. Consequently, this study evaluated the time-temperature gelation process through differential scanning calorimetry (DSC) coupled with scanning electron microscopy (SEM) measurements.

2. Materials and methods

2.1. Sample preparation and experimental setup

The extraction process of the mucilage gum from *O. ficus indica* has been reported elsewhere (Medina-Torres et al., 2000). Food grade commercial preparations of the ι- (Genuvisco Co.) and κ-carrageenan (FMC-911, FMC Co.) were used without further purification. According to the suppliers, the κ-carrageenan sample contained at least 90% of the bio-polymer and approximately 10% of ι-carrageenan, while the ι-carrageenan was 85% pure with the remaining of κ-carrageenan. Details on the gel preparation procedure have also been reported elsewhere (Medina-Torres et al., 2003). Gels with a total hydrocolloid concentration of 2% (w/w) were used at the following κ- or ι-carrageenan/mucilage ratios: 100/0, 80/20, 60/40 and 50/50% (w/w). The mixture of polysaccharides was dispersed through magnetic stirring during 10 min at room temperature and then heated up to 75 °C with agitation for additional 20 min to assure full dispersion. Then, KCl or $CaCl_2$ was added to the dispersion to achieve 12 or 60 mM concentration. KCl was used for the κ-carrageenan/mucilage system and $CaCl_2$ for the ι-carrageenan/mucilage system.

2.2. Differential scanning calorimetry

DSC analysis was done with a DSC-7 calorimeter (Perkin-Elmer, Norwalk, CT, E.U.A.), previously calibrated with Indium (melting temperature = 156.6 °C, melting heat = 28.45 J/g) and equipped with Perkin-Elmer DSC pan cells 02190062. An empty pan was used as reference to develop the baseline form 20 to 140 °C. The sample (18 ± 0.6 mg), previously weighted in aluminum pans, was initially heated to 80 °C for 30 min in the corresponding thermocell of the DSC and then analyzed according to the following program: (a) cooling from 80 to 20 °C and maintaining this temperature for 30 min. (i.e. cooling stage), and (b) heating from 20 to 80 °C and maintaining the system at this temperature for 5 min. (i.e. heating stage). In all stages the heating/cooling rate used was 5 °C/min. Temperatures for the different transitions (i.e. the onset temperature, T_0; peak temperature, T_p; ending temperature, T_e) were determined using the first derivative of the heat capacity calculated with the DSC program library and by comparison with the baseline. On the other hand, enthalpy (ΔH, J/g) for each transition was estimated integrating the corresponding endothermic or exothermic peak, respectively. The limits of integration, X_1 and X_2, were established using the first derivative of the heat capacity as described by Toro-Vazquez, Gomez-Aldapa, Aragon-Pina, Brito-De La Fuente, Dibildox and Charo-Alonso (2003). For the samples containing mixtures of polysaccharides, only one endotherm, (i.e. the peak corresponding to the carregenan 100% used) was considered for the estimation of the enthalpies during the heating cycle.

2.3. Scanning electron microscopy

Sample preparation for SEM measurements has been described elsewhere (Toro-Vazquez et al., 2003). Essentially, after the DSC cooling process the pan was opened and the material transferred to a test tube and frozen quickly to −20 °C with a freezer (Ultra-low temperature, Ultima, Revco). After 3 h at this temperature, the sample was freeze-dried (−40 °C, 50 mbar for 3 h). The lyophilized sample was placed onto an aluminum slide using electrically conductive tape (Bal-Tec, Fürstentum Liechtenstein, Germany) and coated with gold at 10 mbar for 90 s (Polaron SC-7610, Fisson Instruments, CA, USA). Then, samples were examined and photographed with an electron microscope Leica Stereoscan S420i (Cambridge, England).

3. Results and discussion

3.1. Differential scanning calorimetry

Figs. 1 and 2 show the cooling and heating DSC curves as a function of the κ-carrageenan/mucilage ratios at 12 and 60 mM of KCl, respectively. As expected, the thermal response for each blend depended on the hydrocolloid ratio and on the KCl concentration. The thermograms at both 12 and 60 mM KCl showed the evidence of gelation for the κ-carrageenan alone and for the mixtures, i.e. occurrence of an exothermic peak on cooling and an endothermic peak on heating. Independent of the KCl concentration in the system, the cooling thermograms (Figs. 1(a) and 2(a)) showed the presence of just one exotherm at all κ-carrageenan/mucilage ratios investigated. On the other hand, in the heating thermograms at both KCL concentrations (Figs. 1(b) and 2(b)) we obtained a broad single endotherm for κ-carrageenan (i.e. κ-carrageenan/mucilage ratios of 100/0) as well as for the hydrocolloid mixtures. A small apparent exotherm at 52 °C for the mixtures may also be observed (see Fig. 1(a)), but it is so small that it is probably related to the sensitivity of the equipment. This feature was not seen when ι-carrageenan was used.

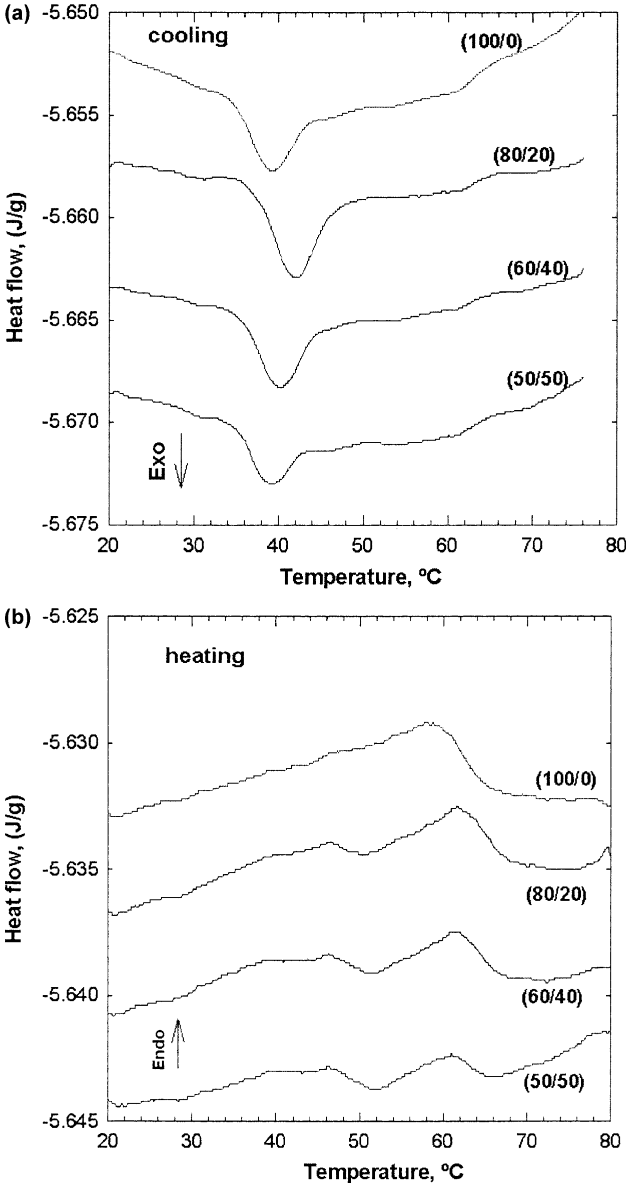

Fig. 1. DSC thermograms during cooling (a) and heating (b) for 2% (w/w) mixtures of κ-carrageenan and mucilage gum at different ratios. The KCl concentration was 12 mM.

These DSC results showed that in the κ-carrageenan/mucilage mixtures the hydrocolloids developed a gel under cooling at the same temperature conditions (i.e. a single exotherm was obtained). These results also suggest that the mucilage increases the transition temperature, effect that can be clearly seen at the 80/20 ratio (see Fig. 1(b)). This last effect is also due to the presence of KCL in the system (compare heating thermograms at

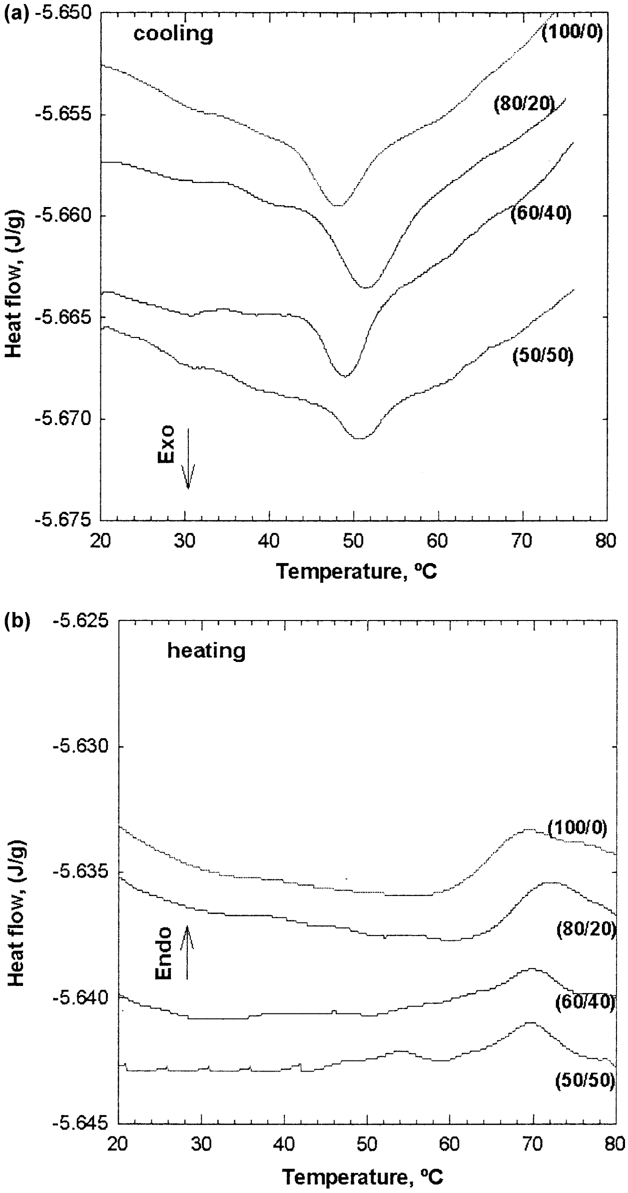

Fig. 2. DSC thermograms during cooling (a) and heating (b) for 2% (w/w) mixtures of κ-carrageenan and mucilage gum at different ratios. The KCl concentration was 60 mM.

12 mM KCl in Fig. 1(b) and at 60 mM KCl in Fig. 2(b)). It is well known that κ-carrageenan is sensitive to K^+. Thus, it has been shown that increases in the KCl concentration raise both the transition temperature and the enthalpy for the sol-to-gel transition for κ-carrageenan (Rochas and Rinaudo., 1984; Standing and Hermansson., 1993; Kara et al., 2003).

The peak temperature for the sol–gel (T_C) and the gel–sol transition (T_H) as a function of the κ-carrageenan concentration in the mixture are shown in Fig. 3. The corresponding heat of transition, ΔH_C and ΔH_H, also as a function of the κ-carrageenan concentration in the mixture are shown in Fig. 4. At all hydrocolloid ratios and at both KCl concentrations investigated the sol–gel peak temperature (T_C), also known as the gel point temperature, was always lower than the corresponding gel–sol transition (T_H) value. This observation shows that the gel developed by the hydrocolloid mixture at the different ratios did not go back to the sol state at the gelation temperature, a phenomenon known as hysteresis and regularly reported for hydrocolloids gelation mechanism (Watase and Nishinari, 1993; Kohyama et al., 1996; Gonçalves et al., 1997).

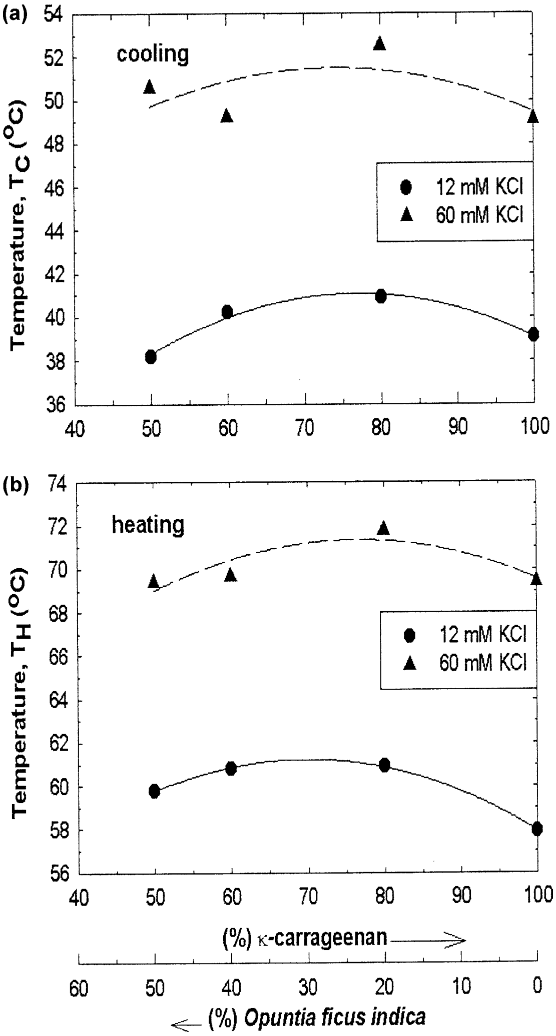

Fig. 3. Transition temperatures during cooling (a) and heating (b) for the 2% (w/w) mixtures of κ-carrageenan and mucilage gum at different ratios and KCl concentrations of 12 and 60 mM. Data points represent the mean of at least two determination. The solid lines is the least square fitting.

On the other hand, independent of the κ-carrageenan concentration higher T_C and T_H values were observed at 60 mM KCl than at 12 mM KCl. These results agree with the ones reported by Kara et al. (2003); Kohyama et al. (1996), who observed higher T_H values in the presence of KCl for κ-carrageenan and mixed systems composed of κ-carrageenan with different molecular weights of konjac glucomannan. In the same way, both T_C and T_H observed a maximum at the 80/20 κ-carrageenan/mucilage ratio, a maximum that was more evident at 60 mM KCl than at 12 mM KCl. These results suggested that at this ratio a

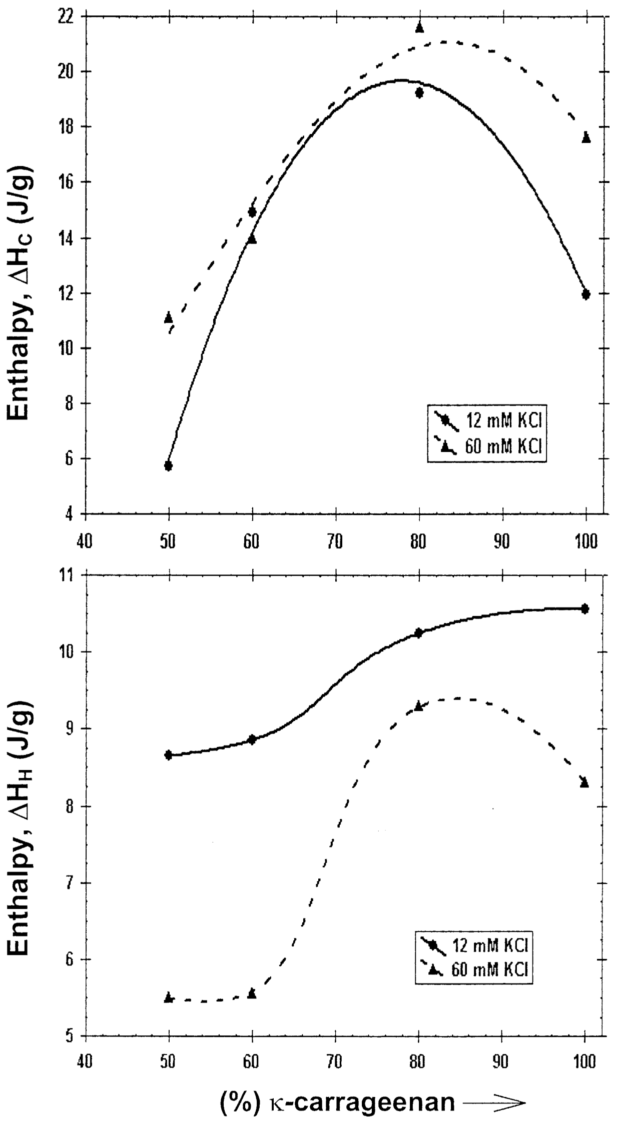

Fig. 4. Transition enthalpy during cooling (a) and heating (b) for the 2% (w/w) mixtures of κ-carrageenan and mucilage gum at different ratios and KCl concentrations of 12 and 60 mM. Data points represent the mean of at least two determination. The solid lines is the least square fitting.

synergistic effect occurred between the hydrocolloids. From theological measurements Medina-Tortes et al., (2003), in κ-carrageenan/*O. ficus indica* mucilage mixtures, and Goncalves et al., (1997) in κ-carrageenan/locus bean gum mixtures, reported synergistic interactions at particular κ-carrageenan/mucilage (i.e. 80/20) and κ-carrageenan/locus bean gum, ratios. In the case of κ-carrageenan/locus bean gum mixtures the authors explained these results considering that locus bean gum, a galactomannan, adsorbs onto carrageenan aggregates (Goncalves et al., 1997). Since the mucilage gum from *O. ficus indica* is also a galactomannan, the enhanced strength of gels developed with an 80/20 mixture of κ-carrageenan/mucilage gum in comparison with gels obtained just with κ-carrageenan (Medina-Torres et al., 2003), might be explained considering that the mucilage gum gets adsorbed onto the carrageenan aggregates resulting in higher rheological properties. This adsorption might be promoted by the presence of KCl in the system.

On the other hand, from an energetic point of view, the sol–gel and gel–sol transitions are characterized by an activation energy known as sol–gel enthalpy (ΔH_C) and gel–sol enthalpy (ΔH_H), respectively. Such values were determined for the hydrocolloid mixtures investigated through the cooling and heating DSC thermograms (Figs. 1 and 2). Fig. 4 shows the ΔH_C and ΔH_H as a function of the κ-carrageenan concentration in the mixture at concentrations of 12 and 60 mM KCl. Overall, it is well known that gel formation (i.e. sol–gel transition) needs more energy than gel melting (i.e. gel–sol transition) (Rochas and Rinaudo, 1984; Kara et al., 2003). Thus, more energy is needed to form double helices and the subsequent aggregates than to destroy them. According to these observations the results obtained showed that all κ-carrageenan/mucilage ratios ΔH_C was greater than ΔH_H, particularly at the highest KCl concentration. From Fig. 4 it is clear that, excepting ΔH_H at 12 mM KCl, both ΔH_C and ΔH_H clearly observed a maximum at the 80/20 κ-carrageenan/mucilage ratio. In ΔH_H at 12 mM KCl the values at the 100/0 and the 80/20 κ-carrageenan/mucilage ratios were statistically the same ($P < 0.05$). In all cases, after the 80/20 κ-carrageenan/mucilage ratio the ΔH_C and ΔH_H values decreased as κ-carrageenan concentration decreased in the mixture, observing values equal or smaller than the ones observed by κ-carrageenan alone (Fig. 4).

From rheology and microstructure studies on κ-carrageenan/LBG systems, Lundin and Hermansson (1997) concluded that LBG did not influence the molecular structure of κ-carrageenan to any large extent at least in the presence of sodium ions. The authors concluded that the ability of LBG to influence the microstructure is dependent on the mannose to galactose ratio of the galactomannans, the relative concentration and the ionic level. In the Lundin and Hermansson study (1997) ΔH_C increased as the κ-carrageenan concentration increased. For our system, both ΔH_C and ΔH_H for the mixtures were smaller than those for κ-carrageenan alone with the exception of the ratio 80/20, this last observing higher values (Fig. 4(a) and (b)). This last observation pointed out again the occurrence of synergism at the 80/20 κ-carrageenan/mucilage ratio.

Regarding the ι-carrageenan/mucilage system Figs. 5 and 6 show the cooling and heating DSC thermograms for the different ι-carrageenan/mucilage ratios at $CaCl_2$ concentrations of 12 and 60 mM respectively. The thermograms show in all cases the evidence of gelation, i.e. an exothermic peak on cooling followed for an endothermic peak on heating, independent of the $CaCl_2$ concentration. The corresponding sol–gel (T_C) and the gel–sol (T_H) transition temperatures as a function of the ι-carrageenan concentration in the hydrocolloid mixture are shown in Fig. 7. As for the κ-carrageenan/mucilage system (Fig. 3) the melting or gel–sol transition

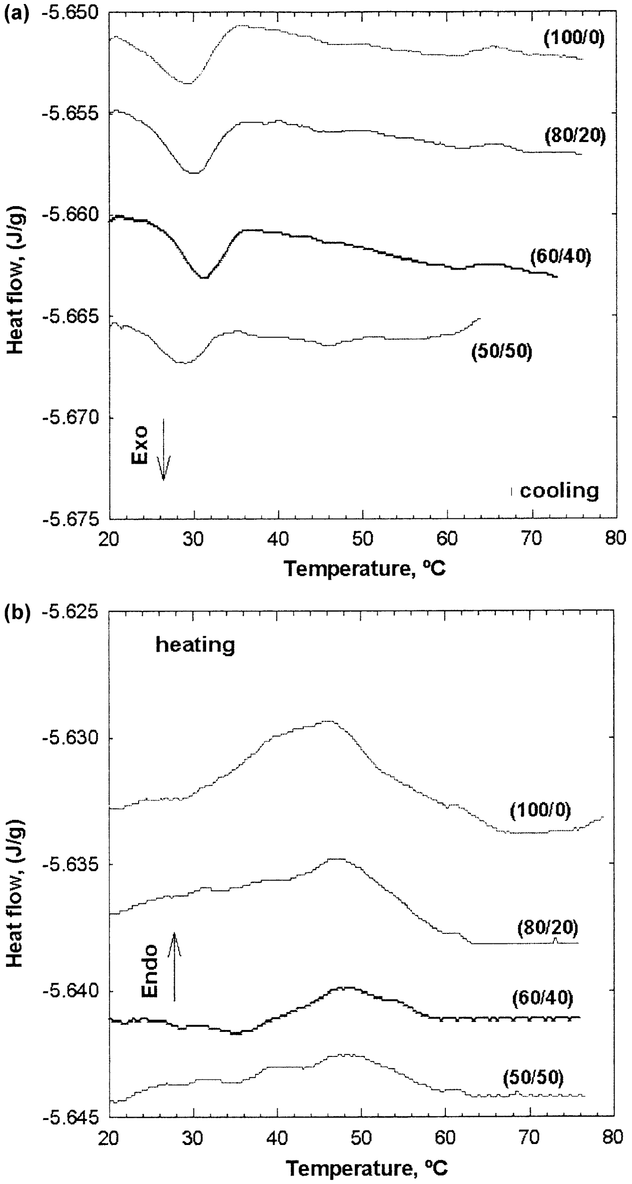

Fig. 5. DSC thermograms during cooling (a) and heating (b) for 2% (w/w) mixtures of ι-carrageenan and mucilage gum at different ratios. The $CaCl_2$ concentration was 12 mM.

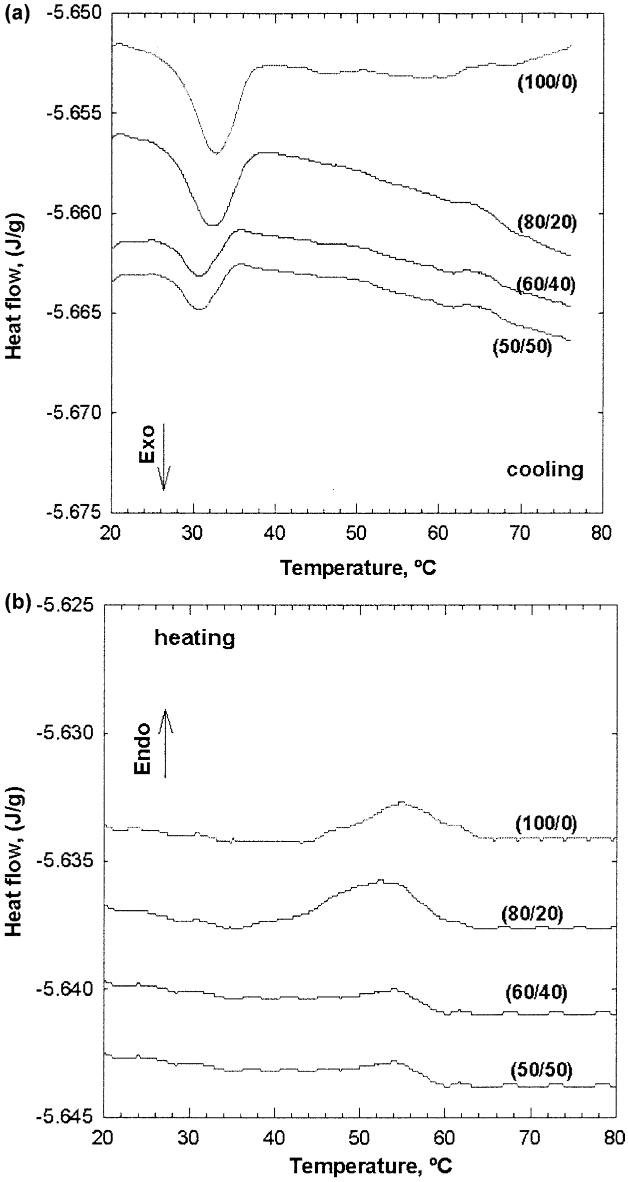

Fig. 6. DSC thermograms during cooling (a) and heating (b) for 2% (w/w) mixtures of ι-carrageenan and mucilage gum at different ratios. The $CaCl_2$ concentration was 60 mM.

temperatures (Fig. 7(b)) were consistently higher than gelling temperatures (Fig. 7(a)), i.e. hysteresis (Gonçalves et al., 1997; Piculell et al., 1998). For these systems, T_C and T_H are almost independent of the concentration of ι-carrageenan present or of the presence of the mucilage gum in the mixture, but dependent on the ionic strength. As the $CaCl_2$ concentration increases, a shift to higher temperature values for both transition temperatures is observed. However, the increase was more pronounced in the case of the gel–sol transition (Fig. 7(b)).

These results are in contrast with the ones reported by van de Velde et al. (2002) who observed no hysterisis with pure ι-carrageenan, suggesting no inter-helical aggregation during gelation. However, comparison of our results with others reported in the literature is difficult, because ι-carrageenan is commonly contaminated with κ-carrageenan. For instance, pure ι-carrageenan shows no cation specificity but some samples may show ion affinity as a consequence of κ-carrageenan impurities (Hugerth and Sundelöf., 2001). In contrast with the κ-carrageenan/mucilage mixture the ι-carrageenan/mucilage system no maximum was observed in the T_C or T_H (Fig. 7). In fact, T_H was practically independent of the ι-carrageenan/mucilage ratio (Fig. 7(b)). However, T_C showed a linear direct relationship ($P<0.10$) with the ι-carrageenan/mucilage ratio (Fig. 7(a)).

The energy involved in the transitions for the ι-carrageenan/mucilage system is shown in Fig. 8. Again,

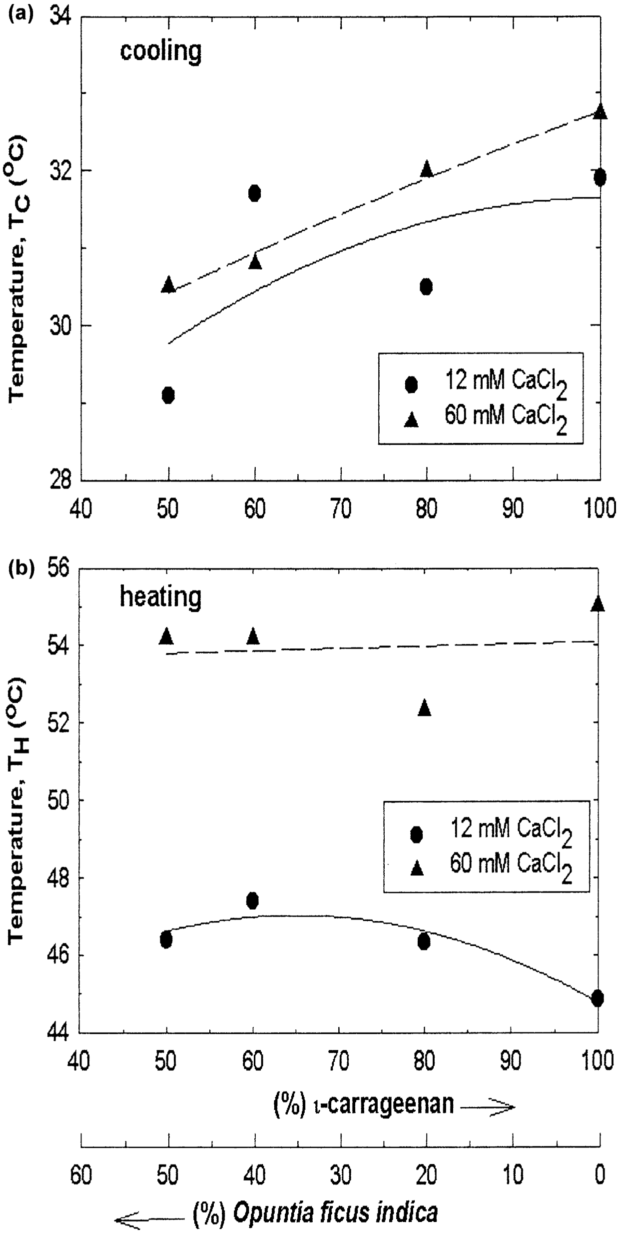

Fig. 7. Transition temperatures during cooling (a) and heating (b) for the 2% (w/w) mixtures of ι-carrageenan and mucilage gum at different ratios and $CaCl_2$ concentrations of 12 and 60 mM. Data points represent the mean of at least two determination. The solid lines is the least square fitting.

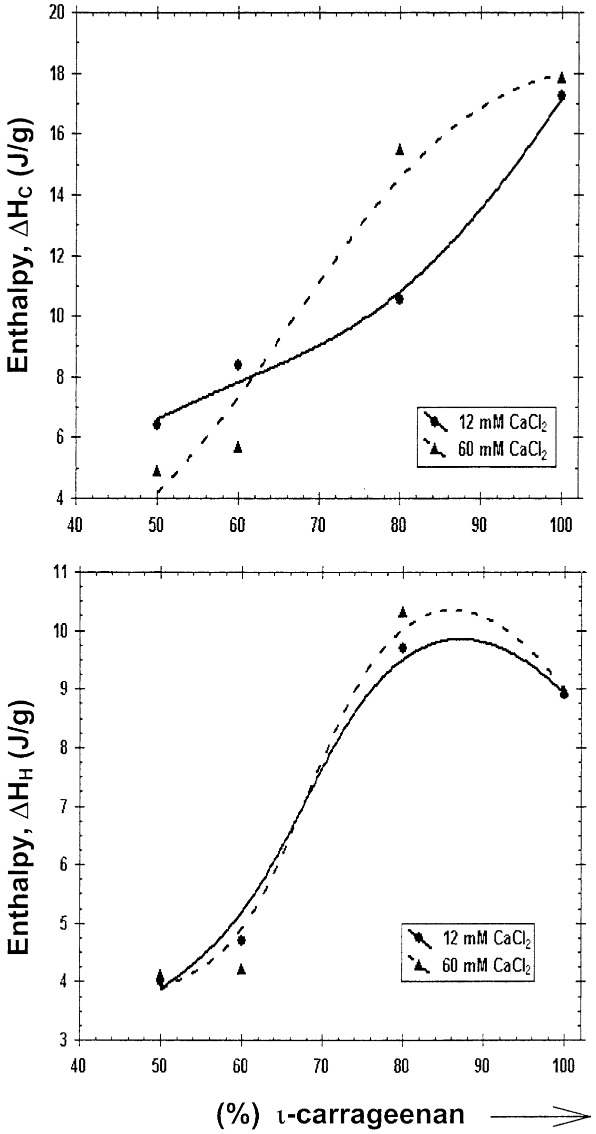

Fig. 8. Transition enthalpy during cooling (a) and heating (b) for the 2% (w/w) mixtures of ι-carrageenan and mucilage gum at different ratios and $CaCl_2$ concentrations of 12 mM and 60 mM. Data points represent the mean of at least two determination. The solid lines is the least square fitting.

as in the κ-carrageenan/mucilage mixture and according to previous reports (Rochas and Rinaudo, 1984; Kara et al., 2003), independent of the $CaCl_2$ concentration ΔH_C was higher than ΔH_H. On the other hand and in contrast with the κ-carrageenan/mucilage mixture, it was evident that at both $CaCl_2$ concentrations investigated ΔH_C decreased as the ι-carrageenan concentration decreased in the mixture (Fig. 8(a)). Then, for the ι-carrageenan/mucilage system, some important differences were noted with respect to the κ-carrageenan/mucilage mixture. First, there was no a significant peak for the 80/20 ι-carrageenan/mucilage ratio as it was observed for the κ-carrageenan/mucilage mixture, and second, the energy required for gelation in the κ-carrageenan/mucilage samples (see Fig. 4(a)) was higher than that for the ι-carrageenan/mucilage mixtures (see Fig. 8(a)).

Comparing the results obtained with the carrageenan/-mucilage systems investigated is evident that T_C, T_H, ΔH_C, and ΔH_H were always higher with κ-carrageenan than with ι-carrageenan, particularly at the 80/20 carrageenan/mucilage ratio. Then, the energy required for gelation of the κ-carrageenan/mucilage mixture is higher than that for the ι-carrageenan/mucilage mixtures. These results agree with previous studies that showed enhancement in the rheological properties of the 80/20 κ-carrageenan/*O. ficus indica* mucilage mixture, while no enhancement was observed in the rheology of gels prepared with mixtures of ι-carrageenan and mucilage gum (Medina-Torres et al., 2003). This behavior pointed out important differences in the conformational transition mechanism between the κ-carrageenan and the ι-carrageenan. Thus, it has been proposed that the sol–gel thermally induced process for the pure ι-carrageenan follows a no specific coil-to-helix cation conformational transition (van de Velde et al., 2002) as for κ-carrageenan. On the other hand, the gel forming mechanism in ι-carrageenan seems to take place at the helical level with further association of double helices. This last process is less marked than in κ-carrageenan, as confirmed from small-angle X-ray scattering studies by Yuguchi et al., (2002, 2003). These last authors also proposed that ι-carrageenan tend to form double-stranded helices in spite of the electrostatic interaction. If the aggregation of the already formed double helices is less marked, then it means that less energy is involved in the gelation process for ι-carrageenan, as compared with that required for κ-carrageenan, where double helices need to be developed. This may explain the lower ΔH_C values and the subsequent lower ΔH_H obtained for the ι-carrageenan/mucilage system as compared with the ones obtained for κ-carrageenan/mucilage system.

3.2. Scanning electron microscopy (SEM)

Scanning electron microphotographs for the carrageenan/*O. ficus indica* mucilage systems investigated are shown in Figs. 9–12. Thus, Fig. 9 shows the corresponding microphotographs for the 2% (w/w) κ-carrageenan gels as a function of the KCl concentration. The microphotographs show clear strands forming a network of open pores, which pore size decreased as KCl concentration increased from 0 to 12 mM up to 60 mM KCl. Then, as KCl concentration increased, the network became more dense. These observations correspond to gels with higher failure stress and failure strain as KCl concentration increased, i.e. gels get stronger and more deformable as compared with those formed without KCl (Medina-Torres et al., 2003). On the other hand, Mao et al. (2000) reported a similar tendency for mixed gellan gels with calcium concentration.

The microphotographs for *O. ficus indica* mucilage gum are shown in Fig. 10. The mucilage gum did not form a clear gel-like structure as the one observed for κ-carrageenan. The microphotographs presented a macromolecular dispersion that became less agglomerated as KCl concentration increased (Fig. 10). These results confirm the non-gelling nature of the *O. ficus indica* mucilage gum, at least in the concentration range used here as previously reported from rheological measurements (Medina et al., 2000). On the other hand, the change in the aggregation state of the mucilage gum as KCL is added can be explained by considering the polyelectrolyte behavior of this polysaccharide, as described by McGarvie & Parolis, (1981). It is interesting to note here that the chemical compositions of Opuntia mucilages have been described by several research groups, with important contradictions as reviewed by Melainine, Dufresne, Dupeyre, Mahrouz, Voung and Vignon (2003). Early in the 1900's, some authors presented evidence on the neutral character of the mucilage but more recent reports have shown it has acidic residues and thus polyelectrolyte behavior (McGarvie & Parolis, 1981; Medina et al., 2000). Finally, some new findings suggest that mucilage may have both neutral and acidic fractions depending on the extraction method used (Melainine et al., 2003).

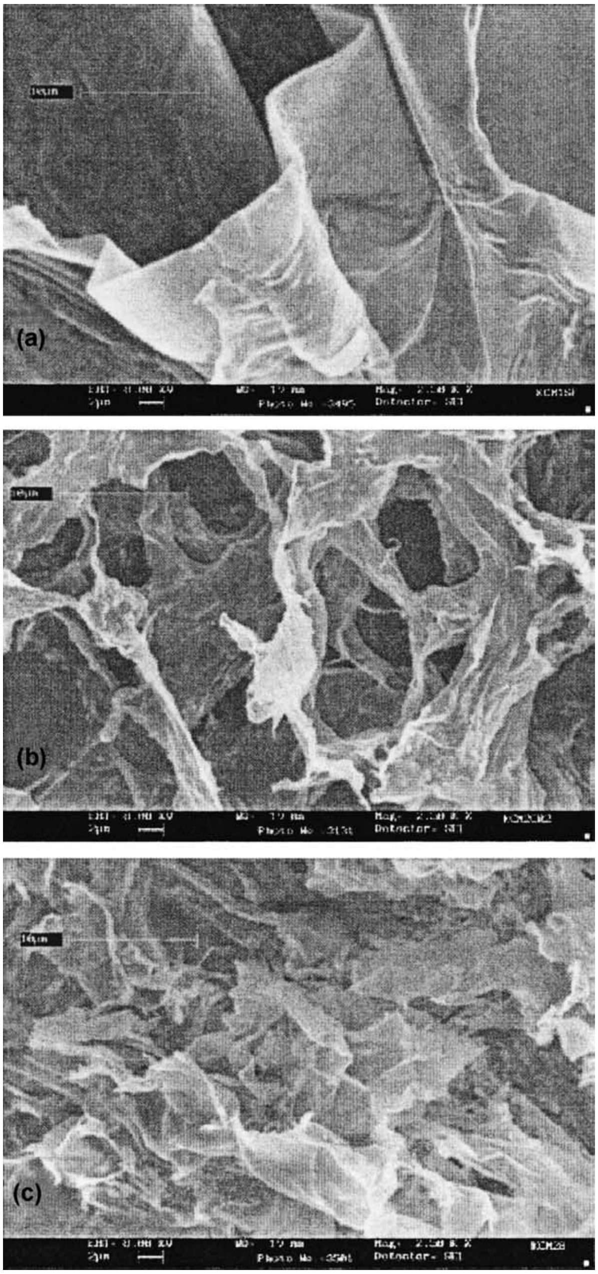

Fig. 9. SEM images (2500×) for 2% (w/w) κ-carrageenan without KCl (a), 12 mM KCl (b), and 60 mM KCl (c).

The mixed hydrocolloid system at 80/20 κ-carrageenan/mucilage ratio presented a completely

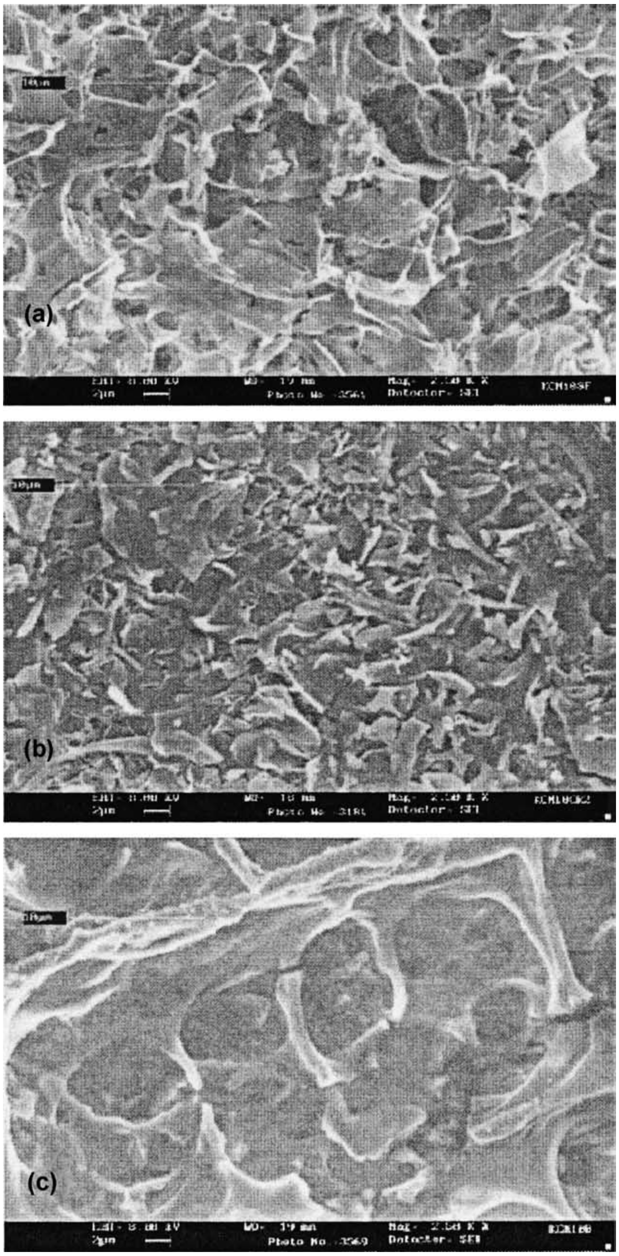

Fig. 10. SEM images (2500×) for 2% (w/w) mucilage gum from *Opuntia ficus indica*. Without KCl (a), 12 mM KCl (b), and 60 mM KCl (c).

different structural organization (Fig. 11) as the one observed with κ-carrageenan or mucilage alone. From the microphotographs it was evident that κ-carrageenan was the dominant structural component, and that mucilage did not form an integral part of the structure. The mucilage was aggregated on the surface of the structure of κ-carrageenan. As the DSC melting studies indicated (Fig. 1(b)), each biopolymer formed its own junction zone and no interaction between chains occurred. Then, the co-existence of two types of microstructure was confirmed by SEM observations. Similar behavior has been reported for a κ-carrageenan/LBG system (Fernandes et al., 1994). The previous stated hypothesis that mucilage gum adsorbs onto the carrageenan aggregates and that the interaction is only controlled by the electrostatic charge of the carrageenan helices, is then supported by these results.

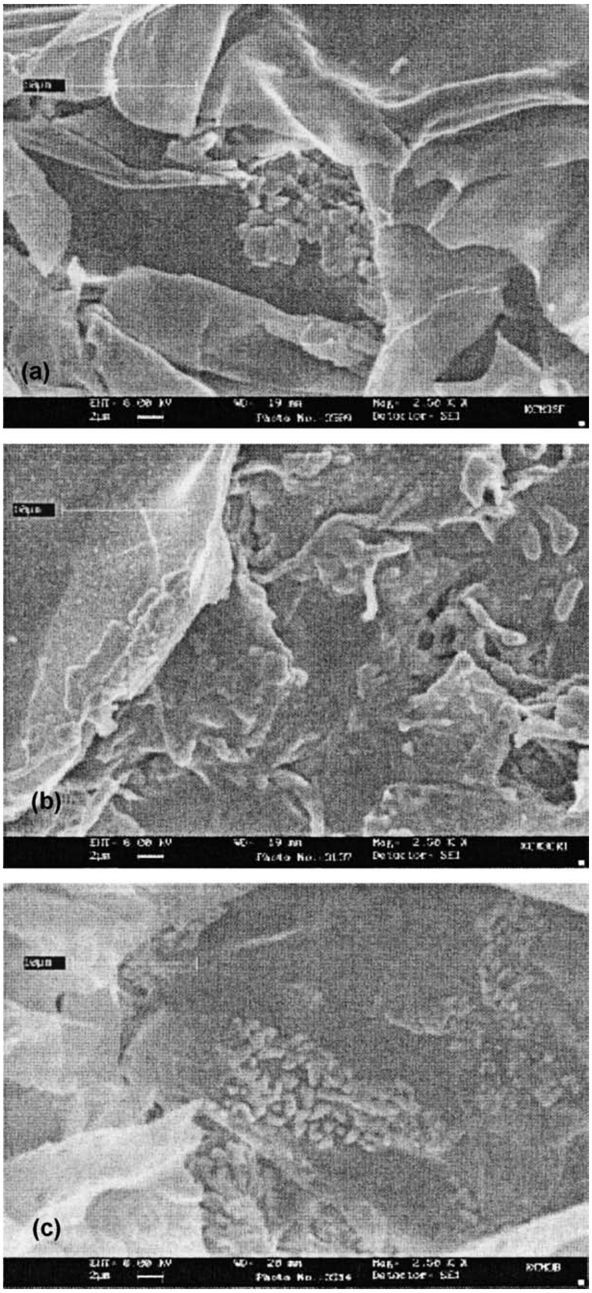

Fig. 11. SEM images (2500×) for 2% (w/w) 80/20 κ-carrageenan/mucilage gum mixture at different KCl concentration. Without KCl (a), 12 mM KCl (b), and 60 mM KCl (c).

SEM micrographs a 80/20 κ-carrageenan/mucilage ratio at different KCl concentration are presented in the Fig. 11, which pore size decreased as KCl concentration increased. The minimum size of pore was observed at gels 0 and 12 mM concentration of KCl. The small pore structure may also be responsible for the stability of the texture properties (Medina et al., 2003). A comparison between Figs. 9 and 11 and 60/40 κ-carrageenan/mucilage ratio (at 60/40 ratio, date

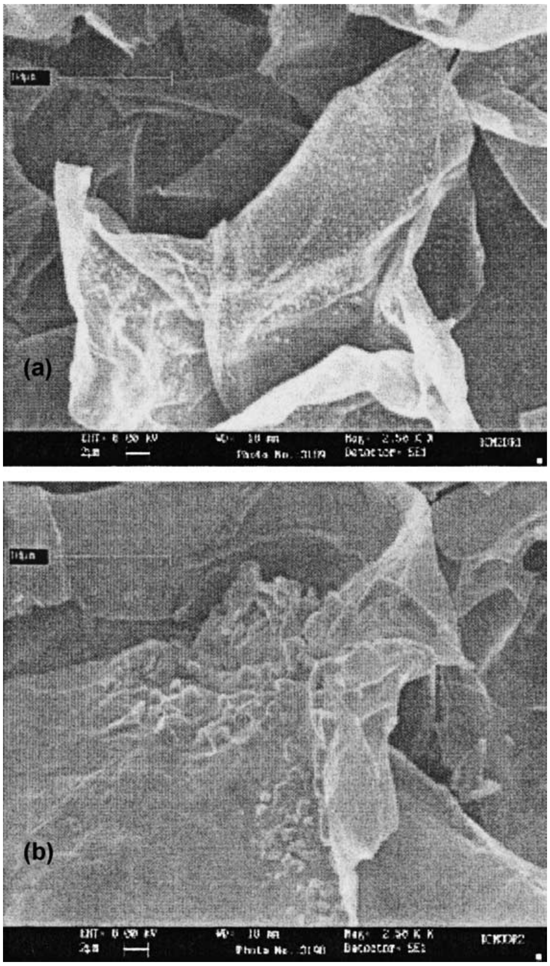

Fig. 12. SEM images (2500×) for gels formed with 2% (w/w) ι-carrageenan alone (a) and 80/20 ι-carrageenan/mucilage gum ratio (b), both at 12 mM $CaCl_2$.

not was presented here) shown a more compact structure when the mucilage gum is not present, thus a decrease in the characteristic pore size of the network as the mucilage gum concentration increases. This behavior is similar to the one observed by Dunstan et al. (2001) for mixed systems of κ-carrageenan and LBG. The synergistic effect of the LBG was explained on the basis of changes in the characteristic pore size of the structure, which decreases as the κ-carrageenan concentration decreases.

Finally, Fig. 12 shows the micrographs for gels formed with both ι-carrageenan and the 80/20 ι-carrageenan/mucilage ratio at 12 mM $CaCl_2$. No clear interaction at structural level between the ι-carrageenan and the mucilage was observed. A modification in the gel micro-structure (Fig. 12(b)) in the presence of the mucilage gum was apparent respect to the one observed just for ι-carrageenan (Fig. 12(a)). Despite this difference in gel micro-structure when the mucilage gum was present, rheological studies did not reported enhancement in the mechanical properties of mixed gels of ι-carrageenan and mucilage gum with respect to the pure ι-carrageenan gels (Medina-Torres et al., 2003).

4. Conclusions

This work presented new experimental evidence on the interactions between mucilage gum from *O. ficus indica* and either κ-or ι-carrageenan. In the case of the κ-carrageenan/mucilage gum mixtures, the thermo-reversible hysteresis slightly increases as the κ-carrageenan concentration in the mixture decreases and it remains practically independent of the KCl concentration. For the ι-carrageenan/mucilage gum system, the hysteresis is independent of the concentration of the carrageenan and it slightly increases as the concentration of ion increases.

From an energetic point of view, mucilage gum does not contribute significantly to either the sol-to-gel or gel-to-sol transitions, except for the 80/20 κ-carrageenan/mucilage ratio. These results corroborate the previously reported synergism in the mechanical properties of gels also observed at the 80/20 κ-carrageenan/mucilage ratio (Medina-Torres, et al., 2003). Based in the calorimetric and the SEM results it is proposed that mucilage gum adsorbs onto the carrageenan aggregates, providing additional strength to the structure. Then, the mucilage does not form an integral part of the molecular κ-carrageenan structure but rather it is aggregated on the surface of this structure. On the other hand, the ι-carrageenan/mucilage mixtures did not show any clear interaction between the two biopolymers during the development of the gel structure.

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