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Effect of galactomannan addition on the thermal behaviour of κ-carrageenan gels

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Gelation/melting cycles of κ-carrageenan/galactomannan (guar, tara and locust bean gums) binary systems have been studied by measuring dynamic rheological parameters. Two experimental conditions were used, (i) the total polysaccharide concentration was kept at 1% and the κ-carrageenan/galactomannan ratio fixed at 4:1 and (ii) the κ-carrageenan concentration was fixed at 0·75% and the galactomannan content varied from 0% to 1·2%. A thermal hysteresis was observed for all mixed systems and was found to depend on the galactomannan used. From a comparison of the gelation temperature (T_g) and melting temperature (T_m) to values obtained with κ-carrageenan alone, it was suggested that galactomannan interferes with gel structure by the formation of a secondary network provided that the M/G ratio is high enough.

INTRODUCTION

Galactomannans are plant seed polysaccharides containing a backbone of $(1\rightarrow4)$-β-D-mannopyranosyl units with attached $(1\rightarrow6)$-α-D-galactose units (Dey, 1978). The proportion of galactose units attached to the mannan chain depends on the species from which the polysaccharide is extracted and varies from approximately 20% for locust bean gum (LBG) to 33% for guar gum (GG) (Hui & Neukom, 1964), and is approximately 25% for tara gum (TG) (Cairns *et al.*, 1986). GG and LBG have found widespread use in the food industry due to their ability to form viscous solutions. However, they do not form gels by themselves.

Carrageenan is a complex mixture of sulphated galactans extracted from certain red marine algae. Kappa-carrageenan (κ-car) is the less sulphated fraction and the easiest to gel. Different models have been proposed for the mechanism of gelation of κ-car (Rees, 1972; Smidsrød, 1980; Rochas & Rinaudo, 1984). This polysaccharide presents a thermally induced conformational change and gelation can occur in the presence of specific ions such as potassium. The role of these cations in the conformation of κ-car at the sol-gel transition has been extensively studied by Rochas & Rinaudo (1982), Norton *et al.* (1983), Plashchina *et al.* (1986), Day *et al.* (1988), Nilsson *et al.* (1989) and Nilsson & Piculell (1989). Recently, Hermansson *et al.* (1991) studied the effects of the cations potassium, sodium and calcium and mixtures of these cations on κ-car gelation by microscopy and viscoelastic measurements. The possibility of aggregation of the ordered helices leads to the appearance of a thermal hysteresis (Morris *et al.*, 1980; Rochas & Rinaudo, 1980). Rochas & Rinaudo (1980, 1982, 1984) found that the inverse of the melting temperature of the ordered conformation was linearly related to the logarithm of the free potassium concentration. According to this relation, these authors described a phase diagram taking into account the free potassium concentration and the temperature in order to characterize the conformation and the gelation of the κ-car. This phase diagram defined three domains: the first one was typical of a coil type macromolecular system; the second domain was characterized by an ordered conformation (helical dimer) and the third one was defined by helical dimers aggregated in a gel phase. For ionic concentrations higher than 7×10^{-3} eq/litre, both values of gelation (T_g) and melting (T_m) temperatures were different and this hysteresis was directly correlated with gel formation.

Gels of κ-car are brittle and prone to syneresis. The tendency to brittleness can be reduced by the addition of LBG. The synergistic effects between κ-car and LBG lead to gelation under conditions at which the pure

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components would not gel. Interestingly, this synergism does not occur with GG because of structural differences. The mechanism of gelation of these binary systems is still a matter of debate and several models have been proposed (Dea & Morrison, 1975; Tako & Nakamura, 1986; Cairns *et al.*, 1987). As in the case of κ-car alone these mixed systems can exhibit thermal hysteresis. Although the region of thermal hysteresis of the κ-gel increases by addition of LBG (Dea *et al.*, 1972; Fernandes *et al.*, 1991*a*), the linear relationship between the logarithm of the free potassium concentration and the inverse of T_g and T_m is still observed for a fixed κ-car/LBG ratio (Fernandes *et al.*, 1991*a*).

In our work initially, κ-car/galactomannan blends at a 4:1 ratio and 1% total polymer concentration were characterized. This ratio corresponds to the maximum of synergistic interaction previously found for these mixed systems (Fernandes *et al.*, 1991*b*). Then, the κ-car content was kept constant at 0·75% and the galactomannan (GG and LBG) was added at a concentration progressively increasing from 0% to 1·2%. The results obtained were compared to those of κ-car alone. The overall objective was a better understanding of the role of the galactomannan in the formation of κ-car/galactomannan mixed systems.

MATERIALS AND METHODS

Materials

Two Portuguese carob flours, galhosa and canela, were obtained at the laboratory scale according to Gonçalves *et al.* (1988) and three commercial grade guar, tara and carob gums were kindly provided by SBI (France), Marine Colloids (USA) and Indal (Portugal), respectively. The κ-car sample was extracted from *Eucheuma cotonii* and was kindly supplied by SBI (France), in the K^+ form. All the galactomannan samples, except guar gum, had to be purified as described by Fernandes *et al.* (1991*b*). The Indal gum was fractionated according to its solubility in water. The procedure was carried out in order to have a final solution of hot water soluble (HWS) fraction at 1% (w/w) as previously described by Fernandes *et al.* (1991*b*). This fraction will be referred to as HWS.

METHODS

Analysis of galactomannan samples

The moisture and ash contents of the gums were obtained by standard methods. The protein content was determined by the Kjeldhal method using a 5·87 factor (Anderson, 1986). The mannose to galactose ratio (M/G) was determined by the procedure of Blakeney *et al.* (1983). In the case of the HWS fraction of Indal gum, the M/G ratio was determined directly from the solution as described in Fernandes *et al.* (1991*b*).

Preparation of the solutions

The galactomannan and κ-car samples were first dispersed in water under moderate agitation for 1 h, at room temperature, and then heated at 90°C for 30 min while stirring. After cooling, the polysaccharide concentration was estimated using the phenol-sulphuric acid method of Dubois *et al.* (1956).

The binary systems were prepared by mixing, at 90°C, the solutions of galactomannan and κ-car at the desired ratio and total polymer concentration: κ-car/galactomannan blends at the 4:1 ratio and 1% total polymer concentration, and κ-car at the constant concentration of 0·75% plus GG or LBG at progressive concentrations until 1·2%. The hot mixture was heated for 15 min at the temperature of 90°C and then cooled to 55°C. In these experiments, no KCl was added, the only K^+ ions were those present in the salt form of the κ-car.

Rheological measurements

Dilute solutions
The viscosity of dilute solutions was measured at 25°C with a Low-Shear 30 viscometer (Contraves) using a concentric cylinder geometry (r_1 = 5·5 mm; r_2 = 6·0 mm; h = 8·0 mm) over the shear rate range of 0·017–128·5 s^{-1}. The intrinsic viscosity, [η], was evaluated from classical Huggins and Kraemer plots. In the case of TG and HWS Indal fraction the intrinsic viscosity was evaluated as described by Fernandes *et al.* (1991*b*).

Cooling–heating cycles
The hot mixture was poured directly onto the plate of the controlled stress rheometer Carri-Med CS-50, at 55°C, and covered with light oil in order to eliminate dehydration problems. The storage modulus, G', and the loss modulus, G'', were obtained from temperature sweep experiments under the same conditions as in Rochas & Rinaudo (1980) and Plashchina *et al.* (1986): by cooling the systems from 55°C to 5°C and then reheating to 55°C, at the rate of 18°C/h, at the constant frequency of 1·0 Hz. A parallel plate geometry was used (gap 4 mm; plate diameter 6 cm) with radial grooves in order to avoid gel slippage. The strain amplitude was fixed at 0·02.

Mechanical spectra and time sweep experiments
The temperatures at which mechanical spectra (0·1–10 Hz) and time sweep experiments (1 Hz) were performed, were attained by decreasing the temperature from 55°C at the rate of 18°C/h. These measurements were performed with the rheometer Carri-Med fitted

with a cone and plate geometry (4° cone angle, 5·0 cm diameter). The strain amplitude was fixed at 0·05.

The sol–gel transition was defined by the cross-over of the rheological moduli, G' and G'', as described in the case of physical gels by Axelos & Kolb (1990), Cuvelier *et al.* (1990) and Lin *et al.* (1991).

RESULTS

The chemical and macromolecular characteristics of the different galactomannan samples are given in Table 1. It is seen that M/G varied to a large extent as expected since galactomannans are of different origins. Also, it can be noticed that M/G varies to a relatively large extent between LBG samples. The intrinsic viscosity which is an indirect measure of the molecular weight varied from 9·4 to 16·0 dl/g. Figure 1 presents the evolution of G' and G'' as a function of temperature for κ-car alone at 1%. The system was cooled from 55°C to 5°C and then reheated. A hysteresis is clearly exhibited with T_g = 24–25°C and T_m = 32°C, taking the sol–gel transition as the cross-over of G' and G''. Typically, the cross-over of G' and G'' takes place at around 1·5–2 Pa. The early beginnings of κ-car gelation were investigated in the vicinity of the sol–gel transition by time sweep experiments at temperatures of 25, 26 and 27°C. For temperatures of 26°C and 27°C, no evidence of gel formation was found during the course of the experiments which lasted for 22 h and 27 h, respectively. But for 25°C, a different situation was observed. Figure 2 shows the kinetics of gelation of κ-car at this temperature. This figure describes the evolution of G' and G'' where G' is seen to increase steadily during the time considered (16 h), while G'' attained a plateau value of the order of 2·2 Pa within about 3 h. The results presented in Fig. 2 indicate that T_g, as obtained from Fig. 1, was determined without ambiguity. Despite the fact that we did not apply the same kind of approach to T_m, it seems reasonable to assume that T_m would show identical behaviour as that described for T_g.

Table 1. Composition and macromolecular characteristics of the galactomannans

Sample	Ash (%)	Protein (%)	M/G	[η] (dl/g)
Guar	0·10	0·22	1·68	12·0
Tara	0·20	0·45	3·10	9·4
Canela	0·33	0·40	3·46	13·8
Galhosa	0·35	0·49	4·04	15·3
Indal	0·31	0·50	4·04	15·4
HWS Indal	—	—	4·80	16·0

[η]: intrinsic viscosity.
M/G: mannose to galactose ratio.

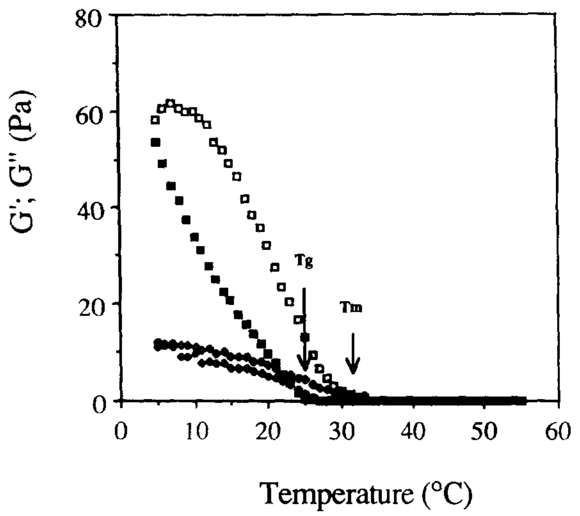

Fig. 1. G' and G'' variation as a function of temperature for κ-car at 1·0% polymer concentration. Frequency: 1 Hz. T_g: gelation temperature; T_m: melting temperature. (■) G', (♦) G'', cooling; (□) G', (◇) G'', heating.

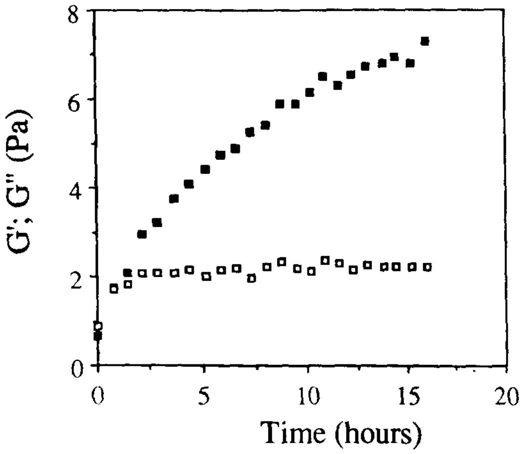

Fig. 2. Kinetics of evolution of κ-car at 1% polymer concentration at the temperature of 25°C. Frequency: 1·0 Hz. ■, G'; □, G''.

It is to be noted that the above data were obtained from measurements at a fixed frequency (1 Hz) while a better understanding of the gelation process would require the description of the entire mechanical spectrum at any time. However, during a cooling–heating cycle, it is difficult to obtain such spectra because the formation or rupture of the gel network is a progressive and continuous process. Figure 3 shows the evolution of G' and G'' as a function of temperature for κ-car/LBG (galhosa) 4:1 at 1%. This mixed system presented a T_g = 26°C and a T_m = 46°C. The thermal hysteresis was thus greatly amplified in comparison to that of κ-car alone (Fig. 1). Figure 4 shows the mechanical spectra that were exhibited by the κ-car/LBG (galhosa) blend at the temperatures of 28°C and 25°C. It is clear that, at 28°C, the viscoelastic behaviour is typical of a macromolecular solution with $G' < G''$ at

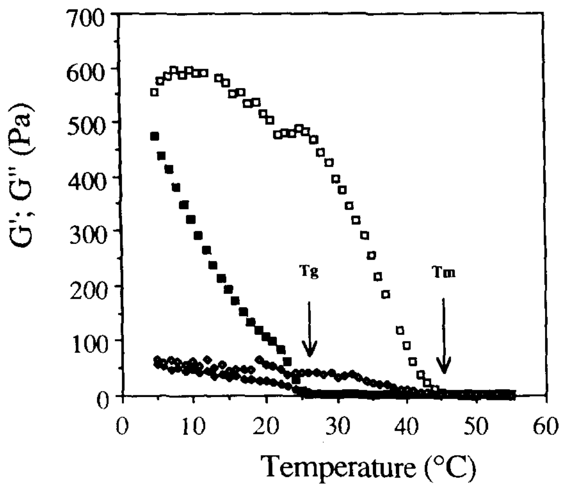

Fig. 3. G' and G'' variation as a function of temperature for 4:1 κ-car/LBG (galhosa) at 1·0% total polymer concentration. Frequency: 1 Hz. T_g: gelation temperature; T_m: melting temperature. (■) G', (♦) G'', cooling; (□) G', (◇) G'', heating.

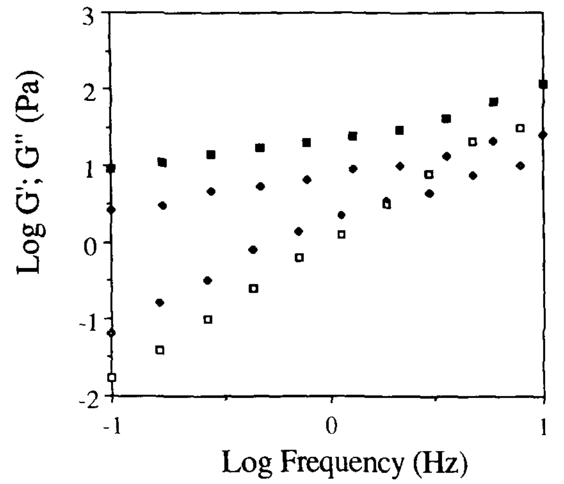

Fig. 4. Mechanical spectra of the 4:1 κ-car/LBG (galhosa) at 1% total polymer concentration at the temperatures of 25°C and 28°C. Frequency: 1 Hz. (■) G', (♦) G'', 25°C; (□) G', (◇) G'', 28°C.

low frequency and $G' > G''$ above 3 Hz. Such behaviour has been reported many times for galactomannan solutions. It is thus likely that it is LBG which plays a major role at this temperature. The behaviour displayed at 25°C is strikingly different. Higher values of G' and G'' were obtained, particularly towards the low frequency range. The fact that $G' > G''$ throughout the frequency range and that G' and G'' vary in a similar way as a function of frequency (in logarithmic coordinates) means that the system corresponds to the usual definition of a weak gel (Clark & Ross-Murphy, 1987). Moreover, the curves $G'(\omega)$ and $G''(\omega)$ are almost parallel and their slope is about 0·5. This value is in close agreement with those found by Cuvelier *et al.* (1990) for iota-carrageenan and xanthan/LBG mixtures at the gel point. The percolation theory predicts the slope of $G'(\omega)$ and $G''(\omega)$ to be 0·7 at the gel point (Axelos & Kolb, 1990). This result shows that we are in close vicinity to the gel point. The gelation temperature, as seen in Fig. 3, was taken as 26°C. It is thus clear that the increase in the values of both moduli between 28°C and 25°C is accompanied by a dramatic change in the rheological characteristics of the system. It may also be of interest to monitor the rheological changes that occur just at the transition temperature. This is illustrated in Fig. 5 where the G' and G'' evolution, at 26°C, of the same mixed system are plotted as a function of time. A continuous increase in G' is clearly seen, meaning that the gelation process is slow under these conditions. However, much higher values of G' and G'' are observed (Fig. 5) when compared to the values obtained for κ-car at 1%, for the temperature of 25°C (Fig. 2). This has to be related to κ-car gelation since T_g of the 4:1 blend is almost coincident with that of κ-car alone. It is clearly seen that the addition of LBG to κ-car induces an important amplification of the hysteresis region. This point will be discussed further. Figure 6 presents the evolution of G' and G'' as a function of temperature, during the cooling-heating cycle, for the κ-car/Guar gum (GG) 4:1 blend at 1%. An hysteresis is observed with T_g = 10°C and T_m = 25°C. However, if we took the increase of the moduli, we would find T'_g = 23°C and T'_m = 38°C. In this case, it is thus clear that the criterion to define the sol–gel transition is critical. In order to investigate more accurately the viscoelastic behaviour of the κ-car/GG mixture, in the vicinity of the gel point, dynamic measurements were also performed. Figure 7 shows the mechanical spectra exhibited by the κ-car/GG 4:1 blend at the temperatures of 7°C and 16°C upon cooling. At 16°C the mechanical spectrum remained typical of a macromolecular solution. In contrast, at 7°C, the spectrum was typical of a weak gel with

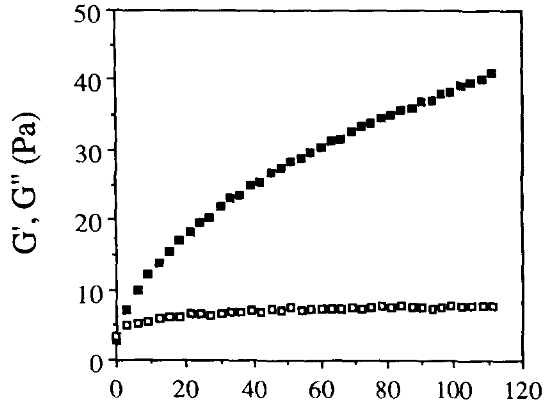

Fig. 5. Kinetics of evolution of the 4:1 κ-car/LBG (galhosa) at 1% total polymer concentration at the temperature of 26°C. Frequency: 1·0 Hz. ■, G'; □, G''.

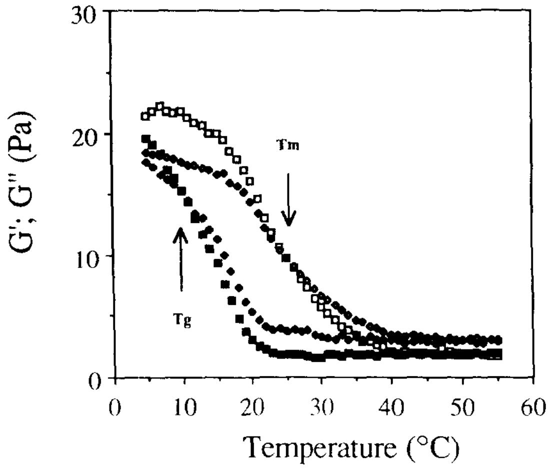

Fig. 6. G' and G'' variation as a function of temperature for 4:1 κ-car/GG at 1·0% total polymer concentration. Frequency: 1 Hz. T_g: gelation temperature; T_m: melting temperature. (■) G' (♦) G'', cooling; (□) G', (◇) G'', heating.

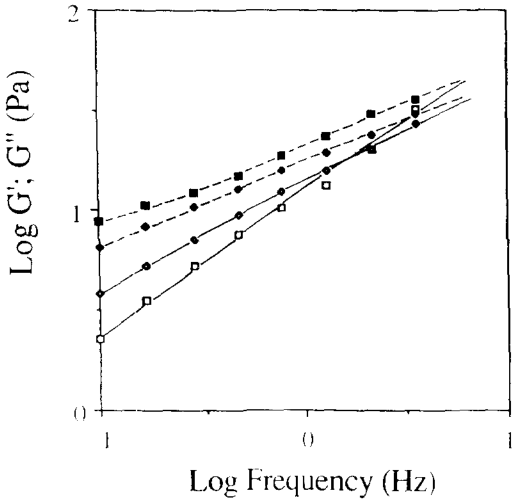

Fig. 7. Mechanical spectra of the 4:1 κ-car/GG at 1% total polymer concentration at the temperatures of 7°C and 16°C. (■) G', (♦) G'', 7°C; (□) G', (◇) G'', 16°C.

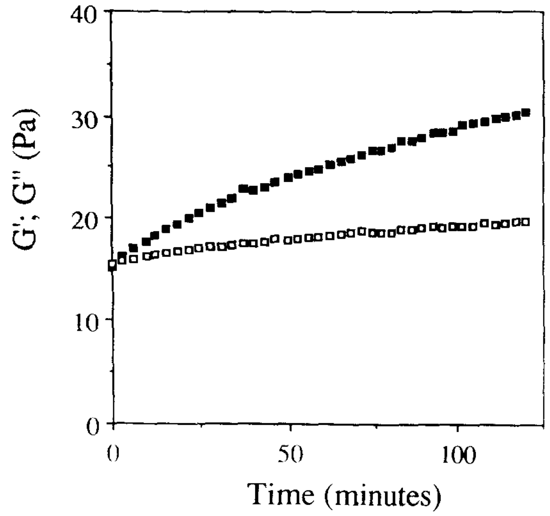

Fig. 8. Kinetics of evolution of the 4:1 κ-car/GG at the 1% total polymer concentration at the temperature of 10°C. Frequency: 1·0 Hz. ■, G'; □, G''.

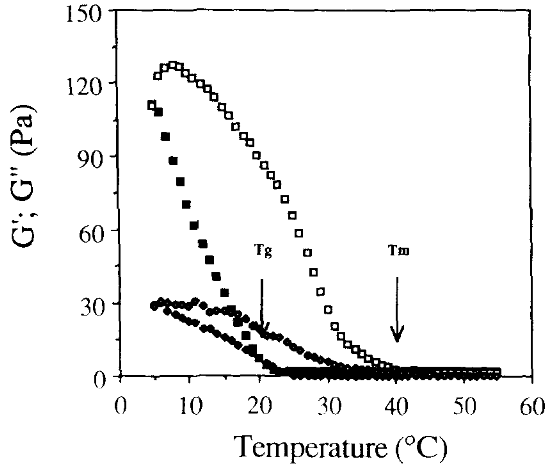

Fig. 9. G' and G'' variation as a function of temperature for the 4:1 κ-car/TG at 1·0% total polymer concentration. Frequency: 1 Hz. T_g: gelation temperature; T_m: melting temperature. (■) G', (♦) G'', cooling; (□) G' (◇) G'', heating.

$G' > G''$, indicating that the system is already beyond the gel point. Figure 8 displays the G' and G'' evolution, at 10°C, as a function of time. The evolution observed in this figure seems to indicate that this temperature is close to the gel point. This is in good agreement with the interception point of G' and G'' as described in Fig. 6. In Fig. 8, the evolution of G' and G'' is much slower than that observed for the κ-car/LBG mixture (Fig. 4). Figure 9 shows the variation of G' and G'' as a function of temperature for the κ-car/Tara gum (TG) 4:1 blend, at 1%. The κ-car/TG mixture presents a T_g = 20°C and a T_m = 40°C as determined by the cross-over of G' and G''. As for GG, there was no coincidence between T_g and the temperature at which G' increased. However, this difference is much less pronounced. The T_m value of κ-car/TG was considerably higher than the T_m value of κ-car alone at 1% (32°C). Thus, the κ-car/TG mixture exhibited thermal behaviour intermediate between those of similar mixtures with GG and LBG, although much closer to that with LBG.

Table 2 presents the data of the cooling–heating cycles for all the systems investigated above. The data obtained with HWS fraction are also shown. The parameters related to the sol–gel transition of the binary systems of κ-car/galactomannan 4:1 at 1% are compared with those of the κ-car alone at concentrations of 0·8% and 1%. It must be remembered that the κ-car content in the 4:1 blend is 0·8%. If we compare the results of κ-car alone at 0·8% with the κ-car/galactomannan blends, an increase in T_g, T_m and ΔT is clearly seen. If we now compare this with κ-car at 1% we

Table 2. Parameters related to the sol–gel transition on binary systems of κ-carrageenan/galactomannan

Galactomannan	M/G	T_g (°C)	T_m (°C)	ΔT ($T_m - T_g$)	G' at 5°C (Pa)
Guar	1·68	10	25	15	20
Tara	3·10	20	40	20	111
Canela	3·46	25	45	20	250
Galhosa	4·04	26	46	20	475
Indal	4·04	26	46	20	480
HWS Indal	4·80	26	46	20	710
κ-car 0·8%	—	5	16	11	6
κ-car 1·0%	—	24	32	8	54

observe some interesting features. The gelation temperatures of the mixtures with LBG and HWS are very close to the T_g of κ-car alone. However, T_g values of κ-car/GG and κ-car/TG blends are lower. With the exception of GG, all the other galactomannan mixtures (TG, LBG and HWS) show significantly higher values of T_m than κ-car at 1%. The thermal hysteresis ($\Delta T = T_m - T_g$) was higher for all the κ-car/galactomannan mixtures than for the κ-car alone. It is noteworthy that ΔT was constant with the exception of GG. On the other hand, values of G' and G'' at 5°C evidenced strong synergistic effects, with the exception of GG. In this latter case, the value lies between those of 0·8% and 1% κ-car alone. It is also clear that the higher the M/G ratio, the higher the storage modulus. However, we previously showed that the molecular weight of galactomannan plays an additional role (Fernandes *et al.*, 1991*b*). The large differences found between LBG samples, from 250 Pa for Canela to 480 Pa for Indal, is further explained by a difference in molecular weight (Table 1).

Figure 10 shows the T_g and T_m of the κ-car at a constant concentration of 0·75%, the galactomannan (GG or LBG) being added at concentrations progressively increasing from 0% to 1·2%. The κ-car alone at 0·75% presented a $T_g = 5$°C and a $T_m = 15$°C (see arrows). With the κ-car/LBG mixture, the T_g and T_m values increased rapidly and steady values of 24–25°C and 44°C were attained, respectively. This plateau was reached at a 0·15% galactomannan content (κ-car:LBG = 0·75:0·15). In the case of κ-car/GG blend, T_g and T_m increased more slowly and steady values of 20°C and 30°C were attained at a GG content of about 0·50 (κ-car:GG = 0·75:0·50). It can be noted that the 4:1 ratio of the κ-car/galactomannan mixtures investigated above corresponds to a 0·75:0·19 ratio as shown by the vertical dashed line. In that case, we have a T_m = 44°C and T_g = 22–23°C for LBG, and T_m = 14°C and T_g = 9°C for GG. These values are slightly lower than those in Table 2 since the total polymer concentration was lower (0·94% against 1%). The variations in T_g and T_m were much less important for the κ-car/GG than for the κ-car/LBG mixture.

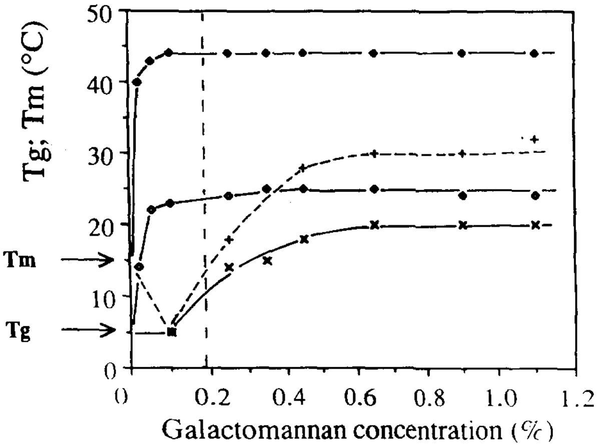

Fig. 10. Effect of the addition of different amounts of guar and carob gums to a 0·75% κ-car gel. (◇) T_g, (◆) T_m, LBG mixture; (×) T_g, (+) T_m, GG mixture. The gelation and melting temperatures of the κ-car alone at 0·75%, are arrowed in the ordinate axis.

DISCUSSION

The data obtained for κ-car alone can be discussed by reference to the phase diagram of Rochas & Rinaudo (1980, 1984). It should be remembered that in the present work no KCl was added, the only K^+ ions present being those introduced by the salt form of the κ-car. On this basis, the free potassium concentration, C_T, can be estimated from (Rochas & Rinaudo, 1980):

$$C_T = \bar{\gamma} C_P$$

with C_P the polymer potassium concentration and $\bar{\gamma}$ the mean activity coefficient. In the present case, C_T was found to be about $10{\cdot}4 \times 10^{-3}$ eq/litre for 1% κ-car and $8{\cdot}3 \times 10^{-3}$ eq/litre for 0·8%. These values are just above the critical concentration, C_T^*, which was found to be 7×10^{-3} eq/litre (Rochas & Rinaudo, 1980, 1984). This explains why a limited thermal hysteresis is seen ($\Delta T \approx 10$°C). The fact that ΔT for 0·8% is of the same order as for 1% may be explained by the fact that we are in the vicinity of C_T^*.

Dynamic rheological measurements have been used to investigate the gelation of κ-car (Rochas & Rinaudo, 1984; Plaschina *et al.*, 1986). Rochas & Rinaudo (1984) described a good correlation between the transition temperature obtained by polarimetry and the evolution of G' as a function of temperature. On the other hand, Plaschina *et al.* (1986) showed a coincidence between T_g (obtained by a rheological method) and the transition temperature determined by polarimetry. On a similar basis, by using viscosimetric and viscoelastic experiments we established a phase diagram (Fernandes *et al.*, 1991*a*) which is very close to that described by

Rochas & Rinaudo (1980, 1984). Upon cooling, the κ-car macromolecules undergo a coil–helix transition as seen by polarimetry. This transition is instantaneous and results in gel formation. The gelation temperature probably coincides with this coil–helix transition. This process is followed by an aggregation of helical dimers of κ-car molecules. This may result in a precipitation and a phase separation (Day *et al*., 1988). This second process is time dependent and quite slow in the vicinity of the transition temperature (Rochas & Landry, 1987). The thermal hysteresis in κ-car systems is classically ascribed to aggregation phenomena. If the aggregation is quantitatively important, the melting of the gel is retarded because the aggregates (established between the carrageenan molecules) must be broken down before the helix–coil transition (T_m) takes place (Morris *et al*., 1980; Rochas & Rinaudo, 1984). In order to interpret the present data, it would be useful to know the mechanisms that take place in the gelation of κ-car/galactomannan systems. A former description by Dea & Morrison (1975) assumed that intermolecular binding occurs between the κ-car double helices and 'smooth' regions of galactomannan chains. This model has been classically accepted and many authors have interpreted their own data on this basis. However, more recently, based on X-ray diffraction of films, Cairns *et al*. (1987, 1991) did not find any experimental evidence for the intermolecular binding between polysaccharides. They suggested another mechanism where a basic κ-car network would contain galactomannan molecules in solution. The participation of the galactomannan in gel formation was thus ruled out. However, our results show that, whatever the galactomannan, there is a stabilization of the κ-car helix and this effect is similar to that described by Tolstoguzov & Braudo (1983) for the stabilization of the gelatin helix by dextran. For higher values of M/G, as in the case of LBG and HWS, the participation of the galactomannan in a network may be considered. This assumption is supported by the high values of G', which seem too high to be due to a simple concentration effect. Moreover, Rochas *et al*. (1991) showed, on the basis of NMR data on LBG, that mannan–mannan interactions could take place in mixed systems. This may imply that two networks are formed which can be interpenetrated. The model proposed by Cairns *et al*. (1987) thus provides a good basis for the interpretation of our results, whatever the type of galactomannan.

The addition of GG to a κ-car is expected not to modify its thermal hysteresis. The first result would be a dilution effect since there is a lowering of the κ-car concentration down to 0·8% in the 4:1 mixed systems at 1% total polymer concentration. In fact, as seen in Table 2, T_g, T_m and G' of the mixture correspond to intermediate behaviour between κ-car concentrations of 0·8% and 1%. This means that GG plays a role in the gelation of the system. This role is more spectacular when looking at Fig. 10. If both polymers did not affect each other, T_g and T_m should not vary upon adding the galactomannan. We suggest that this stabilizing effect of GG is originated by an increase in the actual concentration of κ-car due to volume exclusion, this resulting in a higher actual concentration in a κ-car-enriched phase. This interpretation is consistent with the model proposed by Cairns *et al*. (1987). The same approach can be applied to κ-car/LBG and κ-car/TG mixtures. For TG, the value of G' in Table 2 evidences clearly a synergistic effect in the mixture. The value of T_g (20°C) is slightly lower than that of κ-car at 1% (24°C), suggesting that the actual κ-car concentration is just below 1%. On the other hand, T_m (40°C) is higher than that with κ-car alone at 1% (32°C) meaning that the helical dimers of κ-car are more aggregated. Moreover, TG chains are known to bear 'smooth' galactose-free regions (McCleary & Neukom, 1982) that make their aggregation possible. So, since the effect of TG is also significant in rheological terms, the formation of a secondary galactomannan network can be expected. These trends are still more accentuated with LBG samples and HWS. T_g levels have a value very close to that of κ-car at 1% and may indicate that the actual κ-car concentration is 1%. Also, T_m (46°C) is much higher than that with κ-car alone at 1%. The aggregation of helical dimers thus seems more important. Here also, the galactomannan chains would be involved in a secondary network obtained through mannan–mannan interactions. Here also, the galactomannan chains would be involved in a secondary network obtained through mannan–mannan interactions. The observed values at T_g and T_m should correspond to the gelation and melting temperatures of the κ-car network. If we assume that the galactomannan network is diffuse, its T_g and T_m values should lie somewhere near those for κ-car and could not be detected by our rheological measurements. This kind of effect was recently reported by Williams *et al*. (1992) in mixtures of κ-car and konjac mannan where rheological and calorimetric results were compared. It appears that all the present results are consistent with the model of Cairns *et al*. (1987) although a more important role must be assigned to the galactomannan macromolecules when the M/G ratio is high.

CONCLUSION

It is clearly demonstrated that the thermal behaviour of κ-car/galactomannan mixed systems is influenced by the galactomannan whatever its type. This means that galactomannans interfere in the mechanism of gelation. The hypothesis of volume exclusion provides a convenient means of interpreting most of the present data. The main difference between the galactomannans is related to the M/G ratio; this determines their trend to form aggregates and, eventually, leads to new gel properties by the formation of a secondary network which interpenetrates the primary one formed by the κ-car. However, the present data were obtained from rheological measurements which give information only at a macroscopic level. Indeed, this interpretation has to be confirmed by using other methods to describe the phenomena at the molecular level.

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REFERENCES

Anderson, D.M.W. (1986). Nitrogen conversion factors for the proteinaceous content of gums additives. *Food Addit. Contam.*, 3(3), 231-4.

Axelos, M.A.V. & Kolb, M. (1990). Scaling behavior of the viscoelastic properties of a biopolymer system near the gel point. In *Mat. Res. Soc. Symp. Proc.*, **177**, 199-204.

Blakeney, A.B., Harris, P.J., Henry, R.J. & Stone, B.A. (1983). A simple and rapid preparation of alditol acetates for monosaccharide analysis. *Carbohydr. Res.*, **113**, 291-9.

Cairns, P., Morris, V.J., Miles, M.J. & Brownsey, G.J. (1986). Comparative studies of the mechanical properties of mixed gels formed by kappa-carrageenan and tara gum or carob gum. *Food Hydrocoll.*, **1**(1), 89-93.

Cairns, P., Miles, M.J., Morris, V.J. & Brownsey, G.J. (1987). X-ray fibre diffraction studies of synergistic, binary polysaccharide gels. *Carbohydr. Res.*, **160**, 411-23.

Cairns, P., Atkins, E.D.T., Miles, M.J. & Morris, V.J. (1991). Molecular transforms of kappa-carrageenan and furcellan from mixed gel systems. *Int. J. Biol. Macromol.*, **12**, 353-8.

Clark, A.H. & Ross-Murphy, S.B. (1987). Structural and mechanical properties of biopolymer gels. *Adv. Polym. Sci.*, **83**, 55-192.

Cuvelier, G., Peigney-Nourry, C. & Launay, B. (1990). Viscoelastic properties of physical gels: critical behavior at the gel point. In: *Gums and Stabilizers for the Food Industry – 5*, eds G.O. Phillips, D.J. Wedlock & P.A. Williams. IRL Press, Oxford, pp. 549-52.

Day, D.H., Phillips, G.O. & Williams, P.A. (1988). Gelation of kappa-carrageenan as studied by electron spin resonance spectroscopy. *Food Hydrocoll.*, **2**(1), 19-30.

Dea, I.C.M. & Morrison, A. (1975). Chemistry and interactions of seed galactomannans. *Adv. Carbohydr. Chem. Biochem.*, **31**, 241-312.

Dea, I.C.M., McKinnon, A.A. & Rees, D.A. (1972). Tertiary and quartenary structure in aqueous polysaccharide systems which model cell cohesion: reversible changes in conformation and association of agarose, carrageenan and galactomannans. *J. Mol. Biol.*, **68**, 153-72.

Dey, P.M. (1978). Biochemistry of plant galactomannans. *Adv. Carbohydr. Chem. Biochem.*, **35**, 341-76.

Dubois, M., Gilles, K.A., Hamilton, J.K., Rebers, P.A. & Smith, F. (1956). Colorimetric method for determination of sugars and related substances. *Anal. Chem.*, **28**, 350-6.

Fernandes, P.B., Gonçalves, M.P. & Doublier, J.L. (1991*a*). Phase diagrams in kappa-carrageenan/locust bean gum systems. *Food Hydrocoll.*, **5**(1/2), 71-3.

Fernandes, P.B., Gonçalves, M.P. & Doublier, J.L. (1991*b*). A rheological characterization of kappa-carrageenan/galactomannan mixed gels: a comparison of locust bean gum samples. *Carbohydr. Polym.*, **16**, 253-74.

Gonçalves, M.P., Fernandes, P.B. & Lefebvre, J. (1988). A laboratory-scale preparation and testing of locust bean gum from traditional varieties of Portuguese carob. In: *Proceedings of II International Carob Symposium*, eds. P. Fito & A. Mulet. Generalitat Valenciana, Conselleria d'Agricultura i Pesca, Valencia, pp. 407-17.

Hermansson, A.-M., Eriksson, E. & Jordansson, E. (1991). Effects of potassium, sodium and calcium on the microstructure and rheological behavior of kappa-carrageenan gels. *Carbohydr. Polym.*, **16**, 297-320.

Hui, P.A. & Neukom, H. (1964). Some properties of galactomannans. *TAPPI*, **47**, 39-42.

Lin, Y.G., Mallin, D.T., Chien, J.C.W. & Winter, H.H. (1991). Dynamic mechanical measurement of crystallization-induced gelation in thermoplastic elastomeric poly(propylene). *Macromolecules*, **24**, 850-4.

McCleary, B.V. & Neukom, H. (1982). Effect of enzymic modification on the solution and interaction properties of galactomannans. *Prog. Food Nutr. Sci.*, **6**, 109-18.

Morris, E.R., Rees, D.A. & Robinson, G. (1980). Cation-specific aggregation helices: domain model of polymer gel structure. *J. Mol. Biol.*, **138**, 349-62.

Nilsson, S. & Piculell, L. (1989). Helix-coil transitions of ionic polysaccharides analysed within the Poisson-Boltzmann cell model. 2. Effects of salt concentration on the thermal transition. *Macromolecules*, **22**(7), 3011-17.

Nilsson, S., Piculell, L. & Jonsson, B. (1989). The helix-coil transition of an anionic polysaccharide probed by counterion self-diffusion measurements. *Macromolecules*, **22**(5), 2367-75.

Norton, I.T., Goodall, D.M., Morris, E.R. & Rees, D.A. (1983). Equilibrium and dynamics studies of the disorder-order transition of kappa-carrageenan. *J. Chem. Soc., Faraday Trans. 1*, **79**, 2489-500.

Plashchina, I.G., Muratalieva, I.R., Braudo, E.E. & Tolstoguzov, V.B. (1986). Studies of the gel formation of κ-carrageenan above the coil-helix transition temperature. *Carbohydr. Polym.*, **6**, 15-34.

Rees, D.A. (1972). Shapely polysaccharides. *Biochem. J.*, **126**, 175-73.

Rochas, C. & Landry, S. (1987). Molecular organization of kappa-carrageenan in aqueous solution. *Carbohydr. Polym.*, **7**, 435-47.

Rochas, C. & Rinaudo, M. (1980). Activity coefficients of counterions and conformation in kappa-carrageenan systems. *Biopolymers*, **19**, 1675-87.

Rochas, C. & Rinaudo, M. (1982). Calorimetric determination of the conformational transition of kappa-carrageenan. *Carbohydr. Res.*, **105**, 227-36.

Rochas, C. & Rinaudo, M. (1984). Mechanism of gel formation in κ-carrageenan. *Biopolymers*, **23**, 735-745.

Rochas, C., Taravel, F.-R. & Turquois, T. (1991). N.m.r. studies of synergstic kappa-carrageenan/carob galactomannan gels. *Int. J. Biol. Macromol.*, **12**, 353-8.

Smidsrød, O. (1980). In *IUPAC, 27th International Congress of Pure and Applied Chemistry*, ed. A. Varmavuori. Pergamon Press, New York, pp. 315-27.

Tako, M. & Nakamura, S. (1986). Synergistic interaction between kappa-carrageenan and locust bean gum in aqueous media. *Agric. Biol. Chem.*, **50**(11), 2817-22.

Tolstoguzov, V.B. & Braudo, E.E. (1983). Fabricated foodstuffs as multicomponent gels. *J. Text. Stud.*, **14**, 183–212.

Williams, P.A., Clegg, S.E., Langdon, M.J., Nishinari, K. & Phillips, G.O. (1992). Studies on the synergistic interaction between konjac mannan and locust bean gum with kappa-carrageenan. In *Gums and Stabilizers for the Food Industry – 6*, eds G.O. Phillips, D.J. Wedlock & P.A. Williams. IRL Press, Oxford, 209–16.