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A Rheological Characterization of Kappa-Carrageenan/ Galactomannan Mixed Gels: A Comparison of Locust Bean Gum Samples

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ABSTRACT

Mixed gels of kappa-carrageenan and locust bean gum (LBG) obtained from different varieties of Portuguese carob trees and commercial gums were compared. The viscoelastic properties of the gels were measured using dynamic parallel-plate geometry. Mixed gels at 1·0% of total polysaccharide concentration without addition of KCl showed, whatever the LBG sample, a synergistic maximum when the ratio of kappa-carrageenan to LBG was 80/20. The amplitude of this maximum varied with the LBG sample. The gels prepared at 0·3% total concentration with KCl added, showed a synergistic maximum at the same mixing ratio and the amplitude varied in a similar manner. Each sample was fractionated into the fraction soluble at 25°C and the fraction soluble at 90°C. Mixed

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gels of kappa-carrageenan with cold-water-soluble and hot-water-soluble fractions, and also with tara gum and guar gum were prepared at the 80/20 ratio. It was found that the synergistic maxima were related to the intrinsic viscosity and the M/G ratio. A linear relationship between the storage modulus G'_{max} at the synergistic maximum and the product of the intrinsic viscosity and the square of the mannose to galactose ratio was found, suggesting that the synergistic mechanism can be ascribed to both the unsubstituted (galactose-free) regions of the galactomannan and the molecular weight.

INTRODUCTION

Locust bean, tara and guar gums are plant galactomannans composed of a linear mannan chain bearing side chains of a single galactose unit (Dey, 1978). Guar gum and locust bean gum (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. Considerable work has been carried out on the study of the rheology of aqueous galactomannan solutions (Sabater de Sabates, 1979; Doublier & Launay, 1981; Morris *et al.*, 1981; Robinson *et al.*, 1982). These galactomannans differ essentially in their ratio of mannose to galactose (M/G) which results in different solubilities: the higher M/G, the lower the solubility.

Kappa-carrageenan is one fraction of mixed sulfated polysaccharides extracted from certain red marine algae. Three major fractions are used commercially — kappa, iota and lambda — and they differ essentially in their degree of sulfation. The kappa-carrageenan, being the less sulfated fraction, is the easiest to gel. The mechanism of gelation of kappa-carrageenan has been extensively investigated and different models have been proposed (Anderson *et al.*, 1969; Rees, 1972; Rochas, 1982). Of the three galactomannans, LBG has the lowest galactose content and is the least soluble. Furthermore, LBG can interact with other polysaccharides particularly carrageenan and these blends can gel at total polymer concentrations lower than the concentration at which carrageenan alone will gel (Dea & Morrison, 1975). This is one reason for the widespread application that kappa-carrageenan/LBG mixtures have found in the food industry. The mechanisms underlying the gelling ability of these mixtures is known to be associated with the low G/M ratio and is generally ascribed to poorly substituted mannan backbone regions ('smooth' zones). However, the detailed mechanism is still a matter of debate. Different models have been proposed which assume specific interactions between the smooth zones of the galactomannan molecule and the carrageenan helices (Dea & Morrison, 1975;

Ainsworth & Blanshard, 1978; Tako & Nakamura, 1986). However, attempts to find evidence for intermolecular binding were unsuccessful (Cairns *et al.*, 1987) and an alternative model has been proposed involving a carrageenan network containing the galactomannan within the gel. Most of the rheological work on these systems involved strong gels at a high polysaccharide content ($>0{\cdot}5\%$) and with the addition of alkali ions, especially K^+ (Ainsworth & Blanshard, 1978, 1980; Christensen & Trudsoe, 1980; Miles *et al.*, 1984; Fiszman *et al.*, 1987). Since slippage effects can be encountered with carrageenan gels (Arnaud *et al.*, 1989), compressional testing has been widely preferred to characterize such mixed systems. Slippage, however, can be avoided using special devices with rough surfaces and some work using oscillatory shear has been reported. On the other hand, it is known that softer gels can be obtained by limiting the addition of KCl and the carrageenan concentration. In this way, Tako and Nakamura (1986) investigated kappa-carrageenan/LBG mixed gels using oscillatory measurements and, under their experimental conditions, no slippage was seen.

The aim of the present work was to estimate the variability of LBG samples with respect to the formation of kappa-carrageenan/LBG mixed gels. As far as we are aware, this question had not been investigated yet. For this purpose, we compared LBG samples of commercial origin and gums from Portuguese carob trees. We also investigated mixtures with tara and guar gums. In order to quantify the 'reactivity' of each sample with respect to kappa-carrageenan, we used a relatively simple methodology with no KCl addition at a low temperature (15°C). These conditions correspond to the gel region of kappa-carrageenan alone in the phase diagram described by Rochas and Rinaudo (1980). These are quite similar to those used by Tako and Nakamura (1986) except that in their case the gels were prepared and measured at 0°C which is the freezing temperature of water and may yield practical problems during rheological measurements.

MATERIALS AND METHODS

Materials

Three carob flours were obtained from carob pods that were collected from three selected carob trees, designated according to their variety as 'Mulata', 'Canela' and 'Galhosa', growing in the southern region of Portugal. Two commercial-grade carob flours, 'Indal' and 'Lygomme 6', were provided by Indal (Portugal) and SBI (France), respectively. Two

commercial-grade tara and guar gums were obtained from Marine Colloids (USA) and SBI (France), respectively. The carrageenan sample was extracted from *Eucheuma cottonii* and was kindly provided by SBI (France). It was in the K^+ form. The seed flours of the Portuguese carob varieties were obtained at the laboratory scale using the procedure described by Gonçalves *et al.* (1988). All galactomannan samples, except guar gum and Lygomme 6, had to be purified. For this purpose, galactomannan solutions were prepared and centrifuged at 31 000 ***g*** for 1 h at 25°C. Then they were added to two volumes of isopropanol and the resulting mixture was stirred for 30 min at room temperature. The extract was filtered through filter Duran no. 4 and the operation with isopropanol as described above was repeated twice. Finally, the galactomannan precipitate was dried in the presence of acetone and diethyl ether, successively. After 24 h the dried flour was milled. The kappa-carrageenan sample was used without further purification.

METHODS

Composition of galactomannan flours

The moisture content was determined by drying a sample in an oven, at 103°C, for 12 h. The ash content was obtained from heating dried samples in a muffle furnace at 500°C for 16 h, then at 900°C for 1 h. The protein was determined by the Kjeldahl method using a nitrogen factor of 5·87 (Anderson, 1986).

Fractionation of LBG samples

A known weight of purified polysaccharide was gradually added to strongly stirred distilled water. The dispersion was stirred at 25°C for 1 h, and then was centrifuged at 31 000 ***g*** for 1 h at 25°C. The supernatant was recovered as a solution of the cold-water-soluble fraction (CWS). The pellet obtained from centrifugation was resuspended in distilled water, stirred for 1 h, at room temperature, and then heated to 90°C for 30 min while stirring. All the remaining galactomannnan was recovered. This yielded a solution of the hot-water-soluble fraction (HWS). The procedure was carried out in order to have final solutions of CWS and HWS fractions at 1·0% (w/w). These fractionated samples were not submitted to a drying process. This was chosen in order to avoid subsequent solubilization problems in the preparation of solutions which could be originated by the drying procedure.

Mannose to galactose ratio

Mannose/galactose ratios were determined according to the procedure of Blakeney *et al.* (1983). Polysaccharides were normally treated, at room temperature, with 72% sulfuric acid for 30 min under stirring, and then hydrolysed at 100°C for 2 h with diluted aqueous sulfuric acid. The resulting aldoses were reduced to alditols with sodium borohydride dissolved in dimethylsulfoxide (DMSO). Acetylation was effected with acetic anhydride. Dried alditol acetates were dissolved in dichloromethane and injected (1 μl) onto a GLC capillary column DB-225. The column was operated isothermally (220°C), using a hydrogen carrier gas flow rate of 0·8 ml/min. The injector and detector oven temperatures were maintained at 220°C and 250°C, respectively. Mannose/galactose ratio of the CWS and HWS aqueous fractions were determined directly from the solutions without pretreatment with 72% sulfuric acid. The solutions were hydrolysed at 100°C for 2 h with diluted aqueous sulfuric acid, then the procedure was identical to that described above.

Preparation of the solutions

The galactomannan samples and kappa-carrageenan were first dispersed in water with vigorous stirring. The dispersion was stirred for 1 h at room temperature then heated at 90°C for 30 min while stirring. In the case of kappa-carrageenan solutions in 0·3% KCl, potassium chloride was added to the hot kappa-carrageenan solution, and the solution was stirred at 90°C until dissolution of potassium chloride was completed. Polysaccharide concentration was estimated using the phenol–sulfuric method according to the method of Dubois *et al.* (1956).

Viscosity measurements

Diluted solutions and intrinsic viscosity

The viscosity of dilute solutions of most samples was measured at 25°C with a Low-Shear 30 viscometer (Contraves) using a concentric cylinder geometry ($r_1 = 5{\cdot}5$ mm; $r_2 = 6{\cdot}0$ mm; $h = 8{\cdot}0$ mm) over the shear rate range 0·017–128·5 s^{-1}. The intrinsic viscosity $[\eta]$ was evaluated from classical Huggins' and Kraemer's plots. In the case of tara gum, CWS and HWS carob fractions, the intrinsic viscosity was only roughly evaluated. This was achieved according to the following procedure. Measurements were performed on 1% solutions in order to evaluate the zero shear rate specific viscosity η_{spo} from the flow curve. Using the master curve of zero shear rate specific viscosity η_{spo} versus reduced concentration $c[\eta]$

at 25°C that was established for LBG samples (Fernandes *et al.*, 1989), the intrinsic viscosity $[\eta]$ could be obtained.

Concentrated solutions

Viscosity measurements at 1·0% total polymer concentration were performed at 25·0 ± 0·1°C with a controlled stress rheometer Carri-Med fitted with cone and plate geometry (4° cone angle, 5·0 cm diameter).

Determination of gel characteristics

Preparation of gels

Gels were prepared at 90°C from a mixture of the galactomannan solution with the kappa-carrageenan solution in the desired proportion so as to give an overall concentration of 1·0% (w/w), without KCl, and an overall concentration of 0·3% (w/w), with KCl. The mixture was heated for 15 min at 90°C with stirring and then poured into glass moulds (6·8 cm diameter) in order to give a final thickness of about 4 mm: 20 g of the hot mixture (without KCl) and 23 g of the hot mixture (with KCl) was used. Finally, the glass moulds were covered with parafilm to avoid dehydration problems. Then the mixed gels were allowed to set for approximately 14 h, at 15°C, before use. In the case of the study of the kinetics of gelation, the hot mixture was poured directly onto the rheometer plate. In order to ensure gel stability and eliminate dehydration problems during time sweep experiments, the sample was covered with light oil (paraffin).

Dynamic measurements

Gel cure experiments were performed by the measurement of G' (storage modulus) and G'' (loss modulus) as a function of time at a frequency of 1·0 Hz, in the Carri-Med rheometer at temperatures of 10, 15, 20 and 25°C. The mechanical spectra, $G'(\omega)$ and $G''(\omega)$ were obtained at 15·0 ± 0·1°C over the frequency range 0·01–10 Hz. In both cases 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.

For mixed gels with KCl, which displayed some gel syneresis, exuded water was blotted out from the gel between two Whatman papers no. 1 (12·5 cm diameter) for 1 min just before the gel was placed onto the plate of the rheometer for dynamic measurements. By weighing the gels after the operation, loss of water could be estimated.

RESULTS

Galactomannan characteristics

Table 1 shows the main characteristics of the galactomannan samples. Molecular weights for LBG were estimated from the Mark–Houwink parameters obtained for guar gum (Robinson *et al.*, 1982), as modified in order to take into account the M/G ratio (Doublier, 1975). This procedure assumes that the intrinsic viscosity is determined by the degree of polymerization ($\overline{DP}_v$) of the galactomannan chain. Thus, from the equation of Robinson *et al.* (1982) for guar gum:

$$[\eta] = 3{\cdot}8 \times 10^{-4} \overline{M}_v^{0{\cdot}723} = k' \overline{M}_v^{0{\cdot}723} \tag{1}$$

we obtain

$$[\eta] = 2{\cdot}14 \times 10^{-2} \overline{DP}_v^{0{\cdot}723} = k'' \overline{DP}_v^{a} \tag{2}$$

from

$$k'' = \frac{k' M_o^a}{(1-x)^a}$$

where

$$M_o = 162 \text{ and } x = \frac{G}{M+G}$$

TABLE 1
Composition and Macromolecular Characteristics of the Galactomannans

	Galhosa	*Mulata*	*Canela*	*Indal*	*Lygomme 6*	*Tara*	*Guar*
Ash (%)	0·35	0·40	0·33	0·31	0·10	0·20	0·10
Protein (%)	0·49	0·47	0·40	0·50	0·10	0·45	0·22
M/G[a]	4·04	3·80	3·46	4·04	3·60	3·10	1·68
$[\eta]$[b] (dl/g)	15·30	14·30	13·80	15·40	10·50	9·40	12·00
$\overline{M}_v$[c] ($\times 10^{-6}$)	1·82	1·66	1·58	1·83	1·08	0·97	1·67
$\overline{DP}_v$ ($\times 10^{-3}$)	9·52	8·71	8·34	9·69	5·69	4·86	6·86

[a] M/G: mannose to galactose ratio.
[b] $[\eta]$: intrinsic viscosity.
[c] $\overline{M}_v$: molecular weight obtained from eqn (3).
[d] $\overline{DP}_v$: degree of polymerization obtained from eqn (2).

In eqn (2), k'' is obtained taking $x = 1/2{\cdot}6$ (cf. Table 1). Hence, taking $x = 1/4{\cdot}75$ for LBG, 3·75 being an average value of M/G (Table 1), we obtain from eqn (2):

$$[\eta] = 4{\cdot}56 \times 10^{-4} \overline{M}_v^{0{\cdot}723} \quad (3)$$

The protein and ash contents of all galactomannan samples did not differ to a great extent, and indicate good quality gums (Seaman, 1980) showing that we used an efficient purification procedure. Lygomme 6, tara and guar gums had lower intrinsic viscosity values (~ 10–12 dl/g) than the others (14–15 dl/g). M/G ratios of LBG samples varied between 3·5 and 4 which is in good agreement with the literature: 3·65 (Gaisford *et al.*, 1986); and 3·5–3·8 (MacCleary *et al.*, 1985). The ratio for tara is close to a value reported by Cairns *et al.* (1986) (M/G = 3) and our value for guar gum is also close to that found in the literature (M/G = 1·56) (Robinson *et al.*, 1982).

Gel cure experiments

Figure 1 shows typical kinetics of gelation at 15°C for a kappa-carrageenan gel and a kappa-carrageenan/LBG 80/20 mixed gel, both at

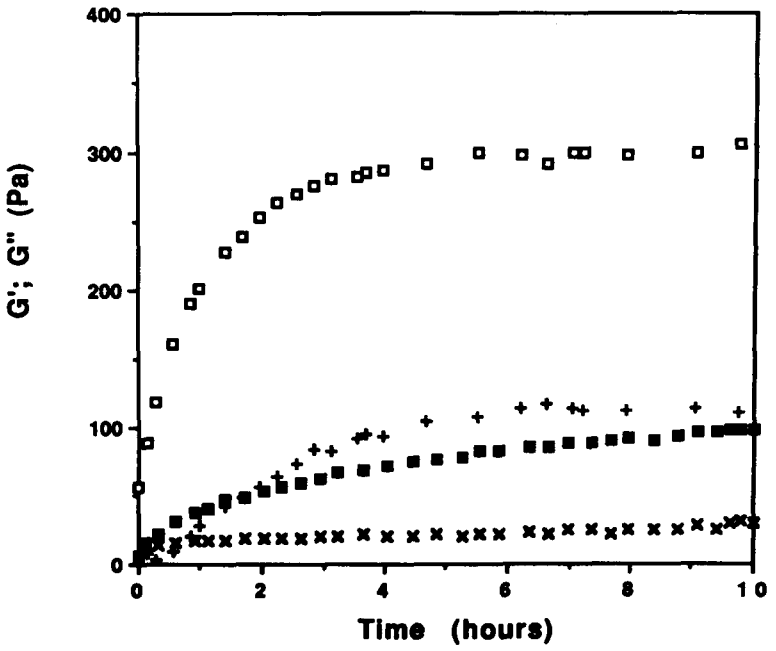

Fig. 1. Kinetics of gelation of a kappa-carrageenan gel and a kappa-carrageenan/Indal mixture 80/20 (total polymer concentration: 1·0%). ■, G'; ×, G'' — kappa-carrageenan gel; □, G'; +, G'' — mixed gel. Frequency: 1·0 Hz; temperature: 15°C.

a 1% total concentration. For carrageenan alone, it is clearly seen that gelation proceeded slowly and 10 h were required in order to reach a steady value of about 90 Pa. In contrast, for the mixed system, the gelation was more rapid and a plateau value of the order of 300 Pa was reached within about 3–4 h. Thus, the final G' value was three times higher for the mixed gel compared with kappa-carrageenan alone.

Figure 2 shows the kinetics of gelation of 80/20 mixed gels at temperatures ranging from 10°C to 25°C. Whatever the temperature, G' was seen to increase rapidly at first for about 2 h and then more slowly. G'' variations are not shown in this figure but it was observed that $G' > G''$ at all times. Figure 3 displays the variation in the loss tangent ($\tan \delta = G''/G'$) variations during the gelation of kappa-carrageenan 1·0% and kappa-carrageenan/LBG 80/20 1·0%, at 15°C. These changes in tan δ suggest that the mechanism of gelation of the kappa-carrageenan and of the mixed gel are very different. For kappa-carrageenan alone, G''/G' is of the order of 0·8 at the beginning and of 0·3 at the end. That means that a sol–gel transition takes place slowly. In contrast, for the mixed system, G' is much higher than G'' from the very beginning. That

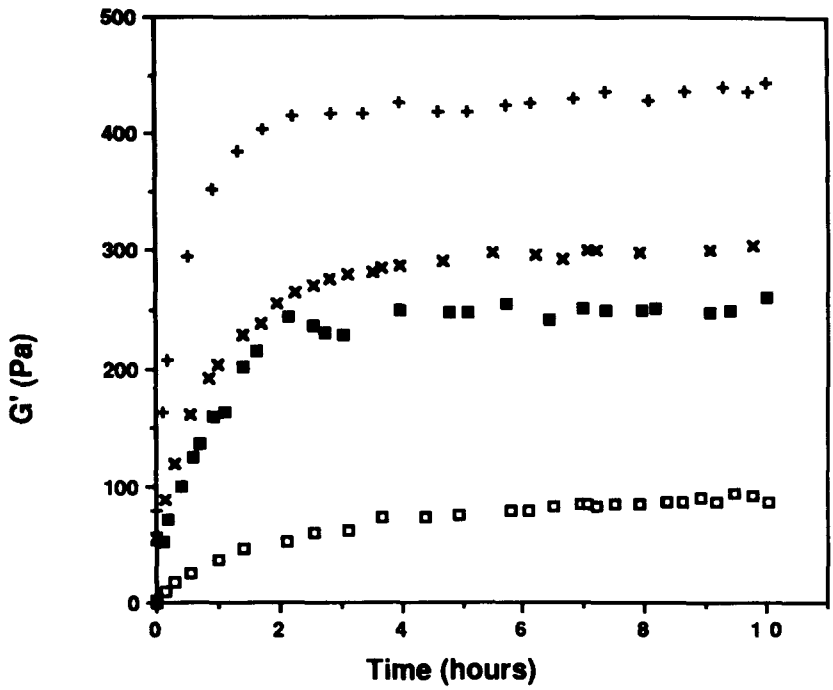

Fig. 2. Kinetics of gelation of a kappa-carrageenan/Indal 80/20 system at different temperatures (total polymer concentration, 1·0%; frequency: 1·0 Hz); temperatures: (+) 10°C; (×) 15°C; (■) 20°C; (□) 25°C.

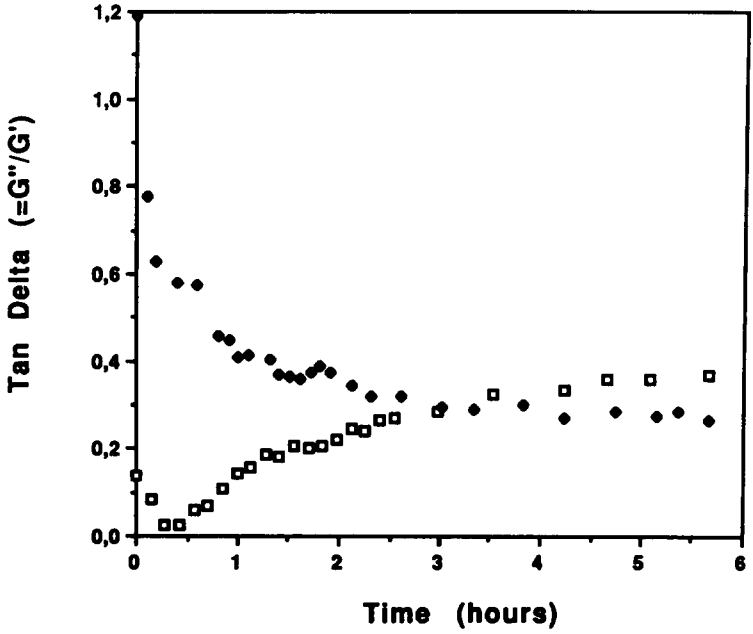

Fig. 3. Tan δ (G''/G') evolution during the gelation of a kappa-carrageenan gel (◆) and a kappa-carrageenan/Indal 80/20 mixture (□). Total polymer concentration: 1·0%; frequency: 1·0 Hz; temperature: 15°C.

means that the gel is formed almost immediately. Surprisingly, the G''/G' ratio tends to increase with time and reaches a final value of the same order as with the carrageenan alone. This suggests that the elastic character of the gel is less pronounced at the end of the curing period than at the beginning. It is to be noted, however, that the present data were obtained from measurements at a fixed frequency (1 Hz) while a better understanding of the curing process would require the description of the entire mechanical spectra at any time.

Characterization of the gels

The mechanical spectra of kappa-carrageenan gels at 1·0% (no KCl added) and 0·3% (0·3% KCl) are shown in Fig. 4. The gels, without KCl addition, showed a slight dependence of G' and G'' with frequency. The G''/G' ratio was of the order of 0·3 over the whole frequency range. These are the characteristics of a weak gel (Clark & Ross-Murphy, 1987). In contrast, for the gels with KCl, G' and G'' remained constant over the whole frequency range with $G' = 10G''$. These characteristics

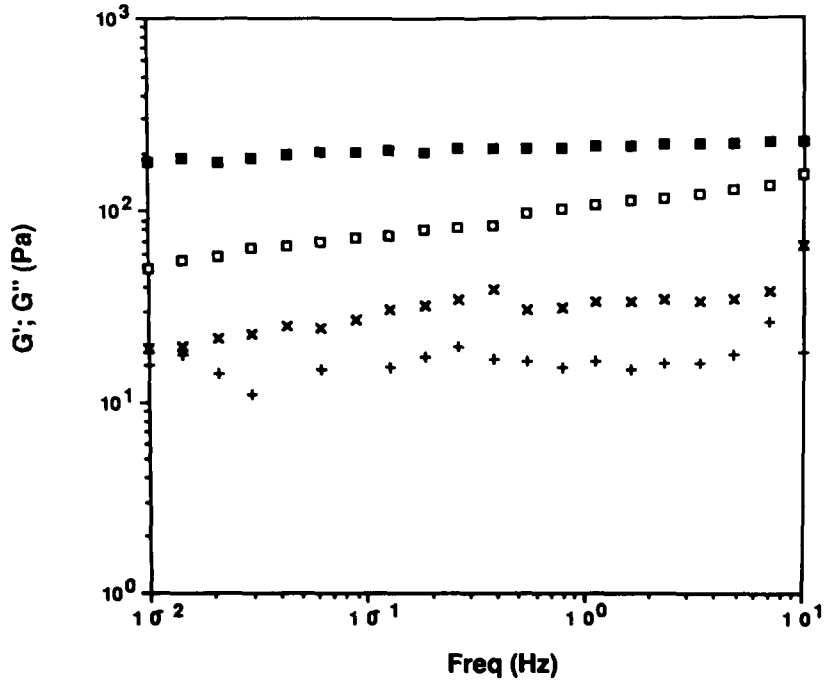

Fig. 4. Mechanical spectra of kappa-carrageenan gels at 1·0% (no KCl added) and 0·3% (0·3% KCl). Frequency: 1·0 Hz; temperature: 15°C. (□) G' and (×) G'', 1·0%; (■) G' and (+) G'', 0·3%.

correspond to those of a true gel. As expected, the addition of K^+, yields stronger gels with a less viscoelastic character.

The mixed gels that were aged inside the rheometer system yielded lower G' values in comparison with the same mixed gel prepared in a glass mould at the same temperature. Apparently, the differences are due to the rate at which the temperature of 15°C was reached in each case. In the rheometer, the hot solution was poured onto the plate at 40°C, and then the desired temperature was automatically attained in a few seconds whereas in the glass mould, the temperature of 15°C would take several minutes to be reached. Since kappa-carrageenan gels are time- and temperature-dependent it is possible that the different temperature histories provide a basis for the interpretation of the differences. However, this assumption has to be confirmed.

In order to compare the effects of the different LBG samples on the properties of mixed kappa-carrageenan/LBG gels, dynamic measurements were performed at variable ratios of the two polysaccharides over a frequency range of 0·01–10 Hz. The G' values

obtained for two LBG samples taken as examples at the frequency of 1·129 Hz are plotted versus the kappa-carrageenan/LBG ratio at a total polysaccharide concentration of 1·0%, no KCl being added, in Fig. 5 and at a total polysaccharide concentration of 0·3% with KCl in Fig. 6. It is seen that a synergistic maximum was obtained when the kappa-carrageenan/LBG ratio was of the order of 80/20. All the LBG samples showed similar patterns. At the polysaccharide ratio corresponding to maximum synergism, marked differences between the LBG samples were in the storage modulus G'. Table 2 displays the G' value at the maximum ratio (G'_{max}) together with the intrinsic viscosity and M/G ratio values for all LBG samples. Results for tara and guar gums are also given. The comparison of the mixed gels from the different LBG samples with and without KCl addition, indicates that two categories of gels can be distinguished: (a) galhosa, mulata (two laboratory samples) and Indal (an industrial gum); and (b) canela (a laboratory sample) and Lygomme 6 (an industrial gum). It is noteworthy that these two groups do not reflect

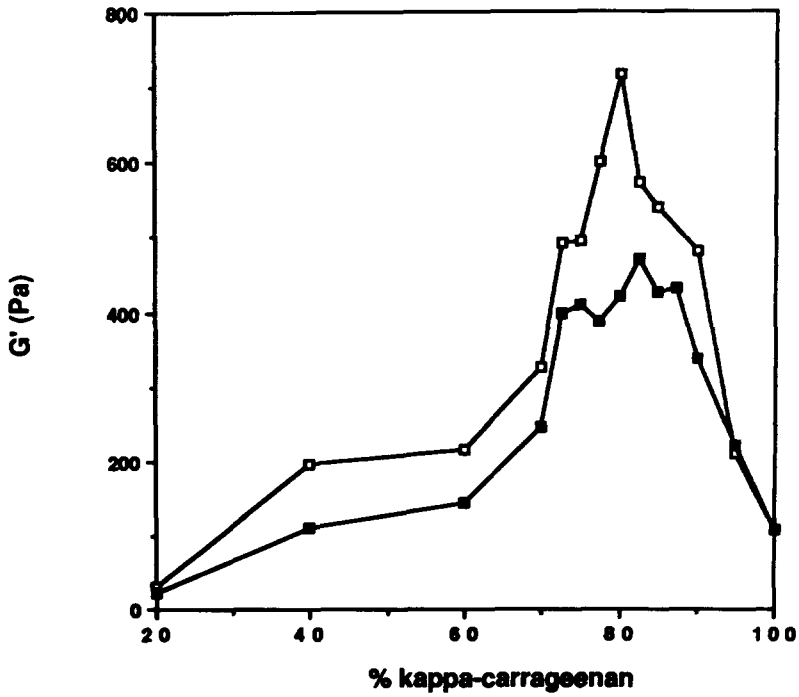

Fig. 5. Effect of LBG carrageenan ratio on the storage modulus (G') of mixed gels (total polymer concentration: 1·0%), at a frequency of 1·13 Hz. Frequency range: 0·01–10 Hz; temperature: 15°C. □, galhosa gum; ■, canela gum.

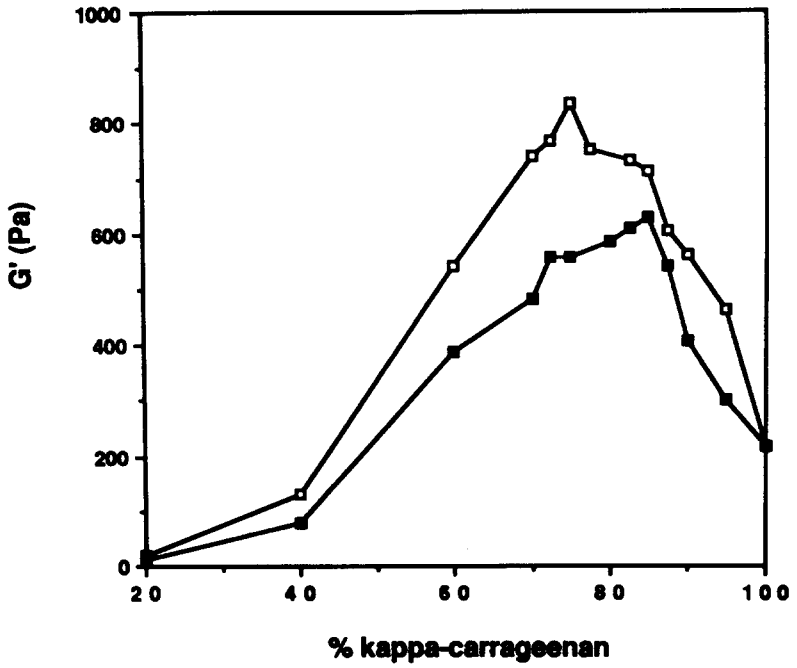

Fig. 6. Effect of LBG carrageenan ratio on the storage modulus (G') of kappa-carrageenan/LBG mixed gels (total polymer concentration: 0·3%) with KCl addition. Frequency: 1·13 Hz; temperature: 15°C. □, galhosa gum; ■, canela gum.

TABLE 2
Characteristic Parameters of Mixed Gels

Samples	*[η]*[a] *(dl/g)*	*M/G*[b]	G'_{max}[c] *without* K^+	*κ-carrag/galact*[d]	G'_{max}[c] *with* K^+	*κ-carrag/galact*[d]
Canela	13·8	3·46	471	82·5/17·5	629	85/15
Galhosa	15·3	4·04	716	80/20	838	75/25
Mulata	14·3	3·80	596	80/20	710	80/20
Indal	15·4	4·04	637	80/20	916	80/20
Lygomme 6	10·5	3·60	477	85/15	601	77·5/22·5
Tara	9·4	3·00	329	80/20	—	—
Guar	12·0	1·68	31·6	—	—	—

[a] $[\eta]$: intrinsic viscosity.
[b] M/G: mannose to galactose ratio.
[c] G'_{max}: maximum value (Pa) at a frequency of 1·129 Hz.
[d] Kappa-carrageenan/galactomannan ratio at G'_{max}.

a distinction between laboratory-scale preparations and industrial gums. Figure 7 displays the mechanical spectrum of a 1% mixed gel of kappa-carrageenan/galhosa 80/20. This contrasts with the mechanical spectrum of kappa-carrageenan systems as seen in Fig. 4 and is typical of a strong gel with $G' = 10\,G''$ and G' and G'' almost independent of frequency.

Gels at a total polymer concentration of 0·3%, with KCl, displayed syneresis. This was quantified by measuring the water loss of kappa-carrageenan/LBG mixed gels. We found identical gel syneresis for the five LBG samples at each carrageenan/LBG ratio. The water loss thus ranged from 24% without LBG to 2% at a 20/80 ratio; it was 12% at 80/20, and 6% at 60/40 and 40/60. The dry matter content should thus be corrected taking into account these values, particularly at a low galactomannan content (Baidon *et al.*, 1987). However, since gel syneresis was limited and identical for all LBG samples, the results on mixed gels containing potassium are presented in Table 2 without correction for water loss.

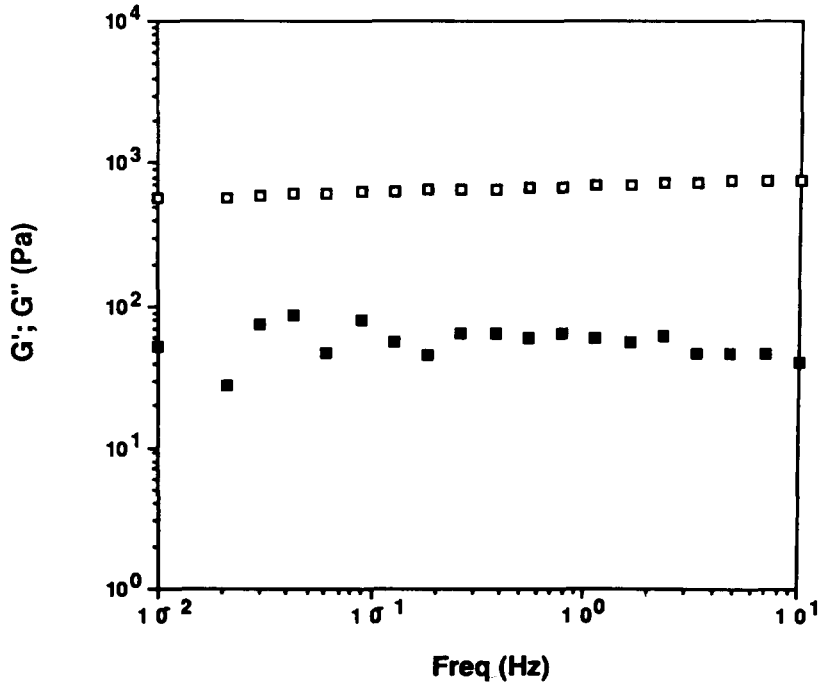

Fig. 7. Mechanical spectrum of a kappa-carrageenan/galhosa 80/20 mixed gel at 1·0% total polymer concentration (no KCl added). Frequency range: 0·01–10 Hz; temperature: 15°C. □, G'; ■, G''.

Interactions of kappa-carrageenan with other galactomannans

It was interesting to check whether other types of galactomannans could display the same type of behavior with kappa-carrageenan. For this purpose, we used a tara gum and a guar gum and also prepared CWS and HWS fractions from the different LBG samples. Figure 8 shows that a similar pattern to the LBG sample was obtained with tara gum which also displayed a synergistic maximum at 80/20, G'_{max} being lower than for LBG. In contrast, guar gum did not display a maximum at 80/20. Unfortunately, we could not prepare large amounts of CWS and HWS fractions of each LBG sample which would allow us to investigate a range of carrageenan/LBG ratios. Gels were prepared at the 80/20 ratio at 1·0% total polymer concentration, with no KCl addition. The results obtained from dynamic measurements are presented in Table 3 together with the molecular characteristics ($[\eta]$ and M/G) of each fraction. Much higher G'_{max} values were obtained with HWS fractions, which are less

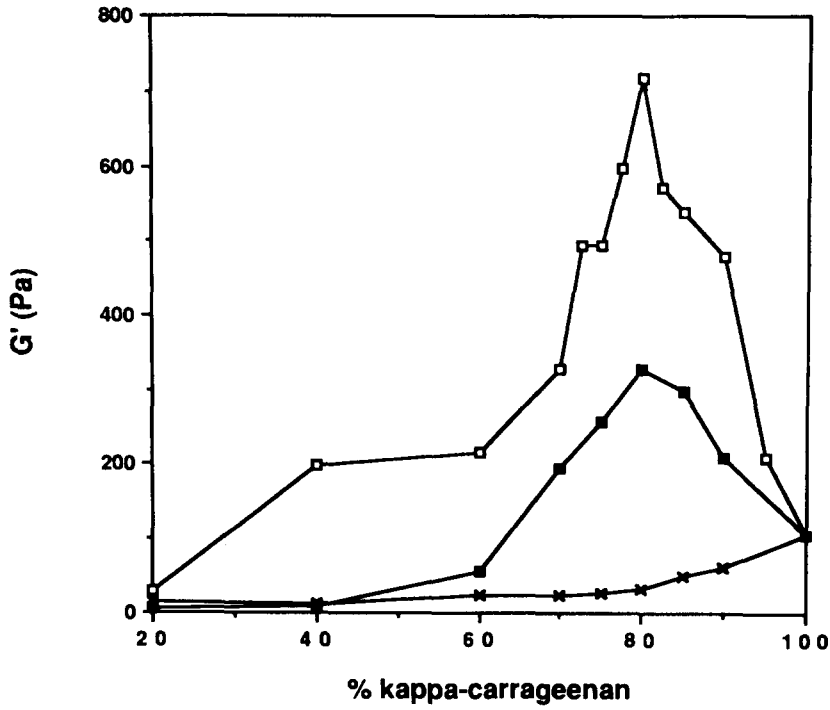

Fig. 8. Effect of galactomannan/carrageenan ratio on the storage modulus (G') of kappa-carrageenan/galactomannan mixed gels (total polymer concentration: 1·0%; frequency: 1·13 Hz; temperature: 15°C. □, galhosa gum; ■, tara gum; ×, guar gum.

TABLE 3
Characteristic Parameters of HWS and CWS Fractions as Related to G'_{max} of Mixed Gels at an 80/20 Carrageenan/Galactomannan Ratio

LBG samples	*CWS*			*HWS*		G'_{max}[a]	
	% extracted	$[\eta]^b$	*M/G*[c]	$[\eta]^b$	*M/G*[c]	*CWS*	*HWS*
Canela	43·4	13·7	3·22	14·5	3·81	420	630
Galhosa	39·0	12·4	3·34	16·0	4·57	455	827
Mulata	42·5	13·4	3·47	16·9	4·25	504	815
Indal	34·5	12·7	3·48	16·0	4·80	460	907
Lygomme 6	46·9	7·6	3·29	12·6	4·18	250	615

[a]G'_{max}: maximum value of G'(Pa) at a frequency of 1·129 Hz.
[b]$[\eta]$: intrinsic viscosity (dl/g).
[c]M/G: mannose to galactose ratio.

galactose substituted than with the CWS ones. Also, these are higher than those of the original LBG samples whereas those of CWS fractions are lower and close to that of tara gum. These results can be explained in terms of the M/G ratio, that is the amount of unsubstituted mannan zones. It is to be noted, however, that there seems to exist a relationship between G'_{max} and the intrinsic viscosity of the samples, suggesting that molecular weight also has an effect on the synergistic interactions.

DISCUSSION

Tables 1 and 3 reflect the differences which can occur between LBG samples. The intrinsic viscosity can vary to a large extent whereas the M/G ratio varies only slightly. The CWS fraction amounts to between 35% and 47% of the total polysaccharide — the highest value being obtained for Lygomme 6 and canela, and the lowest for Indal. All these CWS fractions had the same M/G ratio but differed in molecular weights as measured from the intrinsic viscosity. In contrast, there were significant differences in the M/G ratios of the HWS fractions — Indal gum having the highest M/G ratio and canela gum the lowest one. However, the intrinsic viscosity of HWS fractions did vary to a lower extent than that of the CWS ones. The total gum extracted at 25°C seems thus to be related to the M/G ratio of the initial samples, Canela and Lygomme 6 displaying the lowest M/G values (Table 1).

Gel cure experiments (Fig. 2) showed a strong temperature dependence of the kinetics of gelation. We found a progressive and slow gelation at 25°C yielding a weak gel whereas for the other three temperatures (10, 15 and 20°C) a much more rapid increase in G' was seen. Similar effects were observed for kappa-carrageenan alone. These can be explained on the basis of the phase diagram established by Rochas & Rinaudo (1980) for kappa-carrageenan. The authors showed that below 20°C, even if no KCl is added, these gels lie well within the gel phase, while at 25°C they are located at the boundary between the gel phase and the sol phase.

A maximum synergistic interaction is obtained for a 80/20 carrageenan/LBG ratio both with and without KCl. This ratio was the same for all the LBG samples. It is noteworthy that tara gum gave a similar pattern (Fig. 8). The results for LBG are consistent with those obtained by Tako and Nakamura (1986) who worked with quite similar conditions for gel preparation and characterization (Table 4). Arnaud *et al.* (1989) reported a higher ratio (92/8) for gels without KCl measured in oscillatory shear and compressional testing but the concentration was much higher (3%) than employed in this work. Other data from compressional testing, on the other hand, resulted in a lower ratio for G'_{max} (Table 4) which may reflect strongly the effects of concentration, KCl content and temperature.

The two examples in Figs 5 and 6 reflect the maximum and minimum degrees of synergism found for the different LBG samples. The maximum synergistic interaction can be estimated from the increase of G' of the mixed system with respect to the G' value of kappa-carrageenan alone ($\Delta G'_{max}/G'_{\kappa}$-carrageenan). This varied from 5·7 (galhosa) to 3·4 (canela) without KCl compared with 2·4 (galhosa) and 1·9 (canela) in the presence of KCl. From this comparison, it is clear that the synergistic interactions and the differences between samples are more apparent without KCl addition.

A rapid examination of data in Tables 2 and 3 does not suggest an obvious relationship between G'_{max} and the molecular characteristics of the samples. However, the differences observed in mixed gels with LBG samples, tara gum, and also the CWS and HWS fractions suggest a dependence upon the fine structure of the galactomannan chain. Figures 9 and 10 show the plots of G'_{max} as a function of $[\eta]^*(M/G)^2$. Figure 9 relates to the five LBG samples with and without KCl whereas Fig. 10 is a compilation of all data at 1% total concentration without KCl. Two linear plots are clearly shown in Fig. 9. This finding confirms that the synergy is governed by the size or the number of non-substituted

TABLE 4
Comparison of Data of Kappa-carrageenan/LBG Mixed Gels

Galactomannan	*Maximum synergy*[a,b]	*Experimental testing*	*Measurement temperature (°C)*	*Reference*
LBG	3:1 (0·8%)	Dynamic measurements	0–30	Tako & Nakamura (1986)
LBG, tara	2:1 (1–3%)	Compression testing	20	Cairns *et al.* (1986)
LBG	1:1 (0·5–1·5%)	Compression testing	Room temp.	Fiszman *et al.* (1987)
LBG	92:8 (3%)	Dynamic measurements	4	Arnaud *et al.* (1989)
LBG	92:8 (3%)	Compression testing	4	Arnaud *et al.* (1989)
LBG, tara	4:1 (1%)	Dynamic measurements	15	Present work

[a] κ-Carrageenan/galactomannan ratio at maximum synergism.
[b] Experimental concentrations are given in parentheses.

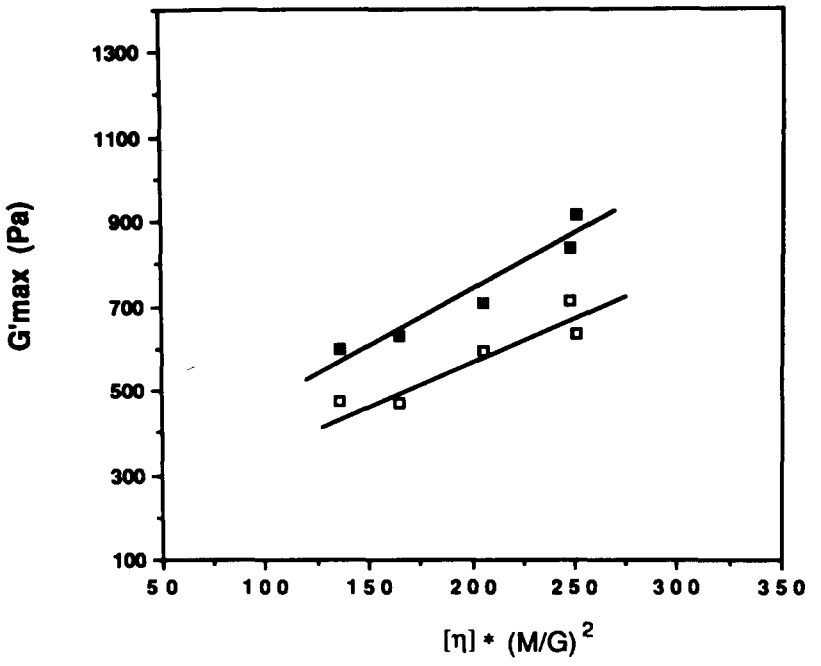

Fig. 9. Maximum G' values (G'_{max}) of kappa-carrageenan/LBG mixed gels as a function of the product $[\eta]^*(M/G)^2$. □, Mixed gels at 1·0%, no KCl added; ■, mixed gels at 0·3% with KCl addition.

mannose chain regions (as roughly estimated by the M/G ratio), but it is also necessary to take into account the molecular size of the main chain of the galactomannan, as estimated from $[\eta]$. This is confirmed in Fig. 10 where it is seen that tara gum, CWS and HWS fractions of LBG samples behave in a similar manner. The relationship can thus be extended to a wider range of $[\eta]^*(M/G)^2$ values. The only exception is the CWS fraction of Lygomme 6 which lies slightly below the straight line ($G'_{max} = 250$ Pa). This fraction did not differ from the other CWS fractions in its M/G ratio but had a lower intrinsic viscosity. This suggests that its lower G'_{max} is to be ascribed to a lower molecular weight. The result obtained for guar gum is also given in Fig. 10 ($G' = 31{\cdot}6$ Pa) although, as shown in Fig. 8, there was no real synergy with this sample because the galactose-free regions in the galactomannan backbone are too short.

Molecular weight and M/G ratio are thus two primary parameters to be taken into account; their effects can be described in the following manner: at a given M/G ratio, the higher the molecular weight, the higher

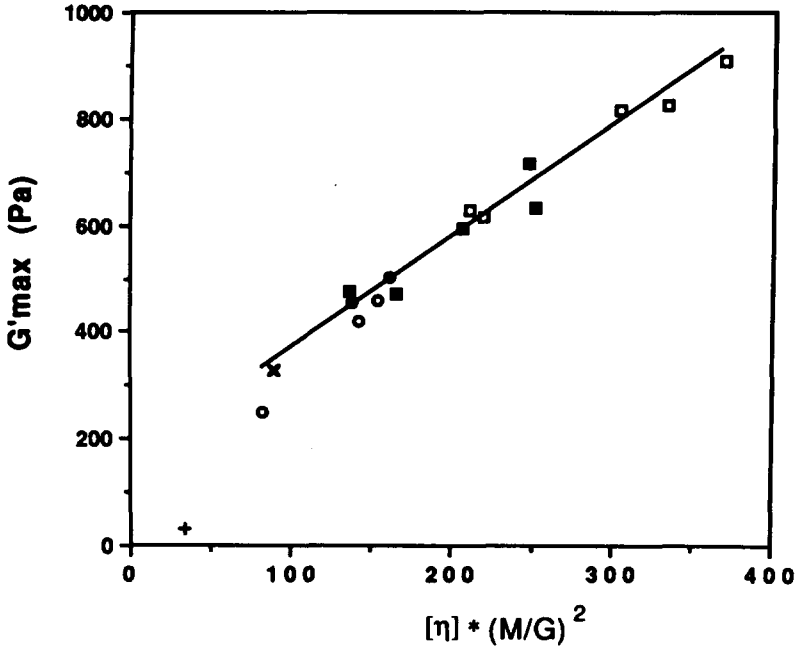

Fig. 10. Maximum G' values (G'_{max}) of mixed gels of kappa-carrageenan/galactomannan mixed gels (no KCl added) as a function of the product $[\eta]^*(M/G)^2$. ×, tara gum; +, guar gum; ■, LBG samples; ○, CWS fractions; □, HWS fractions.

the synergy and, for a given $[\eta]$, G'_{max} is related to M/G^2. The coupled network model described by Dea and Morrison (1975) assumes intermolecular binding between the carrageenan double helices and the smooth regions of the galactomannan chains. Provided that the same macromolecule is long enough to accommodate several carrageenan double helices, it is believed that such a mechanism is able to reinforce the carrageenan network. This imples that the size and the number of the galactose-free regions in the mannan backbone play a major role in the phenomena. This is confirmed in the present work. However, this model would not involve such a strong effect of molecular weight as we observed. Cairns *et al.* (1987) suggest that a basic polymer network is formed by the kappa-carrageenan and this network would contain the galactomannan molecules in solution. They do not ascribe a specific role to the galactomannan. We believe that the present results are consistent with their model. The important role played by the galactomannan molecules does suggest that galactomannan–galactomannan aggregation takes place through galactose-free regions of the mannan backbone giving rise to a secondary network whose rheology would depend on the molecular size. We suggest that both gelation processes yield the

formation of an interpenetrating polymer network whose rheological properties are far removed from those of the original polymers.

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