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Behavior of κ-carrageenan in glycerol and sorbitol solutions

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Abstract

The solubility of κ-carrageenan in low water-content solvents is important in food applications where complete solubilization is required for proper development of structure and rheology. The effect of glycerol and sorbitol on the gelation and conformational helix transition of κ-carrageenan was studied using rheology and optical rotation. Glycerol/water solutions from 0–100 wt% glycerol and sorbitol solutions from 0–100% saturation were studied over the temperature range 0–90°C. The results were analyzed in terms of solvent solubility parameters, water chemical potential, and solvent dielectric constant. Effective cohesive energy density parameters could not be inferred for the carrageenan, but the gelation temperature could be correlated with solvent dielectric constant. Hydrogen bonding interactions control the carrageenan helix formation. The cohesive energy density as a measure of solvent quality accounts for hydrogen bonding but not Coulombic interactions, and the Coulombic interactions scale on dielectric constant. This indicates the dominant role of electrostatics on the gelation process. © 2000 Elsevier Science Ltd. All rights reserved.

Keywords: Solubility of κ-carrageenan; Glycerol and sorbitol solutions; Dielectric constant

1. Introduction

Carrageenans are sulfated galactans extracted from red seaweeds (Fig. 1). At high temperatures, the polymer is present as a random coil. Upon cooling, it undergoes a conformational transition, forming double helices (Hjerde, Smidsrod & Christensen, 1999; Millane, Chandrasekaran, Arnott & Dea, 1988). In the case of κ-carrageenan, these double helices aggregate in the presence of specific cations to form a gel (Borgstrom, Picullel, Viebke & Talmon, 1996; Ciancia, Milas & Rinaudo, 1997; Michel, Mestdagh & Axelos, 1997; Robinson, Morris & Rees, 1980). The biocompatability of these gels makes them valuable as stabilizing, thickening and gelling agents in a variety of food products such as chocolate milk, ice cream and bakery products (Therkelson, 1993). They are also used in the pharmaceutical industry (Guo, Skinner, Harcum & Barnum, 1998; Picker, 1999), and in household products (Guiseley, Stanley & Whitehouse, 1980).

We have studied the gelation and helix–coil transition of κ-carrageenan in aqueous solutions of glycerol and sorbitol (Ramakrishnan & Prud'homme, 2000). We found that the transition shifted to higher temperatures with increasing glycerol content. Also, at higher glycerol concentrations, the lower solubility of carrageenan in glycerol resulted in less hysteresis between the heating and cooling curves for G' and the optical rotation. Here, we compare the effect of glycerol with that of sorbitol on the gelation and conformational transition. We attempt to generalize the effect of solvents in terms of the chemical potential of water, solubility parameters, and the dielectric constant.

2. Experiments

A κ-carrageenan sample containing 5 wt% Na^+ and 0.2 wt% K^+—referred to as Na^+-carrageenan—was supplied by the Food Ingredients Division of FMC Corporation, New Jersey. The melting transitions and gelation of the above samples were studied with dynamic oscillatory measurements during temperature ramps between 0 and 80°C using the Peltier heating option with parallel plate geometry (40 mm diameter) on the Dynamic Stress Rheometer (DSR) (Rheometrics, Inc.).

The conformational transition was followed by measuring the optical rotation with a Rudolph spectropolarimeter, AUTOPOL IV (Rudolph Research Corporation, Flanders, New Jersey). Carrageenan helices are circularly birefringent, and hence retard the right-handed and left-handed components of circularly polarized light by different amounts, resulting in a rotation of the polarization ellipse (Djerassi, 1960). The formation of helices upon cooling is

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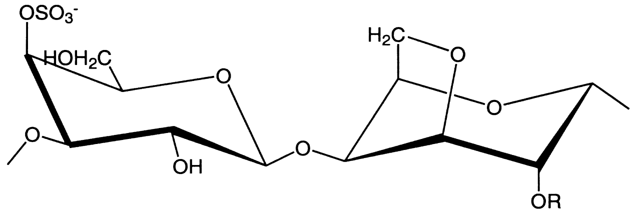

Fig. 1. Repeating disaccharide structures of κ (R = H) and ι ($R = SO_3^-$)-carrageenan.

accompanied by a significant increase in the optical rotation, which allows quantification of the coil to helix transition. The path length of the optical rotation cell was 100 mm and the wavelength of light was 589 nm. A computer controlled circulating bath maintained a ramp rate of 1°C/min.

3. Results and discussion

3.1. Rheology and optical rotation

The melting transition of carrageenan in aqueous solutions of glycerol and sorbitol was studied by measuring the complex moduli and the optical rotation while imposing a temperature ramp. Samples of 1% Na^+-carrageenan with 0.01 M KCl in solutions of water, 50% glycerol, 100% glycerol, 50% saturated sorbitol and 100% saturated (≈70 wt%) sorbitol were melted at 80°C. Melting was verified by the absence of a measurable low frequency modulus and a stable, near baseline value of the optical rotation. These were then cooled to 0°C and re-heated to 80°C at a rate of 1°C/min. Figs. 2 and 3 show the results for glycerol and sorbitol. The OR data for samples in glycerol and sorbitol are plotted in the same graph for better comparison in Fig. 4. The optical rotation was normalized by taking the minimum OR signal at high temperature as zero percent helicity and the maximum OR signal at low temperature as complete helicity. For all samples (except as discussed below), the OR transition is sigmoidal and saturated at high and low temperatures, indicating that the fractional helicity spans zero to one. For 50% glycerol and 50% sorbitol, the temperature axis is normalized by plotting the experimental temperature minus the mid-point temperature of the transition, T_m (i.e the temperature at which the fractional helicity is 0.5).

These results show that the melting transitions are quite

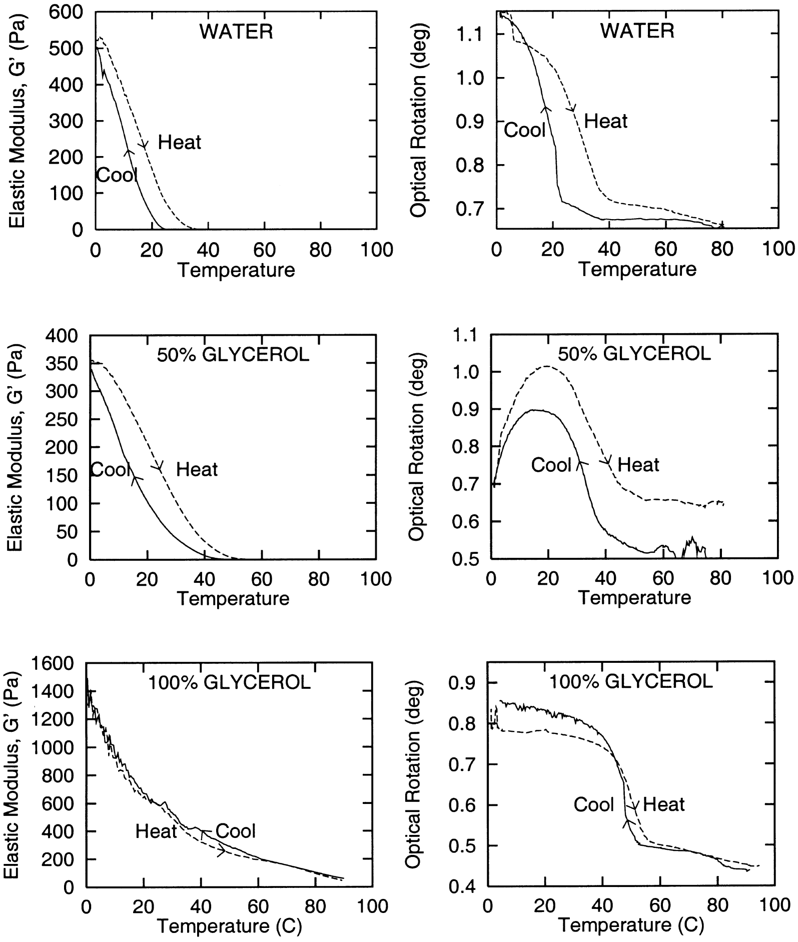

Fig. 2. Rheology (stress of 0.5 Pa at 1 rad/s) and optical rotation (589 nm wavelength, 100 mm cell) during temperature ramps at 1°C/min on samples of 1 wt% Na^+-carrageenan with 0.01 M KCl in water, 50 and 100% glycerol. The samples were initially melted at 80°C, then cooled to 0°C and re-heated to 80°C.

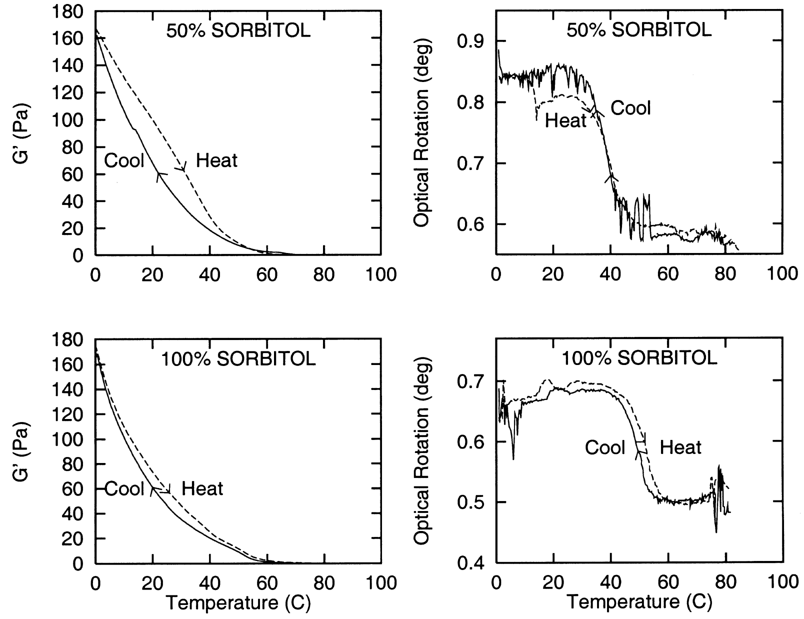

Fig. 3. Rheology (stress of 0.5 Pa at 1 rad/s) and optical rotation (589 nm wavelength, 100 mm cell) during temperature ramps at 1°C/min on samples of 1 wt% Na^+-carrageenan with 0.01 M KCl in water, 50% saturated and 100% saturated sorbitol. The samples were initially melted at 80°C, then cooled to 0°C and re-heated to 80°C.

similar in glycerol and sorbitol. The shapes of the optical rotation curves while cooling and heating are identical, and for the samples in 100% glycerol and 100% saturated sorbitol, the conformational transition occurs at almost the same temperature. Also, as observed in glycerol, there is no hysteresis between the cooling and heating curves for carrageenan in aqueous solutions of sorbitol (Fig. 3). This arises because carrageenan being less soluble in glycerol and sorbitol, comes out of the solution as soon as the double helices are formed (Ramakrishnan & Prud'homme, 2000). Hence, the polymer forms a more random network of double helices than the ordered network of aggregates seen in

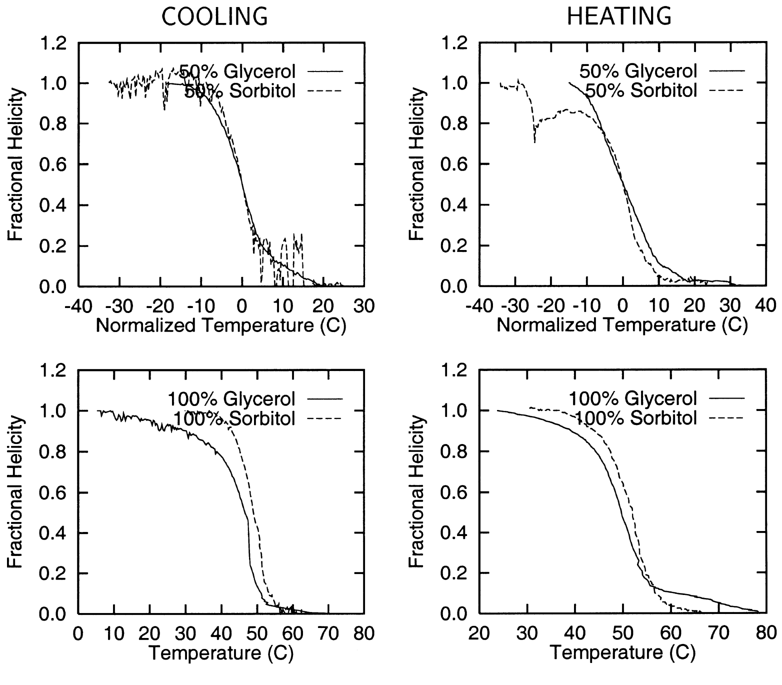

Fig. 4. OR data (589 nm wavelength, 100 mm cell) from Figs. 2 and 3 for 1 wt% Na^+-carrageenan with 0.01 M KCl in 50 and 100% solutions of glycerol and sorbitol (saturated). The optical rotation was normalized by taking the minimum OR signal at high temperature as zero helicity and the maximum OR signal at low temperature as complete helicity. The mid-point temperature of the transition was subtracted from the actual temperature for samples in 50% glycerol and 50% sorbitol for easier comparison. Figures on the left are for cooling, those on the right are for heating.

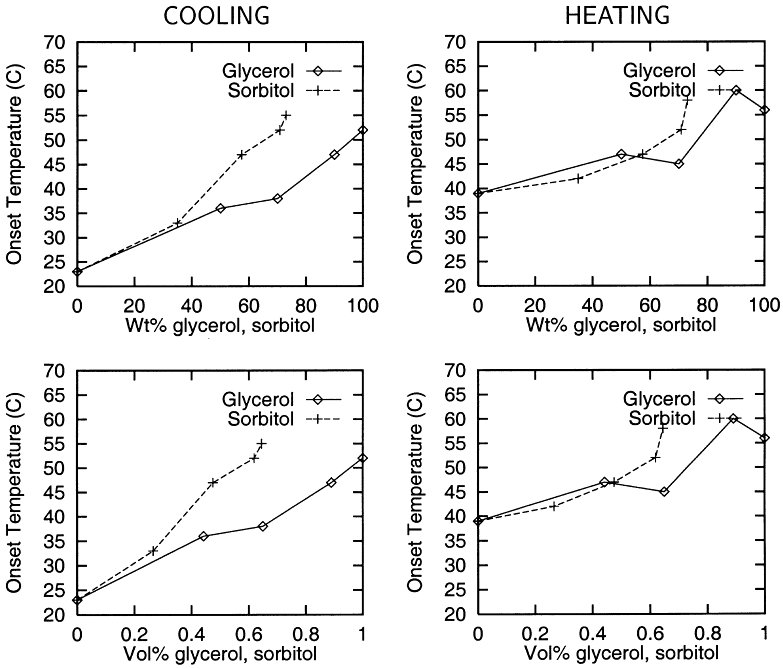

Fig. 5. The onset temperature of the transition for samples of 1% Na$^+$-carrageenan with 0.01 M KCl is plotted as a function of the wt% and vol% of glycerol and sorbitol in the solvent. Figures on the left are for cooling, those on the right are for heating.

water. This is also supported by the fact that the optical rotation does not decrease at low temperatures due to the ordered aggregates rotating light in the opposite direction to that of the individual helices. Arscott, Ma, Wenner and Bloomfield (1995) observed similar behavior in DNA as the dielectric constant of the solvent is decreased. The fluid to gel transition is gradual and occurs over the same temperature range for both glycerol and sorbitol.

The above results indicate that both glycerol and sorbitol affect the conformational transition, and the mechanism of

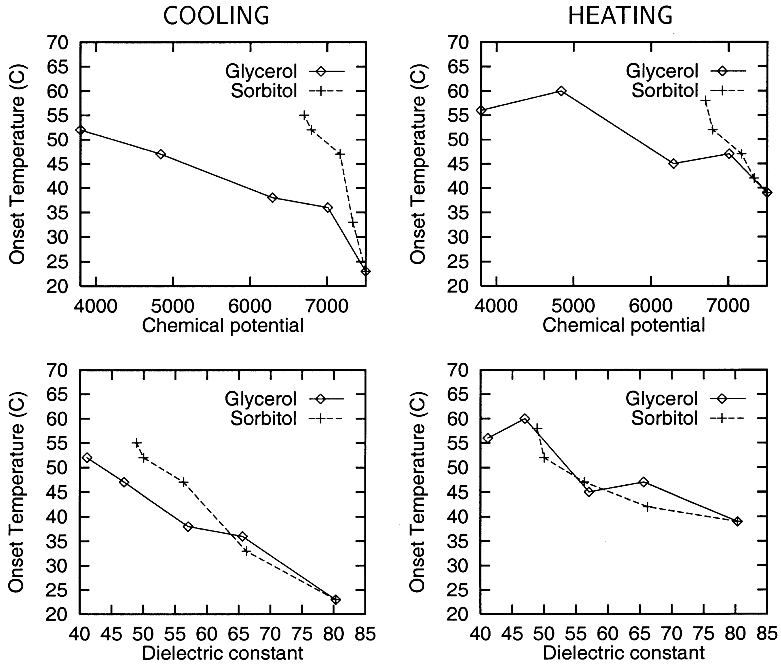

Fig. 6. The onset temperature of the transition for samples of 1% Na$^+$-carrageenan with 0.01 M KCl in aqueous glycerol and sorbitol solutions is plotted as a function of the chemical potential of water (relative to the potential at a pressure of 1 mmHg) and the dielectric constant of the solvent (aqueous solutions of glycerol and sorbitol). The calculations of the chemical potentials and dielectric constants for the solutions as a function of water content are described in the text. Figures on the left are for cooling, those on the right are for heating.

Table 1
Solubility parameters of the aqueous mixtures in which carrageenan was just soluble. The solubility parameters for the pure solvents were obtained from Barton (1983) and those for the aqueous mixtures were calculated using the mixing rule

Solvent	Solubility limit (Wt% Solvent)	δ_d (MPa$^{1/2}$)	δ_p (MPa$^{1/2}$)	δ_h (MPa$^{1/2}$)
THF	50	12.78	16.546	22.54
Methanol	45	11.9	17.8	32.04
MEK	25	12.73	19	31.72
Acetonitrile	57	11	15.45	27.34
Acetone	55	12.68	14.91	22.55
Ethanol	37	12.38	17.85	31.7
Pyridine	70	16	13.86	17.38
1,4-Dioxane	55	15.89	11.42	22.5
Propylene glycol	70	11.92	16.22	29.73

gelation of carrageenan in a similar manner. Fig. 5 illustrates the effect on the the onset temperature of the transition for samples of 1% Na$^+$-carrageenan with 0.01 M KCl. The onset temperature while cooling is the temperature at which there is a sharp increase in the optical rotation indicating the co-operative formation of helices. While heating, the onset temperature refers to the temperature at which helix to coil transition is completed. The onset temperature increased with increasing concentration of both glycerol and sorbitol, though this trend was less well defined in the case of glycerol while heating. However, no direct correspondence could be found between the wt% or vol% of glycerol and sorbitol and the onset temperature. We then attempted to analyze the effect of the solvent in terms of the chemical potential of water, solubility parameters, and the dielectric constant.

3.2. Chemical potential

The chemical potential of water in the gas phase was calculated from its vapor pressure using

$$\mu_w = \mu_{w_0} + RT \ln p_w \tag{1}$$

where p_w is the partial pressure of water, and μ_{w_0} is the chemical potential at unit pressure (taken to be 1 mmHg). Since the vapor phase is in equilibrium with the liquid phase, the chemical potential of water in the solution is the same as that in the vapor. The chemical potentials of water in aqueous solutions of glycerol and sorbitol, relative to the potential at 1 mmHg, were calculated from the vapor pressures of glycerol (Segur, 1953) and sorbitol (Vogel, Merck & Darmstadt, 1994) solutions obtained from literature. The chemical potential of water decreases in a similar manner with the addition of glycerol and sorbitol. However, for the same chemical potential of water, the onset temperature of the transition differs for carrageenan in glycerol/water versus sorbitol/water solutions (Fig. 6); therefore the chemical potential of water does not adequately correlate the transition temperatures.

3.3. Solubility parameters

The Hansen solubility parameters give a quantitative measure of the interactions between molecules in solution and have proven to be useful in predicting polymer/solvent solubility. The cohesive energy density is assumed to be made up of a linear combination of contributions from non-polar or dispersion ($-U_d$), polar ($-U_p$), and hydrogen bonding ($-U_h$) interactions:

$$-U = -U_d - U_p - U_h \tag{2}$$

The corresponding cohesive pressures and cohesion (solubility) parameters are defined as follows:

$$-\frac{U}{V} = -\frac{U_d}{V} - \frac{U_p}{V} - \frac{U_h}{V} \tag{3}$$

$$\delta^2 = \delta_d^2 + \delta_p^2 + \delta_h^2 \tag{4}$$

where V is the molar volume.

The solubility parameters for glycerol are available (Barton, 1983) but not those for sorbitol. In order to better understand how the solubility parameters of the solvent could affect the behavior of carrageenan, we attempted to determine the effective solubility parameters of the carrageenan polymer by constructing a "solubility sphere" in the three-dimensional solubility parameter space. The center of the sphere would then correspond to the solubility parameters for carrageenan, and solvent mixtures at equal distances from the center of the sphere would have equal solvent qualities with respect to carrageenan.

A number of solvents that are miscible in water were chosen. Starting with the pure solvent in which carrageenan was insoluble, water was added until the polymer just became soluble. This yielded a point on the solubility sphere, beyond which carrageenan would not be soluble. Solvents were chosen with as wide a difference in δ_d, δ_p and δ_h as possible. Table 1 lists the solvents used, the aqueous mixture which was just able to dissolve carrageenan, and the solubility parameters. The solubility parameters for the mixture were calculated from those of the pure solvent and water using the mixing rule (Barton, 1983):

$$\bar{\delta} = \frac{\sum_i \phi_i \delta_i}{\sum_i \phi_i} \tag{5}$$

where ϕ_i is the volume fraction, and δ_i is the solubility parameter of component i. The volume fraction of the polymer was assumed to be negligible, and hence $\sum_i \phi_i \approx 1$. A three-dimensional plot of the solubility parameters that are on the boundary of the area within which carrageenan is soluble appeared random and did not enclose a region in space. The lack of any noticeable pattern indicates the inadequacy of solubility parameters for correlating the transition.

3.4. Dielectric constant

The dielectric constant of the solvent plays a major role in processes involving ionization and dipole interactions.

Since the potassium and sulfate ion interactions are involved in gelation, correlation of the onset temperature with the solvent dielectric constant was attempted. The work done to bring two charges together to a distance r from infinite separation is given by

$$\Delta G = \frac{z_1 z_2 e^2}{\epsilon r} \qquad (6)$$

where z_1 and z_2 are the ionic valencies of the two ions, e is the electronic charge, and ϵ is the dielectric constant of the medium. For opposite charges, this work is negative. A decrease in the dielectric constant increases the magnitude of the net free energy change. Hence, it is harder to pull the ions away from the sulphates in glycerol ($\epsilon = 47$) than in water ($\epsilon = 80$). The values of the dielectric constants at room temperature for aqueous glycerol solutions were available in the Landolt–Börstein series (Wohlfarth, 1991). The dielectric constant of pure sorbitol at 353 K is reported as 35.5 (Lide, 1996) but values as a function of water content are not available. The dielectric constants for sorbitol/water solutions were then calculated using the mixing rules developed by Bruggeman (1935):

$$\frac{\epsilon_{\text{mix}} - \epsilon_{\text{s}}}{\epsilon_{\text{w}} - \epsilon_{\text{s}}}\left(\frac{\epsilon_{\text{w}}}{\epsilon_{\text{mix}}}\right)^{1/3} = 1 - \phi_{\text{s}} \qquad (7)$$

where ϵ_{mix}, ϵ_{w}, and ϵ_{s} are the dielectric constants of the sorbitol/water mixture, pure water and pure sorbitol, and ϕ_{s} is the volume fraction of sorbitol in water. Lou, Hatton and Laibnis (1997) compared values predicted by this mixing rule with experimental data for organic solvents and found it to be accurate to within 10% error. Fig. 6 shows that the transition temperatures are remarkably close for aqueous solutions of glycerol and sorbitol that have the same dielectric constant. Therefore, the dielectric constant, which parametrizes the strength of the Coulombic interactions, correlates the gelation transition. It also points to the dominant role of ionic interactions in the gelation process and not merely hydrogen bonding that controls the helix formation process.

4. Conclusions

Both glycerol and sorbitol affect the conformational transition and the mechanism of gelation of carrageenan in a similar manner. The onset temperature of the coil to helix transition increased from 22 to 55°C with increasing concentrations of glycerol and sorbitol. No direct correspondence could be found between the onset temperature and (i) the concentration of glycerol and sorbitol, (ii) the chemical potential of water in the solvent, or (iii) the Hansen solubility parameters. However, the dielectric constants of the glycerol/water and sorbitol/water solutions were found to correlate with the onset temperature. The correlation with the dielectric constant and the lack of correlation with the solubility parameters (which assess hydrogen bonding but not electrostatics) indicate the dominance of Coulombic interactions over short range hydrogen bonding (that controls helix formation) on the mechanism of gelation of carrageenan.

Acknowledgements

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