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Water holding capacity and microstructure of gellan gels

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Abstract

This project studied the water holding capacity of gellan gels as affected by gel composition and microstructure. When not subjected to external forces such as centrifugal force, gellan gel properties including water holding capacity and texture properties were stable at room temperatures. The water losses from gellan gels after four months storage at 4°C were only 1–2%, independent of calcium concentrations. The freeze–thaw stability of gellan gels was poor. Water holding capacity of gellan gels, when subjected to centrifugal forces, was dependent on calcium concentrations, and was related to the texture properties. Two discrete pore-size distributions in gel matrix on the order of 0.1 and 1 μ were observed with scanning electronic microscopy. Large pores were formed with thick strings while the small ones were formed by a thin web structure. Defects in the large pore structure were observed at high calcium concentrations. The small pores may be responsible for the water holding capacity during storage, while large pore structures provide the strength of gels. © 2001 Elsevier Science Ltd. All rights reserved.

Keywords: Gellan gel; Storage; Water holding capacity; Texture; Microstructure

1. Introduction

Carbohydrate polymers are important functional ingredients in many food products including puddings, mousses, jams, jellies and sauces. A major function of these food polymers is to stabilize a large amount of water. A knowledge of water holding capacity of hydrogels made from carbohydrate polymers is important in practical applications. Due to the intrinsic instability of hydrogels, water may be lost after extensive storage time as a result of passive diffusion (syneresis) (Sanderson, 1990). Water in gels may also be lost when the gels are subjected to external forces, or as a result of temperature fluctuation such as freeze-thawing. The loss of water may result in shrinking of the gels, changing texture and reducing quality. Therefore water holding capacity is an important criterion in evaluating the acceptability of food gels.

Syneresis is a common characteristic of polysaccharide gels (Glicksman, 1978). Starch is the most widely used polysaccharide in the food industry. Starch gels experience severe syneresis after prolonged storage (Osman, 1967). κ-carrageenan gels containing calcium ions are brittle and exhibit syneresis. When combined with locust bean gum, κ-carrageenan gels become more elastic and have a reduced degree of syneresis (Glicksman, 1978; Stanley, 1990). Agar gels normally have brittle and crunchy textures, and show extensive syneresis (Glicksman, 1978). The lower the gel strength, the more severe the syneresis of agar gels (Matsuhashi, 1990). Alginates with large proportions of polygulur-onate segments tend to form rigid, brittle gels that are also subject to syneresis, while alginates with predominantly polymannuronic acid units form gels that are more elastic, less brittle and do not exhibit much syneresis (Glicksman, 1978). In general, brittle polysaccharide gels are more susceptible to syneresis than elastic gels.

The stability of gellan gels during storage is not reported in the literature. Gellan is a novel polysaccharide gelling agent that can form both elastic and brittle gels, depending on cation concentrations. Effects of calcium concentrations on the failure stress which indicates the strength, and on the failure strain which indicates the brittleness of gellan gels, were studied by Tang, Lelievre, Tung and Zeng (1994) using compression, torsion and tension tests. The true failure shear stress determined by the three test modes were equivalent and the true failure shear strain by tensile testing was about half that by the other two modes. In all three test modes, two distinctive patterns of mechanical response were observed below and above a critical calcium concentration at which gellan gels exhibited peak strength.

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Gellan gels were soft and easily deformable below the critical calcium concentration, and brittle above. Tang, Tung and Zeng (1995) and Tang, Tung and Zeng (1996) observed that the critical concentrations of divalent cations (Ca^{++}, Mg^{++}) on a molar basis were approximately half of the concentration of carboxylate groups in gellan polymer chains, i.e. approximately at the stoichiometric equivalent point between cations and carboxylate groups. The critical cation concentrations observed in gellan gels formed in pH 3.5 and 5.0 citric buffers were not at stoichiometric equivalent point, which may be explained by different degrees of dissociation of carboxyl groups and different chelating abilities of citrate ions in buffers at different pHs (Mao, Tang & Swanson, 1999).

Although failure stress and strain are frequently used to characterize textural properties of food gels including gellan gels, the mechanisms involved in textural properties related to gel microstructure are not well understood. Tang, Tung and Zeng (1998) observed that the equilibrium stresses following stress-relaxation were correlated to gellan gel strength, and the stress-relaxation rate of gellan gels were correlated to pore-size of the gel network, indicating that water seepage through the polymer matrix was responsible for the stress relaxation. Therefore, textural properties of gellan gels are related to water holding capacity and the microstructure.

The relationship between water holding capacity and textural properties of gellan gels is not reported in the literature. Our objectives were to study the syneresis of gellan gels as affected by calcium concentrations and storage conditions, and to study the relationship among water holding capacity, microstructure and textural properties.

2. Materials and methods

2.1. Preparation of gellan gels for passive diffusion (syneresis) tests

Low acyl gellan gum powder (KELCOGEL F) was provided by Kelco Biopolymers Inc., (San Diego, CA). Composition was supplied by the manufacturer: carbohydrate (dietary fiber), 85%; water, 10%; Na, 0.3%; Ca, 0.4%; Mg, 0.1%; K, 4.9%; and P, 0.2%. One percent gellan powder was dispersed in deionized distilled water in a 400 ml beaker with a magnetic stirrer. The dispersions were heated from 96 to 98°C and the temperature maintained for 1 min to give clear solutions. Calcium chloride at twelve concentrations (2, 4, 6, 8, 14, 20, 30, 40, 50, 60, 70, 80 mM) was added to the hot gellan solutions. The solutions were stirred for 2–3 min. About 50 ml of each solution was transferred into a pre-weighed 100 ml glass beaker and sealed by SARAN wrap film. After the solutions were cooled to room temperature and the gels were firmly set, the beakers were stored in a 4°C incubator for 12 h. Water condensed on the beaker walls was removed with tissue paper and the beakers containing the gels were weighed with an analytical balance. The weight of gels at this stage was used as the initial weight (W_0) in the analysis of water holding capacity by passive diffusion tests. After weighing, the beakers were sealed and incubated at 4°C for further testing. At scheduled time intervals, water released from the gels was removed by tissue paper and gel weight (W) was determined. The weight ratio (W/W_0) was used to characterize the water holding capacity subjected to internal diffusion force. All measurements were conducted in duplicate.

2.2. Preparation of gellan gels for other tests

One percent gellan aqueous solution with 12 calcium concentrations between 2 and 80 mM were prepared by the same procedure as in the previous section. The hot solutions were stirred for 2–3 min and poured into stainless steel tubes (inner diameter 21 mm, length 190 mm) that were preheated to 80°C. The gels were set by cooling the tubes in running tap water at 15°C for 30 min, held for 1 day at room temperature (22°C) before being removed from the tubes for centrifugal dehydration tests, compression tests, or electronic microscopy observations.

2.3. Centrifugal dehydration

Small cylindrical gels (diameter 3.8 mm, length 10 mm) were placed in a Gelman z-spin centrifuge filter (Fisher Scientific) with 0.45 μ pores and centrifuged at 2000 rpm (268*g*) for scheduled time intervals between 5 and 120 min. The gel samples were weighed immediately after the centrifugation runs. Similar to passive diffusion tests, the weight ratio after and before centrifugation (W/W_0) was used to characterize the water holding capacity of the gels. All measurements were made in duplicate.

2.4. Cryo-scanning electron microscopy (Cryo-SEM)

Small sections of gellan gels were mounted in the cryoholder for the Emscope 2000 A cryo-preparation unit (Ashworth, Kent, UK), plunged into liquid propane, and then cooled further with liquid nitrogen. The frozen gel was freeze fractured to expose fresh surfaces, sublimed for 1 h at −80°C, and sputter coated with 30 nm of gold. The gold coated gels were observed and photographed with the Hitachi S-570 scanning electron microscope (Tokyo, Japan) at 10 kV and at temperatures below −135°C.

2.5. Compression tests

After being stored in sealed tubes at room temperature (22°C) for 1, 8, 15, and 60 days gellan gel specimens were compressed to study the effect of storage time on textural properties. Before the compression tests, the gels were removed from the tubes (21 mm inner diameter), cut into cylindrical specimens with length of 21 mm. The specimens were then placed between lubricated flat metal surfaces

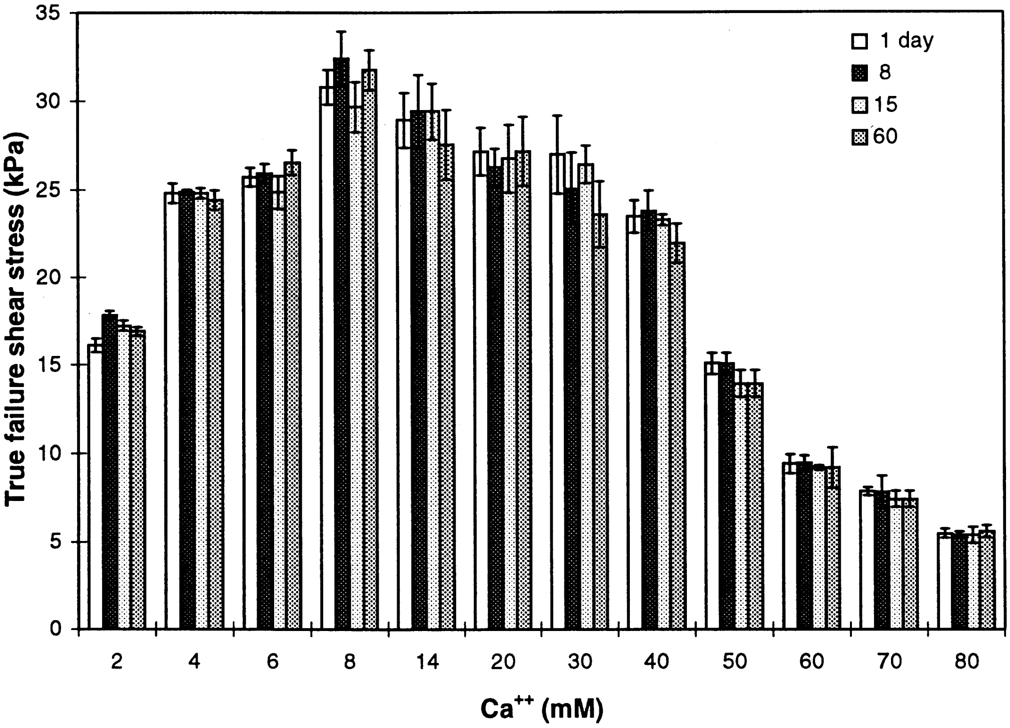

Fig. 1. Influence of storage time on failure stress of gellan gels with various calcium concentrations. The gels were stored at 22°C.

fitted to the TA.XT2 Texture Analyzer (Texture Technologies Corp., Scarsdale, NY/Stable Micro Systems, Godalming, Surrey, UK) with a 5 kg load cell and interfaced with Texture Expert software. The gels were deformed in compression at a constant cross-head speed of 0.3 mm/s until failure. Compression tests were repeated six times. The engineering stress (σ_{eng} = force/original cross-section area) and strain ($\varepsilon_{\mathrm{eng}}$ = reduction of length/original length) corresponding to the maximum force were recorded. Hencky's normal strain (ε_{h}) and corrected stress (σ_{c},

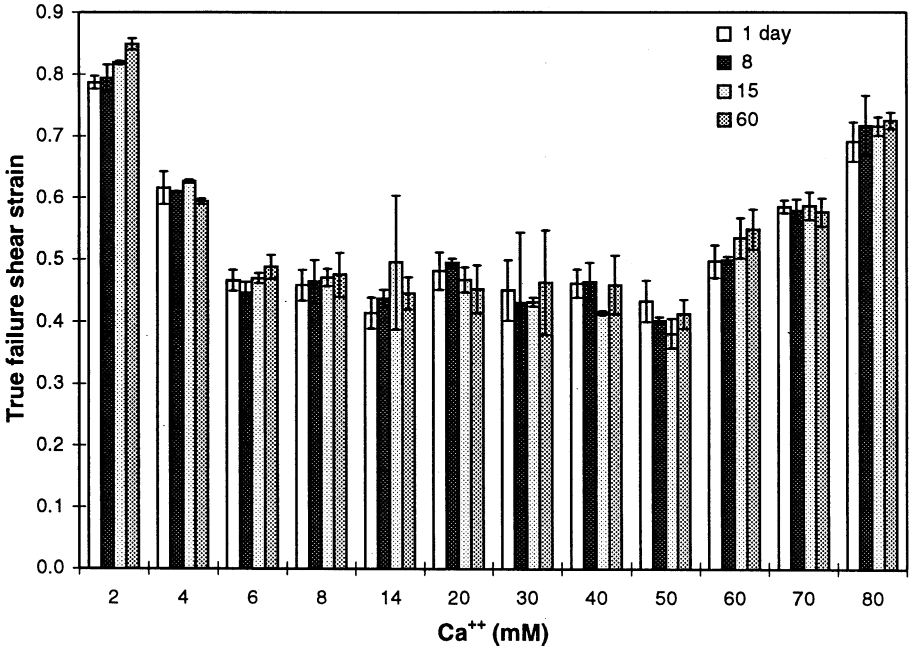

Fig. 2. Influence of storage time on failure strain of gellan gels with various calcium concentrations. The gels were stored at 22°C.

Table 1
Water holding capacity W/W_0 (%) of 1% gellan gels after 125 days storage at 4°C

Ca^{++} (mM)	Average	Standard deviation
2	98.72	0.30
4	98.94	0.02
6	99.10	0.35
8	98.73	0.37
14	99.20	0.05
20	98.91	0.12
30	99.20	0.15
40	99.23	0.13
50	98.34	0.34
60	98.95	0.11
70	99.15	0.06
80	98.95	0.37

corrected for the increase in cross-sectional area of the specimen at the time of failure) were calculated by the following equations (Tang et al., 1996):

$$\varepsilon_h = -\ln(1 - \varepsilon_{eng}) \qquad (1)$$

$$\sigma_c = \sigma_{eng}(1 - \varepsilon_{eng}) \qquad (2)$$

The true failure shear strain (γ) and shear stress (σ) were calculated from (Hamann, 1983):

$$\gamma = (1 + \nu)\epsilon_h \qquad (3)$$

$$\sigma = \frac{\sigma_c}{2} \qquad (4)$$

Gellan gels contained approximately 99% water and can be reasonably assumed to be incompressible under the test conditions. That is, Poisson's ratio (ν) in Eq. (3) is 0.5 (Bourne, 1982).

2.6. Statistical analyses

The ANOVA procedure of Microsoft Excel (Microsoft Corporation, 1995) was used to determine differences among water holding capacities and texture properties of gellan gels after storage. A significant level of $p < 0.05$ was chosen.

3. Results and discussion

3.1. Texture properties after storage

Storage time up to 60 days at 22°C did not significantly affect the texture properties of gellan gels ($p > 0.5$) (Figs. 1 and 2). The appearance of gellan gels after storage did not change either. Similar to gels without storage, a critical calcium concentration was observed in stored gels. The critical calcium level was about 8 mM, based on the trend that the failure stress changed with calcium concentration (Fig. 1), and about 6 mM for failure strain. The failure stress increased with increasing calcium concentration up to $Ca^{++} = 8$ mM, then decreased with further increasing Ca^{++}. The failure strain decreased with increasing calcium concentration to $Ca^{++} = 6$ mM, then remained fairly constant with further increasing Ca^{++}. At very high calcium concentrations, however, the failure strain tended to increase. These results are similar to the previous published results (Tang et al., 1994, 1995, 1996).

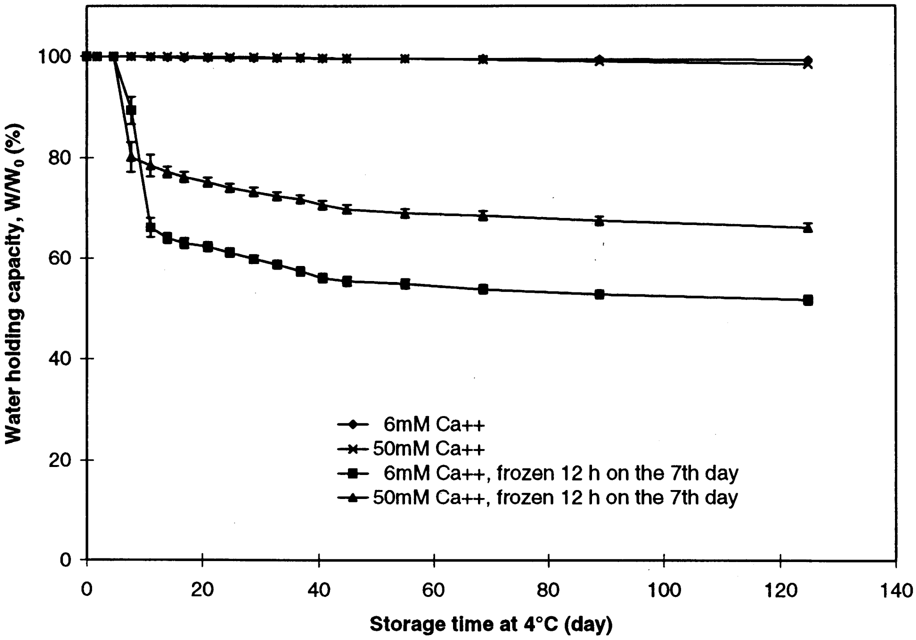

Fig. 3. Water holding capacity of 1% gellan gels with 6 and 50 mM Ca^{++}. The gels were stored at 4°C.

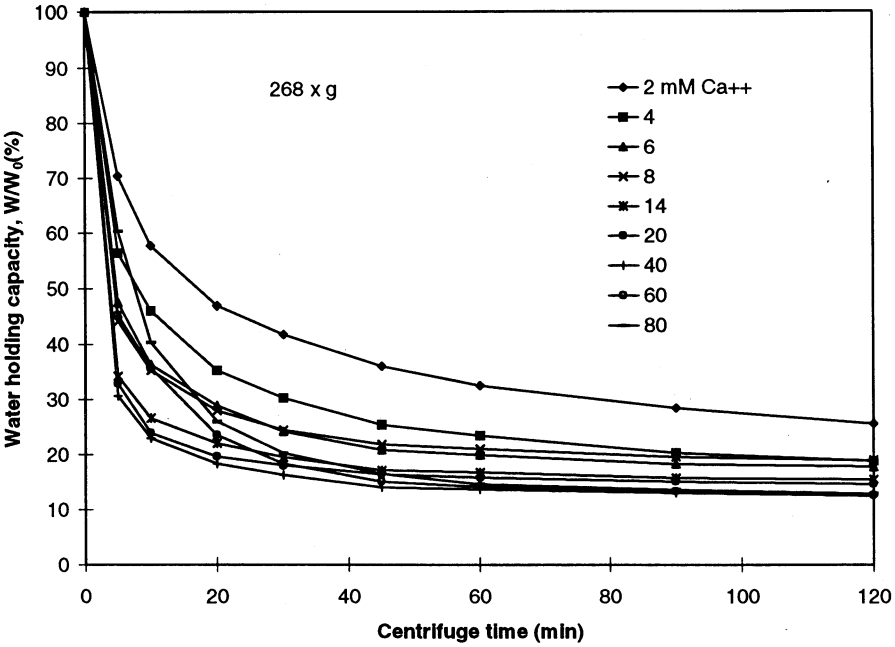

Fig. 4. Water holding capacity of 1% gellan gels subjected to 268*g* centrifuge force.

3.2. Dehydration by passive diffusion

Gellan gels studied in this work were very stable during storage at 4°C. The water loss during storage from passive diffusion (syneresis) was negligible. After four months storage at 4°C, the weight losses for gels were only 1–2% with standard deviation of 0.02–0.4% (Table 1). There was no significant difference in water losses among gels with different calcium concentrations ($p > 0.05$). Water holding capacity of gellan gels with 6 and 50 mM Ca^{++} are presented in Fig. 3. Both brittle and elastic gellan gels exhibited no sign of syneresis. This may be a unique advantage of gellan gels compared to other polysaccharide gels such as carrageenan and alginate gels. The stability of water holding capacity is consistent with the stable texture properties during the storage described in the previous section.

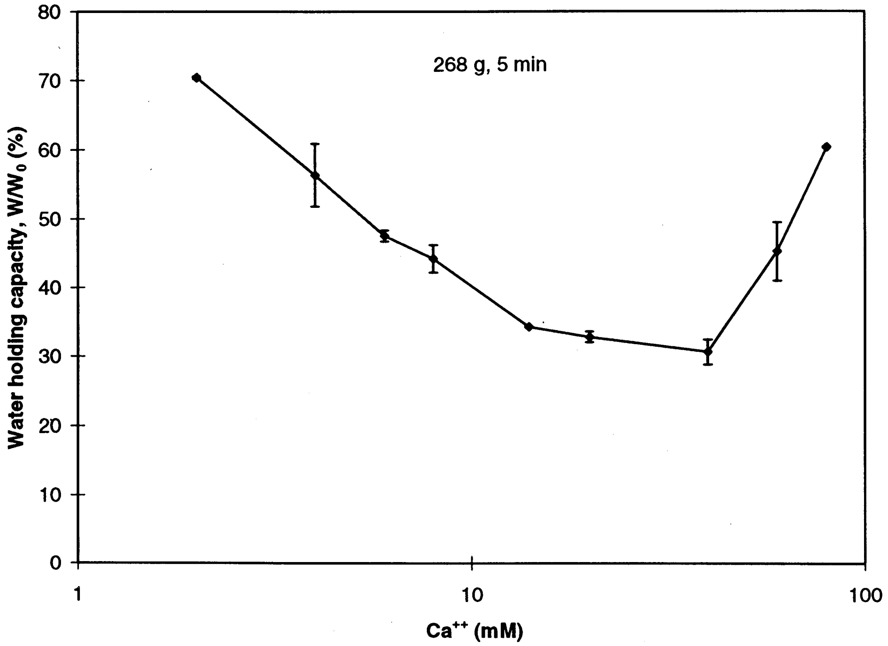

Fig. 5. Effect of calcium concentration on the water holding capacity of gellan gels subjected to centrifugation.

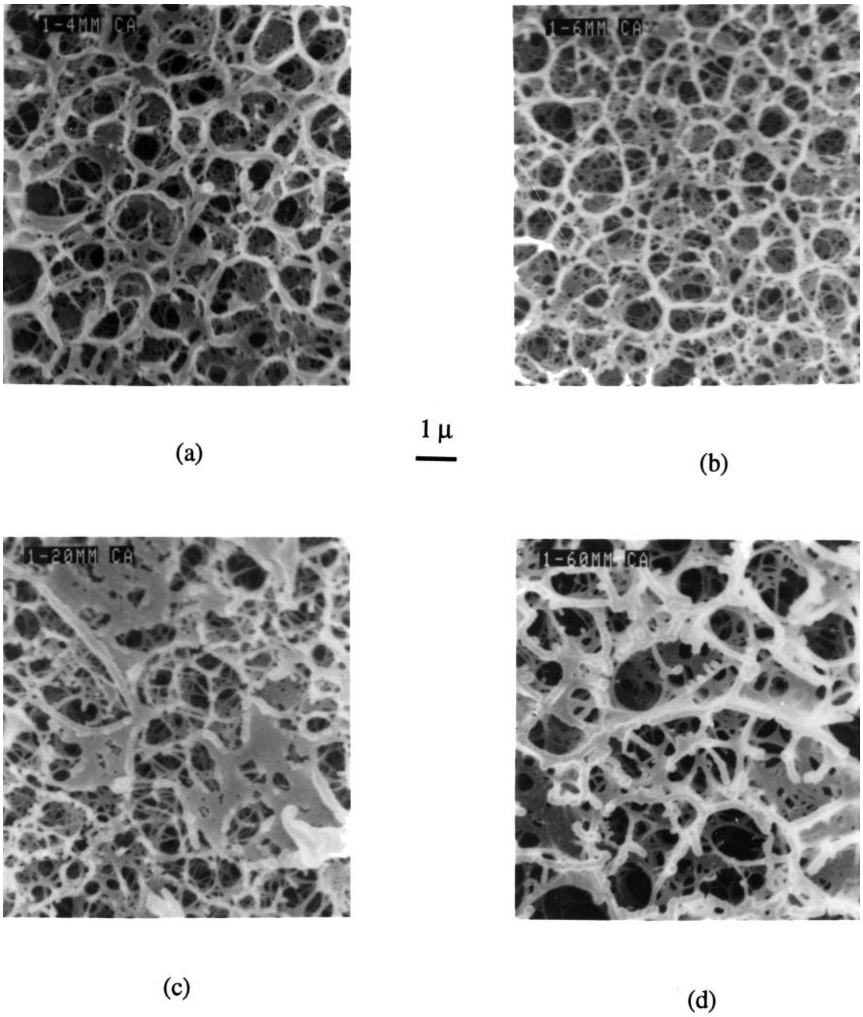

Fig. 6. SEM micrographs of 1% gellan gels with (a) 4, (b) 6, (c) 20, and (d) 60 mM Ca^{++}.

The freeze–thaw stability of gellan gels was poor at tested calcium concentrations. After frozen below 0°C for 12 h, the gels were warmed to 4°C. A large amount of water was lost during this thawing process (Fig. 3), though gels with 50 mM Ca^{++} demonstrated better water holding capacity than the gels with 6 mM Ca^{++}. Thawed gels continued to lose water when stored at 4°C. This suggests that formation of ice in gels during freezing adversely affected the microstructure of gellan gels.

3.3. Dehydration by centrifugation

When subjected to 268*g* centrifugal force, a significant amount of water was expelled quickly from gellan gels, and the gels shrank. Fig. 4 presents a rapid decrease of water in gellan gels with centrifugal time. After 10 min centrifugation, more than 50% of water was lost, except for the gel containing 2 mM Ca^{++}. The gellan gel with 2 mM Ca^{++} exhibited the highest water holding capacity. The water holding capacity (W/W_0) after 5 min centrifugation is presented in Fig. 5. Comparing Figs. 5 and 2 suggest that the effect of calcium concentration on the changes in water holding capacity when subjected to centrifugal force correlated with the effect of calcium concentration on the changes in failure strain. Larger water holding capacity corresponded to larger failure strain. It is possible that the internal structure of brittle gels is easier to be damaged than flexible gels. Thus, the water holding capacity of brittle gels is smaller when subjected to external forces.

3.4. Microstructure of gellan gels

Different water holding behaviors of gellan gels with and without external forces suggest that two types of microstructures may exist simultaneously within gellan gels. One microstructure is responsible for the long time stability of gellan gels, such as water holding capacity determined from passive diffusion tests, and the texture stability after long time storage. This structure is not dependent on calcium concentration. A second microstructure, which should be dependent on calcium concentration, is responsible for the gel behavior when the gels are subjected to external forces, such as water holding capacity measured from centrifugal dehydration tests, and the relationship of failure properties to gel composition. The co-existence of two types of microstructures was confirmed by SEM observations. SEM micrographs of gellan gels with 4, 6, 20, and 60 mM Ca^{++} are presented in Fig. 6. Two discrete pore-size distributions on the order of 0.1 and 1 μ were observed in gellan gels. Discrete pore-size distributions were also observed in a heat treated plant fibre (Hermansson, 1986). In gellan gels, the large pores were formed with thick strings of the gel network while the small pores were formed by a thin web structure. The minimum size of large pores was observed at gels with 6–8 mM Ca^{++} concentration, corresponding to the critical calcium concentration related to the maximum gel strength (Fig. 1) or the transition point between elastic and brittle gels (Fig. 2). Below the critical calcium concentration, the size of the large pores decreased with increasing Ca^{++} concentration. Above critical calcium concentration, the size of the large pores increased with increasing Ca^{++} concentration, and network defects of thick string net were observed. Therefore, the large pore structures with thick strings may be responsible for the stability of gel structure when subjected to external forces, and those thick string networks provided the gel strength. Due to much smaller pore-size, the thin web structure has much higher capillary forces to hold the water in gels than the pores formed by thick string net. The rate of water diffusion inside the gel was also reduced by the thin web structure. The size of the small pores in the thin web structure was not affected by calcium concentrations. This may be the reason that the water holding capacity of gellan gels after long time storage was not affected by calcium concentration. The small pore structure may also be responsible for the stability of the texture properties during storage.

4. Conclusions

Gellan gels with selected calcium concentrations exhibited a stable internal structure and can have a long shelf life under appropriate storage conditions. The water content and texture properties of gellan gels did not change significantly after several months storage. Freeze temperature storage, although common for many food products, must be avoided for gellan gels to prevent severe structural damages and loss of water. External forces may damage the gel structure and reduce the shelf life of gellan gels. Different gel microstructures are responsible for different behaviors of gellan gels with and without external forces. Composition dependent thick string net with large pores provides the gel strength, and determines the water holding capacity when the gels are subjected to external forces. Composition independent thin web structure with small pores determines the water holding capacity and the texture stability when gels are stored for long time but not subjected to external forces.

Acknowledgements

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