Chapter 4
Gelling Properties

Katsuyoshi Nishinari

Abstract Except beverages, soups, and dried foods, most foods are eaten in gel state, and thus it is important to understand the mechanisms of gel formation to improve the food quality. Starting from the definition and classification of gels, this chapter describes the molecular forces responsible for gel formation, how the network structure is formed, rheological determination of gel point, critical molar mass and concentration below which no gelation occurs. Then, characterization methods, gelation kinetics, mechanical spectra, and thermal scanning rheology are described based on studies performed for gelation of polysaccharides and proteins. Various factors influencing structure and properties of polysaccharide and protein gels, in particular, gelation rate, temperature, molar mass, concentration, interaction between long chains and short chains, small molecules such as sugars, acids, salts, and polyphenols are described. Release of molecular chains followed by erosion of gels induced by immersion in solvents is also described. Special categories of gels, microgels, mixed gels, cryogels, and oleogels are also described.

Keywords Definition · Classification · Transition · Polysaccharides · Proteins

1 Introduction

Gels are everywhere around us. In the breakfast, boiled or fried eggs are served. Tofu, yogurt or cheese is also served. They all are gels. Jams on the toast are also gels. There are many food gels, dessert jellies, pudding, marshmallow, etc. When water used to boil fish or meat is concentrated and cooled, a gel is formed. Though raw rice grains are not gels, cooked rice grains, and cooked pasta and noodles might be family members of gels. When dried foods with very low water content are masticated and crushed into small fragments, they are mixed with saliva forming a

K. Nishinari (✉)
Glyn O. Phillips Hydrocolloids Research Centre, School of Food and Biological Engineering, Hubei University of Technology, Wuhan, People's Republic of China
e-mail: katsuyoshi.nishinari@hbut.edu.cn

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bolus and then swallowed. Before swallowing, the solid foods are transformed to gels before forming a swallowable bolus. Our gastric juice is in acidic pH ca. 1–1.5. Why the stomach wall is not injured? It is protected by mucin gels. Acid-induced gelling property of alginate has been used for treating gastroesophageal reflux disorder. When some part of our body is injured, blood comes out and it clots (gels). When we catch a cold, sputum is formed. It is also a gel. They all are formed from a liquid which is solidified or a solid becomes a gel by absorbing liquid. Therefore, apart from two extremes, very dried foods and liquids, most semi-solid foods can be called gels although mainly polysaccharide and protein gels are discussed in this chapter. Some shampoos are called gels although they are not gels in a scientific word. It is necessary to define a gel.

2 What Is a Gel?

The definition of gels has been a problem discussed for a long time. Jordan Lloyd said "The colloidal condition, the 'gel', is one which it is easier to recognize than to define". In the review on structure of gels, she classified gels into (1) heat-reversible gels such as gelatin or agar in water, cellulose acetate in benzoyl alcohol and (2) heat-irreversible gels such as silica in water, colloid in chloroform and alcohol, many metallic sulfides and oxides in water. Since then, many definitions have been done for polymer gels (Djabourov et al. 2013). Recently, low molar mass gelator has been attracting attention in relation to pharmaceutical and cosmetic industry but not so much applications are found in food industry except oleogel. Gels formed from synthetic and biological polymers have many things in common and it is useful to discuss together comparing each other (Djabourov et al. 2013; Nishinari et al. 2016a; Tokita and Nishinari 2009) although the present chapter focuses mainly on polysaccharide and protein gels. Gels can be defined both from a rheological behavior and from a structural feature.

Rheological definition of a gel is that the system does not flow, and it can be characterized by the presence of a plateau region of storage modulus and the low $\tan\delta$ (<0.1) at an angular frequency range from 10^{-3} to 10^{2} rad/s, which is accessible by many commercially available rheometers. This should be called an operational definition and it cannot exclude the possibility to find the violation of this definition if a material obeying this definition shows a liquid-like rheological behavior at further lower frequencies. We should remember a famous saying of a prophetess Deborah "Even the mountains flow before the Lord" or everything flows. We could enjoy to visit the Website of a pitch drop experiment. A drop of the pitch flows (falls) every 9 years or 10 years or 11 years. It shows clearly that the distinction of a liquid and a solid is not simple, and depends on the "patience" of the observer. Since it is difficult to set the reasonable time-scale for both patient and impatient persons, it is more practical to clarify the fluidity by the *yield stress* concept (discussed in the previous Chap. 3) and the large deformation behavior above the yield stress. In this sense, a commonly used tube tilting method is not a good method because of its

dependence on the patience of the observer. Impatient observers will not see whether it flows after a long time under the gravity which does not induce flow during a short time (see the discussion on delayed failure in Chap. 3). If it flows above the yield stress, it is a structured liquid and should not be called a gel as mentioned below. If it is a gel, it will be broken down into separate parts and will not flow. If divided parts recover the initial continuity immediately on contact, such materials could be used in various areas in adhesive, coating, electric conductivity, and tissue engineering. These are called *self-healing* gels (Cordier et al. 2008; Halake and Lee 2017), and it is expected that the principle may be also applied in encapsulation of bioactives and endurable packaging materials in the future.

Structural definition of a gel is based on the connectivity of the system. Gel is a system consisting of molecules, particles, chains, or other structural elements which are partially connected to each other in a fluid medium by crosslinks to the macroscopic dimensions. According to this structural definition, the loss of fluidity is the result of connectivity. Entanglement of long object may be regarded as connected by delocalized crosslinks. The sol-gel transition can be treated by a percolation theory (Tokita 1989). This is also another operational definition, and it cannot exclude the possibility of finding gels whose constituents are not directly connected.

There are two material groups having a name "gel," but it can cause a confusion. These two are a weak gel and a fluid gel. Weak gel was named for structured liquids which show a weak frequency dependence for storage and loss modulus at a commonly accessible angular frequency range from 10^{-3} to 10^{2} rad/s, and the mechanical loss tangent is higher than 0.1. These materials are essentially liquid but it does not flow below its yield stress, thus it is apparently different from other liquids with no yield stress. Since yield stress has attracted further attention recently, it was discussed in the previous chapter for rheology. It may be better to mention here that it is not a good question to ask "which is more viscous, mayonnaise or whip cream and sugar syrup or honey?" The former has a yield stress and the latter not. In the present chapter, this is not called a weak gel but called structured liquid because it is essentially a liquid with non-zero yield stress.

The second class material, fluid gel is against the definition of gels mentioned above in the sense that it flows; fluid includes the meaning of flow. In this case, we should define the length scale of the "phase." Strictly speaking, the so-called fluid gels should be called microgels. Everybody knows that sands "flow." And there is an expression "flowing sands." Everybody also knows that a grain of sand is a solid although a mass of sands "flows." These granular materials like sands, powders, and other solid particles flow, but this flow is different from the liquid flow which has been studied in fluid dynamics. The flow of granular materials stops at the angle of repose of these granular solids when put on the horizontal plane while liquids with no yield stress (described in the previous chapter) flatten showing no angle of repose. Dried sands and powders don't stick each other, and therefore contradict with the connectivity in the above definition, but this situation can be modified by increasing moisture content or adding liquids. Such a material has, however, been called a paste rather than a gel. They are similar to slurries. But, it is difficult to make a clear

distinction between a gel and a paste. It may be impossible to define the highest and the lowest liquid content for gels. This is related to the following problem.

Emulsion gel and oleogel are different from food polymer gels consisting of polysaccharides and proteins. In fats such as butter or margarine, the solid fat content is an index to characterize the state of the material. This index decreases with increasing the temperature. The difference between the gel-sol transition and the melting of fat is that the latter is more crystalline than the former. What is the border of the crystallinity or the orderliness of the structure when we define the gel? There may be gels very close to crystalline state and also other gels very close to amorphous state. It may be impossible to define a borderline which depends on many factors, which is discussed later.

It is therefore difficult to propose the simple definitive definition of gels, and the present author has a similar feeling with his good friend, Nijenhuis who says *A gel is a gel, as long as one cannot prove that it is not a gel.* The difficulty arises when we define a border between a gel and non-gel as pointed above such as liquid content, and the degree of the order, and glass. If the border can be found as a discontinuous transition, it can be clearly defined, but if the border is not discontinuous and vague, it might be difficult to distinguish the materials on both sides, gel and non-gel. The above tentatively proposed definition of gels could be operationally practical, but the author will be happy if more persuasive definition is proposed because the science is the never-ending endeavor of human beings.

3 Classification of Food Gels

Gels can be classified from various points of views based on mechanical properties, molecular forces, constituting ingredients, temperature dependence of elastic modulus, transparence, electric charges, etc.

3.1 Classification of Food Gels Based on Mechanical Properties

When we hear that it is easier to recognize than to define a gel, we expect that a gel may wobble which is easy to recognize. The amplitude we observe is related to the elastic modulus (Nishinari 1976) or simply we can push a gel with a finger. Gels may not have the elastic modulus of common solids such as metals, stones, glass, or plastics which have the moduli $>10^{9\sim11}$ Pa. Moduli of most rubbers are of the order of 10^6 Pa. What is the maximum and the minimum moduli for gels? There is no consensus about this because there may be a very firm gel which is very close to solids, and a very tenuous or sloppy gel very close to liquids. The minimum modulus

necessary for a gel to be self-standing is ca10^3 Pa, and this and yield stress are recognized important in 3D printing (Nan et al. 2020).

Beyond the small deformation range, gels show a failure or fracture. Gels close to liquids show failure but not clear fracture. The strain at failure or fracture is an important property in the application of gels. Generally, gels consisting of long chains show a deformable behavior while gels consisting of short chains or small molecules show a failure at small strain and are brittle. Obviously, textures of deformable gels and brittle gels are very different (Nishinari 2000a).

Soft gels are not chewed by teeth but compressed/squeezed between the tongue and the hard palate. The transition of the mastication mode from the tongue-palate squeezing to the chewing by teeth could be well predicted by a simulating compression experiment of a gel between an artificial tongue and the base plate of the uniaxial compression machine, and depended on the deformability of gels (Ishihara et al. 2013, 2014).

3.2 Classification of Food Gels Based on Molecular Forces

Molecular forces which form gel network are covalent and non-covalent bonds such as hydrogen bonds, hydrophobic interactions, ionic bonds. Gels formed by non-covalent weak secondary forces such as hydrogen bonds, hydrophobic interactions, van der Waals forces are called *physical gels*, while gels formed by covalent bonds are called *chemical gels*. In many food protein gels, physical and chemical bonds coexist (Djabourov et al. 2013).

It seems that it is still not possible to understand the structure-property relation of all different gels in common language which is aimed in this chapter. Researchers studying polysaccharide gels consisting of linear chain molecules use the network theory derived from rubber elasticity and the ordering process is related with coil-helix transition and aggregation processes while globular protein gels are studied from the viewpoint of molecular rearrangement (Nishinari and Takahashi 2003; Nishinari et al. 2000). Gelatin is a fibrous molecule and its gelation has been studied extensively, but in food area, particle gels formed from globular proteins are another important material. Helix-coil transition is also a molecular rearrangement, but this term is not so much used among researchers interested in polysaccharide gels or gelatin gels which are jokingly called an honorary polysaccharide. Both of these gels are common in a sense that structural elements are connected or in modern parlance, after de Gennes' introduction, "percolated" to span the space. The term percolation originates from a coffee percolator (Djabourov et al. 2013).

3.3 Classification of Food Gels Based on Molar Mass of Network Elements

Most food gels are made from polysaccharides or proteins. When they are degraded into lower mass compounds, they cease to form a gel. Everybody knows that sucrose solutions don't form a gel even if they are concentrated. They precipitate above a certain concentration at each temperature. An important difference between lower molar mass compounds such as sugar or salt and higher molar mass compounds (polymer) is that the former solutions precipitate when the temperature or pressure or concentration is changed so that the extrinsic condition is outside of the condition of saturated solution while the latter materials have plural conformations such as helix and coil and their aggregates which lead to network structure called commonly gels. Precipitated lower molar mass materials such as butter or margarine are usually not called gels. However, lower molar mass gelators (Weiss and Terech 2006) have been attracting much attention and especially oleogels in food area (Singh et al. 2017). Pullulan is one of most soluble polysaccharides and its films are used in food packaging, but its gels are not studied in gel technology so much, because materials called gelling agents form a gel at lower concentrations.

3.4 Classification of Food Gels Based on Ingredients

In foods, many polysaccharide gels and protein gels have been known, and gels consisting of mixed polysaccharides and proteins have also been studied. Emulsion gels which consist of polysaccharide or protein gels containing oil droplet have been attracting much attention in the past two decades. Aerated gels have been also studied recently because it can reduce the calorie inducing satiety thus can be a convenient tool to prevent the obesity. Recently, gelation of oils has attracted much attention, and is described later.

3.5 Classification of Food Gels Based on Origin

Gels can be also classified by the origin of materials, plant or animals, sea weed, seed, bean, legume, fish, meat, egg, milk, and microbial origin such as curdlan, gellan, and so on. This viewpoint is important in designing food products (Phillips and Williams 2020).

3.6 Classification of Food Gels Based on Temperature Dependence of Elastic Modulus

Some biopolymer solutions of agar, carrageenan, gellan, and gelatin form a gel on cooling, but egg white, milk proteins, solutions of methylcellulose and other cellulose derivatives, and curdlan form a gel on heating (Nishinari 2000b; Nishinari and Zhang 2004). They are often called cold-set gels and heat-set gels, respectively. Some of them are thermoreversible and the others are thermo-irreversible; gels of agar, carrageenan, gellan, gelatin are formed on cooling, return to sol state on heating, thus called thermoreversible gels, while boiled egg white or tofu or konjac is a typical thermo-irreversible gel because they do not show gel-sol transition after gel formation. However, some thermoreversible gelation of ovalbumin (OVA), one of the main proteins in egg white, is well-known and studied extensively (Doi 1993).

3.7 Classification of Food Gels Based on Optical Properties

Gels of gellan and gelatin are transparent while egg white gels, starch gels, and many other gels are opaque. But it is well established to make a transparent egg white protein gel (Doi 1993). Most pigments are added artificially but some gels have a color in gels where naturally binding pigments exist.

3.8 Classification of Food Gels Based on Shape of Network Elements or Crystallinity

Fibrous gel, particle gels, or mixture of these may exist, since rearrangements or ageing always occurs and these may be related to syneresis. Partially ordered structured gels or liquid crystalline gels also exist. Even in apparently homogeneous gels, an internal structure consists of ordered domains and amorphous domains. It may be impossible to define the upper and lower limit of the degree of order.

3.9 Classification of Food Gels Based on Electric Charges

Most gelling polysaccharides have anionic groups such as carboxyl group (gellan (Morris et al. 2012)) or sulfate group (carrageenan) while chitosan molecule has a positive charge. Gels of agarose, curdlan, and starch are electrically neutral without electric charges although most commercially available samples contain some ions. Since amino acids are positively or negatively charged, gelling behavior of proteins is strongly influenced by pH and ionic strength.

3.10 Classification Based on Other Criteria

It is also possible to classify gels based on size (microgels and bulk gel), pH dependence, water holding capacity, thermal conductivity, liquid (water, oil, alcohol), or gas content. This viewpoint is useful in food processing and preservation. Macroscopic shape such as spherical, rod, tetrahedron, rectangular can be controlled if it is moldable, but these geometrically regular shaped gels may not be naturally occurring gels. It is an important problem to keep a shape in food processing and flavor release.

4 Gel-Sol Transition

4.1 Molecular Forces

Suzuki et al. (1972) examined whether the pressure would prompt the gel formation in which hydrogen bonds play a main role, while the pressure would retard it in which hydrophobic interactions play a main role. Investigating the dependence of equilibrium constant K in the gel-sol phase transition on the temperature and pressure $(\partial 1\mathrm{nK}/\partial T)_{\mathrm{p}} = \Delta H/\mathrm{R}T^2$ and $(\partial 1\mathrm{nK}/\partial P)_{\mathrm{T}} = -\Delta V/\mathrm{R}T$, respectively, they studied the effect of pressure and temperature on the gelation of gelatin and methylcellulose (MC). They found that the pressure prompted the gel formation of gelatin, while the pressure retarded that of MC, and therefore, it is concluded that hydrogen bonds and hydrophobic interactions are dominating molecular forces for gelatin and MC, respectively. However, they noticed that ΔV changes the sign above 6000 atm, and stated that both hydrophobic interaction and hydrogen bonds concern in the gelation of MC.

Kometani et al. (2015) studied the pressure effect on the gelation of agarose and MC (Fig. 4.1). The cloud-point temperature, T_{cloud}, was determined from cooling curves by the temperature of 75% transmittance because it corresponds to the steepest point of decreasing transmittance in all curves. In the same way, the "transparency temperature," T_{trans}, at which the solution returns to a transparent sol state was determined from heating curves by the temperature of 75% transmittance. From the Clapeyron-Clausius relation $\mathrm{d}T/\mathrm{d}P = \mathrm{T}\Delta V/\Delta H$ together with the enthalpy change determined by thermal measurements, the volume change for agarose was determined as $\Delta V = -4.57 \times 10^{-6}$ m^3/kg which is an order of magnitude larger than that for gelatin (Fig. 4.1a). This suggests that the gels of agarose and gelatin are stabilized under high pressure, but their stabilization mechanism may be different. In addition, it should be mentioned that both κ- and ι-carrageenan gels formed also by hydrogen bonds are destabilized by pressure (Gekko and Kasuya 1985).

The slope $\mathrm{d}T/\mathrm{d}P = 2.97 \times 10^{-2}$ K/MPa determined from Fig. 4.1b for MC is consistent with the reported values $\Delta V = 0.80 \times 10^{-6}$ m^3/kg and $\Delta H = 11.8$ kJ/kg

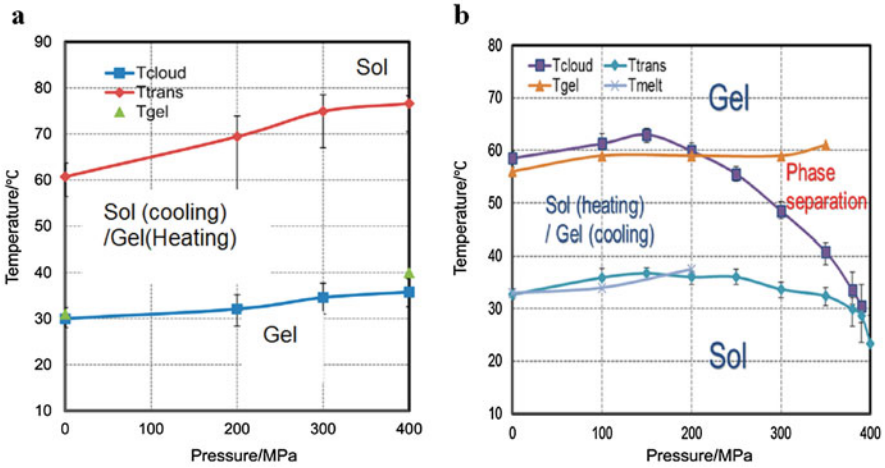

Fig. 4.1 Plots of T_{cloud} (square) and T_{trans} (diamond) as a function of pressure for 1 wt % solutions of agarose (Fig. 4.1a) and methylcellulose (Fig. 4.1b). Triangles and cross show the gelling temperature T_{gel} and melting temperature T_{melt}, respectively, measured by the falling-ball method. Reproduced with permission from Kometani et al. (2015), Copyright 2015ACS

taken from literatures, and indicates that MC gel is destabilized by compression as contrasted with agarose (Kometani et al. 2015).

The main molecular forces responsible for gel formation in agarose and MC are hydrogen bonds and hydrophobic interaction, respectively, and both appear as an endothermic peak on heating in DSC. In the former, gel-to-sol transition occurs, while in the latter sol-to-gel transition occurs. However, in the latter, at some intermediate concentration range, an exothermic peak appears on heating which is attributed to the appearance of anisotropic phase (Yin et al. 2006). This anisotropic phase formation at higher concentration for stiff molecules is also known for xanthan (Lee and Brant 2002), gellan (Nitta et al. 2010), schizophyllan (Fang et al. 2004), and other biopolymers (Djabourov et al. 2013).

Shirakawa et al. (1998a, b) found that xyloglucan from which galactose residues were removed formed a gel on heating and returned into sol state on further heating and that this transition was thermally reversible (Fig. 4.2). Enthalpy change ΔH accompanying gel to sol transition on heating was reported 24.2 J/g for gelatin (Gekko et al. 1992), 33.2 J/g for κ-carrageenan (Watase and Nishinari 1987a), 40 J/g for agarose (Watase and Nishinari 1987b) while ΔH accompanying sol to gel transition on heating is 16.0 J/g (Haque and Morris 1993) or 10–17 J/g (Funami et al. 2007) for methylcellulose, 6.9 J/g for methylhydroxypropylcellulose (Yuguchi et al. 1995). ΔH found for the degalactosylated xyloglucan was 4.4 J/g, which is the same order of magnitude to the latter group where hydrophobic interaction plays a main role. The gelation of xyloglucan by galactose removal is still studied recently (Sakakibara et al. 2017).

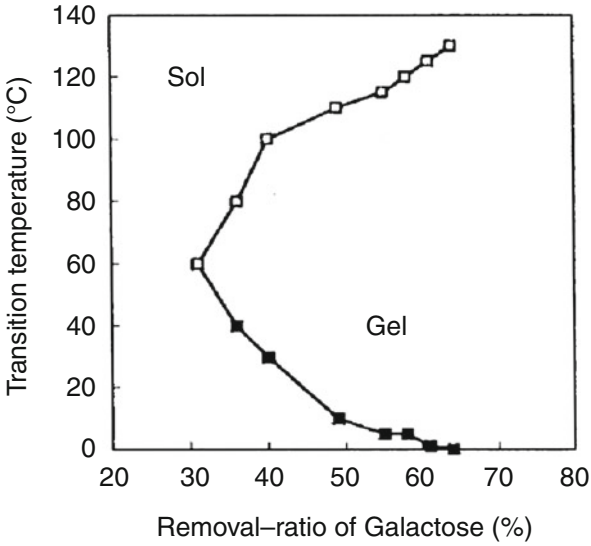

Fig. 4.2 Sol-gel transition temperature for xyloglucan as a function of galactose removal ratio. ■ lower temperature transition point; □ higher temperature transition point. Reproduced with permission from Shirakawa et al. (1998a), Copyright 1998 Elsevier

As is seen in the gelation of MC and xyloglucan, the chemical modification changes the solubility or gel forming ability. Gel forming ability is the intermediate between the insolubility and solubility. Carboxymethylation of curdlan confers curdlan water solubility but makes lose its gelling ability (Jin et al. 2006; Nishinari and Zhang 2004).

On the other hand, molecular forces in the gelation of globular protein gels are more complicated (Djabourov et al. 2013; Walstra 2003).

4.2 Rheological Determination of Sol-Gel Transition

One of the most widely used methods for the determination of sol-gel transition is that proposed by Winter and Chambon (1986). These authors examined the gelation process of polydimethylsiloxane (PDMS) to determine the gel point. PDMS was chosen for its well-defined chemical nature, the need of catalysis, the availability of prepolymers with different molecular weights, and the elastomeric nature of the samples after crosslinking. The advantage of this method was to stop the catalyst action instantaneously to prevent the further crosslinking reaction. Since the measurement of the frequency dependence of G' and G'' takes a time, it was examined before and after the crossover of G' and G'', which was possible by stopping the crosslinking reaction by stopping the catalytic action by stopping agent (sulfur). By this method, they could observe the storage modulus G' and loss modulus G'' as a function of frequency near the crossover point of G' and G'' observed as a function of time at a constant frequency. Before the gelation point GP, $G' < G''$ and after GP, $G' > G''$. The frequency dependence of G' and G'' was observed from 6 min before

the time of crossover of G' and G'', t_0, to the time after 6 min of t_0. At an earlier stage of the crosslinking reaction, both G' and G'' are strongly frequency dependent and $G' < G''$, and at time t_0, both G' and G'' show a similar frequency dependence, and after the time t_0, G' tends to show a plateau which is a characteristic mechanical behavior for rubbers. They proposed the following criterion for the critical gelation point.

$$G' \sim G'' \sim \omega^n \tag{1}$$

$$\tan\delta = G''/G' = \tan(n\pi/2) \tag{2}$$

which is now called Winter-Chambon criterion. At the critical point, both of these two equations should be satisfied simultaneously. The exponent n in the two equations should be the same. These equations are valid at times longer than a certain characteristic time t_0 or at lower frequencies than $1/t_0$. First, this criterion was proposed for such a chemical gel, but later, Nijenhuis and Winter (1989) found that this criterion is valid also for physical gels of polyvinyl chloride (PVC). Since then many papers have been published to study the critical gelation point, and although initially the exponent was proposed as 0.5, other values between 0.3 and 0.7 have been reported for various gelation processes (Winter 2016). Figure 4.3a shows the mechanical spectra, i.e., the frequency dependence of G' and G'' of a 2.5 wt %

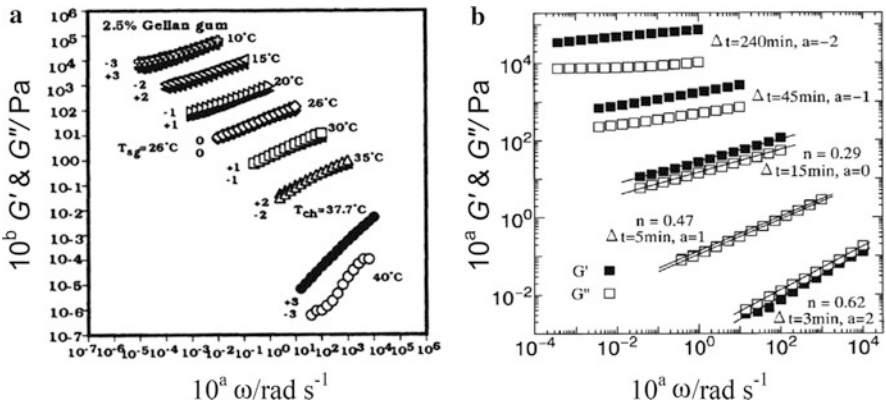

Fig. 4.3 (**a**) Angular frequency dependence of G' (open symbols) and G'' (filled) for a 2.5% gellan gum solution at various temperatures shown on the right side of each curve. The data are shifted along both the horizontal and the vertical axes by shift factors "a" and "b," respectively, to avoid overlapping. Numbers on the left side of each curves represent "a" and "b," respectively. Reproduced with permission from Miyoshi and Nishinari (1999), Copyright 1999 Springer. (**b**) Angular frequency dependence of G' (filled symbols) and G'' (open) for a 50mg cm^{-3} OVA solutions heated at 80 °C for 60 min and then reheated after addition of 60 mM NaCl for various times Δt =3, 5, 15, 45, and 240 min. The data are shifted along both the horizontal and the vertical axes by shift factor "a" to avoid overlapping. Reproduced with permission from Koike et al. (1998), Copyright 1998 Elsevier

sodium type gellan solution at temperatures from 40 °C to 10 °C (Miyoshi and Nishinari 1999). At higher temperatures, G' and G'' of the solution shows strong frequency dependence and $G' < G''$ while at low temperatures $G' > G''$ and both moduli tend to show a plateau. At 26 °C, the solution satisfies the above equations, and is in the critical state.

Figure 4.3b shows the mechanical spectra of G' and G'' of a 50 mg cm^{-3} OVA solution prepared by a two-step heating method; first heating at 80 °C for 60 min and then cooled, and then reheated at the same temperature for various heating time periods Δt (Koike et al. 1998). Critical gels satisfying the eqs. (1) and (2) were observed for Δt from 3 to 30 min indicating the fractal structure persisted in this range of the heating time. Koike et al. (1998) had reported that OVA formed linear aggregates, worm-like cylinder, diameter 12 nm, persistence length 23 nm, and the molar mass per contour length 16,000 nm^{-1} after heating the solution at 80 °C by dynamic laser scattering (Nemoto et al. 1993) (See Fig. 4.13). Transparent gels were formed by both one-step and two-step heating in a certain condition of the salt concentration and heating time, and these gels were found to satisfy the critical condition eqs. (1) and (2). However, such a critical gel behavior was not observed for translucent gels. Although these authors ascribed the main molecular forces for the gel formation are hydrophobic interaction, they found the necessity of further structural study based on neutron scattering and SEM, and this is discussed again in Sect. 7.

Hossain et al. (1997) studied the gelation of iota-carrageenan solutions, and found that $G' = G''$ at 57°C and n =0.42. The value of n was found to decrease with increasing concentration of iota- carrageenan. The crossover of G' and G'' as a function of time or temperature has often been taken as an indication for the gelation point, but this is only right for a critical gel with the relaxation exponent $n = 0.5$. Very small values 0.1 for the relaxation exponent were reported for bacterial poly-ester, poly(β -hydroxyoctanoate), and gelatin. The critical gelation point at which G' and G'' show the same frequency dependence may lie in between the entangled solution behavior and a weak gel behavior described before. To determine the gelation point by Winter-Chambon criterion, it is convenient to plot the tan δ observed at different frequencies as a function of time.

Figure 4.4 shows gelation process of carboxymethyl cellulose crosslinked with polyfunctional glycidyl ether.

However, this method of determination of gel point was found not applicable in some systems. In the gelation of pluronic (called also poloxamer, PEO-PPO-PEO triblock copolymer, where PEO = poly(ethylene oxide), PPO = poly(propylene oxide)) used in drug delivery, it was found that at lower temperatures (33 °C) a liquid-like behavior $G'<G''$ was found, on further heating gel is formed at >34 °C, however, at higher temperatures it was found that $G'>G''$ at high frequencies and $G'<G''$ at low frequencies, which is characteristic for entangled polymer solutions (Nyström and Walderhaug 1996).

Globular protein solutions show mechanical spectra similar to those of structured liquids such as xanthan solutions, that is, $G'>G''$ at the angular frequency range from 10^{-1} to 10^{2} rad s^{-1} and tan $\delta \geq 0.1$ (Ikeda and Nishinari 2000; Ikeda and Nishinari

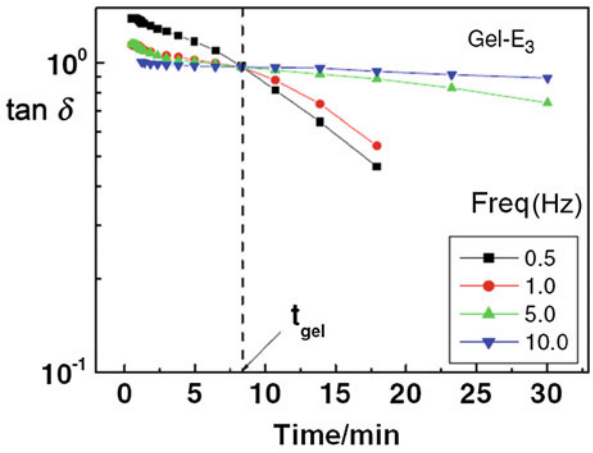

Fig. 4.4 Loss tangent tan δ as a function of time for the hydrogels at various fixed frequencies (0.5, 1, 5, 10 Hz) measured at constant temperature 60 °C. t_{gel} is the gel point. Reproduced with permission from Lawal et al. (2011), Copyright 2011 Springer

2001a, b; Matsumoto and Inoue 1993). During isothermal heating at 70°C, 5% w/w β-lactoglobulin in a 0.1 mol/dm^3 NaCl aqueous solution shows a gelation behavior (Ikeda and Nishinari 2001a). It was observed that $G' > G''$ all the time, and the gelation occurred at around 3000 s, which was also confirmed by tilting tube. Thus, the Winter-Chambon criterion cannot be used. For such a case, the temperature or the time at which G' begins to increase steeply is generally taken as the gelation point (Tobitani and Ross-Murphy 1997).

4.3 *Spinodal Decomposition or Nucleation and Growth?*

Gelation of agarose was thought to progress through spinodal transition based on Cahn's theory (Feke and Prins 1974). San Biagio et al. (1996a) recognized the characteristic or "signature" behavior of spinodal decomposition in the gelation of agarose (concentration $< 1\%$); the occurrence of a low-angle scattering ring, the exponential increase of scattered light, its typical dependence upon the scattering vector represented by linear Cahn's plot, the occurrence of a transient viscosity peak. Summarizing their data, they constructed a *C-T* phase diagram according to which at higher concentrations ($>1.5\%$), only direct gelation occurs at 60 ± 10 °C, and below this temperature range both demixing mediated and direct gelation are thought to be possible. They conclude that this approach is useful to understand the gelation beginning from the break of symmetry, the formation of inhomogeneous structure from a homogeneous sol state through the spinodal decomposition, and the possibility to control the final gel structure. San Biagio et al. (1996b) studied the gelation of bovine serum albumin (BSA) by the same approach and found the occurrence of spinodal demixing of sol after the unfolding of the native BSA.

Morita et al. (2013) constructed a *C-T* phase diagram of agarose, and found that an agarose solution formed a gel on cooling and then the phase separation occurred, which was detected as a cloud point. The spinodal points are found below these

temperatures. They also recognize characteristic features of spinodal decomposition mentioned above. They attributed the opacity of agarose gels to the frozen concentration fluctuation within the gel already formed. They pointed out that the sol-gel transition and the phase separation are independent phenomena, and showed a porous structure of a quenched gel. They finally emphasize the importance to take into account this porous structure induced by spinodal decomposition in addition to the formation of the crosslinking points (junction zones) to understand the macroscopic properties of gels.

MC gelation has been studied by many research groups. Group of Lodge (Arvidson et al. 2013) studied gelation and phase separation of MC with three different Mw using Winter-Chambon criteria and light transmittance. The gelation point was determined by frequency independent tan δ for concentrated solutions ($C \geq 10C^*$, where C^* is the coil-overlap concentration). They found a high correlation between the T_{gel} and the cloud point which is different from previous papers reporting the phase separation and the gelation are distinct events. In addition, they showed that gelation of MC has strong dependence on heating rate while the melting of the gel has little dependence on cooling rate, and thus suggested that thermogelation of MC proceeded by a nucleation and growth mechanism (McAllister et al. 2015) rather than spinodal decomposition although they cited several papers which propose the different mechanism that the gelation of MC is induced by spinodal decomposition.

4.4 *Jamming Transition: Another Molecular Rearrangement Induced by Shear*

In relation to shear thickening of suspensions, shear induced close packing formation has been attracting much attention. Jamming transition is functionally defined to occur when, with increasing packing fraction or decreasing applied shear stress, a yield stress is first observed (Liu and Nagel 1998). Tanaka (2011) classifies jamming gels with delocalized crosslinks as viscoelastic fluids with long relaxation times.

The minimum packing fraction where a yield stress is observed depends on a number of suspension properties, including shape, polydispersity, friction coefficient, roughness, density matching, temperature, and attractive forces (Brown and Jaeger 2014).

4.5 *Zippering and the Size of Junction Zone*

To correlate the microscopic states of a macroscopic system with thermodynamic state variables such as the temperature, volume, and pressure, it is necessary to determine the partition function in the statistical mechanics. A simple **zipper model**

for thermoreversible gels consists of aggregated ordered structures such as helices and stiff chains which can be opened from both ends (Nishinari et al. 1990). From the partition function including the number of parallel links N, the number of zippers **N**, binding energy ε and the degeneracy (rotational freedom) g, the heat capacity C can be calculated.

In the heating DSC measurements, dT/dt is positive, then the endothermic peak is equivalent to the maximum of the heat capacity C. In the cooling DSC measurements, dT/dt is negative, then the exothermic peak is again equivalent to the maximum of C. When the concentration of gels increases, the mobility of chain molecules decreases, and then the degeneracy g will decrease. As a result, the peak of the heat capacity shifts to higher temperatures. This is in accordance with Eldridge-Ferry's empirical formula. This zipper model approach can also explain that the transition is sharper in cooling than in heating as has been observed for many polysaccharide gels such as agarose, κ-carrageenan, and gellan (Watase and Nishinari 1987b; Miyoshi and Nishinari 1999).

A single DSC endothermic peak for a κ-carrageenan gel on heating was successfully fitted by choosing appropriate structural parameters. This treatment was extended to multi-component systems in which junction zones consist of associations with different kinds of zippers. A broader DSC endothermic peak for 0.42% κ-carrageenan /0.186% konjac glucomannan gel was thus fitted (Nishinari et al. 1990).

Using van't Hoff's law, Eldridge and Ferry (1954) proposed a method to determine the heat ΔH_m absorbed on forming a mole of crosslinks—$d\ln C / dT_m = \Delta H_m / RT_m^2$, where R is the gas constant, T_m (gel melting/K) is the transition temperature, and C is the gel concentration. Integration of this equation leads to $\ln C = \Delta H_m / RT_m$ + const. This EF equation is based on the following chemical equilibrium: *2* moles crosslinking loci → 1 mole crosslinks.

Tanaka and Nishinari (1996) modified the EF equation taking into account the molar mass dependence and proposed a method to estimate the bonding energy ε (i.e., the enthalpy change for binding a single repeat unit into the network junction), the number ζ of repeating unit in the molecular chain constituting a crosslink, and the junction multiplicity s (the number of polymer chains combined in a single junction) (Nishinari et al. 1996). It is expected that this method is applied for food polymers although it is not so easy to get food polymers with different molar masses (Nishinari and Fang 2021).

4.6 Critical Molar Mass and Concentration for Gelation

For thermoreversible gels, the term "junction zone" has been frequently used to describe the crosslink because each crosslink involves aggregates of ordered molecular chains like helices or extended stiff chains. It is required to know the lower limit of the molar mass below which a helix is not formed for each polymers. The minimum concentration necessary for gelation might be related to a persistent length of the chain. It has never been found that gels are formed in a dilute solution of flexible polymers such as polyethylene oxide or pullulan which only entangle each

other but don't form junction zones. Such a system cannot retain water solvent, and the system flows if the concentration is not high enough. Koga and Tanaka reported that the critical concentration for gelation shifted to lower concentrations with increasing persistence length of linear chain molecules by Monte Carlo simulation. Solutions of flexible chains, like pullulan, polyethylene oxide, don't form a gel even at quite a high concentration. However, since it is known that these polymer solutions produce a film when the solvent water is evaporated they should form a gel before becoming a solid film. It is evident that solutions of monomers of these polymers, glucose, or ethylene glycol do not form a gel even if solvents are evaporated. These substances with low molar mass are known to form a crystal when the solution is concentrated. Therefore, there should be a critical molar mass of polymers below which no gelation occurs.

Ogawa et al. (2006) prepared 6 gellan gum samples with different molar masses converted to sodium form and examined the helix-coil transition. The weight average molar mass for higher molar mass samples at 25 °C ($M_{w,25}$) determined by DLS was two times higher than that determined at 40 °C ($M_{w,40}$), indicating the double helix formation while the ratio $M_{w,25}/M_{w,40}$ is becoming smaller with decreasing molar mass, and was 1 for the lowest molar mass sample (Mw = 17,000) indicating that the lowest molar mass sample does not form a double helix. This is consistent with results obtained by circular dichroism and DSC. The minimum molar mass for gelation of pectin, κ-carrageenan, and alginate has been discussed recently (Nishinari and Fang 2021).

The critical gelation concentration has been determined by scaling in which the shear modulus G' near the gelation threshold is plotted against the distance of the concentration from the critical concentration: $G' \sim (C - C_{cr})^t$. The critical exponent $t \approx 2$ has been reported by many workers (Djabourov et al. 2013). Joly-Duhamel et al. (2002) reported that the G' of gelatin gels could be represented well by the helix concentration for all the gelatin extracted from various fish although gelatin extracted from warm water fish begins to form a gel at a higher temperature than that from cold water fish.

Concentration dependence of the elastic modulus of gels has been studied for a long time, and it has been known empirically that the modulus is represented by a power law. The exponent for agarose has been known larger ~4 at lower concentrations and smaller ~2 at higher concentrations. A cascade treatment and a modified rubber elasticity theory were applied to fit the concentration dependence of moduli for agarose, amylose, pectin, κ-carrageenan, etc. Both these theories predict that the exponent is larger at lower concentration region and smaller at higher concentration region. However, an opposite tendency was found in heated ovalbumin gels: the exponent is about 4 at lower concentration region and about 10 at higher concentration region (Koike et al. 1996).

Generally, the minimum concentration required for gelation is found lower for linear polymers but gelatin needs higher concentration than agarose or gellan (Fig. 4.5). This is not a rigorous statement because it may be possible to make the opposite situation by preparing gels with short chain gellan samples which need

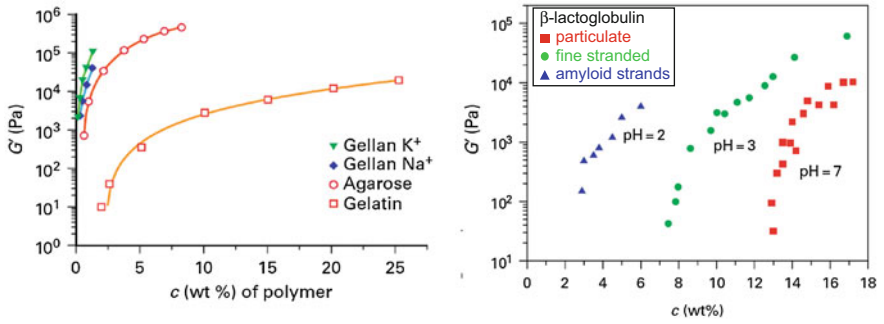

Fig. 4.5 Storage modulus of linear chain polymers and globular protein gels as a function of concentration. This comparison is not strict because the molar masses and other molecular parameters are not the same. Taken from Djabourov et al. (2013) collecting the data from Clark et al. (1983); Milas, Rinaudo (1996); Kavanagh et al. (2000); Veerman et al. (2002). Reproduced with permission from Djabourov et al. (2013), Copyright 2013 Cambridge University Press

higher concentration than agarose. Djabourov et al. (1988, 2013) plotted G' of gelatin gels as a function of helix content χ and found $G' = 0$ at $\chi < 8\%$.

4.7 Rigid Network Chains

Common characteristics in physical gels are that network chains are very stiff. Stiff chains tend to align to form a liquid crystal. Structure formation has been discussed in relation to Flory's phase diagram (1956) for rod-like polymers; agarose (Hayashi et al. 1978), MC (McAllister et al. 2015), xanthan (Lee and Brant 2002). Since stiff chains are expected to form a gel at a low concentration, it has been attracting much attention.

Fibrils have been attracting much attention in relation to amyloidosis diseases such as Alzheimer's, Creutzfeldt-Jakob disease (CJD), and type II diabetes (van der Linden 2012). Recently, β-lactoglobulin (β-lg) fibrils formed after heating at 80 °C and pH 2 for 10 h were observed by atomic force microscopy (AFM) and it was reported that the fibrils have a multi-stranded helical shape with twisted ribbon-like structures having persistence length from 1000 to 3000 nm (Adamcik et al. 2010). Since the fibrils of globular proteins have a similar structure to those causing amyloidosis diseases, it is necessary to confirm the safety of these materials. Bateman et al. (2010) reported that β-lg fibrils could be digested in simulated gastric fluid. They found that the fibrils were digested completely by pepsin within 2 min. Fibrils of β-lg are known to form a gel above a certain concentration depending on pH, ionic strength (I), and temperature. A phase diagram of solutions of β-lg fibrils at pH 2 was constructed as a function of concentration (0–2 wt%) and ionic strength (0–800 mM NaCl) at 20 °C (Bolisetty et al. 2012). Gel phase is found in the region $C = 0.3$–2.0 wt% and $I =$ ca 40–100 mM. Recently, Cao et al. (2018) found that the

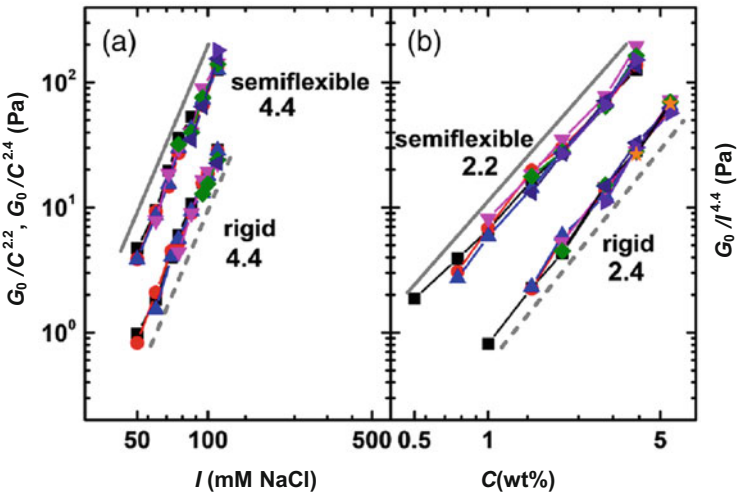

Fig. 4.6 Normalized G_0 of BLG fibril networks as a function of (**a**) ionic strength I (mM NaCl) and (**b**) polymer concentration C (wt%). Reproduced with permission from Cao et al. (2018), Copyright 2018 APS

dependence of plateau modulus G_0 on the polymer concentration C scales as $G_0 \sim C^{2.2}$ for semiflexible fibrils or $G_0 \sim C^{2.4}$ for rigid fibrils (Fig. 4.6b).

Based on the recent theoretical treatment on the stiff chain networks by groups of MacKintosh, Janmey, Weitz, Cao et al. deduced $G_0 \sim C^{11/5}$ for semiflexible fibrils network and $G_0 \sim C^{5/2}$ for rigid fibrils network, which coincided well with their experimental observation (Fig. 4.6b). As for the dependence of plateau modulus G_0 on the ionic strength I, Cao et al. (2018) found $G_0 \sim I^{4.4}$ for both semiflexible fibrils and rigid fibrils (Fig. 4.6a). They explained this high exponent taking into account the DLVO theory which led to the exponent 4.1.

It was shown recently that β-lg fibrils in the presence of transglutaminase (TGase) form a gel at a much lower β-lg concentration (Wu et al. 2016) because TGase has a crosslinking ability to form isopeptide bonds between glutamine and lysine residues in proteins, thus introducing both inter- and intramolecular covalent crosslinks.

4.8 Interaction of Short Chains and Long Chains

Fang et al. (2007) re-examined the so-called egg-box model which has been proposed for the gelation of alginates and pectins, where two or more chains are involved in cooperative binding, forming the egg-box structure, by isothermal titration calorimetry (ITC). Three distinct and successive steps in the binding of calcium to alginate were identified with increasing concentration of Ca ions. They were assigned to (1) interaction of Ca^{2+} with a single guluronate unit forming monocomplexes; (2) propagation and formation of egg-box dimers via pairing of

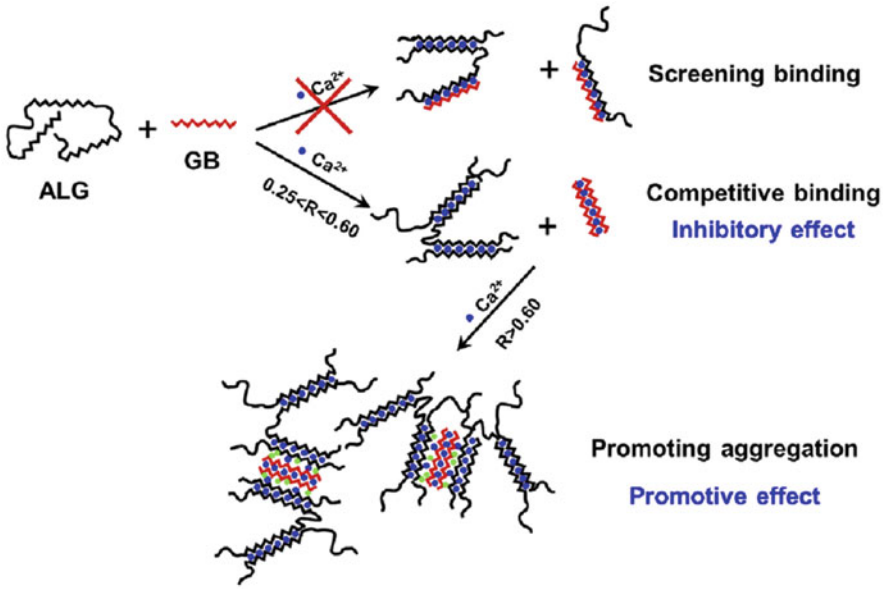

Fig. 4.7 Coexistence of oligoguluronate (GB), short chain guluronate, with alginates promotes the aggregation and strengthens the gel in the presence of sufficient calcium while at low Ca^{2+} it inhibits the network growth by binding with calcium. The blue dots represent Ca^{2+} and the green dots represent Na^{+} and H_2O etc. Reproduced with permission from Liao et al. (2015), Copyright 2015 Elsevier

these monocomplexes; and (3) lateral association of the egg-box dimers, generating multimers.

Since alginates and pectins both are known to have a similar structure and form gels in the presence of calcium, it is expected to see the similar multiple step gelation process also in pectin. The different behaviors in alginates and pectin gelation were attributed to the structural difference; alginates are block-copolymer while arrangement of copolymer units in pectins is more random (Fang et al. 2008).

Recently, effects of the addition of oligoguluronate, guluronate block (GB) to alginates with and without salt on rheological properties were studied, and the ratio of Ca to guluronate G, R(Ca/G) was found to play an important role (Liao et al. 2015). The addition of GB was inhibitive in the range of $0.25 < R\ (Ca/G) < 0.60$ and promotive in the range of $R > 0.60$. These two effects were shown to be associated with the different molecular events that dominate the gelation of alginate, namely, egg-box dimerization and lateral aggregation. Quantitative analysis indicates a competitive binding rather than a screening binding during egg-box dimerization, which led to the inhibitory effect in the lower Ca concentration regime. On the other hand, in the higher Ca concentration regime where alginate gelation is predominated by chain lateral aggregation, the dimers formed by GB could act as a binder to enhance the aggregation of alginate dimers, resulting in a promotive effect on alginate gelation (Fig. 4.7). The results are consistent with the microstructures observed by AFM.

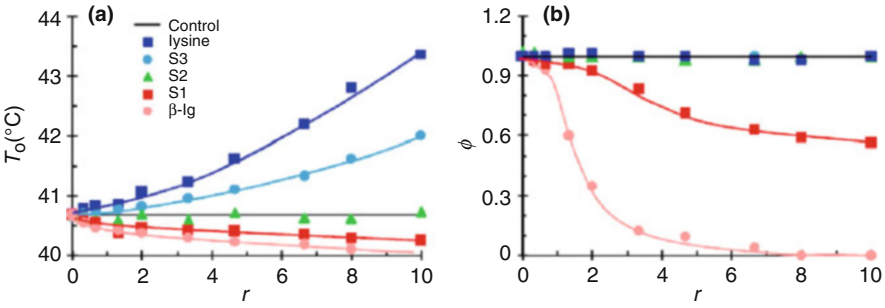

Fig. 4.8 The effect of β-lg or its hydrolysates on the onset DSC temperature (T_0) of helix formation of κ-car (**a**) and relative extent (φ) of the conformational transition of κ-car (**b**) as a function of the mixing ratio r = β-lg/κ-car. The concentration of κ-car 0.15%; pH = 4.7. Reproduced with permission from Cao et al. (2016b), Copyright 2016 ACS

Cao et al. (2016a) studied the mixture of κ-carrageenan (κ-car) and β-lg with different mixing ratios r = β-lg/κ-car, and found that the exothermic peak enthalpy of κ-car accompanying coil to helix transition detected by DSC decreased with increasing r. It was analyzed quantitatively based on a theory of McGhee and von Hippel (1974) which was used to explain the binding of protein to DNA. The relative extent of conformational transition ϕ (r) could be experimentally measured as the ratio of the enthalpy change of conformational transition of protein/polyelectrolyte mixture (ΔH (r)) to that of pure polyelectrolyte (ΔH ($r = 0$)). The inhibiting degree ϕ of κ-car helix formation by β-lg was quantified by the enthalpy change as a function of r. Electrostatic complexation in soluble state had subtle effect on the coil-to-helix transition of κ-car, which is due to the relatively high freedom of κ-car at the initial stage of protein binding. Electrostatic complexation in insoluble state however greatly suppressed the conformational transition, which is due to the high physical hindrance imposed by β-lg upon extensive protein binding. The effect of protein/polyelectrolyte electrostatic complexation on the conformation transition of polyelectrolyte was described quantitatively. The effect was closely associated with the molar mass of β-lg or its hydrolysates: Larger hydrolysates S1 (>2000 Da) had an inhibitory effect on the conformational transition of κ-car, smaller hydrolysates S3 (<1000 Da) tended to promote it (Fig. 4.8).

4.9 Cation- or Acid-Induced Gelation

Gelation of globular proteins can be induced either by heating (heat induced gelation) or by cold gelation. In the cold gelation, a low concentration solution of native proteins is heated or subjected to high pressure and soluble aggregates are formed. The aggregates remain soluble on cooling. Then, gelation is induced either by adding salt or by changing the pH toward the isoelectric point of the proteins in order to reduce the electrostatic repulsion. Cold-set gels have been used as a vehicle

to deliver thermally labile and water insoluble functional ingredients (Chen et al. 2006), and recently cold-set emulsion gels are also attracting much attention (McClements 2017; Khalesi et al. 2019).

Ion-induced gelation of alginate has been used to produce an imitation *ikra* (salmon roe). When a droplet of non-gelling sodium alginate falls into calcium lactate solution, a thin film is formed on the surface of droplet. This process was patented by a Japanese chemical industry (Nishinari 1988). This very fast gelation can be used to produce artificial berries and onion rings (Draget 2021). Since this gelation of sodium alginate is too fast and leads to inhomogeneous gel formation, slow release of calcium ions from insoluble calcium salts is used by chelating agents EDTA, citrate) or glucono-delta-lactone to control the gelation rate and to form homogeneous gels (Draget 2021). Alginate is also used to treat heart burn (acid reflux) since alginate solution forms a gel raft on the top surface in the stomach because of its low pH (ca 1.5) (Gaviscon has been used).

5 Characterization Methods

5.1 *Gelation Kinetics: Time Dependence of Storage and Loss Moduli G′ and G″ of a Solution at a Constant Temperature and a Frequency*

When a solution prepared at a non-gelling temperature is kept at a certain gelling temperature, G* begins to increase with lapse of time. Generally employed condition of the measurements are as follows: the frequency and the amplitude should be as low as possible so that the structure being formed might not be broken, however, the frequency ca l Hz has been chosen in most cases. The geometry of the apparatus for viscoelastic measurements should be chosen so that the injected sample solution can be quenched (cooled rapidly) or can be heated rapidly. This can be satisfied generally if the required volume of sample solution is small.

Generally, the gelation proceeds faster at higher temperatures for a heat-setting system whilst it proceeds faster at lower temperatures for a cold-setting system. It is easier to carry out the rheological measurement at the temperature where the gelation proceeds slowly for the analysis of kinetics. In the first order kinetics, the storage modulus is written as follows:

$$G'(t) = G'_{\text{sat}} \left[1 - \exp\left(-k\left(t - t_0\right)\right)\right],$$

where t_0 is the latent time (gelation time), k is the rate constant, G'_{sat} is the saturated value of the storage modulus after a long time. Solutions of some biopolymers such as methyl cellulose, degalactosylated xyloglucan, curdlan, glycinin, and β-conglycinin form a gel on heating, while solutions of other biopolymers such as agarose, carrageenan, gelatin, and gellan form a gel on cooling (All these were cited

in Nishinari 1997). When the gelling polymer is composed of fast gelling and slow gelling components, the above kinetic equation can be modified using two rate constants and two latent times for each components. More kinetic models have been proposed, see p. 386 in Lapasin and Pricl (1999). Recently, the gelation of β-lactoglobulin was analyzed by a simple eq. $G'(t) = G'_{sat} \exp(-\beta/t)$ proposed by Scott Blair in 1963.

Lapasin et al. (1990) reported that both moduli increased first and then decreased during gelation of calcium pectate. The decrease of the moduli was attributed to the structure breakdown into smaller flow units.

Rennet-induced gels have been studied extensively by many groups as a basis for cheese production. The dependence of casein micelle size on the gelation has been studied using micelles with different sizes prepared by differential centrifugation, and it was found that smaller casein micelles form a stronger gel faster (Niki et al. 1994). It was interpreted on the basis that the κ-casein, which plays a main role in rennet-induced casein gelation, is abundant on the surface of casein micelles.

As in the enzymatic modification of casein, partial hydrolysis has a great potential to control the structure and property of proteins. Doi et al. (1987) performed a limited proteolysis of ovalbumin (OVA) (Mw = 45,000) using a pepsin, and obtained an intermediate OVA (Mw = 42,500), called p-OVA having a similar physicochemical properties with native OVA, and compared the gelation. This will be described later in Sect. 7.

To control the rheological and structural properties of proteins, transglutaminase (TGase) was used by several research groups. Jaros et al. (2010) reported G'_{max} in GDL-induced gelation process of TGase-treated casein solution at 30 min after the GDL addition, and G'_{max} depended strongly on the incubation time by TGase. G'_{max} increased and then deceased with the incubation time, and was attributed to rearrangement and not due to the slippage frequently observed in gelation experiments. Syneresis and tanδ showed the minimum at 30 min after the GDL addition when G' showed the maximum (Fig. 4.9). In the processing of yogurt, various polysaccharides or microparticulated whey proteins are added to increase G', which reduces the syneresis and increases the creaminess (Jørgensen et al. 2019).

Soy protein gels are used not only for tofu but also attracted attention as raw materials for cheese and yogurt to replace animal milk, and the optimization condition has been studied by many research groups (Nishinari et al. 2018).

5.2 *Mechanical Spectra: Frequency Dependence of* G′ *and* G″ *at a Constant Temperature*

Figure 4.10 shows the frequency dependence of gellan solutions with a small and large amount of NaCl. Most commonly available commercial gellan samples contain salts to enhance the gelling ability. This figure shows the behavior of gellan which is converted into pure sodium type to see the effect of salts on the gelation behavior.

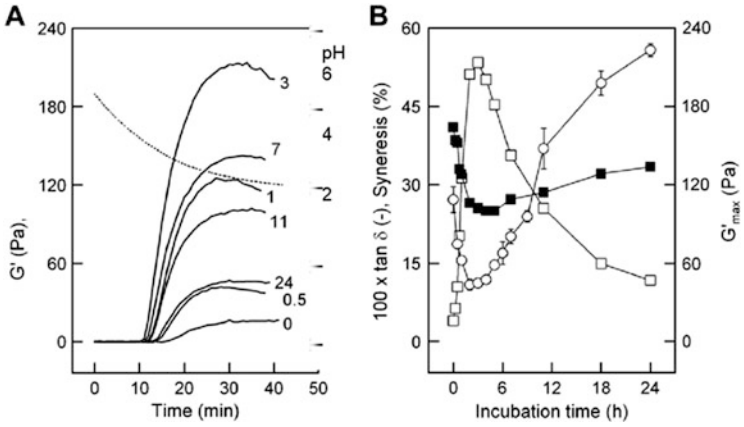

Fig. 4.9 (**a**) Averaged gelation curves of three enzyme treated casein solutions (27 g kg^{-1}). TGase treatment was with 3U TGase g^{-1} protein at 40 °C, the numbers refer to incubation times in hours. Inactivation was with 1 g L^{-1} N-Ethylmaleimide, and gelation was induced by 40 g L^{-1} GDL at 30 °C. Dotted line shows the corresponding pH decay. (**b**) G'_{max} (□) with the corresponding tan δ (■) and syneresis (○) of cross-linked casein gels as affected by enzyme incubation time. Data are mean values ± standard deviations of three individual experiments. Reproduced with permission from Jaros et al. (2010), Copyright 2010 Elsevier

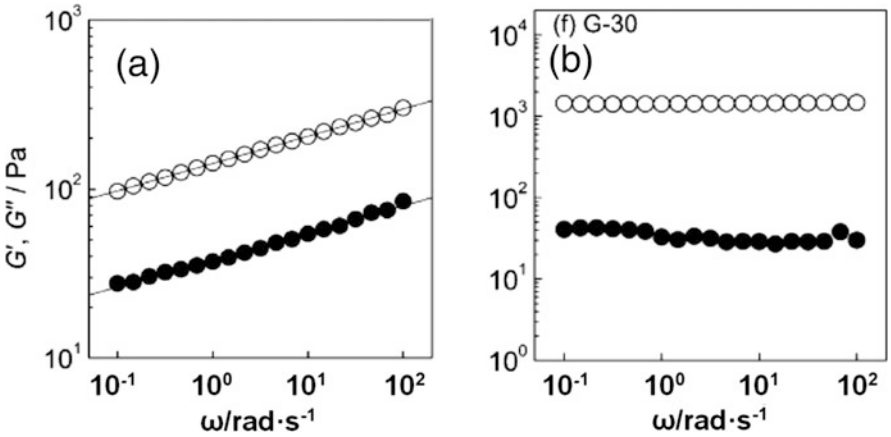

Fig. 4.10 Frequency dependence of G' (open) and G'' (closed) of 2 wt% sodium type gellan with 30 mM NaCl at 5 °C (**a**) and 0.3 wt% sodium type gellan with 100 mM NaCl at 35 °C (**b**) Reproduced with permission from Nitta et al. (2010), Copyright 2010 ACS

Both storage modulus G' and loss modulus G'' show a weak frequency dependence in Fig. 4.10a, while both G' and G'' show a plateau in Fig. 4.10b although the polymer concentration is much lower in the latter.

The behavior in Fig. 4.10a is seen in a structured liquid (often called a "weak gel," but this term was not to be used because it is essentially a liquid), and is different

from a "true gel" or "elastic gel" behavior shown in Fig. 4.10b. The difference between a structured liquid and an elastic gel appears in a larger value of tangent delta for the former because of its liquid nature. The large deformation behavior shows a more clear difference between a structured fluid and an elastic gel; the former flows while the latter fractures above the yield stress. For example, see the strain dependence of G′ and G″ in Fig. 4.4 and the frequency dependence G′ and G″ in Fig. 4.3 for κ-carrageenan in a paper by Ikeda and Nishinari (2001d).

In the globular protein such as soybean β-conglycinin, the gelation commences only at higher temperatures than the denaturation temperature. The frequency dependence for G' and G'' of β-conglycinin or glycinin solution begins to show a plateau at 68 °C or 80 °C, respectively (Nagano et al. 1994a; Nishinari et al. 2014).

5.3 *Thermal Scanning Rheology*

Temperature dependence of storage and loss moduli G' and G'' of a solution at a constant frequency.

Figure 4.11 shows the gelation process of a 15% soybean β-conglycinin solution. At a constant temperature of 80 °C, G' increased, and continued to increase when the temperature was lowered from 80 °C to 20 °C at the rate of 2 °C/min. G' decreased with increasing temperature from 20 °C to 80 °C at the same rate. Values of G' are symmetrical about the vertical line at $t = 3600$ s. The increase in G' after 1800 s when heating is stopped and the temperature is lowered is attributed to the further formation of network structure by hydrogen bonding which may be broken by subsequent heating from 20 °C to 80 °C. Although it is almost impossible to evaluate the contribution of hydrophobic interactions, hydrogen bonding, ionic interactions, and covalent bonding quantitatively because these interactions operate

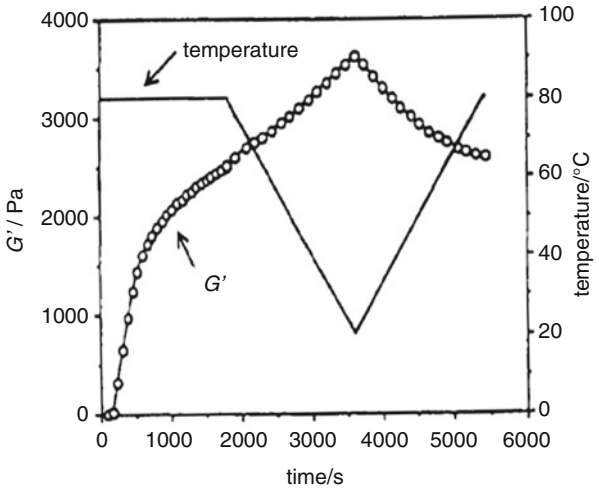

Fig. 4.11 Gelation process of 15% soybean β-conglycinin solution at pH 7.6. The solution was heated at 80 °C for 30 min (=1800 s), and then cooled to 20 °C at 2 °C /min, and heated again to 80 °C at the same rate. Reproduced with permission from Nagano et al. (1994b), Copyright 1994 ACS

simultaneously, Fig. 4.11 clearly shows the important contribution of hydrogen bonding to the gel formation of β-conglycinin.

At higher temperatures for a 2 wt% solution of sodium type gellan, $G' < G''$ while on cooling moduli show a steep increase at ca 35 °C (T_{ch}) which is attributed to coil-to-helix transition, which coincides with the temperature at which the molecular ellipticity shows a transition. On further cooling, G' and G'' show a crossover at 8 °C (T_{sg}) which is ascribed to sol-gel transition (Miyoshi and Nishinari 1999). This sol-gel transition was found not the formation of a true gel but a structured liquid. Strictly speaking, this procedure to determine the gelation point is not precise because the crossover point depends on the frequency of measurement in addition to scan rate.

For gelling polysaccharides, it is necessary to pay attention to the slippage which sometimes leads to a serious misinterpretation. The cooling curve of G' and G'' using a normal cone and plate geometry showed a peak for both G' and G'' in the cooling process of carrageenan solutions, while that using perforated cylinders which prevent the slippage, no peak of G' and G'' was observed (Richardson and Goycoolea 1994). Zhang et al. (2001) also found the peak of G' and G'' as a function of time for 2% dispersions of KGM at higher temperatures at $\omega = 1$rad/s. It is known that gelation of KGM is faster at higher temperatures. Zhang et al. (2001) measured the compressive force during gelation of KGM which is free from slippage, and then they did not observe the peak even at higher temperatures confirming that the peak of G' and G'' observed was caused by the slippage.

6 Physical Properties of Gels

6.1 Effect of Gelling Rate on Gel Properties

The elastic modulus of gelatin gels increased with time at lower temperatures not reaching an equilibrium value even after 100 h (Nijenhuis 1981, 1997). The thermal stability of gelatin gels increased with increasing storage time at the temperature which is a little higher than gelation temperature (Michon et al. 1997). For agarose gels, which are other helix-forming thermoreversible gels, the effect of the storage temperature near the gelation temperature on rheological and structural properties has been studied and it was found to differ from the behavior of gelatin gels. Aymard et al. (2001) found that holding solutions of agarose for long times at high temperatures decreased the strength of the gels formed on cooling. They interpreted this by structural observation that during the holding period the un-gelled solutions resolved progressively into regions of high and low polymer concentration, and that the resulting heterogeneity gave weaker networks when the solutions were gelled by cooling. On the other hand, Mohammed et al. (1998) reported that agarose gels showed larger elastic modulus and were more thermally stabilized by cooling more slowly. Recently, it was shown that storage Young's modulus and the fracture stress and strain of gellan gels increased with decreasing cooling rate (Nitta et al. 2014).

As mentioned before, a gel may be formed before a solid film is formed when the solvent is evaporated from, for example, solutions of pullulan which is usually not called a gelling polysaccharide because it forms a gel only at higher concentrations. The evaporation speed influences the length scale of the structure being formed, but this problem has only been tackled recently (Schaefer et al. 2015) mainly in relation to spin coating. This may be called a kind of concentration quench, as temperature quench influenced the formation of gel structure.

6.2 *Temperature Dependence of Elasticity of Gels*

Sol-gel transition in polysaccharides can be classified into four groups: (1) cold-set gels like agarose, kappa- and iota-carrageenans, and gellans which form a gel on cooling the solution; (2) heat-set gels like some cellulose derivatives such as methyl cellulose (MC), curdlan, konjac glucomannan (KGM) which form a gel on heating the solution; (3) inverse reentrant gels like a mixed solution of methyl cellulose and gelatin, which forms a gel at higher and lower temperatures and stays a sol state at an intermediate temperature range. Gelatin may be replaced by some po1ysaccharides which form a gel on cooling; and (4) reentrant gels like xyloglucan from which some galactose residues are removed. It forms a gel at an intermediate specific temperature range and remains in the sol state at higher and lower temperatures outside of this specific temperature range. Some copolymer gels show a complex temperature dependence: Sol-gel transition is observed at two or three different temperatures, i.e. they show reentrant transition (Nishinari 2000a, b, 2001, 2009).

There have been many investigations and debate on the temperature dependence of the elastic modulus or gels relating with the entropic and energetic contribution. Since it was difficult to observe the elastic modulus of thermoreversible gels as a function of temperature from a low temperature, the increase in modulus with increasing temperature was not observed. It was concluded that the gel elasticity is energetic elasticity in previous studies while the opposite was also asserted. To understand this problem it is effective by dividing the elastic modulus into entropic and energetic contribution from the temperature dependence of the elastic modulus of gels of agarose with different molar masses (Nishinari et al. 1984).

Reel-chain model was proposed to explain the temperature dependence of thermoreversible gels (Nishinari et al. 1985). In this model, network consists of Langevin chains which allow to treat large deformation. On heating, some segments are released from junction zones consisting of aggregated helices. There should be a limit for number of segments released before gel-to-sol transition occurs. Some statistical calculations gave the elastic modulus as a function of the binding energy, upper limit number of segments released from the junction zone, average distance between junction zones. It predicts the monotonic increase of the modulus for a high binding energy as expected from rubber elasticity theory, and the monotonic decrease for a low binding energy, and an intermediate behavior showing the

maximum of the modulus as a function of temperature. This theory was applied to many thermoreversible gels to explain the temperature dependence of the modulus.

6.3 Molar Mass Dependence of Elastic Modulus

Since it is difficult to prepare samples with different molar masses with a narrow molar mass distribution, there have not been so many studies on the molar mass dependence of elastic modulus of gels. Saunders and Ward (1955) studied rheological properties of gelatin gels with different molar masses and showed that the elastic modulus increased with increasing molar mass up to a certain value and then leveled off whilst the breaking stress continued to increase with increasing molar mass. Similar tendency was observed for gels of alginate (Smidsrød 1974) and κ-carrageenan (Rochas et al. 1990). The gelation kinetics of dispersions of konjac glucomannan with different molar masses has been studied by measurements of storage shear modulus and it was shown that the storage modulus of the dispersion of the same concentration increased with increasing molar mass (Yoshimura and Nishinari 1999).

Effects of molar mass on the stress-strain curve and on the temperature dependence of the modulus of agarose gels are shown in Fig. 4.12. Both the stress and strain at break increased (Fig. 4.12a), and the entropic behavior of elastic moduli was enhanced (Fig. 4.12b) with increasing molar mass of agarose.

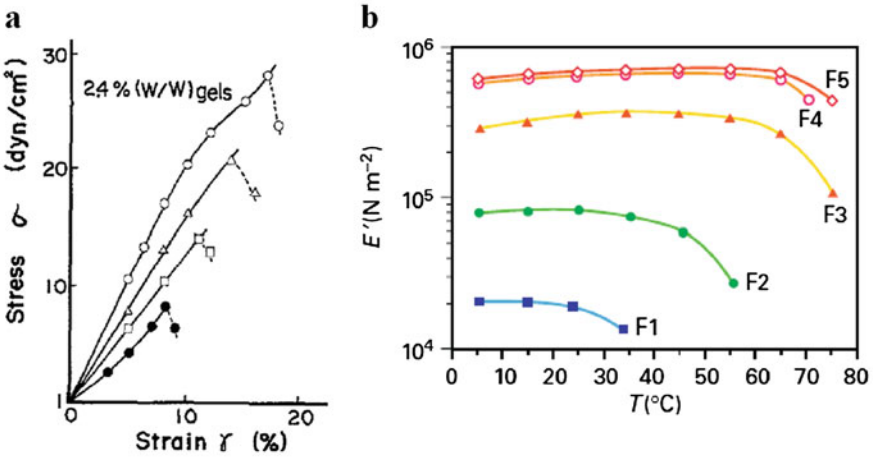

Fig. 4.12 (**a**) Stress-strain relation of 2.4% (w/w) gels of agarose with different molar masses (• < □ < △ < ○) at 25 °C. Reproduced with permission from Watase and Nishinari (1983), Copyright 1983 Springer. (**b**) Temperature dependence of storage modulus E' for five agarose fractions with M_w from 3.4×10^4 g mol^{-1} (F1) to 48.5×10^4 g mol^{-1} (F5) (Nishinari and Watase 1993)

Molar mass dependence of the moduli was found sometimes contradictory (Nishinari and Fang 2021). While from a figure of G' as a function of concentration in amylose gels showed a greater G' for a higher molar mass amylose (Clark et al. 1989), a similar plot for oat β-glucan showed a greater G' for a lower molar mass (Lazaridou et al. 2003). This apparent inconsistency is originated from the non-equilibrium nature of these physical gels. In both amylose and oat β-glucan, the gelation proceeded faster for lower molar mass because of the higher mobility. This slower pace of gelation is different in amylose and β-glucan, and therefore, great care is required to compare the molar mass dependence of the modulus. Gel formation of higher molar mass fractions is so slow that the modulus at a time not long enough is smaller than that for lower mass fractions. Gelation of gelatin is also known to take a long time as mentioned before (Nijenhuis 1981, 1997).

6.4 Molecular Motion (Rearrangement) in Gels

Agarose forms a gel at a very low concentration < 0.1 wt% depending on the molar mass, sulfate content (purified agarose or idealized agarose contains no sulfate groups but most commercially available agarose contains a small amount of sulfate groups). Even after gelation it is not in the equilibrium state. Syneresis, which is an exudation process of water from the network, is also observed in globular protein gels such as cheese (syneresis is necessary) and yogurt (syneresis is not preferable). This process is not so fast, and therefore gel engineers study small and large deformation rheological behavior of these gels in pseudo-equilibrium state. Agarose gels as well as other polysaccharide gels are believed to be formed by aggregated double helices connected by flexible chains. Since the concentration is low, there are free spaces where molecular motion is occurring.

A step-like decrease of tan δ was observed while the endothermic peak in DSC and the steep change in the specific ellipticity in CD were observed at 25 °C on heating a 1.6% gellan gel, and these changes are attributed to helix-coil transition (Nitta et al. 2001; Nishinari et al. 2001). A molecular motion or molecular rearrangement such as the helix-coil transition occurs in an apparently solid material with the elastic modulus of ca 10^5 Pa. This was a surprise for authors because of the appearance of a solid of the gellan gels. From the viewpoint of the solid content, most part of the gel consists of water and thus there must be much enough space where large-scale molecular motion or rearrangement can occur.

Helix-coil transition can occur in different ways. Segments can be released from junction zones on heating as was discussed in reel-chain model approach (See Sect. 6.2). On cooling, these released segments are reeled into junction zones so that junction zones become longer and/or thicker. If the elastic modulus is mainly determined by the number of elastically active chains which connect junction zones as in the theory of rubber elasticity, the increase in elastic modulus should be attributed to the increase in the number of elastically active chains rather than to the thickening or lengthening of junction zones. It is quite possible that long

segments released from junction zones form a new junction by helix formation. Helix-coil transition does not necessarily occur only at the chain ends, but it also may occur at the intermediate point of long chains.

There may be different junction zones with different thermal stabilities. Weak junction zones with low thermal stabilities may disintegrate at lower temperatures which arc directly related to helix-coil transitions, and these chains produced from the disintegration of helices may form helices on cooling only at lower temperatures. The distribution of thermal stability may cause a broad endo- and exo-thermic peak in DSC heating and cooling curves as well as gradual increase and decrease of the specific ellipticity in heating and cooling CD measurements. According to a fibrous model (Morris et al. 2000), the elasticity of gels arises mainly from stretching and bending of fibers that consist of aggregated helices. When some ends of these fibers arc converted into flexible chains by helix-coil transition, these ends will cease to contribute to the elasticity and then the elastic modulus will decrease. Even if one of these conformational changes occurs, it does not necessarily lead to the gel-to-sol transition. When these changes are only local, only less ordered helices are converted into coils, and the whole network structure is not broken down, and keeps the size and shape of the self-supporting gel.

6.5 Chain Release and Erosion of Gels

Most gel technologists using chemical gels formed by covalent bonds believe that a polymer gel consists of a three-dimensional crosslinked network and swells in a solvent to a certain finite extent, but does not dissolve even in a good solvent. It was found that some molecular chains in a gellan gel release out from the gel when it is immersed in a solvent such as water or salt solutions (Tanaka and Nishinari 2007; Hossain and Nishinari 2009). After a long time, the gel swells infinitely, and disperses completely in a great amount of water. On immersion of gels in water, cations and non-crosslinked chains diffuse out from the gel into the surrounding medium, and the gel structure is weakened. Salt diffusion from the gels into the surrounding medium is faster than chain release; chains which lose condensed or bound ions cannot retain a helical conformation, and so they then diffuse out. Analysis of the released chains revealed that the shorter chains are released predominantly. The modulus increase by immersion was observed at first, and it was suggested by stiffening of elastically active chains. Potassium ions in the external solvent were found to strengthen the gel structure of gellan, yet tetramethylammonium (TMA) ions in the external solvent, which are known to inhibit the gelation though not inhibiting helix formation, were also found to increase the modulus.

A mass loss in gellan gels prepared by calcium ions were also found, and after 28 days these gels maintained constant composition without further mass loss during the remainder of the test period between 28 and 164 days (De Silva et al. 2013). This shows the difference between monovalent cations and divalent cations as will be discussed in Sect. 6.7.

6.6 Syneresis/Water Holding Capacity (WHC)

Syneresis is a phenomenon in which water oozes out from the gel network, and is known from long time ago but its mechanism is not so well understood although the importance has been recognized because of its close relation with flavor release and juiciness perception. Many methods of measurements have been proposed: centrifugation, compression, wiping the surface, measuring water on the top of a test tube containing the gel, etc. As Mizrahi described (Mizrahi 2010), the syneresis may be governed by the swelling pressure which is the difference between the osmotic pressure and the network pressure $\Pi_{sw} = \Pi_{os} - \Pi_{net}$, where the osmotic pressure is produced by polymer chains in the gel and it drives water into the gel, thus causing the swelling and stretching of polymer network. Stretched chains tend to contract to compensate the decrease of the entropy caused by the chain stretching, thus giving rise to an internal pressure which is called a network pressure. Mizrahi (2010) suggested several methods to prevent the syneresis by increasing the osmotic pressure, controlling network pressure and crosslinking.

Miwa et al. (1994) compared the syneresis of 3–4 wt % gels of curdlan, carrageenan, agar, konjac after kept at 4 °C for 20 h, and the syneresis of both curdlan and konjac was 10.3% which was much larger than that of carrageenan (1.4%) and agar (0.6%). However, after freezing and thawing, syneresis of both curdlan and konjac increased to ca 21% while it was much more than that for carrageenan and agar because these gels became sponge-like and water ran out through larger pores. This encouraged food industry to use curdlan and konjac in frozen foods such as surimi to improve the texture. Syneresis of agar gels was reported to decrease with increasing agar concentration/added sucrose/sulfate residue. It should be reminded that the order of the addition of sucrose to make polysaccharide gels, before or after gelation, changes the structure/mechanical properties and syneresis. For example, the addition of a higher sucrose content (>55 wt%) in 1 wt% agar gelation before heating was found to make the gel inhomogeneous and led to a weaker gel with more syneresis (Yang et al. 2015). It was found that the elastic modulus decreased after syneresis although the concentration was increased by syneresis, and it was attributed to the loosening of the network structure caused by syneresis (Nagasaka and Taneya 2000; Nishinari and Fang 2016, 2017). It was also shown that syneresis was reduced by adding sucrose or starch to curdlan though it also reduced the gel strength. While this practical manipulation may be sometimes useful, the basic understanding of syneresis in polysaccharide gels is not yet reached (Ako 2017; Divoux et al. 2015).

Syneresis in globular protein gels may be understood in the first approximation by permeability B which is proportional to the area and the fraction of pores in a cross section of the gel (Walstra 2003). Walstra explains the great difference in the syneresis in ground coffee ($B \sim 10^{-8}$ m^2), rennet milk gel ($B \sim 10^{-12}$ m^2), and polysaccharide gel ($B \sim 10^{-17}$ m^2) by the approximate value of the permeability. The difference of B values roughly explains that some polysaccharide gels retain water while globular protein gels show a much more syneresis. While syneresis is undesirable in most gels, it is an essential process in cheese making, where most of the liquid (whey) in the rennet gel has to be removed (Walstra 2003). In these protein

gels, rearrangements of strands consisting of globular protein aggregates lead to loose ends which in turn form junctions thus becoming more compact expelling liquids. When junctions are broken, a pressure is built up, which is called "endogenous syneresis pressure," that is causing syneresis even without external pressure such as gravity. However, the experimental determination of the endogenous syneresis pressure seems to be difficult, and thus the complete understanding of the syneresis of protein gels is still a challenge.

Nieuwland et al. (2016) found a good correlation between the structure, mechanical properties, and WHC for ovalbumin (OVA) gels at a pH range from 5.8 to 6.8: the microstructure of OVA gels became less dense based on SEM and CLSM, and the Young's modulus decreased, in parallel with the decrease in WHC with decreasing pH.

Urbonaite et al. (2016) prepared 14 w/w% WPI gels at pH 7.2 with different pore sizes ranging from 10^{-2} to 10^{0} μm by changing NaCl content from 0 to 300 mM to examine the effect of coarseness (pore size) and the mechanical properties, Young's modulus on WHC. They found that, depending on the lower or higher than 100 mM NaCl, fine stranded gels (length scale <0.1μm) or coarse stranded gels (>0.1μm) were formed. They found the Young's modulus showed the maximum at 100 mM NaCl, while the WHC decreased with increasing concentration of added NaCl, and concluded that coarseness was more dominant than stiffness (Young's modulus) for water holding.

Dai et al. (2016) examined the effects of the addition of 0.5% KGM on the physicochemical properties of yogurt, and found that the syneresis was decreased while retaining the structural stability and the firmness of full-fat yogurt. They found the suppression of syneresis was equivalent to previous reports in which inulin or orange fiber was added to skimmed yogurt and noticed that total solid content was also responsible for the reduction of syneresis.

To improve the texture and water holding capacity of myofibrillar protein gels, dietary fibers are widely used. Effects of the addition of insoluble fibers (microcrystalline cellulose, oat fiber, walnut shell flour) and native and modified starches (potato, tapioca) on liquid loss, hardness and resilience (recoverable deformation) of chicken breast myofibrillar proteins were measured (Gravelle et al. 2017). Since fat and connective tissues were removed, the samples were myofibrillar protein gels, and liquid loss was mainly from water. Among the fillers tested, native potato starch was most effective to reduce the liquid loss, which was attributed to the difference in hydration estimated by T_2 relaxation time of water.

6.7 *Effect of Sugars, Salt, Acid, and Polyphenols on Rheological Behavior of Gels*

It has been empirically known that the addition of sucrose to polysaccharide gels make them transparent, less brittle and reduce the syneresis. The Young's modulus,

fracture stress, and melting enthalpy increased and then decreased with increasing sucrose or glucose in agarose or κ-carrageenan gels (Nishinari and Fang 2016). The different strengthening effect of sugars has been correlated with a number of equatorial hydroxyl residues in sugars. Kawai et al. (2007, 2008) extended the reel-chain model in which flexible chains are released from junction zones constituting aggregated helices to explain the observed strain hardening and then softening in uniaxial compression. As the end-to-end distance increases due to the deformation, more and more segments are reeled out from the junction zone. Finally, one end of the chain is liberated from the junction and the chain becomes dangling. The appearance of dangling chains causes the strain softening because they cease to contribute to the elasticity.

Drying of gels has been studied from various viewpoints, and recently the effect of sugars on drying kinetics of agarose gels was studied (Mao et al. 2017).

Epiga1locatechin gallate (EGCG), a polyphenol abundant in tea leaves was found to induce gelation of non-gelling xyloglucan (Nitta et al. 2004). Yan et al. (2016) further reported that other gallate analogs also induce gelation of xyloglucan.

Generally, salt does not influence so much the gelation of neutral polysaccharides such as agarose but sometimes enhances the gelation of KGM or xyloglucan. Effects of salts on gelation of charged polysaccharides have been extensively studied. While the monovalent cations shield the electrostatic repulsion of carboxyl groups in gellan, divalent cations bind different molecular chains of gellan, and therefore once the gel is formed it does not return to the sol state on heating up to 90 °C (Miyoshi et al. 1994; Djabourov et al. 2013).

7 Particulate Disordered Structure: Globular Protein Gels

When globular proteins are denatured by heating, high pressure or denaturants such as urea or guanidine hydrochloride, hydrophobic portion folded and buried inside were exposed outside, which is called unfolding, and then unfolded polypeptide chains form a three-dimensional network (Doi 1993). In comparison with network structure formed from long chain molecules which are predominantly entropic and have been successfully elucidated to some extent by developing rubber elasticity theory, particulate disordered gels are less well understood. Rheology and structure of rennet-induced or acid-induced casein or whey protein gels (Nicolai et al. 2011), ovalbumin gels (Nemoto 2000; Nieuwland et al. 2016) as well as soybean proteins (Nishinari et al. 2014), and other plant proteins have been extensively studied.

A great difference from thermoreversible polysaccharide gels such as agarose, kappa-carrageenan, gellan is that the minimum concentration required is much higher for globular protein gels, but recently, gelation of fibrils made from globular protein changed this common knowledge. This was described in “4.7 Rigid network chains.” Most polysaccharide gels except cellulose derivatives such as methyl cellulose form a gel by hydrogen bonds and thus form a gel on cooling while globular protein gels are formed on heating and it is thought that the denaturation

of the native protein structure is prerequisite for gel formation. The main force responsible for gelation is thought to be hydrophobic interaction, but other secondary forces are also contributing.

Dutch school employed a fractal model and measured the permeability and rheology to correlate the structure and property of milk protein gels. Bremer et al. (1989) concluded that acid casein gels can be described very well by a fractal model with a fractal dimension 2.3, while it was not applicable for rennet-induced casein gels because of microsyneresis. The modulus decreased with increasing temperature which contradicts some earlier publications affirming that the casein gels are entropic like rubber elasticity (van Vliet and Walstra 1985). The increase of the aging temperature to 50 °C and extending the aging time to 157 h changed the gel to a more solid like material; the storage modulus tends to show a rubber like plateau and the loss tangent decreased. Storage modulus G' as a function of measuring temperature T_m for acid sodium caseinate gels aged at different temperatures increased with increasing aging temperature, but decreased with the temperature of measurement (Roefs and van Vliet 1990). It is generally accepted that in casein gels, an ultimate effect of structural rearrangement is syneresis: expulsion of liquid. Syneresis is desired for manufacturing of cheese but not for yogurt.

Doi (1993) classified the gelation of globular protein into random aggregates and string beads network. His group studied the gelation of ovalbumin (OVA) by rheology, dynamic light scattering (DLS), CD, TEM, SEM, CLSM based on the conformational studies reporting that OVA molecules are worm-like chains, linear aggregates, with cylindrical diameter 12 nm and the persistence length 23 nm (Doi et al. 1987; Nemoto 2000). As a result of heat denaturation, hydrophobic regions are exposed on the surface, and linear polymer or aggregate is formed depending on the balance of hydrophobic interaction and electrostatic repulsion (Fig. 4.13).

They found that the heat induced gelation of 39–59% OVA solution showed a critical structure obeying the Winter-Chambon criteria (Koike et al. 1996), and found a very low exponent 0.09–0.14. Gels formed with concentrations lower than 25% and higher than 59% did not show critical structure. The exponent of concentration dependence was found to change from 4 below 59% to 10 above the concentration range from 59% to 89%, where the modulus an order of 10^9 Pa close to the glass modulus was found. This is totally different from common concentration dependence of modulus for polysaccharide gels where the exponent is ca 4 at lower concentration range, but ca 2 at a higher concentration range as described in "4.6 Critical molar mass and concentration for gelation."

A two-step heating was employed in addition to one-step heating; the OVA solution was first heated without NaCl for 60 min, and quickly quenched to room temperature where linear aggregates molar mass higher than several millions were formed. After addition of NaCl (20, 60, and 100 mM) to the viscous solutions, they were reheated for various time periods from 2 to 240 min and then immediately cooled (Koike et al. 1998). Transparent gels were obtained by both the one-step and the two-step heating. Doi (1993) described methods to obtain transparent egg white or milk whey protein gels controlling the pH and ionic strength by two-step heating.

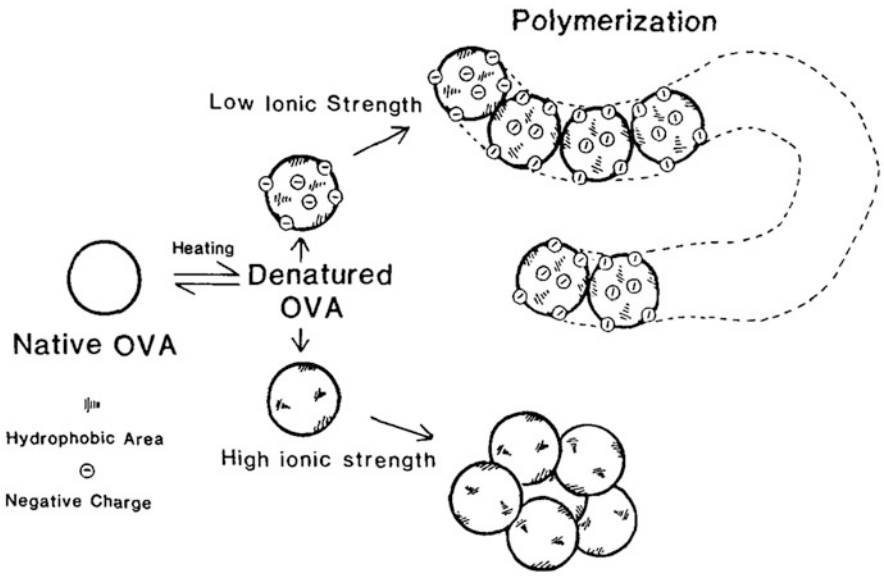

Fig. 4.13 Schematic representation of the heat denaturation of OVA, leading to the formation of linear polymer or aggregate (Doi et al. 1987). Reproduced with permission from Doi et al. (1987), Copyright 1987 John Wiley & Sons

More recently, gelling behavior of OVA and p-OVA (formed by a limited hydrolysis with pepsin) was analyzed based on SANS (small angle neutron scattering) in addition to DLS (Hiroi et al. 2016). In spite of the similarity of physicochemical properties, intrinsic viscosity, secondary structure, denaturation temperature, OVA and p-OVA showed significantly different gelling behaviors on heating. Heating 6 wt% solutions at neutral pH without salt led to a transparent gel for OVA while a turbid gel was formed for p-OVA. On heating OVA solution, the resulted aggregate size was found proportional to the protein concentration judging from the independence of the peak position in the SANS intensity as a function of scattering angle. On the contrary, an increase in the amount of large clusters formed on heating was thought to disturb the correlation of p-OVA dimers resulting in wide distribution of the distance between each solute and thus a two-phase separated structure was formed. The fact that the cleavage of only 22 N-terminal residues from OVA led to a very different gelling behavior is expected to be a hint to develop further utilization of OVA.

Globular protein solutions show a "solid-like" mechanical spectra (shown in Chap. 3) even before gelation probably because they form a colloidal crystalline structure (Matsumoto and Inoue 1993; Ikeda and Nishinari 2001a, b, c). Tobitani and Ross-Murphy (1997) made a temperature-concentration diagram constructed by the gelation temperature for each concentration of globular protein which is determined

by interpolation and extrapolation of the data set, a locus of nominally infinite gelation time.

8 Mixed Gels

Just as in alloys of metals and plastics, many mixed gels have been prepared to respond to the demand of various functionalities (Morris 1986). Many papers have been published on the mixed gels of xanthan, locust bean gum, konjac mannan, etc., and some common features are recognized for synergistic interaction, which means that the only one polysaccharide does not form a gel at that concentration, but forms a gel by mixing with other polysaccharide. 1) One polysaccharide undergoes coil-helix conformational transition, such as agarose, carrageenan, furcellaran, gellan, and xanthan gum. 2) The backbone linkage of the other polysaccharide in the mixture is β(1,4) linkage, such as in konjac glucomannan or galactomannan, or xyloglucan (Morris 1994).

Polysaccharide mixture gels have been extensively studied, and Unilever model and Norwich model have been proposed and examined. Gels of the mixture of xanthan and galactomannan or konjac glucomannan have been studied, and it was proposed that helical xanthan associates with the smooth region of galactomannan devoid of galactose side chains (Unilever model, Dea et al. 1977) while Norwich model proposed that the mixed gel was formed between coiled xanthan and galactomannan or KGM because gels were formed only after the helical xanthan was denatured into coil state (Brownsey et al. 1988). The latter statement was affirmed by adding helix enhancing cations to raise the helix-coil transition and in such a condition the mixture did not form a gel on cooling. However, the gelation in the mixture of pyruvate free xanthan (PFX) and KGM was observed to start about 40 °C more than 60 °C lower than coil-to-helix transition temperature of xanthan at acidic pH (3.5 and 4) higher than 100 °C, and that the synergistic gelation of KGM with PFX and commercial xanthan was not so different. Thus, the above-mentioned prerequisite of Norwich group for the gel formation of the mixture, the conformation of xanthan should be in coil state, was refuted (Agoub et al. 2007).

The effect of salt is important to study the mixtures. Annable et al. (1994) studied the mixture of xanthan and KGM in the presence of different salts using dynamic viscoelasticity, DSC, and ESR. They showed that the gelation temperature shifted to lower temperatures when electrolyte is present, with divalent cations having a greater effect than monovalent cations. These observations are explained by the fact that electrolyte promotes xanthan self-association at the expense of xanthan/KGM interaction, and thus the temperature for gelation on cooling shifted to lower temperatures with increasing xanthan self-association. The effect of salt follows the Hofmeister series. The strength of the cationic effect was as follows: $K^+ \sim Cs^+ < Na^+ \sim NH^{4+} \sim Ba^{2+} < <Mg^{2+} \sim Ca^{2+}$ (Annable et al. 1994).

Recently, Takemasa and Nishinari (2016) found the nuclear Overhauser effect (NOE) between low molar mass galactomannan and xanthan, which is a direct

evidence for the synergistic interaction because NOE is seen only for nuclei closer than about 0.5 nm.

Although synergistic interaction leading to an industrially interesting phenomenon such as enhancement of thickening and gelling ability has been attracting great interest, the most common phenomenon occurring in the mixing of different biopolymers is phase separation. Fang et al. (2006) constructed a state diagram of gelatin/κ-carrageenan aqueous mixtures based on turbidity measurement, confocal scanning laser microscopy (CSLM), DSC, and zeta potential measurements. They found a coexistence of associative and segregative (associative-*co*-segregative) phase separations at low temperature and low NaCl concentration in addition to compatible, associative, and segregative phase separation behaviors. A coexistence of associative and segregative phase separations was observed and it was attributed to a kinetically trapped state by gelation.

The effect of the addition of insoluble fibers and starches (discussed in Sect. 6.6 Syneresis) on mechanical properties of myofibrillar protein gels (Gravelle et al. 2017) is discussed here. When the addition of fiber was low (mass fraction m_f of filler <0.1–0.15), the hardness increased with increasing m_f. While the resilience, estimated from the immediately recovered deformation, was decreased with increasing m_f in myofibrillar protein gels with insoluble fibers, that of starch-filled protein gels was almost independent of m_f. This was interpreted that swelling of starch granules made the filler more flexible and diminished the stress concentration at the interface. The stress concentration S_c as a function of m_f estimated for completely bonded interface was shown to decrease, and thus the relative strength R_s which is an inverse of the stress concentration, $R_s = 1/S_c$, increased with increasing m_f (Gao and Lelievre 1994).

The fundamental problems and many examples of mixed gels are discussed in Chap. 10 of Djabourov et al. (2013) Cao et al. (2016a, b) and in Nicolai (2019).

9 Fracture of Food Gels

Fracture of gels can be divided into brittle fracture and ductile fracture. Fracture occurs at the structural defects. To avoid the uncertain distribution of structural defects, the fracture test is sometimes done for notched samples (van Vliet and Walstra 1995). While Young's modulus obtained at the small deformation is not so influenced by the depth of the notch *l,* the fracture stress decreased with increasing *l.*

In the extension test, the gel tends to fracture at the clamps or slips if the gel is not clamped tightly or too loosely, therefore it is not easy to obtain reproducible results. A memorable record in food gels was reported by van Kleef (1986) who could use *224 experiments* which were successfully performed free from slippage or fracture of the gels at the clamps among more than several hundreds of experiments for OVA gels. He got the relation between the fracture stress and the protein concentration C: $\sigma_b = 102C^{2.6}$ for the pH 10 gels and $\sigma_b = 5.89C^{3.2}$ for the pH 5 gels.

Fragment size distribution of masticated fish sausages was analyzed, and it was found that the distribution was fitted well by combination of the lognormal

distribution with exponential tail indicating that the fragmentation process has a size-segregation-structure between large and small parts. For large fragments population, incorporation of exponential distribution $N(s) = Ae^{-s/B}$ in addition to lognormal distribution was effective. In the mastication, the segregation of fragments into larger and smaller fragments occurs, but the number of large fragments decreases with the progress of mastication and the value of B decreases with the increasing number of mastication (Kobayashi et al. 2010). Recently, the fragmentation process in the mastication was studied using agarose gels with different sizes and with different molar masses. The effects of gel size and molar mass were recognized in the early stage of mastication. The average particle size showed a high correlation with the hardness (Moritaka et al. 2019).

Soft gels are known to be crushed between the tongue and the hard palate (Nishinari et al. 2020). Many such food gels have recently been produced for disadvantaged persons. Ishihara et al. (2013, 2014) studied the compression of agar gels and gellan gels using an artificial tongue made from silicone gel, and the transition from the tongue only breaking to chewing by teeth was found to be predicted from the instrumental uniaxial compression.

10 Microgels (Fluid Gels)

Microgels can be produced by shearing the gelling polymer solutions undergoing sol-gel transitions (Norton et al. 1999) or other method using emulsion in which gelling polysaccharides are dispersed. These microgels have been used as texture modifiers, stabilizers and especially for reduced fat foods. With a finite yield stress, microgels behave as a solid at rest but flow when subjected to a stress above the yield stress. In addition to many kind of polysaccharides from agar (Farres and Norton 2015), carrageenan (Garrec et al. 2013), and other origins, whey protein isolate (Moakes et al. 2015) was also used to make microgels.

Microgels of alginates were studied to encapsulate probiotics such as lactobacillus which are labile in severe gastrointestinal environments. In comparison with external gelation where alginate solution is dripping into calcium chloride solution, internal gelation of alginate is performed in alginate emulsion containing insoluble calcium carbonate particles by slowly lowering pH using GDL, which can produce small alginate beads ca 1μm or even smaller than 200 nm (Paques et al. 2013). Smaller gel beads are advantageous because larger particles than 25μm (this value depends on the shape of particles, hard and irregular particles are perceived as gritty than soft and round particles of similar size; Engelen et al. 2005) are detected on the palate, and thus can extend the application. Cai et al. (2014) compared alginate beads prepared using calcium carbonate and calcium disodium ethylenediamine- tetra-acetate and found that encapsulation using the former protected *L. acidophilus* more effectively in the survival test, which was related to the mechanical strength of the microcapsules.

Protein microgels can be also produced by ultrasonication of heated β-lg at low pH (Murphy et al. 2017, 2018). Produced microgels of β-lg with a diameter ca

230 nm were shown to be stable without coalescence over 6 weeks storage; however, emulsions containing limonen stabilized by the microgels were susceptible to creaming, flocculation, and limonen was lost during storage, which remain as future problems.

11 Cryogels

A cryogel is formed by the cryogenic treatment, freezing–frozen storage–thawing of the precursor system. Scientific research became active since the discovery of cryogels of poly(vinyl alcohol) (Lozinsky and Okay 2014). According to this definition, traditional Japanese foods such as *koori-tofu* (dried tofu after freezing), *bo kanten* (agar-stick, resulting from freezing–thawing) are cryogels but since they are xerogels and are eaten after absorbing solvent (water) by cooking, the characteristics of these xerogels are quite different from those of swollen food gels. Most gels lose their elastic characteristics once they are frozen and thawed. Most of them lose their solvents and only dried framework remains like agar-stick and koori-tofu. Cryogels keeping the rubber-like texture and water even after freeze–thaw cycles have been studied extensively including PVA and hyaluronan (Djabourov et al. 2013; Zhang et al. 2013). Other cryogels of amylopectin, gelatin, maltodextrin, potato starch, oat β-glucan have also been studied, but most of them are sponge-like cryogels (Lozinsky and Okay 2014). LBG solution was found to form a gel after the repeating cycles of freezing and thawing (Tanaka et al. 1998). It was found that LBG does gel from solutions containing in excess of 1% solids at room temperature on a time-scale of several months. The gelation mechanism of LBG in concentrated sucrose solutions was shown to be governed by frustrated crystallization process with nucleation and growth stages rather than reversible pairwise crosslinking and the gelation rate became maximum at −5 °C (Richardson and Norton 1998). This experimental finding may give a clue to explain the well-known fact that LBG forms a gel via a freeze–thaw cycle.

Giannouli and Morris (2003) reported that ca. 0.5% xanthan solution forms a cryogel when subjected to a freeze–thaw cycle. As has been known, xanthan solution is a structured liquid and can suspend solid particles. This network is known to be strengthened by calcium ions. Giannouli and Morris reported that the cryogelation was abolished by adding sucrose higher than 30% and also by adding 0.4 mM calcium ions. The inhibiting role of sucrose was attributed to the freezing point depression, and thus ice crystallization was inhibited leading to the insufficient alignment and association of xanthan chains. The weakening of cryogelation of xanthan by calcium ions was suggested that the strengthening the structure of xanthan by calcium restricts the alignment during freezing, thus hampering the cryogel formation.

Doyle et al. (2006) studied the effect of sugars (sucrose, glucose and fructose) and sorbitol on the cryogelation of galactomannans. They prepared galactomannans with different M/G ratios from 2.65 to 4.16 by enzymatic modification of guar gum

(M/G ~ 1.6), and in addition they used LBG with much higher molar mass. They found that Young's modulus determined from the initial slope of the compression curve, the stress at break, increased up to ca 50% sugar and then decreased with increasing concentration of sugars. On the other hand, the strain at break was found to decrease monotonically with increasing sugar concentration. In addition, they found that Young's modulus, stress at break as a function of M/G increased steeply around M/G = 4, and the chain length did not affect so much for these behaviors. They raise again the freezing point depression as one of the reasons. Since the concentration of sugars used in this study was 40–60% and was lower to make the system glassy state where conformational ordering of polysaccharide chains was inhibited. Then, the inhibition of polymer–polymer association by binding of sugar molecules to polysaccharide chains was thought to be the main reason for the decrease in Young's modulus, and stress at break after reaching the maximum.

It was found that cereal β-glucans from oat, barley, and wheat (molar mass ca. 200×10^3) form cryogels, and both elastic modulus and the fracture stress increased with increasing freeze–thaw cycles (Lazaridou and Biliaderis 2004). They further examined the effects of sugars and polyols (fructose, glucose, sorbitol, sucrose, and xylose) on rheological and thermal properties of cryogels of barley β-glucans (Lazaridou et al. 2008). While both G' and G'' decreased by adding fructose, glucose, sucrose, and xylose, both modulus increased by adding sorbitol. On heating, G' of cryogels slightly decreased up to 50–60 °C, then decreased steeply indicating the melting. The melting temperature shifted to lower temperatures except cryogels with sorbitol. Heating DSC curves also showed the endothermic peak accompanying the melting, and the melting peak temperature shifted to lower temperatures in the presence of polyols. However, the endothermic enthalpy was increased by the addition of polyols. They interpreted these experimental results by the freezing point depression of the β-glucan solutions by adding of sugars that may decrease the ice crystallization leading to the weaker structural formation of cryogels. They attributed the difference between rheological behavior and thermal behavior to the difference in the global structure and local structure.

Though some trial cryogel production of food hydrocolloids such as hydroxyethyl cellulose irradiated with UV-visible light at room temperature and in the frozen state or chitosan cryogels formed in moderately frozen aqueous solutions using glutaraldehyde as a crosslinker (Okay 2014), they are not still acceptable in food use. It is expected that other polysaccharide or protein cryogels are developed.

12 Oleogels

Since the findings of adverse effect of trans fatty acids on blood lipids and coronary heart disease risk in the early 1990s, food industry tries to find a better way to modify the texture-property of oil/fat products without using a large amount of crystalline triacylglycerol molecules which are rich in saturated and or trans fatty acids. Solid fat foods such as butter and margarine keep a shape but are spreadable when

subjected to shear stress higher than their yield stress. One of the most important differences between polysaccharide and protein gels and solid fat is that the linear elastic range is very narrow in the latter in comparison with the former because of the crystalline nature in the latter. The most widely employed method of producing oleogels is dispersing a gelator into liquid oil. The well-known gelators include waxes such as rice bran waxes, beeswax, shellac, and ethylcellulose (Singh et al. 2017; Patel and Dewettinck 2016). All the edible oleogels reported show a structured liquid behavior as described earlier, that is, both G' and G'' are almost independent of frequency and $G' > G''$, tan $\delta > 0.1$ (de Vries et al. 2017; Moschakis et al. 2016; Zetzl et al. 2014; Patel et al. 2013).

The finding that the slowly formed gels have a stronger structure is widely observed for polymer gels but the different behavior is reported for **oleogels** made by dispersing gelator in edible oil. Oleogels of shellac, a natural resin secreted by lac insect, were prepared in the following way: the dispersion of shellac in the rapeseed oil was heated above melting temperature of shellac (>85 °C), and then cooled to room temperature resulting in oleogels. The onset of nucleation and crystal formation was delayed on slower cooling resulting in larger and less dense crystals. The storage modulus was found to decrease with lowering the cooling rate (Patel et al. 2013). This was explained by the increase in total effective area for smaller crystals which leads to the stronger network formation due to the higher crystal–crystal interactions.

13 Applications of Gelling Agents in Food Industry

Nata de coco, a kind of bacterial cellulose produced from coconut water, has been a favorite dessert jelly, and boomed in the 1990s especially in Japan, and still continued to be consumed all over the world, may be because of its health benefits, lowering cholesterol, and effective for diabetes and obesity in addition to its peculiar textural characteristics (Fontana et al. 2017). It has an anisotropic fibrous texture and thus expected to be a suitable matrix as vegetarian "meat," since it can have various colors and meat-like flavors (Fontana et al. 2017). The addition of small amount of bacterial cellulose increased the gel strength of tofu and *kamaboko* and thus improved the sensory evaluation (Okiyama et al. 1992, 1993; Shi et al. 2014).

In the preparation of mixed gels of κ-carrageenan, KGM, and LBG, the small deformation rheology was governed by κ-carrageenan, and Young's modulus decreased with decreasing κ-carrageenan concentration. The large deformation behavior examined by gel ring extension test, however, was dominated by KGM, and the rupture strain increased with increasing KGM concentration (Brenner et al. 2013). These two opposing trends led to a maximum in rupture stress. It should be noted that only κ-carrageenan has a gelling ability in this combination at neutral or lower pH, and KGM gels only at higher pH. This result may be generalized to design texture of food gels consisting of a deformable component and a brittle component.

The instantaneous gelation ability of alginate is applied to make an imitation *ikura* (salmon roe). A drop of salad oil is introduced into a sol of carrageenan or

xanthan which does not form a gel. This sol is wrapped by a film of alginate or pectin. The appearance resembles a natural salmon roe so even a fisherman cannot distinguish between them (Ueda 1985; Kishi 1977). This is quite often used for *sushi* and some other snacks. This became a popular demonstration in open campus event in universities.

Fish paste gels (*surimi* or *kamaboko*) show rubber-like behavior and frequently added starch plays a role of filler reducing the entropic nature of elasticity (Nishinari 1988). Many trials to improve the texture of surimi or meat products have been done by adding cellulose, KGM, curdlan, and other polysaccharides (Kaur and Sharma 2019; Zhuang et al. 2020). Anisotropy is an important factor controlling the fibrous texture, and has been successfully introduced in softened meat and string cheese (Nishinari 2020). A Japanese invention, crab stick (*kani-kama*) is consumed internationally, and the fibrousness is conferred by making the structure anisotropic sometimes adding alginates or other polysaccharides.

Frozen tofu containing curdlan keeps the smooth texture after thawing (Nakao et al. 1994) whilst when usual tofu is frozen, the texture becomes rough, which has been used as *koori-tofu* in Japanese and Chinese dishes.

Agar has been used widely in Japanese cuisine, and higher molar mass agar ($M_w \sim 5{\cdot}10^5$) is used for *tokoroten* (noodle-shaped agar gel served with vinegar and soy sauce or brown sugar syrup), while medium molar mass agar ($M_w \sim 3{\cdot}10^5$) is used for sweets and yogurt, and lower molar mass agar ($M_w < 10^5$) gels with low gel strength break down into microgels by a weak force, and thus suitable for mixing with honey to make it less sticky and easy to spread on toasted bread (Nishinari and Shiba 2007). Strictly speaking, agar is not a molecule but a mixture of agarose and agaropectin, and thus the molar mass is a kind of the average. This concept of the average molar mass has also been used for alginate, starch, and other biopolymers.

Melt-in-the mouth sensation of foods are liked in chocolate, ice cream and jellies. This means that most humans like the sensation or feeling when chocolate, ice cream and jellies are melted. Fish gelatin exploitation was pursued by many hydrocolloids research groups to overcome the BSE issue or religious issues. Xanthan cryogel (0.5%) (Giannouli and Morris 2003) and mixed gels of xanthan and KGM at acidic pH (3.5 and 4) (Agoub et al. 2007) were found to melt in the mouth temperature range, which might be useful for fruit jellies.

Aerogels have attracted much attention because of the light mouth feel and low calorie. Marshmallow, soufflé, and *hanpen* are typical examples and have been enjoyed before a short note with colloids science view appeared in Nature (Kistler 1931). Endeavor to develop aerogels more systematically to apply them in delivery carrier of active compounds using starch-based aerogels containing agar or microcrystalline cellulose (Dogenski et al. 2020) or in food packaging as absorber matrix (da Silva et al. 2020) is in progress.

Gelling polysaccharides agar, alginate, KGM, curdlan, carrageenan, gellan, pectin, starch are used in controlling the texture, water holding capacity of meat and fish products, and are also used to make dessert jellies (Nussinovitch and Hirashima 2014; Nishinari 2020).

References

Adamcik J, Jung JM, Flakowski J, De Los RP, Dietler G, Mezzenga R (2010) Understanding amyloid aggregation by statistical analysis of atomic force microscopy images. Nat Nanotechnol 5:423–428. https://doi.org/10.1038/nnano.2010.59

Agoub AA, Smith AM, Giannouli P, Richardson RK, Morris ER (2007) "Melt-in-the-mouth" gels from mixtures of xanthan and konjac glucomannan under acidic conditions: a rheological and calorimetric study of the mechanism of synergistic gelation. Carbohydr Polym 69:713–724. https://doi.org/10.1016/j.carbpol.2007.02.014

Ako K (2017) Yield study with the release property of polysaccharide-based physical hydrogels. Int J Biol Macromol 101:660–667. https://doi.org/10.1016/j.ijbiomac.2017.03.145

Annable P, Williams PA, Nishinari K (1994) Interaction in xanthan-glucomannan mixtures and the influence of electrolyte. Macromolecules 27:4204–4211. https://doi.org/10.1021/ma00093a023

Arvidson SA, Lott JR, McAllister JW, Zhang J, Bates FS, Lodge TP, Sammler RL, Li Y, Brackhagen M (2013) Interplay of phase separation and thermoreversible gelation in aqueous methylcellulose solutions. Macromolecules 46:300–309. https://doi.org/10.1021/ma3019359

Aymard P, Martin DR, Plucknett K, Foster TJ, Clark AH, Norton IT (2001) Influence of thermal history on the structural and mechanical properties of agarose gels. Biopolymers 59:131–144. https://doi.org/10.1002/1097-0282(200109)59:3<131::AID-BIP1013>3.0.CO;2-8

Bateman L, Ye A, Singh H (2010) In vitro digestion of β-lactoglobulin fibrils formed by heat treatment at low pH. J Agric Food Chem 58:9800–9808. https://doi.org/10.1021/jf101722t

Bolisetty S, Harnau L, Jung JM, Mezzenga R (2012) Gelation, phase behavior, and dynamics of β-lactoglobulin amyloid fibrils at varying concentrations and ionic strengths. Biomacromolecules 13:3241–3252. https://doi.org/10.1021/bm301005w

Bremer LGB, van Vliet T, Walstra (1989) Theoretical and experimental study of the fractal nature of the structure of casein gels. J Chem Soc Faraday Trans 1 Phys Chem Condensed Phases 85:3359–3372. https://doi.org/10.1039/F19898503359

Brenner T, Achayuthakan P, Nishinari K (2013) Linear and nonlinear rheology of mixed polysaccharide gels. Pt. I. Young's modulus, ring extension and uniaxial compression tests. J Texture Stud 44:66–74. https://doi.org/10.1111/j.1745-4603.2012.00366.x

Brown E, Jaeger HM (2014) Shear thickening in concentrated suspensions: phenomenology, mechanisms and relations to jamming. Rep Prog Phys 77:046602. https://doi.org/10.1088/0034-4885/77/4/046602

Brownsey G, Cairns P, Miles MJ, Morris VJ (1988) Evidence for intermolecular binding between xanthan and the glucomannan konjac mannan. Carbohydr Res 176:329–334. https://doi.org/10.1016/0008-6215(88)80146-0

Cai S, Zhao M, Fang Y, Nishinari K, Phillips GO, Jiang F et al (2014) Microencapsulation of lactobacillus acidophilus CGMCC1.2686 via emulsification/internal gelation of alginate using Ca-EDTA and $CaCO_3$ as calcium sources. Food Hydrocoll 39:295–300. https://doi.org/10.1016/j.foodhyd.2014.01.021

Cao Y, Fang Y, Nishinari K, Phillips GO (2016a) Effects of conformational ordering on protein/polyelectrolyte electrostatic Complexation: ionic binding and chain stiffening. Sci Rep 6:23739. https://doi.org/10.1038/srep23739

Cao Y, Li S, Fang Y, Nishinari K, Phillips GO (2016b) Conformational transition of polyelectrolyte as influenced by electrostatic complexation with protein. Biomacromolecules 17:3949–3956. https://doi.org/10.1021/acs.biomac.6b01335

Cao Y, Bolisetty S, Adamcik J, Mezzenga R (2018) Elasticity in physically cross-linked amyloid fibril networks. Phys Rev Lett 120:158103. https://doi.org/10.1103/PhysRevLett.120.158103

Chen LY, Remondetto GE, Subirade M (2006) Food protein-based materials as nutraceutical delivery systems. Trends Food Sci Technol 17:272–283. https://doi.org/10.1016/j.tifs.2005.12.011

Clark AH, Gidley MJ, Richardson RK, Ross-Murphy SB (1989) Rheological studies of aqueous amylose gels: the effect of chain length and concentration on gel modulus. Macromolecules 22:346–351. https://doi.org/10.1021/ma00191a063

Clark AH, Richardson RK, Ross-Murphy SB, Stubbs JM (1983) Structural and mechanical properties of agar/gelatin co-gels. Small-deformation studies. Macromolecules 16:1367–1374. https://doi.org/10.1021/ma00242a019

Cordier P, Tournilhac F, Soulié-Ziakovic C, Leibler L (2008) Self-healing and thermoreversible rubber from supramolecular assembly. Nature 451:977–980. https://doi.org/10.1038/nature06669

da Silva FT, de Oliveira JP, Fonseca LM, Bruni GP, da Rosa Zavareze E, Dias ARG (2020) Physically cross-linked aerogels based on germinated and nongerminated wheat starch and PEO for application as water absorbers for food packaging. Int J Biol Macromol 155:6–13. https://doi.org/10.1016/j.ijbiomac.2020.03.123

Dai S, Corke H, Shah NP (2016) Utilization of konjac glucomannan as a fat replacer in low-fat and skimmed yogurt. J Dairy Sci 99:7063–7074. https://doi.org/10.3168/jds.2016-11131

De Silva DA, Poole-Warren LA, Martens PJ, Panhuis M (2013) Mechanical characteristics of swollen gellan gum hydrogels. J Appl Polym Sci 130:3374–3383. https://doi.org/10.1002/app.39583

Dea ICM, Morris ER, Rees DA, Welsh EJ, Barnes HA, Price J (1977) Associations of like and unlike polysaccharides: mechanism and specificity in galactomannans, interacting bacterial polysaccharides, and related systems. Carbohydr Res 57:249–272. https://doi.org/10.1016/S0008-6215(00)81935-7

de Vries A, Wesseling A, van der Linden E, Scholten E (2017) Protein oleogels from heat-set whey protein aggregates. J Colloid Interface Sci 486:75–83. https://doi.org/10.1016/j.jcis.2016.09.043

Divoux T, Mao B, Snabre P (2015) Syneresis and delayed detachment in agar plates. Soft Matter 11:3677–3685. https://doi.org/10.1039/C5SM00433K

Djabourov M, Leblond J, Papon P (1988) Gelation of aqueous gelatin solutions. II. Rheology of the sol-gel transition. J de Phys 49:333–343. https://doi.org/10.1051/jphys:01988004902033300

Djabourov M, Nishinari K, Ross-Murphy SB (2013) Physical gels from biological and synthetic polymers. Cambridge University Press, Cambridge. https://doi.org/10.1017/CBO9781139024136

Dogenski M, Navarro-Díaz HJ, de Oliveira JV, Ferreira SRS (2020) Properties of starch-based aerogels incorporated with agar or microcrystalline cellulose. Food Hydrocoll 108:106033. https://doi.org/10.1016/j.foodhyd.2020.106033

Doi E (1993) Gels and gelling of globular proteins. Trends Food Sci Technol 4:1–5. https://doi.org/10.1016/S0924-2244(05)80003-2

Doi E, Koseki T, Kitabatake N (1987) Effects of limited proteolysis on functional properties of ovalbumin. J Am Oil Chem Soc 64:1697–1703. https://doi.org/10.1007/BF02542506

Doyle JP, Giannouli P, Martin EJ, Brooks M, Morris ER (2006) Effects of sugars, Galactose content and chain length on freeze–thaw gelation of galactomannans. Carbohydr Polym 64:391–401. https://doi.org/10.1016/j.carbpol.2005.12.019

Draget K (2021) Alginates. In: Phillips GO, Williams PA (eds) Handbook of hydrocolloids, 3rd edn. Woodhead Publishing, pp 805–829. https://doi.org/10.1016/B978-0-12-820104-6.00007-3

Eldridge JE, Ferry JD (1954) Studies of the cross-linking process in gelatin gels. III. Dependence of melting point on concentration and molecular weight. J Phys Chem 58:992–995. https://doi.org/10.1021/j150521a013

Engelen L, van der Bilt A, Schipper M, Bosman F (2005) Oral size perception of particles; effect of size, type, viscosity and method. J Texture Stud 39:83–113. https://doi.org/10.1111/j.1745-4603.2005.00022.x

Fang Y, Al-Assaf S, Phillips GO, Nishinari K, Funami T, Williams PA (2008) Binding behavior of calcium to polyuronates: comparison of pectin with alginate. Carbohydr Polym 72:334–341. https://doi.org/10.1016/j.carbpol.2007.08.021

Fang Y, Al-Assaf S, Phillips GO, Nishinari K, Funami T, Williams PA, Li L (2007) Multiple steps and critical behaviors of the binding of calcium to alginate. J Phys Chem B111:2456–2462. https://doi.org/10.1021/jp0689870

Fang Y, Li L, Inoue C, Leif Lundin L, Appelqvist I (2006) Associative and segregative phase separations of gelatin/κ-carrageenan aqueous mixtures. Langmuir 22:9532–9537. https://doi.org/10.1021/la061865e

Fang Y, Takemasa M, Katsuta K, Nishinari K (2004) Rheology of Schizophyllan solutions in isotropic and anisotropic phase regions. J Rheol 48:1147–1166. https://doi.org/10.1122/1.1781170

Farres IF, Norton IT (2015) The influence of co-solutes on tribology of agar fluid gels. Food Hydrocoll 45:186–195. https://doi.org/10.1016/j.foodhyd.2014.11.014

Feke GT, Prins W (1974) Spinodal phase separation in a macromolecular sol-gel transition. Macromolecules 7:527–530. https://doi.org/10.1021/ma60040a022

Fontana JD, Koop HS, Tiboni M, Grzybowski A, Pereira A, Kruger CD, da Silva MGR, Wielewski LP (2017) Chapter 7 New insights on bacterial cellulose. In: Grumezescu AM, Hlban AM (eds) Food biosynthesis, handbook of food bioengineering, vol 1. Academic Press, Elsevier, Amsterdam. https://doi.org/10.1016/B978-0-12-811372-1.00007-5

Funami T, Kataoka Y, Hiroe M, Asai I, Takahashi R, Nishinari K (2007) Thermal aggregation of methylcellulose with different molecular weights. Food Hydrocoll 21:46–58. https://doi.org/10.1016/j.foodhyd.2006.01.008

Gao YC, Lelievre J (1994) A theoretical analysis of the strength of composite gels with rigid filler particles. Polym Eng Sci 34:1369–1376. https://doi.org/10.1002/pen.760341802

Garrec DA, Guthrie B, Norton IT (2013) Kappa carrageenan fluid gel material properties. Part 1: rheology. Food Hydrocoll 33:151–159. https://doi.org/10.1016/j.foodhyd.2013.02.014

Gekko K, Kasuya K (1985) Effect of pressure on the sol-gel transition of carrageenans. Int J Biol Macromol 7:299–306. https://doi.org/10.1016/0141-8130(85)90028-5

Gekko K, Li X, Makino S (1992) Effects of polyols and sugars on the sol-gel transition of gelatin. Biosci Biotechnol Biochem 56:1279–1284. https://doi.org/10.1271/bbb.56.1279

Giannouli P, Morris ER (2003) Cryogelation of xanthan. Food Hydrocoll 17:495–501. https://doi.org/10.1016/S0268-005X(03)00019-5

Gravelle AJ, Barbut S, Marangoni AG (2017) Food-grade filler particles as an alternative method to modify the texture and stability of myofibrillar gels. Sci Rep 7:11544. https://doi.org/10.1038/s41598-017-11711-1

Halake K, Lee J (2017) Functional hyaluronic acid conjugates based on natural polyphenols exhibit antioxidant, adhesive, gelation, and self-healing properties. J Ind Eng Chem 54:44–51. https://doi.org/10.1016/j.jiec.2017.04.018

Haque A, Morris ER (1993) Thermogelation of methylcellulose. Part I: molecular structures and processes. Carbohydr Polym 22:161–173. https://doi.org/10.1016/0144-8617(93)90137-S

Hayashi A, Kinoshita K, Kuwano M, Nose A (1978) Studies on the agarose gelling system by the fluorescence polarization method II. Polym J 10:485–494. https://doi.org/10.1295/polymj.10.485

Hiroi T, Okazumi Y, Littrell KC, Narita Y, Tanaka N, Shibayama M (2016) Mechanism of heat-induced gelation for ovalbumin and its N-terminus cleaved form. Polymer 93:152–158. https://doi.org/10.1016/j.polymer.2016.04.036

Hossain KS, Nemoto N, Nishinari K (1997) Dynamic viscoelasticity of iota carrageenan gelling system near sol-gel transition. Nihon Reoroji Gakkaishi 25:135–142. https://doi.org/10.1678/rheology1973.25.3_135

Hossain KS, Nishinari K (2009) Chain release behavior of gellan gels. Progr Colloid Polym Sci 136:177–186. https://doi.org/10.1007/978-3-642-00865-8_25

Ikeda S, Nishinari K (2000) Intermolecular forces in bovine serum albumin solutions exhibiting solidlike mechanical behaviors. Biomacromolecules 1:757–63. https://doi.org/10.1021/bm005587o

Ikeda S, Nishinari K (2001a) On solid-like rheological behaviors of globular protein solutions. Food Hydrocoll 15:401–406. https://doi.org/10.1016/S0268-005X(01)00052-2

Ikeda S, Nishinari K (2001b) Solid-like mechanical behaviors of ovalbumin aqueous solutions. Int J Biol Macromol 28:315–320. https://doi.org/10.1016/S0141-8130(01)00128-3

Ikeda S, Nishinari K (2001c) Structural changes during heat-induced gelation of globular protein dispersions. Biopolymers 59:87–102. https://doi.org/10.1002/1097-0282(200108)59:2<87::AID-BIP1008>3.0.CO;2-H

Ikeda S, Nishinari K (2001d) "Weak gel"-type rheological properties of aqueous dispersions of nonaggregated κ-carrageenan helices. J Agric Food Chem 49:4436–4441. https://doi.org/10.1021/jf0103065

Ishihara S, Isono M, Nakao S, Nakauma M, Funami T, Hori K, Ono T, Kohyama K, Nishinari K (2014) Instrumental uniaxial compression test of Gellan gels of various mechanical properties using artificial tongue and its comparison with human Oral strategy for the first size reduction. J Texture Stud 45:354–366. https://doi.org/10.1111/jtxs.12080

Ishihara S, Nakao S, Nakauma M, Funami T, Hori K, Ono T, Kohyama K, Nishinari K (2013) Compression test of food gels on artificial tongue and its comparison with human test. J Texture Stud 44:104–114. https://doi.org/10.1111/jtxs.12002

Jaros D, Jacob M, Otto C, Rohm H (2010) Excessive cross-linking of caseins by microbial transglutaminase and its impact on physical properties of acidified milk gels. Int Dairy J 20:321–327. https://doi.org/10.1016/j.idairyj.2009.11.021

Jin Y, Zhang H, Yin Y, Nishinari K (2006) Comparison of curdlan and its carboxymethylated derivative by means of rheology, DSC, and AFM. Carbohydr Res 341:90–99. https://doi.org/10.1016/j.carres.2005.11.003

Joly-Duhamel C, Hellio D, Ajdari A, Djabourov M (2002) All gelatin networks: 2. The master curve for elasticity. Langmuir 18:7158–7166. https://doi.org/10.1021/la020190m

Jørgensen CE, Abrahamsen RK, Rukke E-O, Hoffmann TK, Johansen A-G, Skeie SB (2019) Processing of high-protein yoghurt - a review. Int Dairy J 88:42–59. https://doi.org/10.1016/j.idairyj.2018.08.002

Kaur R, Sharma M (2019) Cereal polysaccharides as sources of functional ingredient for reformulation of meat products: a review. J Funct Foods 62:103527. https://doi.org/10.1016/j.jff.2019.103527

Kavanagh GM, Clark AH, Ross-Murphy SB (2000) Heat-induced gelation of globular proteins: 4. Gelation kinetics of low pH b-lactoglobulin gels. Langmuir 16:9584–9594. https://doi.org/10.1021/la0004698

Kawai S, Nitta Y, Nishinari K (2007) Large deformation analysis of gellan gels. J Appl Phys 102:043507. https://doi.org/10.1063/1.2769143

Kawai S, Nitta Y, Nishinari K (2008) Model study for large deformation of physical polymeric gels. J Chem Phys 128:134903. https://doi.org/10.1063/1.2894845

Khalesi H, Emadzadeh B, Kadkhodaee R, Fang Y (2019) Effect of Persian gum on whey protein concentrate cold-set emulsion gel: Structure and rheology study. Int J Biol Macromol 125:17–26. https://doi.org/10.1016/j.ijbiomac.2018.12.051

Kishi S (1977) Nippon Carbide Kogyo Co. Ltd., Jap. Pat. 52-59079

Kistler SS (1931) Coherent expanded aerogels and jellies. Nature 127:741. https://doi.org/10.1038/127741a0

Kobayashi N, Kohyama K, Shiozawa K (2010) Fragmentation of a viscoelastic food by human mastication. J Phys Soc Jpn 79:044801. https://doi.org/10.1143/JPSJ.79.044801

Koike A, Nemoto N, Doi E (1996) Structure and dynamics of ovalbumin gels. I. Gel induced by high temperature heat treatment. Polymer 37:587–593. https://doi.org/10.1016/0032-3861(96)83145-4

Koike A, Takada A, Nemoto N (1998) Structure and dynamics of ovalbumin gels: II. Gels induced by heat treatment at 80°C. Polym Gels Networks 6:257–271. https://doi.org/10.1016/S0966-7822(98)00018-5

Kometani K, Tanabe M, Su L, Yang K, Nishinari K (2015) In situ observations of Thermoreversible gelation and phase separation of agarose and methylcellulose solutions under high pressure. J Phys Chem B 119(22):6878–6883. https://doi.org/10.1021/acs.jpcb.5b03632

Lapasin R, Pricl S (1999) Rheology of industrial polysaccharides: theory and applications. Springer, Berlin

Lapasin R, Pricl S, Paoletti S, Zanetti F (1990) Novel rheological model for the gelatin kinetics of ionic polysaccharides. J Appl Polym Sci 41:1395–1410. https://doi.org/10.1002/app.1990.070410704

Lawal OS, Yoshimura M, Fukae R, Nishinari K (2011) Microporous hydrogels of cellulose ether cross-linked with di- or polyfunctional glycidyl ether made for the delivery of bioactive substances. Colloid Polym Sci 289:1261–1272. https://doi.org/10.1007/s00396-011-2458-0

Lazaridou A, Biliaderis CG (2004) Cryogelation of cereal β-glucans: structure and molecular size effects. Food Hydrocoll 18:933–947. https://doi.org/10.1016/j.foodhyd.2004.03.003

Lazaridou A, Biliaderis CG, Izydorczyk MS (2003) Molecular size effects on rheological properties of oat β- glucans in solutions and gels. Food Hydrocoll 17:693–712. https://doi.org/10.1016/S0268-005X(03)00036-5

Lazaridou A, Vaikousi H, Biliaderis CG (2008) Effects of polyols on cryostructurization of barley β-glucans. Food Hydrocoll 22:263–277. https://doi.org/10.1016/j.foodhyd.2006.11.012

Lee HC, Brant DA (2002) Rheology of concentrated isotropic and anisotropic xanthan solutions.1. A rodlike low molecular weight sample. Macromolecules 35:2212–2222. https://doi.org/10.1021/ma011526m

Liao H, Ai W, Zhang K, Nakauma M, Funami T, Fang Y, Nishinari K, Draget KI, Phillips GO (2015) Mechanisms of oligoguluronate modulating the calcium-induced gelation of alginate. Polymer 74:166–175. https://doi.org/10.1016/j.polymer.2015.08.007

Liu AJ, Nagel SR (1998) Jamming is not just cool any more. Nature 396:21–22. https://doi.org/10.1038/23819

Lozinsky VI, Okay O (2014) Basic principles of cryotropic gelation. In: Oaky O (ed) Polymeric cryogels-macroporous gels with remarkable properties. Springer International Publishing, Heidelberg, pp 49–101. https://doi.org/10.1007/978-3-319-05846-7_2

Mao B, Divoux T, Snabre P (2017) Impact of saccharides on the drying kinetics of agarose gels measured by *In-situ* interferometry. Sci Rep 7:41185. https://doi.org/10.1038/srep41185

Matsumoto T, Inoue H (1993) Analysis of a novel phenomenon in a solidlike structure in ovalbumin aqueous colloids using the Yukawa potential. J Appl Phys 74:2415–2419. https://doi.org/10.1063/1.355323

McAllister JW, Schmidt PW, Dorfman KD, Lodge TP, Bates FS (2015) Thermodynamics of aqueous methylcellulose solutions. Macromolecules 48:7205–7215. https://doi.org/10.1021/acs.macromol.5b01544

McClements DJ (2017) Designing biopolymer microgels to encapsulate, protect and deliver bioactive components: physicochem aspects. Adv Colloid Interf Sci 240:31–59. https://doi.org/10.1016/j.cis.2016.12.005

McGhee JD, von Hippel PH (1974) Theoretical aspects of DNA-protein interactions: co-operative and non-co-operative binding of large ligands to a one-dimensional homogeneous lattice. J Mol Biol 86:469–489. https://doi.org/10.1016/0022-2836(74)90031-X

Michon C, Cuvelier G, Relkin P, Launay B (1997) Influence of thermal history on the stability of gelatin gels. Int J Biol Macromol 20: 259–264. doi: 10.1016/S0141-8130(97)00024-X

Milas M, Rinaudo M (1996) The gellan sol-gel transition. Carbohydr Polym 30:177–184. https://doi.org/10.1016/S0144-8617(96)00090-2

Miwa M, Nakao Y, Nara K (1994) Food applications of curdlan. In: Nishinari K, Doi E (eds) Food hydrocolloids: structures, properties, and functions. Plenum Press, New York, pp 307–312

Miyoshi E, Nishinari K (1999) Rheological and thermal properties near the sol-gel transition of gellan gum aqueous solutions. Progress Colloid Polym Sci 114:68–82. https://doi.org/10.1007/3-540-48349-7_11

Miyoshi E, Takaya T, Nishinari K (1994) Gel-sol transition in gellan gum solutions. I. Rheological studies on the effects of salts. Food Hydrocollois 8:505–527. https://doi.org/10.1016/S0268-005X(09)80062-3

Mizrahi S (2010) 11. Syneresis in food gels and its implications for food quality. In: Skibsted LH, Risbo J, Andersen ML (eds) Chemical deterioration and physical instability of food and beverages. Woodhead Publication, Oxford, pp 324–348

Moakes RJA, Sullo A, Norton IT (2015) Preparation and characterisation of whey protein fluid gels: the effects of shear and thermal history. Food Hydrocoll 45:227–235. https://doi.org/10.1016/j.foodhyd.2014.11.024

Mohammed ZH, Hember MWN, Richardson RK, Morris ER (1998) Kinetic and equilibrium processes in the formation and melting of agarose gels. Carbohydr Polym 36:15–26. https://doi.org/10.1016/S0144-8617(98)00011-3

Morita T, Narita T, Mukai S, Tanagisawa M, Tokita M (2013) Phase behaviors of agarose gel. AIP Adv 3:042128. https://doi.org/10.1063/1.4802968

Moritaka H, Yamanaka K, Kobayashi N, Ishihara M, Nishinari K (2019) Effects of the gel size before ingestion and Agarose molecular weight on the textural properties of a gel bolus. Food Hydrocoll 89:892–900. https://doi.org/10.1016/j.foodhyd.2018.11.043

Morris ER (1994) Polysaccharide synergism - more questions than answers? In: Harding SE, Hill SE, Mitchell JR (eds) Biopolymer mixtures. Nottingham University Press, Nottingham, pp 247–288

Morris ER, Nishinari K, Rinaudo M (2012) Gelation of gellan - a review. Food Hydrocoll 28:373–411. https://doi.org/10.1016/j.foodhyd.2012.01.004

Morris VJ (1986) Multicomponent gels. In: Philips GO, Wedlock DJ, Williams PA (eds) Gums and stabilisers for the food industry, 3. Elsevier, London, pp 87–99

Morris VJ, Gurming AP, Kirby AR, Mackie AR, Wilde PJ (2000) Viewing biopolymer networks, their formation and breakdown by AFM. In: Nishinari K (ed) Hydrocolloids, part l: physical chemistry and industrial application of gels, polysaccharides, and proteins. Elsevier, Amsterdam, pp 99–109. https://doi.org/10.1016/B978-044450178-3/50012-3

Moschakis T, Panagiotopoulou E, Katsanidis E (2016) Sunflower oil organogels and organogel-in-water emulsions (part I): microstructure and mechanical properties. LWT Food Sci Technol 73:153–161. https://doi.org/10.1016/j.lwt.2016.03.004

Murphy R, Farkas B, Jones O (2017) Effect of crosslinking on the physical and chemical properties of β-lactoglobulin (BLG) microgels. J Colloid Interface Sci 505:736–744. https://doi.org/10.1016/j.jcis.2017.06.061

Murphy R, Zhu L, Narsimhan G, Jones OG (2018) Impacts of size and deformability of β-lactoglobulin microgels on the colloidal stability and volatile flavor release of microgel-stabilized emulsions. Gels 4:79. https://doi.org/10.3390/gels4030079

Nagano T, Akasaka T, Nishinari K (1994a) Dynamic viscoelastic properties of glycinin and β-conglycinin gels from soybeans. Biopolymers 34:1303–1309. https://doi.org/10.1002/bip.360341003

Nagano T, Mori H, Nishinari K (1994b) Effect of heating and cooling on the gelation kinetics of 7S globulin from soybeans. J Agric Food Chem 42:1415–1419. https://doi.org/10.1021/jf00043a005

Nagasaka K, Taneya S (2000) Effects of sugars on syneresis of agarose gel. Nippon Shokuhin Kagaku Kogaku Kaishi 47:670–678. https://doi.org/10.3136/nskkk.47.670

Nakao Y, Yamaguchi T, Taguchi T (1994) Preparations of freezable processed tofu and freeze-dried tofu by using curdlan. Nippon Shokuhin Kogyo Gakkaishi 41:141–147. https://doi.org/10.3136/nskkk1962.41.141

Nan B, Galindo-Rosales FJ, Ferreira JMF (2020) 3D printing vertically: direct ink writing free-standing pillar arrays. Mater Today 35:16–24. https://doi.org/10.1016/j.mattod.2020.01.003

Nemoto N (2000) Structure and dynamics of ovalbumin gels. In: Nishinari K (ed) Hydrocolloids - Part 1 physical chemistry and industrial application of gels, polysaccharides, and proteins.

Elsevier Science B.V, Amsterdam, pp 45–52. https://doi.org/10.1016/b978-0-444-50178-3.x5000-8
Nemoto N, Koike A, Osaki K, Koski T, Doi E (1993) Dynamic light scattering of aqueous solutions of linear aggregates induced by thermal denaturation of ovalbumin. Biopolymers 33:551–559. https://doi.org/10.1002/bip.360330405
Nicolai T (2019) Gelation of food protein-protein mixtures. Adv Colloid Interf Sci 270:147–164. https://doi.org/10.1016/j.cis.2019.06.006
Nicolai T, Britten M, Schmitt C (2011) β-lactoglobulin and WPI aggregates: formation, structure and applications. Food Hydrocoll 25: 1945–1962. doi: https://doi.org/10.1016/j.foodhyd.2011.02.006
Nieuwland M, Bouwman WG, Pouvreau L, Martin AH, de Jongh HHJ (2016) Relating water holding of ovalbumin gels to aggregate structure. Food Hydrocoll 52:87–94. https://doi.org/10.1016/j.foodhyd.2015.06.018
Nijenhuis KT (1981) Investigation into the ageing process in gels of gelatin/water systems by the measurement of their dynamic moduli part I – phenomenology. Colloid Polym Sci 259:522–535. https://doi.org/10.1007/BF01397890
Nijenhuis KT (1997) Thermoreversible networks. Viscoelastic properties and structure of gels. Adv Polym Sci 130:160–193. https://doi.org/10.1007/BFb0008699
Nijenhuis KT, Winter HH (1989) Mechanical properties at the gel point of a crystallizing poly (vinyl chloride) solution. Macromolecules 22:411–414. https://doi.org/10.1021/ma00191a074
Niki R, Kohyama K, Sano Y, Nishinari K (1994) Rheological study on the rennet-induced gelation of casein micelles with different sizes. Polym Gels Networks 2:105–118. https://doi.org/10.1016/0966-7822(94)90030-2
Nishinari K (1976) Longitudinal vibrations of high-elastic gels as a method for determining viscoelastic constants. Jpn J Appl Phys 15:1263–1270. https://doi.org/10.1143/JJAP.15.1263
Nishinari K (1988) Food hydrocolloids in Japan. In: Phillips GO, Wedlock DJ, Williams PA (eds) Gums and stabilisers for the food industry 4. IRL Press, Oxford, pp 373–390
Nishinari K (1997) Rheological and DSC study of sol-gel transition in aqueous dispersions of industrially important polymers and colloids. Colloid Polym Sci 275:1093–1107. https://doi.org/10.1007/s003960050189
Nishinari K (2000a) Rheology of physical gels and gelling processes. Rep Progress Polym Phys Jpn 43:163–192
Nishinari K (2000b) Sol-gel transition of biopolymer dispersions. Macromol Symp 159:205–214. https://doi.org/10.1002/1521-3900(200010)159:1<205::AID-MASY205>3.0.CO;2-B
Nishinari K (2001) Gel-sol transition in biopolymers. Trans Mater Res Soc Jpn 26:541–544
Nishinari K (2009) Some thoughts on the definition of a gel. Progr Colloid Polym Sci 136:87–94. https://doi.org/10.1007/978-3-642-00865-8_12
Nishinari K (2020) Textural characteristics of world foods. Wiley
Nishinari K, Fang Y (2016) Sucrose release from polysaccharide gels - a review. Food Funct 7:2130–2146. https://doi.org/10.1039/C5FO01400J
Nishinari K, Fang Y (2017) Relation between structure and rheological and thermal properties of agar. A mini-review on the effect of alkali treatment and the role of Agaropectin. Food Struct 13:24–34. https://doi.org/10.1016/j.foostr.2016.10.003
Nishinari K, Fang Y (2021) Molar mass effect in food and health. Food Hydrocoloids 112:106110. https://doi.org/10.1016/j.foodhyd.2020.106110
Nishinari K, Fang Y, Guo S, Phillips GO (2014) Soy proteins: a review on composition, aggregation and emulsification. Food Hydrocoll 39:301–318. https://doi.org/10.1016/j.foodhyd.2014.01.013
Nishinari K, Fang Y, Nagano T, Guo S, Wang R (2018) Soy as a food ingredient. In: Yada RY (ed) Proteins in food processing, 2nd edn. Woodhead Publishing, Elsevier. https://doi.org/10.1016/B978-0-08-100722-8.00007-3
Nishinari K, Ishihara S, Hori K, Fang Y (2020) Tongue-palate squeezing of soft gels in food oral processing. Trends Food Sci Technol 99:117–132. https://doi.org/10.1016/j.tifs.2020.02.023

Nishinari K, Kajiwara K, Nagasaki Y, Kaneda I (2016a) Utilization of gels in food, cosmetics and medicine (popular Ed., in Japanese), 245 p, CMC Publ, Tokyo

Nishinari K, Koide S, Ogino K (1985) On the temperature dependence of elasticity of thermoreversible gels. J de Phys 46:793–797. https://doi.org/10.1051/jphys:01985004605079300

Nishinari K, Koide S, Williams PA, Phillips GO (1990) A zipper model approach to the Thermoreversible gel-sol transition. J de Phys (France) 51:1759–1768. https://doi.org/10.1051/jphys:0199000510160175900

Nishinari K, Nitta Y, Miyoshi E, Ikeda S, Takaya T (2001) Helix-coil transition in the thermoreversible gels. In: Williams PA, Phillips GO (eds) Gums & stabilisers for the food industry 11. Royal Society of Chemistry, pp 85–94

Nishinari K, Shiba K (2007) Polysaccharides from red seaweeds. In: Nishinari K (ed) Food hydrocolloids: development and applications. CMC Publ, Tokyo, pp 196–211. (In Japanese)

Nishinari K, Takahashi R (2003) Interaction in polysaccharide solutions and gels. Curr Opin Colloid Interface Sci 8:396–400. https://doi.org/10.1016/S1359-0294(03)00099-2

Nishinari K, Watase M (1993) Temperature dependence of elastic modulus of agarose gels with different molecular weights. Rep Progress Polym Phys Jpn 36:661–664

Nishinari K, Watase M, Tanaka F (1996) Structure of junction zones in poly(vinyl alcohol) gels by rheological and thermal studies. J Chim Phys Physicochim Biol 93:880–886. https://doi.org/10.1051/jcp/1996930880

Nishinari K, Watase M, Ogino K (1984) On the temperature dependence of elasticity of agarose gels. Die Makromolekulare Chem 185:2663–2668. https://doi.org/10.1002/macp.1984.021851216

Nishinari K, Zhang H (2004) Recent advances in the understanding of heat set gelling polysaccharides. Trends Food Sci Technol 15:305–312. https://doi.org/10.1016/j.tifs.2003.05.001

Nishinari K, Zhang H, Ikeda S (2000) Hydrocolloid gels of polysaccharides and proteins. Curr Opin Colloid Interface Sci 5:195–201. https://doi.org/10.1016/S1359-0294(00)00053-4

Nitta Y, Fang Y, Takemasa M, Nishinari K (2004) Gelation of Xyloglucan by addition of epigallocatechin gallate as studied by rheology and differential scanning Calorimetry. Biomacromolecules 5:1206–1213. https://doi.org/10.1021/bm034526y

Nitta Y, Ikeda S, Takaya T, Nishinari K (2001) Helix-coil transition in gellan gum gels. Trans MRS-J 26(2):621–624

Nitta Y, Takahashi R, Nishinari K (2010) Viscoelasticity and phase separation of aqueous Na-type Gellan solution. Biomacromolecules 11:187–191. https://doi.org/10.1021/bm901063k

Nitta Y, Yoshimura M, Nishinari K (2014) The effect of thermal history on the elasticity of K-type Gellan gels. Carbohydrate Polym 113:189–193. https://doi.org/10.1016/j.carbpol.2014.06.084

Norton IT, Jarvis DA, Foster TJ (1999) A molecular model for the formation and properties of fluid gels. Int J Biol Macromol 26:255–261. https://doi.org/10.1016/S0141-8130(99)00091-4

Nussinovitch A, Hirashima M (2014) Cooking innovations –using hydrocolloids for thickening, gelling and emulsification. CRC Press

Nyström B, Walderhaug H (1996) Dynamic viscoelasticity of an aqueous system of a poly(ethylene oxide)−poly(propylene oxide)−poly(ethylene oxide) triblock copolymer during gelation. J Phys Chem 100:5433–5439. https://doi.org/10.1021/jp952486p

Ogawa E, Takahashi R, Yajima H, Nishinari K (2006) Effects of molar mass on the coil to Helix transition of sodium-type gellan gums in aqueous solutions. Food Hydrocoll 20:378–385. https://doi.org/10.1016/j.foodhyd.2005.03.016

Okay O (2014) Polymeric Cryogels-macroporous gels with remarkable properties. Springer, Berlin

Okiyama A, Motoki M, Yamanaka S (1992) Bacterial cellulose II. Processing of the gelatinous cellulose for food materials. Food Hydrocoll 6:479–487. https://doi.org/10.1016/S0268-005X(09)80033-7

Okiyama A, Motoki M, Yamanaka S (1993) Bacterial cellulose IV. Application to processed foods. Food Hydrocoll 6:503–511. https://doi.org/10.1016/S0268-005X(09)80074-X

Paques JP, van der Linden E, van Rijn CJM, Sagis LMC (2013) Alginate submicron beads prepared through w/o emulsification and gelation with $CaCl_2$ nanoparticles. Food Hydrocoll 31:428–434. https://doi.org/10.1016/j.foodhyd.2012.11.012

Patel AR, Dewettinck K (2016) Edible oil structuring: an overview and recent updates. Food Funct 7:20–29. https://doi.org/10.1039/C5FO01006C

Patel AR, Schatteman D, Vos D, Lesaffer A, Dewettinck K (2013) Preparation and rheological characterization of shellac oleogels and oleogel-based emulsions. J Colloid Interface Sci 411:114–121. https://doi.org/10.1016/j.jcis.2013.08.039

Phillips GO, Williams PA (eds) (2020) Handbook of hydrocolloids, 3rd edn. Woodhead Publ, Cambridge

Richardson RK, Goycoolea FM (1994) Rheological measurement of κ-carrageenan during gelation. Carbohydr Polym 24:223–225. https://doi.org/10.1016/0144-8617(94)90134-1

Richardson RK, Norton IT (1998) Gelation behavior of concentrated locust bean gum solutions. Macromolecules 31:1575–1583. https://doi.org/10.1021/ma970550q

Rochas C, Rinaudo M, Landry S (1990) Role of the molecular weight on the mechanical properties of kappa carrageenan gels. Carbohydrate Polym 12:255–266. https://doi.org/10.1016/0144-8617(90)90067-3

Roefs SPFM, van Vliet T (1990) Structure of acid casein gels 2. Dynamic measurements and type of interaction forces. Colloids Surf 50:161–175. https://doi.org/10.1016/0166-6622(90)80260-B

Sakakibara CN, Sierakowski MR, Chassenieux C, Nicolai T, de Freitas RA (2017) Xyloglucan gelation induced by enzymatic degalactosylation; kinetics and the effect of the molar mass. Carbohydr Polym 174:517–523. https://doi.org/10.1016/j.carbpol.2017.06.118

San Biagio PL, Bulone D, Emanuele A, Palma-Vittorelli MB, Palma MU (1996a) Spontaneous symmetry-breaking pathways: time-resolved study of agarose gelation. Food Hydrocoll 10:91–97. https://doi.org/10.1016/S0268-005X(96)80059-2

San Biagio PL, Bulone D, Emanuele A, Palma MU (1996b) Self-assembly of biopolymeric structures below the threshold of random cross-link percolation. Biophys J 70:494–499. https://doi.org/10.1016/S0006-3495(96)79595-4

Saunders PR, Ward AG (1955) Mechanical properties of degraded gelatins. Nature 176:26–27. https://doi.org/10.1038/176026a0

Schaefer C, van der Shoot P, Michels JJ (2015) Structuring of polymer solutions upon solvent evaporation. Phys Rev E 91:022602. https://doi.org/10.1103/PhysRevE.91.022602

Shi Z, Zhang Y, Phillips GO, Yang G (2014) Utilization of bacterial cellulose in food. Food Hydrocoll 35:539–545. https://doi.org/10.1016/j.foodhyd.2013.07.012

Shirakawa M, Yamatoya K, Nishinari K (1998a) Tailoring of xyloglucan properties using an enzyme. Food Hydrocoll 12:25–28. https://doi.org/10.1016/S0268-005X(98)00052-6

Shirakawa M, Uno Y, Yamatoya K, Nishinari K (1998b) Rheological and thermal studies on the sol-gel transition of aqueous solutions of enzymatically modified xyloglucan. In: Williams PA, Phillips GO (eds) Gums and stabilisers for the food industry 9. The Royal Society of Chemistry, pp 94–103

Singh A, Auzanneaub F-I, Rogers MA (2017) Advances in edible oleogel technologies – a decade in review. Food Res Int 97:307–317. https://doi.org/10.1016/j.foodres.2017.04.022

Smidsrød O (1974) Molecular basis for some physical properties of alginates in the gel state. Faraday Dis Chem Soc pp 263–274. https://doi.org/10.1039/DC9745700263

Suzuki K, Taniguchi Y, Enomoto T (1972) The effect of pressure on the sol-gel transformation of macromolecules. Bull Chem Soc Jpn 45:336–338. https://doi.org/10.1246/bcsj.45.336

Takemasa M, Nishinari K (2016) Solution structure of molecular associations investigated using NMR for polysaccharides: xanthan/Galactomannan mixtures. J Phys Chem B 120:3027–3037. https://doi.org/10.1021/acs.jpcb.5b11665

Tanaka F (2011) Polymer physics: applications to molecular association and Thermoreversible gelation. Cambridge University Press, Cambridge. https://doi.org/10.1017/CBO9780511975691

Tanaka F, Nishinari K (1996) Junction multiplicity in thermoreversible gelation. Macromolecules 29:3625–3628. https://doi.org/10.1021/ma951577h
Tanaka R, Hatakeyama T, Hatakeyama H (1998) Formation of locust bean gum hydrogel by freezing-thawing. Polym Int 45:118–126. https://doi.org/10.1002/(SICI)1097-0126(199801)45:1<118::AID-PI908>3.0.CO;2-T
Tanaka S, Nishinari K (2007) Unassociated molecular chains in physically crosslinked gellan gels. Polym J 39:397403. https://doi.org/10.1295/polymj.PJ2006149
Tobitani A, Ross-Murphy SB (1997) Heat-induced gelation of globular proteins. 1. Model for the effects of time and temperature on the gelation time of BSA gels. Macromolecules 30:4845–4854. https://doi.org/10.1021/ma970112j
Tokita M (1989) Gelation mechanism and percolation. Food Hydrocoll 3:263–274. https://doi.org/10.1016/S0268-005X(89)80038-4
Tokita M, Nishinari K (2009) Gels: structures, properties and functions: fundamentals and applications. In: Progress in colloid and polymer science, vol 136. Springer, Berlin
Ueda T (1985) (QP Co. Ltd.) Jap Pat. 60-83570
Urbonaite V, van der Kaaij S, de Jongh HHJ, Scholten E, Ako K, van der Linden E, Pouvreau L (2016) Relation between gel stiffness and water holding for coarse and fine-stranded protein gels. Food Hydrocoll 56:334–343. https://doi.org/10.1016/j.foodhyd.2015.12.011
van der Linden E (2012) From peptides and proteins to micro-structure mechanics and rheological properties of fibril systems. Food Hydrocoll 26:421–426. https://doi.org/10.1016/j.foodhyd.2010.11.019
van Kleef FSM (1986) Thermally induced protein gelation: gelation and rheological characterization of highly concentrated ovalbumin and soybean protein gels. Biopolymers 25:31–59. https://doi.org/10.1002/bip.360250105
van Vliet T, Walstra P (1985) Note on the shear modulus of rennet-induced milk gels. Neth Milk Dairy J 39:115–118
van Vliet T, Walstra P (1995) Large deformation and fracture behaviour of gels. Faraday Dis Chem Soc Gels 101:359–370. https://doi.org/10.1039/FD9950100359
Veerman C, Ruis H, Sagis LMC, van der Linden E (2002) Effect of electrostatic interactions on the percolation concentration of fibrillar β-lactoglobulin gels. Biomacromolecules 3:869–873. https://doi.org/10.1021/bm025533+
Walstra P (2003) Physical chemistry of foods. Marcel Dekker, New York
Watase M, Nishinari K (1983) Rheological properties of agarose gels with different molecular weights. Rheol Acta 22:580–587. https://doi.org/10.1007/BF01351404
Watase M, Nishinari K (1987a) Rheological and thermal properties of carrageenan gels. Die Makromolekulare Chem 188:2213–2221. https://doi.org/10.1002/macp.1987.021880918
Watase M, Nishinari K (1987b) Dynamic viscoelasticity and anomalous thermal behaviour of concentrated agarose gels. Die Makromolekulare Chem 188:1177–1186. https://doi.org/10.1002/macp.1987.021880520
Weiss RG, Terech P (2006) Molecular gels. Materials with self-assembled fibrillar networks. Springer, Dordrecht
Winter HH (2016) Gel point. In: Peterca M (ed) Encyclopedia of polymer science and technology. Wiley
Winter H, Chambon F (1986) Analysis of linear viscoelasticity of a crosslinking polymer at the gel point. J Rheol 30:367–382. https://doi.org/10.1122/1.549853
Wu X, Nishinari K, Gao Z, Zhao M, Zhang K, Fang Y, Phillips GO, Jiang F (2016) Gelation of β-lactoglobulin and its fibrils in the presence of transglutaminase. Food Hydrocoll 52:942–951. https://doi.org/10.1016/j.foodhyd.2015.09.012
Yan Y, Takemasa M, Zhao C, Yu L, Nishinari K (2016) Structure-gelation research on gallate analogs and xyloglucan by rheology, thermal analysis and NMR. Food Hydrocoll 52:447–459. https://doi.org/10.1016/j.foodhyd.2015.07.012

Yang K, Wang Z, Brenner T, Kikuzaki H, Fang Y, Nishinari K (2015) Sucrose release from agar gels: effects of dissolution order and the network inhomogeneity. Food Hydrocoll 43:100–106. https://doi.org/10.1016/j.foodhyd.2014.05.005

Yin Y, Nishinari K, Zhang H, Funami T (2006) A novel liquid crystalline phase in dilute aqueous solutions of methyl cellulose. Macromol Rapid Commun 27:971–975. https://doi.org/10.1002/marc.200600099

Yoshimura M, Nishinari K (1999) Dynamic viscoelastic study on the gelation of konjac glucomannan with different molecular weights. Food Hydrocoll 13:227–233. https://doi.org/10.1016/S0268-005X(99)00003-X

Yuguchi Y, Urakawa H, Kitamura S, Ohno S, Kajiwara K (1995) Gelation mechanism of methylhydroxypropylcellulose in aqueous solution. Food Hydrocoll 9:173–179. https://doi.org/10.1016/S0268-005X(09)80213-0

Zetzl AK, Gravelle AJ, Kurylowicz M, Dutcher J, Barbut S, Marangoni A (2014) Microstructure of ethylcellulose oleogels and its relationship to mechanical properties. Food Struct 2(1–2):27–40. https://doi.org/10.1016/j.foostr.2014.07.002

Zhang H, Yoshimura M, Nishinari K, Williams MAK, Foster TJ, Norton IT (2001) Gelation behaviour of konjac glucomannan with different molecular weights. Biopolymers 59:38–50. https://doi.org/10.1002/1097-0282(200107)59:1<38::AID-BIP1004>3.0.CO;2-A

Zhang H, Zhang F, Wu J (2013) Physically crosslinked hydrogels from polysaccharides prepared by freeze–thaw technique. React Funct Polym 73:923–928. https://doi.org/10.1016/j.reactfunctpolym.2012.12.014

Zhuang X, Jiang X, Zhou H, Chen Y, Zhao Y, Yang H, Zhou G (2020) Insight into the mechanism of physicochemical influence by three polysaccharides on myofibrillar protein gelation. Carbohydr Polym 229:115449. https://doi.org/10.1016/j.carbpol.2019.115449