

Kinetic Study of Thermally Induced Inulin Gel

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ABSTRACT: Heated inulin solution undergoes a sol-gel transition during cooling and forms a white smooth gel under optimized conditions. The degree of gel formation is negatively related to hydrolysis of inulin during heating. Heating makes inulin soluble; overheating causes hydrolysis of dissolved inulin into smaller molecules. Using a 2-step model including a solubilization step and a hydrolysis of inulin step, we found that both steps follow pseudo first-order kinetics. Separate studies on initial rates of the solubilization and hydrolysis of inulin steps at constant temperatures show that the solubilization rate of inulin is much faster than that of the hydrolysis of inulin. Gel formation of inulin may be related to the average chain length of inulin after heating.

Keywords: inulin, gel, kinetics, solubilization, hydrolysis

Introduction

NOWADAYS, WITH GROWING ATTENTION ON DIETS WITH healthy foods, people are more interested in eating low-fat or low-caloric foods than in the past. According to the chemical nature of fat substitutes, we can categorize them as fat-, carbohydrate-, and protein-based substitutes (Iyengar and Gross 1991). Among carbohydrate-based fat replacers, starch (Harkema 1996), microcrystalline cellulose (Humpherys 1996), and maltodextrin (Roller 1996) have been studied. Yackel and Cox (1992) and Cho (1999) have reviewed the examples of starch-based and carbohydrate-based fat substitutes. Using carbohydrates as fat substitutes, the formation of gel structure is an important attribute for making fat mimetic texture.

Apart from starch-based materials, inulin is another good candidate for carbohydrate-based fat substitute since it generates relatively low calories when it is metabolized (Roberfroid 1993). In terms of chemical structure, inulin consists of a long chain made up of 22 to 50 fructose molecules connected by β -(2-1) bonds and 1 glucose molecule at one end (Niness 1999). From a nutritional point of view, inulin, a dietary fiber as well as a prebiotic food material, exhibits several physiological benefits to humans such as reducing lipid level and enhancing the growth of *Bifidobacterium* species inside the colon (Havenaar and others 1999).

So far, most works on inulin have been done from nutritional or sensory points of view (Roberfroid 1999; Schaller-Povolny and Smith 1999). Very little has been reported on the physical requirement of using inulin as a fat substitute from an engineering point of view. High shearing process has been employed to make the fat mimetic gel structure from inulin powders (Leenheer 1993). Kim and others (2001) have found that thermal treatment of an inulin-water mixture followed by cooling can also make a gel. They showed that inulin concentration, heating temperature, and pH affected the degree of gel formation. They also found that gel strength of inulin is a function of particle size of inulin made through different processing history. Furthermore, they postulated that after inulin was dissolved, inulin could be hydrolyzed into smaller molecules leading to decreased degrees of gel formation.

Gelation is a common characteristic of synthetic and nat-

ural polymers. Generally speaking, gel formation (gelation) of food materials can be represented by sol-gel transition of polysaccharides or proteins. Gels made from biopolymers, including food gels, can be classified into either associative gel or particulate gel according to the mechanism of networking between polymer molecules (Clark 1996).

Most kinetic studies of food gels have been done with phenomenological changes like rheological change (Sarkar 1995; Aguilera and Rojas 1997; Böhm and Kulicke 1999) during gelation. As for kinetic models reported for the gelation of polymer, there are simple stochastic models (Marangoni and others 2000), networking/cross-linking models (Pauletti and others 1996), recrystallization models (Böhm and Kulicke 1999), and coagulation models (Somvársky and others 1998).

So far, the kinetics of inulin gel formation has not been reported yet. The main objective of this research is to study the kinetics of gel formation of inulin through thermal processes (heating followed by cooling) to facilitate the optimization of gel formation of inulin and the use of the gel as a fat substitute in the future.

Materials and Methods

Materials

Inulin (Raftilin® HP) from Orafit (Malvern, Pa., U.S.A.) was used throughout the experiments. According to the company (Niness 1999), Raftilin® HP was extracted from chicory root, and its average degree of polymerization is 25. The average particle size of inulin is $30 \pm 5 \mu\text{m}$ based on our measurement under a microscope (Kim and others 2001).

Thermally Induced Gel Formation of Inulin

First, a specified amount of inulin powder was dissolved in water at a particular temperature with stirring (150 RPM in a beaker). The dissolved inulin was poured into a volumetric flask and made up to 100 mL. After mixing it again, the inulin solution was poured into a bottle and allowed to cool down for a d at 25 °C for the gel structure to develop.

Volumetric Gel Index (VGI)

The volumetric gel index (VGI) is expressed as the volume

of gel (V_G) over the total volume (V_T) multiplied by 100. We used VGI as a parameter for optimizing the formation of inulin gel.

Measurement of Reducing Sugar

The concentrations of soluble reducing sugars were measured using a 3,5-dinitrosalicylic acid (DNS) method (Chaplin 1986). Immediately after heating, the solution was diluted with water 10 times, and 0.2 mL of the diluted sample was transferred to a test tube where it was mixed with 2 mL of the DNS reagent. After mixing it with a vortex, the samples were heated in the water bath at 100 °C for 10 min followed by rapid cooling in iced water. The optical densities (OD) of the samples were measured at 570 nm using a Bausch & Lomb (Rochester, N.Y., U.S.A.) spectrophotometer 601. Fructose was used for the establishment of a standard curve.

Kinetic Study on Solubilization During Heating (Measurement of the Rate of Solubilization of Inulin During Heating)

During heating of a specified concentration of inulin-water mixture at 70, 80, and 90 °C, a sample each was taken at 10, 20, 30, 40, and 50 s and 1, 2, 3, 4, and 5 min of heating and then filtered with Whatman No. 4 filter paper. The inulin on the filter was dried at 105 °C and weighted to calculate the degree of solubilization during heating. Log value of amounts of insoluble inulin during heating were plotted against heating time to show the degree of solubilization with respect to time. To minimize possible thermal lag during transfer of water and samples, the beaker and the water were preheated at a specific experimental temperature in an extra water bath before starting solubilization kinetic studies. During the experiment, temperature inside the solution was monitored with a thermocouple, which indicated that the temperature stayed at a desired temperature throughout our experiments.

Kinetic Study on Hydrolysis of Inulin During Heating

The rates of hydrolysis of inulin at 70, 80, and 90 °C were studied based on measured reducing sugar contents in inulin solutions after heating. The reaction rate constant and the reaction order for hydrolysis of inulin were calculated using the kinetic equations described in the discussion section.

Development of a 2-Step Model for Thermally Induced Inulin Gel

Kim and others (2001) have found that, with relatively low shearing at 250 RPM in a beaker, inulin suspensions with 5%

to 25% (w/v) concentrations could not form 100% gel. But for thermally induced gel, they can make a 100% inulin gel even with a 10% (w/v) solution. This fact suggests that there are at least 2 different paths that can be followed for inulin gel formation, that is, with shearing or with heating and followed by cooling.

Now we consider developing the kinetic model for the thermally induced gel formation in this paper. Kim and others (2001) also found that solubilization is the preliminary step that is required for forming the thermally induced gel and that hydrolysis occurring during heating is negatively affecting gel formation.

Based on these 2 observations, we are proposing a model with 2 steps as shown in Figure 1. The main steps of this study are boxed in Figure 1. According to this model, inulin powder dissolves into the gel-forming components (solubilized inulin), and then it can be hydrolyzed into smaller fragments with further heating, which do not form gel. We also assume that both steps are irreversible under the experimental condition studied. Once the inulin molecules are separated from one another after dissolution with the help of thermal energy, they could form a gel network through association among the molecular chains during cooling. But the non-gel-forming components (smaller molecules) would remain in the liquid portion at least partly without association with the gel structure.

For a 2-step model, referring to Figure 1, the rate expressions for the degradation of inulin and generation of gel-forming components and non-gel-forming components can be expressed using the following system of equations:

$$-\frac{dC_i}{dt} = k_1 C_i^{n_1} \quad (1)$$

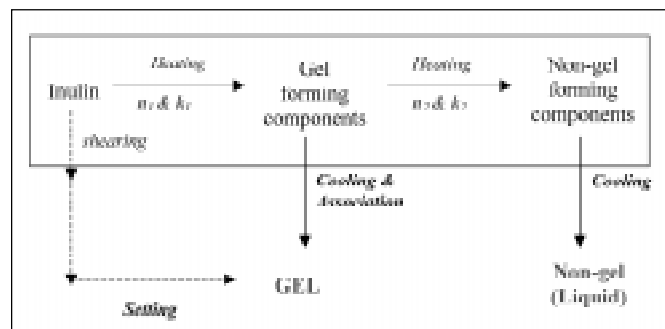


Figure 1—A model for the formation of thermal-induced inulin gel

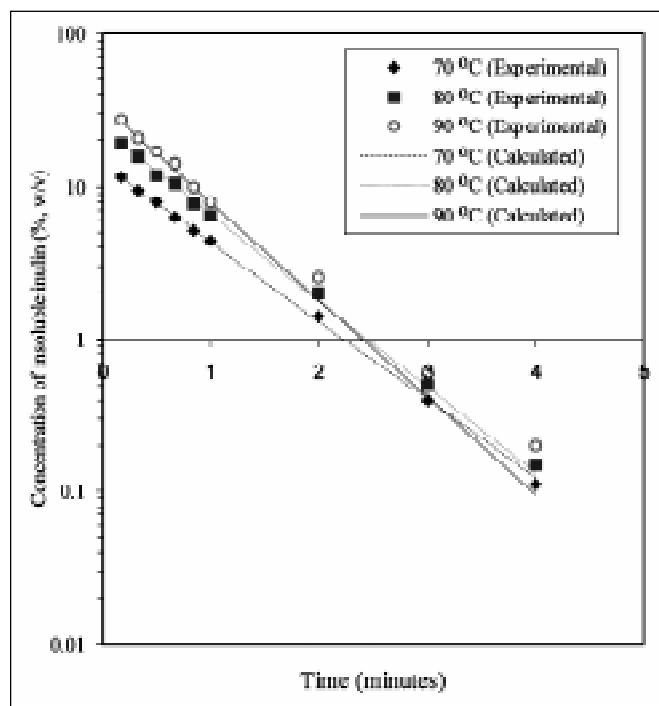


Figure 2—Kinetics on the solubilization of inulin at 70, 80, and 90 °C

$$\frac{dC_G}{dt} = k_1 C_I^{n_1} - k_2 C_G^{n_2} \quad (2)$$

$$\frac{dC_{NG}}{dt} = k_2 C_G^{n_2} \quad (3)$$

where C is the concentration; t, the reaction time; k, the reaction rate constant; and n, the reaction order. Subscript I denotes inulin; G, gel-forming component; NG, non-gel-forming component; and 1 and 2, the 1st and 2nd reactions, respectively, in Figure 1. Some modifications of system equations are discussed in the Results and Discussion section. As mentioned in the introduction, most kinetic studies have focused on gel formation during cooling. In this study, we divert the angle of research to be more on the thermal changes of inulin during heating, which would essentially affect the formation of gel.

Calculation of Kinetic Parameters

Experimental data from separate kinetic studies of solubilization and hydrolysis of inulin were fitted with system kinetic equations. Computer software called "origin" from Origin Lab (Northampton, Mass., U.S.A.) was used for the calculation of these kinetic parameters (the reaction rate constant and the reaction order).

Computer Simulation of the Concentrations of Various Components During Heating

Computer simulation using software called "polymath" was performed to model the trend of the concentrations of gel-forming and non-gel-forming components. Details of the computer simulation are described below.

Results and Discussion

Kinetic Studies on Solubilization of Inulin

In the model proposed in Figure 1, we need to pay special attention to Eq. 1 carefully. Dissolution of solute in solvent is often studied using a mass transfer or diffusion model. In both models, mass transfer constant is dependent on the

speed of mixing (stirring). In this study, we used a constant rate of stirring in a standard container (150 RPM in a beaker) to avoid variation of solubilization rate with mixing. We then assume that the apparent rate constant (k_1) of solubilization of inulin is influenced mainly by temperature of heating under our experimental conditions. In essence, Eq. 1 states that the rate of dissolution of inulin powder is proportional to the rate constant multiplied by the concentration of the powder raised to the n th power. We also used the powder that has the same average particle size (30 μm) to avoid variation of solubilization rate due to differences in the surface to volume ratio of the powders.

Figure 2 is a semi-log plot of the amount of insolubles during the solubilization step within 4 min of heating. Kim and others (2001) reported that water solubility of inulin at 70, 80, and 90 °C are 14%, 24%, and 34% (w/v), respectively. Figure 2 shows the kinetics of dissolution of inulin powder in water at different temperatures. These curves show that saturation is reached quickly within 3 to 4 min of heating. Data were fitted with Eq. 1 to determine the reaction rate constant and reaction order.

Table 1 shows the results of curve fitting of the solubilization data at 70, 80, and 90 °C. The table consists of 2 parts. First, the best fitting shows the result of curve fitting using a computer. The reaction order of solubilization at 70, 80, and 90 °C follows 1.01, 0.93, and 0.96 order, respectively. The reaction rate constants are 1.11 (g/mL)^{-0.01} min⁻¹, 1.49 (g/mL)^{0.07} min⁻¹, and 1.61 (g/mL)^{0.04} min⁻¹ at 70, 80, and 90 °C, respectively. The differences among these calculated reaction orders may not come from the nature of reaction but from combinations of experimental errors. Generally speaking, elementary reactions having the same mechanism should have the same reaction order, even though reaction order is an experimentally obtained number. Fractional orders have been reported by many studies. Nevertheless, comparisons of the reaction rate constants of reactions with different reaction orders are difficult because they have different units. For the convenience of comparing, we need to unify the reaction order with a reasonable reaction order. Since the 3 reaction orders obtained from computer fitting in Table 1 are all close to 1, we concluded that the solubilization process of inulin follows a pseudo first-order reaction.

The second part of Table 1 shows the recalculated reaction rate constants assuming that the solubilization process follows the pseudo first-order kinetics. The calculated reaction rate constants for the process are 1.18, 1.30, and 1.47 min⁻¹ at 70, 80, and 90 °C, respectively. As expected, by assuming a first-order kinetic to the data, smaller correlation coefficients (R^2) are obtained. These reaction rate constants under the assumption of pseudo first-order reaction are used for further simulation studies of inulin gel formation.

Kinetic Studies on Hydrolysis of Inulin

Figure 3 shows the changes of reducing sugar content in inulin solution with heating time. Dissolution of inulin in water was almost completed within 3 to 4 min of heating as shown in Figure 2. But as seen in Figure 3, the reducing sugar content in solution kept increasing continuously with heating without leveling even after 2 h of heating.

Experimentally measuring the degrees of conversion of gel-forming components into non-gel-forming components is not an easy task because non-gel-forming components may consist of various molecules with different degrees of polymerization (DP). However, it is fair to say that an increase in the non-gel-forming component is related to the extent of hy-

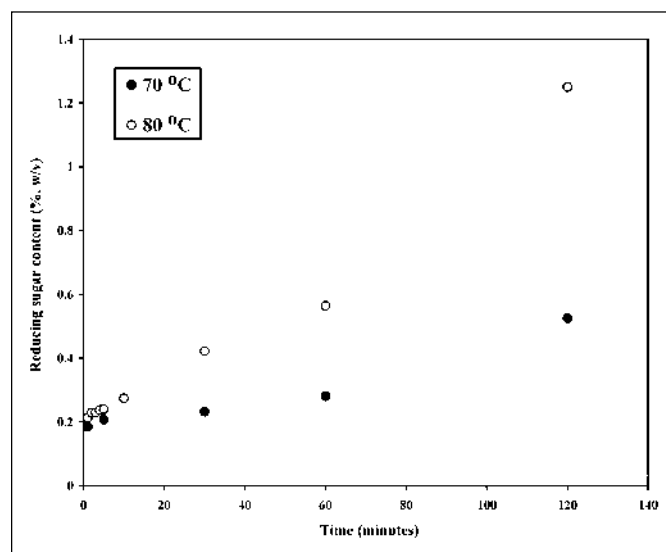


Figure 3—Reducing sugar contents with heating of dissolved inulin at 70 and 80 °C

Table 1—The best curve fitting of initial solubilization rate with Eq. 1 and the reaction constant from curve fitting of initial solubilization rate with the pseudo 1st-order kinetic model

Temperature (°C)	n*	The best fitting k_1^*	R ²	SD *	The pseudo 1st-order k_1 (min ⁻¹)	R ²
70	1.01	1.11 (g/mL) ^{-0.01} min ⁻¹	0.9970	0.165	1.18	0.9966
80	0.93	1.49 (g/mL) ^{-0.07} min ⁻¹	0.9962	0.170	1.30	0.9935
90	0.96	1.61 (g/mL) ^{-0.04} min ⁻¹	0.9965	0.169	1.47	0.9949

* n and k₁ denote the reaction order and the reaction rate constant, respectively.
** SD stands for standard deviation.

drolysis of inulin during heating. Each hydrolysis reaction causes an increase in soluble reducing sugar content. So in the absence of a direct way of measurement, one of the indirect ways of determining the reaction order for the formation of non-gel-forming components is to measure the reducing sugar contents during heating of inulin solution. Based on this rationale, Eq. 3 can be rewritten as Eq 4.

$$\frac{dC_{\text{reducing sugar}}}{dt} = k_2' C_G^n \tag{4}$$

where

$$k_2' = k_2 \left(\frac{C_{\text{reducing sugar}}}{C_{NG}} \right)$$

Furthermore, because the solubilization of inulin powder into gel-forming components was much faster than that of hydrolysis, the concentration of gel-forming component (C_G) in Eq. 4 can be substituted with the initial concentration of inulin (C_{I0}) during the very early stage of heating. So Eq. 4 can be rewritten as Eq. 5. Also, in all experiments, inulin concentration used for kinetic study was below the saturation concentration at each temperature. So that when all inulin powders are dissolved, C_{I0} = C_G, assuming that very little hydrolysis has occurred in this early stage of the heating process. Referring to Figure 2 and 3, this early stage encompasses the 1st 3 to 5 min of heating.

$$\frac{dC_{\text{reducing sugar}}}{dt} = k_2' C_{I0}^n \text{ (for initial rates)} \tag{5}$$

In other words, the initial reaction rate for the production of reducing sugar is expressed by a product of an apparent reaction rate constant, k₂', and initial inulin concentration in the system. Figure 4 shows the first-order plots of initial rate of hydrolysis (first 5 min), d(the reducing sugar)/dt compared with initial inulin concentration at different temperatures. The slope represents the apparent first-order reaction rate constant, k₂'. As seen in Figure 4, the correlation coefficients (R²s) are all above 0.990.

However, using d(the reducing sugar)/dt instead of the amount of reducing sugar may generate more error in the calculation of kinetic parameters. But this was the only possible way to determine the reaction rate constant under our experimental conditions. So with this limitation on the data analysis, we concluded that the hydrolysis reaction of inulin followed pseudo first-order kinetics at the 3 temperature studies. The apparent reaction rate constants, k₂', for hydrolysis of inulin were 0.0026, 0.0029, and 0.0038 min⁻¹ at 70, 80, and 90 °C, respectively.

As mentioned in the introduction, most kinetic studies on the gelation of food materials are based on the measured rheological changes during phase transition from a solution to 100% gel. With different sets of models and instruments, researchers monitored the rheological changes occurring during the gelation step (Sarkar 1995; Aguilera and Rojas

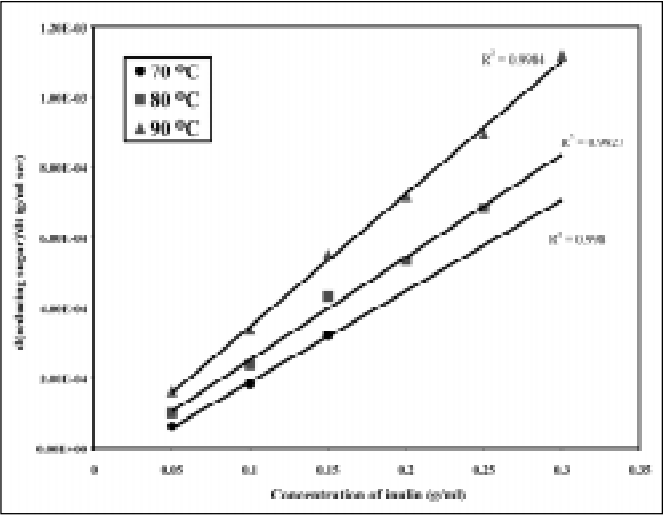
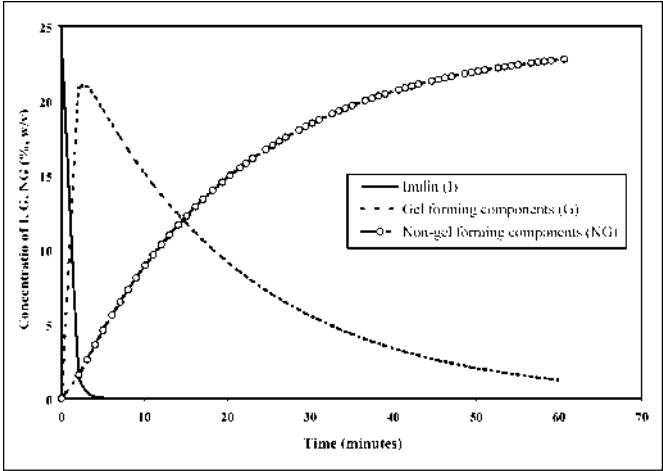


Figure 4—First-order kinetics plots of hydrolysis of inulin at different temperatures



**Figure 5—Simulation of formation of gel-forming and non-gel-forming component during heating; concentrations of I, G, and NG (k₁ = 1.3 min⁻¹, *k₂ = 0.05 min⁻¹, and initial inulin concentration = 24%)
*k₂ value was arbitrarily chosen.**

1997; Böhm and Kulicke 1999).

As for the kinetic studies of gelation, the overall reaction rate parameters were mostly evaluated using a 1-step model. A typical first-order equation (Aguilera and Rojas 1997), nth-order equation (Pauletti and others 1996), and Avrami equation (Böhm and Kulicke 1999) were used for the modeling of gelation of milk protein, whey protein, and β -glucan, respectively. But in our studies, we have found either complete or partial formation of inulin gel according to the inulin concentrations and heating temperatures used (Kim and others 2001). The partial gel formation of inulin leads to phenomenological phase separation of inulin-water mixture into a solid (gel) and a liquid phase (solution complex of non-gel-forming components). This prompted the kinetic studies and the model building reported here. We developed a 2-step model to explain the complete or partial gel formation depending on the processing conditions. These calculated reaction rate constants were used for the computer simulation of gel formation in the next section.

Computer Simulation of Concentration Change of Gel-Forming Components During the Heating Process

The parameter useful for predicting the degree of gel formation of inulin is the concentration of gel-forming components in the system. Computer simulations were performed using software called "polymath" to see the effect of hydrolysis on the gel-forming component.

So far, we have determined the reaction orders ($n_1 = n_2 = 1$) and reaction rate constant (k_1 and k_2) for the proposed 2-step model. Using these parameters, we can model the trends of I, G, and NG with the system Eq. 1, 2, and 3 and polymath software. Figure 5 is an example of the computer simulation showing the trend in concentrations of I, G, and NG during heating and reaction when k_1 and k_2 are chosen as 1.3 min^{-1} and 0.05 min^{-1} , respectively, with 24% (C_{I0}) of inulin. Since k_1 is large, inulin concentration quickly dropped down to 0. Concentration of gel-forming component reached the maximum value in 3 min along the reaction coordinates, but it started to drop with further heating of inulin

solution. Levels of gel-forming and non-gel-forming components are highly dependent on the value of the rate constant of hydrolysis, k_2 . To see how the gel-forming component concentration (C_G) is affected by the hydrolysis rate constant (k_2), 4 arbitrary chosen k_2 values were used ($0.5, 0.05, 0.005$, and 0.0005 min^{-1}) along with 1.3 min^{-1} as the value of k_1 and 24%, the initial inulin concentration. Figure 6 shows the sensitivity of C_G value as influenced by different rates of hydrolysis of inulin. Obviously, as k_2 increases, the level of gel-forming components quickly decreases. In all cases, the maximum values of gel-forming components are reached in less than 5 min of heating. Therefore, heating time should be carefully controlled in inulin gel making.

Effect of Heating Time on Gel Formation

Figures 7a, 7b, and 7c show the effects of heating time on gel formation at different combinations of inulin concentrations and heating temperatures. Each graph shows the experimental volumetric gel indices (circles) and the theoretical concentrations of gel-forming components calculated from computer simulation (solid line). The main purpose of this work is (1) to see the relationship between initial concentration of inulin and heating time for gel formation at a specific temperature and (2) to compare our model with experimental results.

As seen in Figure 7a, experimental results (circles) show that 10% inulin solution started to form gel after 1 min of heating at 70°C . The volumetric gel index (VGI) stayed at 100% for 30 min of heating and dropped to a lower value with further heating. But in Figure 7b with 15% of inulin at 70°C , VGI stayed at 100% even after 1 h of heating. The difference between these 2 experiments is that one has 10% inulin (Figure 7a) and the other 15% inulin (Figure 7b). Therefore, the differences between Figures 7a and 7b are suggesting that the density or concentration of polymer chain with a critical chain length is also an important factor in deciding the VGI. Since we did not include this "density" factor in the model used for simulation, the calculated (simulated) solid line did not show the same pattern as the experimental VGI data in Figure 7a and Figure 7b. In Figure 7c with 15% of inulin at 80°C , VGI only stayed at 100% after about 3 or 4 min of heating. As heating time increased, VGI decreased gradually to reach a value around 30%. In Figure 3, we see that reducing sugar concentration increases with respect to heating time. This is correlating well with the decrease in VGIs as seen in Figure 7c. Again, hydrolysis brings about increases in reducing sugar concentration, decreases in average polymer chain lengths, and also decreases in gel-forming ability. Even at the same concentration of inulin, 2 different trends of volumetric gel index were observed at 70 and 80°C , respectively. We can see that gel formation of inulin is more sensitive to heating time at 80°C (Figure 7c) as compared to that at 70°C (Figure 7b). Higher temperature increases the rate of hydrolysis exponentially according to the Arrhenius equation. These are also shown in Figures 3 and 4. Considering that the same concentration of inulin was used in Figures 7b and 7c, the differences of experimental VGIs at 70 and 80°C are due to different hydrolysis rates at these temperatures. Because of a faster rate of hydrolysis at 80°C , the expected average molecular weight or average DP of inulin chain may be smaller at 80°C as compared to that at 70°C . The smaller average DP of inulin chain may lead to a lower degree of gel formation. Therefore, the differences between Figures 7b and 7c are suggesting the importance of a critical average DP of inulin chain for gel formation.

As reported earlier, a minimal concentration of inulin is

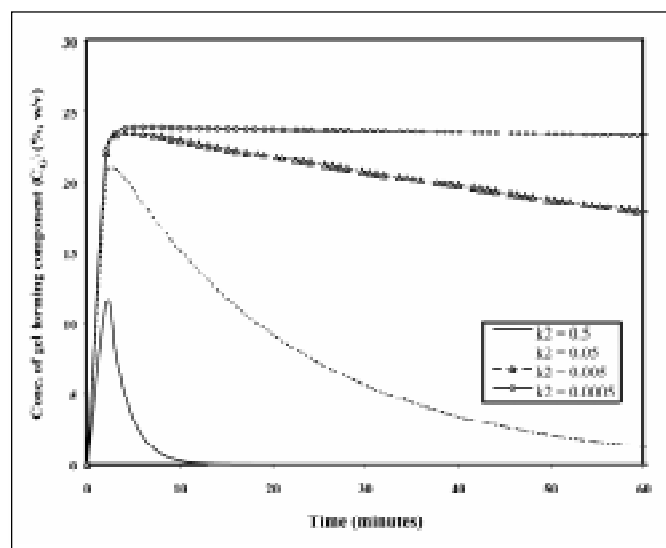


Figure 6—Effect of k_2 on the concentration of gel-forming component during heating

needed to form thermally induced inulin gel through sol-gel transition (Kim and others 2001). In addition to the minimum concentration, the system may also need a minimum

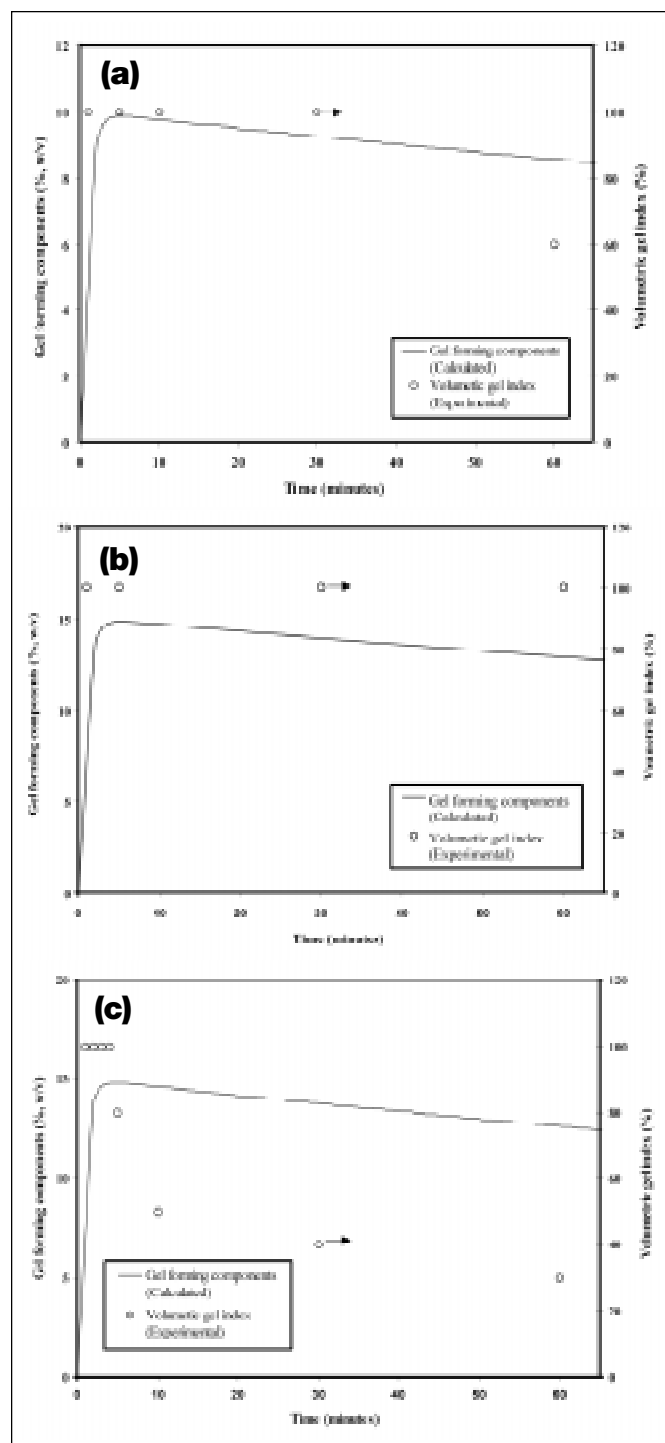


Figure 7—Effect of heating time on the volumetric gel index (comparison between calculated gel-forming component concentration and experimental volumetric gel index) (a) 70 °C with 10% ($k_1 = 1.18 \text{ min}^{-1}$, $k_2 = 0.0026 \text{ min}^{-1}$), (b) 70 °C with 15% ($k_1 = 1.18 \text{ min}^{-1}$, $k_2 = 0.0026 \text{ min}^{-1}$), and (c) 80 °C with 15% inulin ($k_1 = 1.3 \text{ min}^{-1}$, $k_2 = 0.0029 \text{ min}^{-1}$)*
*Values in parentheses are the reaction rate constants used for computer simulation.

length of inulin chain. It is proposed that without a certain minimal length of inulin chain, gel can not be formed. As a simple example, glucose or sucrose solution never forms a gel structure even with very high concentrations and at low temperatures, except that crystallization may happen. At this moment, we do not have the data for the required minimal (critical) chain length or average molecular weight required for gel formation. But based on reducing sugar data and the changes of VGI with heating time, we are proposing that the average molecular weight of inulin in the solution is a very important factor in deciding whether a gel is to be formed or not. The said average molecular weights can be obtained by using HPLC (high performance liquid chromatography) or other analytical methods to analyze the prepared inulin gels. It would also be beneficial to establish the relationships between the average DP in inulin solution and the minimal concentration at each DP for 100% gel formation of inulin for the optimization of gel-making process. By integrating information on this “relationship between concentration and DP” into a 2-step kinetic model, we may predict the degree of gel formation more accurately.

In the model proposed here, we assumed that the formation of gel from inulin solution follows the same kinetics regardless of the heating temperature and the concentration of inulin used and the hydrolyzed products formed during the process. This assumption should be verified with further studies since kinetics of gel formation may be affected by the composition of inulin and by-products produced during heating. So, more kinetic studies of gel formation from inulin solution may also be a key to better prediction of gel formation in computer simulation. Coupling the kinetic model in this paper with other published models could be another way of improving the simulation effort to fit the experimental data even better.

Conclusions

THERMALLY INDUCED GEL FORMATION OF INULIN WAS STUDIED using a 2-step model. Rate of hydrolysis of inulin during heating is a major factor in causing the different degrees of polymerization (DP) of inulin chain to be formed and thus leading to different degrees of gel formation in the inulin-water mixtures. Density or concentration of inulin with a critical chain length may be an important factor affecting thermally induced gel formation of inulin.

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