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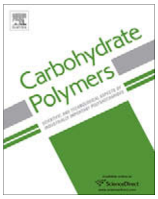

On the applicability of Flory–Huggins theory to ternary starch–water–solute systems

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ABSTRACT

The effects of glucose and glycerol on gelatinization of highly concentrated starch mixtures were investigated with wide-angle X-ray scattering and differential scanning calorimetry. The gelatinization/melting of starch was found to be a two step process. In the first step the granule swells at low temperatures (i.e., 30–50 °C), which is followed by a solvent–temperature cooperative step that induces loss of crystallinity. The results were interpreted with an extended form of the adapted Flory equation. The values of the model parameters (T_m^0, ΔH_u, χ_{12}, χ_{13}, and χ_{23}) obtained were similar to the values reported in the literature. Ternary phase diagrams were constructed with melting lines representing fully gelatinized starch. The crystalline region of starch with glucose was larger than with glycerol. This could be understood from the differences in χ_{13} (solute–solvent interaction). The extended form of Flory–Huggins model somewhat under predicts the experimental values of the gelatinization process. Comparing the Flory–Huggins model with experiments led to the conclusion that Flory equation is a useful tool to interpret and predict the gelatinization and melting behaviour of ternary starch-based systems. But the experiments are complex, the systems are often not in true equilibrium and other disturbing effects are easily encountered. Therefore one should be cautious in the translation of experimental results to the thermodynamics of gelatinization in multicomponent systems.

1. Introduction

Starch is one of the most common and available biopolymers. Starch is used as a food ingredient and as raw material for biodegradable plastics (Rouilly & Rigal, 2002). Granular starch is a partially crystalline, granular solid (i.e., 15–39% crystallinity) (Jenkins & Donald, 1998; Slade & Levine, 1987). It consists of approximately 25% w/w linear amylose and 75% w/w branched amylopectin, which are based on chains of (1 → 4)-linked α-D-glucose (Imberty, Buleon, Tran, & Perez, 1991). When starch is heated above a certain temperature in the presence of excess water (>66% w/w), it undergoes an irreversible order–disorder transition, which is identified as gelatinization (Ratnayake & Jackson, 2007). On the other hand, this transition is known as melting of starch in case of low water conditions. During gelatinization or melting, various changes occur in the starch granules: loss of order, swelling, exudation of amylose, granule disruption, enhanced solubility, and increased viscosity (Ratnayake & Jackson, 2007; Sopade, Halley, & Junming, 2004).

Gelatinization and melting often take place in mixtures containing low-molecular-weight solutes. For example, starch-based plastic materials, known as thermoplastic starch (TPS), are usually produced below the excess of water in the presence of plasticizers such as glycerol using heat and shear (Van Soest, Benes, De Wit, & Vliegenthart, 1996). In the food industry, starch is often gelatinized in mixtures containing sugars, for example in the production of candies, snacks, breakfast cereals, baked goods, and ready-to-eat foods (Eliasson, 2004). Many authors have shown the strong influence of low-molecular-weight solutes on depressing the glass transition temperature, plasticizing the material, and modifying its mechanical properties (Myllarinen, Partanen, Seppälä, & Forssell, 2002; Nashed, Rutgers, & Sopade, 2003; Rodriguez-Gonzalez, Ramsay, & Favis, 2004; Sopade et al., 2004). Moreover, low-molecular-weight solutes affect the melting and gelatinization temperatures (Lelievre, 1976; Nashed et al., 2003; Perry & Donald, 2002; Smits, Kruiskamp, Van Soest, & Vliegenthart, 2003; Tan, Wee, Sopade, & Halley, 2004; Van Soest, Bezemer, De Wit, & Vliegenthart, 1996; Van Soest, de Wit, Tournois, & Vliegenthart, 1994; Van Soest & Knooren, 1997).

To understand and quantify the effect of the plasticizer on the gelatinization and melting of starch from a theoretical point of view, we propose to explore the use of an approach introduced earlier and inspired by (synthetic) polymer science. The analogies between synthetic polymers and biopolymers can be used to predict the functional properties of biopolymers during processing and storage with well-established theories from the field of synthetic

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polymers. For example, the structure of starch granules shows some similarities to that of the spherulites formed in crystalline synthetic polymers. The Flory–Huggins theory is often used to describe crystalline–amorphous phase transitions in polymer–diluent mixtures, and to study the effect of water and solutes on gelatinization and melting of starch granules (Baks, Ngene, Van Soest, Janssen, & Boom, 2007; Donovan, 1979; Donovan, Lorenz, & Kulp, 1983; Donovan & Mapes, 1980; Farhat & Blanshard, 1998; Lelievre, 1976; Moates, Parker, & Ring, 1998; Parker & Ring, 2001; Russell, 1987; Van den Berg, 1981; Whittam, Noel, & Ring, 1990). In spite of the non-equilibrium nature of starch gelatinization (Slade & Levine, 1987), the Flory–Huggins theory applied to the crystalline melting of starch seems important as a means to describe and quantify the effect of low-molecular-weight components in starch gelatinization (Farhat & Blanshard, 1998). Several authors have used the Flory–Huggins theory to model phase diagrams in ternary systems: acetone–cellulose acetate–water (Altena & Smolders, 1982), dioxane–cellulose acetate–water (Altena, Schroder, Van de Huls, & Smolders, 1986), water–dextran–ethanol (Neuchl & Mersmann, 1995), water–dextran–agarose (Clark, 2000), among others.

Most literature use the standard form of the Flory–Huggins theory developed for semi-crystalline polymers. However, a number of starch properties differ from those crystalline polymers; for example, starch contains a large fraction of amorphous material in addition to the crystalline component. Therefore, a distinction between the original amorphous and crystalline fractions is suggested (Whittam et al., 1990). In addition, inclusion of the temperature dependency of the heat of fusion of the polymer repeat unit and the Flory–Huggins interaction parameters improves the predictions. Recently, Baks et al. (2007) reported a first attempt with some of these modifications included. The authors adapted the Flory–Huggins equation to provide a quantitative description of the degree of starch gelatinization in the binary water–starch system as a function of the starch–water ratio and the temperature.

This paper extends the approach of (Baks et al., 2007) to three-component systems (water–starch–solute). An extended form of the Flory–Huggins theory is used to analyze the effect of a low-molecular-weight solute on the gelatinization and the melting of starch mixture, to provide a quantitative description of the dependence of the degree of starch gelatinization on the starch–water–solute composition and the temperature. Since the purpose of this study is to monitor the extent of the gelatinization process at different stages, we made use of offline measurements. The advantages of this approach are that different techniques (i.e., DSC, WAXS, etc.) can be applied to stable materials and kinetics effects can be suppressed.

2. Flory–Huggins theory of melting in polymers

The Flory–Huggins model can be used to describe the equilibrium between a crystalline polymer and a solution of polymer in solvent at temperature T (Flory, 1964). Defining water as component 1, the polymer as component 2, and the low-molecular-weight solute as component 3, the Flory–Huggins model applied to a tertiary system in equilibrium can be expressed as follows:

$$\frac{1}{T_\mathrm{m}}-\frac{1}{T_\mathrm{m}^0}=\left(\frac{R}{\Delta H_\mathrm{u}}\frac{V_\mathrm{u}}{V_1}\right)\left[\phi_1+\left(\frac{\phi_3}{x_3}\right)+\chi_{13}\phi_1\phi_3 - \left[\chi_{12}\phi_1+\chi_{32}\left(\frac{\phi_3}{x_3}\right)\right](\phi_1+\phi_3)\right] \tag{1}$$

where ΔH_u is the heat of fusion of a polymer repeat unit, T is the absolute temperature, T_m^0 is the melting temperature of pure polymer, R is the gas constant, V_1 and V_u are the molar volumes of the diluents and the repeating unit of the polymer; φ_1, φ_2, and φ_3 are the volume fractions; x_3 is the number of segments per molecule of solute, and χ_{12}, χ_{13}, and χ_{32} are the (enthalpic) interaction parameters. The values of the interaction parameters are assumed to be independent of the composition. Eq. (1) was derived assuming that ΔH and ΔS, in the thermodynamic relation $\Delta G = \Delta H - T\Delta S$, are constant with temperature, and equal to ΔH_u and $\Delta H_\mathrm{u}/T_\mathrm{m}^0$, respectively. However, ΔH and ΔS are expected to depend on temperature (Hoffman, 1958). Therefore, we included a correction based on Hoffman's approximation (Hoffman, 1958):

$$\frac{1}{T_\mathrm{m}}-\frac{1}{T_\mathrm{m}^0}=\left(\frac{R}{\Delta H_\mathrm{u}}\frac{V_\mathrm{u}}{V_1}\right)\left(\frac{T_\mathrm{m}^0}{T_\mathrm{m}}\right)\left[\phi_1+\left(\frac{\phi_3}{x_3}\right)+\chi_{13}\phi_1\phi_3 - \left[\chi_{12}\phi_1+\chi_{32}\left(\frac{\phi_3}{x_3}\right)\right](\phi_1+\phi_3)\right] \tag{2}$$

The resulting expression relates the melting temperature to the composition of the liquid surrounding the crystals. This can be rewritten as:

$$T_\mathrm{m}=T_\mathrm{m}^0-\left(\frac{R}{\Delta H_\mathrm{u}}\frac{V_\mathrm{u}}{V_1}\right)\left(T_\mathrm{m}^0\right)^2\left[\phi_1+\left(\frac{\phi_3}{x_3}\right)+\chi_{13}\phi_1\phi_3 - \left[\chi_{12}\phi_1+\chi_{32}\left(\frac{\phi_3}{x_3}\right)\right](\phi_1+\phi_3)\right] \tag{3}$$

We can now derive a relation for the dependence of the degree of crystallization as a function of the temperature. Following a similar procedure to that proposed earlier by Baks et al. (2007) but extended to three components, we define:

The degree of starch crystallinity (α_c):

$$\alpha_\mathrm{c}=\frac{v_2^\mathrm{c}}{v_2^\mathrm{a}+v_2^\mathrm{c}} \quad \text{or} \quad v_2^\mathrm{a}=(v_2^\mathrm{a}+v_2^\mathrm{c})(1-\alpha_\mathrm{c}) \tag{4}$$

where v_2^a and v_2^c are the volume of the amorphous and crystalline parts of starch, respectively.

The volume fractions of the solvent (1) and solute (3) in the amorphous part of the system (ϕ_1, ϕ_3) are:

$$\phi_1=\frac{v_1}{v_1+v_2^\mathrm{a}+v_3} \tag{5a}$$

$$\phi_3=\frac{v_3}{v_1+v_2^\mathrm{a}+v_3} \tag{5b}$$

where v_1 and v_3 are the volume occupied by the solvent and solute, respectively.

The overall volume fraction of the solvent and solute ($\phi_1^\mathrm{T}, \phi_3^\mathrm{T}$) are:

$$\phi_1^\mathrm{T}=\frac{v_1}{v_1+v_2^\mathrm{a}+v_2^\mathrm{c}+v_3} \tag{6a}$$

$$\phi_3^\mathrm{T}=\frac{v_3}{v_1+v_2^\mathrm{a}+v_2^\mathrm{c}+v_3} \tag{6b}$$

By substituting Eq. (4) into Eq. (5a), dividing the denominator and numerator by $v_1+v_2^\mathrm{a}+v_2^\mathrm{c}+v_3$, then substituting Eqs. (6a) and (6b), and using $\phi_3^\mathrm{T}=1-\phi_1^\mathrm{T}-\phi_2^\mathrm{T}$, we obtain:

$$\phi_1=\frac{\phi_1^\mathrm{T}}{1-\phi_2^\mathrm{T}\alpha_\mathrm{c}} \tag{7a}$$

Analogously, combining Eqs. (4), (5b), (6a), and (6b), gives:

$$\phi_3=\frac{1-\phi_1^\mathrm{T}-\phi_2^\mathrm{T}}{1-\phi_2^\mathrm{T}\alpha_\mathrm{c}} \tag{7b}$$

Substituting Eqs. (7a) and (7b) into Eq. (3) gives:

$$T_m = T_m^0 - \left(\frac{R}{\Delta H_u}\frac{V_u}{V_1}\right)\left(T_m^0\right)^2\left(\left(\frac{\phi_1^T}{1-\phi_2^T\alpha_c}\right) + \left(\frac{1}{x_3}\right)\left(\frac{1-\phi_1^T-\phi_2^T}{1-\phi_2^T\alpha_c}\right) + \chi_{13}\left(\frac{\phi_1^T}{1-\phi_2^T\alpha_c}\right)\left(\frac{1-\phi_1^T-\phi_2^T}{1-\phi_2^T\alpha_c}\right) - \left[\chi_{12}\left(\frac{\phi_1^T}{1-\phi_2^T\alpha_c}\right) + \frac{\chi_{32}}{x_3}\left(\frac{1-\phi_1^T-\phi_2^T}{1-\phi_2^T\alpha_c}\right)\right]\left(\frac{\phi_1^T}{1-\phi_2^T\alpha_c} + \frac{1-\phi_1^T-\phi_2^T}{1-\phi_2^T\alpha_c}\right)\right) \tag{8}$$

If we assume that the non-crystalline fraction of starch is an inert component in the system, we can state that the amount of crystallinity far below the gelatinization/melting temperature represents 100% crystallinity. Therefore, the relative crystallinity can be used instead of the degree of crystallinity (Baks et al., 2007). Thus, α_c can be replaced by $1 - \mathrm{DG}$, where DG stands for the degree of gelatinization/melting. The resulting expression is:

$$T_m = T_m^0 - \left(\frac{R}{\Delta H_u}\frac{V_u}{V_1}\right)\left(T_m^0\right)^2\left(\left(\frac{\phi_1^T}{1-\phi_2^T(1-\mathrm{DG})}\right) + \left(\frac{1}{x_3}\right)\left(\frac{1-\phi_1^T-\phi_2^T}{1-\phi_2^T(1-\mathrm{DG})}\right) + \chi_{13}\left(\frac{\phi_1^T}{1-\phi_2^T(1-\mathrm{DG})}\right) \times \left(\frac{1-\phi_1^T-\phi_2^T}{1-\phi_2^T(1-\mathrm{DG})}\right) - \left[\chi_{12}\left(\frac{\phi_1^T}{1-\phi_2^T(1-\mathrm{DG})}\right) + \frac{\chi_{32}}{x_3}\left(\frac{1-\phi_1^T-\phi_2^T}{1-\phi_2^T(1-\mathrm{DG})}\right)\right]\left(\frac{\phi_1^T}{1-\phi_2^T(1-\mathrm{DG})} + \frac{1-\phi_1^T-\phi_2^T}{1-\phi_2^T(1-\mathrm{DG})}\right)\right) \tag{9}$$

Eq. (9) describes the relationship between the melting temperature, the total concentration of plasticizer, and the degree of gelatinization of starch in a starch–water–solute system.

3. Experimental

3.1. Materials

Native wheat starch (<12% H_2O) (S5127) and D-glucose (<0.5% H_2O) (G7528) were purchased from Sigma–Aldrich (Steinheim, Germany). Glycerol (<0.1% H_2O) was obtained from Acros Organics (Geel, Belgium). The moisture content of the most important powder materials was determined using an infrared heating balance (Sartorius MA30 Moisture Balance) heated to 130 °C. The water content of the starch, glucose, and glycerol was taken into account in all experiments.

Known amounts of native wheat starch, water, and solute (i.e., glycerol or glucose) were prepared and thoroughly mixed manually with a spatula before equilibration for at least 12 h at room temperature. The composition of the mixtures and the range of the treatment temperature are presented in Table 1. In addition, a 60% w/w starch–water mixture was prepared to compare with the ternary mixtures and the literature data.

3.2. Compression molding

Compression molding was used to investigate the effect of the solute during the gelatinization of starch, which leads to a reduction in starch crystallinity according to the compression molding conditions. To obtain the heat-treated sample, 30 g of the starch–water–solute mixture were put into the mold, which consisted of three stainless steel plates (300 × 350 mm^2). The upper and lower plates were 4 mm thick. The middle plate had a rectangular hole (150 × 100 × 1 mm^3), into which the mixture was placed. The samples were covered with plastic overhead transparencies (printable grade) on both sides of the middle plate to facilitate removal of the sample from the mold. The mold was then placed in a hydraulic PHI press (City of Industry, California). The mold was heated to the desired temperature at a rate of 10 °C min^{-1}, and a pressure of 0.86 MPa was applied. After heating, the starch mixture was compressed at the set temperature for an additional 45 min. The mold was cooled to room temperature at a cooling rate of 10–15 °C min^{-1}, and the material was then released from the mold.

3.3. Handling of samples

To minimize re-crystallization, samples taken from the compression molder were immediately transferred to a vessel containing liquid nitrogen and stored at −80 °C until further use.

After storage, the samples were freeze-dried in a Christ Epsilon 2-6D freeze-dryer (Osterode am Harz, Germany) prior to analysis. Freeze-drying was started at −20 °C and 1.030 mbar for 20 h followed by a second drying stage at −20 °C and 0.050 mbar for 20.5 h, and a third drying stage −5 °C and 0.001 mbar for 29.5 h. The freeze-dried samples were ground in an analytical mill (type A10) from IKA (Staufen, Germany) to obtain a fine powder that dissolved easily. For comparison, each unprocessed starch–water–solute mixture was freeze-dried and ground under the same conditions described above.

3.4. Wide-angle X-ray scattering (WAXS)

Diffractograms were recorded with a Philips PC-APD diffractometer. The scattered X-ray radiation was recorded by a proportional moving detector over a 4° and 40° (2θ) angular range with an angular scanning velocity of 1.2 min^{-1} and a measurement frequency of 1 s^{-1}. The Cu Ka radiation from the anode operating at 40 kV and 50 mA was monochromated using a 15-mm thick Ni foil. The diffractograms were smoothed with the computer program Table Curve 2D (Jandel Scientific, 1994, version 2.0, San Rafael, USA) by applying Savitsky–Golay data smoothing (5% smoothing). The procedure of (Van Soest, Tournois, De Wit, & Vliegenthart, 1995) was used to determine the relative crystallinity. For wheat starch, the characteristic peak at $2\theta = 22.9°$ was selected. A straight line was used to approach the baseline below the characteristic peak. Following the procedure described by Baks et al. (2007), the height of the characteristic peak (H_t), and the total height of the characteristic peak minus the height of the baseline at the diffraction angle at $2\theta = 22.9°$ (H_c) were measured. The ratio $R_H = H_c/H_t$ was obtained for native wheat starch (100% relative crystallinity). The relative crystallinity of a sample based on peak heights (X_{rH}) was determined using the following equation:

Table 1
Summary of thermal treatment and composition of the water, starch, and solute mixtures.

Starch:water ratio	Water (% w/w)	Starch (% w/w)	Glucose (% w/w)	Glycerol (% w/w)	Thermal treatment [°C]
3:2	40	60			30, 40, 50, 60, 70, 80, 90, 100, 110, 120
2:1	30	60	10		30, 40, 50, 70, 83, 90, 100, 110, 120, 130, 140, 150
	26	50	24		30, 40, 50, 70, 83, 100, 120, 130, 140, 150
	20	40	40		70, 83, 90, 100, 110, 120, 130, 140, 150
3:1	17	60		23	30, 40, 50, 60, 70, 80, 83, 90, 100, 110, 120, 130, 140, 150
2:1	26	50		24	30, 40, 50, 60, 70, 80, 83, 90, 100, 110, 120, 130, 140, 150
	20	40		40	70, 83, 100, 120, 130, 140, 150

$$X_{rH} = \frac{(R_H)_s}{(R_H)_n} \tag{10}$$

where the indices s and n indicate sample and native wheat starch, respectively. The relative crystallinity was used as a measure for the degree of gelatinization/melting by applying the following equation:

$$DG = 1 - X_{rH} \tag{11}$$

3.5. Differential scanning calorimetry (DSC)

Freeze-dried starch samples (5 mg) with 50 µl of Milli-Q water (i.e., 10% w/w starch–solute mixture) were hermetically sealed in stainless steel DSC pans (60 µl). The mixtures were left overnight to equilibrate the starch–water–solute mixture. Next, the samples were scanned against a blank (empty pan) using a Perkin Elmer Diamond DSC (Perkin–Elmer Co., Norwalk, CT). Before the actual measurement, the samples were held at 0 °C for 5 min in the DSC measuring cell. Subsequently, the sample was heated at 10 °C min^{-1} from 0 to 150 °C. The raw data were processed with Pyris version 8.0 (Perkin–Elmer Co., Norwalk, CT) to obtain the enthalpy changes needed to gelatinize the crystalline part of the starch in the sample (ΔH_s). For simplicity we will use enthalpy to refer to the change of enthalpy. The characteristic gelatinization endotherms obtained were corrected for the amount of solvent and solute present in the sample after the freeze-drying step. Using the gelatinization enthalpy of dried native wheat starch (ΔH_n), the degree of gelatinization/melting of the sample was determined:

$$DG = 1 - \frac{\Delta H_s}{\Delta H_n} \tag{12}$$

To eliminate any thermal effect on the gelatinization enthalpy of native wheat, untreated starch–water–solute mixtures were freeze-dried, re-suspended in approximately 10% w/w water, and analyzed. For each starch–water–solute mixture at least two independent DSC traces were recorded. The average DSC enthalpies of each starch–water–solute mixtures were used as reference for the estimation of DG.

3.6. Finding model parameter values

Eq. (9) was used to calculate the melting temperature as a function of the degree of gelatinization/melting, starch concentration, and the total concentration of plasticizers. The results from the model were compared to the experimental data in the temperature range 60–140 °C using Mathcad (Parametric Technology Corporation, version 14.0, USA). The partial molar volumes were 18.1 cm^3 mol^{-1} for water (Baks et al., 2007), 97.5 cm^3 mol^{-1} for wheat starch (Shahidi, Farrell, & Edward, 1976), 112.2 cm^3 mol^{-1} for glucose (Shahidi et al., 1976), and 73.0 cm^3 mol^{-1} for glycerol (Habeych, Van der Goot, & Boom, 2007), respectively. It was assumed that these partial molar volumes are constant and do not depend on temperature. In addition, it was assumed that the densities of the crystalline and the amorphous wheat starch are equal. Two interaction parameters (χ_{13} and χ_{23}) were determined by minimizing the sum of the squared residuals.

4. Results

The gelatinization enthalpies obtained for each untreated starch–water–solute mixture are given in Table 2. The gelatinization enthalpy obtained for freeze-dried native wheat starch: 13.4 ± 0.6 J g^{-1} (95% confidence interval) is in good agreement with the value reported elsewhere (Baks et al., 2007; Chiotelli & Le Meste, 2002; Ratnayake & Jackson, 2007). Moreover, average DSC enthalpies of the binary starch–water system treated at low temperature (i.e., 30–50 °C) increased from ~16.2 Jg^{-1} at 30 °C to 18.1 Jg^{-1} at 50 °C. Ratnayake and Jackson (2007) observed similar results in starch–water system. The authors explained this phenomenon due to the rearrangement of amylose in the amorphous domains of the starch granules. During the first part of the starch gelatinization/melting process, the water absorbed by the granules increases the mobility, especially of amylose, in the amorphous domains, leading to realignment and formation of new intermolecular bonds (Ratnayake & Jackson, 2007). Overall, it leads to an additional enthalpy bonus, which however is not directly related to crystallinity. We therefore took zero values for DG at low temperature in the fitting estimations. Furthermore, the addition of glycerol decreases the average DSC enthalpy of untreated samples especially for mixtures containing a high glycerol concentration (i.e., 40% w/w glycerol) (Table 2). Mixtures containing glucose did not show a statistically significant difference with only starch in water.

Table 2
Enthalpy (ΔH) of the gelatinization or melting for different untreated freeze-dried water–starch–solute mixtures (i.e., 10% w/w starch–solute) determined by DSC measurements with a heating rate of 10 °C min^{-1} (native wheat starch 13.4 ± 0.6 J g^{-1}).

Starch:water ratio	Glucose (% w/w)	Glycerol (% w/w)	ΔH (J g^{-1})[a]
2:1	10	–	13.4 ± 1.0[b]
	24	–	14.9 ± 0.9[b]
	40	–	12.4 ± 1.1[b]
3:1	–	23	12.8 ± 0.8[b]
2:1	–	24	11.2 ± 0.3[b]
	–	40	8.6 ± 1.9[b]

[a] Gelatinization enthalpy in J g^{-1} dry starch.
[b] Confidence interval (95%).

Fig. 1 shows the most characteristic WAXS diffractograms obtained from each starch–water–solute mixture. WAXS data showed three crystal structures depending on the processing conditions and composition. The V_H ($2\theta \approx 19.8°$) and E_H ($2\theta \approx 18.5°$) type peaks are generally associated with the single helical amylose during processing and have been reported to appear after compression molding of wheat starch containing water and glycerol (Van Soest, Hulleman, De Wit, & Vliegenthart, 1996). The relative crystallinity of wheat starch gradually decreased with the heat treatment starting at 30 °C (see Fig. 1). Similar results were reported by Ratnayake and Jackson (2007) for 6% w/v mixture of different starches heated in the range of 35–85 °C at 5 °C intervals for 30 min. In contrast, 26:50:24 water:starch:glycerol sample showed an opposite trend that agree with the DSC data collected for low temperate treated samples (Fig. 1-BII). Furthermore, WAXS for mixtures with 40% w/w glucose shown a overlapped signal with glucose and were therefore not use in the fitting procedure.

Fig. 2 shows the degree of gelatinization of a starch–water–solute mixture obtained from WAXS and DSC measurements after various treatments at constant temperature for 45 min. The start and the end of the gelatinization/melting process used in the fitting procedure are marked with vertical lines. Within this range the DSC measurements yield a higher estimation of the gelatinization temperature than WAXS for mixtures containing glucose; for mixtures containing glycerol, DSC gave comparable results as WAXS. Furthermore, the temperature span of the gelatinization process (i.e., start to end) decreases by increasing the total amount of plasticizer.

Fig. 2 compares the experimental values with those predicted by the model. The predicted and experimental values are close although there are some deviations at relatively large quantities of solute. In general, the modified Flory–Huggins equation used here somewhat under predicts the degree of gelatinization process and gave more accurate results when the solute concentrations

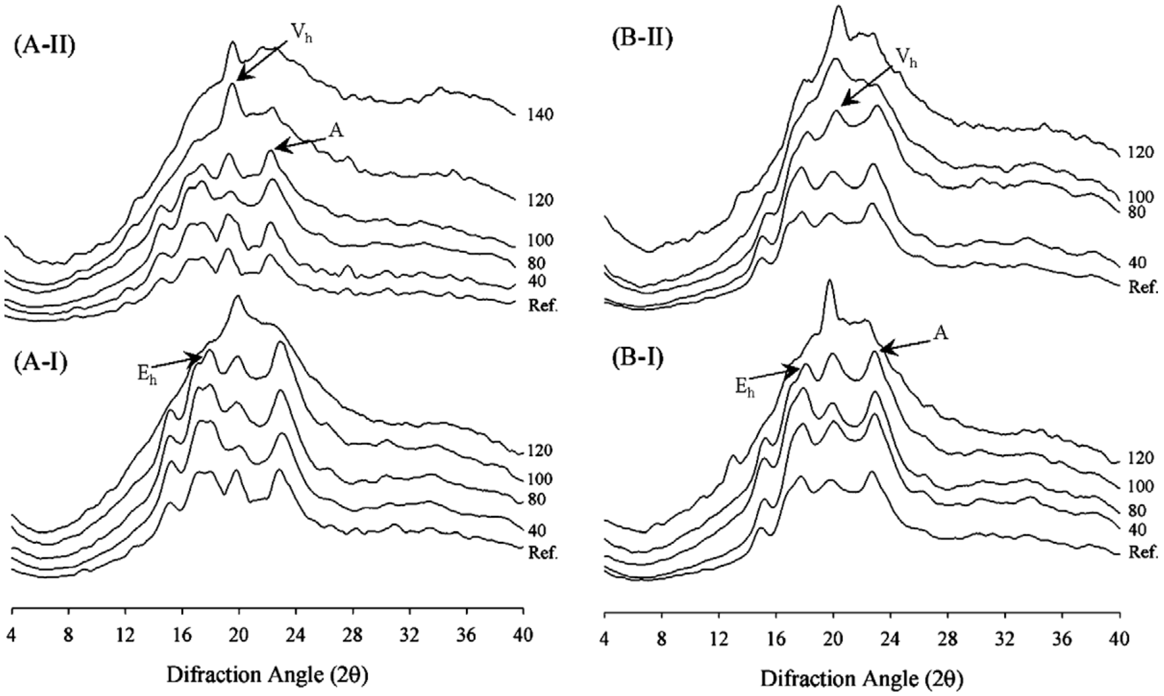

Fig. 1. WAXS from starch–water–solute samples treated at different temperatures. Water:starch:glucose: (A-I) 30:60:10, (A-II) 26:50:24. Water:starch:glycerol: (B-I) 17:60:23, (B-II) 26:50:24. Bottom: untreated native wheat starch. Numbers to the right of each profile represent the corresponding treatment temperature (°C).

were relatively low. Relative to the values obtained for the binary system (Fig. 2A), the addition of solute at constant starch concentration shifted the end of the gelatinization curve (i.e., DG = 100%) upwards in temperature (see dash line Fig. 2A, B, and E). This situation is observed for every starch:water ratio and has a direct relation with the molecular weight of the solute. The presence of glucose seems to shift the onset of the gelatinization/melting process (i.e., DG = 0) to a higher temperature (see straight line Fig. 2B–D), glycerol had a different effect: the onset gelatinization temperature now remains approximately constant at 70 °C (cf. Fig. 2E–G).

Table 3 presents the predicted Flory–Huggins parameters obtained using the DSC and WAXS values. The starch–water interaction parameter (χ_{12}), the heat of fusion of the starch repeat unit (ΔH_u), and the melting temperature of pure polymer (T_m^0) were derived from the results obtained for the binary system (60% starch) in combination with data reported earlier (Baks et al., 2007). The values are within the confidence interval of those reported earlier. χ_{12} is in agreement with the value obtained by others using different methods of measurement (Lelievre, 1976; Moates, Noel, Parker, & Ring, 1997), ΔH_u is quite close to the enthalpy of fusion of D-glucose, which has been reported to be around 30 kJ mol^{-1} (Moates et al., 1997). The value of T_m^0 is close to the value of 259.85 °C obtained by van den Berg (Van den Berg, 1981).

The interaction parameter between starch and the solute (χ_{23}) was found to be negative in both cases, indicating strong enthalpic interaction. Similar results were reported earlier for starch–glucose and starch–D-glucitol (polyol) (Moates et al., 1998). The interaction parameter between water and glucose ($\chi_{13} \approx 0.30$) appeared to be slightly lower than the value obtained for starch–water ($\chi_{12} \approx 0.48$) and the values reported earlier ($\chi_{13} \approx 0.5$) (Baks et al., 2007; Lelievre, 1976; Van den Berg, 1981).

In order to evaluate the plasticizing capability of water, glycerol, and glucose; Eq. (3) can be rearranged to estimate the effect of each plasticizer on the change in melting temperature of starch (i.e., $T_m^0 - T_m$) as a function of the volume fraction of the plasticizers, as follows:

$$T_m^0 - T_m = \left[(T_m^0)^2 \frac{R}{\Delta H_u} \frac{V_u}{V_1}\right] \left[\phi_1 + \left(\frac{\phi_3}{x_3}\right) + \chi_{13}\phi_1\phi_3 - \chi_{32}\left(\frac{\phi_3}{x_3}\right)(\phi_1 + \phi_3) - \chi_{12}\phi_1(\phi_1 + \phi_3)\right] \tag{13}$$

Fig. 3 shows the results of the simulation for the onset of melting (i.e., DG is 100%), which can be seen as a measure of the effectiveness as plasticizer of each molecule. From this figure results one can see that water has the highest effectiveness in terms of change in melting temperature compared to glycerol and glucose with glycerol being more effective than glucose. The contributions of each term in the second bracket at the right side of the Eq. (13) can be analyzed as well. In this equation the first and second terms arise from the entropy of mixing and the other three terms are related to the enthalpy of mixing and depend of the interaction parameters χ_{13}, χ_{32}, and χ_{12} (Lelievre, 1976; Moates et al., 1997). Fig. 4 shows the change in the magnitude of each term when 10% of the volume fraction of water is replaced by either glycerol or glucose. In both cases, the main contributions to the change in melting of starch come from Term 1 and Term 5, meaning that waters plays a major role in the equation because the entropy of mixing given by water and the interaction between starch and water mainly. Replacing water for a larger molecule (i.e., glycerol or glucose) results in a lower entropy of mixing and hence, (as can be seen with the reduction in Term 1), leading to a smaller change in melting temperature of starch (Fig. 3). In summary, the depression of starch gelatinization is mainly affected by the entropic (Term 1) and enthalpic (Term 5) contribution of water with little influence of the other terms.

5. Discussion

To study the gelatinization/melting process one can follow an on-line or an off-line approach (Cooke & Gidley, 1992). The on-line approach implies an on-line direct registration of the physical–chemical changes during gelatinization/melting by for example DSC, light scattering, and/or optical microscopy. In this study, we

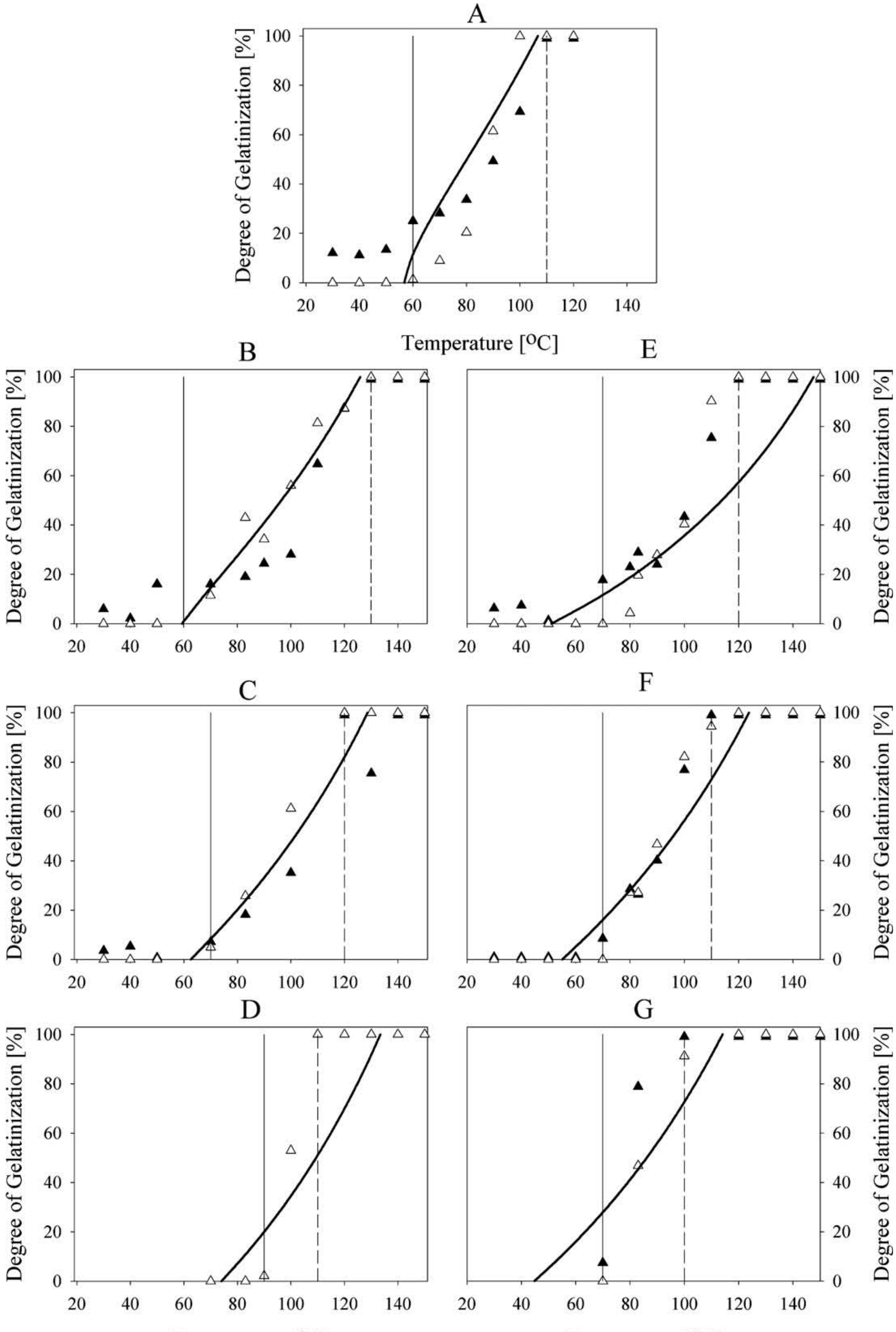

Fig. 2. Degree of gelatinization of starch as a function of the treatment temperature (treatment time 45 min). Solid line represents the model prediction, DSC data (△), and X-ray data (▲). (A) 60% w/w wheat starch–water mixture. Water:starch:glucose: (B) 30:60:10, (C) 26:50:24, (D) 20:40:40. Water:starch:glycerol: (E) 17:60:23, (F) 26:50:24, (G) 20:40:40. Vertical solid and dash line represent the onset and the end of the gelatinization process obtained during the isolation of the samples.

have used off-line analysis to obtain detailed information on the crystalline order at different temperatures and water–starch–solute concentrations. During the off-line approach samples were isolated during gelatinization/melting and analyzed afterwards. The principal disadvantage of this approach is the possibility of structural changes during the isolation process, which probably ex-

Table 3
Flory–Huggins parameter relevant to water–starch–solute used by different authors.

System	ΔH_u (kJ mol^{-1})	T_m^0 (°C)	χ_{12} (–)	χ_{13} (–)	χ_{32} (–)	Reference
Water:starch	32.83	258.20	0.48	–	–	This article
Water:starch:glycerol	32.83	258.20	0.48	−0.21	−1.64	This article
Water:starch:glucose	32.83	258.20	0.48	0.30	−1.00	This article
Water:starch:glucose	25.12	221.85	0.50	0.50	0.00	Lelievre (1976)
Water:starch:glucose	22.00	262.85	0.57	0.74	−1.63	Moates et al. (1998)
Water:starch:D-glucitol	22.00	262.85	0.57	0.74	−1.42	Moates et al. (1998)

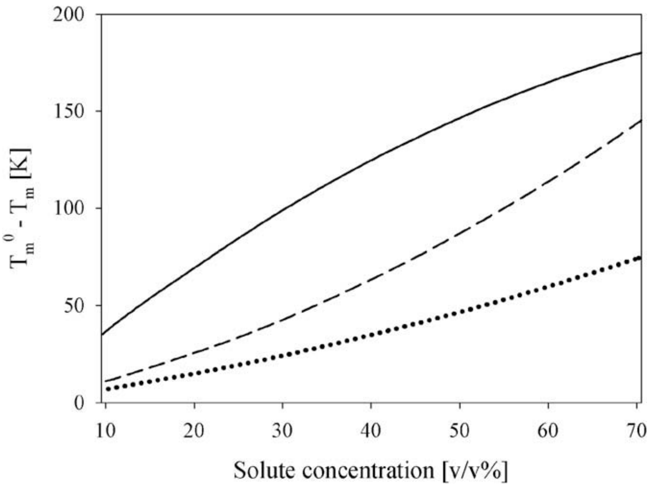

Fig. 3. Theoretical prediction of the effect of plasticizer concentration on the change in melting temperature. Solid line: water, dash line: glycerol, and dot line: glucose.

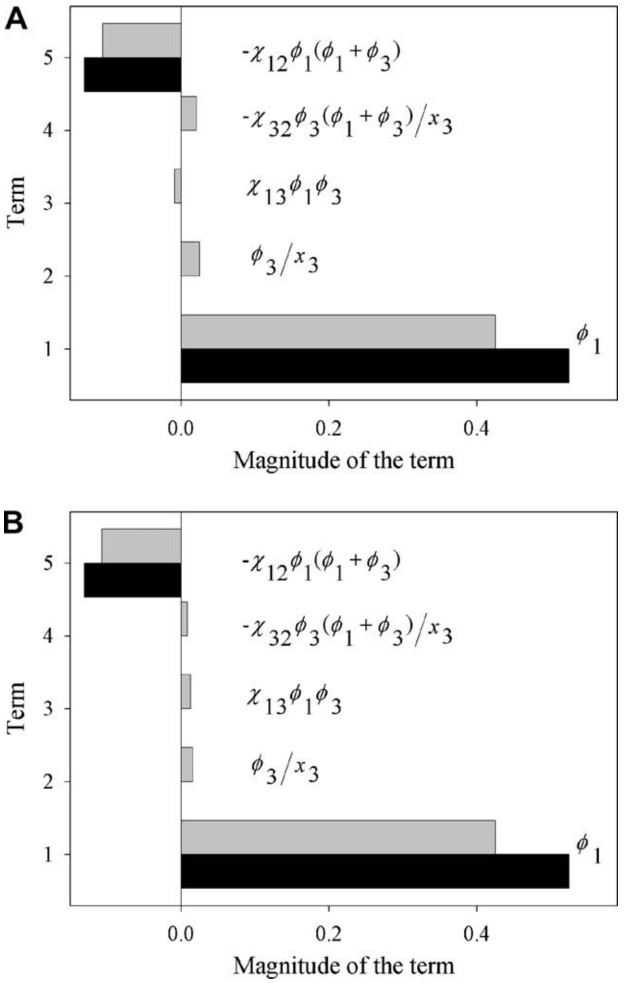

Fig. 4. Contributions of the four composition terms in Eq. (13) to the change in melting temperature in a 60% w/w starch–water mixture due to replacement of 10% v/v water by solute as predicted by the extended Flory–Huggins's theory (see Eq. (13)). (A) Effect on glycerol addition. (B) Effect on glucose addition.

plains a relatively large spread in WAXS and DSC data (Fig. 2) but it allows much more extensive analysis. Nevertheless, an offline approach is the most appropriate to one obtained data where kinetic effects can be suppressed.

The DSC endothermic enthalpy values reflect both the loss of double-helical order and crystallinity. WAXS however only detects crystallinity (Cooke & Gidley, 1992). Cooke and Gidley (1992) found that the total amount of ordering is almost twice as large as the amount of crystallinity. This explains why DSC gives higher values for the loss of crystallinity compared to WAXS.

The results presented above show that the start and the end of the gelatinization/melting process depend on the amount and type of plasticizers. Compared to systems with only water, the presence of another solute during the gelatinization/melting process shifts the end of the gelatinization curve (i.e., DG = 100%) upwards in temperature. An increase in the total amount of plasticizers reduced the temperature span of gelatinization process. Therefore, this reduction of the temperature span of the gelatinization process gives a probe that what fundamentally matters is the properties of the mixture as a whole. Similar findings were reported earlier (Perry & Donald, 2000; Perry & Donald, 2002), who proposed a *solvent–temperature–time superposition* model envisaged in two steps. The first step was referred to as a swelling-driven step in which a minimum degree of plasticization is required to induce mobility in the granule. Thereafter, the combined action of solvent and temperature prompts the loss of crystallinity. Our results agree with this model: changes occurred prior to the gelatinization, as was observed with increase in enthalpy changes at low temperature. Furthermore, at higher temperatures the crystalline regions are disrupted due to the cooperative action of solvent and heat.

Strong enthalpic interactions were found between starch and the solutes glucose and glycerol ($\chi_{23} < 0$). Previous work showed a strong exothermic peak during DSC measurements of dry starch in the presence of glycerol and sugars (Perry & Donald, 2000; Van Soest, Bezemer, et al., 1996). Van Soest, Bezemer, et al. (1996) attributed the exothermic peak to the stronger interaction between starch–glycerol compared to the starch–water interaction. Perry and Donald (2000) gave this a physical interpretation by suggesting that the exothermic peak is indicative of reorganization in the crystalline lamellae. Before the ingress of the solvent, the lamellar and crystalline organization is imperfect. Due to solvent absorption, rearrangement of the crystalline lamella can occur giving an enthalpic bonus derived from crystallization. In addition, the diffusion of the solute in between the starch molecules takes time, but will be faster at higher temperatures. Therefore, at high solute concentration, the temperature to which the starch–solute mixture must be heated to obtain plasticization increases. In addition, higher molecular weight solutes will need much more time to penetrate into the starch granule, because they diffuse much more slowly in the granule matrix, and are less effective plasticizers due to their lower contribution to the entropy of mixing. In consequence, when only water acts as the solvent, it is proposed that hydration, plasticization and resulting lamellar arrangement crystallization occurs faster, while the effects of glucose and glycerol are slower and smaller (see Fig. 2).

The Flory–Huggins model was developed for concentrated polymer mixtures (Lelievre, 1976; Moates et al., 1998). Therefore, it is not surprising that the (modified) Flory–Huggins equation shows some deviation at high solute concentration. Moreover, starch contains other components, such as lipids, which influence the behaviour of the amylose and amylopectin by complex formation. A further additional problem is that starch is only partially crystalline; situation that is not included in the model. Finally, the initial plasticizing step previous to gelatinization is not taken into account in the model.

The applicability of the extended Flory–Huggins equations has pros and cons that should be taken into account before using it as a predictive tool. Fig. 5 compares the binary model with the extended form for ternary systems. The extended form gives closer predictions even in the case of assuming a higher starch concentration as a result of the lower water availability due to the presence of the solute. One can thus see from Fig. 5 that the binary model does not improve the prediction and gives significant over prediction the onset (start) of the process. In addition, in spite of the many assumptions and simplifications made, the extended form can be interpreted physically, which has been proven to work well in ternary mixtures (Clark, 2000). Finally, the validity of the extended form of the Flory–Huggins equation allows to reduce the amount of experimental work, which is not possible with the binary model. However, the extended form of the model does require the finding of two additional parameters (i.e., χ_{13} and χ_{23}), which however do not contribute strongly to the prediction (Fig. 4), and therefore might be estimated. The predictability decreases specially at high solute concentration, so its applicability seems mainly in systems that are more concentrated in starch. In summary, the use of the extended form has some advantages that make it fair to consider in spite of the deviation obtained.

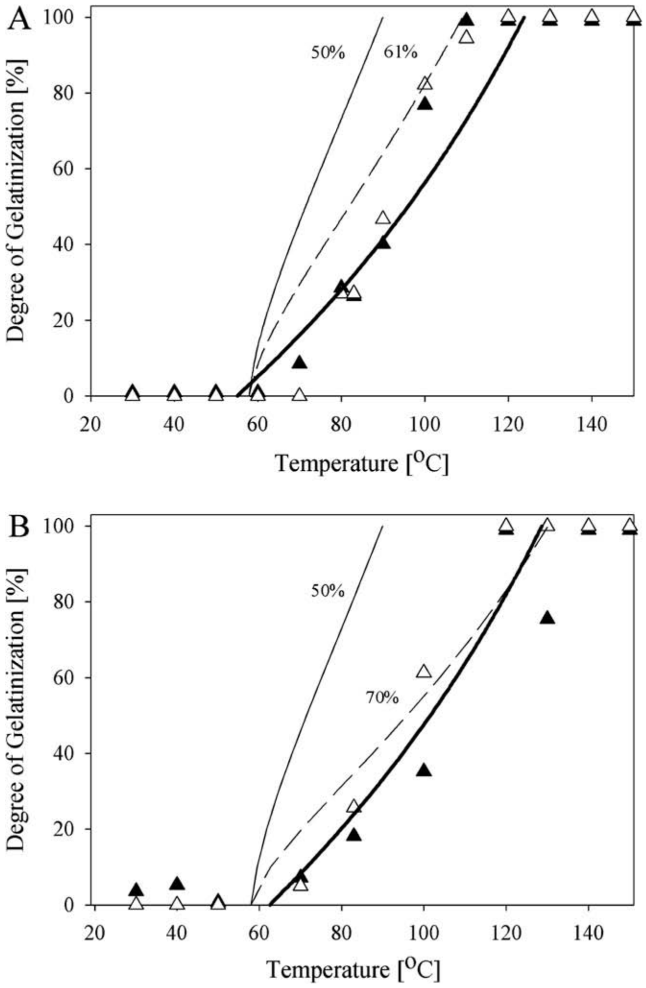

Fig. 5. Degree of gelatinization of starch as a function of the treatment temperature (treatment time 45 min). Solid bold line represents the model prediction using the extended model, percentage indicate the predicted value using the binary model, DSC data (△), and X-ray data (▲). (A) 26:50:24 water:starch:glycerol. (B) 26:50:24 water:starch:glucose.

The Flory–Huggins theory applied to starch has a number of shortcomings. In the original theory, a solvent molecule was assumed to occupy the same volume as a monomer unit of the polymer. In the case of molecules with large size differences such as starch and water, this assumption is not true (Gustafsson, Wennerstrom, & Tjerneld, 1986). For polymers capable of hydrogen bonding with water, the inter-polymer H-bonds may be disrupted by replacement of these bonds by polymer–water H-bonds (Slade & Levine, 1987), leading to a large increase in molecular mobility. Many solvent molecules will interact with the monomeric unit making this interaction more complex than the pair-wise nearest-neighbor interactions assumed in the Flory–Huggins theory (Flory, 1964). Nevertheless, the application of this theory to uncharged biopolymer–water systems for the description of ternary diagrams has proven to be no worse than for synthetic polymer–solvent systems, and is sometimes even better (Clark, 2000). Obtaining the correct experimental equilibrium conditions and thereby the correct analytical data is difficult. For the case of stiff, high-molecular-weight polysaccharides, Clark (2000) stated that this is of greater importance than incorrect assumptions in the model.

In spite of the limitations outlined above, the Flory–Huggins theory can be used to derive ternary phase diagrams using the set of parameters obtained for the water–starch–solute system (see Table 3), and setting DG = 100%, and Eqs. (7) and (9). Figs. 6 and 7 show the solid–liquid transition phase diagrams for the water–starch–solute systems. The curves represent the composi-

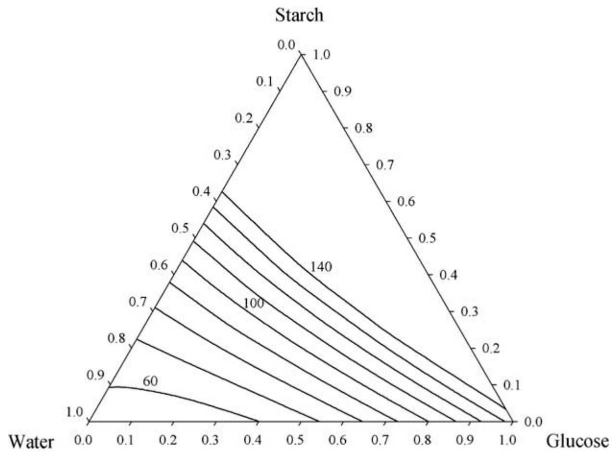

Fig. 6. Volumetric ternary phase diagram for the gelatinization/melting of the water–starch–glucose system. The solid lines refer to the corresponding predicted equilibrium gelatinization/melting isotherm. Numbers in the graph represent the corresponding isotherm in ascending order starting from T = 60 °C to T = 140 °C.

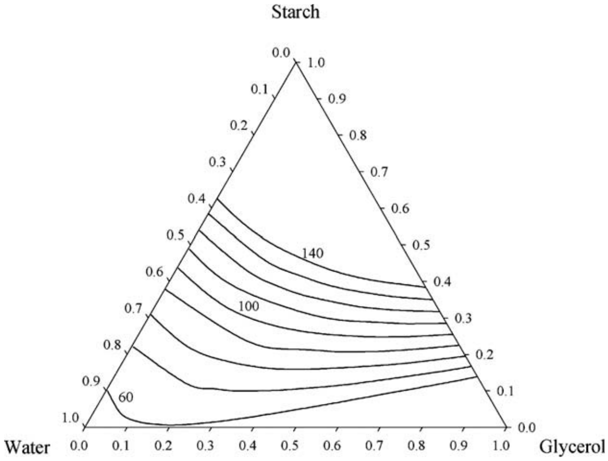

Fig. 7. Volumetric ternary phase diagram for the gelatinization/melting of the water–starch–glycerol system. The solid lines refer to the corresponding predicted equilibrium gelatinization/melting isotherm. Numbers in the graph represent the corresponding isotherm in ascending order starting from $T = 60$ °C to $T = 140$ °C.

tions at equilibrium with (pure starch) crystals at specific, indicated temperatures. Comparing Fig. 6 with Fig. 7, it can be seen that the water–starch–glycerol system shows a smaller crystallization region than water–starch–glucose, due to the stronger water–glycerol interaction and lower molar volume relative to water–glucose (see χ_{13} in Table 3). Since the extended form of the Flory–Huggins model under predicts the experimental values of the gelatinization process, the values reported in Figs. 6 and 7 give a reasonable estimation of the amount of plasticizer and heat treatment required to obtain a fully gelatinized starch.

6. Conclusion

The effects of glucose and glycerol on gelatinization/melting of highly concentrated starch mixtures were investigated with WAXS and DSC. The results suggest that the gelatinization/melting of starch can be envisaged in two steps, similar to the solvent–temperature–time superposition model proposed earlier (Perry & Donald, 2000; Perry & Donald, 2002). An adapted version of the Flory equation provided a quantitative description of the degree of starch gelatinization/melting as a function of the starch–water–solute ratio and temperature. The applicability of this extended model showed that it is useful to consider the theory as a mean to interpret and predict the gelatinization/melting behaviour of ternary starch-based systems. However, the theory does not capture all changes occurring in the ternary starch system.

Fitting the model to experimental data resulted in T_m^0, ΔH_u, χ_{12}, χ_{13}, and χ_{32} values that are reasonable and in agreement with values reported in the literature. The adapted Flory equation can be used as a means to estimate the temperature that is needed to completely gelatinize starch in a starch–water mixture over the whole concentration range. Ternary phase diagrams were constructed with crystallization lines representing fully gelatinized starch. The crystalline region with glucose was larger than with glycerol. This could be understood from the differences in χ_{13} (solute–solvent interaction) and molar volumes.

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