

Glass Transition Temperature of Honey as a Function of Water Content As Determined by Differential Scanning Calorimetry

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The glass transition of pure and diluted honey and the glass transition of the maximally freeze-concentrated solution of honey were investigated by differential scanning calorimetry (DSC). The glass transition temperature, T_g , of the pure honey samples accepted as unadulterated varied between -42 and -51 °C. Dilution of honey to 90 wt % honey content resulted in a shift of the glass transition temperature by -13 to -20 °C. The concentration of the maximally freeze-concentrated honey solutions, C_g , as expressed in terms of honey content is approximately 102–103%, i.e., slightly more concentrated in sugars than honey itself. The application of DSC measurements of T_g and C_g in characterization of honey may be considered, but requires systematic study on a number of honeys.

Keywords: Honey; glass transition; freeze concentration; differential scanning calorimetry, DSC

INTRODUCTION

Rate- and direction-dependent temperature shifts and low peak resolution make differential scanning calorimetry (DSC) inappropriate for the determination of the real nature of phase transitions (first-order vs continuous phase transitions, etc.). On the other hand, DSC characterization is quite sensitive; it offers a fairly good determination of the heat of transitions and reveals the occurrence of thermal events. Due to these advantages, DSC is widely used in research and industrial laboratories as well.

Depending on concentration and heating and cooling rates, aqueous carbohydrate solutions possess several thermal events (Franks, 1982). In real thermodynamic equilibrium, below the liquidus, crystallization of water and/or of the solute should occur. In practice, due to the not infinitely slow temperature changes (e.g., in industrial food processing, or in a DSC experiment), several other dynamic thermal events are observed. From a practical aspect, one of the most important of these is the glass transition (Blanshard and Lillford, 1993), i.e., the transition of a concentrated solution into an amorphous solid upon cooling at the glass transition temperature, T_g . The diffusion-limited processes, which are extremely slowed in the glassy state, influence the stability, and thereby, the shelf life of food products. Molecular movements, crystallization, etc., are practically blocked in a glass, at least on the possible time scales of laboratory investigations. Among the sharp changes in the trends of several materials properties (specific volume, viscosity, etc.) at T_g , the heat capacity undergoes a steplike change, appearing as a (blurred)

step in the DSC heat flow vs temperature curves [e.g., Chapter 1 in Franks (1982) and Wunderlich (1981)]. Addition of water to the mixture normally lowers the glass transition temperature.

When the solution freezes, water separates out as pure ice, leading to an increase in the concentration of the unfrozen solution (freeze concentration). The concentration of the solute reaches a maximum level, C_g , at and above which further crystallization of water is no longer possible even at lower temperatures. Upon further cooling, the maximally freeze-concentrated solution undergoes a glass transition at T_g . Since both the actual freezing point of water and the glass transition temperature are fundamentally determined by the concentration, both T_g and C_g are largely independent of both the original concentration of the solution and the cooling rate (or power) and are typical of the kind of solute.

It should be noted that in DSC characterization of aqueous sugary systems, the glass transition of the maximally freeze-concentrated solution appears as a double-step feature in the heating scan data: a step in the heat flow (or heat capacity) curve at T_g , preceded by another step related to coexisting less concentrated glass due to limitation in molecular transport at low temperatures and high sugar concentrations.

In the application of nuclear magnetic resonance (NMR) for the investigation of vitrification, Rubin et al. used honey as a model carbohydrate system (Rubin et al., 1990), in parallel with DSC measurements. As one of their conclusions, they found DSC to be a reliable tool for the investigation of the glass transition events in pure and diluted honey.

Because of the importance of the water content in the preservation of honey, Dadarlat et al. recommended the application of photopyroelectric (PPE) calorimetry in the determination of the moisture content (Dadarlat et al., 1998). Clearly, the PPE signals changed characteristi-

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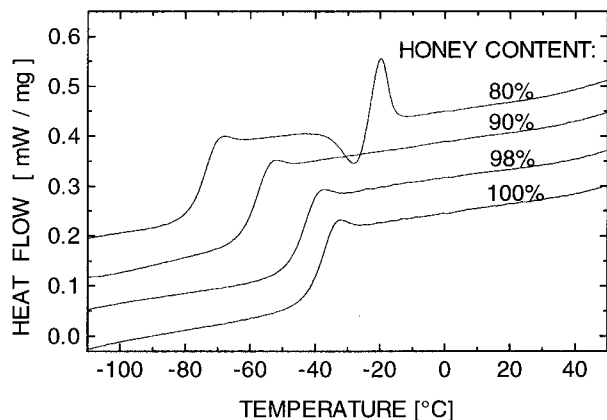


Figure 1. DSC heat flow curves as a function of temperature of pure and diluted samples of honey 1 with honey content between 80 and 100% recorded during heating from -130 to 50 °C at a temperature scanning speed of 10 °C/min. The heating scans were preceded by cooling at -10 °C rate. In these samples water did not crystallize upon cooling. For clarity reasons, the curves are vertically displaced. Positive changes correspond to endothermal events.

cally with added water content, which may be of practical importance, even though the interpretation of the thermal event, observed close to 0 °C for pure honey and at significantly lower temperatures for the diluted samples, as *crystallization*, may require further substantiation.

There are several methods for testing the water content of honey, including mechanical methods (viscosimetry and picnometry), oven drying, chemical titration, and as the fastest and widely used test, refractometry.

MATERIALS AND METHODS

Natural tree honeys from a German and a Dutch source (honeys 1 and 2, respectively), acacia honey (3), and mixed floral honey from a Hungarian source (4) were investigated. Before the experiments, the samples were kept in the laboratory (21 – 22 °C) in well-sealed containers, so moisture exchange with the ambient air was practically avoided. Diluted samples of honey 1 with 98, 90, 80, 70, 60, 50, and 30% (weight percent) honey content were prepared by mixing with distilled water. The absolute error of the determination of the honey content was less than 0.3%. All undiluted and diluted samples were encapsulated into sealed stainless steel sample pans for use in a commercial DSC apparatus. The weight of the samples was between 23 and 35 mg, determined with 0.5% accuracy.

The phase transitions in the undiluted and diluted samples were detected by a Perkin-Elmer differential scanning calorimeter (DSC 7) working on the principle of power compensation. The calorimeter's furnace was cooled to approximately -150 °C with liquid nitrogen. The temperature changing rate (scanning speed) in the DSC was 10 K/min both in heating and cooling, as a compromise between noise and temperature determination accuracy. Before cooling scans, the samples were kept at $+50$ °C during 3–5 min to reach thermodynamic equilibrium. The samples were routinely cooled to approximately -130 °C and then heated to $+50$ °C again.

For each scan, the obtained power difference data was evaluated by subtracting a baseline and dividing by the mass of the sample, resulting in a heat flow per unit mass (referred as heat flow). For the better evaluation of certain phase transition phenomena, the derivative of heat flow with respect to temperature was also determined. The temperature scale was calibrated for $+10$ K/min heating rate using the onset temperature of melting of acetone (-95.35 °C), chlorobenzene (-45.6 °C), and water. With chlorobenzene, amplitude calibra-

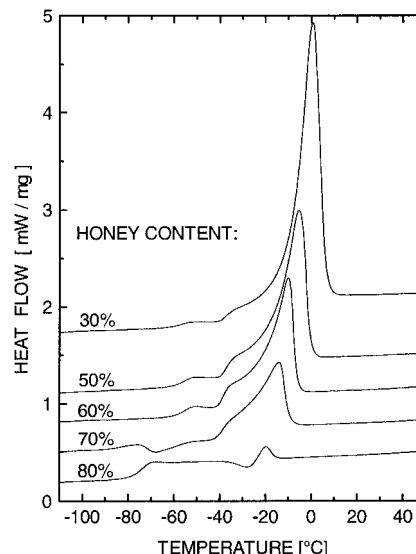


Figure 2. DSC heat flow curves as a function of temperature of pure and diluted samples of honey 1 with honey content between 30 and 80% recorded during heating from -130 to 50 °C at a temperature scanning speed of 10 °C/min. The heating scans were preceded by cooling at -10 °C rate. In these samples water crystallized upon cooling. For clarity reasons, the curves are vertically displaced. Positive changes correspond to endothermal events.

tion was also performed. For the cooling scans, reliable calibration of the temperature scale could not be performed because of supercooling of the temperature standards. In the following, all displayed curves and temperature data will refer to the DSC data obtained from heating scans. In cooling, only the cooling rate data was used.

The glass transition temperature, T_g was consistently determined in the heating scans as the onset temperature of the steplike increase in the heat flow. It should be noted that this is different from the common practice, i.e., defining T_g as the midpoint of the step. The reason for this is the more precise definition of T_g in case of double-step structures, or when the melting of ice renders the exact determination of the height of the heat capacity step unreliable. This practice is also favored by Shalaev et al. (Shalaev and Franks, 1995).

Although first-order phase transitions can hardly be treated by DSC, the determination of the melting point of frozen solutions was attempted by detecting the abrupt change in the slope of the first derivative of the DSC curves at the high-temperature end of the melting range, assuming that the remixing of the melted water with the solution is immediate. The melting-point data obtained this way (which may possess some error as compared to the real equilibrium values) are presented for approximate information only.

RESULTS AND DISCUSSION

The DSC curves recorded in heating of the pure and diluted tree honey (honey 1) samples are shown in Figure 1 (100, 98, 90, and 80% honey content) and in Figure 2 (80, 70, 60, 50, and 30% honey content). For clarity reasons, the curves have been vertically displaced. Because of instrument limitations, there is always an unknown additive constant in the recorded differential power data that consequently renders the determination of absolute heat capacity values in this setup impossible. However, changes in the heat capacity within narrow temperature domains can be quantitatively determined.

The glass transition temperature, T_g , of the undiluted sample as determined in the heating scans as the onset temperature of the increase in the heat flow was ap-

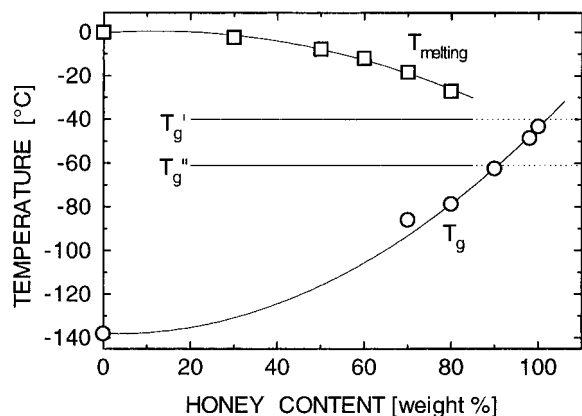


Figure 3. State diagram of the water–honey 1 system obtained from DSC data at 10 °C/min heating rate. T_{melting} and T_g indicate the breakdown point of the melting peaks (see Figure 2) and the onset of the heat-flow step associated with the glass transition (see Figure 1). The onset temperatures of the glass transitions of the freeze-concentrated solutions (T_g' and T_g'') are also indicated.

Table 1. Glass Transition Temperatures of Pure ($T_g^{100\%}$) and Diluted ($T_g^{90\%}$) Honeys and of the Freeze-Concentrated Solutions (T_g' , T_g'') and the Concentration of the Maximally Freeze-Concentrated Solution (C_g) As Expressed in Terms of Honey Content

honey	$T_g^{100\%}$ (°C)	$T_g^{90\%}$ (°C)	T_g' (°C)	T_g'' (°C)	C_g (%)
1	-42.5	-62.6	-40	~ -60.5	~102%
2	-50.7	-63.6			
3	-46.2	-59.1			
4	-47.9	-65.3	-41.9	~ -62.5	~103%
data from Rubin (1990)	-49	-65	~-44	~-62	~103%

proximately -42.5 °C (see Figure 1, bottom curve, represented in Figure 3 and Table 1). The diluted samples possessed significantly different glass transition temperatures of -48.2 , -62.6 , and -78.8 °C for 98, 90, and 80% honey contents, respectively (Figure 1 and Table 1). For the samples with at least 90% honey content, there was no indication of crystallization of water (Figure 1). The first sign of water crystallization was observed at 80% honey content (Figure 2, bottom curve), only in the *heating* scans, after devitrification. During cooling, crystallization was not detected for this sample, even in repeated thermal cycling, because of supercooling of the solution beneath the temperature at which water could crystallize at the actual cooling rate.

With decreasing honey content below 80%, the melting point of the solution moved toward 0 °C (Figure 2), with the concomitant decrease of the change of enthalpy upon melting as a function of honey content (Figure 4) as approximately calculated by the integration of the phase transition peak of the curves in Figure 2. The extrapolation of the data toward higher concentrations revealed that water crystallization from mixtures with more than approximately 85% honey content is rather unlikely under any circumstances.

The glass transition temperature of the maximally freeze-concentrated solution, T_g remained constant at approximately -40 °C for all dilutions with honey content between 70 and 30% (Figure 2). Between approximately -61 and -59 °C (see Figure 2), there is another step in the heat flow curves. The onset temperature of this change, T_g'' , is also seemingly invariant of concentration. This double-step feature appearing in the DSC curves of the freezable honey solutions was also

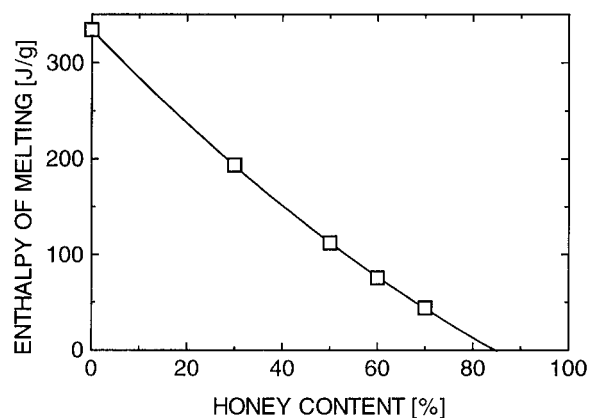


Figure 4. Enthalpy of melting (i.e., the integrated area of the melting peaks of the curves of Figure 2) as a function of honey content.

demonstrated for another type of honey by Rubin et al. (1990) and for binary sugar–water systems by several authors [see, e.g., Simatos and Blond (1993), Shalaev and Frank, (1995), Ablett et al. (1992), etc.].

In the DSC heating curve of the 70% solution, one can observe a small increase at approximately -86 °C (liquefaction of the glass) and then an exothermic bump (water crystallization) between approximately -72 and -62 °C.

The state diagram of the honey–water system (Figure 3) allows for the determination of the concentration of the maximally freeze-concentrated solution. From the crossing point of the glass transition temperature curve with the T_g' -line, for C_g , we obtain approximately “102%”, which can be interpreted by considering the intrinsic water content of genuine honey (in the order of 20%), or in other words, the pure honey, after the extraction of water of approximately 2 wt %, will be similar to its maximally freeze-concentrated solution. For honey 4, the glass transition temperature for the samples with 100 and 90% honey content were -47.9 and -65.3 °C, respectively, and $T_g = -41.9$ °C (Table 1) and for C_g a value of 103% could be estimated. The same value, approximately 103% can be estimated from the T_g values measured by Rubin et al. for a different (commercially available) honey (Rubin et al., 1990).

It should be emphasized again, that while the glass transition temperature, T_g is a function of both moisture content and kind of solute, the temperature of the glass transition of the maximally freeze-concentrated solution, T_g' , is characteristic of the kind of solute only. The measurement of T_g alone of the honey sample is thus not sufficient for its characterization; C_g as derived from the state diagram is indicative of moisture content, and T_g is typical of the sugar profile of the given honey sample. While T_g varied in a wide range (between -42 and -51 °C for the measured samples accepted as unadulterated), C_g appeared to be practically constant.

It is usually desirable to determine the moisture content of honey as a quality factor. Clearly, the value of C_g measured by means of a calorimeter can be translated into moisture content data, since for mono- and disaccharides, almost independent of the kind of material, the solute content of the maximally freeze-concentrated solution lies in a narrow range between approximately 80 and 81.6% [see, e.g., Roos (1993)]. For example, for a honey characterized by $C_g = 102\%$, the

moisture content is approximately 20–21.6%. So far, it is not clear whether the above-described calorimetric method can compete with other methods used for moisture content determination, e.g., viscosimetry, gravimetry, or refractometry.

CONCLUSIONS

The glass transition of pure and diluted honey and the glass transition of the maximally freeze-concentrated solution of honey were investigated by differential scanning calorimetry (DSC). The glass transition temperature of the pure honey samples accepted as unadulterated varied between -42 and -51 °C. Dilution of honey to 90 wt % honey content resulted in a shift of the glass transition temperature by -13 to -20 °C. The concentration of the maximally freeze-concentrated honey solutions as expressed in terms of honey content is approximately 102–103%, i.e., slightly more concentrated in sugars than honey itself. The application of DSC measurements of T_g and C_g in characterization of honey may be considered, but requires systematic study on a number of honeys.

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