

Edible Wheat Gluten Film: Influence of Water Content on Glass Transition Temperature

N. Gontard^{*,†} and S. Ring[‡]

ENSIA-SIARC/CIRAD-SAR, B.P. 5098, 34032 Montpellier Cedex, France, and IFR/BBSR 2, Colney Lane, Norwich NR47 UA, United Kingdom

The depression of the glass transition temperature of a wheat gluten film by water was studied by dynamic mechanical thermal analysis and differential scanning calorimetry. Wheat gluten film behaved as a typical amorphous polymer, and the large plasticizing effect of water did not fit simply on the prediction of the Couchman–Karasz equation. Temperature and water mass fraction, at which large changes of film mechanical and barrier properties were previously observed, are well correlated with glass transition.

Keywords: *Edible film; wheat gluten; glass transition*

INTRODUCTION

Functional properties of edible or biodegradable films from food biopolymers are highly influenced by numerous parameters, such as film-forming technology, solvent characteristic, additives content, etc. (Guilbert, 1986; Kester and Fennema, 1986; Guilbert and Biquet, 1989). Because of their hydrophilicity, film properties are also highly dependent on usage conditions such as relative humidity and temperature (Kamper and Fennema, 1984; Biquet and Labuza, 1988; Rico Pena and Torres, 1991; Gontard et al., 1993). Water is the most ubiquitous and uncontrollable plasticizer. Until recently, this temperature and water content dependence was only interpreted in terms of water activity or disruptive water–polymer hydrogen bonding in a polymer hydrogen-bonded network.

The structure–properties relationships of hydrated biopolymers could be better understood with the theories of glass transition used in polymer science; that is, in terms of critical variables of time, temperature, and diluent content (Slade, 1984; Levine and Slade, 1987, 1991; Slade et al., 1989). Glass transition of an amorphous or partially amorphous polymer is typically described as a transition from a brittle glass to a highly viscous or a liquid-like rubber (Levine and Slade, 1991; Allen, 1993). The glass transition separates two domains according to clear structural differences, and polymer properties are seriously modified when the temperature rises above the glass transition temperature (T_g). Theoretically, the addition of plasticizers such as water under isothermic conditions has the same effect as increased temperature on molecular mobility. Water, which is a low molecular weight component, increases free volume and thereby allows increased backbone chain segmental mobility.

In spite of their wide variety of structure, reflecting the diversity of the aminoacids from which they are built, it is well established that most proteins undergo a glass transition. Plasticization by water affects the T_g of amorphous or partially amorphous proteins such as caseine (Kalichevsky et al., 1993), gelatin (Yannas,

1972; Marshall and Petrie, 1980), collagen (Batzner and Kreibich, 1981), or elastin (Kakivaya and Hoeve, 1975; Scandola et al., 1981), resulting in a drop of T_g . A variety of techniques including differential scanning calorimetry (DSC), dynamical mechanical thermal analysis (DMTA), or pulsed nuclear magnetic resonance, have been used to show that wheat gluten is a highly amorphous multipolymer system that is water plasticizable but not water soluble (Hoseney et al., 1986; Slade, 1984; Slade et al., 1989; Kalichevsky et al., 1992).

In previous investigations (Gontard et al., 1991, 1992, 1993), an edible wheat gluten film was developed, and the effects of film-forming conditions, plasticizer content (glycerol), additives, relative humidity, and temperature on various film properties (water vapor permeability, mechanical, sorption properties) were studied. In the present study, the T_g of wheat gluten film is studied as a function of water content by DSC and DMTA to gain a better understanding and control of the plasticizing effect of water on the film.

MATERIALS AND METHODS

Wheat Gluten Film Preparation. A film-forming solution was prepared with gluten (Amylum Aquitaine, F 33000 Bordeaux), glycerol (95%, Merck, Darmstadt, Germany), acetic acid, ethanol (Aldrich Chemie, Steinheim, Germany), and water. The gluten concentration (7.5 g/100 mL solution), glycerol concentration (20 g/100 g of gluten), ethanol concentration (45 mL/100 mL solution), and pH of the solution (4.0 adjusted with acetic acid) were chosen on the basis of results from previous investigations (Gontard et al., 1992, 1993). All components were mixed vigorously at 40 °C, and the film-forming solution was then immediately poured on a plexiglass-leveled surface and dispersed with a thin-layer chromatography (TLC) spreader (Braive, 4000 Liège, Belgium) adjusted to 0.8 mm height. The film-forming solution was dried in a ventilated oven at 30 °C to evaporate volatile solvents (ethanol, acetic acid, and water). This procedure resulted in the formation of a transparent film with a constant controlled thickness (0.050 ± 0.003 mm) that was measured at several positions with a micrometer (Roch).

Disks (for DSC measurements) or rectangles (for DMTA measurements) of film were equilibrated for 5 days at 25 °C in atmospheres with controlled relative humidities (using saturated salt solutions of known water activity ranging from 0.06 to 0.97 at 30 °C (Spiess and Wolf, 1987; Multon, 1984; Stamp et al., 1984)). The moisture content of each film was evaluated with previously determined sorption isotherms (Gontard et al., 1993).

* Author to whom correspondence should be addressed (fax 33 67 61 70 55; e-mail gontard@cirad.fr).

† ENSIA-SARC/CIRAD-SAR.

‡ IFR/BBSR 2.

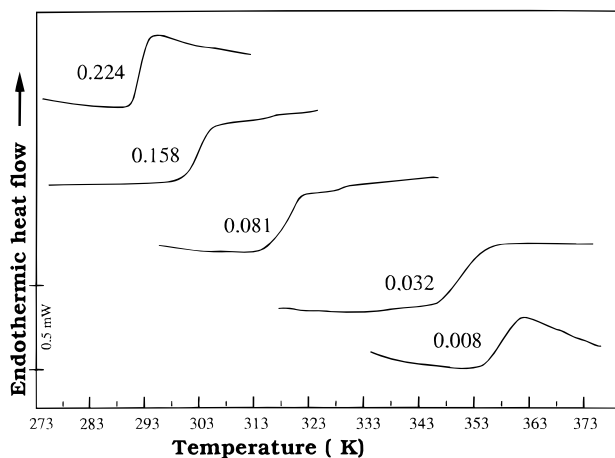


Figure 1. DSC thermogram of wheat gluten film stored at 0–90% relative humidity (water mass fraction, 0.008–0.224).

DSC Measurements. Ten milligrams of small film disks of known dry matter were piled up in open stainless-steel pans, which were designed to withstand high pressures and suppress the volatilization of water, and equilibrated at the desired relative humidity for 5 days. After equilibration, the pans were immediately and hermetically sealed. Calorimetric measurements were carried out with a Perkin Elmer DSC-4 calorimeter equipped with a thermal analysis data station. The temperature was calibrated daily with the melting points of Indium and cyclohexane II. An empty aluminium pan was used as an inert reference.

The T_g was determined from the midpoint of the heat capacity change observed at a heating rate of $10\text{ }^\circ\text{C}\cdot\text{min}^{-1}$ on at least triplicate samples, and verified from the second run after cooling to eliminate the eventual effects of sample history.

Dynamic Mechanical Thermal Analysis (DMTA) Measurements. DMTA measurements were made with a Polymer Laboratories PL-DMTA mark I apparatus (Loughborough, U.K.). Sample rectangles of film (12.0×6 mm) of known dry matter were previously equilibrated at the desired relative humidity for 5 days. After equilibration, they were immediately coated with silicon grease to avoid water evaporation and subjected to the tensile mode at a frequency of 1 Hz while the temperature was increased from -20 to $160\text{ }^\circ\text{C}$ at $2\text{ }^\circ\text{C}\cdot\text{min}^{-1}$. Sample displacement gave the storage modulus (E'), the loss modulus (E''), and $\tan \delta (= E''/E')$. The storage modulus is a measure of the stiffness or rigidity of the material and is calculated as the ratio of the stress to the applied strain. The loss modulus reflects the ability of a material to dissipate mechanical energy by converting it to heat through molecular motion. As the specific molecular groups undergo rotation at a given temperature, a peak is observed in the loss modulus data due to the absorption of the energy applied by the mechanical spectrometer. $\tan \delta$ is a useful index of material viscoelasticity (Urzendowski and Pechak, 1992).

The T_g was defined as usual by the $\tan \delta$ peak or the inflexion point of the decrease in E' on at least triplicate samples.

RESULTS AND DISCUSSION

The T_g of wheat gluten films was determined by DSC as well as by DMTA measurements. DSC measurements (Figure 1) show only a change of the baseline toward a higher value, indicating that gluten is a highly amorphous polymer undergoing a glass transition phenomenon. But the heat capacity change is relatively small (~ 5 cal/g) and induced a 10–20% experimental error (compared with a 5–8% error for DMTA measurements). The T_g , which was taken in the curves as the inflection point, decreases with increasing water content.

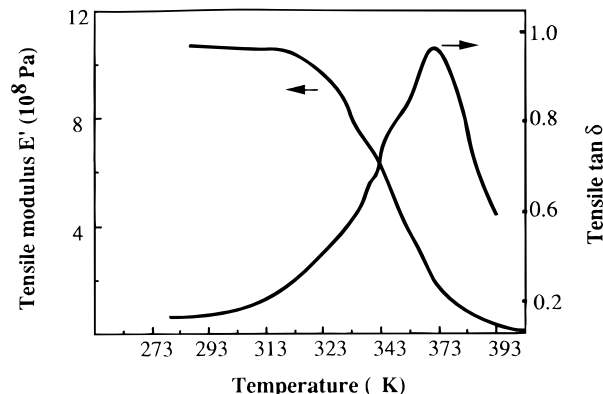


Figure 2. Typical DMTA plot for a wheat gluten film stored under 43% relative humidity (0.032 water mass fraction) showing tensile modulus (E') and $\tan \delta$ as a function of temperature.

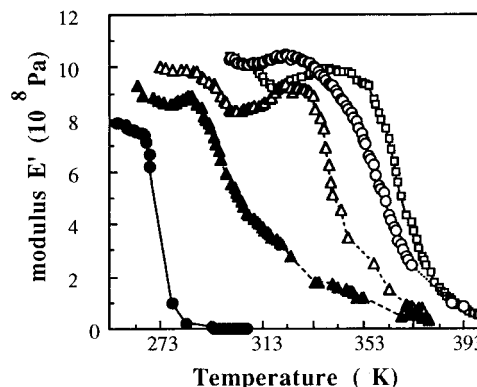


Figure 3. DMTA tensile modulus (E') as a function of temperature for wheat gluten film with 0.08 (\square), 0.032 (\circ), 0.057 (\triangle), 0.194 (\blacktriangle) and 0.292 (\bullet) water mass fraction.

Glass transition phenomenon in gluten film was confirmed by DMTA. A typical DMTA graph showing E' (elastic modulus) and $\tan \delta$ as a function of temperature is shown in Figure 2. Wheat gluten film clearly displays the behavior of an amorphous polymer: with increasing temperature, this amorphous protein traverses the glassy region, the transition region ($\tan \delta$ peak and modulus drop), and a rubbery plateau. The size of the $\tan \delta$ peak that is thought to reflect the volume fraction of the material undergoing the transition (Wetton, 1986) is similar to the typical $\tan \delta$ peak of other totally amorphous biopolymers such as elastin (Lillie and Gosline, 1993), amylopectin, gelatin, or native wheat gluten (Kalichevsky et al., 1993).

In the glassy state, the storage modulus E' displays a value of $\sim 10^9$ Pa and is insensitive to moisture content changes (Figure 3). At the glass-to-rubber transition, a characteristic decrease of 10^3 Pa is observed. These results are consistent with those for native wheat gluten or other biopolymers, such as casein (Kalichevsky et al., 1993), starch, gelatin, or ovalbumin (Kalichevsky et al., 1993; Yannas, 1972). The absence of a flow region precludes the presence of intermolecular covalent cross-bondings, such as disulfide bonds. Gluten has been described as a thermosetting polymer (Slade et al., 1989) and glutenin subunits (the higher molecular weight component of gluten) are linked by interchain disulfide bonding of cysteine residues which maintains high rubber-like modulus.

Glycerol, which was used as plasticizer, appears to be miscible with gluten at the composition used because only one transition is observed. However, the broaden-

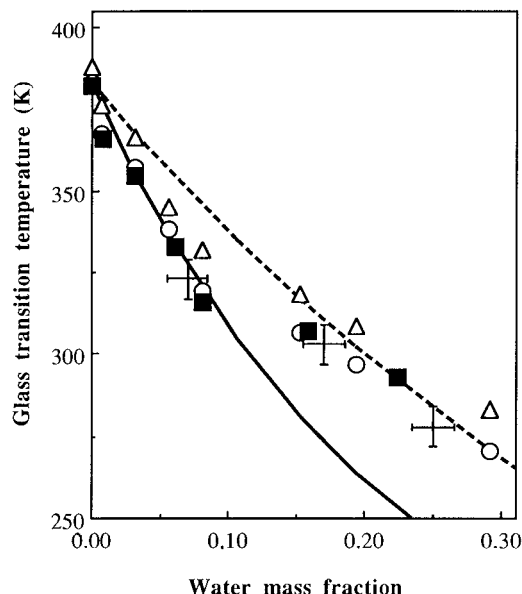


Figure 4. T_g of wheat gluten film determined by DMTA E' inflexion point (○) and $\tan \delta$ peak (Δ), and by DSC (■) as a function of water content. The solid and dashed lines are the eq 2 theoretical curve obtained for $K_1 = 2.1$ (---) and $K_1 = 4.0$ (—). The crosses indicate water mass fraction and temperature at which wheat gluten film changes of mechanical and barrier properties occur, according to Gontard et al. (1994).

ing of the transition (a few tens of °C) could suggest a lack of miscibility or simply reflect the high heterogeneity of gluten proteins molecular weights ranging from 30 000 to millions (Graveland et al., 1985; Bietz and Wall, 1972).

The T_g decreased when the water content of the film increased (Figure 3). It is interesting to note that, as observed with wheat gluten (Kalichevsky et al., 1992) or gelatin (Yannas, 1972), increasing the water content decreases the rubbery modulus (high-temperature value of E'). It has been observed that E' values above T_g are highly dependent on the crosslink density of a polymer; thus, the reduction of the rubbery modulus in the presence of a plasticizer implies reduced cross-linking. In this case, the reduction in E' may be due to polymer-polymer hydrogen bonds being replaced by labile polymer-water hydrogen bonds (Kalichevsky et al., 1992).

The inflexion point in the drop in E' and $\tan \delta$ peak are plotted again water content in Figure 4 and can be compared with DSC results. A good correspondence is obtained when DSC results are compared with the E' inflexion point. As previously reported for wheat gluten (Kalichevsky et al., 1992) or other biopolymers such as casein (Kalichevsky et al., 1993), the $\tan \delta$ peak falls above the E' inflexion point and the midpoint in the change in heat capacity observed by DSC. A simple comparison between mechanical and calorimetric determinations of T_g is not easy. Techniques are sensitive to different degrees of molecular mobility. Glass transition is not observed at a unique temperature, but is related to the frequency and nature of the measurement technique. The T_g measured by DMTA is dependent on frequency, and T_g measured by DSC depends on the heating rate used because it is a kinetically determined transition. However, the DMTA method is a more sensitive method and the glass transitions were easier to detect. Thus, DMTA results are very reproducible for high water contents (5% of experimental error), where glass transition occurred at a low temperature avoiding water loss. DSC results could be more reliable

at low water content because no water loss occurred. These two methods could thus be considered as complementary.

On increasing water content, the value of the T_g drops, at first rapidly (5 °C/1% water for 0–0.1 water mass fraction) and then more slowly (3.5 °C/1% water for 0.1–0.3 water mass fraction), indicating a large plasticizing effect of water on wheat gluten film. The sensitivity of T_g to polar diluent such as water makes it very likely that this transformation involves the regions that contain large polar residues such as Glu, Asp, Arg, and Lys. This lowering of T_g is similar to the value observed for gelatin/glycerol system (5 °C/1% water; Yannas, 1972), starch (~5 °C/1% water; Orford et al., 1989), elastin (15 °C/1% for low water content and 5 °C/1% water for 0.1–0.2 water mass fraction; Kakivaya and Hoeve, 1975), and other biopolymers (generally ~5–10 °C/1% water; Slade and Levine, 1989).

The depression of T_g by the addition of a diluent or plasticizer may be explained by a number of theoretical approaches such as free volume theories or classical thermodynamic theories proposed by Couchman and Karasz (1978). A thermodynamic consideration of the entropies of the components at the T_g of the mixtures leads to the following equation for a three-component mixture:

$$T_g(\text{mixture}) = \frac{W_1 \Delta C_{p1} T_{g1} + W_2 \Delta C_{p2} T_{g2} + W_3 \Delta C_{p3} T_{g3}}{W_1 \Delta C_{p1} + W_2 \Delta C_{p2} + W_3 \Delta C_{p3}} \quad (1)$$

In eq 1, the subscript 1 refers to water, the subscript 2 refers to wheat gluten, and the subscript 3 refers to glycerol, W is the weight fraction of each component, and ΔC_p is the change in heat capacity observed at T_g . This equation requires values for the ΔC_p of water that have been subject of considerable debate (Kalichevsky et al., 1992). Because there is one less constant and no need for water ΔC_p , the Gordon–Taylor equation (Gordon and Taylor, 1952) has been more easily and successfully used to fit T_g data of various biopolymers such as starch (Roos and Karel, 1991). But, this latter equation cannot be extended to more than bicomponent system. Thus, to obtain a better fit to the predicted T_g of mixture, the following equation, which is equivalent to eq 1 but avoids the use of water ΔC_p , was used:

$$T_g = \frac{W_1 K_1 T_{g1} + W_2 T_{g2} + 0.2 W_2 K_2 T_{g3}}{W_1 K_1 + W_2 + 0.2 W_2 K_2} \quad (2)$$

In eq 2, $0.2 = W_3/W_2$ (equal to the constant weight ratio of glycerol/gluten), $K_1 = \Delta C_{p1}/\Delta C_{p2}$, and $K_2 = \Delta C_{p3}/\Delta C_{p2}$. In addition, K is a constant that is proportional to the plasticizing effect of diluent (water for subscript 1 or glycerol for subscript 3) on the polymer (wheat gluten with the subscript 2).

For dry film, $W_1 = 0$, and eq 2 becomes

$$T_{g(\text{mixture})} = T_{g2} + 0.2 K_2 T_{g3} / 1 + 0.2 K_2 \quad (3)$$

Using $T_{g2} = 435$ K and $T_{g3} = 180$ K (from Kalichevsky et al., 1992), and $T_{g(\text{mixture})} = 384$ K (dry film T_g determined in the present study), K_2 was calculated from eq 3. The K_2 value thus obtained was 1.28, which is a lower value than the one calculated (2.26) with dry gluten and glycerol ΔC_p (respectively, $\Delta C_{p2} = 0.39$ and $\Delta C_{p3} = 0.88$ J·g⁻¹·K⁻¹) from Kalichevsky et al. (1992). This result means that wheat gluten proteins were

plasticized by glycerol but not as much as predicted. Similarly, but to a greater extent, observations have been made on the plasticizing effects of fructose on sodium caseinate (Kalichevsky et al., 1993), or of fructose, sucrose, and glucose on wheat gluten (Kalichevsky et al., 1992). It seems that the incomplete compatibility of sugar and proteins was responsible for this phenomenon. Glycerol appears to be more miscible with gluten than sugars, because of its lower molecular weight, but the compatibility is not complete and could explain the deviation from the Couchman–Karasz theory.

The DSC and DMTA data were fitted with eq 2, with K_1 as a variable. As shown in Figure 4, eq 2 could not be easily correlated with the experimental curve with a unique K_1 that represents the ratio of water ΔC_p to wheat gluten ΔC_p . With $K_1 = 4.0$, a very good correlation was obtained for water mass fraction of <0.1 . This relatively high K_1 value required for eq 2 to fit for low water content is consistent with results obtained by Kalichevsky et al. (1992) who found a ratio equal to 5 for wheat gluten. This value is comparable with the ratio of (4.55 for amylopectin (Orford et al., 1989), indicating that the wheat gluten film is highly water plasticizable.

The model seriously overestimates the plasticizing effect of water for higher water fraction. Use of $K_1 = 2.1$ provided a better fit to the data for water mass fraction between 0.15 and 0.3 (Figure 4). This value is equivalent to the results obtained by Kalichevsky et al. (1993) for slightly water plasticizable proteins such as casein (2.3) or sodium caseinate (1.3).

During film hydration, the T_g became progressively more independent of water content increase. A simple correlation with Couchman–Karasz theory is expected when the diluent mixes homogeneously with the polymer to form a single-phase system at any diluent content, and thus when polymer is soluble in the diluent to fit the equation for very high water content. The formation of various degree of protein/water interactions during hydration and the wheat gluten film insolubility in water (Gontard et al., 1994) could explain the discrepancy between the theory and the experimental results.

Recently, Schneider et al. (1993) demonstrated that in the depression of hydrophilic polyurethanes, T_g is controlled by the amount of nonfreezing bound water, with very little contribution from the other forms of water. For one specific type of polyurethane, the simple Fox mixing equation was more reliable than the available free volume approach.

But, as efficiently demonstrated by Slade et al. (1989), the “free” and “bound” physical states of water in food materials are ill-defined and oversimplified, especially in the context of proteins hydration. Many experimental methods (such as DSC or nuclear magnetic resonance) lead to fundamental disparity between results, and two, three, or more discrete physical state of water could be observed for a same product. This discrepancy indicates the need for more information on the state of water and the depression of the T_g in such complex biopolymers. In addition to DSC and DMTA measurements, solid-state nuclear magnetic resonance could be useful in defining the state of water and its effect on polymer mobility.

In a previous investigation (Gontard et al., 1993), the effect of water and temperature on the functional

properties of the same wheat gluten film were studied. During the hydration of the film, a dramatic decrease in puncture strength and elasticity, and an increase of extensibility and water vapor transmission rate were observed at 5, 30, and 50 °C for water mass fractions of ~ 0.25 , ~ 0.15 , and ~ 0.07 , respectively. These results were discussed in terms of disruptive water–polymer hydrogen bonding, but the highly discontinuous behavior could not be explained.

The values in Figure 4 are well correlated with glass transition, especially with DSC measurements and E' inflection points for DMTA measurements. This correlation suggests that the important change of film functional properties could be attributed to a change from the glassy to the rubbery state. The anomalous diffusion behavior of glassy polymers may be directly related to the influence of the changing polymer structure on the solubility of the penetrant and its diffusional mobility (Crank, 1975), and the dependence of the ultimate mechanical properties (rupture stress and strain) on temperature and rate of deformation is similar to that of viscoelastic properties and therefore reflects the configurational rearrangement rates (Ferry, 1980).

A glass transition could thus explain the discontinuous behavior of the gluten film properties in response to changes in moisture content and/or temperature. Below the glass transition, wheat gluten films retain relatively good mechanical and barrier properties and should thus be used under these conditions of temperature and water content.

LITERATURE CITED

- Allen, G. A history of the glassy state. In *The Glassy State in Foods*; Blanshard, J. M. V., Lillford P. J., Eds.; Nottingham University: Nottingham, U.K., 1993; pp 1–12.
- Batzer, H.; Kreibich, U. T. Influence of water on thermal transitions in natural polymers and synthetic polyamides. *Polym. Bull.* **1981**, *5*, 585–589.
- Bietz, J. A.; Wall, J. S. Wheat gluten subunits: molecular weights determined by sodium dodecyl sulfate polyacrylamide gel electrophoresis. *Cereal Chem.* **1972**, *49*, 416–430.
- Biquet, B.; Labuza, T. P. Evaluation of the moisture permeability of chocolate films as an edible moisture barrier. *J. Food Sci.* **1988**, *53*, 989.
- Crank, J. Non-fickian diffusion. In *The Mathematics of Diffusion*, 2nd ed.; Clarendon Press: Oxford, U.K., 1975.
- Couchman, P. R.; Karasz, F. E. A classical thermodynamic discussion of the effect of composition on glass transition temperatures. *Macromolecules* **1978**, *11*, 117–119.
- Ferry, J. D. Ultimate mechanical properties. In *Viscoelastic Properties of Polymers*; Ferry, J. D., Ed.; Wiley: New York, 1980; pp 583–587.
- Graveland, A.; Bosveld, P.; Lichtendonk, W. J.; Marseille, J. P.; Moonen, J. H. E.; Scheepstra, A. A model for the molecular structure of the glutenins from wheat flour. *J. Cereal Sci.* **1985**, *3*, 1–16.
- Gontard, N. Films et enrobages comestibles: étude et amélioration des propriétés filmogènes du gluten. Ph.D. Thesis, Université de Montpellier II, France, 1991.
- Gontard, N.; Guilbert, S.; Cuq, J. L. Edible wheat gluten films: influence of the main process variables on film properties using response surface methodology. *J. Food Sci.* **1992**, *57* (1), 190–195.
- Gontard, N.; Guilbert, S.; Cuq, J. L. Water and glycerol as plasticizers affect mechanical and water vapor barrier properties of an edible wheat gluten film. *J. Food Sci.* **1993**, *58* (1), 206–211.
- Gordon, M.; Taylor, J. S. Ideal polymer and the second order transition of synthetic rubber. I. Non-crystallin copolymers. *J. Appl. Chem.* **1952**, *21*, 447–451.

- Guilbert, S. Technology and application of edible protective films. In *Food packaging and Preservation: Theory and Practice*; Mathlouthi, M., Ed.; Elsevier Applied Science: New York, 1986; Chapter 19, pp 371–394.
- Guilbert, S.; Biquet, B. Les films et enrobages comestibles. In *L'Emballage des Denrées Alimentaires de Grande Consommation*; Bureau, G., Multon, J. L., Eds.; Technique et Documentation, Lavoisier, Apria, 1989; Chapter 22.
- Hoseney, R. C.; Zeleznak, K.; Lai, C. S. Wheat gluten: a glassy polymer. *Cereal Chem.* **1986**, *63*, 285–286.
- Kakivaya, S. R.; Hoeve, C. A. The glass point of elastin. *Proc. Natl. Acad. Sci. U.S.A.* **1975**, *72*, 9, 3505–3507.
- Kalichevsky, M. T.; Jaroszkiewicz, E. M.; Blanshard, J. M. V. Glass transition of gluten. 1: Gluten and gluten-sugar mixtures. *Int. J. Biol. Macromol.* **1992**, *14*, 257–266.
- Kalichevsky, M.; Blanshard, J.; Tokarczuk, P. Effect of water and sugars on the glass transition of casein and sodium caseinate. *Int. J. Food Sci. Technol.* **1993**, *28*, 139–151.
- Kamper, S. L.; Fennema, O. R. Water vapor permeability of an edible, fatty acid, bilayer film. *J. Food Sci.* **1984**, *49*, 1482–1485.
- Kester, J. J.; Fennema, O. Edible films and coatings: a review. *Food Technol.* **1986**, *40*, 12–47.
- Levine, H.; Slade, L. Structural stability of intermediate moisture foods—a new understanding? In *Food Structure—Its Creation and Evaluation*; Mitchell, J. R., Blanshard, J. M. V., Eds.; Butterworth: London, 1987; pp 115–123.
- Levine, H.; Slade, L. Beyond water activity: recent advances based on an alternative approach to the assessment of food quality and safety. *Crit. Rev. Food Sci. Nutr.* **1991**, *30*, 115–360.
- Levine, H.; Slade, L. Glass transition in foods. In *Physical Chemistry of Foods*; Schwartzberg, H. G., Hartel, R. W., Eds.; Dekker: New York, 1992; pp 83–221.
- Lillie, M. A.; Gosline, J. M. The effect of swelling solvents on the glass transition in elastin and others proteins. In *The Glassy State in Foods*; Blanshard, J. M. V., Lillford, P. J., Eds.; Nottingham University Press: Nottingham, U.K., 1993; pp 281–302.
- Marshall, A. S.; Petrie, S. E. Thermal transition in gelatin and aqueous gelatin solutions. *J. Photogr. Sci.* **1980**, *28*, 128–136.
- Multon, J. L. Conclusions provisoires des travaux de la commission: Aliments à Humidité Intermédiaires du CNER-NA. *Ind. Aliment. Agric.* **1984**, *11*, 1125–1128.
- Orford, P. D.; Parker, R.; Ring, S. G.; Smith, A. C. Effect of water as a diluent on the glass transition behaviour of malto-oligosaccharides, amylose and amylopectin. *Int. J. Biol. Macromol.* **1989**, *11*, 91–96.
- Ricco-Peña, D. C.; Torres, J. A. Sorbic acid and potassium sorbate permeability of an edible methylcellulose-palmitic acid film: water activity and pH effects. *J. Food Sci.* **1991**, *56*, 497–499.
- Roos, Y.; Karel, M. Plasticizing effect of water on thermal behavior and crystallisation of amorphous food models. *J. Food Sci.* **1991**, *56*, 38–43.
- Scandola, M.; Cecorulli, G.; Pizzoli, M. Water clusters in elastin. *Int. J. Biol. Macromol.* **1981**, *3*, 147–149.
- Schneider, N. S.; Illinger, J.; Karasz, F. E. Effect of water on the glass transition temperature of hydrophilic polyurethanes. *J. Appl. Polym. Sci.* **1993**, *48*, 1723–1729.
- Slade, L. Starch properties in processed foods: staling of starch-based products. Presented at *69th Annual Meeting of American Association of Cereal Chemists*, Washington, DC, 1984; paper 112.
- Slade, L.; Levine, H.; Finlay, J. W. Protein-water interactions: water as a plasticizer of gluten and other protein polymers. In *Protein Quality and the Effects of Processing*; Phillips, R. D., Finlay, J. W., Eds.; Dekker: New York, 1989; pp 9–23.
- Spieß, W. E.; Wolf, W. Critical evaluation of methods to determine moisture sorption isotherms. In *Water Activity: Theory and Applications to Food*; Rockland, L. B., Beuchat, L. R., Eds.; Dekker: New York, 1987; pp 215–217.
- Stamp, J. A.; Linscott, S.; Lomauro, C.; Labuza, T. P. Measurement of water activity of salt solutions and foods by several electronic methods as compared to direct vapor measurements. *J. Food Sci.* **1984**, *49*, 1139–1145.
- Urzendowsky, I. R.; Pechak, D. G. Characterization of food packaging materials by microscopic, spectrophotometric, thermal and dynamic mechanical analysis. *Food Struct.* **1992**, *11*, 301–314.
- Wetton, R. E. Dynamic mechanical thermal analysis of polymers and related systems. In *Developments in Polymer Characterization*; Dawkins, J. V., Ed.; Elsevier Science Publishers: 1986; pp 179–221.
- Yannas, I. V. Collagen and gelatin in the solid state. *J. Macromol. Sci.—Rev. Macromol. Chem.* **1972**, *47*–49.

Received for review April 11, 1996. Accepted July 30, 1996.®

JF960230Q

® Abstract published in *Advance ACS Abstracts*, September 15, 1996.