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Mechanical *versus* calorimetric glass transition temperature in the diffusion of nicotinic acid from a condensed gelatin/glucose syrup system

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ABSTRACT

Condensed systems of 25% (w/w) bovine gelatin (Mw = 173 kDa) or 25% (w/w) fish gelatin (Mw = 60 kDa) with 59% (w/w) glucose syrup and 1% (w/w) nicotinic acid to a total solids level of 85% (w/w) were prepared in this study. Analysis was carried out using Fourier transform infrared spectroscopy (FTIR), wide angle X-ray diffraction (WAXD), scanning electron microscopy (SEM), differential scanning calorimetry (DSC), and small deformation dynamic oscillation in-shear. Work allowed elucidation of the micromolecular and macromolecular properties focusing on the estimation of the mechanical (T_{gm}) and calorimetric (T_{gc}) glass transition temperature of these systems. A combined protocol of free volume and reaction rate theories was considered to rationalise results in the vicinity of the glass transition region. Following this, a diffusion procedure of nicotinic acid was set up over a wide temperature range (from 5 to −40 °C) using UV–*vis* spectroscopy and an appropriate chromogenic reaction to detect the presence of the bioactive compound. Time dependence of the mass transfer of nicotinic acid throughout the delivery vehicle of gelatin/co-solute was followed using diffusion theory. The treatise unveiled the correlation between structural relaxation of the vitrified matrix and release kinetics of the diffusant.

1. Introduction

Often, processed food is regarded as amorphous, making the transformation from the rubbery to glassy consistency a key factor affecting the stability and quality of the end product (Le Meste, Champion, Roudaut, Blond, & Simatos, 2002; Slade & Levine, 1991). Rapid cooling encourages vitrification around the so-called glass transition temperature (T_g), which results in limited physicochemical, enzymatic and biological activity in materials (Gray, Bowen, Farhat, & Hill, 2008; Roudaut, Simatos, Champion, Contreras-Lopez, & Le Meste, 2004). Characterisation of the molecular mechanisms during vitrification is carried out by several experimental tecnhiques that can lead to a variation in the obtained estimates of the glass transiton temperatrure (Chaudhary, Panyonyai, Small, Shanks, & Kasapis, 2017; Zhong, Tan, & Langrish, 2019). To provide a bird's eye view of all the molecular mechanims contributing to this type of physical behaviour, parallel application of mechanical and calorimetric techniques is desirable (Abiad, Carvajal, & Campanella, 2009).

Adoption of a mechanical approach requires monitoring the viscoelastic response of a polymer arising from motions of distinct molecular moieties. These can encompass long length scales due to the entire molecular chain and its association with neighbouring molecules to form a three-dimensional structure (Kasapis, Al-Marhoobi, & Mitchell, 2003a; Paramita, Bannikova, & Kasapis, 2016). A polymer with a high molecular weight will have a low specific volume and thus results in a high glass transition temperature (Abiad et al., 2009). Differential scanning calorimetry, on the other hand, monitors the heat flow of a material at a given heating rate, which can be used to estimate its heat capacity. This is then sensitive to the total concentration of solids the sample is made of, which determines the amount of heat needed to raise its temperature one degree centigrade (Kasapis & Sablani, 2005). The functionality of a material to form active "structural ties" or inactive "dead ends", according to the cascade formalism in the context of gelation, is a secondary consideration in the derivation of the calorimetric glass transition temperature.

Amongst the hydrocolloids used in the food and pharmaceutical industries, gelatin maintains its popularity due to biocompatibility, unique melt-in-the-mouth property and formation of a rubbery/glassy network with vivid clarity (Deszczynski, Kasapis, MacNaughton, & Mitchell, 2002; Kasapis, Al-Marhoobi, Deszczynski, Mitchell, & Abeysekera, 2003b). Besides the mammalian gelatin, fish gelatin produced by the partial hydrolysis of collagen-rich fish skin or scale, has been utilised in

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product development to address dietary (vegetarian/vegan) or religious (in particular, Muslim and Hindu) habits/concerns, and allow utilisation of some of the by-products of the fishing industry. It contains a high content of imino acids and is commonly produced at a relatively low molecular weight yielding distinct physical properties from the mammalian counterpart (Avena-Bustillos et al., 2006; Pang, Deeth, Yang, Prakash, & Bansal, 2017; Sila, Martinez-Alvarez, Krichen, Gómez-Guillén, & Bougatef, 2017; Zhou, Mulvaney, & Regenstein, 2006).

In addition to the structural characteristics of amorphous polymers, there is an increasing interest in the rubber-to-glass dispersion with emphasis on the release of bioactive compounds. These are delivery systems in the shape of slabs, cylinders and spheres used in the controlled release of drugs, nutrients, antioxidants, fragrances, etc. (Hoare & Kohane, 2008). They have been researched in terms of the time and temperature of transport, and the chemistry, size, morphology and swellability/solubility of the material (Paramita & Kasapis, 2019; Zarzycki, Modrzejewska, & Nawrotek, 2010). Thus, the effect of molecular weight on the retardation of caffeine release has been reported for protein-polysaccharide matrices under acid, bile and yogurt storage conditions (Hernández-Marín, Lobato-Calleros, Román-Guerrero, Alvarez-Ramirez, & Vernon-Carter, 2016).

Nicotinic acid (also known as vitamin B3) is an essential vitamin, which plays a significant role in regulating metabolism, including the digestive system, human skin and mental health (Gehring, 2004). Lack of nicotinic acid in daily intake leads to severe photosensitive dermatitis, called pellagra (Bui, Small, & Coad, 2013; Gehring, 2004). Nicotinic acid can be found in both animal and plant products, such as milk, fish, beef, eggs, cereals and fruits (Lešková et al., 2006; Panyonyai, Bannikova, Small, Shanks, & Kasapis, 2016). However, a proportion of nicotinic acid is lost during boiling, pressure cooking, microwaving and stir-frying. Upon food preparation, retention of the vitamin is reported to be from 45 to 60% (Lešková et al., 2006) and is one of the most commonly added "fortification vitamins" found in foods.

The present work researches two types of gelatin with distinct molecular weight and physicochemical fingerprints impacting structural properties. They are utilised in a high-solid preparation in the presence of glucose syrup to function as entrapping and then delivery vehicles of nicotinic acid. The working protocol allows elucidation of the fundamental relationship between kinetics of the bioactive compound release and physicochemical characteristics of the polymeric system. This is achieved in terms of the concepts of glass transition temperature, apparent diffusion coefficient and coupling parameter between supporting matrix and diffusant.

2. Materials and methods

2.1. Materials

Type B bovine gelatin (SKU G9382), bloom = 225, $M_w \approx 173$ kDa, and fish gelatin from cold-water fish skin (SKU G7041), bloom = 85, $M_w \approx 60$ kDa, were purchased from Sigma-Aldrich Co. (Sydney, Australia). Glucose syrup, as the co-solute, was supplied by Edlyn Foods Pty. Ltd. (Victoria, Australia). The product has a total solids level of 81% (w/w), with the dextrose equivalent (DE) being ≈ 42. Nicotinic acid ($C_6H_5NO_2$, M_w = 123.11 Da, purity ≥ 99.5%) and ethanol (≥99.5%) were both obtained from Sigma-Aldrich (Sydney, Australia) and used as the diffusant and release medium, respectively.

2.2. Sample preparation

Gelatin solutions were prepared by hydrating 10% (w/w) bovine or fish gelatin in Milli-Q water type II overnight at 4 °C. The protein mixture was heated to 55 °C with constant stirring for 30 min until a clear solution was obtained. Meanwhile, the glucose syrup was tempered at 55 °C before adding to the gelatin solution. The gelatin/glucose syrup mixture was continuously stirred at 55 °C for another 10 min to obtain a homogenous mixture with the desired level of solid being 25% (w/w) gelatin plus 60% (w/w) glucose syrup. Similarly, samples were made with 1% (w/w) nicotinic acid, 25% (w/w) gelatin plus 59% (w/w) glucose syrup to reach a total level of solids of 85% (w/w). Single preparations of 25% (w/w) bovine or fish gelatin and 85% (w/w) glucose syrup were also prepared to serve as the reference systems.

2.3. Experimental analysis

Fourier transform infrared spectroscopy (FTIR): Measurements were performed by using Spectrum Two GladiATR-Fourier transform infrared spectroscopy (Perkin Elmer, Pike Technologies, Norwalk, US). A single preparation of bovine or fish gelatin, glucose syrup and their mixtures with nicotinic acid were scanned at 600-4000 cm^{-1} with resolution of 4 cm^{-1} averaged over 64 scans. Each experiment was repeated three times resulting in overlapping interferograms.

Wide angle X-ray diffraction (WAXD): Measurements were performed with a Bruker D4 Endeavour (Karlsruhe, Germany). Samples of gelatin, glucose syrup and their mixtures with nicotinic acid were freeze-dried prior to analysis. They were then loaded onto the observation chamber and exposed to an accelerated voltage of 40 kV and current at 40 mA. Data obtained were in the 2θ range of 5–90° with measuring intervals of 0.1° and were processed using DIFFRACplus Evaluation (Eva), version 10.0, revision 1. Each test was repeated three times yielding identical diffractograms.

Differential scanning calorimetry (DSC): Measurements were performed on the gelatin/glucose syrup/nicotinic acid systems using Q2000 (TA instruments, New Castle, DE) with a refrigerated cooling system (RCS90). About ten mg of the sample was loaded onto the T_{zero} aluminium pans and sealed hermetically with an empty pan serving as the reference. Samples were equilibrated at 60 °C for 20 min followed by cooling from 60 to −90 °C and heating to 60 °C. The scan rate was 1 °C/min with a modulation rate of 0.53 °C for every 40s. During experimentation, nitrogen gas was purged into the cell chamber at a rate of 50 mL/min. Each run was repeated three times yielding consistent thermograms.

Dynamic oscillatory measurements: These were conducted on AR-G2 rheometer (TA Instruments, New Castle, DE). Single preparations and mixtures of gelatin with glucose syrup were loaded onto the preheated Peltier plate at 60 °C using a 10 mm parallel-plate measuring geometry. Silicone oil (BDH, 50 cS) was applied at the edges of the measuring geometry to minimize moisture loss. Samples were cooled to −50 °C at a constant rate of 1 °C/min, angular frequency of 1 rad/s and 0.01% of strain, which is within the polymer's linear viscoelastic region (LVR). Once the cooling run was complete, they were heated at the same rate and frequency sweeps were taken at 0.1–100 rad/s every 3 °C. Mechanical spectra were subjected to a time-temperature superposition (TTS) to construct the master curve of viscoelasticity for these systems. Isochronal and isothermal routines were carried out in triplicate returning consistent results.

Scanning electron microscopy (SEM): Single systems of gelatin and their mixtures with glucose syrup and nicotinic acid were freeze-dried prior to experimentation. They were coated with iridium and analyzed under the intense electron beam of FEI Quanta 200 SEM (Hillsboro, Oregon, USA). Analysis was carried out under high vacuum conditions of 0.6 Torr with a working distance of 10 mm at 1500 times magnification. Topography images were taken using accelerated medium voltage of 15 kV and spot size of 3.5. Multiple microscopy images were attempted and typical outcomes are shown here.

Diffusion studies: Two grams of samples containing gelatin, glucose syrup and nicotinic acid were prepared in beakers with 4 cm diameter and 0.5 cm thickness. These, and 4 ml of ethanol in a separate test tube, were equilibrated overnight at the temperatures of −40, −30, −20, −10, 0, and 5 °C; internal sample temperatures were confirmed using a digital

syrup to a total level of solids of 85% (w/w). Samples were both cooled and heated at a constant rate of 1 °C/min to unveil step changes in the variation of heat flow over an extensive temperature range of about eighty degrees centigrade. We have defined the calorimetric glass transition temperature (T_{gc}), an empirical index of convenience, as the midpoint of the sigmoidal curve of the exothermic or endothermic energy transition obtained from the cooling and heating runs.

The T_{gc} values of gelatin/glucose syrup were higher than those in the presence of nicotinic acid. Thus, they dropped from −25 °C to −31 °C in the mixtures of bovine gelatin with the bioactive compound, and from −39 °C to −43 °C in the corresponding mixtures of fish gelatin (Fig. 2). It appears that the relatively low molecular weight of nicotinic acid (123.11 Da) has a plasticizing effect that increases the overall mobility of the system leading to a decrease in its glass transition temperature. In addition, the higher molecular weight of bovine gelatin resulted in a T_{gc} value of −25 °C, which is fourteen degrees centigrade above that of fish gelatin in the presence of glucose syrup (−39 °C). The antiplasticising effect of increasing molecular weight of a polymer is known in the literature and in the case of gelatin that would restrict the rotation and vibration of the side chains of the protein (Abiad et al., 2009; Jadhav, Gaikwad, Nair, & Kadam, 2009).

As for the earlier physicochemical characterisation, we pursued in this section a complimentary methodology in the form of dynamic oscillation in-shear. Fig. 3a illustrates the viscoelastic transformation of 25% (w/w) bovine and fish gelatin with 60% (w/w) glucose syrup over a wide temperature range of 50 to −40 °C. In both cases, the storage (G') and loss (G'') modulus are recorded at the same scan rate as for the DSC work (1 °C/min). Flow occurs at temperatures above 50 °C in the preparations of bovine gelatin followed by a steep rise in both viscoelastic functions. Storage modulus trace is relatively flat and well over that of loss modulus at temperatures above 0 °C signifying the rubbery plateau in the fish gelatin system. The considerable rise in both moduli at lower temperatures is the mechanical signature of the glass transition region. Finally, at the end of the cooling ramp, the G' trace levels off to almost 10^8 Pa and that of G'' descends rapidly. Clearly, there is qualitative evidence in here of the earlier vitrification of the bovine gelatin/glucose syrup matrix compared to the fish gelatin counterpart.

To progress from the qualitative treatment of the preceding paragraph to the modeling of viscoelastic data, we implemented the principle of time-temperature superposition (*TTS* in Al-Ruqaie, Kasapis, Richadson, & Gordon, 1997). In doing so, a series of frequency sweeps from 0.1 to 100 rad/s were collected every three degrees centigrade, and

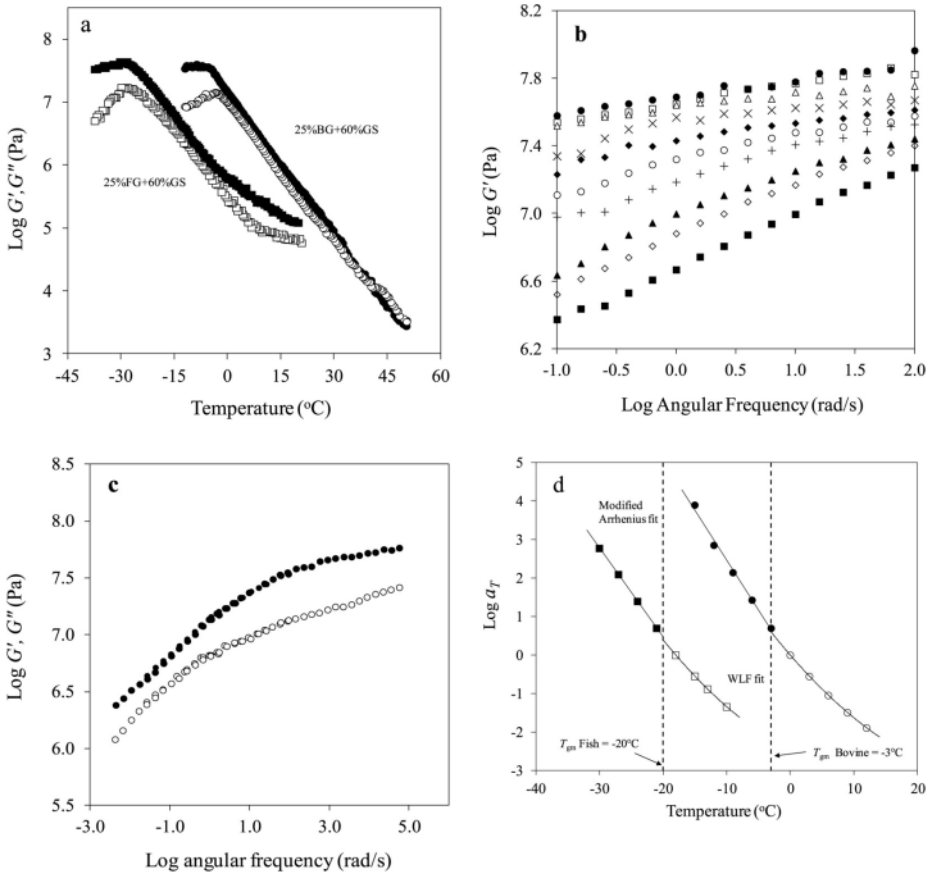

Fig. 3. **(a)** Cooling profiles of storage (G', closed symbol) and loss (G'', open symbol) modulus for 25% fish gelatin with 60% glucose syrup (square) and 25% bovine gelatin with 60% glucose syrup (circle), scanned at 1 °C/min, frequency of 1 rad/s and strain 0.01%; **(b)** mechanical spectra of 25% fish gelatin with 60% glucose syrup as a function of angular frequency (0.1–100 rad/s) for storage modulus (G') at the experimental temperatures of −10 (■), −13 (◇), −15 (▲), −18 (+), −21 (○), −24 (◆), −27 (x), −30 (Δ), −33 (●), −36 (□), arranged successively upward; **(c)** master curve of reduced shear modulus (G', ● and G'', ○) for 25% fish gelatin with 60% glucose syrup as a function of reduced angular frequency; **(d)** WLF and modified Arrhenius fits of the shift factors (a_T) within the glass transition region (open symbols) and glassy state (closed symbols) for 25% fish gelatin with 60% glucose syrup (square) and 25% bovine gelatin with 60% glucose syrup (circle), with the dash lines showing the predictions of the mechanical glass transition temperature.

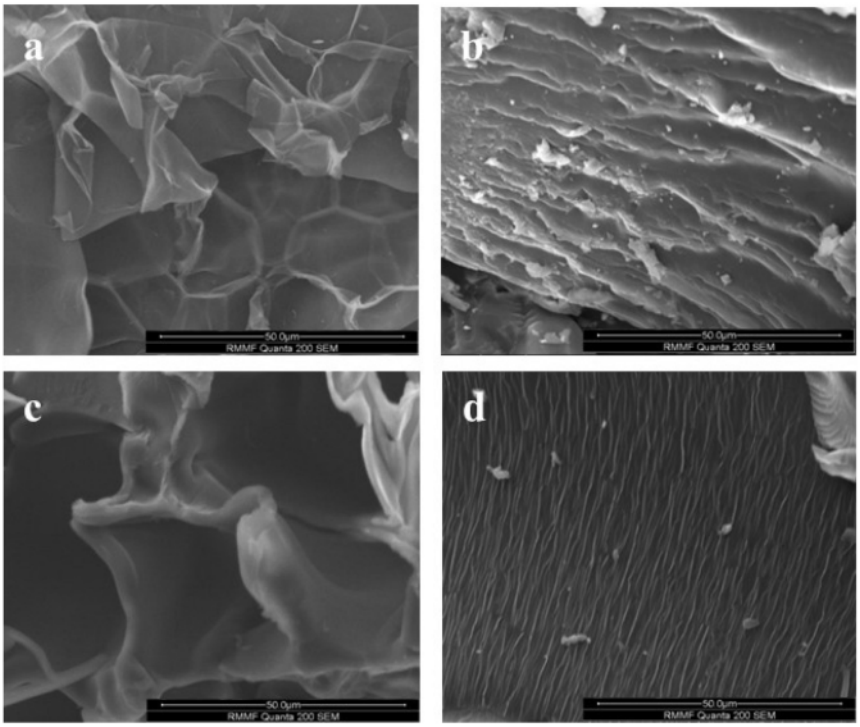

Fig. 4. SEM micrographs of **(a)** 25% bovine gelatin, **(b)** 25% bovine gelatin with 59% glucose syrup and 1% nicotinic acid, **(c)** 25% fish gelatin and **(d)** 25% fish gelatin with 59% glucose syrup and 1% nicotinic acid.

a_T, as follows (Kasapis & Shrinivas, 2010):

$$\log a_T = \log \frac{k_0}{k} \tag{4}$$

where, k_0 represents the rate constant at the reference temperature of −13 °C and −33 °C for bovine and fish gelatin, respectively. The series of spectroscopic shift factors were plotted against temperature to allow estimation, *via* the modified Arrhenius equation, of activation energies for the diffusion of nicotinic acid in bovine (25.2 kJ/mol) and fish (19.7 kJ/mol) gelatin in Table 1. These are about an order of magnitude below the tabulated E_a values of structural relaxation of the two polymeric matrices hence reflecting the high freedom of nicotinic acid mobility that underpins diffusion.

To advance further the direct comparison of molecular dynamics between polymeric matrices with glassy consistency and bioactive compound transport, we made a start with the power-law equation of Ritger and Peppas (1987):

$$\frac{M_t}{M_\infty} = kt^n \tag{5}$$

where, M_t and M_∞ denote the absolute amounts of the diffusant during experimentation and its completion, k is a constant characteristic of the bioactive compound-polymer system, t is given in seconds, and n is the kinetic diffusion exponent. For the cylindrical disk of the present investigation, n values of 0.45 and 0.89 describe Fickian and Case II diffusion kinetics, with those below, above and in-between being known as Less Fickian, Super Case II and anomalous transport (Ozdemir & Floros, 2001; Siepmann & Siepmann, 2008).

Application of equation (5) to our data, in a double logarithmic form, allows estimation of the kinetic diffusion exponent in Table 2. High n values at the low temperature end, e.g. 1.31 at −30 °C, indicate a Super Case II mechanism in the glassy state of the bovine gelatin mixture, where release kinetics are controlled by the rate-limiting polymeric relaxation. On the other hand, at 5 °C, which lies well above the bovine gelatin T_{gm} (−14 °C), an n estimate of 0.44 is obtained arguing for a Fickian diffusion as the rate-controlling process. Between the two bounds, an anomalous transport is recorded, where both mechanisms contribute equally to the release kinetics ($n = 0.66$ at −10 °C). Qualitatively, a similar profile is obtained for the fish gelatin/glucose syrup mixture, but with the anomalous transport that includes a Fickian diffusion contribution, making inroads into temperatures as low as −20 °C due to the lower T_{gm} (−34 °C). As before, within the glassy state, structural relaxation of the matrix, as opposed to Fickian diffusion, is the predominant mechanism of nicotinic acid (Super Case II) transport.

To estimate the apparent diffusion coefficient, D, absorbance readings were converted to diffused nicotinic-acid concentrations and Fick's second law was employed for $M_t/M_\infty \leq 0.4$ (Busk & Labuza, 1979; Siepmann & Siepmann, 2012):

$$\frac{M_t}{M_\infty} = 4\left(\frac{Dt}{\pi R^2}\right)^{1/2} - \frac{Dt}{R^2} \tag{6}$$

where, R represents the radius of the cylindrical disk. We were then able to utilise the apparent diffusion coefficient of the bioactive compound in a recently derived mathematical expression that argues for a linear relationship with a function of the fractional free volume of the polymeric matrix, as follows (Panyoyai & Kasapis, 2016):

$$\log D_T = \log D_{T_g} + \frac{\xi}{2.303}\left[\frac{1}{f_g} - \frac{1}{f}\right] \tag{7}$$

where, D_T, f and D_{Tg}, f_g are the apparent diffusion coefficient and fractional free volume at T and T_g, respectively, and ξ represents the critical

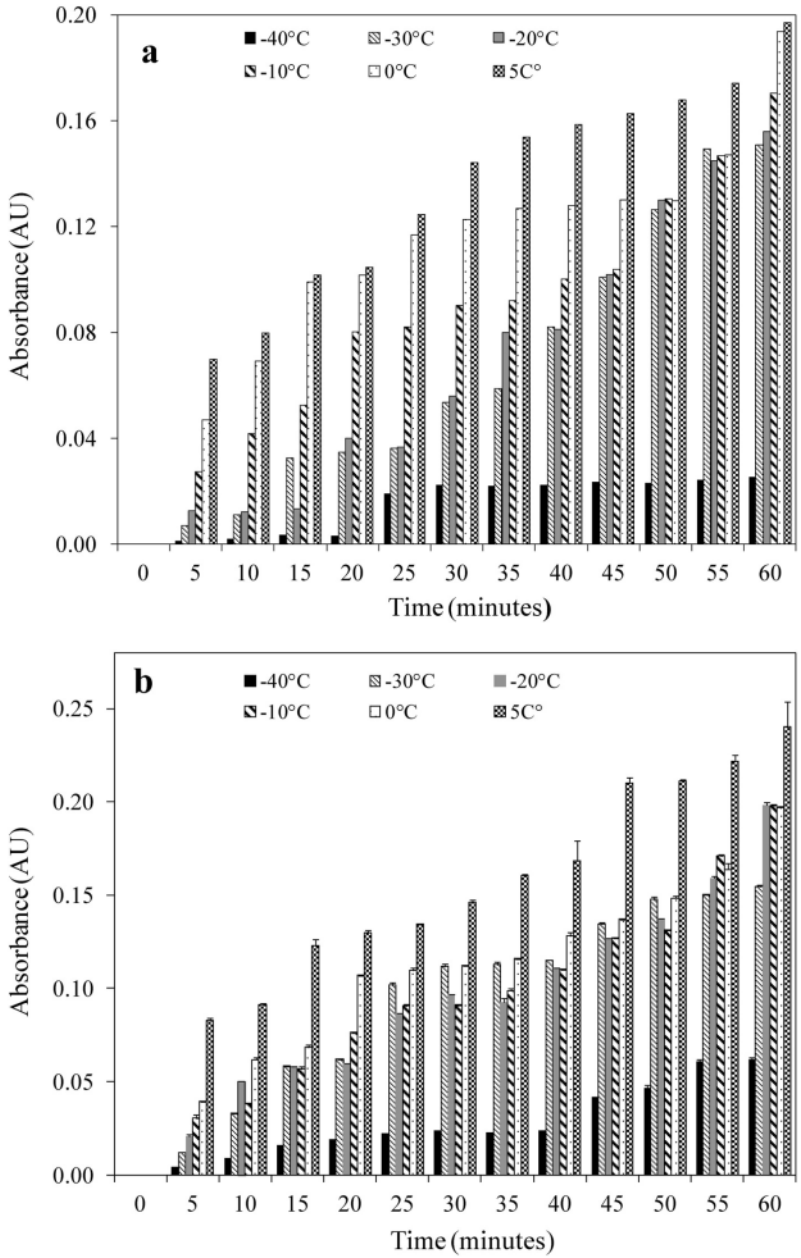

Fig. 5. Absorbance of 1% nicotinic acid following diffusion from **(a)** 25% bovine gelatin with 59% glucose syrup and **(b)** 25% fish gelatin with 59% glucose syrup as function of time at the experimental temperatures of −40 (■), −30 (▨), −20 (▩), −10 (▧), 0 (□) and 5(▩) °C obtained at 262 nm.

molecular volume of the jumping unit of a bioactive compound to that of the polymeric matrix. This relationship was found to be valid within the glass transition region of spray dried whey protein used as an encapsulant and delivery vehicle of nicotinic acid.

Utilisation of the combined protocol of equations (6) and (7) allows derivation of the apparent diffusion coefficients of nicotinic acid, which are then plotted in logarithmic form against a reciprocal function of free volume for the bovine/and fish gelatin/glucose syrup matrices (Fig. 6). Good linear relationships are obtained ($r^2 = 0.980$), which allow estimation of gradients being 0.346 and 0.137 for bovine and fish gelatin, respectively. These values reflect effectively a coupling parameter, ξ, that increases with nontrivial interactions between polymeric network and bioactive compound. They argue for a fundamental relationship between free volume, which diminishes in the connective bovine-gelatin network and high estimates of its coupling parameter. The corresponding term, 1-ξ, denotes a decoupling parameter, its values for the high-solid bovine and fish gelatin systems are 0.654 and 0.863, encompassing an estimate of 0.70 for molecularly interacting hybrid polyurethanes in biodegradable stent coating (Guo, Knight, & Mather, 2009).

Table 2
Diffusion kinetics and parameters for nicotinic acid in the gelatin/glucose syrup matrix.

Temperature (°C)	Bovine gelatin			Fish gelatin		
	Diffusion exponent (n)	Characteristic constant (k)	Diffusion mechanism	Diffusion exponent (n)	Characteristic constant (k)	Diffusion mechanism
−40	1.41	1.24E-05	Super Case II	1.02	2.06E-04	Super Case II
−30	1.31	2.16E-05	Super Case II	1.02	3.00E-04	Super Case II
−20	1.21	4.43E-05	Super Case II	0.86	7.29E-04	Anomalous
−10	0.66	3.61E-03	Anomalous	0.73	1.99E-03	Anomalous
0	0.46	1.91E-02	Fickian	0.57	9.42E-03	Anomalous
5	0.44	2.79E-02	Fickian	0.44	2.47E-02	Fickian

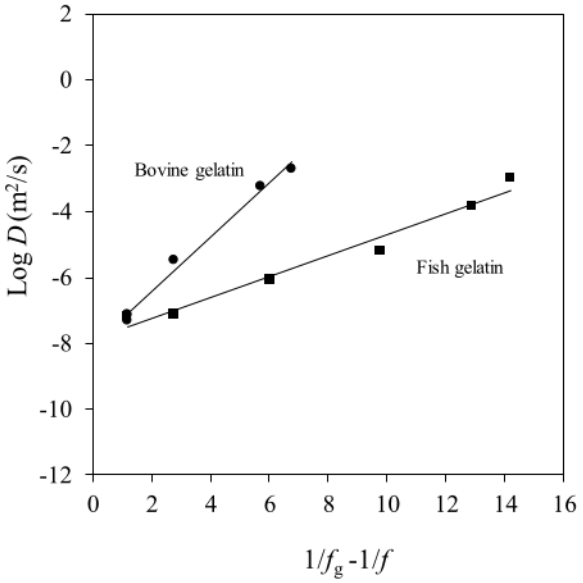

Fig. 6. Apparent diffusion coefficient against an inverse function of fractional free volume at the glass transition region for bovine gelatin (●) and fish gelatin (■).

Finally, values of the apparent diffusion coefficient and fractional free volume were plotted against the experimental temperature range covering the glass transition region and the glassy state. As shown in Fig. 7a, there is a dramatic decrease with the systematic drop in temperature in the latter, levelling off at the mechanical glass transition temperature of the bovine gelatin/glucose syrup mixture (−14 °C) and remaining constant thereafter at about 0.042, which is the predicted value of f_g. Interestingly, a similar pattern is observed for the apparent diffusion coefficient with decreasing temperature dropping from a log value of −2.90 (1.26×10^{-3} m^2/s) at 5 °C to −7.32 (4.84×10^{-8} m^2/s) at −20 °C and remaining flat at lower temperatures. Similarly, for the fish gelatin/glucose syrup counterpart, diffusion of the vitamin increased dramatically from the log value of −7.12 (7.60×10^{-8} m^2/s) at −30 °C, i.e. near its T_{gm} (−34 °C), to −3.0 (9.90×10^{-4} m^2/s) at 5 °C with a congruent increase in the fractional free volume from 0.042 to 0.052. It appears that in both cases, the calorimetric glass transition temperature, having lower values of −31 °C and −43 °C for the bovine and fish protein system with co-solute at 85% (w/w) total solids, does not influence the dynamics of the molecular processes related to volume shrinkage in the polymeric matrix and diffusion kinetics of the entrapped bioactive compound.

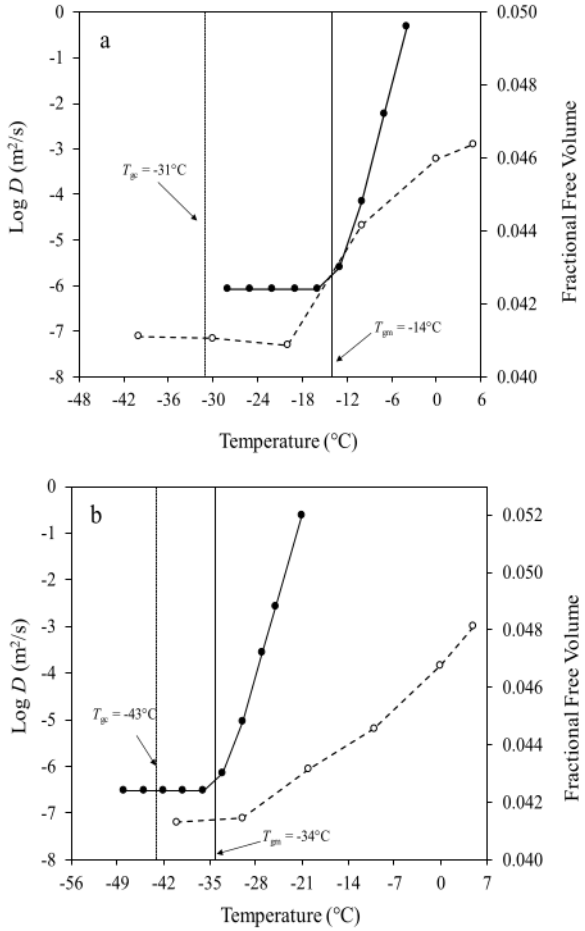

Fig. 7. Apparent diffusion coefficient (○, left y-axis) and fractional free volume (●, right y-axis) for mixtures of **(a)** 1% nicotinic acid, 25% bovine gelatin and 59% glucose syrup, and **(b)** 1% nicotinic acid, 25% fish gelatin and 59% glucose syrup.

4. Conclusions

Small deformation dynamic oscillation in-shear is an increasingly popular technique to measure the glass transition temperature of biomaterials, alongside the established practice of obtaining T_g values from differential scanning calorimetry. We have discussed in this communication the nature of mechanical and DSC T_g for condensed systems of bovine and fish gelatin in mixture with glucose syrup. The structure-forming ability of high molecular weight bovine gelatin is reflected in

a higher mechanical T_g, as compared to the fish gelatin counterpart. In both types of gelatin, estimates of the mechanical glass transition temperature are well above those of the calorimetric glass transition temperature for the chosen level of total solids in the mixture and experimental settings. The mechanical manifestation of vitrification relates to the ability of gelatin to form a network that governs the patten of nicotinic acid transport. This increasingly departs from concentration gradient (i.e. Fickian) considerations in the glassy state of the polymer and, instead, exhibits anomalous to Super Case II kinetics. Structural cooperativity between vitamin and polymer was quantified *via* the recently introduced concept of coupling parameter arguing for a strong interaction between the two constituents in the high-solid system. Rheology is well qualified to monitor network connectivity, which appears to govern molecular dynamics in the diffusion of nicotinic acid, as opposed to the calorimetric T_g.

Declaration of competing interest

The authors declare no conflict of interest.

CRediT authorship contribution statement

Diah Ikasari: Methodology, Writing - original draft. **Vilia Darma Paramita:** Supervision, Writing - review & editing. **Stefan Kasapis:** Funding acquisition, Conceptualization, Supervision, Writing - review & editing.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.foodhyd.2020.106046.

References

Abiad, M. G., Carvajal, M. T., & Campanella, O. H. (2009). A review on methods and theories to describe the glass transition phenomenon: Applications in food and pharmaceutical products. *Food Engineering Reviews, 1*(2), 105–132.
Al-Ruqaie, I. M., Kasapis, S., Richardson, R. K., & Gordon, M. (1997). The glass transition zone in high solids pectin and gellan preparations. *Polymer, 38*, 5685–5694.
Almargh, O., George, P., Bannikova, A., Katopo, L., Chaudhary, D., & Kasapis, S. (2012). Phase behaviour of gelatin/polydextrose mixtures at high levels of solids. *Food Chemistry, 134*(4), 1938–1946.
Arridge, R. G. C. (1975). The glass transition. In R. G. C. Arridge (Ed.), *Mechanics of polymers* (pp. 24–50). Oxford: Clarendon Press.
Asma, C., Meriem, E., Mahmoud, B., & Djaafer, B. (2014). Physicochemical characterization of gelatin-cmc composite edibles films from polyion-complex hydrogels. *Journal of the Chilean Chemical Society, 59*(1), 2279–2283.
Avena-Bustillos, R. J., Olsen, C. W., Chiou, B., Yee, E., Bechtel, P. J., & McHugh, T. H. (2006). Water vapor permeability of mammalian and fish gelatin films. *Journal of Food Science, 71*, E202–E207.
Bui, L. T. T., Small, D. M., & Coad, R. (2013). The stability of water-soluble vitamins and issues in the fortification of foods. In V. R. Preedy, V. R. Srirajaskanthan, & V. B. Patel (Eds.), *Handbook of food fortification and health: From concepts to public health applications* (Vol. 1, pp. 199–211). New York, NY: Humana Press, Springer.
Busk, G. C., JR., & Labuza, T. P. (1979). A dye diffusion technique to evaluate gel properties. *Journal of Food Science, 44*, 1369–1372.
Chaudhary, V., Panyoyai, N., Small, D. M., Shanks, R. A., & Kasapis, S. (2017). Effect of the glass transition temperature on alpha-amylase activity in a starch matrix. *Carbohydrate Polymers, 157*, 1531–1537.
Deszczynski, M., Kasapis, S., MacNaughton, W., & Mitchell, J. R. (2002). High sugar/polysaccharide glasses: Resolving the role of water molecules in structure formation. *International Journal of Biological Macromolecules, 30*, 279–282.
Ferry, J. D. (1980). *Viscoelastic properties of polymers*. New York: John Wiley & Sons.
Gehring, W. (2004). Nicotinic acid/niacinamide and the skin. *Journal of Cosmetic Dermatology, 3*, 88–93.
Gray, D. A., Bowen, S. E., Farhat, I., & Hill, S. E. (2008). Lipid oxidation in glassy and rubbery-state starch extrudates. *Food Chemistry, 106*, 227–234.
Guo, Q., Knight, P. T., & Mather, P. T. (2009). Tailored drug release from biodegradable stent coatings based on hybrid polyurethanes. *Journal of Controlled Release, 137*, 224–233.
Hernández-Marín, N. Y., Lobato-Calleros, C., Román-Guerrero, A., Alvarez-Ramirez, J., & Vernon-Carter, E. J. (2016). Physical properties and release behaviour of caffeine multiple emulsions stabilised by binary or ternary biopolymer soluble complexes under acid, bile and yogurt storage conditions. *Food Hydrocolloids, 58*, 42–48.
Hoare, T. R., & Kohane, D. S. (2008). Hydrogels in drug delivery: Progress and challenges. *Polymer, 49*(8), 1993–2007.
Jadhav, N. R., Gaikwad, V. L., Nair, K. J., & Kadam, H. M. (2009). Glass transition temperature: Basics and application in pharmaceutical sector. *Asian Journal of Pharmaceutics, 3*(2), 82–89.
Kasapis, S., Al-Marhoobi, I. M., & Mitchell, J. R. (2003a). Testing the validity of comparisons between the rheological and the calorimetric glass transition temperatures. *Carbohydrate Research, 338*(8), 787–794.
Kasapis, S., Al-Marhoobi, I. M., Deszczynski, M., Mitchell, J. R., & Abeysekera, R. (2003b). Gelatin vs polysaccharide in mixture with sugar. *Biomacromolecules, 4*(5), 1142–1149.
Kasapis, S., & Sablani, S. S. (2005). A fundamental approach for the estimation of the mechanical glass transition temperature in gelatin. *International Journal of Biological Macromolecules, 36*(1–2), 71–78.
Kasapis, S., & Shrinivas, P. (2010). Combined use of thermomechanics and UV spectroscopy to rationalize the kinetics of bioactive compound (caffeine) mobility in a high solids matrix. *Journal of Agricultural and Food Chemistry, 58*(6), 3825–3832.
Kavanagh, G. M., Clark, A. H., & Ross-Murphy, S. B. (2000). Heat-induced gelation of globular proteins: Part 3. Molecular studies on low pH β-lactoglobulin gels. *International Journal of Biological Macromolecules, 28*(1), 41–50.
Le Meste, M., Champion, D., Roudaut, G., Blond, G., & Simatos, D. (2002). Glass transition and food technology: A critical appraisal. *Journal of Food Science, 67*(7), 2444–2458.
Lešková, E., Kubíková, J., Kováčiková, E., Košická, M., Porubská, J., & Holčíková, K. (2006). Vitamin losses: Retention during heat treatment and continual changes expressed by mathematical models: A review. *Journal of Food Composition and Analysis, 19*, 252–276.
Nguyen, H. T. l., Katopo, L., Pang, E., Mantri, N., & Kasapis, S. (2019). Structural variation in gelatin networks from low to high-solid system affected by honey addition. *Food Research International, 121*, 319–325.
Ooi, S. Y., Ahmad, I., & Amin, M. C. I. M. (2016). Cellulose nanocrystals extracted from rice husks as a reinforcing material in gelatin hydrogels for use in controlled drug delivery systems. *Industrial Crops and Products, 93*, 227–234.
Ozdemir, M., & Floros, J. D. (2001). Analysis and modeling of potassium sorbate diffusion through edible whey protein films. *Journal of Food Engineering, 47*(2), 149–155.
Pang, Z., Deeth, H., Yang, H., Prakash, S., & Bansal, N. (2017). Evaluation of tilapia skin gelatin as a mammalian gelatin replacer in acid milk gels and low-fat stirred yogurt. *Journal of Dairy Science, 100*, 3436–3447.
Panyoyai, N., Bannikova, A., Small, D. M., & Kasapis, S. (2015). Control release of thiamin in a glassy κ-carrageenan/glucose syrup matrix. *Carbohydrate Polymers, 115*, 723–731.
Panyoyai, N., Bannikova, A., Small, D. M., Shanks, R. A., & Kasapis, S. (2016). Diffusion of nicotinic acid in spray-dried capsules of whey protein isolate. *Food Hydrocolloids, 52*, 811–819.
Panyoyai, N., & Kasapis, S. (2016). A free-volume interpretation of the decoupling parameter in bioactive-compound diffusion from a glassy polymer. *Food Hydrocolloids, 54*, 338–341.
Paramita, V. D., Bannikova, A., & Kasapis, S. (2015). Release mechanism of omega-3 fatty acid in k-carrageenan/polydextrose system undergoing glass transition. *Carbohydrate Polymers, 126*, 141–149.
Paramita, V. D., Bannikova, A., & Kasapis, S. (2016). Preservation of oleic acid entrapped in a condensed matrix of high-methoxy pectin with glucose syrup. *Food Hydrocolloids, 53*, 284–292.
Paramita, V. D., & Kasapis, S. (2019). Molecular dynamics of the diffusion of natural bioactive compounds from high-solid biopolymer matrices for the design of functional foods. *Food Hydrocolloids, 88*, 301–319.
Paramita, V. D., Piccolo, J. D. L., & Kasapis, S. (2017). Effect of co-solute concentration on the diffusion of linoleic acid from whey protein matrices. *Food Hydrocolloids, 70*, 277–285.
Ritger, P. L., & Peppas, N. A. (1987). A simple equation for description of solute release_ II. Fickian and anomalous release from swellable devices. *Journal of Controlled Release, 5*, 37–42.
Roos, Y. H. (2006). Phase transitions and transformations in food systems. In D. R. Heldman, & D. B. Lund (Eds.), *Handbook of food engineering* (2nd ed., pp. 287–352). Boca Raton: CRC Press, Taylor & Francis Group, 601.
Roudaut, G., Simatos, D., Champion, D., Contreras-Lopez, E., & Le Meste, M. (2004). Molecular mobility around the glass transition temperature: A mini review. *Innovative Food Science & Emerging Technologies, 5*(2), 127–134.
Siepmann, J., & Siepmann, F. (2008). Mathematical modeling of drug delivery. *International Journal of Pharmaceutics, 364*, 328–343.
Siepmann, J., & Siepmann, F. (2012). Modeling of diffusion controlled drug delivery. *Journal of Controlled Release, 161*(2), 351–362.
Sila, A., Martinez-Alvarez, O., Krichen, F., Gómez-Guillén, M. C., & Bougatef, A. (2017). Gelatin prepared from European eel (*Anguilla anguilla*) skin: Physicochemical, textural, viscoelastic and surface properties. *Colloids and Surfaces A, 529*, 643–650.
Slade, L., & Levine, H. (1991). Beyond water activity: Recent advances based on alternative approach to the assessment of food quality and safety. *Critical Reviews in Food Science and Nutrition, 30*(2–3), 115–360.

Teimouri, S., Morrish, C., Panyoyai, N., Small, D. M., & Kasapis, S. (2019). Diffusion and relaxation contributions in the release of vitamin B6 from a moving boundary of genipin crosslinked gelatin matrices. *Food Hydrocolloids, 87*, 839–846.
Zarzycki, R., Modrzejewska, Z., & Nawrotek, K. (2010). Drug release from hydrogel matrices. *Ecological Chemistry and Engineering, 17*, 117–136.
Zhong, C., Tan, S., & Langrish, T. (2019). Redness generation via Maillard reactions of whey protein isolate (WPI) and ascorbic acid (vitamin C) in spray-dried powders. *Journal of Food Engineering, 244*, 11–20.
Zhou, P., Mulvaney, S. J., & Regenstein, J. M. (2006). Properties of Alaska pollock skin gelatin: A comparison with Tilapia and pork skin gelatins. *Journal of Food Science, 71*, C313–C321.

Paper 12

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