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The effect of $MgCl_2$ on the kinetics of the Maillard reaction in both aqueous and dehydrated systems

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ABSTRACT

The modification of Maillard reaction kinetics induced by $MgCl_2$ was evaluated in both liquid and dehydrated model systems with special emphasis on the interactions of the salt with water and/or the sugars. In liquid trehalose systems, browning is accelerated by the presence of $MgCl_2$ due to the increased sugar hydrolysis and to the reduction of water mobility caused by the salt (shown by the decrease in ^1H NMR relaxation times T_2), counteracting the inhibitory effect of water on the reaction. In water restricted trehalose systems, $MgCl_2$ inhibited the Maillard reaction. The salt–sugar interactions, manifested by the delayed sugar crystallization, decreased the reaction rate by affecting the reactivity of reducing sugars. Molecular and supramolecular effects in the presence of $MgCl_2$ have been observed in the present work, and must be taken into account considering high technological interest in finding strategies to either inhibit or enhance the Maillard reaction depending on the application.

1. Introduction

The rate of the Maillard reaction and the nature of its products are governed by the immediate chemical environment of the reactants, defined by the chemical composition of the system (water content, pH, presence and type of buffer salts, temperature and exposure to light) (Baisier & Labuza, 1992; Bell, 1997; Cerruti, Resnik, Seldes, & Ferro Fontán, 1985; Petriella, Chirife, Resnik, & Lozano, 1988). The development of fluorescent products from the Maillard reaction (excitation 340–370 nm/emission 420–475 nm) was described during the storage of different types of food products and model systems. The application of fluorescence measurement is considered a potential tool for addressing key problems of food deterioration as it is an early marker or index of the biomolecular damage (Matiacevich, Santagapita, & Buera, 2005). The behaviour of model systems in the presence of divalent cations (Mg^{2+} or Ca^{2+}) was previously studied. Salts may affect important properties of sugar systems, including fluorescence and browning development from the Maillard reaction, the kinetics of sugar crystallization and sugar hydrolysis (Matiacevich & Buera, 2006; Santagapita & Buera, 2006; Santagapita & Buera, 2008; Schebor, Burin, Buera, & Chirife, 1999). Gökmen & Şenyuva (2007) showed that acrylamide formation is prevented by divalent cations during the Maillard reaction in fructose-asparagine model systems; however, ionic associations between the charged groups of asparagine and Ca^{2+} were not observed. Sugar–cation (Morel-Desrosier, Lhermet, & Morel, 1991) and brown pigment-cation complexes (O'Brien & Morrissey, 1997) have been shown to form in solution. The development of strategies for inhibiting the Maillard reaction employing additives other than sulphites (Lester, 1995) has a high technological interest. Alternatively, in other applications it may be beneficial to enhance the Maillard reaction. However, there has been little work carried out on the role of food-compatible salts in the kinetics of the Maillard reaction. The effect of cations can be analysed through their impact on the modification of solid–water interactions, and water availability, which govern and modulate the kinetics of the Maillard reaction (Acevedo, Schebor, & Buera, 2006).

Nuclear magnetic resonance (NMR) is a very useful technique for investigating the behaviour of water mobility in foods (Schmidt, 2007). The differences in relaxation times of protons from different environments have been exploited in NMR studies to measure the relative amounts of water with different degrees of interaction with solids and consequently, with different mobility (Acevedo et al., 2006; Farroni, Matiacevich, Guerrero, Alzamora, & Buera, 2008). The spin–spin or transversal relaxation time constants of protons (T_2) are related to the molecular mobility of protons in the system. The relaxation processes are affected by the chemical exchange taking place in sites of different mobility. Protons of water molecules which interact tightly with solids, and therefore have reduced mobility (although they are still exchangeable), show small T_2 values, whereas protons which are readily

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mobile have relatively long T_2 values (Kuo, Gunasekaran, Johnson, & Chen, 2001).

The purpose of the present work was to analyse the modification of Maillard reaction kinetics induced by the presence of $MgCl_2$ in both liquid and dehydrated model systems, with special emphasis on the interactions of the salt with water and/or the sugars.

2. Materials and methods

2.1. Preparation of model systems

2.1.1. Liquid systems

Liquid systems were prepared from 5% to 70% w/v of D(+)glucose (Merck, Darmstadt, Germany) or α-α-trehalose (Hayashibara Co, Ltd., Okayama, Japan) and 0.5% L-glycine (Merck) with and without $MgCl_2$ (Mallinckrodt Chemical Works, St. Louis, USA), in a 5:1 sugar:salt molar ratio; controls without $MgCl_2$ and glycine were also prepared. Aliquots (2.5 ml) of each model system were placed in 5 ml vials. The systems were prepared in 0.1 M phosphate buffer, pH 5 (Merck, Darmstadt, Germany). All reactants were analytical grade. The vials were hermetically sealed and stored at 70 ± 1 °C in a forced air convection oven.

2.1.2. Solid systems

Solid systems consisted of freeze-dried solutions containing 20% w/v trehalose and 1% w/v L-glycine. The systems were prepared in 0.1 M phosphate buffer at pH 5. The salt $MgCl_2$ was added in a 5:1 sugar:salt molar ratio; controls without $MgCl_2$ and without glycine were also prepared. Aliquots (2.5 ml) of each model were placed in 5 ml vials, frozen at −26 °C for 24 h and immersed in liquid nitrogen immediately before freeze-drying. Freeze-drying was performed in a Heto Holten A/S, cooling trap model CT 110 freeze-dryer (Heto Lab Equipment, Denmark) operating at a condenser plate temperature of −111 °C and a chamber pressure of 4×10^{-4} mbar. After freeze-drying, the systems were transferred to vacuum desiccators and exposed to different relative vapour pressure conditions (RVP): 22%, 43%, 52%, 84% and 97% (Greenspan, 1977) at 25 ± 1 °C for 2 weeks. After humidification, the vials were hermetically sealed and stored at 70 ± 1 °C in a forced air convection oven.

2.2. Determination of browning and fluorescence development

The progress of the Maillard reaction was followed by a browning index defined as absorbance units at 445 nm (UV–vis 1203 spectrophotometer, Shimadzu, Kyoto, Japan) multiplied by the dilution factor, and by fluorescence intensity with excitation at 340 nm/emission 492 nm (model USB 2000 spectrofluorometer, Ocean Optics Inc., FL, USA). The samples employed for the determination of fluorescence were diluted in order to avoid inner filter effect, with absorbance values lower than 0.1 at excitation wavelength (340 nm). Since trehalose is a non-reducing sugar, its participation in Maillard reaction can only occur after hydrolysis.

Browning and fluorescence development as a function of reaction time followed a quadratic dependence, which indicates a half order reaction, which results from the combination of two steps of zero and first order (Matiacevich & Buera, 2006).

The confidence intervals for browning and fluorescence values were 3% and 5% for 95% certainty, respectively.

2.3. Disaccharide hydrolysis

Sugar hydrolysis was analysed by measuring the amount of glucose released during the incubation time by means of an enzymatic method (Wiener Lab. S.A.I.C., Rosario, Argentina) described previously (Bergmeyer & Grassi, 1983).

2.4. Fluorescence quantum yield

Sugar systems solutions (70% w/v) were diluted in order to obtain absorbance below 0.05 to avoid inner filter effects (Lakowicz, 1999). Four dilutions (in duplicate) were prepared for each time of incubation. Fluorescence quantum yields (Φ_F) were determined using quinine sulphate in 0.1 N sulphuric acid as a reference fluorophore of known quantum yield ($\Phi_F = 0.546$) (Lakowicz, 1999), using the classical Eq. (2.1):

$$\Phi_F^S = \Phi_F^R \cdot \frac{I_\lambda^S}{I_\lambda^R} \cdot \frac{A_\lambda^R}{A_\lambda^S} \cdot \frac{n_S^2}{n_R^2} \tag{2.1}$$

where Φ_λ^S and Φ_λ^R are the fluorescence quantum yield of the sample (S) and the reference (R), respectively; I_λ^S and I_λ^R are the integrated areas of their fluorescence spectra; A_λ^S and A_λ^R are their absorbances at the excitation wavelength λ, and n_S and n_R are the refractive index of each solution. In Eq. (2.1), it is assumed that the sample and reference are excited at the same wavelength.

2.5. Nuclear magnetic resonance (NMR) measurements

Transversal or spin–spin relaxation times (T_2) were measured by time resolved proton nuclear magnetic resonance (^{1}H NMR) in a Bruker Minispec mq20 (Bruker Biospin Gmbh, Rheinstetten, Germany) with a 0.47 T magnetic field operating at a resonance frequency of 20 MHz. Proton populations of different mobility were measured using two spin-echo sequences: (a) Hahn (Hahn, 1950), for protons from solids or from water strongly interacting with the solid matrix, and (b) Carr–Purcell–Meiboom–Gill (CPMG) (Carr & Purcell, 1954; Meiboom & Gill, 1958), for more mobile protons. All samples were previously equilibrated at 25.00 ± 0.01 °C in a thermal bath (Haake, model Phoenix II C35P, Thermo Electron Corporation Gmbh, Karlsruhe, Germany).

2.5.1. Measurements using a Hahn sequence

Hahn spin-echo consists of a (90°–τ–180°) sequence. This method allows for analysis of the less mobile protons interacting with the solid matrix; the more mobile protons could not be analysed using this sequence due to diffusion problems during measurement at long times, caused by inhomogeneities in the magnetic field. Trehalose dehydrated systems humidified between 22% and 97% RVP were analysed using this sequence. The following settings were used: scans = 4, recycle delay = 1.5 s, gain = 75 dB, number of points = 20, time for decay curve display = 1 s and interpulse (τ) range of 0.001–0.5 ms. The interpulse range was selected in order to record the complete relaxation of the signal. No phase cycling was used. A polyvinylpyrrolidone (PVP of 58 000 Da) system equilibrated at 43% RVP was used for the automatic update of the equipment which tunes the pulse duration, detection angles, gain and magnetic field homogeneity.

2.5.2. Measurements using a CPMG sequence

Liquid systems were analysed using a CPMG sequence, which consists of $90^\circ_x - \tau - \left[180^\circ_y - \tau - \text{echo} - \tau\right]_n$ sequence, with the following setting: $\tau = 0.5$, scans = 4, number points = 256, dummy shots = 15, gain = 68 dB; phase cycling was used. The automatic update of the equipment was performed employing a 40% w/v trehalose system without thermal treatment.

For both sequence measurements, an exponential function (as stated below in Eq. (2.2)) was found to fit the experimental data adequately, from which transverse relaxation time constant (T_2) was obtained.

$$A = A^{(-t/T_2)} \quad (2.2)$$

where A is the signal amplitude that is proportional to the amount of protons in the sample relaxing after the pulse sequence. This mono-exponential model was used for the liquid samples and for most of the dehydrated systems, obtaining the contributions of protons in the most representative fraction. A bi-exponential model fitted some dehydrated systems with high water content (corresponding to samples at 97% and 84% RVP containing $MgCl_2$), but it is important to note that, for these samples, the signal amplitude of the detected second proton fraction represented a very small contribution to the total proton population, and the mono-exponential approach was selected as the best model for comparative purposes.

2.6. Determination of water content

The total water content of the trehalose solid rehumidified systems was determined gravimetrically by difference in weight before and after drying in a vacuum oven for 48 h at 96 ± 2 °C. These drying conditions were selected in previous studies with samples of similar composition (Mazzobre, Longinotti, Corti, & Buera, 2001) and they were adequate to determine water content in the studied systems with a confidence interval of 6% for a 95% certainty. Duplicate analyses were performed and the average of the results is reported on a dry basis (db).

2.7. Thermal transitions measurements

Melting events were determined by dynamic differential scanning calorimetry (DSC), in the temperature range from 20 to 120 °C, by means of a Mettler Toledo model 822 equipment (Mettler Toledo AG, Greifensee, Switzerland) at a heating rate of 10 °C/min. Melting temperature values were taken as peak temperatures. All measurements were made in duplicate with 10–25 mg sample mass using hermetically sealed aluminium pans of 40 μl inner volume (Mettler Toledo AG). An empty pan was used as a reference and average values are reported. The instrument was calibrated using standard compounds (indium and zinc) of defined melting point and heat of melting (ΔH_m). All thermograms were analysed using STAR^e Software v. 6.1 (Mettler Toledo AG). The samples were initially amorphous as determined by the absence of endothermal peaks in the DSC scans. The amount of trehalose dihydrate formed (degree of crystallization, ϕ) during the storage time was calculated from the ratio of the area of the endothermic melting peak of the sample and the calorimetric enthalpy of the melting of pure trehalose dihydrate measured under the same conditions in a dynamic DSC run, as Eq. (2.3).

$$\phi = \frac{\Delta H_m}{\Delta H_{mT}} * 100 \quad (2.3)$$

where ΔH_m is the heat of melting of trehalose in the sample and ΔH_{mT} is the heat of melting of pure trehalose (139 J/g) measured under the same conditions (Santagapita & Buera, 2006). The relative error for ϕ (calculated with a 95% confidence interval) was about 10% of the ϕ value.

3. Results and discussion

3.1. Maillard reaction development: fluorescence and absorbance data

3.1.1. Liquid systems

Browning and fluorescence development of the liquid systems increased with increasing storage time at 70 °C, for the whole composition range studied (5–70% w/v), as shown in Fig. 1 (a–d). The 70% w/v sugar systems, for example, showed corresponding visual colour changes: uncoloured → yellow → golden → cinnamon → reddish brown. The type and sugar concentration and the presence of $MgCl_2$ were critical for the rate of pigment and fluorescent compounds formation. In the model systems without amino acid (control samples), no increase in the absorbance or the fluorescence was detected, indicating that there was no contribution to colour or fluorescence due to caramelization in the experimental time frame. The decreasing order for browning development was found according to sugar reactivity/stability: glucose > trehalose (Burton, Mc Weeny, Pandhi, & Biltcliffe, 1962; O'Brien & Morrissey, 1997) in all analysed sugar concentrations, as shown in Fig. 1a and c for the 70% w/v samples. In liquid glucose systems, inhibitory effects of $MgCl_2$ on browning and development of fluorescent products, observed for the 70% w/v samples in Fig. 1a and b (respectively), were also noticed in the whole composition range studied (5–70% w/v), extending our previous results for 5% and 40% w/v samples (Matiacevich & Buera, 2006; Santagapita & Buera, 2006). However, as shown in Fig. 1c and d for the 70% w/v systems, an accelerating effect of $MgCl_2$ was observed for both browning and development of fluorescence in liquid systems containing trehalose, in the whole composition range studied. Fig. 2 shows the kinetic constants for browning development in trehalose systems, calculated through a quadratic model, plotted as a function of different water contents. Although trehalose is very stable to hydrolysis (Santagapita & Buera, 2006; Schebor et al., 1999), it was hydrolysed during heat treatment at 70 °C and the hydrolysis rate was accelerated by the presence of $MgCl_2$ (Santagapita & Buera, 2006), as confirmed by glucose analysis (data not shown). For example, for the range between 5% and 40% w/v trehalose system, the rate of trehalose hydrolysis was lower than 1% in systems without salt and increased to 2.3–2.5% in the presence of $MgCl_2$. Pigment development through the Maillard increased accordingly, as observed in Fig. 1c and d.

3.1.2. Solid systems

In the freeze-dried trehalose samples humidified at relative vapour pressures (RVPs) in the range 22–97%, the development of fluorescence was parallel to the development of browning, and the hydrolysis rate increased in the presence of salt as was seen in the analysed liquid systems (also in agreement with Santagapita & Buera, 2006). In the samples containing $MgCl_2$ at 10% (db) water content, only 1.1% trehalose was hydrolysed after 24 h at 70 °C. Under the same conditions, disaccharide hydrolysis was 0.13% for trehalose samples without salt. However, contrary to the results observed in trehalose liquid samples, in solid trehalose samples the presence of $MgCl_2$ delayed browning development, as shown in Fig. 2. It is clearly shown that $MgCl_2$ inhibited the Maillard reaction at low water contents in solid trehalose systems (Fig. 2, left side of the dotted line), but in liquid systems, the presence of the salt increased the reaction rate (Fig. 2, right side of the dotted line, and Fig. 1c). Thus, the effect of $MgCl_2$ on browning development in trehalose samples was dependent on the state of the systems.

3.2. Fluorescence characteristics and quantum yield study

In order to analyse the effects associated with sugar type and their interactions with $MgCl_2$ on the Maillard reaction kinetics in liquid systems shown in Fig. 1, fluorescence characteristics of the samples were studied. In agreement with previous studies (Matiacevich et al., 2005), the maximum wavelength for emission was 450 nm in all systems, and the fluorescence development at zero time of incubation was negligible. Fig. 3 shows the fluorescence emission spectra of trehalose or glucose systems with and without $MgCl_2$, after 0 and 120 h of storage at 70 °C. As seen in this figure, the spectral characteristics were independent of the studied

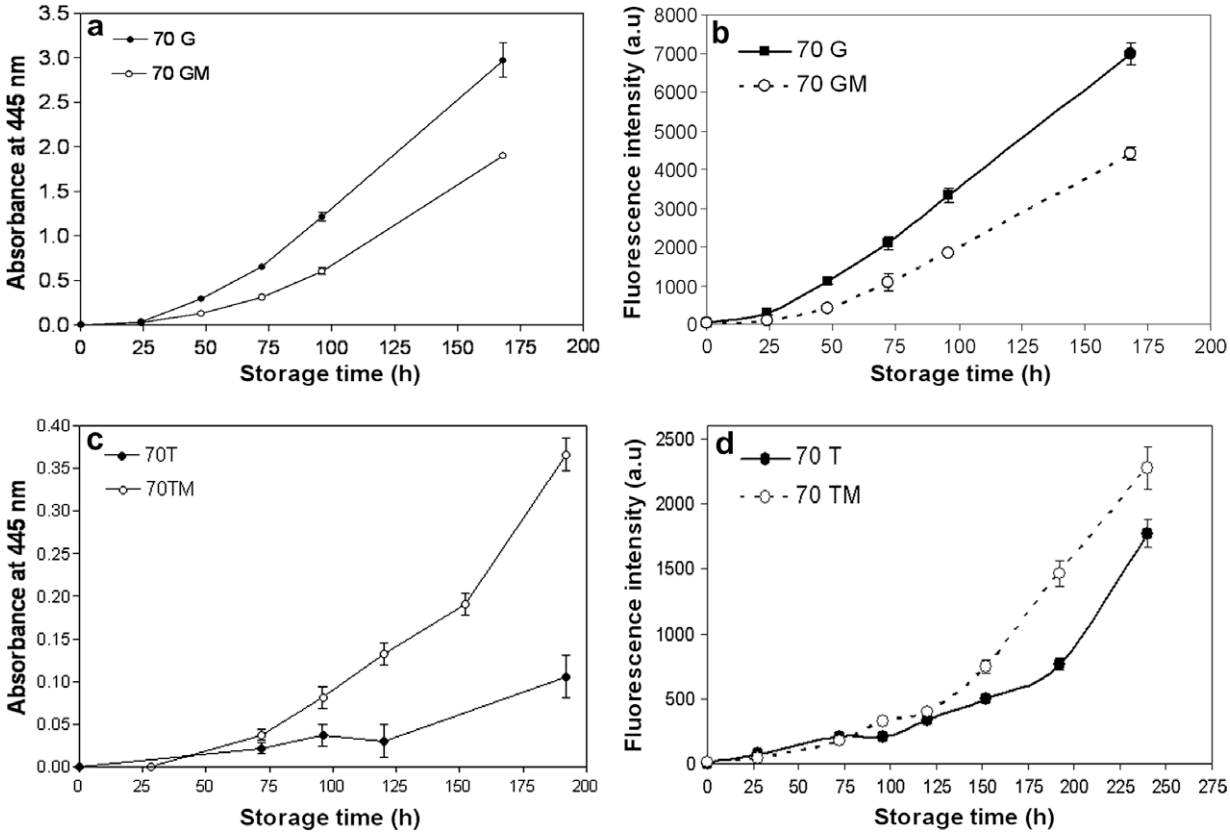

Fig. 1. Browning and fluorescence development for 70% w/v glucose (a, b), or trehalose (c, d) systems with $MgCl_2$ (open symbols) and without $MgCl_2$ (closed symbols) as a function of storage time at 70 °C. Excitation/emission 340/450 nm. T, trehalose; G, glucose; M, $MgCl_2$. Bars on symbols represent the 95% confidence interval.

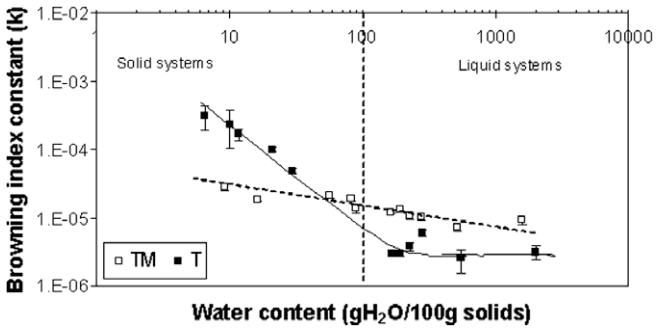

Fig. 2. Kinetic constants for the Maillard reaction (obtained by a quadratic model) as a function of water content for trehalose systems. Solid lines correspond to sugar systems without salt and dashed lines to $MgCl_2$ -containing systems. T, trehalose; M, $MgCl_2$. Bars on symbols represent the 95% confidence interval.

system, indicating that neither the type of sugar nor the presence of salt affected the type of fluorophore obtained in each system.

The fluorescence quantum yield, which is defined as the ratio between the number of photons involved in the emission and the total number of excited photons, may provide information on molecular structural properties of the fluorophores and their interactions. The fluorescence quantum yield was calculated for the samples as a function of heating time at 70 °C, in order to further analyse the effect of $MgCl_2$ on the Maillard reaction (shown in Figs. 1 and 2). As shown in Fig. 4, the fluorescence quantum yield was constant as a function of heating time in glucose systems after a slight initial increase. It is important to note that the presence of $MgCl_2$ did not affect the quantum yield. Thus, the increase of fluorescence observed in Fig. 1b is due to the increasing concentration of fluorophores with increased incubation time, and not to an increase of the fluorophores quantum yield. The same conclusion could be obtained for trehalose systems (Fig. 1d), considering that trehalose must be hydrolysed to two glucose units in order to participate of the Maillard reaction.

Therefore, in agreement with previous studies, the presence of $MgCl_2$ affected the kinetics of the reaction without affecting the chromatic and fluorescent spectral characteristics of samples (Matiacevich & Buera, 2006; Santagapita, Matiacevich, & Buera, in press) or the fluorescence quantum yield properties of the fluorophores.

3.3. Proton mobility studies by NMR

In order to evaluate the possible contribution of water–salt interactions in the results observed in Figs. 1 and 2, proton mobility in each liquid system was studied by ^1H NMR transversal relaxation times (T_2). In liquid systems, T_2 values were calculated after the spin-echo CPMG sequence by a mono-exponential decay model and the results are shown in Fig. 5. With increasing sugar concentration, T_2 values diminished for all sugar systems, indicating a reduced mobility of water protons. As also observed in Fig. 5, T_2 values were not affected by $MgCl_2$ in systems containing glucose (i.e., water–glucose interaction was unmodified by salt), whilst in trehalose systems they were between 6% and 14% lower in the presence of salt, reflecting a reduced mobility. The T_2 values obtained indicated that the $MgCl_2$ interaction affects water mobility in the trehalose samples, but not in glucose systems. Since an inhibitory effect of water has been observed on the Maillard reaction (Acevedo et al., 2006; Labuza, 1994), the accelerating effect of $MgCl_2$ on the reaction rate observed in the disaccharide samples in Figs. 1 and 2 can be associated with water–salt interactions. According to Miller and de Pablo (2000), the local environment

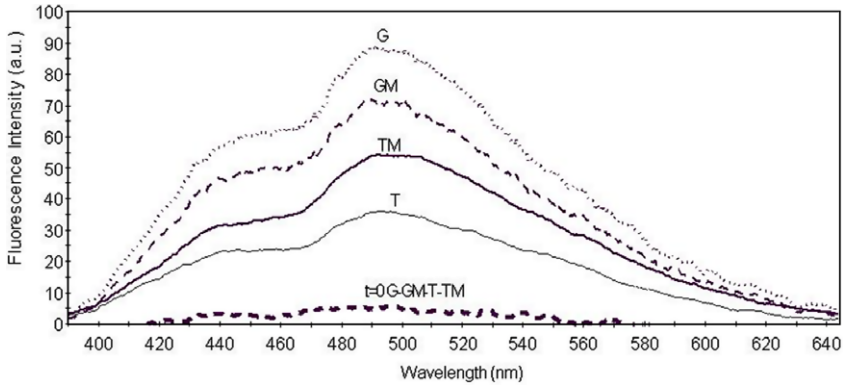

Fig. 3. Fluorescence emission spectra of 70% w/v sugar systems heated at 70 °C for 120 h (-.-.-.-.-. glucose, and —— trehalose systems; grey curves correspond to $MgCl_2$ containing systems). The curve labelled as $t = 0$ h, G or T corresponds to the fluorescence for zero time of incubation for all sugar systems. T, trehalose; G, glucose; M, $MgCl_2$.

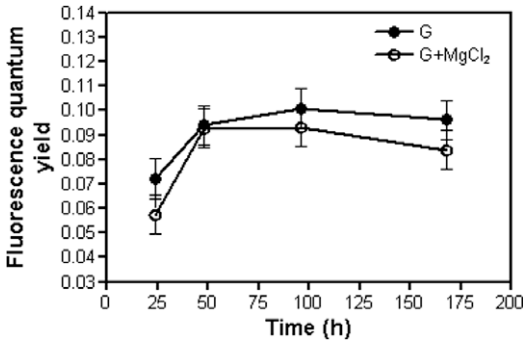

Fig. 4. Fluorescence quantum yield as a function of time of incubation for glucose systems with (open symbols) and without (closed symbols) $MgCl_2$. Excitation/emission 340/450 nm. G, glucose; M, $MgCl_2$. Bars on symbols represent the 95% confidence interval.

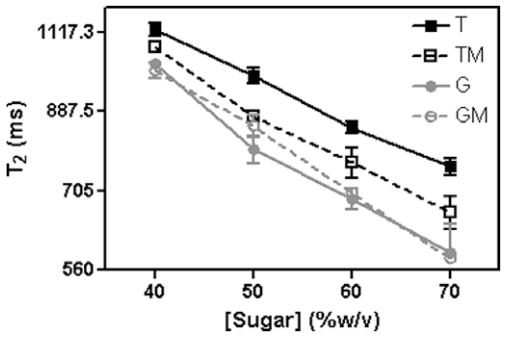

Fig. 5. ^1H NMR spin–spin relaxation times (T_2) obtained by a mono-exponential decay model, using a CPMG sequence as a function of sugar concentration for liquid systems. T, trehalose; G, glucose; M, $MgCl_2$. Bars on symbols represent the 95% confidence interval.

of the ions contained in a trehalose system has more water molecules compared to those uniformly distributed in water, which can explain the lower T_2 values obtained in the disaccharide-salt containing systems.

In the case of glucose liquid systems (Fig. 1a and b) the retarding effect of $MgCl_2$ can be attributed to sugar–salt interactions. The formation of sugar-cation complexes, which may affect the sugar availability for the reaction, has been previously reported and depends on the tautomeric forms and spatial position of the hydroxyl groups of the sugar (Angyal, 1973). It is to be noted that in trehalose systems, the reactant participating in the Maillard reaction is glucose, as in the pure glucose systems. However, in the trehalose

Table 1
Degree of trehalose crystallization (ϕ) obtained by differential scanning calorimetry for systems of different water contents (w.c., in % of dry basis) with or without $MgCl_2$.

W.C. ± 2 (% db)	ϕ (%)*	
	w.o. $MgCl_2$	with $MgCl_2$
11	38	6
24	86	61
27	88	54
32	80	35

* $\phi = \frac{\Delta H_m}{\Delta H_{mT}} * 100$; where ΔH_m is the melting enthalpy of trehalose at a given time and ΔH_{mT} is the melting enthalpy of pure trehalose (Santagapita & Buera, 2006). The relative error for ϕ (calculated for a 95% confidence interval) was about 10% of the ϕ value.

systems the reaction is accelerated by $MgCl_2$, whilst it is delayed in the glucose systems in the presence of salt. Thus, it is not an effect of the type of reactants involved in the reaction, but of the kind of water-matrix interactions: whilst in the glucose systems the water–glucose interactions were not affected by the presence of $MgCl_2$ (as shown by ^1H NMR relaxation times), in the trehalose systems a decreased relaxation time indicated lower water mobility in the presence of salt. A probable interaction between Mg^{2+} and phosphate buffer, which may cause the inhibition of Maillard reaction, has been reported (Akagawa, Miura, & Suyama, 2002) and must also be taken into account.

In other set of experiments, in the trehalose solid systems (with and without $MgCl_2$), the spin–spin ^1H NMR relaxation times were obtained by the spin-echo Hahn sequence (indicated as T_{2H}) in samples at different RVP. The T_{2H} values in samples of similar water content (between 10% and 20% db) were 25 ± 2 μs either in the presence of $MgCl_2$ or without the salt, showing no differences between them. The T_{2H} values obtained correspond to matrix and water protons displaying strong interactions with the matrix, and no changes in their mobility could be detected by the presence of $MgCl_2$. Therefore, the water–salt interactions manifested by the relaxation times from ^1H NMR in the trehalose solid systems could not explain the inhibitory effect of salts on the browning kinetics. Of the dehydrated systems, only the trehalose–$MgCl_2$ systems at RVP 84% and 97% and the trehalose systems at 97% showed a second set of T_{2H} with values higher than 1 ms, which correspond to water molecules of weak interactions with the solid matrix. It is to be noted that the effect of $MgCl_2$ in these solid systems (in which mobile water molecules were detected) was not as important on the browning kinetic constants of these systems as at lower water contents (Fig. 4).

Another effect of the salt interactions in restricted water environments could be manifested by the modification of the sugar

crystallization kinetics by $MgCl_2$. Table 1 shows that the degree of trehalose crystallization (ϕ) in solid trehalose systems containing $MgCl_2$ was lower than in samples without salt. The delay of sugar crystallization by the presence of salts in supercooled systems has been previously reported (Longinotti, Mazzobre, Buera, & Corti, 2002; Santagapita & Buera, 2008) and may be explained by dynamic water–salt–sugar interactions, which take place at a molecular level and are related to the charge/mass ratio of the cation present. An interaction/complexation between the magnesium cation and the Maillard reaction products has also been reported (O'Brien and Morrissey, 1997) and is not discarded as another factor influencing molecular mobility.

In conclusion, the Maillard reaction kinetics can be affected by the presence of salts to a different degree, depending on the type of sugar present (either acting as reactant or as part of the solid matrix) and the state of the system. Molecular and supramolecular effects of the presence of $MgCl_2$ have been observed in this work.

Due to the inhibitory effect of water on the Maillard reaction, modifications in water–solids interactions promoted by salts could be responsible for changes in the reaction rates. In liquid trehalose systems (or when mobile water protons were detected), the browning reaction is accelerated by the presence of $MgCl_2$ due to the increased sugar hydrolysis and the reduction of water mobility caused by the salt, counteracting the inhibitory effect of water on the Maillard reaction. On the other hand, in water restricted (freeze-dried) trehalose systems, $MgCl_2$ inhibited the Maillard reaction. In this case, the salt–sugar interactions, manifested by the delayed sugar crystallization, decreased the reaction rate by affecting the reactivity of reducing sugars.

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