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研究成果報告(精簡版)

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ABSTRACT

Fructose/glycine model systems of different sugar concentrations were incubated at 60, 75, and 90^{o}C separately for studying the reaction kinetics of color development, pH change, and antioxidant activity change in the early stage of Maillard reactions. The results indicated that in the Maillard reactions, systems' color development, pH decreasing, and TEAC increasing all followed first-ordered kinetics on galactose concentration. The Q_{10} values at temperature between 60 and 90˚C were 4.76, 2.81, and 3.59 for color development, pH decreasing, and TEAC increasing, respectively. And activation energies were 151l, 101, and 123 kJ/mol for color development, pH decreasing, and TEAC changing, respectively.

Key words: *Maillard reactions, Reaction kinetics, Antioxidant activity*

INTRODUCTION

Maillard reaction is one of the reactions that attract the very curiosities of food chemists and technologists. This is not only because that the reaction, or reactions, initiate with the consumption of two macronutrients common in foods and terminate with the formation of brown pigments and roasted aroma. But also, its outputs are much complicate that we can hardly define a real image whether or not these reactions occur in foods are good for food quality or our health.

The antioxidant activity derived from Maillard reactions is one of such confusions that food scientists would want to resolve. Generally speaking, foods at antioxidant conditions would be beneficial for both food quality itself and consumers' health. However, in studies concerning this properties in foods, some authors indicated that Maillard reaction products (MRPs) could act as antioxidants (Osada & Shibamoto, 2006; Benjakul, Lertittikul, & Bauer, 2005; Wagner, Derkits, Herr, Schuh, & Elmadfa, 2002); while others indicated MRPs were pro-oxidant (Odani *et al.*, 1998; Anese, Manzocco, Nicoli, & Lerici, 1999; Yen & Liao, 2002; Puscasu and Birlouez-Aragon, 2002); and still others reported MRPs to exhibit both antioxidant and pro-oxidant activity (Wijewickreme & Kitts, 1997; Calligaris, Manzocco, Anese, & Nicoli, 2004; Yilmaz & Toledo, 2005). By reviewing these literatures, we can see such contradictory findings were differentiated from each other by the stages of Maillard reactions progress. At the earlier stage, MRPs of smaller molecule such as glyoxal, methylglyoxal, some redutones, and other dicarbonyls are formed (Hodge, 1953; Yaylayan & Heffenden, 2003, Chen, Jin, & Chen, 2006). Since these compounds are high in oxidative potential and chemical activity, MRPs at this stage, *i.e.* the intermediate MRPs, tended to be pro-oxidant. The high chemical activity of these compounds then drives Maillard reactions progress to the later stage during which MRPs with high molecular weight and brown color are formed via series of condensations and polymerizations. Complex of MRPs at the later stage had been proved to be antioxidant (Wagner *et al.*, 2002) and were named collectively as melanoidins. It is clear that the pro-oxidant MRPs are formed prior to the antioxidant ones in Maillard reactions. Consequently, it would be more healthful for any food processing dealing with Maillard reactions to take antioxidant activity into consideration. To reduce pro-oxidant activity and promote antioxidant activity in Maillard foods, food technologists need to know the reaction kinetics of Maillard reactions, *i.e.* the time and temperature required for foods to reach the optimal antioxidant activity.

Literatures pertaining to the reaction kinetics of Maillard reactions mainly

focused on the reactant consumption (Chen *et al.*, 2006), the color development (Rapusas & Driscoll, 1995; Peterson, Tong, Ho, & Welt, 1994; Ajandouz & Puigserver, 1999; Chen *et al.*, 2006; Turkmen, Sari, Poyrazoglu, & Velioglu, 2006), and the formation of intermediate MRPs (Martins & Van Boekel, 2005[a,b], Chen *et al.*, 2006). However, since its existence in foods is so common, its influences on foods are so significant, its reaction pathways are so complicate, and its product compounds are so diverse that Maillard reactions have become quite a series of reactions worthy of far more exploration. In addition to reactant consumption, color development, and MRPs formation, there remain the reaction kinetics of still other resultant properties such as antioxidant activity, mutagenic activity, volatile formation, system pH, *etc.* to be investigated.

In the current study, fructose/glycine model systems were used for constructing the kinetic parameters of color development, pH change, and antioxidant activity change at early Maillard reaction stage.

MATERIALS AND METHODS

Galactose/glycine model systems:

α-D(+)-Fructose (Sigma Chemical Co., USA) and L-glycine (Sigma Chemical Co., USA) were dissolved in de-ionized water to prepare galactose and glycine solutions separately. Concentrations of prepared fructose solution were 1.25, 2.5, 5.0, and 10% , where that of glycine solution was 10%. In the beginning of experiments, equal volume of galactose solution and glycine solution were mixed and adjusted pHs to 8.0 by using 1.0 N NaOH and 1.0 N HCl solutions. The final concentrations of galactose and glycine in the mixtures would be half of the original solutions.

The mixtures were dispensed in screw-capped glass tubes and incubated at 60, 75, or 90ºC water bath immediately. Model solutions would be sampled at intervals of half an hour to 24 hours and determined pH, browning, and anti-oxidative activity.

Measurement of pH: A pH meter (Model SP-71, Suntex Inc., Taiwan) was used to determine the pH value of the model solutions.

Measurement of color change: The color change of model solutions was determined the absorbance at wavelength 420 nm (A_{420}) as the browning indicator (Chen *et al*., 2005). Instrument for determining A_{420} was a spectrophotometer (Spectro UV-Vis Auto, Culver City, CA, USA).

Measurement of antioxidative activity:

Trolox equivalent antioxidant capacity (TEAC)

The modified method was according to Miller et al., (1995) and Arnao et al., (1996). ABTS solution was prepared by peroxidase, 2,2′-Azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS), H_2O_2 and the final concentration was 4.4U/ml, 100mM, and 50 μM, respectively. The ABTS solution was incubated at dark and 30℃ for 1 hour. 30μl samples and 300μl ABTS solutions were added into each well to react for 3 minutes then measured the absorbance at 734nm.

Scavenging effect (%) = $[A_b-(A-A_s)] /A_b$ x 100

Where A_b was the absorbance of blank, A was the absorbance of sample, and A_s was the absorbance of background of sample.

Different Trolox concentrations were determined by the same procedure and the antioxidant capacities of samples were calculate as the Trolox equivalent antioxidant capacity

Data analysis :

Rates of reaction including color, pH, and anti-oxidative activity change were

calculated using equation 2 derived from Laidler (1987):

$$v = d\,P / d\,t = k[F]^n \text{ ----------(2)}$$

where v was reaction rate, P was the intensity of response, $[F]$ was initial fructose concentration, t was time, k was reaction constant, and n was the order of reaction.

For figuring out the effects of temperature on the reactions, the temperature coefficient (Q_{10}) and activation energy (E_a) were calculated using Equations 3 and 4 respectively.

$$\log k = \log (Q_{10}) \times 10^{-1} T_C + C_1 \text{ ---------- (3)}$$

$$\ln k = C_2 + (E_a/R) \cdot T_K^{-1} \text{ ---------- (4)}$$

Where T_C and T_K were temperatures in Celsius and Kelvins respectively, E_a was activation energy in kJ/mol, C_1 and C_2 were constants, and R was gas constant which was 8.31 x 10^{-3} kJ/K·mol. Equation (5) which was widely used for calculating the reaction rate at different temperature by known temperature coefficient was derived from Equation (3).

$$k_{T1} = k_{T0} * Q_{10}^{(T1-T0)/10} \text{ ---------- (5).}$$

Where k_{T1} and k_{T0} were the reaction rate constants at temperature T1 and T0 respectively. Arrhenius equation (Equation 6) was derived from Equation (4) (Laidler, 1987) by taking natural logarithm on both sides and the natural logarithm of frequency factor, ln A as the constant.

$$k = A * e^{-Ea/RT} \text{ ---------- (6)}$$

2.6 Statistics analysis:

All experiments were done in triplicate. Stepwise simple linear regression analysis, on the basis of least sum of square of residuals and visual examination on distribution of residuals, was used to describe the reaction rate.

RESULTS AND DISCUSSIONS

Changes of color

The progress of color development in Maillard browning could be divided into three linear stages. The first linear stage which was the change of A_{420} during the beginning of browning reaction was not obvious and named the induction stage. A sharp linearly increasing browning which was named the zero-ordered stage followed the first linear stage. Then, the change slowed down in A_{420} that was the third linear stage which was the brown color came gradually to saturation. The zero-ordered stage was concerned about most researchers. The linear slope of A_{420} by time during the zero-ordered stage had been used as the browning rates in most studies. In this study, the same parameter was employed as the reaction rate of color development in model systems.

The changes of color in fructose/glycine model systems were dependent not only on temperature but also on fructose concentration (Figure 1). The linear regressions of each curve at the zero-ordered stage in figure 1 were summarized at table 1. Browning rates of the model systems (v_B) were the range of 0.09×10^{-5}~0.31×10^{-5}, 0.48×10^{-5}~2.19×10^{-5}, 1.90×10^{-5}~9.98×10^{-5}, and 9.16×10^{-5}~ 49.3×10^{-5} ΔA_{420}/s at temperature 45, 60, 75, and 90ºC in sequence, depending on fructose concentration (Table 1). The correlations between browning rate and initial fructose concentration were revealed a first-ordered empirical equation of color development on fructose concentration (Figure 2):

$$v_B = C1 + k_B [Fru]^1 \text{---------- (7).}$$

Where C1 was the constant, v_B was browning rate in ΔA_{420}/s, the slope k_B was rate constant of browning, which was 1.07×10^{-6}, 6.84×10^{-5}, 3.22×10^{-4}, and 1.57×10^{-3} ΔA_{420}/s·M for systems at 45, 60, 75, and 90ºC in sequence. The coefficients of determination (R^2) for Equation 7 were between 0.9032 and 0.9934 (Figure 2). And the browning reactions in fructose/glycine model systems were justified in the first-ordered kinetics.

The effect of temperature on browning reaction was figured out by taking linear regression, then Equation (3) and (4) were concluded by Equation (8) and (9):

$$\log k_B = 0.0678\, T_C - 8.68 \text{ ---------- (8).}$$

$$\ln (k_B) = 44.06 - 18178 T_K^{-1} \text{ ---------- (9).}$$

The slope of Equation 8 was equivalent to $\log Q_{10}$, and then the Q_{10} of browning reaction in fructose/glycine model systems was 4.76 at temperature range between 45 and 90ºC. And the slope in Equation 9 was equivalent to Ea/R, thereby the activation energy of browning reaction in the fructose/glycine model systems was 151 kJ/mol.

The reaction kinetics of color development in Maillard reactions had been reported to be zero- or pseudo-zero-ordered previously (Rapusas & Driscoll, 1995; Peterson *et al.*, 1994; Ajandouz & Puigserver, 1999; Chen *et al.*, 2005; Turkmen *et al.*, 2006). But the browning rates among different sugar concentrations at the same temperature and pH have had significant difference. Where the results indicated by Chen *et al.* (2005), who employed four sugar concentrations in their model systems, had shown the concentration-dependence of browning rate by fructose and glucose. And Liu *et al*. (2007) had shown the concentration-dependence of browning rate by galactose model solution. Then the current study had the similar results. Consequently, it would be much more suitable to define the kinetics of the color development in Maillard reactions to be first-ordered rather than zero- or pseudo-zero- ordered ones.

For the effects of temperature, the activation energy and Q_{10} values of Maillard browning were 121 - 139 kJ/mol and 4 - 5 in real food system, respectively (Rapusas & Driscoll, 1995; Turkmen *et al.,* 2006) and were 69.0KJ/mol and 1.98 in galactose/glycine model solution, respective (Liu *et al*., 2007). Reyes *et al*. (1982) reported that fructose/glycine systems browned at a rate faster than glucose/glycine and sucrose/glycine systems at initial stage. de Man (1990) indicated that the relative reactivity of galactose was higher than glucose in model solutions. In the current study, the activation energy and Q_{10} value were both higher than galactose/glycine ones. The result was shown that the relative reactivity of fructose was higher than galactose in sugar/glycine model solution. But the activation energies were the same values (~120KJmol^{-1}) in glucose and lactose / caseinate model system (Morales and van Boekel; 1998). It is an interesting thing to find the interaction between amino acids and sugars.

Changes of pH values.

The pH values in all model solutions decreased linearly with time (Figure 4). The linear regressions of each curve at the zero-ordered stage in figure 3 were summarized at table 2. The pH value decreasing rates of the model solutions (v_p) were the range of -0.26 x 10^{-6} ~ -1.29 x 10^{-6}, -1.15 x 10^{-6} ~ -8.72 x 10^{-6}, -4.83 x 10^{-6} ~ -35.62 x 10^{-6}, and -21.18 x 10^{-6} ~ -133.10 x 10^{-6} ΔpH/s at temperature 45, 60, 75, and 90ºC in sequence, depending on fructose concentration (Table 2). These initial pH decreasing trends could be indicated by a linear empirical equation as:

$$A_p = v_p\, t + C2 \quad \text{---------- (10).}$$

Where A_p was value in pH, C2 was the constant, t was time in hour, and v_p was the decreasing rate of pH in ΔpH/s. The R^2 values of these regression lines were almost more than 0.95, only the R^2 values of 0.04 and 0.14M fructose concentrations

incubated at 70ºC were no less than 0.90 (Table 2). The correlation between initial fructose concentration and pH decreasing rate followed a first-ordered reaction kinetics as the following equation (Figure 4):

$$v_p = C3 - k_p [Fru] \quad \text{---------- (11).}$$

Where C3 was the constant, and k_p indicated the rate constant of pH decreasing which were 4.2 x 10^{-6}, 3.15 x 10^{-5}, 1.2 x 10^{-4}, and 4.7 x10^{-4} ΔpH/s·M for systems incubated at 45, 60, 75 and 90ºC separately. These temperature-rate constants sets were further taken linear regression, and concluded with Equation (12) and (13).

$$\log k_p = 0.0449\ T_C - 7.3048 \quad \text{---------- (12).}$$

$$\ln (k_p) = 21.37 - 12141.35\ T_K^{-1} \quad \text{---------- (13).}$$

The slope in Equation (12) was equivalent to a Q_{10} of 2.81 for the pH decreasing in fructose/glycine model systems at temperature ranged between 60 and 90ºC. And the slope in Equation (13) was equivalent to an activation energy 101 kJ/mol for pH reduction in the fructose/glycine model systems.

Consumption of this group by Maillard reaction therefore would shift systems into more acidic condition (DeMan, 1999). In addition, some acidic compounds with buffering capacities including formic acid, acetic acid, methylglyoxal, glyoxal *etc.* (Martins, Marcelis, & Van Boekel, 2003; Chen *et al.*, 2005) had been reported to present in the intermediate MRPs, whereas none of any basic intermediate MRPs had yet been found during the earlier stage. The activation energy and Q_{10} values of pH value in fructose / glycine model solution were higher than the ones in galactose / glycine model solution.

Antioxidative activity.

The reaction of antioxidative activity of Maillard reaction products in model fructose/glycine systems, similar to that of browning reaction and pH decreasing, followed first ordered kinetics, too. Figure 5 revealed that the TEAC value increased linearly during the experimental period and was summarized in Table 3. The rate of TEAC increasing, as shown in Table 3 can be further figured as the following equation:

$$v_T = C4 - k_T [Fru] \quad \text{---------- (14).}$$

Where C4 was the constant, and k_T indicated the rate constant of TEAC increasing which were 2.58 x 10^{-5}, 3.09 x 10^{-4}, 2.22 x 10^{-3}, and 7.99 x10^{-3} Δ%/s·M for systems incubated at 45, 60, 75 and 90ºC separately. These temperature-rate constants sets were further taken linear regression, and concluded with Equation (15) and (16).

$$\log k_T = 0.0555\ T_C - 6.594 \quad \text{---------- (15).}$$

$$\ln (k_T) = 36.24 - 14822\ T_K^{-1} \quad \text{---------- (16).}$$

The slope in Equation (15) was equivalent to a Q_{10} of 3.59 for the TEAC increasing in fructose/glycine model systems at temperature ranged between 45 and 90ºC. And the slope in Equation (16) was equivalent to an activation energy 123 kJ/mol for TEAC increasing the fructose/glycine model systems.

LITERATURES

Ahmed, M.U., Thorpe, S.R., Baynes, J.W., (1986). Identification of N-ε-carboxymethyllysine as a degradation product of fructoselysine in glycated protein. *J. Biol. Chem., 61: 4889-4894.*

Amrani-Hemaimi, M., Cerny, C., and Fay, L. B., (1995). Mechanisms of formation of alkylpyrazines in the Maillard reaction. *J. Agric. Food Chem., 43: 2818-2822.*

Anese, M., Manzocco, L., Nicoli, M.C., and Lerici, C.R., (1999). Antioxidant properties of tomato juice as affected by heating. *J. Sci. Food Agric., 79: 750-754.*

Bailey, M.E. (1988). Inhibition of warmed-over flavor, with emphasis on Maillard reaction products. *Food Technol., 42(6): 123-126.*

Baptista, J.A.B., and Carvalho, R.C.B., (2004). Indirect determination of Amadori compounds in milk-based products by HPLC/ELSED/UV as an index of protein deterioration. *Food Research International, 37: 739-747.*

Baisier,W.M., and Labuza,T.P.(1992). Maillard browning kinetics in a liquid model system. *J. Agric. Food Chem., 40(5): 707-713.*

Brands, C.M.J., and van Boekel, M.A.J.S., (2003). Kinetic modeling of reactions in heated disaccharide-casein sein systems. *Food Chem., 83:13-26.*

Chen, S.-L., Jin, S.-Y., and Chen, C.-S., (2005). Relative reactivities of glucose and galactose in browning and pyruvaldehyde formation in sugar/glycine model systems . *Food Chemistry, 92: 597-605.*

Dutta, U., Cohenford, M.A., and Dain, J.A. (2005) Nonenzymatic glycation of DNA nucleosides with reducing sugars. *Analytical Biochemistry, 345: 171-180.*

Furth, A.J.. (1997) Glycated proteins in diabetes. *British Journal of Biomedical Science, 54:192- 200.*

Hodge,J.E. (1953). Chemistry of browning reaction in model system. *J. Agric. Food Chem. 1, 928-943.*

Hayashi,T., and Namiki,M. (1986). Role of sugar fragmentation in an early stage browning of amino-carbonyl reaction of sugar with amino acid. *Agric. Biol. Chem., 50(8), 1965-1970.*

Jing, H., and Kitts, D.D., (2000). Comparison of the antioxidative and cytotoxic properties of glucose-lysine and fructose-lysine Maillard reaction products. *Food*

Research International, 33: 509-516.

Jing, H., and Kitts, D.D., (2004a). Antioxidant activity of sugar-lysine Maillard reaction products in cell free and cell culture systems. *Archives of Biochem. Biophys., 429: 154-163.*

Jing, H., and Kitts, D.D., (2004b). Chemical characterization of different sugar-casein Maillard reaction products and protective effects on chemical-induced cytotoxicity of Caco-2 cells. *Food and Chemical Toxicity, 42: 1833-1844.*

Lertittikul, W., Benjakul, S., and Tanaka, M., (2005). Characteristics and antioxidative activity of Maillard reaction products from a porcine plasma protein-glucose model system as influenced by pH. *Food Chem.* (in press)

Liu, S.-C., Chang, H.-M., and Wu, J. S.-B., (2003). A study on the mechanism of browning in mei liqueur using model solutions. *Food Research International, 36: 579-585.*

López-Galilea, I., Andueza, S., di Leonard, I., de Peña, M.P., and Cid, C., (2006). Influence of torrefacto roast on antioxidant and pro-oxidant activity of coffee. *Food Chemistry, 94: 75-80.*

Maku, C., Shibamoto, T., (1991) Antioxidative activity of volatile heterocyclic compounds. *J. Agric. Food Chem, 39: 1990-1993.*

Miller, L. N., Rice-Evans, C. A., Davies, M. J., Gopinathan, V., Milner A. 1993. A noval method for measuring antioxidant status in premature neonates. *Clin, Sci. 84: 407-412.*

Nakayama, T., Hayase, F., Kato, H., (1980). Formation of ε-(2-formyl-4-hydroxymethyl-pyrrol-1-yl)- L-norleucine in the Maillard reaction between D-glucose and L-lysine. *Agric. Biol. Chem., 44: 1201-1210.*

Osada, Y., and Shibamoto, T., (2006) Antioxidative activity of volatile extracts from Maillard model systems. *Food Chem.* (in press).

Oyaizu, M. 1986. Studies on products of browning reaction: Antioxidant activities of products of browning reaction prepared from glucosamine. *Jpn. J. Nut. 44: 307-315.*

Rufian-Henares, J.A., and Morales, F.J., (2006). A new application of a commercial microtiter plate-based assay for assessing the antimicrobial activity of Maillard reaction products. *Food Research International, 39: 33-39.*

Schieberle, P., and Engel, W., (2002). Characterization of novel, sulfur-containing Maillard flavor compounds. *International Congress Series, 1245: 229-233).*

Shimada, K., Fujikawa, K., Yahara, K., Nakamura, T. (1992). Antioxidative properties of Xanthan on the autoxidation of soybean oil in cyclodextrin emulsion. *J. Agric. Food Chem. 40: 945-948*.

Shipanova, I.N., Glomb, M.A., and Nagaraj, R.H. (1997). Protein modification by methylgloxal: chemical nature and synthetic mechanism of a major fluorescent adduct. *Arch. Biophys., 344(1): 29-33.*

Wagner, K-H., Derkits, S., Herr, M., Schuh, W., and Elmadfa, I., (2002). Antioxidative potential of melanoidins isolated from a roasted glucose-glycine model. *Food Chem., 78: 375-382.*

Yanagimoto, K., Lee, K.-G., Ochi, H., and Shibamoto, T., (2002). Antitoxidative activity of heterocyclic compounds formed in Maillard reaction products. *International Congress Series, 1245: 335-340.*

Yaylayan, V.A., and Haffenden, L.J.W., (2003a). Mechanism of pyrazole formation in [^{13}C-2] labeled glycine model systems: N-N bond formation during Maillard reaction. *Food Research International, 36:571-577.*

Yaylayan, V.A., and Haffenden, L.J.W., (2003b). Mechanism of imidazole and oxazole formation in [^{13}C-2] labeled glycine and alanine model systems. *Food Chem., 81: 403-409.*

Yen,G.-C., Chau,C.-F., and Lii,J.-D. (1993). Isolation and charaterization of the most antimutagenic Maillard reaction products derived from xylose and lysine. *J. Agric. Food Chem., 41:771-776*

Yen, G-C., and Tsai, L.-C., (1993). Antimutagenicity of a partially fractionated Maillard reaction product. *Food Chemistry, 47(1): 11-15.*

Yilmaz, Y., and Toledo, R., (2005). Antioxidant activity of water-soluble Maillard reaction products. *Food Chem., 93: 273-278.*

Yim,H.S., Kang,S.O., Hah,Y.C., Chock,P.B., and Yim,M.B. , (1995). Free radicals generated during the glycation reaction of amino acids by methylglyoxal. A model study of protein-cross-linked free radicals. *J. Biol. Chem., 270(47): 28228-28233.*

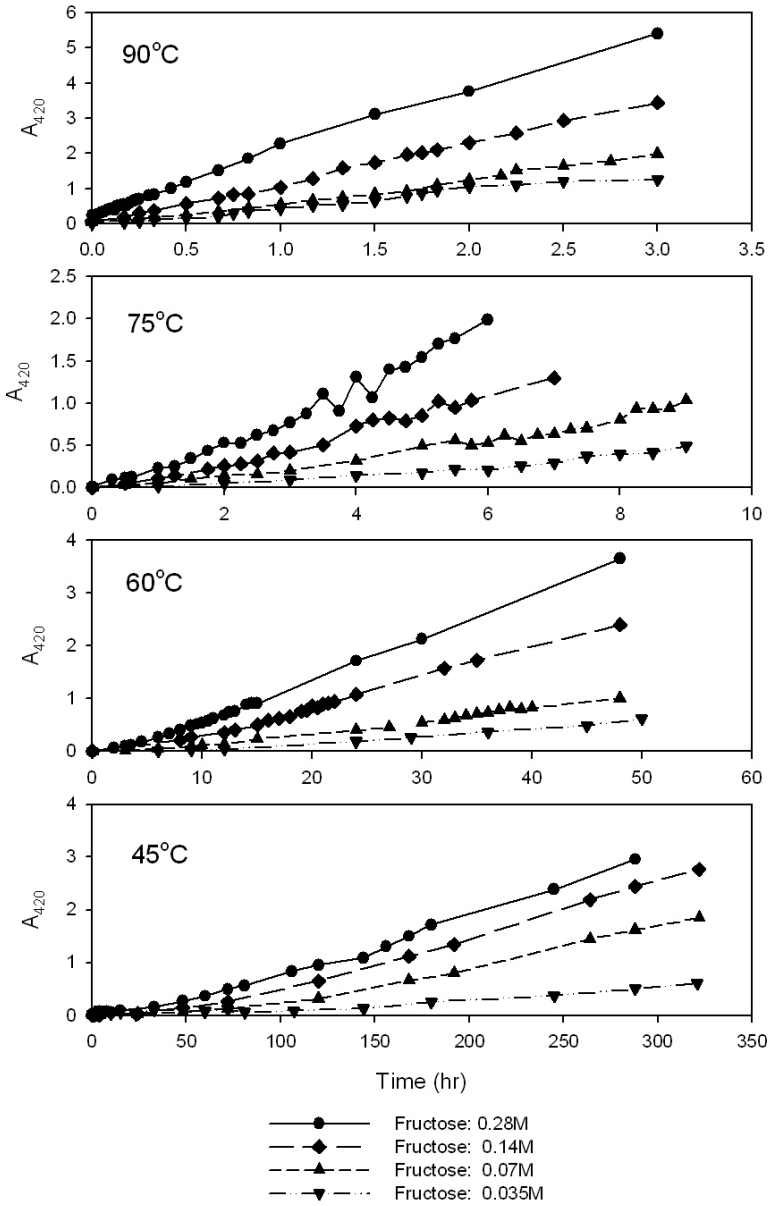

Figure 1. Development of brown color in fructose/glycine model systems incubated at 45, 60, 75, and 90°C.

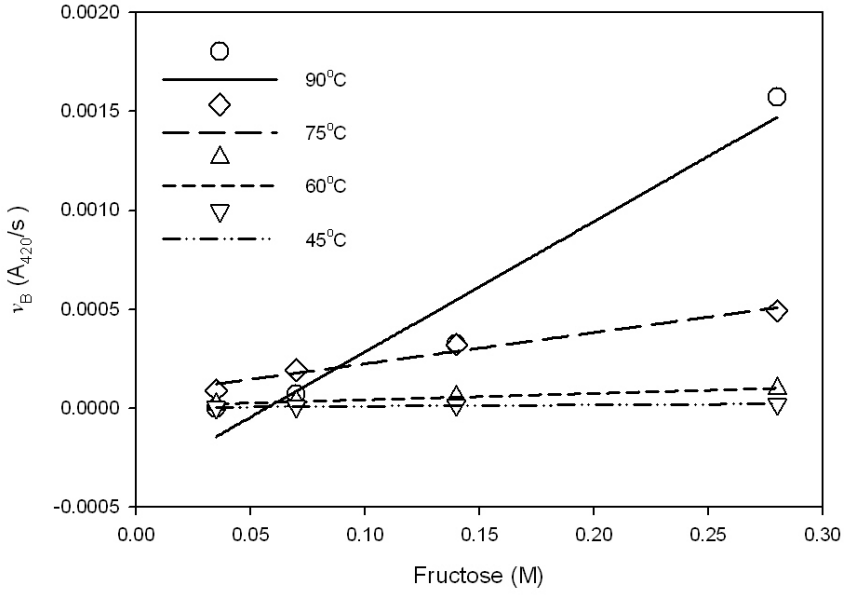

Figure 2. Effects of fructose concentration and temperature on the browning rate of fructose/glycine model systems.

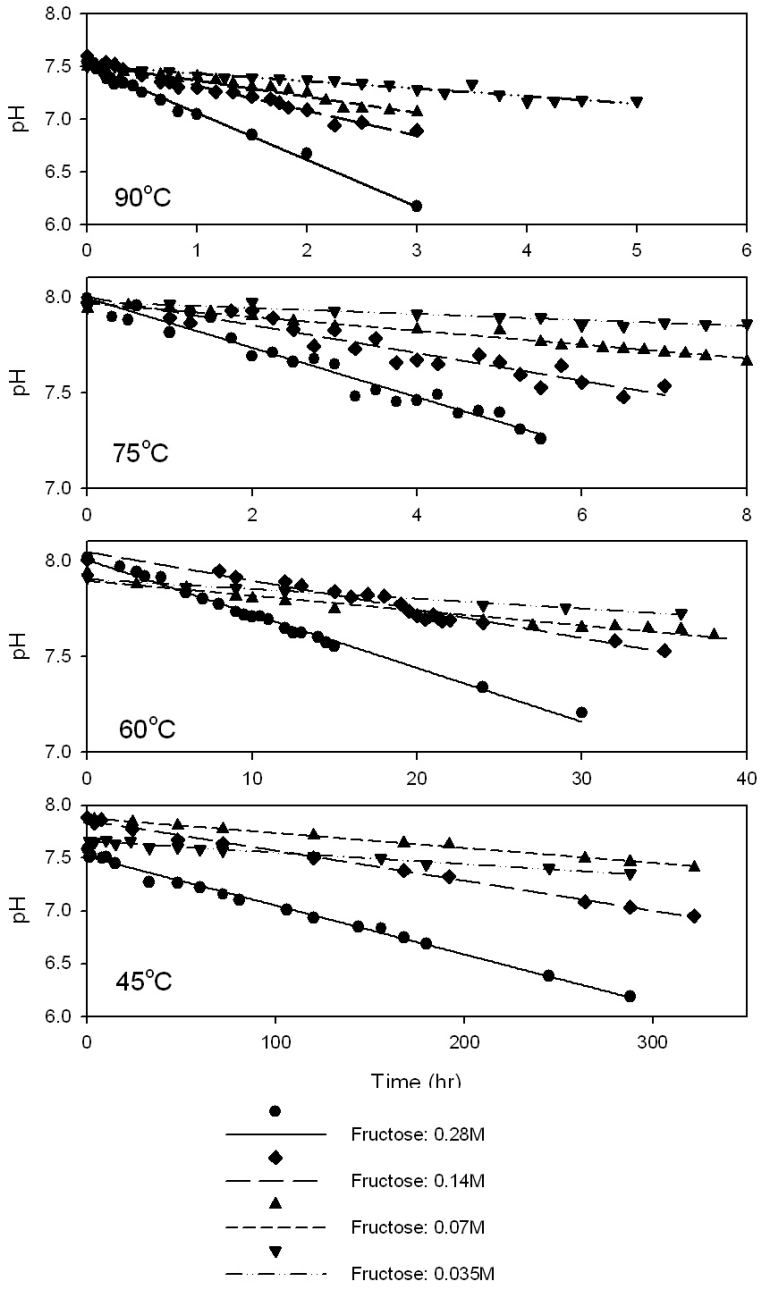

Figure 3. Change of pH in model fructose/glycien systems incubate at 45, 60, 70, and 90°C.

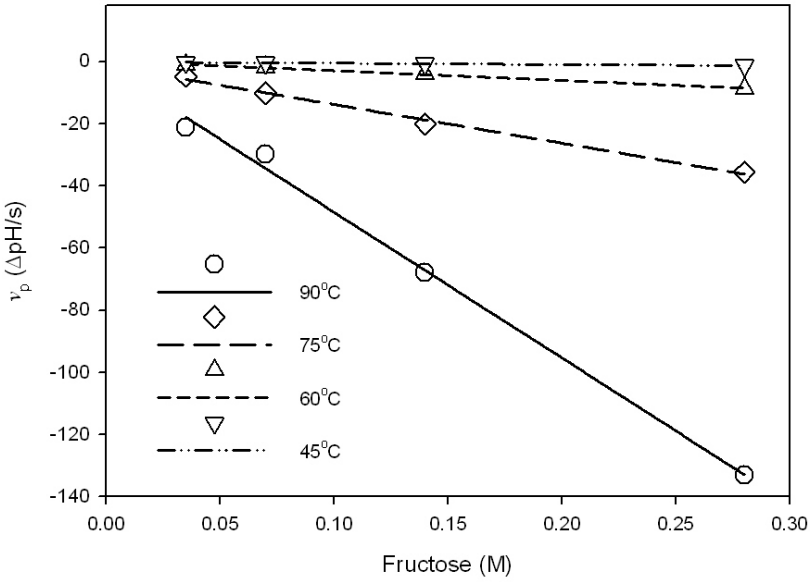

Figure 4. Effects of fructose concentration and temperature on the rates of pH decreasing in fructose/glycine model systems.

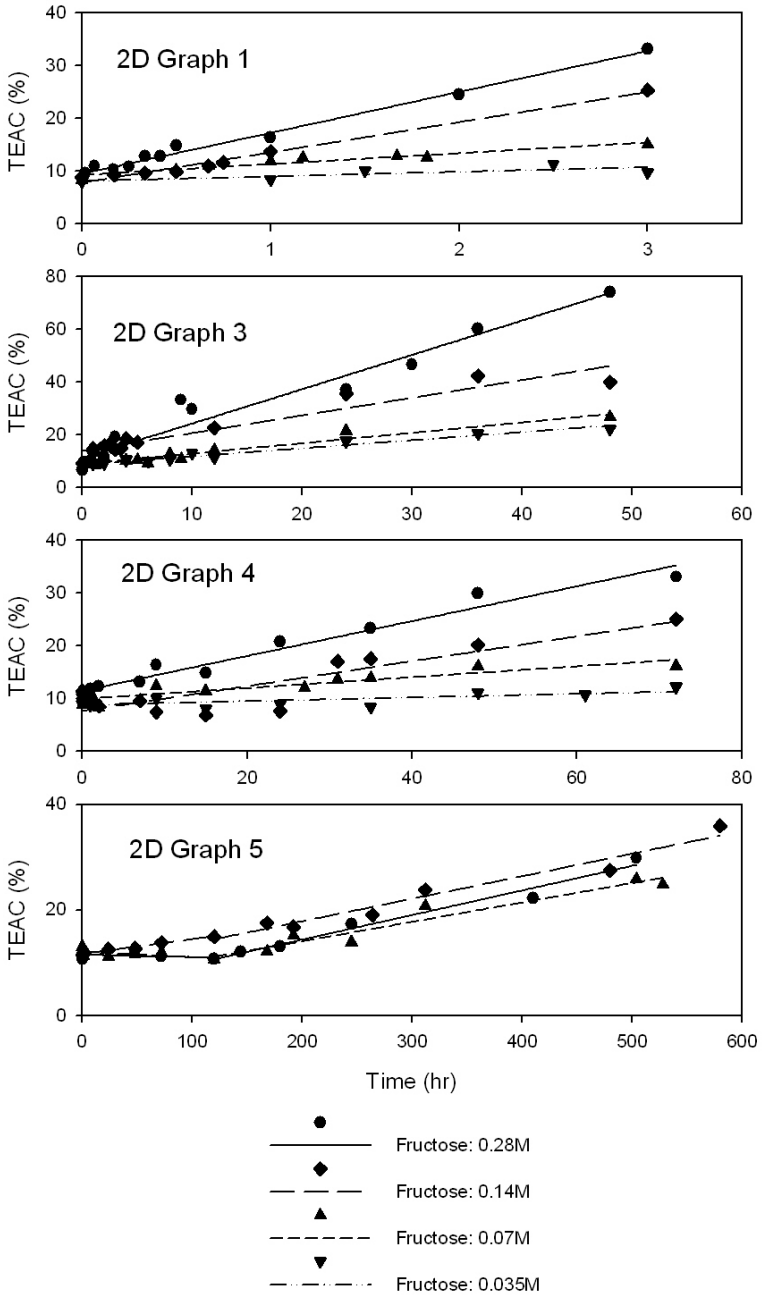

Fig 5. TEAC changes of model fructose/glycine systems at 45°C, 60°C, 75°C, and 90°C.

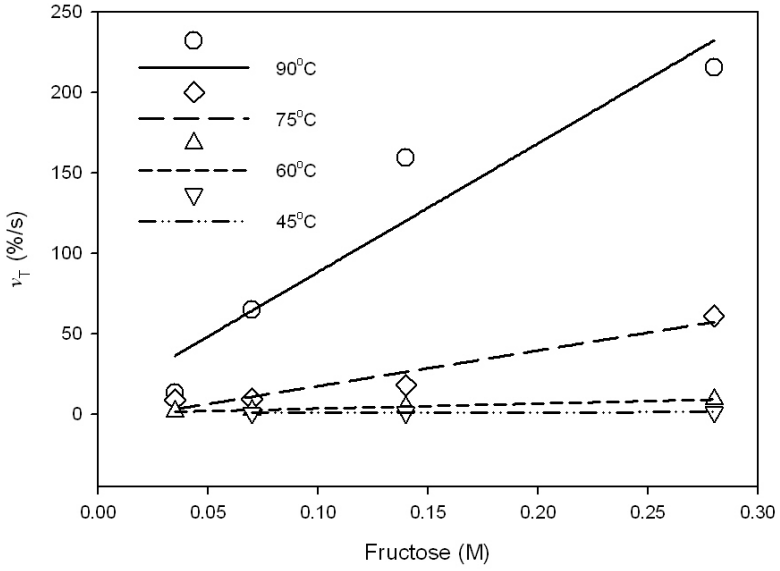

Figure 6. Effects of fructose concentration and temperature on the rate of TEAC changes in fructose/glycine model systems.

Table I Browning rate of model fructose/glycine systems incubated at 45, 60, 75, and 90˚C

Fructose	v_B (x10^{-5} ΔA_{420}/s)				intercept (A_{420})				R^2			
(M)	90˚C	75˚C	60˚C	45˚C	90˚C	75˚C	60˚C	45˚C	90˚C	75˚C	60˚C	45˚C
0.28	49.30	9.98	2.19	0.31	0.262	-0.248	-0.233	-0.348	0.996	0.971	0.997	0.992
0.14	32.09	5.95	1.64	0.29	-0.013	-0.184	-0.358	-0.563	0.998	0.981	0.998	0.997
0.07	19.31	3.56	0.87	0.22	-0.133	-0.197	-0.388	-0.647	0.992	0.958	0.979	0.997
0.035	9.16	1.90	0.48	0.09	-0.222	-0.155	-0.244	-0.338	0.996	0.978	0.994	0.976

Table 2. Changes of pH in model fructose/glycine systems incubated at 45, 60, 75, and 90˚C.

Fructose	v_{pH} (x10^{-6} △pH/s)				intercept (pH)				R^2			
(M)	90˚C	75˚C	60˚C	45˚C	90˚C	75˚C	60˚C	45˚C	90˚C	75˚C	60˚C	45˚C
0.28	-133.10	-35.62	-8.72	-1.29	7.51	7.99	8.03	7.51	0.969	0.959	0.995	0.992
0.14	-68.08	-20.19	-3.98	-0.79	7.56	8.00	8.03	7.85	0.971	0.894	0.968	0.997
0.07	-29.98	-10.27	-1.93	-0.39	7.46	7.97	7.87	7.88	0.993	0.987	0.979	0.997
0.035	-21.18	-4.83	-1.15	-0.26	7.52	7.98	7.89	7.66	0.980	0.904	0.977	0.987

Table 3. Rate of TEAC changes of model fructose/glycine systems at 45˚C, 60˚C, 75˚C, and 90˚C.

Fructose	v_T (x10^{-5}%s)				intercept (%)				R^2			
(M)	90˚C	75˚C	60˚C	45˚C	90˚C	75˚C	60˚C	45˚C	90˚C	75˚C	60˚C	45˚C
0.28	215.40	61.10	9.17	1.44	9.49	7.27	11.50	3.56	0.989	0.939	0.958	0.994
0.14	159.00	18.12	4.85	1.17	7.80	14.11	11.76	10.44	0.986	0.946	0.994	0.969
0.07	65.10	9.48	2.60	0.88	9.00	9.54	10.95	6.22	0.878	0.983	0.992	0.953
0.035	13.50	9.06	1.65	NS	8.81	8.91	7.98	NS	0.850	0.976	0.974	NS