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ScienceDirect
Food Chemistry 103 (2007) 1297–1304
Food Chemistry
www.elsevier.com/locate/foodchem

Antioxidant formation by γ-irradiation of glucose–amino acid model systems

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Received 17 July 2006; received in revised form 17 August 2006; accepted 17 October 2006

Abstract

The efficacy of γ-irradiation for the formation of Maillard reaction products (MRPs) from glucose and lysine/glycine and the antioxidant potential of MRPs thus formed were examined. Formation of MRPs was observed by monitoring absorbance at 284 nm and 420 nm. Upon irradiation, there was a dose-dependent increase in absorbance ($r^2 = 0.99$) at both the wavelengths. Irradiation of glucose/lysine solution resulted in higher absorbance at 284 nm than did that of glucose/glycine solution. Similarly, increase in absorbance at 420 nm was observed upon irradiation in both the systems. No significant absorbance was observed with unirradiated solution of glucose and lysine/glycine. These findings thus clearly revealed the formation of intermediate products and brown complexes (of Maillard reaction) upon irradiation of glucose/amino acid solution. A fluorescence was also observed in irradiated glucose/amino acid solution, whereas, none was seen in non-irradiated solution. These observations further confirmed the formation of MRPs, as fluorescent compounds are known to be precursors of brown pigments formed during the Maillard reaction. These MRPs exhibited excellent antioxidant activity, as measured by 1,1-diphenyl-2-picrylhydrazyl (DPPH) and β-carotene bleaching assays. MRPs, formed at a 40 kGy dose, scavenged up to 62% of DPPH radical and 82% of β-carotene was protected from bleaching. Reducing power of MRPs, estimated using the ferricyanide, method was also increased as compared to non-irradiated solutions. Further, these MRPs were able to scavenge hydroxyl radical and superoxide anion radical to the extents of 33% and 58%, respectively. These MRPs could chelate iron to an extent of 32% under in vitro conditions. Thus, these studies clearly demonstrated that radiation technology could be employed for obtaining novel antioxidants from sugar and amino acid combinations.

Keywords: Maillard reaction; γ-Irradiation; Antioxidant; Free radicals

1. Introduction

The Maillard reaction involves the formation of brown pigments by condensation between carbonyl groups of reducing sugars, aldehydes or ketones and amine groups of amino acids, peptides or proteins or other nitrogenous compounds (Jing & Kitts, 2002; Yoo, Kim, Kim, & Kang, 2004). The Maillard reaction, produced from an amino acid–sugar model system, has been known to be associated with the formation of compounds with pronounced antioxidant activity (Yoshimura, Ujima, Watanabe, & Nakazawa, 1997). It is one of the major reactions taking place during thermal processing, cooking and storage of foods. A myriad of products is formed, which have direct impact on nutritional and sensory qualities of foods. The antioxidative properties of MRPs produced by heat treatment of amino acid–sugar have been studied in model systems by a number of investigators (Jayathilakan & Sharma, 2006; Kirigaya, Kato, & Fujimake, 1968; Lingnert & Eriksson, 1980; Yamaguchi, Koyama, & Fujimake, 1981). Similarly, antimutagenic characteristics of MRPs have also been reported (Yun & Tsai, 1993). Radiation processing enhances shelf-life and/or improves the microbiological safety of raw and processed food materials without

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doi:10.1016/j.foodchem.2006.10.035

compromising nutritional quality (WHO, 1999). Many of the chemical changes caused by radiation processing of food are similar to those of other preservation methods (Diehl, 1995). During radiation processing of food, chemical changes arise as a result of direct action on the food components or by indirect action through reactive intermediates formed by radiolysis of water (Diehl, 1995). Most of these chemical changes are similar to those produced by heat treatment. However, information on formation of MRPs by radiation processing is scanty. Non-enzymatic browning in γ-irradiated aqueous solutions of different sugars with lysine has recently been reported (Oh et al., 2006). However, there is no report on assessment of antioxidant potential of sugar–amino acid solutions upon irradiation. The present studies were therefore carried out to investigate formation of MRPs by radiation treatment of sugar and amino acid solutions and to examine their antioxidant activity. In a model system, the sugar chosen for the present study was glucose and amino acids were glycine or lysine.

2. Materials and methods

2.1. Chemicals

β-Carotene, 2,2,diphenyl-1-picryl hydrazyl (DPPH), thiobarbituric acid (TBA), nitroblue tetrazolium (NBT) and linoleic acid were purchased from Sigma Chemical Co. (St. Louis, MO). All other chemicals used were of analytical grade and procured from Himedia Laboratories (Mumbai, India) or Sisco Research Laboratories (Mumbai, India).

2.2. Preparation of radiation-induced MRPs

Glucose and amino acids (glycine, lysine) solutions were prepared separately in sodium phosphate buffer (100 mM, pH 7.0) to get a final concentration of 200 mM. Glucose (10.0 ml) of was then mixed with 10.0 ml of either amino acid solution in a screw-cap tube. Similar mixtures were prepared in distilled water and Tris–HCl buffer (100 mM, pH 7.0) to study the effect of buffer type on radiation-induced changes.

The sugar amino acids mixtures thus prepared were subjected to different doses of γ-irradiation (0–100 kGy) in a Gamma-cell 5000 (BRIT, Mumbai) at a dose rate of 9.87 kGy/h. Dosimetry was performed by a ceric-cerous dosimeter calibrated against Fricke's dosimeter. Dosimetry intercomparison was carried out with National Standards established by the Radiological Physics and Advisory Division (RP&AD), Bhabha Atomic Research Centre (BARC), Mumbai, India.

2.3. Spectrophotometric analyses

Absorbance at 284 nm (early Maillard reaction products) and 420 nm (late Maillard reaction products) of radiation-treated sugar amino acid solutions appropriately diluted, was measured. Fluorescence of samples was determined after 100-fold dilution. The fluorescence intensity was measured at an excitation wavelength of 365 nm and emission wavelength of 440 nm, using a fluorescence spectrophotometer (Matiacevich & Buera, 2006).

2.4. Determination of electron-donating ability

The DPPH radical-scavenging assay (Blois, 1958) was employed to determine electron-donating ability of radiation-induced MRPs. To a 1 ml aliquot of appropriately diluted solution, 1 ml of ethanolic DPPH$^{\cdot}$ solution (0.2 mM) was added. The mixture was vortexed and left to stand at ambient temperature for 30 min. Reaction mixture containing 1 ml distilled water and 1 ml of ethanolic DPPH$^{\cdot}$ solution (0.2 mM) served as the control. The absorbance of the solution was measured spectrophotometrically at 517 nm. The percentage of DPPH$^{\cdot}$-scavenging was calculated from the equation:

$$\text{Radical scavenging activity } (\%) = (A_{\text{Control}} - A_{\text{Sample}})/(A_{\text{Control}}) \times 100.$$

where A_{Control} is absorbance of control and A_{Sample} absorbance of sample.

2.5. Determination of antioxidant activity by β-carotene bleaching assay

Antioxidant activity of the aqueous solution was determined by a β-carotene/linoleic acid system, as described by Matthaus (2002). Briefly, 1 ml of β-carotene solution (1 mg/ml in chloroform), 40 μl of linoleic acid and 400 μl of Tween 80 were transferred to a round-bottom flask. Chloroform from the sample was evaporated using a stream of nitrogen. Then, 100 ml of distilled water were added slowly to the residue and vigorously agitated to give a stable emulsion. To an aliquot of 4.5 ml of this emulsion, 500 μl of appropriately diluted samples were added. To the control reaction mixtures, 500 μl of distilled water were added. Absorbance was measured, immediately, at 470 nm. The tubes were placed in a water bath at 50 °C and the absorbance was measured after 120 min. Antioxidant activity index (AAI) was calculated as:

$$\text{AAI} = \frac{A_{s(0)} - A_{s(120)}}{A_{b(0)} - A_{b(120)}} \times 100$$

where $A_{s(0)}$ is absorbance of sample at 0 min, $A_{s(120)}$ is absorbance of sample at 120 min, $A_{b(0)}$ is absorbance of control at 0 min, $A_{b(120)}$ is absorbance of control at 120 min.

2.6. Determination of reducing power

Reducing power was determined by the ferricyanide method of Yen and Duh (1993). Appropriately diluted

sample (1 ml) was added to 2.5 ml of phosphate buffer (200 mM, pH 6.6), followed by 2.5 ml of 1% potassium fericyanide. The reaction mixture was incubated for 20 min. in a water bath at 50 °C. After incubation, 2.5 ml of 10% trichloroacetic acid (TCA) were added, followed by centrifugation at 3000 rpm for 10 min. The upper layer (5 ml) was mixed with 5 ml distilled water and 1 ml of 0.1% ferric chloride. Absorbance of the resultant solution was measured at 700 nm.

2.7. Determination of hydroxyl radical scavenging activity

Hydroxyl radical-scavenging activities of radiation-induced MRPs were determined according to the modified method of Halliwell, Gutteridge, and Aruoma (1987). To 1 ml of the appropriately diluted sample, 1 ml of phosphate buffer (0.1 M, pH 7.4) containing 1 mM ferric chloride, 1 mM EDTA, 1 mM ascorbic acid, 30 mM deoxyribose and 20 mM hydrogen peroxide, was added. After incubation at 37 °C for 90 min, 2 ml of 2% (w/v) TCA and 2 ml of 1% (w/v) TBA were added. The reaction mixture was heated in a boiling water bath 15 min. The absorbance of the pink colour developed was measured at 532 nm, using a spectrophotometer. The percentage of hydroxyl radical-scavenging activity was calculated as:

$$\%\ \text{Inhibition} = [(A_{\text{Control}} - A_{\text{Sample}})/A_{\text{Control}}] \times 100.$$

where A_{Control} is absorbance of control and A_{Sample} absorbance of sample

2.8. Measurement of superoxide anion-scavenging activity

Superoxide anion-scavenging activities of radiation-induced MRPs were determined according to the method of Liu, Ooi, and Chang (1997) with some modifications. The reaction mixture consisted of 1 ml of NBT (156 μM in 0.1 M potassium phosphate buffer pH 7.4), 1.0 ml of reduced nicotinamide adenine dinucleotide (NADH 468 μM in 0.1 M potassium phosphate buffer pH 7.4) and 0.5 ml of appropriately diluted sample. The reaction was initiated by addition of 100 μl of phenazine methosulphate (PMS 60 μM in 0.1 M potassium phosphate buffer pH 7.4) to the mixture. The tubes were incubated at ambient temperature for 5 min and the absorbance was measured at 560 nm. Decreased absorbance of the reaction mixture indicated increased superoxide anion-scavenging activity. The percentage inhibition of superoxide anion generation was calculated using the following formula:

$$\%\ \text{Inhibition} = [(A_0 - A_s)/A_0] \times 100$$

where A_o is absorbance of the control and A_s is absorbance of the sample.

2.9. Determination of iron-chelation activity

The ferrous ion-chelation potentials of radiation induced MRPs were investigated by estimating the ferrous iron–ferrozine complex at 562 nm (Decker & Welch, 1990). Briefly, the reaction mixture consisted of 1.0 ml of appropriately diluted sample, 3.7 ml distilled water, 0.1 ml ferrous chloride (2 mM) and 0.2 ml ferrozine (5 mM). The reaction mixture, containing 1 ml of distilled water instead of sample, served as control. Tubes were incubated at ambient temperature for 20 min. The absorbance of the colour developed was measured at 562 nm. The ability of sample to chelate ferrous ions was calculated using the following equation:

$$\text{Chelation activity}\ (\%) = [(A_0 - A_s)/A_0] \times 100$$

where A_o is absorbance of the control and A_s is absorbance of the sample.

2.10. Statistical analysis

All results given in figures are means ± standard deviation. Differences between the variables were tested for significance by one-way ANOVA with Turkey's post test, using GraphPad InStat version 3.05 for windows 95, GraphPad Software, San Diego California USA www.graphpad.com. Differences at $p < 0.05$ were considered to be significant.

3. Results and discussion

In the present study, the formation of Maillard reaction products (MRPs) by γ-irradiation of a model system, consisting of a sugar (glucose) and an amino acid (glycine or lysine), was investigated. A sharp increase in UV absorbance at 284 nm of glucose/glycine and glucose/lysine solutions was observed with irradiation dose ($r^2 = 0.99$) (Fig. 1). In samples irradiated at 100 kGy, these values, after correcting for dilution factor, were 38.9 and 46.8 for glucose/glycine and glucose/lysine solutions, respectively. These

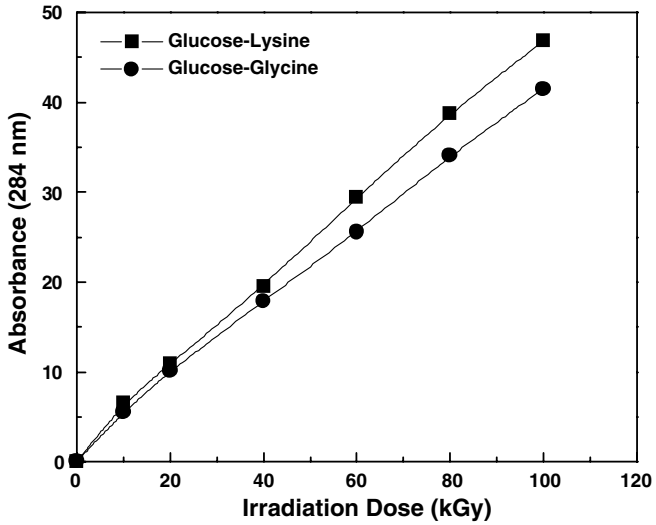

Fig. 1. UV absorbance (A_{284}) of glucose/amino acid solution as a function of irradiation dose. Values represented after correcting for dilution factor.

findings suggested formation of intermediate products upon irradiation. The Maillard reaction is associated with development of UV-absorbing intermediate compounds prior to generation of brown pigments. This increased UV absorbance is attributed to decomposition of sugars by dehydration and sugar fragmentation (Hodge, 1953). Similarly, Oh et al. (2006) investigated effects of γ-irradiation (up to 30 kGy) on UV absorbance, in aqueous solutions of different sugars and sugar/lysine mixtures and observed dose-dependent increase in UV absorbance. The UV absorbance of glucose/lysine solution was much greater than that of glucose/glycine irradiated at the same dose. UV-absorbing intermediate compounds are formed during heat-induced formation of MRPs (Ajandouz, Tchiakpe, Ore, Benajiba, & Puigserver, 2001; Moreno, Molina, Olano, & Lopez-Fandino, 2003).

The extent of browning, as a function of irradiation dose, is depicted in Fig. 2. It can be seen that the browning intensity, as measured by A_{420} nm for glucose/glycine and glucose/lysine solutions increased ($r^2 = 0.99$) from 0.002 to 3.56 and 3.81, respectively. Lysine was found to be more reactive than glycine when compared at the same irradiation dose. By irradiating a solution of glucose alone, negligible absorbance was observed, confirming that, in the sugar/amino acid solution, browning is due to the Maillard reaction and not due to caramelisation of the sugar. A similar finding with heat-treated sugar solution in the presence and absence of amino acid has been earlier reported (Morales & Jimenez-Perez, 2001). Our findings are in agreement with a recent report where dose dependent increase in A_{420} nm of aqueous solutions of different sugars with lysine was observed (Oh et al., 2006). However, the authors investigated doses up to 30 kGy; in the present study we observed increase in browning up to 100 kGy. Our findings are in concurrence with other studies where browning of sugar/amino acid solution due to heat-induced Maillard reaction increased with heating time (Cammemer & Kroh, 1996; Morales & Jimenez-Perez, 2001).

To evaluate the free radical-scavenging, glucose and glucose/amino acid solutions subjected to different dose of γ-irradiation, were allowed to react with stable DPPH free radical. The scavenging of DPPH free radical, indicating a positive antiradical activity, was followed by monitoring reduction in absorbance at 517 nm. DPPH free radical-scavenging activity increased with irradiation dose in glucose/lysine ($r^2 = 0.95$), as well as in glucose/glycine ($r^2 = 0.88$). At 40 kGy, in the case of glucose/lysine solution, it was observed to be 62.8% (Fig. 3). Upon further irradiation to 80 kGy, the increase was not linear and tended to plateau at higher irradiation dose. Similarly, radical-scavenging activity of glucose/glycine increased with the dose but was lower than that of glucose/lysine. No significant DPPH free radical-scavenging activity was observed in sugar solution, irrespective of irradiation dose. Capability of heat-induced MRPs to scavenge DPPH radical has been reported in a number of studies (Jing & Kitts, 2002; Benjakul, Lertittikul, & Bauer, 2005; Morales & Jimenez-Perez, 2001; Murakami et al., 2002; Yen & Hsieh, 1995). However, there are no reports showing radical-scavenging activity by radiation induced MRPs. The radical-scavenging activities of radiation-induced MRPs were saturated with irradiation dose of 40 kGy. Similar findings have been reported with heat-induced MRPs, where radical-scavenging activities tend to saturate after certain heating times (Morales & Jimenez-Perez, 2001; Jing & Kitts, 2002).

It was observed that β-carotene bleaching was significantly ($P < 0.05$) inhibited in the presence of irradiated sugar/amino acid solution, whereas no protection was offered by non-irradiated solution (Fig. 4). Thus, these findings indicated that compounds formed upon irradiation of sugar/amino acid solution have significant antioxidant potential. The mechanism of bleaching of β-carotene

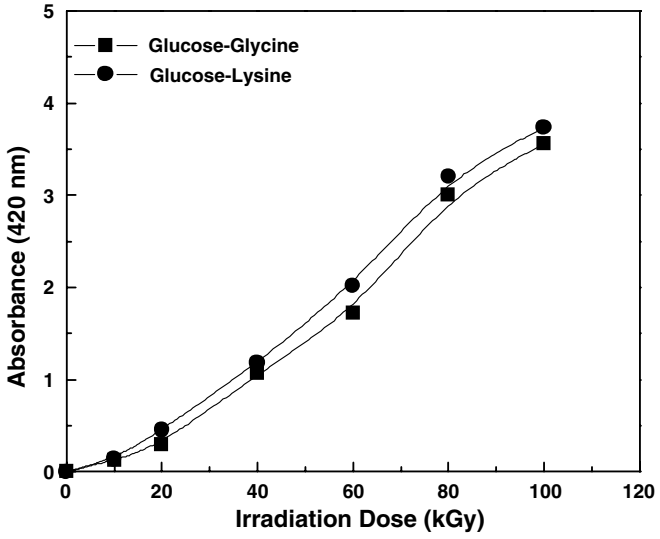

Fig. 2. Browning (A_{420}) of glucose/amino acid solution as a function of irradiation dose. Values represented after correcting for dilution factor.

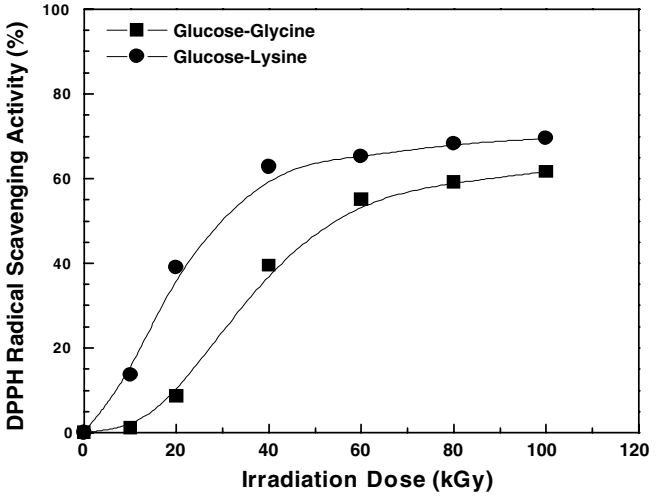

Fig. 3. DPPH radical-scavenging activity of glucose/amino acid solution as a function of irradiation dose.

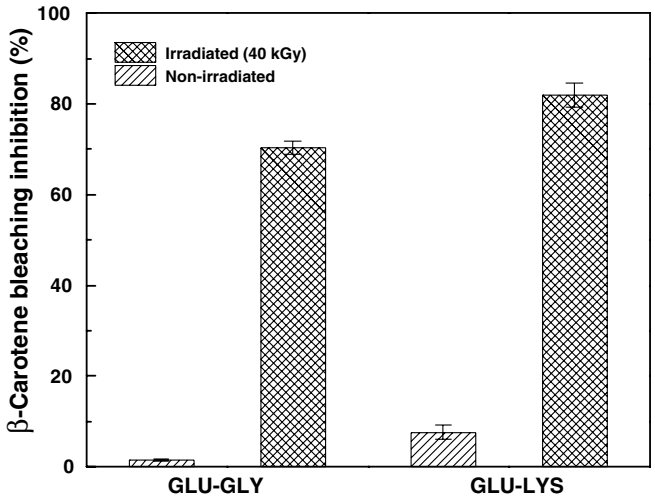

Fig. 4. Effect of γ-irradiation (40 kGy) on β-carotene bleaching inhibition activity of glucose/amino acid solution.

is a free radical-mediated phenomenon, resulting from the hydroperoxides formed from linoleic acid. In this model system, β-carotene undergoes rapid discoloration due to attack of free radicals formed upon abstraction of a hydrogen atom from the diallylic methylene group of linoleic acid. The presence of an antioxidant in the reaction mixture hinders the rate of bleaching by neutralizing free radicals formed in the system during incubation at 50 °C (Wettasinghe & Shahidi, 1999). Synthesis of antioxidative compounds upon heat treatment of sugar/amino acid solutions is reported in a number of studies (Jayathilakan & Sharma, 2006; Jing & Kitts, 2002; Lingnert & Eriksson, 1980; Yamaguchi et al., 1981; Yoshimura et al., 1997; Morales & Jimenez-Perez, 2001).

Iron reducing power of sugar/amino acid solution before and after subjecting it to γ-irradiation is shown in Fig. 5. It was seen that non-irradiated sugar/amino acid solutions had negligible reducing power, which increased significantly ($P < 0.05$) upon irradiation treatment. As observed in the case of radical-scavenging and β-carotene bleaching assay, reducing power of glucose/lysine was significantly higher than that of glucose/glycine when a comparison was made at the same irradiation dose. It has been reported that compounds responsible for reducing activity are formed during thermolysis of Amadori products in the primary phase of Maillard reactions (Hwang, Shue, & Chang, 2001) or could be heterocyclic products of Maillard reaction or caramelisation of sugars (Charurin, Ames, & Castiello, 2002). Possibly, γ-irradiation induces similar changes in sugar/amino acid solution, resulting in formation of products which contribute toward the reducing power. Heat-induced MRPs from xylose–lysine (Yen & Hsieh, 1995), glucose–glycine (Yoshimura et al., 1997), sugar–lysine (Wijewickreme, Krejpcio, & Kitts, 1999) and a porcine plasma protein–glucose model (Benjakul et al., 2005) possessed reducing power.

Development of fluorescent compounds has been reported to be associated with heat-induced Maillard reaction (Jing & Kitts, 2002, 2000). In the present study, formation of fluorescent compounds was observed in irradiated glucose/amino acid solution. The fluorescences of irradiated glucose/glycine and glucose/lysine solution were 7- and 10-fold higher than that of corresponding non-irradiated control solutions (Fig. 6). These fluorescent compounds are reported to be precursors of brown pigments formed during Maillard reaction (Leclere & Birlouez-Aragon, 2001).

The Fenton reaction system was used in deoxyribose degradation, by generating hydroxyl radicals. The treatment of deoxyribose with the Fenton reaction reagent used in this experiment resulted in a high rate of deoxyribose degradation. The hydroxyl radical is the most reactive of species and induces the most severe damage to adjacent biomolecules, resulting in lipid peroxidation in biological systems (Gutteridge, 1984). Hydroxyl radical-scavenging

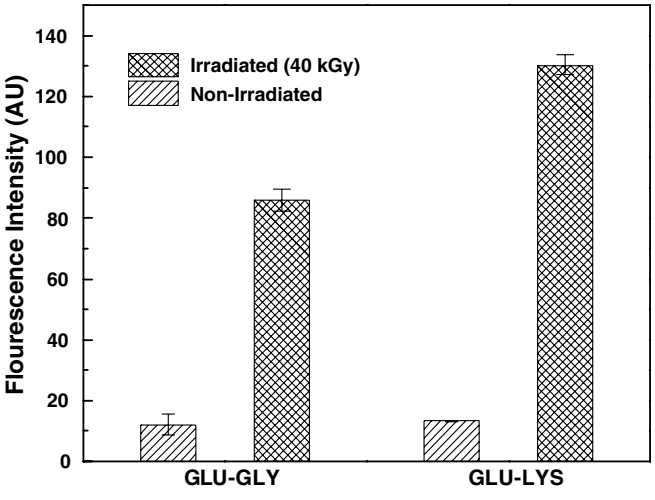

Fig. 5. Effect of γ-irradiation (40 kGy) on florescence of glucose/amino acid solution.

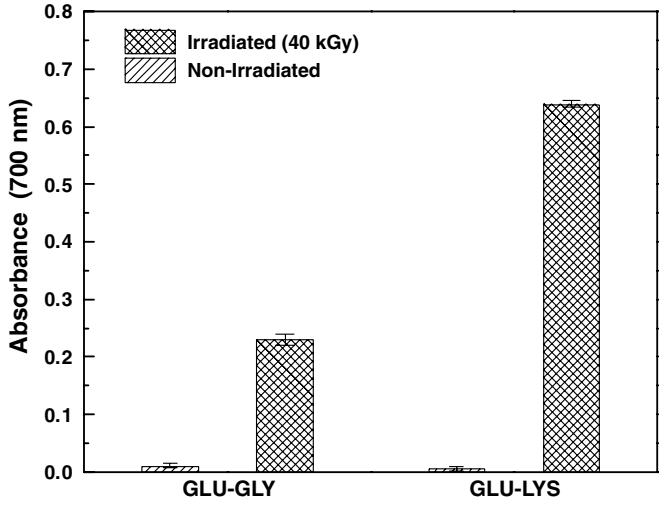

Fig. 6. Effect of γ-irradiation (40 kGy) on reducing power of glucose/amino acid solution.

activity of irradiated glucose/amino acid solution was threefold higher than that of their non-irradiated counterparts (Fig. 7). These findings revealed that compounds formed upon irradiation treatment of glucose/amino acid solutions have potential as antioxidants in biological systems. Development of compounds capable of scavenging hydroxyl radical and utility of this test in studies to demonstrate in vitro hydroxyl radical-scavenging activity of heat-induced MRPs has been reported (Aruoma, 1994; Jing & Kitts, 2002, 2000; Wijewickreme et al., 1999).

The superoxide radicals are generated by numbers of biological reactions. Although they do not directly initiate lipid oxidation, superoxide radical anions are precursors of highly reactive hydroxyl radical, which contributes toward lipid peroxidation in biological systems. Thus, superoxide anion-scavenging activity indirectly contributes toward antioxidant potential. Irradiated glucose/glycine and glucose/lysine exhibited $43.3 \pm 1.5\%$ and $56.7 \pm 1.3\%$ superoxide anion radical-scavenging whereas their non-irradiated counterparts exhibited just $3.5 \pm 0.5\%$ and $1.9 \pm 0.1\%$ of activity (Fig. 8). Our results are in agreement with a previous study on heat-induced MRPs formed from glucose/glycine that were reported to scavenge superoxide anion radical (Yoshimura et al., 1997).

The Fe^{2+} ion is the most powerful pro-oxidant among various species of metal ion (Yomauchi, Tatsumi, Asano, Kato, & Ueno, 1988). Metal chelation activity plays an important role in the antioxidant action as it results in reduction in the concentration of the transition metal required for lipid peroxidation. Ferrous ion-chelating activities of sugar/amino acid solutions are shown in Fig. 9. It can be seen that iron-chelation activity of sugar/amino acid solutions significantly ($P < 0.05$) increased upon irradiation. Irradiated glucose/glycine and glucose/lysine exhibited $29.3 \pm 0.5\%$ and $32.2 \pm 1.2\%$ iron-chelation. In contrast, non-irradiated samples exhibited only $4.5 \pm 1.5\%$ and $9.9 \pm 1.1\%$ of activity. Our finding are in concurrence with an earlier report, where iron-chelation activity was observed in a glucose/glycine model system as a result of MRPs formed due to heat treatment (Yoshimura et al., 1997).

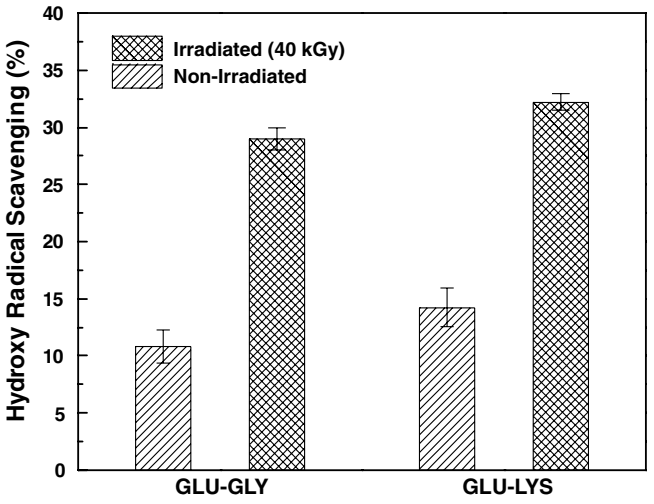

Fig. 8. Effect of γ-irradiation (40 kGy) on hydroxyl-scavenging activity of glucose/amino acid solution.

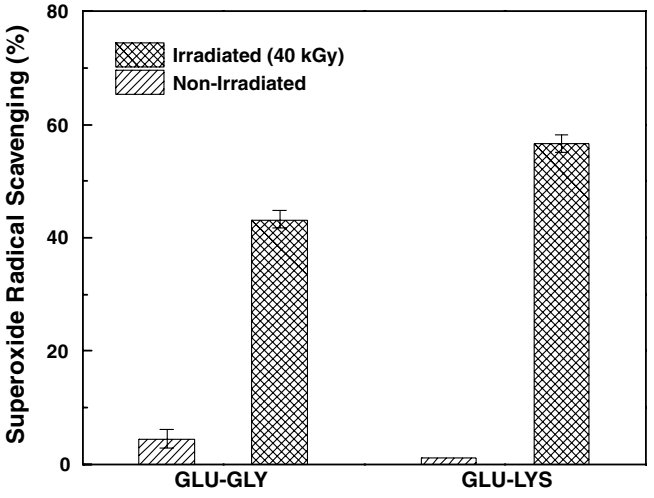

Fig. 9. Effect of γ-irradiation (40 kGy) on superoxide radical-scavenging activity of glucose/amino acid solution.

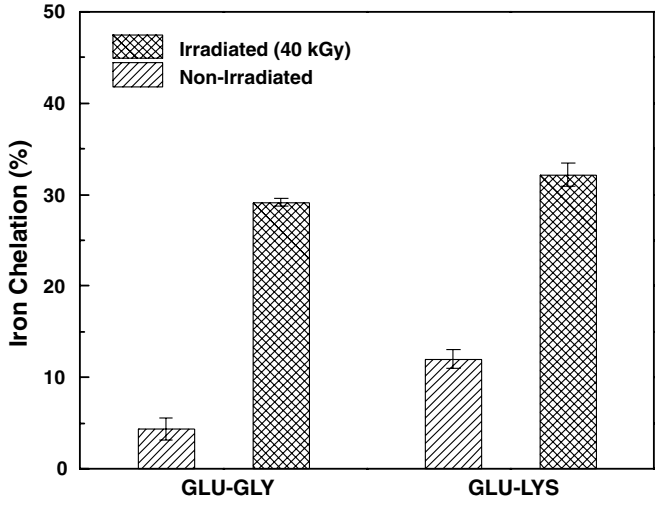

Fig. 7. Effect of γ-irradiation (40 kGy) on iron-chelation activity of glucose/amino acid solution.

The pH of the reaction mixture significantly influences both the reaction rate and type of products formed during heat-induced Maillard reaction; generally, the reaction rate increases as pH increases (Apriyantono & Ames, 1993; Ashoor & Zent, 1984; Bell, 1997). Therefore, a comparison of formation of MRPs was made by preparing reaction mixtures in distilled water, phosphate buffer (100 mM, pH 7) and Tris–HCl (100 mM, pH 7) and irradiating the samples at a 40 kGy dose. It was observed that pH of the reaction mixture, after irradiation, was not affected when it was prepared in buffer but significantly decreased when distilled water was used instead of buffer. UV absorbance, browning and radical-scavenging activity were found to be

the highest in the phosphate buffer, followed by Tris–HCl buffer, and lowest in distilled water (Table 1). Our results are in agreement with the earlier studies where the catalytic role of phosphate buffer in heat-induced model systems has been reported (Potman & Van Wijk, 1989). The role of phosphate anion as a bifunctional catalyst for nucleophilic reaction of the amine with carbonyl, due to its ability to simultaneously donate and accept the proton necessary for conversion, was suggested earlier (Bell, 1997; Watkins et al., 1987). Most of the foods have neutral pH; proteins and amino acids present in the food systems act as natural buffers. Presence of phosphates in food can enhance yields of MRPs. In the present study, yield of radiation-induced MRPs was enhanced by phosphate in a model system. In these studies, conditions for maximal formation of MRPs by γ-irradiation were determined. These preformed MRPs may find applications in providing chemical stability to the food system that is being investigated.

Irradiated glucose/lysine showed more UV absorbance, browning and fluorescence than did that of glucose/glycine when comparison was made at the same irradiation dose. It showed higher radical-scavenging activity in all the model systems tested, suggesting higher reactivity of lysine than of glycine. Higher reactivity of lysine in heat-induced Maillard reaction has been reported in earlier studies (Ajandouz & Puigserver, 1999; Morales & Jimenez-Perez, 2001).

Addition of preformed heat-induced MRPs to a model food system has been shown to inhibit lipid peroxidation. MRPs formed by heating protein hydrolysate and glucose inhibited rancid flavour development in cooked ground beef (Alfawaz, Smith, & Jeon, 1994). Similarly, MRPs formed by heating amino acid and glucose enhanced the stability of a methyl linoleate model system (Jayathilakan & Sharma, 2006), as well as of fluidised bed-dried mutton (Jayathilakan, Sharma, Radhakrishna, & Bawa, 2006). However, information on antioxidant potential of MRPs formed by γ-irradiation is not available. Further studies are being carried out to ascertain the antioxidant potential of MRPs formed by γ-irradiation in an actual food system.

Table 1
Effect of solvent and γ-irradiation (40 kGy) on pH, absorbance and DPPH radical-scavenging activity (DPPH RSA) of glucose/amino acid solution

	pH	A_{284}[a]	A_{420}[a]	DPPH RSA %
DW GGC	6.89	0.036	0.031	0.5 ± 0.03
DW GGI	5.43	4.60	0.070	2.2 ± 0.15
Tris GGC	7.07	0.078	0.022	0.7 ± 0.03
Tris GGI	7.02	10.60	0.87	3.6 ± 0.19
PB GGC	7.10	0.050	0.004	0.6 ± 0.03
PB GGI	7.08	14.40	1.11	12.4 ± 1.3
DW GLC	6.56	0.075	0.015	0.7 ± 0.03
DW GLI	4.93	7.20	0.11	5.1 ± 0.53
Tris GLC	7.01	0.085	0.002	0.5 ± 0.03
Tris GLI	6.98	12.60	0.39	7.8 ± 0.53
DW GLC	6.98	0.095	0.005	0.6 ± 0.03
DW GLI	6.94	18.10	1.271	31.1 ± 3.3

DW – distilled water, Tris–Tris–HCl buffer (0.1 M, pH 7.0), PB-sodium phosphate buffer (0.1 M, pH 7.0), GG – glucose/glycine, GL – glucose/lysine, C – non-irradiated, I – irradiated (40 kGy).
[a] Values after correcting for dilution factor.

4. Conclusion

Ionizing radiation, with γ rays from a Co^{60} source, was found to produce MRPs in aqueous glucose/amino acid solutions, probably due to carbonyl-amine reaction. Similarly, heat-treatment results in formation of Maillard reaction products. To the best of our knowledge, this is the first report to demonstrate antioxidant potential of compounds formed upon irradiation of glucose/amino acid solutions. Further studies are needed to elucidate the mechanism and identification of compounds formed during radiation processing of aqueous sugar/amino acid solutions. Also, efforts are needed to investigate the factors affecting the reaction and to generate information when applied to a complex food system.

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