NOTE

Radiation-Induced Sol–Gel Transition of Protein: Effects of Metal Ions on Thermal Property

Ionizing radiation influences strongly the chemical and conformational properties of biological macromolecules.[1,2] Some biological macromolecules, such as proteins, form a thermoreversible gel[3–5] and interact with metal ions, such as Cu^{2+} ions.[6] Since the thermal property changes that accompany a change of cross-links of polypeptide chains in a gel pose a problem of considerable interest, it was decided to investigate the effect of metal ions (Cu^{2+} and Fe^{2+}) on the cross-links of an irradiated protein molecule. Changes in the cross-links can be followed conveniently by measuring the setting point of irradiated protein hydrosol as a function of concentration of metal ions (Cu^{2+} and Fe^{2+}).

The solid gelatin (the Kanto Chemical Co.), as the model protein, was irradiated with ^{60}Co gamma rays in air at room temperature at dose rates of 6.0×10^4 to 1.3×10^5 rad/h. The irradiated solid gelatin was dissolved in distilled water or metal ion (Cu^{2+} as $CuSO_4$ or Fe^{2+} as $FeSO_4$) solution at about 80°C and held at 30°C for 1 h. Then, the gelatin hydrosol was cooled at a rate of 0.2°C/min and the setting point measured. The heat energy required to associate cross-links of the gelatin hydrosol was calculated using the setting point given by the equation of Eldridge and Ferry.

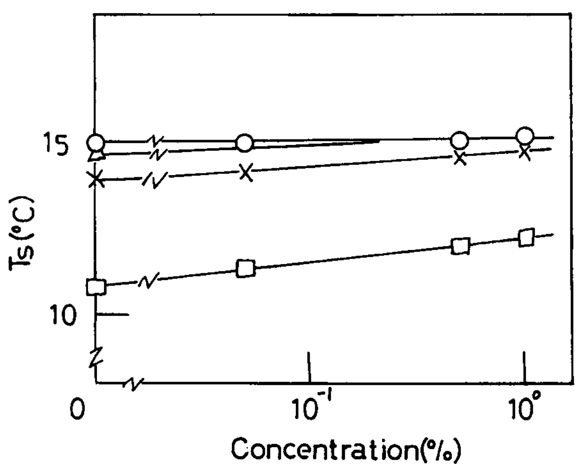

Figure 1 Setting point vs. concentration of $CuSO_4$ for various radiation doses: (○) 0 rad; (△) 10^5 rad; (**x**) 10^6 rad; (□) 10^7 rad. Concentrations: 8% gelatin hydrosol.

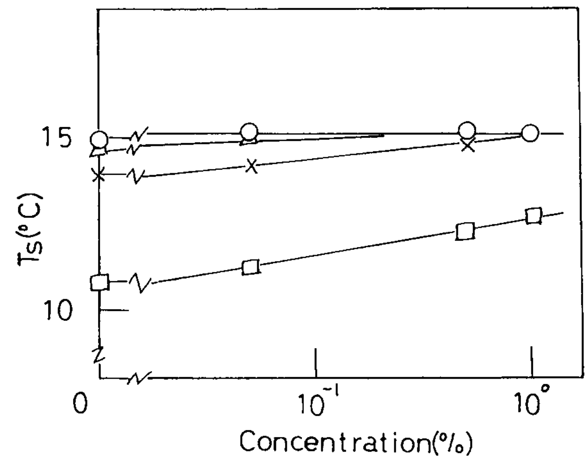

Figure 2 Setting point vs. concentration of $FeSO_4$ for various radiation doses: (○) 0 rad; (△) 10^5 rad; (×) 10^6 rad; (□) 10^7 rad. Conditions: 8% gelatin hydrosol.

The changes in setting point of irradiated gelatin with and without metal ions (Cu^{2+} and Fe^{2+}) were studied with 3–10% gelatin and by the irradiation of 0, 10^5, 10^6, and 10^7 rad. Figures 1 and 2 show some of the relations between the values of the setting point and the concentration of metal ions (Cu^{2+} and Fe^{2+}) for various radiation doses.

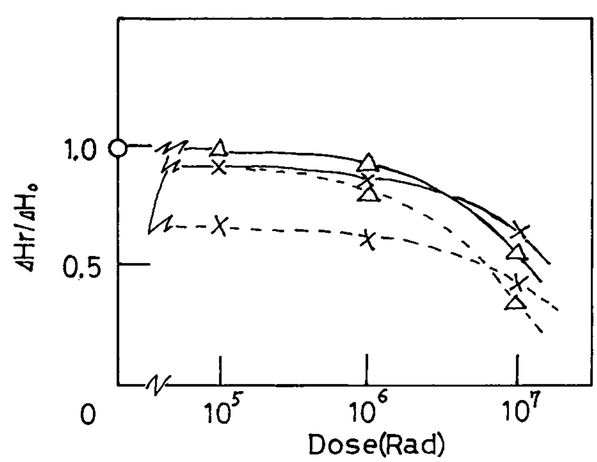

Figure 3 Heat of reaction vs. radiation dose with and without $CuSO_4$ for various cross-linking processes: (△) Gel_I; (×) Gel_{II}; (—) 0.5% $CuSO_4$; (----) no $CuSO_4$.

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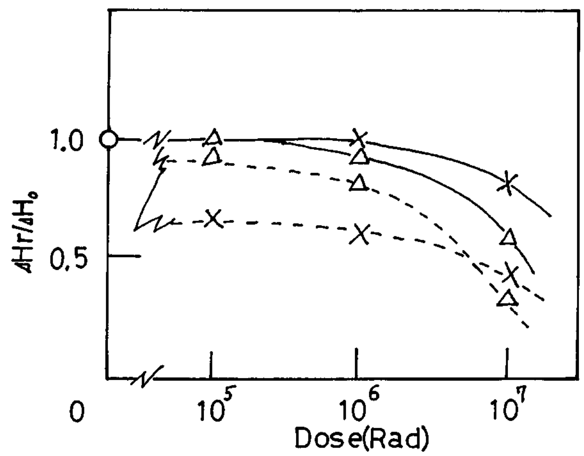

Figure 4 Heat of reaction vs. radiation dose with and without $FeSO_4$ for various cross-linking processes: (Δ) Gel_I; (×) Gel_{II}; (—) 0.5% $FeSO_4$; (----) no $FeSO_4$.

From these results, it is clear that the restoration of the T_s value is related to an increase of the cross-links of the irradiated gelatin molecule by adding the metal ions. As a result, the T_s value is recovered up to a certain extent.

Also, the changes in the heat energy required to associate cross-links of irradiated gelatin hydrosol with and without metal ions (Cu^{2+} and Fe^{2+}) were estimated by the equation of Eldridge and Ferry. If such changes in setting point and heat of reaction are compared to changes in melting point and heat of reaction, which were reported in a previous paper,[7] then the changes in the cross-linking processes of irradiated gelatin are obtained by the combination of the setting- and melting-point data, since the setting point or the melting point in a cross-linking system is considered to be the point at which a 3-dimensional network first appears or disappears, respectively. In the case of this particular gelatin sol–gel transition, the 3-dimensional networks must be entrapped in considerable amounts of water and formed by cross-links between polypeptide chains. Suppose that on cooling the gelatin hydrosol (Sol), the polypeptide chain segments in the random coil state associate in the helices to cross-link the chains in a 3-dimensional network (Gel_I) and, subsequently or simultaneously, the helices could combine into large aggregates (Gel_{II}). Then, the relations between the values of heat of reactions (of setting and of difference of melting − setting) and the radiation dose with and without metal ions are obtained for Gel_I and Gel_{II} and shown in Figures 3 and 4. With increasing the radiation dose, the heat of reaction of Gel_{II} is higher than that of Gel_I in the presence of the metal ions (Cu^{2+} and Fe^{2+}). It is understood that the effect of the metal ions on the cross-links in the cross-linking process II (Gel_{II}) is higher than that in the cross-linking process I (Gel_I).

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