Radiation-Induced Sol–Gel Transition of Protein: Effects of Radiation on Setting Property

Ionizing radiation influences strongly the chemical and conformational properties of biological macromolecules.[1,2] Some biological macromolecules, such as proteins, form a hydrogel.[3-5] Since the mechanism of the sol–gel transition of protein is a problem of general interest, it was decided to investigate the effects of radiation on the setting property of protein. Gelatin was selected as the protein molecule, since it was described in a previous paper.[6]

The changes in the setting property can be followed conveniently by measuring the setting point of the irradiated protein.

EXPERIMENTAL

Material

Gelatin used in this work was the same as that described in a previous paper.[6]

Apparatus and Procedure

In irradiation, the solid gelatin was irradiated by ^{60}Co gamma rays in air at room temperature at dose rates of 6.0×10^4–1.3×10^5 rad/h.

In thermometry, the irradiated solid gelatin was dissolved in distilled water 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 measured the setting point by the method of Kauzman et al.

In calculation of heat of reaction for crosslinking process, the heat energy required to associate crosslinks of the gelatin hydrosol was calculated by the use of the setting point given by the equation of Eldridge and Ferry:

$$\log_{10} C = \Delta H/2.303RT + \text{const} \qquad (1)$$

where C is the gelatin concentration (g/L), ΔH is the heat of reaction for the crosslinking process of the gelatin hydrosol (kcal/mol of crosslinks), R is the gas constant, and T is the setting point of the gelatin hydrosol (K). Equation (1) is converted to

$$\Delta H = (k \log_{10} C_1/C_2)(1/T_1 - 1/T_2) \qquad (2)$$

$$k = 2.303 \times R$$

RESULTS AND DISCUSSION

The changes in setting point of gelatin at various radiation doses were studied with 3–10% gelatin. Figure 1(a) shows the relation between the values of the setting point and the radiation dose. Also, the changes in heat energy required to associate crosslinks of gelatin with the irradiation were estimated by the equation of Eldridge and Ferry. Figure 2(a) shows the relation between the values of the heat of reaction and the radiation dose.

However,the changes in setting point of gelatin at different times after irradiation (3×10^6 rad) were studied with 3–10% gelatin. Figure 1(b) shows the relation between the values of setting point and the time after irradiation. Also Figure 2(b) shows the relation between the values of heat of reaction and the time after irradiation.

From these results, the setting point and the heat of reaction decreased depending upon the irradiation and, while recovered, depending upon the elapsed time after irradiation until certain values. If such changes in setting point and heat of reaction are compared to changes in melting

Journal of Applied Polymer Science, Vol. 38, 579–581 (1989)

CCC 0021-8995/89/030579-03$04.00

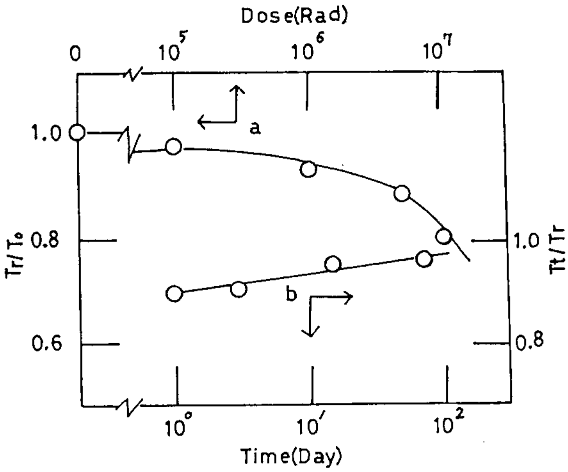

Fig. 1. (a) Setting point vs. radiation dose (10% gelatin hydrosol). (b) Setting point vs. time after irradiation (10% gelatin hydrosol and 3 × 10^6 rad).

point and heat of reaction, which were reported in a previous paper,[6] then the changes in the crosslinking processes of gelatin are obtained by the combination of the setting and melting point data, since the setting point or the melting point in a crosslinking 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 crosslinks between polypeptide chains. Suppose that on cooling gelatin hydrosol (sol) the polypeptide chain segments in the random coil state associate in the helices to crosslink the chains in a 3-dimensional network (gel_{I}) and, subsequently or simultaneously, the helices could combine into large aggregate (gel_{II}). Then the relations between the values of heat of reactions (of melting, setting, and its difference) and the radiation dose are obtained for $\text{gel}_{\text{I+II}}$, gel_{I}, and gel_{II}, and are shown in Figure 3. Figure 4 shows the relations between the values of heat of reactions (of melting, setting, and its difference) and the time after irradiation for $\text{gel}_{\text{I+II}}$, gel_{I}, and gel_{II}. With increasing the radiation dose or the elapsed time after irradiation, the heat of reaction of gel_{II} is lower or higher than that of gel_{I}, respectively. It is understood that radiation resistance or radiation recovery of crosslinks in the crosslinking process II (gel_{II}) is lower or higher than that in the crosslinking process I (gel_{I}), respectively.

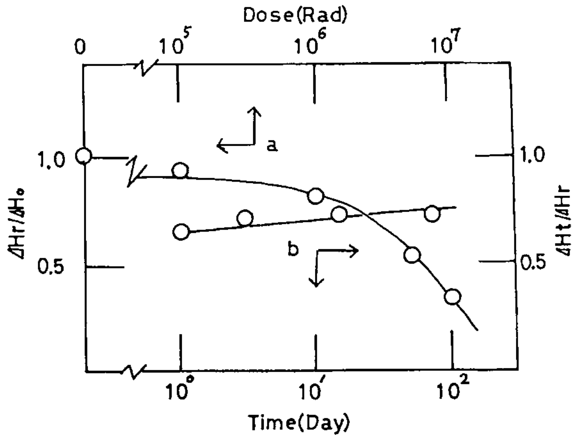

Fig. 2. (a) Heat of reaction vs. radiation dose. (b) Heat of reaction vs. time after irradiation (3 × 10^6 rad).

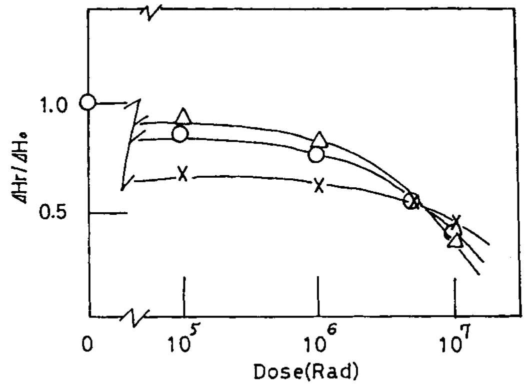

Fig. 3. Heat of reaction vs. radiation dose for various crosslinking processes: (○) gel_{I+II}; (△) gel_I; and (×) gel_{II}.

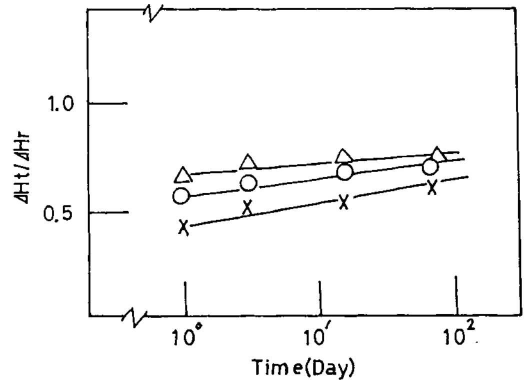

Fig. 4. Heat of reaction vs. time after irradiation (3×10^6 rad) for various crosslinking processes: (○) gel_{I+II}; (△) gel_I; (×) gel_{II}.

The author wishes to thank Dr. T. Hirano of Tokyo University of Fisheries for many suggestions.

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Received April 21, 1988
Accepted July 20, 1988