Note

Improvement of Gelling Properties of Ovalbumin by Heating in Dry State

Naotoshi MATSUDOMI, Youko ISHIMURA
and Akio KATO

Department of Agricultural Chemistry,
Yamaguchi University,
Yamaguchi 753, Japan

Received September 12, 1990

Heat-induced gelation of egg white is one of its important functional properties. Kato *et al.*^{1,2)} have reported that when spray-dried egg white was heated in a controlled dry state at 80°C for various periods of time, the heat-induced gels greatly increased in firmness. As ovalbumin is the main constituent of egg white, the formation of a firm gel made from egg white heated in the dry state might be due to some conformational changes of ovalbumin. Ovalbumin gives a transparent solution, transparent gel, turbid gel, or turbid suspension, depending on the pH and ionic strength of the heating medium.^{3–6)} We examined the gelling properties of ovalbumin heated at various pHs and NaCl concentrations to identify the effects of prior dry heat treatment on the gelling behavior, particularly the turbidity and hardness of the gels.

Ovalbumin was prepared and purified by the method of Kekwich and Cannan.⁷⁾ Dry heat treatment at 80°C for varying times was done by placing 0.5 g of the freeze-dried ovalbumin (5.65% moisture content) in 50-ml serum bottles and sealing it with a rubber stopper, and then removed at specified times (days). The heat-treated samples were then stored at 4°C pending analysis.

Measurements of the hardness and turbidity of gels were done as follows⁸⁾: the proteins (the untreated and dry heat-treated ovalbumin) were dissolved in 20 mM NaCl solution at a protein concentration of 8% (w/v) and the pH was adjusted by adding small amounts of either NaOH or HCl. One-ml samples of the protein solution were placed in Sigmacoated tubes (6.0 mm in diameter) and centrifuged to remove any air bubbles. The tubes were sealed, heated at 80°C for 30 min, and stored for 15 hr at 4°C. After tempering at 25°C for 30 min, the gels were removed from the tubes without disrupting the surface of the gels. Gel hardness was measured on the gel sections (6.0 mm diameter × 5.0 mm height) with a tensile tester (Tensilon UTM-II, Toyo Baldwin Co.) and calculated from the force peak (grams) when compressed up to 2.0 mm from its original height. The turbidity of gel sections was measured by their optical densities at 440 nm on a densitometer

(Densitol DMU-33, Toyo Kagaku Co.) against the cutting surface (5-mm light-path lengths) of gel sections. The hardness and turbidity are the average values of three experiments.

The effects of prior dry heat treatment at 80°C for varying times on the hardness of heat-induced gels of a 8% ovalbumin solution (pH 6.5) in 20 mM NaCl is shown in Fig. 1. The hardness of the gels was greatly increased within the duration of storage treatment at 80°C for 5 days and decreased after 7 days because of a slight loss of solubility. The turbidity of ovalbumin gels drastically decreased after 3 days of heating in the dry state and transparent gels were formed. However, the transparency of gels made from ovalbumin heated in the dry state over 7 days was slightly lowered by a slight loss of solubility

Fig. 1. Effects of Storage Time at 80°C in the Dry State of Ovalbumin on the Gel Hardness.

Fig. 2. Effects of pH on the Hardness and Turbidity of Ovalbumin Gels.

○, control; ●, dry-heated; —, hardness; ---, turbidity.

Fig. 3. Effects of NaCl Concentration on the Hardness and Turbidity of Ovalbumin Gels Made at pH 3.0 (A) and 6.5 (B).

○, control; ●, dry-heated; —, hardness; ---, turbidity.

(see photograph in Fig. 1). Therefore, the sample stored at 80°C for 5 days was used in the following experiments.

The pH profiles of the turbidity and hardness of ovalbumin gels are shown in Fig. 2. The untreated ovalbumin formed turbid gels between pH 3.5 and 6.5, and below pH 3.0 and above pH 7.0 the gels were transparent. This result was consistent with the result reported by Hatta *et al.*⁵⁾ The pH profile of turbidity of the dry heat-treated ovalbumin gels was different from that of gels made from untreated ovalbumin; the degree of the turbidity was much lower between pH 6.0 and 7.0, and above pH 6.5 transparent gels were obtained. The pH profile of hardness of untreated ovalbumin gels was bimodal with a maximum gel strength at both sides (at pH 3.0 and 6.5) of the isoelectric point (pI 4.6), in essential conformity with the results reported elsewhere.^{4,5,9)} The hardness of the dry heat-treated ovalbumin gels greatly increased at all pHs and had higher value at pH 6.5 than at pH 3.0, compared with that of untreated ovalbumin gels. Thus, although the hardness and turbidity of both ovalbumins gels varied depending on the pH in the presence of 20 mM NaCl, the dry heat-treated ovalbumin could form firmer and transparent gels at the alkaline side of the isoelectric point, compared with the untreated ovalbumin.

The effects of NaCl concentration on the hardness and turbidity of gels prepared at pH 3.0 and 6.5 was examined (Fig. 3). At pH 3.0 (Fig. 3A), both ovalbumins formed turbid gels with an increase of NaCl concentration, and the degree of the turbidity of gels made from the treated ovalbumin was higher. The hardness of gels from both ovalbumins depended on the NaCl concentration and gave maximum values at 40 mM NaCl, and the hardness of the treated ovalbumin gels had much higher values than that of untreated ovalbumin gels. At pH 6.5 (Fig. 3B), in the presence of 40 mM NaCl the untreated ovalbumin formed an opaque gel, while the gel made from treated ovalbumin

remained transparent. This shows that some conformational changes in the ovalbumin occurred during heating in the dry state lowered the sensitivity to NaCl. The maximal hardness of gels from both ovalbumins was obtained at 40 mM NaCl, and the treated ovalbumin formed firmer gels, compared with untreated ovalbumin.

Thus, it was found that transparent and firm ovalbumin gels could be obtained within wider ranges of pH and ionic strength by heating in the dry state, particularly at the alkaline side of the isoelectric point. These results indicate that the controlled heating in the dry state was an effective method to improve the gelling properties of ovalbumin, particularly the transparency and hardness of the gel.

Hatta *et al.*⁵⁾ and Kitabatake *et al.*⁶⁾ have reported that the protein network and coagulum give the rigidity and turbidity, respectively, of the heat-induced ovalbumin gel. They have also suggested that a turbid gel seems to comprise a network structure and coagulum, and that a transparent gel seems to comprise a protein network but does not contain coagulum. In addition, Kitabatake *et al.*⁶⁾ have reported that a transparent gel from ovalbumin having high sensitivity to NaCl during heating could be prepared even in a high NaCl concentration using the two-step heating method, and that the transparent gel seemed to be formed by hydrophobic interaction among the soluble linear aggregates made by the first salt-free heating. In consideration of their reported results,^{5,6)} a possible elucidation is that the ovalbumin molecules partially unfolded by the dry heat treatment may gain a suitable balance between attractive and repulsive forces and facilitate the formation of the soluble linear aggregates to form a gel network during heating, resulting in the formation of the transparent, firm ovalbumin gels. The effectiveness of the dry-heating method is that the firmer gel of ovalbumin can easily be prepared from the same amounts of the protein within the ranges of pH 3 to 8. The sensitivity to NaCl of ovalbumin was slightly lowered by heating in the dry state, however, a transparent gel of ovalbumin could not be made in the presence of NaCl concentrations above 100 mM, differing from transparent ovalbumin gels formed by the two-step heating method of Kitabatake *et al.*⁶⁾

Further experiments are required to elucidate such gelling properties of ovalbumin heated in the dry state.

References

- 1) A. Kato, H. R. Ibrahim, H. Watanabe, K. Honma and K. Kobayashi, *J. Agric. Food Chem.*, **37**, 433 (1989).
- 2) A. Kato, H. R. Ibrahim, H. Watanabe, K. Honma and K. Kobayashi, *J. Agric. Food Chem.*, **38**, 32 (1990).
- 3) P.-O. Hegg, *J. Food Sci.*, **47**, 1241 (1982).
- 4) B. Egeland, *J. Food Sci.*, **45**, 570 (1980).
- 5) H. Hatta, N. Kitabatake and E. Doi, *Agric. Biol. Chem.*, **50**, 2083 (1986).

- 6) N. Kitabatake, H. Hatta and E. Doi, *Agric. Biol. Chem.*, **51**, 771 (1987).
 - 7) R. A. Kekwich and R. K. Cannan, *Biochem. J.*, **30**, 227 (1936).
 - 8) N. Matsudomi, D. Rector and J. E. Kinsella, *Food Chem.*, (submitted).
 - 9) R. Nakamura, T. Fukano and M. Taniguchi, *J. Food Sci.*, **47**, 1449 (1982).
-