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# Effect of Dry-Heated Egg White on Wheat Starch Gel and Gluten Dough

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**Characterization of the mechanisms of improvement of Chinese noodles by dry-heated egg white, were estimated by rheological and spectroscopic data of the effects of dry-heated egg white on wheat starch gel and gluten dough. The dry-heated egg white significantly increased breaking stress and elasticity of the starch gel but had little effect on retrogradation of the gel and distribution of proteins in the gel. On the other hand, the dry-heated egg white had a negligible effect on creep curves and IR spectra of gluten dough. From these data, improvement of Chinese noodles by the dry-heated egg white is believed to be due to interaction between the dry-heated egg white proteins and gelatinized starch molecules rather than gluten proteins. However, the effect of the dry-heated egg white on starch gel was unexpectedly small compared to the effect on the noodles.**

Keywords: dry-heated egg white, wheat starch, wheat gluten, gelatinization

## Introduction

Egg whites are widely used in food processing because of their excellent properties of emulsifying, foaming and gelling. Further, controlled heating in a dry state (for example, 80°C for 5–10 days) was reported to improve the functionalities of egg white without any loss in solubility (Kato *et al.*, 1989, 1990a, 1990b, Mine, 1996, 1997). Xu *et al.* (1998) reported the potential value of dry-heating, which is a heating process of food materials, especially egg white, at over 100°C for several hours with very low water content. The dry-heating allowed egg white powder to be soluble in water during reheating of solution of the dry-heated egg white. They also showed an inhibitory effect on heat-induced coagulation of fresh egg white to improve the pasteurization of egg white. Watanabe *et al.* (1999) reported that the dry-heated egg white inhibits heat aggregation and coagulation of ovotransferrin and lysozyme in freeze-dried egg white resulting in a transparent solution after reheating (60°C, 5 min). Further study on dry-heating showed that ovalbumin formed soluble aggregates and the aggregates inhibited coagulation of ovotransferrin (Watanabe *et al.*, 2000). Features of the dry-heating recognized by Xu *et al.* (1998) and Watanabe *et al.* (1999 and 2000) were that the dry-heating allowed most water to evaporate from protein powder over 100°C at the first stage and proteins were thermally denatured under very low water content.

As for other properties, Tachi *et al.* (2004) reported an improvement in Chinese noodles (Chukamen in Japanese), made from wheat flour and alkaline solution (Kansui), by the addition of dry-heated egg white. The dry-heated egg white increased breaking stress, instantaneous elasticity and tensile strain of the noodles, resulting in greater acceptability. Spray-dried egg white powder has long been added to wheat flour to improve Chinese noodles. However, sprayed egg white has a problem in that its addition made the noodles shorter as well as firmer than noodles to which there had been no such addition. Addition of the dry-heated egg white instead of the spray-dried powder significantly improved the Chinese noodles by sensory evaluation as well as rheological properties. We therefore wanted to clarify the mechanism of improvement of the noodles by the dry-heated egg white. In this study, we evaluated the effects of the dry-heated egg white on wheat starch gel and gluten dough and discussed interactions between egg white protein and starch or gluten.

## Materials and Methods

*Materials* Commercially available wheat starch (special grade, NCA Ryori no Moto K.K., Japan) and gluten (A-Glu SS, Glico Foods Co., Ltd., Japan) were used for this study.

*Dry-heating of egg white powder* Dry-heated egg white powder was prepared according to previous paper (Xu *et al.*, 1998). Namely, the spray-dried egg white powder was dissolved in distilled water and lyophilized. Dried egg white powder was spread thinly on paper and heated at 120°C for 6 h uncovered in an oven. The dry-heated egg white was then put in a desiccator to cool.

*Preparation of gluten dough and starch gel* Wheat starch powder (3 g) and egg white powder (0.12 g; 4% to starch) was mixed with water (7 g) and put into a casing tube (12 mm i.d.). Both edges of the tube were fastened and the tube was put in a water bath and first partially gelatinized at 60°C for 3 min and then completely gelatinized at 100°C for 30 min (Amano *et al.*, 1995). The starch gel was sliced into pieces 10 mm height.

Wheat gluten powder (2.88 g), egg white powder (spray-

Abbreviations: FT-IR, Fourier transform infrared; NMR, nuclear magnetic resonance
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dried or dry-heated; 0.12 g) and 2 ml of water or alkaline solution (kansui; 2.5 g of $K_2HPO_4$, 1.7 g of $Na_2HPO_4$ in 100 ml of water) was mixed and stirred for 1 min with a glass rod to furnish raw gluten dough. The dough was put into a casing tube and both edges tightly fastened and heated in boiling water for 5 min.

*Rheological measurements* Rheoner RE-3305 (Yamaden Co., Ltd., Tokyo) was used for measurements of breaking strength and creep behavior of starch gel and gluten dough containing the dry-heated egg white. Stress – strain curves were obtained under conditions as follows: sample diameter, 12 mm; compressive speed, 1.0 mm/s; and deformation ratio, 95%. Creep behavior under compression was also analyzed as follows: sample diameter, 12 mm. Initial speed of the sample plate was set at 1.0 mm/s. Elevation of the plate was continuously adjusted to produce a constant stress of 60 g-force by the creep meter (5200 Pa), and the displacement of the gel after the initial stress (60 s) was recorded. The data were stored in a diskette and analyzed using software (CA-3305-16) designed for creep analysis (Yamaden Co., Ltd.).

*Fourier transform infrared (FT-IR) spectra* IR spectra were recorded with a System 2000 FT-IR spectrometer (Perkin Elmer, Inc.) by Horizontal ATR method.

*$^{13}C$ nuclear magnetic resonance (NMR) spectra* $^{13}C$ NMR spectra were recorded on a Unity Inova 400 spectrometer ($^{13}C$ at 100 MHz; Varian, Inc.). Relative peak height of each signal of glucose residue to signals of pyridine (an internal standard) was used to characterize the retrogradation. The peak height derived from the starch gel immediately after preparation was set to 1 and decrease of the relative peak intensity indicated retrogradation of the starch gel.

*Distribution of proteins in starch gel* Starch gel was fixed with 3.7% formaldehyde solution (pH 7) for 24 h, and then sliced by a microtome (0.15 mm thickness). Each piece of sliced disk was dyed by acrolein Schiff reaction and applied to a BHS optical microscope (Olympus Optical Co., Ltd.).

## Results and Discussion

*Effect of the dry-heated egg white on the starch gel* The dry-heated egg white effectively increased breaking stress, instantaneous elasticity, tensile strain and sensory properties of Chinese noodles compared to the spray-dried egg white, as well as the noodles containing no egg white (Tachi *et al.*, 2004). In order to clarify the mechanism, we evaluated the effect of the dry-heated egg white on starch gel. Stress – strain curves of the starch gel containing two kinds of egg white are shown in Fig. 1. Breaking stress of the starch gel was increased by addition of the spray-dried egg white powder, and further increment in the stress was observed by addition of the dry-heated egg white powder. The dry-heated egg white showed more effective enhancement of the breaking stress, and these data coincided with the previous result (Tachi *et al.*, 2004).

Next, the starch gel was applied to a creep experiment (Fig. 2). Creep compliance of the starch gels with the spray-dried egg white was less than that without the egg white. Further, the dry-heated egg white decreased the compliance compared with the spray-dried egg white. These data showed the dry-heated egg white enhanced gel strength of the starch gel more than the spray-dried, similar to the above data on breaking stress. From these curves, viscoelastic moduli were calculated using the software (CA-3305-16, Yamaden Co., Ltd.) according to a Voigt-type model (Fig. 2 inset). The starch gel containing the spray-dried egg white was well demonstrated by the Voigt-type four-element model. However, the gel containing no egg white and the dry-heated egg white was calculated by the six-element model because the four-element model demonstrated the gel insufficiently. As a result, the modulus of instantaneous elasticity ($E_0$) of the starch gel containing the dry-heated egg white was significantly higher than other samples (Fig. 3 and Table 1). However, there was no significant difference in moduli of retarded elasticity. On the other hand, the moduli of viscosity in steady state ($\eta_0$) and retarded state ($\eta_1$) varied widely and had no significant differences, so no significant effect was detected (Table 1). Hardness and elasticity of the starch gel increased by addition of the dry-heated egg white compared with the spray-dry egg white as well as that with no addition. In previous papers (Xu *et al.*, 1998, Watanabe *et al*, 1999 and 2000), ovalbumin molecules were reported to form soluble aggregates during the dry-heating process, and the

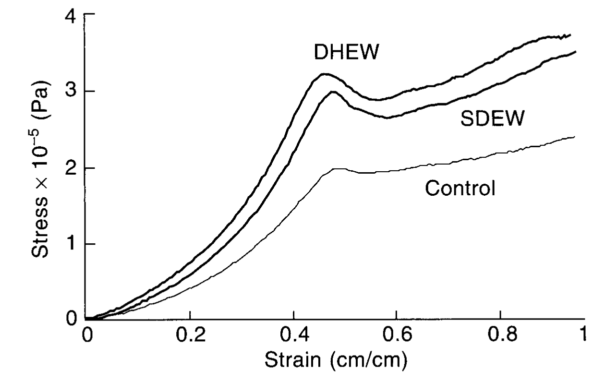


**Fig. 1.** Stress – strain curves of starch gel containing dry-heated egg white. Control, No addition of egg white; SDEW, addition of spray-dried egg white; DHEW, addition of dry-heated egg white.

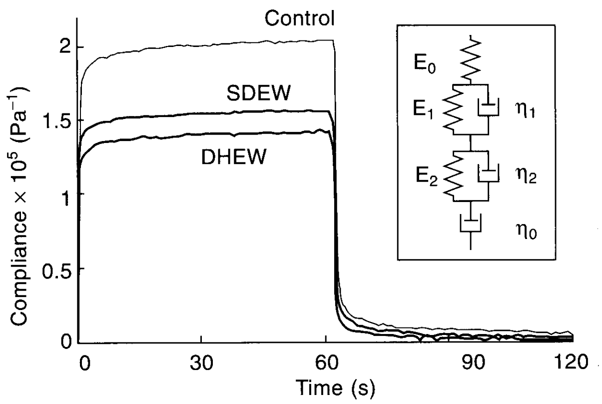


**Fig. 2.** Creep curves of starch gel containing dry-heated egg white. Abbreviations, see Fig. 1. Inset, Voigt type six-element model.

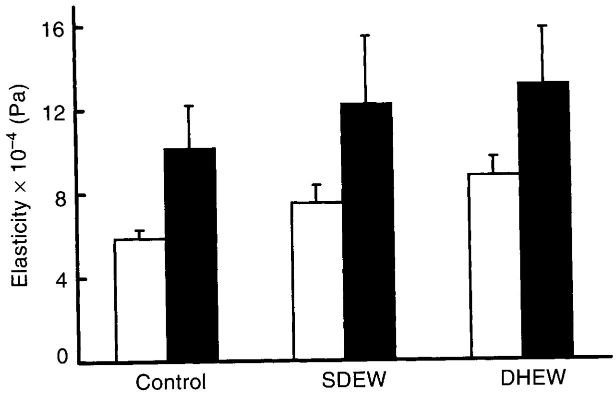


**Fig. 3.** Elasticity of starch gel containing dry-heated egg white and effect of refrigeration. Abbreviations, see Fig. 1. Open square, Immediately after preparation; Hatched square, After 1 day of refrigeration.

resulting aggregates were believed to interact with ovotransferrin to inhibit formation of insoluble aggregates of ovotransferrin under thermal denaturation and to interact with fresh egg white proteins to form a more rigid gel structure during re-heating. Considering these reports, the soluble aggregate of ovalbumin in the dry-heated egg white is considered to be more effective for interactions with starch gel matrix, possibly because of hydrogen bonding and other interactions. These improved interactions may allow the gel network to be dense and tight. Further, the more rigid structure of protein aggregate compared with the starch gel matrix is probably responsible for high elasiticity of the starch gel containing the dry-heated egg white.

Then, we evaluated the effect on retrogradation of the starch gel. Creep compliance was measured, because retrogradation generally allowed the starch gel to be harder (Fig. 3). After 1 day of refrigeration at 4°C, the starch gel was applied to a creep experiment. Before refrigeration, the elasticity of the dry-heated egg white added gel was significantly higher than both the control and the spray-dried egg white added starch gel. After refrigeration, the elasticity of each sample increased, but the differences between before and after refrigeration were reduced by addition of the dry-heated egg white and there was no significant difference among 3 kinds of gels despite the egg white addition. Increments of moduli of the elasticity of each starch gel (no addition, spray-dried egg white and dry-heated egg white) from 0 day to 1 day were calculated to be 1.73, 1.63 and 1.49, respectively. The dry-heated egg white might have some suppressive effect on increase of the elasticity by retrogradation of the starch gel during refrigeration but there was no clear evidence of retrogradation of the gel containing the dry-heated egg white.

To estimate the decrease in gelatinization degree of starch molecules by retrogradation, the starch gel was applied to $^{13}$C-NMR measurements. Kainosho and Ajisaka (1978) reported that glucose residues in starch showed narrow $^{13}$C signals by gelatinization. This narrowing of the signals is believed to be caused by higher mobility of glucose residues in gelatinized starch molecules through the breaking of inter-residual hydrogen bondings by thermal energy. Fuke and Matsuoka (1984) evaluated the retrogradation degree of kiwi fruit starch by measuring signal intensities of glucose carbons in starch molecules. Partial recrystallization of starch molecule during retrogradation limited the mobility of glucose residues and allowed only low signal intensities. As for signals assigned to glucose residue in the starch gel, ratios of signal intensities before and after 1 day of refrigeration were calculated on the starch gel, the spray dried egg white added and the dry-heated egg white added gels (Fig. 4), where the intensity immediately after preparation of the gel was set to 1. The starch gel (no addition of egg white) showed about 0.68 of relative intensity, while starch gels containing the spray-dried egg white or the dry-heated egg white was 0.86. Addition of the egg white to the starch gel effectively suppressed the decrease of signal intensity, but there was no significant difference between the spray-dried and the dry-heated egg white. Thus, these results showed no significant differences between the spray-dried and the dry-heated egg whites on the elasticities and $^{13}$C NMR signal intensities of the refrigerated gels.

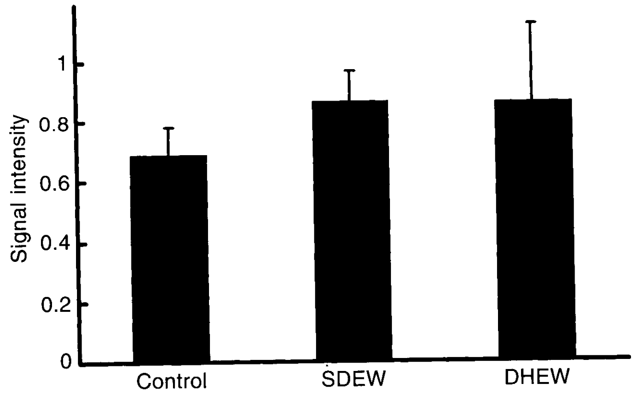


**Fig. 4.** Signal intensity of $^{13}$C-NMR spectra of starch gel containing dry-heated egg white after refrigeration. Abbreviations, see Fig. 1. Each signal intensity was relatively shown against the intensity measured immediately after preparation of the gel.

**Table 1.** Creep parameters of starch gel containing dry-heated egg white.

| | $E_0 \times 10^{-4}$ (Pa) | $E_1 \times 10^{-5}$ (Pa) | $E_2 \times 10^{-6}$ (Pa) | $\eta_1 \times 10^{-6}$ (Pa · s) | $\eta_2 \times 10^{-6}$ (Pa · s) | $\eta_0 \times 10^{-8}$ (Pa · s) |
|---|---|---|---|---|---|---|
| None | 5.84 ± 0.45 | 8.27 ± 1.38 | 1.11 ± 0.17 | 5.70 ± 1.82 | 1.63 ± 0.64 | 1.07 ± 0.17 |
| SDEW | 7.49 ± 0.90 | 8.74 ± 1.98 | – | 2.07 ± 0.66 | – | 1.60 ± 0.70 |
| DHEW | 8.82 ± 0.86 | 8.60 ± 1.74 | 1.70 ± 0.16 | 3.60 ± 1.89 | 9.07 ± 4.22 | 1.26 ± 0.21 |

None, noodle with no addition of egg white
SDEW, spray dried egg white
DHEW, dry-heated egg white

Both of the spray-dried and dry-heated egg whites were shown to suppress retrogradation of starch gel effectively, but there were no significant differences between them. Egg white proteins were believed to interact with glucose residues in starch gel matrix after partial breaking of inter-residual hydrogen bondings by thermal treatment and to inhibit recrystallization of gelatinized starch molecules by suppressing reconstitution of inter-residual hydrogen bondings during refrigeration of the starch gel. But this effect of protein is likely to be independent of the dispersion state of proteins such as the molecular mass of protein aggregates.

We characterized distribution of egg white protein in the starch gel and compared the spray-dried and the dry-heated egg white. Sections of starch gel showed granules of the protein and there were no significant differences in protein distribution (Fig. 5). These data may imply that effect of the dry-heated egg white on the starch gel was not caused by the distribution of egg white protein molecules in the gel, but by interaction between egg white proteins and starch molecules.

*Effect of the dry-heated egg white on gluten dough* Then, in order to identify the interaction between the dry-heated egg white and gluten, the effect of the former on gluten dough was evaluated. Creep behavior of the dough containing the dry-heated egg white was compared with the dough containing the spray-dried egg white (Fig. 6). There were no differences between these two samples, and there were also no differences even if the gluten was dispersed in alkaline solution (Kansui) instead of water (data not shown).

In order to evaluate the effect on secondary structure of gluten protein, the gluten dough containing the dry-heated egg white was subjected to FT-IR spectra and the deconvoluted spectra obtained are shown in Fig. 7. Absorbing peaks ranging from ca. 1700 to 1580 $cm^{-1}$ (the amide band I) and from 1580 to 1450 $cm^{-1}$ (the amide band II) reflect the secondary structures (e.g., α-helix, β-sheet and random structures) (Surewicz *et al.*, 1993). Addition of egg white allowed the intensity at ca1630 $cm^{-1}$, which is assigned to the β-sheet structure (Surewicz *et al.*, 1993) to become rel-

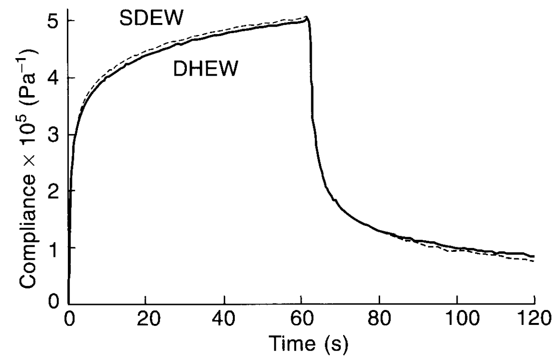


**Fig. 6.** Creep curves of gluten dough containing dry-heated egg white. Fine line, spray-dried egg white; Bold line, dry-heated egg white.

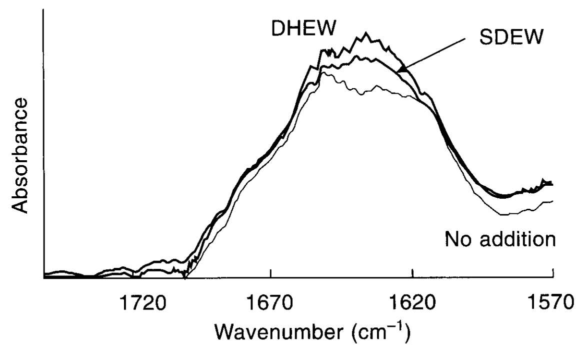


**Fig. 7.** Deconvoluted FT-IR spectra of gluten dough containing spray-dried or dry-heated egg white. Abbreviations, see Fig. 1.

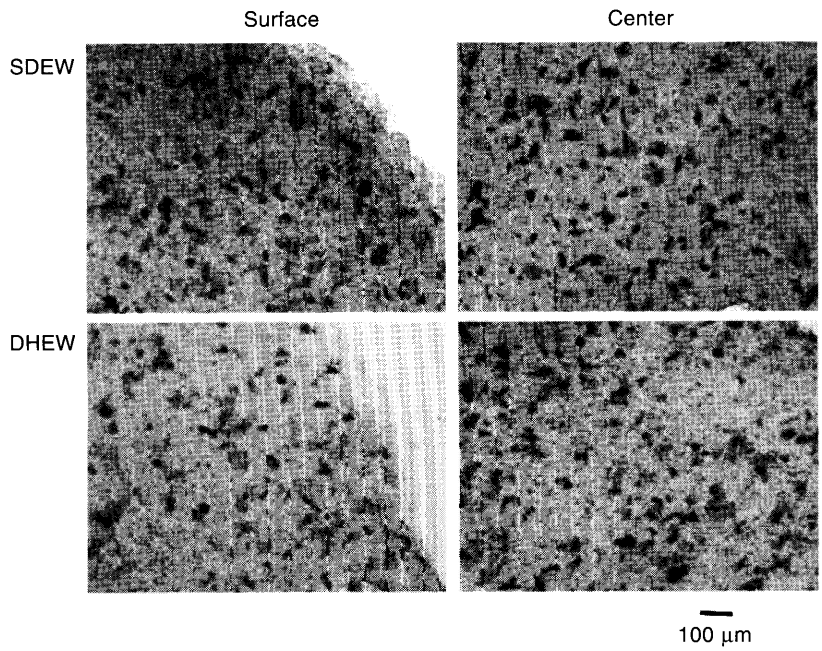


**Fig. 5.** Distribution of proteins in starch gel containing dry-heated egg white. Abbreviations, see Fig. 1.

atively larger. These changes are thought to be responsible for differences in structures between gluten and egg white proteins. Comparing the spray-dried and dry-heated egg whites, the deconvoluted spectra had almost equal patterns with only a slight difference in peak top position of amide band I of each sample, namely 1639 $cm^{-1}$ (spray-dry egg white) and 1936 $cm^{-1}$ (dry-heated egg white). So these data may show that both egg whites have almost the same effects on the gluten protein structures.

There may be no differences in interactions between egg white and wheat proteins, even if we use the spray-dried or the dry-heated egg whites. These data suggest that the dry-heated egg white protein interact with starch molecules and/or the starch gel network more than wheat proteins, and there were some differences in interactions between the spray-dried and the dry-heated egg whites. However, Tachi *et al.* (2004) reported much greater improvement of rheological and sensory properties of Chinese noodle. These data seem to show that the dry-heated egg white proteins affect the interaction between starch and gluten and the effect brings about an improvement in the rheological property of the noodles.

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