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Molecular characteristics of egg white protein-dextran conjugates

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Abstract

Egg white protein-dextran conjugates were prepared by using the Maillard reaction, and gelling properties, sulfhydryl content, hydrophobicity and molecular flexibility of the conjugates were determined. The results showed that gel hardness and water holding capacity of egg white proteins increased by 112.51% and 18.89%, respectively, after glycosylation. During the Maillard reaction, the conjugate molecules partially unfolded, which made hydrophobic groups in the molecules exposed. On the other hand, disulfide bonds formed between the molecules or in the molecules, so the total sulfhydryl content decreased.

Key words: Egg white powder, Maillard reaction, gelling properties, hydrophobicity, molecular flexibility.

Introduction

Egg white powder is an ideal substitute of egg white liquid for its long shelf life, safety, convenient to transportation and storage, etc., because its functional properties such as gelling properties, foaming ability, emulsification, etc., it is widely used in food industry [1, 2]. In order to improve gelling properties of egg white powder, previous scholars used a variety of methods, and in these methods the Maillard reaction was proved to be a safe and effective method [3-7].

Most previous studies have been carried out on the physicochemical properties of egg white protein-dextran conjugates. However, much less work has been reported on the molecular properties of egg white protein-dextran conjugates. In this paper, egg white protein-dextran conjugates were prepared by using the Maillard reaction, and gelling properties, sulfhydryl content, hydrophobicity and molecular flexibility were determined.

Materials and Methods

Egg white powder and dextran were purchased from Dalian Han Wei food limited company, and other reagents were purchased from Sinopharm Chemical Reagent Co., ltd..

The equipments included SHD type constant temperature and humidity test chamber (Shanghai is one equipment factory); FDU-1100 freeze dryer (EYELA); PHS-3C acidity meter (Shanghai Precision Instrument Company Limited); TA-XTplus2 texture analyzer (SMS company, UK); LD4-2A centrifuge (Beijing medical centrifuge plant); F-4500 fluorescence spectrophotometer (Japan Hitachi).

Egg white protein-dextran conjugates were prepared and gelling properties were determined by the method of Yu and Chi [3]. Sulfhydryl content, hydrophobicity and molecular flexibility were determined according to the methods of Plancken [8], Nakai and Li-Chan [9] and Meste *et al*. [10].

Statistical analysis: Each experiment was performed in triplicate. Collected data were expressed as mean ± SD. ANOVA was used to determine the significance of differences by SPSS PASW Statistics 18.0 (SPSS Inc, Chicago, IL, USA). The upper level of significance was set at $p<0.05$ [11].

Results and Discussion

Gelling properties of egg white protein-dextran conjugates is shown in Fig. 1. Both gel hardness and water holding capacity changed significantly, gel hardness increased by 112.51% and water holding capacity by 18.89% in the first 4 days and dropped by the end of the reaction time.

Disulfide bond is one important force to maintain protein structure, so the variation of sulfhydryl content can reveal protein extensibility. Sulfhydryl content of egg white protein-dextran conjugates is shown in Fig. 2. With the prolongation of time, surface sulfhydryl content increased, while the total sulfhydryl content decreased, which illustrated that the change of egg white protein-dextran conjugates conformation in the incubation included that sulfhydryl in the interior exposed because partially protein molecules unfolding and disulfide bonds formed intermolecular and intramolecular made the total sulfhydryl content decrease.

Hydrophobicity is an important factor to influence physicochemical and functional properties of protein, and the surface hydrophobicity reflected hydrophobic amino acid number on the protein surface. Because hydrophobicity can reveal protein conformation changes, it was used as an important parameter to investigate protein structure [12]. In this study, ANS method was used to determinate the protein surface hydrophobicity, and the result is shown in Fig. 3.

During the formation of protein gel, hydrophobicity is an important force to maintain gel structure, and surface

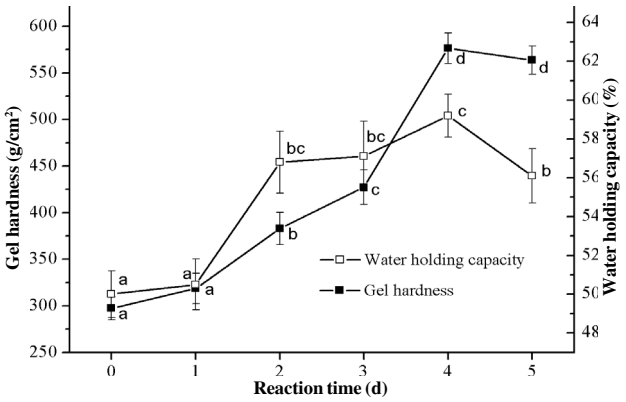

Figure 1. Gel hardness and water holding capacity of egg white protein-dextran conjugates. Error bars are standard deviations of mean values. Different values (a-d) within the same line mean differ significantly (p<0.05).

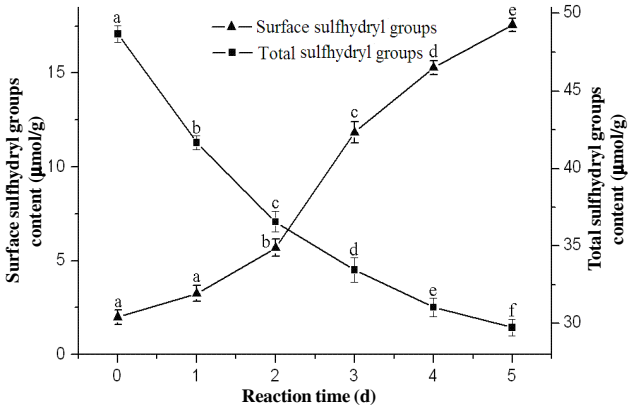

Figure 2. Change of sulfhydryl content of egg white protein-dextran conjugates. Error bars are standard deviations of mean values. Different values (a-f) within the same line mean differ significantly (p<0.05).

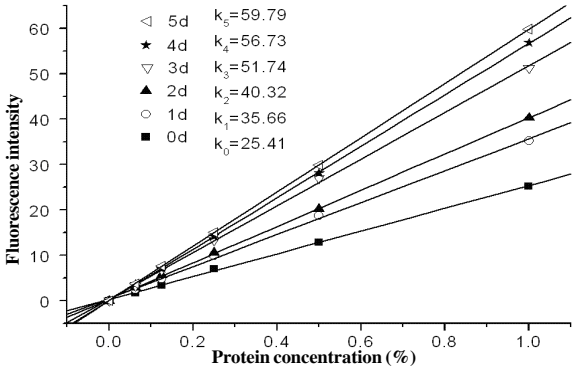

Figure 3. Change in hydrophobicity of egg white protein-dextran conjugates. Error bars are standard deviations of mean values. Different values (a-f) within the same line mean differ significantly (p<0.05).

hydrophobicity is closely related to molecular hydrophobic interaction. Fig. 3 showed that with protein concentration increasing, fluorescence intensity increased linearly, it can reflect hydrophobicity of the conjugates. Protein hydrophobicity increased with reaction time prolonging, and this variation was similar with gel hardness, it suggested that hydrophobicity is one important factor to influence gel hardness. According to view of Townsen and Nakai [13], ANS connected to the aromatic amino acids such as phenylalanine, tyrosine and tryptophan, so fluorescence intensity was directly related to protein hydrophobicity. Most hydrophobic groups of egg white protein distributed in the molecular structure [11], it indicated that protein hydrophobic groups in the internal of natural egg white protein exposed and the protein structure extended during the reaction.

Flexibility is one indicator to investigate protein molecules activity and structure, it is closely related to protein intramolecular hydrophobicity and protein shapes. During the reaction, protein molecular structure changed constantly, so the conjugates space extensibility changed. This research determined the flexibility of egg white protein-dextran conjugates and the result is shown in Fig. 4.

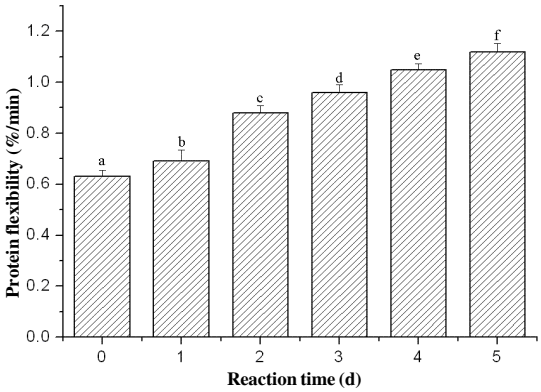

Figure 4. Change of egg white protein-dextran conjugates flexibility. Error bars are standard deviations of mean values. Different values (a-f) within the same line mean differ significantly (p<0.05).

Flexibility reflected proteins extensibility, and it is closely related to protein functional properties. Fig. 4 shows that with treatment time prolonging, protein flexibility increased ceaselessly, but there was no significantly difference (p>0.05) between sample (0 d) and sample (1d). This may be because Maillard reaction reduced the number of lysine, and protein conformation extending made activity center of trypsin easier to connect with the protein hydrolysis position, so the flexibility of samples (0d and 1d) had no significant differences, while samples (2-5d) were more accessible for enzymatic hydrolysis and the protein flexibility increased.

Conclusions

(1) During the Maillard reaction, partially protein molecules unfolding made sulfhydryl in the interior of egg white protein-dextran conjugates expose, and disulfide bonds forming made the total sulfhydryl content decrease.

(2) The samples of 0 d and 1d had no significant differences on flexibility, while samples of 2-5 d were more accessible for enzymatic hydrolysis and the protein flexibility increased.

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