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Preparation and characterization of adhesive from spent hen proteins

Chanchan Wang, Jianping Wu *

4-10 Agricultural and Forestry Center, Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, AB, Canada T6G 2P5

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ABSTRACT

Spent hens, a by-product of the poultry industry, are of little economic value while their disposal is an environmental concern. In this study, spent hen proteins were modified by sodium dodecyl sulfate (SDS) or urea to develop adhesive, and the adhesive properties on wood veneers were investigated. Adhesives prepared with 3 M urea or 3% SDS show dry strengths of 7.99 ± 0.17 MPa and 9.35 ± 0.17 MPa, wet strengths of 3.35 ± 0.10 MPa and 2.90 ± 0.59 MPa, and soaked strengths of 5.21 ± 0.04 MPa and 8.89 ± 0.14 MPa. The morphologies of the adhesives on the wood veneers after curing were investigated by scanning electron microscope. Effects of modifications on protein structural and thermal properties were studied with Fourier transform infrared spectroscopy and differential scanning calorimetry, respectively. Adhesive mechanisms include protein unfolding to reveal secondary structures that can interact with wood substance, interaction between protein and the modification agents that enhance the strength in the protein bulk, and indispensable mechanical interlocking. The wood adhesives prepared in this study from spent hen proteins can be used in dry and wet applications.

1. Introduction

Proteins have been used for centuries to prepare adhesives, while petroleum-based adhesives started to dominate the market for decades due to their affordable cost and satisfactory performance [1]. However, limited reserves of petroleum and concerns about emission of volatile organic compounds (especially formaldehyde, a carcinogenic compound from petroleum-based adhesives), have revived attempts to develop biobased adhesives from renewable resources [2,3]. Adhesives prepared from soybean proteins have been extensively reported; adhesives developed from animal blood, gluten, and sorghum proteins have also been reported [3,6,10]. Biobased adhesives tend to perform low adhesive strength and poor water resistance compared to petroleum-based adhesives [3,4]. Recent research on protein-based adhesives focused on improving their adhesive strength and water resistance, and on reducing cost [1]. The adhesive strength and water resistance of soy proteins were improved by modifying proteins with sodium dodecyl sulfate (SDS), sodium dodecyl benzene sulfonate, sodium bisulfite, urea, or guanidine hydrochloride [4–6], or by developing soy protein biomimetics [1,7,8], or using a combination of protein and petroleum products [9].

Spent hens, a by-product, are of little economic value in the poultry industry. Traditional markets for spent hens, like food uses and feedstuffs, are no longer available due to little revenue and concerns over the safety of using animal by-product ingredients [11]. Thus an increasing number of birds are being composted or buried, resulting in increased waste volume and concentration; consequently, disposal of spent hens in landfill has become less acceptable due to concerns about environment [12]. Therefore, finding new methods to utilize spent hens are critical to eliminate those concerns while yield residual value to the poultry industry.

Spent hen contains around 25% crude protein by dry weight [13]. The primary protein in spent hens is myofibrillar; approximately 45% of the myofibrillar protein in muscle tissues is myosin [14]. Native myosin has a double-headed globular region and a helical tail. The objective of this study was to study the suitability of preparing adhesives from spent hen proteins. The potential of spent hen proteins in the development of adhesives is exploited in this study, where adhesives are prepared from spent hens and characterized with calorimetry and spectroscopy.

2. Experimental sections

2.1. Materials

Spent hens were purchased from a local supermarket (Edmonton, AB, Canada). Natural wood veneers (birch, 0.6 mm thick) were products of Windsor Plywood Co. (Edmonton, AB, Canada)

* Corresponding author. Tel.: +1 780 492 6885.
E-mail address: jianping.wu@ualberta.ca (J. Wu).

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and used as received. SDS and urea were purchased from Sigma Chemical Co. (St. Louis, MO, USA).

2.2. Protein extraction

Spent hen proteins were extracted at alkaline pH and recovered at acidic pH according to the pH-shifting method [14] with slight modifications. All procedures were performed at 4 °C. Spent hen meats were separated from the freezing carcasses. External fat and heavy connective tissue were removed from spent hen meat prior to blender grinding for myofibril isolation; ground spent hen meat was mixed with four times water (w/v) and stirred for 10 min. After filtration using a 200-mesh sieve, the slurry was centrifuged at 10,000 rpm at 4 °C for 25 min (Avanti J-E Centrifuge, Beckman, Mississauga, Ontario, Canada). Three layers: fat, myofibrillar, and collagen, were obtained in the order of top to bottom. The myofibrillar layer was collected, and the pH of the slurry was adjusted to 11 with 2 M NaOH. The solution was centrifuged at 10,000 rpm at 4 °C for 25 min. The protein rich supernatant was collected. The pH of the supernatant was adjusted to 5 with 2 M HCl. After centrifugation, the precipitate was collected and washed three times with deionized water. The collected precipitate was freeze dried and stored in −20 °C.

2.3. Protein modification

To modify proteins, spent hen protein was mixed with SDS (0.5%, 1%, 3%, and 5% of protein weight), urea (1, 3, 5, and 8 M), and alkaline solution (adjusted to pH 10 with 2 M NaOH) at a concentration of 10% (w/v), and stirred at room temperature for 4 h. These protein solutions were considered as protein adhesives. The adhesive strengths of the adhesives were investigated immediately after preparation. Freeze dried samples were used for investigating the effect of the protein modification.

2.4. Effect of protein modifications on protein structure

The effects of protein modifications on protein structure were studied using a Nicolet Magna 750 FTIR spectrometer (Madison, WI, USA). Protein samples prepared above were milled with potassium bromide (KBr) in a mortar to form a very fine powder, and then compressed into a thin sheet prior to analysis. Each spectrum was collected by accumulating 10 scans at a resolution of 8 cm^{-1}. FTIR spectra of the adhesives in the amide III modes from 1150 to 1350 cm^{-1}, which are attributed to N–H bending coupled to C–N stretching, were compared. The FTIR spectra were acquired using Bruker OPUS software (version 5.5).

2.5. Effect of protein modifications on thermal properties

The thermal properties of protein samples were studied using a heat flux differential scanning calorimetry (DSC, PerkinElmer, Norwalk, CT, USA). About 4 mg of freeze dried protein samples were placed in a large DSC stainless steel pan. Samples were held at 25 °C for 1 min and then scanned from 25 °C to 220 °C at a heating rate of 10 °C min^{-1}.

2.6. Wood specimen preparation

The dimension of each wood veneer was 0.6 mm × 20 mm × 120 mm (thickness, width, length, respectively). The wood veneers were conditioned in a humidity and temperature controlled chamber (ETS 5518, Glenside, PA, USA) at 50% humidity and 25 °C for seven days. The modified protein adhesive slurry (protein content was 0.1 g mL^{-1} with a standard deviation of 0.04 g mL^{-1}) was spread onto one end of each piece of wood veneer over an area of 5 mm × 20 mm. The two wood pieces with the adhesive coating were allowed to rest at room temperature for 5 min before they were assembled by hand and then hot-pressed under different conditions as in Section 2.7.

2.7. Adhesive strength

Adhesive strength was determined by an automated bonding evaluation system (ABES, Corvallis, OK, USA). Adhesive strength is the force required to cause a separation of two bonded surfaces. Dry strength was tested with the ABES immediately after cooling the bonding area by air jet (combined in the ABES) for five seconds. Water resistance of the adhesive was measured according to the ASTM Standard D1151-00 using the wet strength and soaked strength. Bonded wood specimens were soaked in tap water at 23 °C for 48 h and tested for wet strength immediately right after soaking. Soaked strength was measured after the soaked specimens were conditioned at 23 °C and 50% humidity for seven days in the chamber. Effects of curing conditions such as time (60, 120, 180, and 240 s at 110 °C and 3.5 MPa), temperature (80, 110, and 140 °C at 3.5 MPa for 240 s), and pressure (2.5, 3.5, and 4.5 MPa at 110 °C for 240 s) on adhesive strength were studied. Effects of concentrations of urea and SDS on adhesive strength of samples cured at 110 °C and 3.5 MPa for 240 s were studied. Data consisted of an average of at least three replicates.

2.8. Fracture morphology of the bonding area

The cross sections of pulled bonding surfaces of wood specimens were observed with a Hitachi S-2500 scanning electron microscope (SEM, Nissei Sangyo America Ltd, CA, USA). Specimens were coated with a thin layer of gold with a Gold Sputter Unit (Denton Vacuum, Moorestown, NJ, USA) before observation.

2.9. Statistical analysis

Univariate linear correlation analyses were performed to identify the best modification concentration and the major curing factors affecting adhesion properties. The statistical analysis was carried out with the statistical package for the social sciences (SPSS) software. The correlation coefficient (r) was used for linear estimations of the strength and direction of linear correlations between two parameters. The coefficient r is always between -1 and $+1$, where $r=-1$ or $+1$ means a perfect correlation and 0 means the absence of a relationship. If $r<-0.75$ or $r>0.75$ there is a strong correlation between two parameters. Correlations are considered statistically significant at a 95% confidence interval ($p<0.05$).

3. Results and discussion

3.1. Effect of urea and SDS modifications on spent hen protein adhesive properties

At increasing urea concentrations from 1 to 8 M, dry strength of the spent hen protein adhesive increased from 6.57 ± 0.34 MPa to 8.55 ± 0.56 MPa but the highest water resistance was reached at around 3 M urea, which was 3.35 ± 0.10 MPa, then started to decrease as urea concentration was further increased (Fig. 1A). At 1 M urea, a cooperative destruction of the tertiary protein structure might occur, which will result in a mildly denatured state that contains some secondary structure [5]. At 3–5 M urea, the denatured state gradually loses its residual secondary structure; the radius of gyration increases to a near maximum value, and the polypeptide chain becomes disordered with highly mobile side chains [15]. The unfolding of proteins increased their