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Paper

Preparation and Mechanical Properties of Wheat Protein Isolate Films Cross-linked with Resorcinol

M. Chandrasekhar*, M.N. Prabhakar*, Jung-Il Song*†

ABSTRACT: The purpose of the present work was to preparation and study of full biodegradable Eco-friendly bio-composites by using renewable resources. In this study, wheat protein isolate (WPI) films were formed by cross linking with resorcinol through solution casting method for packaging applications. By varying the resorcinol content (10, 20, 30, 40, and 50 wt %), its effect on mechanical properties of the wheat protein isolate film was measured. The addition of 20% resorcinol led to an overall increase in the tensile strength from 5.2 to 18.6 MPa and modulus increase from 780 to 1132 MPa than WPI films. The % elongation was increased from 2.8 to 9.05 when compared to unmodified WPI film. A thermal phase transition of the prepared WPI was assessed by means of DSC. FTIR is evident that the characteristic WPI spectral IR bands shifted on cross-linking with resorcinol.

Key Words: Wheat protein isolate, Resorcinol, Mechanical properties, Biodegradable films

1. INTRODUCTION

Petroleum-based plastics dominate today's plastics market because of their high strength, lightweight, low cost, easy processability, and good water barrier properties. Plastic materials may be degraded by naturally occurring microorganisms in the environment, but the process may take about 150 years (low density polyethylene), while paper can be naturally biodegraded in about one year [1]. Inert and non-biodegradable plastic materials account for about 30% by weight of municipal solid wastes, but due to their low density they represent 2/3 of the wastes volume. There has been a growing interest for edible films and coatings in recent years trying to reduce the amount of wastes, capable of protecting the food once the primary packaging is open, and because of public concerns about environmental protection [2]. However, most of the synthetic polymers are not biodegradable. Synthetic biodegradable polymers, such as poly(lactic acid), polycaprolactone, and poly (hydroxy butyrate) have high production costs. With the increasing concerns of environmental pollution caused by non-biodegradable, petroleum based plastics; increasing efforts have been made to utilize the polymeric materials derived from agricultural products.

Environmental concern about plastic materials is leading to and an increasing interest towards the development of ecological products. The use of disposable plastic material increases in understandable waste portion and for this reason it is necessary to develop more recyclable and/or biodegradable plastics to reduce the amount of plastic waste. The recent trend is to develop fully sustainable, biodegradable, eco-friendly and easily disposable materials. Therefore, in the past decades, extensive studies have been made for the potential use of polymeric materials derived from renewable resources, such as carbohydrate, starch and proteins [3-5]. Plant proteins from soy [6,7], corn [8], why protein [9], cotton seed [10], and wheat [11], have been studied because of their abundance, low cost, good biodegradability and sustainable properties for usage as films and plastics. Natural matrices are biodegradable, renewable and cheaper. Further, these materials are expected to lower the usage of synthetic polymers which are non-degradable and derived from precious depleting fossil fuels. In addition to its use as a food ingredient, non-food applications of wheat protein as polymeric materials have attracted increasing attention in recent years. The main attractive features of wheat protein-based plastics are that they are biodegradable, environmentally friendly, and from an abundant renewable

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*Department of Mechanical Engineering, Changwon National University
*†Department of Mechanical Engineering, Changwon National University, Corresponding author (E-mail: jisong@changwon.ac.kr)

resource. Use of wheat protein as a plastic material can be traced back to the 1930s. Brother and Mckinney studied the formaldehyde-hardened wheat plastics and their compounding with the phenolic resin. The formaldehyde-treated wheat plastics compounded with phenolic resin were reported to display very low water absorption.

However, in 1995 about 62% of total whey production was used in some application, but the remaining 38% of whey proteins (270,000 tons) were still available as valuable food ingredients. In the USA about 51.2% of the liquid whey produced was used by the food industry [12]. The whey protein is high quality, since it has all essential amino acids, and a biological value higher than egg or casein proteins [13]. Whey protein has also some functional properties of interest to the food industry, such as solubility, emulsification, foaming, gelation, and viscosity development [14,15].

Biopolymer films are generally designed from biological resources such as polysaccharides, proteins, lipids, and their derivatives. Proteins (plants or animal derived) are ideal material for bio-plastics production due to the presence of many polar and non-polar amino acid groups providing a broad spectrum of functional and structural properties [16-20].

Various researchers have been investigating the film properties of proteins from soy protein, cottonseed, wheat gluten, pea proteins, maize zein, sunflower proteins, collagen [21-23], corn [24], Faba Bean protein isolate [25] and whey protein [26]. In general, protein films are very brittle due to the extensive interactions that exist between the protein chains. One of the effective ways to overcome this problem is the addition of plasticizers during film casting. Plasticizer is a substance, which changes certain properties of the materials by reducing the intermolecular forces among polymer chains, brittleness of the films, causing a decrease in film strength and increase in film extensibility which can facilitate polymer processing [28]. Several theories have been proposed to explain the mechanism of plasticization action [27].

Protein-based film materials have a three-dimensional network structure. Such structure is formed through the three steps: first, rupture of low-energy intermolecular bonds using rupture agents, secondly rearrangement and orientation of polymer chains (shaping) and finally, the formation of a three dimensional network structure by forming new interactions and bonds. Generally, protein based biodegradable materials are manufactured by two technological processes: a wet process technology, which is based on dispersion or solubilisation of proteins and a dry process, which is based on the thermoplastic properties of proteins under low-moisture conditions [28,29]. Wheat protein isolate (WPI) WPI is a highly refined or purified form of wheat gluten (76.5% protein) with a minimum 90% protein content on a moisture-free basis. The United States is one of the major producers of wheat seed in the world. Wheat protein isolate containing approximately 90% protein, 4% fat, about 5% ash and 1% remaining other constituent. Several researchers modified wheat protein to improve its mechanical and physical properties and thermal stability, to reduce the moisture absorption, and to improve processability. WPI was modified by glutaraldehyde [30-32], to improve its physical and mechanical properties, and in the present study, glycerol used as plasticizers.

Although, very limited information is available on the production and characterization of protein isolate prepared from wheat, commercial production of wheat protein isolate, offers an inexpensive protein source for use in various applications ranging from food products to packaging films. Wheat protein isolate (WPI) is a protein obtained from wheat seeds of an annual plant with a protein content of 90%. Wheat protein isolate (WPI) has been studied as a film former due to its functional, cohesive and elastic properties [33-36] and they are non toxic and safe; hence, it provides possible applications in food packaging industry.

In this study, the author used resorcinol as a cross linking agent to prepare WPI films and then studied their tensile properties. Resorcinol has two hydroxyl groups, which can crosslink with carboxyl, amine, and amide groups present in WPI. External plasticizer was not used in the present case. According to material safety data sheet, both glutaraldehyde and resorcinol have a health risk index of 2 which is moderate on a scale of 0-4 and resorcinol-formaldehyde mixture is widely used in endodontic therapy [21]. As resorcinol has a 127.28C flash point and a 607.8°C auto ignition temperature the cross linking reactions were carried out at temperatures below 110°C.

2. MATERIALS AND METHODS

2.1 Processing and Modification of Wheat Protein Isolate Resin (MWPI)

WPI powder was obtained from Honeyville Food Products, Salt Lake City, Utah, USA. According to the supplier, the WPI used in the present case consisted 90% protein, 4%, about 5% ash and 1% remaining unknown constituents. Analytical grade sodium hydroxide (NaOH) was purchased from MERCK Chemicals, Mumbai, India and resorcinol was obtained from FINAR- Chemicals Ltd., Ahmadabad, India.

WPI resin films were prepared using a casting method. To prepare these films; WPI powder was mixed with distilled water (15 times by weight of WPI powder) and the mixture was stirred in a water bath at 75°C for 20 min, at the end 1N NaOH solution was added to adjust the pH value to 10. The stirring was continued at 75°C for another 20 min. Then 10-50% resorcinol was added and stirred for 5 min at 75°C. This stir-heating process denatures the WPI and is called 'pre-curing'. To obtain cured resin films, the pre-cured suspension was poured onto the Teflon-covered glass mould ($150 \times 150 \times 3\ mm^3$) to form a sheet with desired thickness. The glass mould with the pre-cured WPI was dried for 24 h at room temperature. The dried WPI film was peeled from the mould, sandwiched between two aluminium sheets and cured by hot pressing at 100°C for 20 min under a pressure of 2 MPa. The hot-pressing was carried out on a hydraulic hot press (Model PFM15, Technosearch Instruments, Kolbad, Maharashtra, India). It was very difficult to form a pristine WPI film due to its brittle nature.

2.2 Fourier Transform Infrared (FTIR) Spectra

The normalized infrared spectra of the WPI and MSPI film samples were recorded on an Analect RFX-65A Fourier transform infrared (FTIR) spectrophotometer. The samples were dried in an oven and ground into powder. In each case 2 mg of the powdered samples were mixed with 98 mg of KBr to prepare sample pellets. FT-IR analysis was performed with a resolution of 2 cm^{-1} in the range of 4000-400 cm^{-1}.

2.3 Moisture Content

The moisture content of the specimens was determined as per the ASTM D 1576-90 method, a procedure recommended for protein (wool) fibers. In this method, pre weighed samples in aluminium foil were dried in a circulating air oven at 105°C for 24 h, and the weight loss was then calculated.

2.4 Differential Scanning Calorimetry (DSC)

The differential scanning calorimetry analysis of wheat protein isolate (WPI), modified wheat protein isolates (MWPI) and resorcinol samples were scanned using SDT-Q 600, TA instrument under a nitrogen atmosphere. The samples were scanned from 30°C to 400°C at heating rate of 10°C/min.

2.5 Tensile Test

In order to study the composite films mechanical strength, its tensile properties are measured. The tensile test was performed as per ASTM D 882-02 standard method. Specimens with dimensions of 100 × 20 × 0.2 mm^3 were selected. The tensile properties such as maximum stress, Young's modulus and percent elongation at break were determined using an INSTRON 3369 Universal Testing Machine at a crosshead speed of 20 mm/min maintaining a gauge length of 50 mm. For tensile tests and for each category, five samples were tested at 50% RH and 23°C and the average values are reported.

3. RESULTS AND DISCUSSION

3.1 Characterization of wheat protein isolate with resorcinol

Fourier transform infrared spectrometer is an effective method to assess the structural changes in protein. In order to probe the cross-linking of the WPI molecules with resorcinol, the Fourier transform infrared spectra of pure WPI, cross-linked WPI with different resorcinol content and pure resorcinol are presented in the Fig. 1.

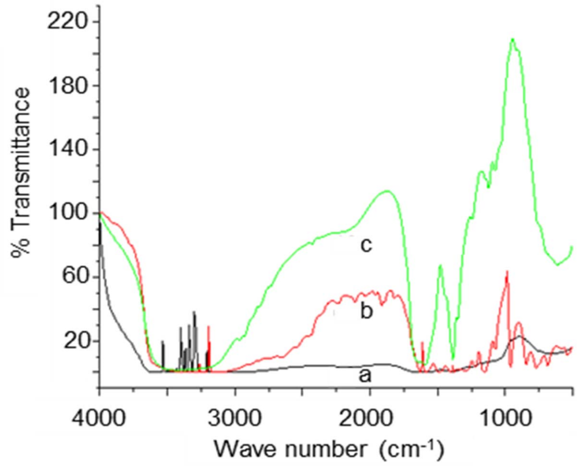

Fig. 1. FTIR spectra of (a) Pure WPI (b) pure Resorcinol (c) WPI 20% resorcinol

From Fig. 1, it is evident that the characteristic WPI IR bands shifted on cross-linking with resorcinol. The band position at 3355.9 cm^{-1} correspond to N-H stretching of WPI and the band at 3270 cm^{-1} correspond to O-H stretching of the phenolic group in resorcinol. Then characteristic peak position at 1651 and 1581.5 corresponds to C=O stretching and N-H stretching of amide I and amide II of unmodified WPI films, respectively. In case of resorcinol modified WPI films the band at 1582 cm^{-1} decreased its intensity and shifted slightly when compared to unmodified WPI film. This indicates that the cross-linking was affected by the NH_2 group of WPI and OH groups of resorcinol by hydrogen bonding.

3.2 Moisture Content

The moisture content values of the films for various resorcinol loadings are presented in Fig. 2.

From Fig. 2, it is evident that the moisture content was lower when the resorcinol content was 20%. For other loadings, the moisture content increased. It can also be observed from Fig. 3 that the moisture content of the resorcinol modified WPI films

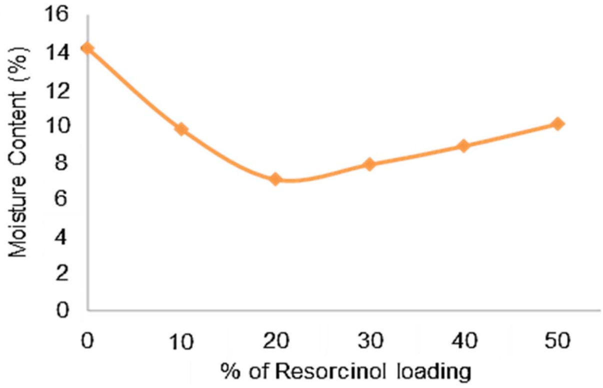

Fig. 2. Moisture content of resorcinol modified wheat protein isolate films

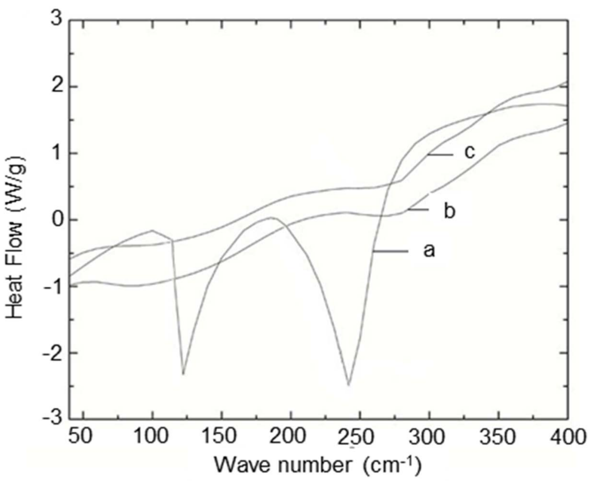

Fig. 3. DSC curves of (a) pure resorcinol (b) pure WPI (c) WPI 20% resorcinol

was lower than that of the unmodified ones. A similar observation was made by Zhichao *et al.* [23] in the case of N-hydroxymethyl-acrylamide cross linked with resorcinol. They observed that hydrophilicity of the polymeric chains decreased with increasing cross-linking that induced the attraction between cross-linked chains until the integral network was formed.

3.3 Differential Scanning Calorimetry (DSC)

The DSC endothermic curves of pure WPI, MWPI and pure resorcinol samples are presented in the Fig. 3. DSC thermograms showed two thermal transitions for WPI films, irrespective of their resorcinol content; a glass transition for the amorphous fraction and a melting transition for the crystalline one.

The Thermogram for pure resorcinol depicts two endotherms, the first one starting at approximately 110°C (corresponding to its melting point) and another at approximately 240°C (corresponding to its boiling point). In the case of pure WPI and modified WPI these two peaks are missing, indicating the plasticizing and crosslinking nature of resorcinol in the MWPI and the absence of free resorcinol in pure WPI.

3.4 Tensile Properties

The variation of ultimate tensile strength, modulus and % elongation at break of WPI films with resorcinol loading is shown in Fig. 4(a), Fig. 4(b) and Fig. 4(c) respectively. From Fig. 4(a) and Fig. 4(b), it is evident that both tensile strength and modulus of WPI films were at a maximum when resorcinol content was 20%. Thus the tensile studies indicate that the resorcinol content of 20% (w/w) was required in WPI films for optimum cross-linking.

From Fig. 4(c), it is observed that the percent of elongation at break is increasing with resorcinol content, due to the fact that, incorporation of resorcinol reduces make crosslink between protein chains, thus increasing the chain mobility. This behaviour indicates that resorcinol is also acting as a plasticizer.

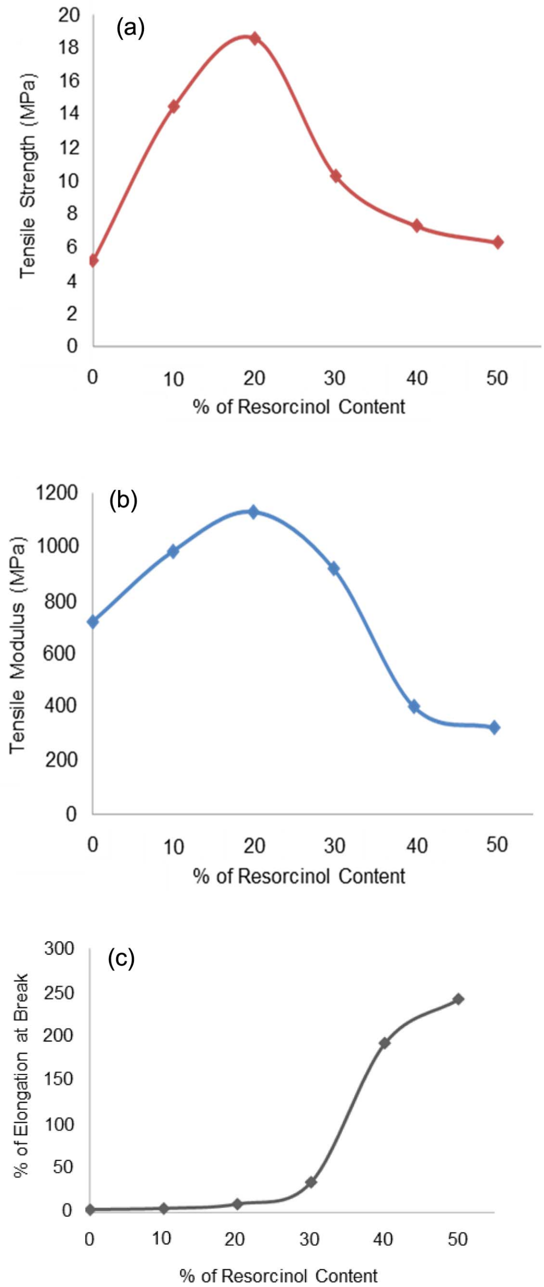

Fig. 4. (a): Tensile strength of wheat protein isolate films with resorcinol content, (b) Tensile modulus of wheat protein isolate films with resorcinol content, and (c). % Elongation at break of wheat protein isolate films with resorcinol content

3.5 Morphology (SEM) Analysis of WPI Films

The crosslinked WPI films were dried in vacuum and stored in a desiccator before the SEM micrographs recorded. Some specimens were cryogenically cooled and brittle fractured. Fig.

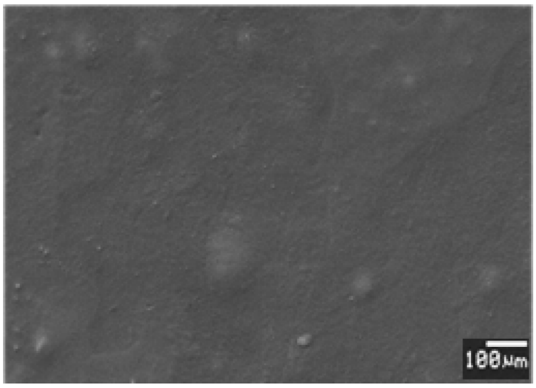

Fig. 5. The scanning electron microscope (SEM) photographs of 10 wt% of resorcinol plasticizer WPI film

5 shows the cross-sectional images of crosslinked WPI films and the effect of resorcinol level on films. The film obtained was very clear and uniform in thickness. At relatively less than 10 wt % of resorcinol, the films were brittle and difficult to peel from the casting surface. The author used resorcinol as a cross linking agent and no external plasticizer was used. Resorcinol was homogeneously incorporated within a network of hydrogen bonds between the WPI molecular chains, which makes the film more flexible, soft and transparent. The resorcinol apparently affects the intra and intermolecular interactions of WPI films. The rides and valleys were likely formed due to the extensive local deformation of the "protein-matrix" formed after consolidation of individual protein particles.

4. CONCLUSION

Fully bio-degradable eco-friendly WPI films were prepared with resorcinol as a cross linking agent. In this case, no external plasticizer was used. The optimum resorcinol level required for effective cross linking was determined using the tensile strength, tensile modulus and % elongation at break. The data show that the best combination was obtained when the resorcinol content was 20% by weight of WPI. Further, the moisture content of these modified films was at a minimum when the resorcinol content was 20% by weight of WPI. DSC analysis indicated the absence of free resorcinol present in the MWPI films. The transparent resorcinol crosslinked WPI films showed superior tensile strength and modulus, % of elongation and thermal stability. In conclusion, the prepared transparent films can be applicable for food packaging especially medical applications. Preparation of wheat protein isolate composites by using resorcinol as crosslinker is an innovative study in the field which the most employed technique to prepare films is a low cost solution casting.

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