Carbohydrate Polymers 97 (2013) 817–824

Contents lists available at SciVerse ScienceDirect

Carbohydrate Polymers

journal homepage: www.elsevier.com/locate/carbpol

Soluble soybean polysaccharide: A new carbohydrate to make a biodegradable film for sustainable green packaging

Sima Tajik[a], Yahya Maghsoudlou[a,*], Faramarz Khodaiyan[b], Seid Mahdi Jafari[a], Mehran Ghasemlou[b], Mehran Aalami[a]

[a] Department of Food Science and Technology, Gorgan University of Agricultural Sciences and Natural Resources, Beheshti Avenue, Gorgan 49138-15739, Iran

[b] Department of Food Science, Engineering and Technology, Faculty of Agricultural Engineering and Technology, Campus of Agriculture and Natural Resources, University of Tehran, Karaj, Iran

ARTICLE INFO

Article history:
Received 30 March 2013
Received in revised form 20 April 2013
Accepted 17 May 2013
Available online 24 May 2013

Keywords:
Soluble soybean polysaccharide
Plasticizer
Atomic force microscopy
Optical properties

ABSTRACT

Biodegradable edible films based on soluble soybean polysaccharide (SSPS), a new film-forming material, and three levels of glycerol (20%, 30% and 40%, w/w) as plasticizer, were developed and evaluated in terms of physical, mechanical, barrier and optical properties as well as their microstructure. SSPS-based films with a concentration of 20% glycerol possessed the lowest water vapor permeability. Increasing the glycerol content increased ($P<0.05$) values for elongation at break, but decreased ($P<0.05$) tensile-strength values. It was found that plasticizer concentration significantly affected the films' glass-transition temperature; however, it had no significant effect on their melting point. Color measurement showed that increasing the glycerol concentration caused the b and L values to increase, while ΔE value decreased. These results were explained by the film's microstructure, which was analyzed by atomic force microscopy and scanning electron microscopy. The results indicated that SSPS could be a promising food-packaging material.

1. Introduction

Nowadays, millions of tons of plastics are produced annually all over the world, and their production and consumption continue to increase; this increase has created serious environmental problems due to the materials' inability to biodegrade (Debeaufort, Quezada-Gallo, & Voilley, 1998). In order to solve the problems generated by plastic waste, much research effort has been done to obtain an environmental friendly material. Most of the research is focused on substitution of petro-based plastics by biodegradable materials with similar properties and low cost. Significant effort to extend the shelf life and enhance food quality while reducing packaging waste has encouraged food and packaging industries to explore for new biobased packaging materials that can reduce waste-disposal problems. Biopolymers derived from natural resources, which have attracted a great deal of attention in recent years, are considered as potential substitutes for traditional nonbiodegradable plastic films owing to their low cost, easy availability from reproducible resources and biodegradability (Janjarasskul & Krochta, 2010). Edible, biodegradable films, by acting as barriers to control the transfer of moisture, oxygen, lipids and flavors, can prevent quality deterioration and increase the shelf life of food products (Gontard, Duchez, Cuq, & Guilbert, 1994).

Several studies have reported the use of polysaccharides from different sources to prepare films and coatings with different properties, and have indicated that these carbohydrates are promising materials (Averous & Boquillon, 2004; Ghanbarzadeh, Almasi, & Entezami, 2010; Mali, Sakanaka, Yamashita, & Grossmann, 2005). Polysaccharide-based films are relatively stiff, and have a strong hydrophilic character compared with synthetic packaging films. Therefore, to overcome these films' brittleness and increase their workability and flexibility, various plasticizers, usually polyols, have been widely used. Glycerol, one of the most studied and popular, is a hydrophilic plasticizer; when added at the correct level with respect to the biopolymer content, it can reduce intermolecular forces and increase the mobility of polymer chains, a process generally used to improve the mechanical properties of edible films (Sobral, Menegalli, Hubinger, & Roques, 2001; Sothornvit & Krochta, 2001). In recent decade, a novel polysaccharide – soluble soybean polysaccharide (SSPS) – has been extracted from cell-wall material of the cotyledon of soybeans. SSPS has a pectin-like structure composed of a galacturonan backbone of homogalacturonan (α-1,4-galacturonan) and rhamnogalacturonan (repeating units being comprised of α-1,2-rhamunose and α-1,4-galacturonic acid),

* Corresponding author. Tel.: +98 171 442 6432; fax: +98 171 442 6432.
E-mail address: y.maghsoudlou@gau.ac.ir (Y. Maghsoudlou).

0144-8617/$ – see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.carbpol.2013.05.037

branched by β-1,4-galactan and α-1,3- or α-1,5-arabinan chains (Maeda, 2000; Nakamura, Furuta, Maeda, Takao, & Nagamatsu, 2002). SSPS has been reported to provide health benefits to humans through lowering blood cholesterol, improving laxation and reducing the risk of diabetes (Shorey, Willis, Lo, & Steinke, 1985; Tsai, Vinik, Lasichak, & Lo, 1987). Apart from its nutritional value, it has various functions such as dispersion, stabilization, emulsification and adhesion (Asai et al., 1994). Previous study have found that SSPS can be used as a functional ingredient in fortified foods (Furuta & Maeda, 1999). The literature data and preliminary studies in our laboratory have shown that SSPS can produce films with good appearance and satisfactory mechanical properties: it appears to have excellent potential as a film-forming agent. However, to the best of the authors' knowledge, there is no specific study on the film characteristics of SSPS. In the recent years, a major emphasis has been placed on the search for new film-forming materials with different compositions and properties. This study aims to develop a novel edible film based on SSPS, with potential applications as an edible film and a biodegradable food-packaging material, and to examine in detail the physical, mechanical, thermal, barrier and microstructural properties of the resulting films as a function of varying concentrations of glycerol as plasticizer.

2. Materials and methods

2.1. Materials

Soluble soybean polysaccharide (abbreviated as SSPS) was supplied by Fuji Oil (Osaka, Japan). Magnesium nitrate and sodium chloride (used to equilibrate films at 50% RH and 75% RH, respectively) together with sorbitol and glycerol were purchased from Sigma Chemical Co (St. Louis, MO). All other reagents used were of analytical grade.

2.2. Preparation of films

The films were prepared according to the so-called casting technique. SSPS film-forming solutions (3%, w/v) were prepared by dissolving 3 g SSPS in 100 ml distilled water under magnetic stirring (500 rpm), followed by heating to 90 °C on a hot plate for 30 min. The solution was then mixed with plasticizer (50% w/w of the polysaccharide). Both glycerol and sorbitol were tested as possible plasticizers, but the former gave better results. Following the addition of glycerol, stirring was continued for a further 15 min at 82 °C. SSPS films were cast by pouring the mixture onto polystyrene Petri dishes (14 cm in diameter) placed on a leveled granite surface for approximately 18 h at room temperature and room relative humidity. Films without any plasticizers were also prepared for later comparison. The film solution was left for several minutes to naturally remove most of the air bubbles incorporated during stirring. Dried films were peeled off the casting surface and stored inside desiccators at 25 ± 1 °C and 53% RH until evaluation.

2.3. Determination of physical properties of films

2.3.1. Film thickness

Thickness of the films was measured using a manual digital micrometer (Mitutoyo No. 293-766, Tokyo, Japan) to the nearest 0.001 mm. Measurements were made in at least ten random locations for each film, and an average value was calculated. The average value was used in calculations for tensile properties and WVP tests.

2.3.2. Moisture content

The films' moisture content was determined by measuring the weight loss of films, upon drying in an oven at 110 °C until a constant weight was reached (dry sample weight). Three replications of each film treatment were used for calculating the moisture content.

2.3.3. Film solubility in water

Solubility of SSPS films in water was measured following the method of Shojaee-Aliabadi et al. (2013). Film samples were dried at 110 °C for 24 h in a laboratory oven and then weighted to determine initial solid content. Pre-weighed film samples (1 cm × 3 cm) were immersed under constant agitation in 50 ml of distilled water for 6 h at 25 °C. After that period, the remaining pieces of films were filtered and dried at 110 °C to constant weight (final dry weight). The water solubility (%) of the film was calculated according to the equation WS (%) = $((W_i - W_f)/W_i) \times 100$, where W_i is the initial weight of the film expressed as dry matter and W_f is the weight of the desiccated undissolved film.

2.3.4. Moisture uptake

Moisture uptake was measured by following the method of Ghanbarzadeh and Almasi (2011) instead of the classical technique (immersion in water), because SSPS is very sensitive to liquid water and can partially dissolve after long time exposure to water. Prior to testing all specimens were first conditioned using anhydrous $CaSO_4$ at 0% RH for 48 h. The film samples were cut to a square piece of 2.0 cm × 2.0 cm and accurately weighed to give the dried film (W_i). After weighing, they were conditioned at 25 °C in a desiccator containing K_2SO_4 saturated solution to ensure a relative humidity of 97%. The samples were removed from the desiccator after 48 h and weighed (W_1). The moisture uptake values of the samples were calculated using the following equation:

$$\%\,\text{moisture uptake} = \frac{(W_1 - W_i)}{W_i} \times 100 \quad (1)$$

where W_1 is the weight of the film sample after exposure to 97% RH for 48 h, and W_i is the initial weight of the sample. Each test consisted of triplicate measurements and expressed as the mean value.

2.4. Water vapor permeability

Standard method E96/E96M (ASTM, 2012a) was used to determine water vapor transmission rate, with a 75% RH gradient at 25 °C. Diffusion cells containing anhydrous calcium chloride desiccant (0% RH, assay cup) were sealed by the test film (0.00287 m^2 film area). To maintain a 75% RH gradient across the film, a sodium-chloride-saturated solution (75% RH) was used in the desiccators. The RH inside the cell was always lower than outside, and water vapor transport was determined from the weight gain of the diffusion cell at a steady state of transfer. Changes in the weight of the cell were recorded to the nearest 0.0001 g and plotted as a function of time. The slope of each line was calculated by linear regression ($r^2 > 0.99$), and the water vapor transmission rate was calculated from the slope of the straight line (g/s) divided by the test area (m^2). All values for water vapor transmission rate (WVTR) were corrected for air-gap distance between the calcium chloride and the film surface according to the equations of Gennadios, Weller, and Testin, 1993. After the permeation tests, the film thickness was measured, and water vapor permeability (WVP) (g Pa^{-1} s^{-1} m^{-1}) was calculated as:

$$\text{WVP} = \frac{\Delta m}{A\Delta t}\frac{X}{\Delta p} \quad (2)$$

where $\Delta m/\Delta t$ is the weight of moisture gain per unit of time (g/s), X is the average film thickness (mm), A is the area of the exposed film surface (m^2), and Δp is the water vapor pressure difference

between the two sides of the film (Pa). WVP was measured for three replicated samples for each type of film.

2.5. Color measurement

A CIE colorimeter (Minolta CR 300 Series, Minolta Camera Co. Ltd., Osaka, Japan) was used to evaluate the color of SSPS films. The colorimeter was calibrated using a standard white plate (L^* = 93.49, a^* = −0.25, b^* = −0.09). Then, the color measurements were performed by placing the film specimens over colorimeter. At least three points of each sample were selected randomly to measure color properties of SSPS films. The following Eqs. (3)–(5) were used to calculate the total color difference (ΔE), whiteness (WI) and yellowness (YI) indexes of samples, respectively (Bolin & Huxsoll, 1991).

$$\Delta E = \sqrt{(L^* - L)^2 + (a^* - a)^2 + (b^* - b)^2} \quad (3)$$

$$\mathrm{WI} = 100 - \sqrt{(100 - L)^2 + a^2 + b^2} \quad (4)$$

$$YI = \frac{142.86\, b}{L} \quad (5)$$

where L^*, a^*, and b^* are the color parameter values of the standard and L, a, and b are the color parameter values of the sample.

2.6. Mechanical properties

According to ASTM standard method D882 (ASTM, 2012b) a Testometric Machine M350-10CT (Testometric Co. Ltd., Rochdale, Lancs., England) was used to measure tensile strength (TS) elongation at break (EB) and Young's modulus (YM) of the preconditioned SSPS films (with dimensions of 1 cm × 10 cm). The initial grip separation and cross-head speed were set to 50 mm and 50 mm/min, respectively. A microcomputer was used to record the stress–strain curves. Tensile properties included TS and EB were calculated as outlined in ASTM D882 (2001). At least five replicates of each film were tested.

2.7. Thermal properties

The thermal properties of the films were carried out using DSC equipment (TA Instrument, New Castle, Del., USA). Samples of 2–4 mg were sealed in standard aluminum dishes, using a sealed empty aluminum dish as the reference sample. All Samples were scanned at a heating rate of 10 °C/min between temperature ranges of −50 and 250 °C. Nitrogen was used as the purge gas at a flow rate of 20 ml/min. The glass transition temperatures (T_g) of the different films were determined from resulting thermograms as the midpoint temperature of a step-down shift in baseline, due to the discontinuity of the specific heat of the sample. The melting point (T_m) was calculated as the temperature where the peak of the endotherm occurs. The enthalpy (ΔH_m) of the sol–gel transition was determined as the area over the endothermic peak. All these properties were determined in duplicates and the results were averaged.

2.8. Scanning electron microscopy (SEM)

Microstructural analysis of the surface and cross-sections of the dried films was conducted by scanning electron microscopy (Cam Scan MV2300). The samples were fractured in liquid nitrogen, mounted on aluminum stubs using a double-sided adhesive tape, and sputtered with a fine gold layer before obtaining the micrographs. All samples were examined using an accelerating voltage of 15 kV. Samples were photographed at an angle of 90° to the surface to allow observation of the films' cross-section.

2.9. Atomic force microscopy (AFM)

Before testing, samples were preconditioned at 50% RH and at room temperature for at least 48 h. The AFM was carried out by a Autoprobe CP Research instrument (Veeco Instruments) in contact mode with a 125 μm × 125 μm scan size and a 6 μm vertical range. The resulting data were transformed into a 3D image. Measurements were taken from several areas of the film surface (50 μm × 50 μm). According to method ASME B46.1 (ASME, 2009), the following two statistical parameters related with sample roughness were calculated: average roughness (R_a: average of the absolute value of the height deviations from a mean surface), root-mean-square roughness (R_q: root-mean-square average of height deviations taken from the mean data plane). A minimum of three replicates were considered to obtain these parameters.

2.10. Statistical analysis

The experiments were factorial with a completely randomized design. Data was analyzed by the analysis of variance (ANOVA) procedure using SAS software (Version 9.1; Statistical Analysis System Institute Inc., Cary, NC, USA). Duncan's Multiple Range tests were used to compare the difference among the mean values for the films' properties at the level of 0.05.

3. Results and discussion

3.1. Film formulation

Based on a preliminary study using different concentrations of SSPS up to 6%, 3% SSPS was chosen as a suitable polysaccharide concentration for the film-forming solution. The SSPS films were prepared using glycerol and sorbitol as plasticizers at concentrations between 20% and 40% (w/w). The sorbitol-plasticized films showed white stains and irregular thickness, and they broke easily during peeling from plates. Thus, we discontinued working with this type of plasticizer. In contrast, the films cast from solutions containing glycerol were found to be flexible and could be easily peeled from the casting plate. Films formulated without plasticizer were brittle, and it was necessary to handle them carefully, and it was not possible to evaluate their mechanical and optical properties. As glycerol concentration increased, films became more flexible and sticky. At macroscopic scale, all films were homogenous and transparent and apparently without pores or cracks, even the ones prepared without plasticizer, as shown in Fig. 1.

3.2. Physical properties of films

The films' thicknesses were found to be similar (Table 1). The thicknesses were controlled well because all film-forming solutions were weighed to the same mass prior to casting.

Packaging films should maintain moisture levels within the packaged product. Therefore, the films' moisture content and total soluble matter are the most important parameters in food-packaging applications.

Table 1 shows the variations of the moisture content in SSPS films for increasing concentrations of glycerol. Increasing the level of glycerol led to increases in the films' moisture content. However, the values present a small difference ($p < 0.05$) between SSPS films with different glycerol concentrations. The presence of water in SSPS films is highly dependent on glycerol concentration. Glycerol, due to its hydrophilic nature, retains water in the film matrix. Higher concentrations of plasticizer favor the adsorption of water

Table 1
Physical properties of SSPS films incorporated with various concentration of glycerol as plasticizer[a].

Glycerol conc. (% w/w) (based on SSPS content)	Thickness (mm)	Moisture content (% d.b.)	Solubility in water (%)	Water uptake (%)	WVP ($g\,s^{-1}\,m^{-1}\,Pa^{-1} \times 10^{-10}$)
0	0.077 ± 0.006 a	11.24 ± 0.26 c	86.03 ± 3.36 b	6.98 ± 0.32 c	0.93 ± 0.43c
20	0.071 ± 0.007 a	11.26 ± 0.17 c	89.01 ± 2.10 ab	7.22 ± 0.11 c	1.29 ± 0.13 bc
30	0.073 ± 0.005 a	12.05 ± 0.21 b	90.12 ± 1.89 ab	7.96 ± 0.27 b	2.04 ± 0.70 b
40	0.081 ± 0.002 a	13.57 ± 0.41 a	91.01 ± 1.24 a	8.56 ± 0.29 a	3.28 ± 0.48 a

[a] Values are given as mean ± standard deviation. Different letters in the same column indicate significantly different ($p < 0.05$) when analyzed by Duncan's Multiple Range Test.

molecules, which is mainly attributed to the predisposition of plasticizers to form hydrogen bonds (O—H).

The water solubility of SSPS films is shown in Table 1. Results demonstrated that increasing glycerol from 20% to 40% increased film solubility; however, the differences observed were not significant ($P > 0.05$). The results obtained are directly attributable to the hydrophilic behavior of glycerol in SSPS films. Generally, high solubility would indicate lower water resistance; however, for some applications, such as packaging wrap, high solubility is an indicator of biodegradability, which could be an advantage (Stuchell & Krochta, 1994). This is the case when the film or coating will be consumed simultaneously with the food. Similar results were also observed by Ghasemlou, Khodaiyan, Oromiehie, and Yarmand (2011) and Ahmadi, Kalbasi-Ashtari, Oromiehie, Yarmand, and Jahandideh (2012), who suggested that increasing the glycerol content in films increased the water-soluble dry content. The addition of glycerol also had a strong influence over the water uptake. The unplasticized films had lower moisture absorption than the glycerol-plasticized films. However, water uptake increased significantly ($P < 0.05$) with increased glycerol concentration (Table 1). These results were in accordance with those of Ghanbarzadeh and Almasi (2011), who reported that higher concentrations of glycerol in the carboxymethyl cellulose-based films gave a higher hydrophilicity, leading to the films' increased water uptake.

3.3. Water vapor permeability (WVP)

Since a main function of a food packaging is often to avoid, or at least to decrease, moisture transfer between the food and the surrounding atmosphere, WVP should be as low as possible (Hosseini, Rezaei, Zandi, & Ghavi, 2013). The WVP of SSPS films containing various concentrations of glycerol as plasticizer are shown in Table 1. There was a significant difference between the WVP values of films made with different glycerol concentrations ($p < 0.05$). The increase in WVP with increasing hygroscopic plasticizer concentration is common in edible films (Cuq, Gontard, Cuq, & Guilbert, 1997; Sobral et al., 2001). The same behavior was shown in the study of Cuq et al. (1997), who suggested that the increased WVP in films as a consequence of adding plasticizer was due to the reorganization of the polymeric network, thereby making the structure less dense and increasing free volume. Consequently, there is greater space for water and other molecules to migrate through the film structure. As a result, it promotes the sorption of water to relative humidities above 80%. Under these conditions, the plasticizing effect of water is additional to that of glycerol, thus explaining the films' higher WVP (Gontard et al., 1994). Cuq et al. (1997) also mentioned that WVP and water content in films tended to increase with increasing glycerol concentration due to higher numbers of polar groups present in the films (hydrophilic plasticizer being compatible with hydrophilic film-forming materials like SSPS, thereby enhancing sorption of polar molecules like water). Consequently, this would again increase water diffusion within the polymer matrix of the film. The WVP value of SSPS films was in the range of 0.93×10^{-10} g m/m^2 s Pa–3.28×10^{-10} g m/m^2 s Pa, and was less permeable to water vapor than the films from psyllium-seed gum examined by Ahmadi et al. (2012), who found WVPs in the range of 1.16 and 1.88×10^{-10} g m^{-1} s^{-1} Pa. Additionally, the moisture-barrier properties of the SSPS films were slightly higher than those of cellophane (0.84×10^{-10} g m^{-1} s^{-1} Pa) (Shellhammer & Krochta, 1997), suggesting their application in some food products.

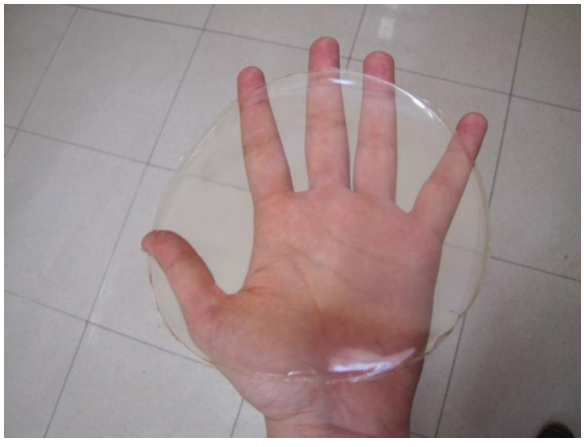

Fig. 1. Appearance of soluble soybean polysaccharide based film (30% w/w glycerol was used in its formulation).

3.4. Optical properties

Color parameters of studied SSPS films are presented in Table 2. Our results showed that all of the color parameters of SSPS films, with the exception of the a value, were significantly ($p < 0.05$) changed when glycerol concentration increased. Incorporation of glycerol increased film lightness, as evidenced by higher ($P < 0.05$) L values. Conversely, yellowness (b values) decreased with incorporation of glycerol (Table 2).

The SSPS films with 40% glycerol had the highest L, and were lighter than the 20%-glycerol films. The overall color of the films was lightened with incorporation of glycerol. Therefore, addition of plasticizer to SSPS films can improve their optical properties. Color functions such as ΔE, which indicates the degree of total color difference from the standard color plate, YI, which indicates degree of yellowness, and WI, which indicates degree of whiteness can be described as a result of increasing glycerol concentration in SSPS films. While WI increased significantly ($p < 0.05$) as a result of increase in glycerol concentration, YI decreased considerably. ΔE showed the same pattern as YI, indicating that the color difference of glycerol-incorporated SSPS film was mainly due to changes in yellowness. These results agreed with visual observations. Similar effects on color parameters from adding glycerol to the film matrix have been previously observed by Ghasemlou et al. (2011) and Ahmadi et al. (2012). The films obtained in this work have the advantage of providing light-barrier properties, which can be desirable depending on the film's application in food products.

Table 2
Hunter color parameters (L, a, b), total color differences (ΔE), whiteness index (WI) and yellowness index (YI) for the studied film samples[a].

Glycerol conc. (% w/w) (based on SSPS content)	L	a	b	ΔE	WI	YI
20%	94.21 ± 2.66 b	−1.41 ± 0.15 b	17.86 ± 1.64 a	17.56 ± 2.04 a	80.91 ± 3.01 b	26.41 ± 1.07 a
30%	94.43 ± 1.56 b	−1.33 ± 0.12 a	15.60 ± 2.46ab	15.31 ± 1.26 ab	83.21 ± 4.31 ab	25.44 ± 2.45 a
40%	96.26 ± 0.88 a	−1.40 ± 0.15 b	14.53 ± 2.71b	13.62 ± 2.55 b	84.83 ± 3.57 a	22.19 ± 1.22 a

[a] Values are given as mean ± standard deviation. Different letters in the same column indicate significantly different ($p < 0.05$) when analyzed by Duncan's Multiple Range Test.

3.5. Mechanical properties

The mechanical properties of films have been characterized by the TS and EB values, which are indicators of the film's strength and flexibility, respectively. The effect of plasticizer concentration on the mechanical properties of SSPS-based films is presented in Fig. 2.

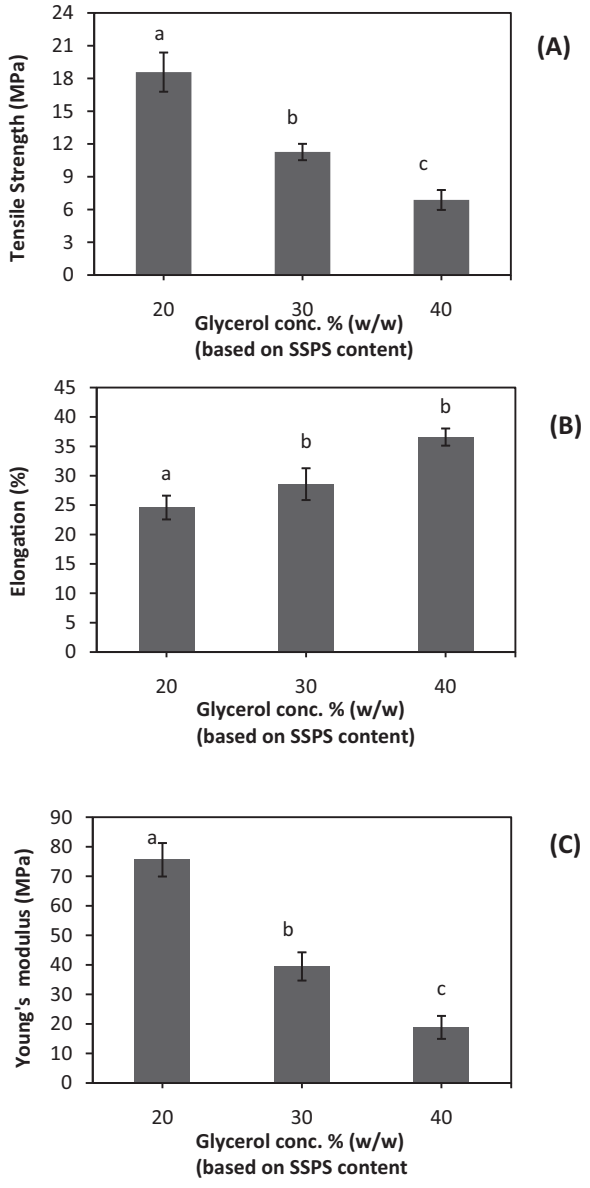

Fig. 2. Tensile strength (A), elongation at break (B) and Young's modulus (C) of the soluble soybean polysaccharide based films incorporated with various glycerol concentrations as a plasticizer. *Note*: *a*, *b* and *c* are different letters represent significant differences ($p < 0.05$) between the means obtained in Duncan's test.

The TS of plasticized SSPS films decreased ($P < 0.05$) when glycerol content increased from 20% (18.58 MPa) to 30% (11.27 MPa). For the same amount of added glycerol, the EB values of the films improved ($P < 0.05$) when the quantities of plasticizer were increased. Furthermore, when glycerol concentration increased from 30% to 40%, this behavior was more evident, with a decrease of TS value of approximately 62% and an increase of EB value of 48%. Based on these results, it can be concluded that increasing the glycerol concentration in SSPS films improved film extensibility and reduced its resistance. The typical effects on mechanical properties of adding plasticizers, such as increases in elongation and decreases in tensile strength, have been broadly discussed in the literature (Abdorreza, Cheng, & Karim, 2011; Lourdin, Coignard, Bizot, & Colonna, 1997). Plasticizers interfere with polymer chains: they decrease intermolecular forces, soften the rigidity of the film's structure and facilitate the sliding of chains, and thus help to improve the overall flexibility of the films. A comparison of the tensile test results obtained in this study to those reported for synthetic polymers shows comparable TS values between SPSS films and both low- and high-density polyethylene, reported between 9–17 MPa and 16–41 MPa, respectively (Smith, 1986). EB values for the SPSS films tested in this study were better than those for cellophane and polystyrene, which have been reported between 15–25% and 1–10%, respectively (Gennadios et al., 1993).

3.6. Thermal properties

Table 3 shows the glass-transition temperatures (T_g), melting temperatures (T_m), and enthalpies of fusion (ΔH_m). T_g of a polymer blend can reveal useful information about miscibility between its constituent components (Xiao, Lim, & Tong, 2012). Unplasticized SSPS films had a T_g of 30.82 °C, which was higher than those of glycerol-plasticized films. A comparison of the results of different plasticized films showed that increasing the glycerol concentration decreased the T_g of the films from 24.67 °C (20% (w/w) glycerol) to 21.92 (30% (w/w) glycerol) and 19.73 °C (40% (w/w) glycerol) (Table 3). Similar results were obtained by Arvanitoyannis, Psomiadou, Nakayama, Aiba, and Yamamoto (1997) with films of gelatin/starch blends plasticized with glycerol or sorbitol, and by Carvalho and Grosso (2004) with films based on gelatin plasticized with glycerol. Glycerol, due to its plasticizing effect, decreases glass-transition temperatures of polysaccharide films, which is in agreement with the free-volume theory of plasticization.

Table 3 also shows the changes in ΔH_m and T_m of SSPS films with increases in glycerol concentration. Increases in glycerol concentration lead to an increase in ΔH_m, and to a decrease in T_m values. As a result, the observed single glass transition for all glycerol-plasticized films was attributed to the whole polymer matrix. They remained homogeneous throughout the heating cycle, as no phase separation was observed (data not shown). If blends of polymer and plasticizers are immiscible, the mixture will exhibit two T_g and T_m corresponding to the two pure phases (Ghasemlou, Khodaiyan, & Oromiehie, 2011a).

Table 3
DSC data of SSPS-based film as a function of various glycerol concentrations[a].

Glycerol conc. (% w/w) (based on SSPS content)	T_g (°C)	T_m (°C)	ΔH_m (J/g)
0	30.83 ± 2.89 a	98.37 ± 1.78 a	129.31 ± 2.20 c
20	24.67 ± 1.53 ab	94.29 ± 0.64 a	151.73 ± 4.73 b
30	21.92 ± 1.49 ab	93.35 ± 1.52 a	169.07 ± 1.31 a
40	19.73 ± 3.78 b	88.92 ± 2.75 a	180.44 ± 3.84 a

[a] Values are given as mean ± standard deviation. Different letters in the same column indicate significantly different ($p < 0.05$) when analyzed by Duncan's Multiple Range Test.

3.7. Film microstructure

Scanning electron microscopy (SEM) has been widely used as a tool for the study and characterization of the microstructure of edible films.

Fig. 3 shows the SEM micrographs of the surface and cross-section for both the control and the glycerol-containing SSPS films. The surface of the control film appeared smooth. However, plasticized SSPS films showed remarkable differences in terms of surface microstructure compared to the control film, probably due to the hydrophilicity of glycerol, which may absorb higher water content. As the concentration of glycerol increased from 20 to 40%, the film surface appeared to be much rougher. The voids on the surface of the films could be the site for water binding during moisture absorption, which may explain the slightly higher WVP of the films. Cross-section images revealed that control films had a rough and uneven structure, while glycerol-plasticized films showed a smooth and uniform internal-structure morphology. The appearance of all the glycerol-plasticized films was generally uniform and homogeneous, and SEM observations of films with different glycerol concentrations did not present any obvious differences in structure.

3.8. Atomic force microscopy (AFM)

Atomic force microscopy (AFM) is a powerful tool for studying surfaces, and has been used to provide qualitative and quantitative information about biopolymers at the nanometer scale that are often inaccessible by any other experimental technique. It has been used previously to study isolated edible films (Ghasemlou, Khodaiyan, & Oromiehie, 2011b). AFM was used to obtain roughness parameters such as R_a and R_q; these values as calculated for the surfaces of films are shown in Fig. 4.

Pure SSPS films were the smoothest and showed the lowest values of all roughness parameters. The film surfaces became increasingly rough as glycerol was incorporated. All the films showed good structural integrity, and no pores or cracks were observed.

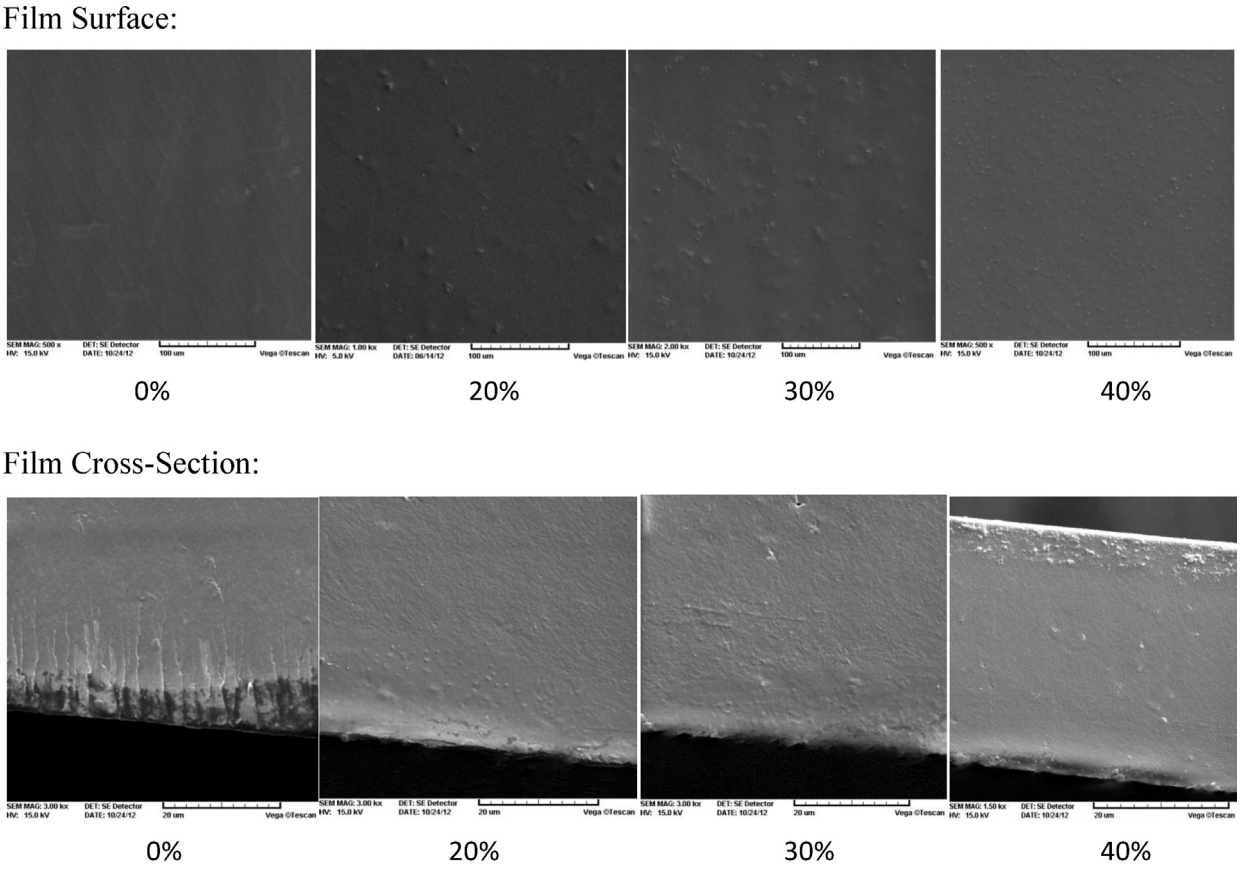

Fig. 3. Scanning electron microscopic images of surface and cross-section of the soluble soybean polysaccharide films plasticized with various glycerol concentration.

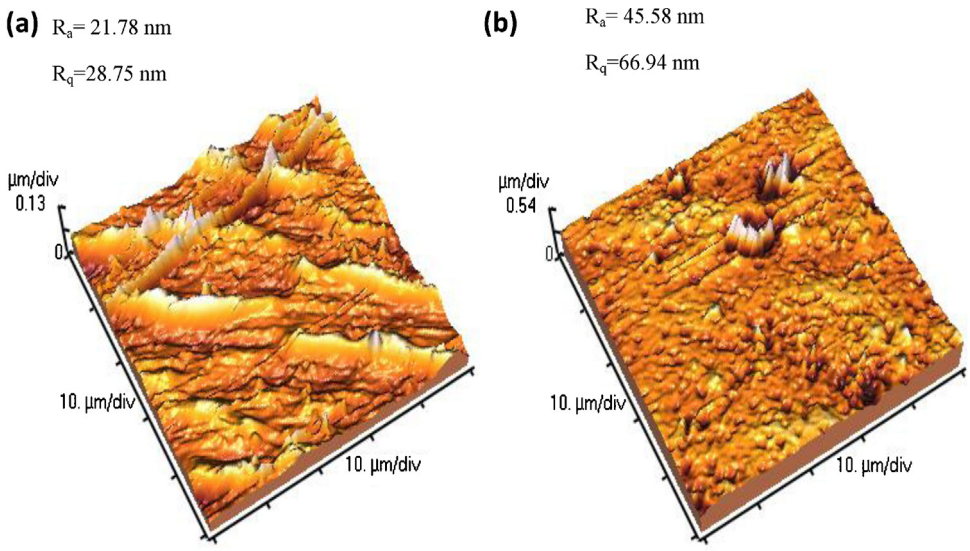

Fig. 4. AFM topographic images of soluble soybean polysaccharide films; (a) unplasticized and (b) plasticized with 30% (w/w) glycerol.

4. Conclusion

This is the first report on the feasibility of SSPS to form biodegradable edible film. The results of the present study showed that SSPS could be a very interesting raw material for the preparation of edible films and coatings. It was clearly demonstrated in this study that the use of different concentrations of glycerol affected physical, mechanical, optical and barrier properties of all SSPS-based films. SSPS films with a glycerol concentration of 20% generally possessed the lowest WVP values, and may have some commercial potential for food-packaging applications. Our results clearly showed that glycerol content in film formulations had dramatic effects on various properties of the SSPS films; these effects should be considered carefully when used in film formation. Further studies should be performed to modify SSPS films' properties, such as improvement of WVP by lipid addition.

Acknowledgements

The authors are thankful to Fuji Oil Co. for supplying raw material for this research. The financial assistance provided by the Department of Food Science and Technology, Gorgan University of Agricultural Sciences is gratefully acknowledged. The authors also wish to thank the Department of Food Science, Engineering and Technology, University of Tehran for the generous use of its facilities.

References

Abdorreza, M. N., Cheng, L. H., & Karim, A. A. (2011). Effects of plasticizers on thermal properties and heat sealability of sago starch films. *Food Hydrocolloids*, *25*, 56–60.

Ahmadi, R., Kalbasi-Ashtari, A., Oromiehie, A., Yarmand, M. S., & Jahandideh, F. (2012). Development and characterization of a novel biodegradable edible film obtained from psyllium seed (*Plantago ovata Forsk*). *Journal of Food Engineering*, *109*, 745–751.

Arvanitoyannis, I., Psomiadou, E., Nakayama, A., Aiba, S., & Yamamoto, N. (1997). Edible films made from gelatin, soluble starch and polyols. Part 3. *Food Chemistry*, *60*(4), 593–604.

Asai, I., Watari, Y., Iida, H., Masutake, K., Ochi, T., Ohashi, S., et al. (1994). Effect of soluble soybean polysaccharide on dispersion stability of acidified milk protein. In K. Nishinari, & E. Doi (Eds.), *Food hydrocolloids: Structure, properties and functions* (pp. 151–156). New York: Plenum Press.

ASME B46.1. (2009). *Surface texture (surface roughness, waviness, and lay). An American National Standard*. New York: ASME.

ASTM. (2012a). *Standard test methods for water vapor transmission of material, E96/E96M. Annual book of ASTM*. Philadelphia, PA: American Society for Testing and Materials.

ASTM. (2012b). *Standard test method for tensile properties of thin plastic sheeting. Standard D882. Annual book of ASTM*. Philadelphia, PA: American Society for Testing and Materials.

Averous, L., & Boquillon, N. (2004). Biocomposites based on plasticized starch: Thermal and mechanical behaviors. *Carbohydrate Polymers*, *56*, 111–122.

Bolin, H. R., & Huxsoll, C. C. (1991). Control of minimally processed carrot (*Dascus carota*) surface discoloration caused by abrasion peeling. *Journal of Food Science*, *56*, 416–418.

Carvalho, R. A., & Grosso, C. R. F. (2004). Characterization of gelatin based films modified with transglutaminase, glyoxal and formaldehyde. *Food Hydrocolloids*, *18*, 717–726.

Cuq, B., Gontard, N., Cuq, J. L., & Guilbert, S. (1997). Select functional properties of fish myofibrilar protein-based films as affect by hydrophilic plasticizer. *Journal of Agricultural and Food Chemistry*, *45*(3), 622–626.

Debeaufort, F., Quezada-Gallo, J. A., & Voilley, A. (1998). Edible films and coatings: Tomorrow's packagings: A review. *Critical Reviews in Food Science and Nutrition*, *38*(4), 299–313.

Furuta, H., & Maeda, H. (1999). Rheological properties of water-soluble soybean polysaccharides extracted under weak acidic condition. *Food Hydrocolloids*, *13*, 267–274.

Gennadios, A., Weller, C. L., & Testin, R. F. (1993). Temperature effect on oxygen permeability of edible protein-based films. *Journal of Food Science*, *58*, 212–214.

Ghanbarzadeh, B., & Almasi, H. (2011). Physical properties of edible emulsified films based on carboxymethyl cellulose and oleic acid. *International Journal of Biological Macromolecules*, *48*(1), 44–49.

Ghanbarzadeh, B., Almasi, H., & Entezami, A. A. (2010). Physical properties of edible modified starch/carboxymethyl cellulose films. *Innovative Food Science and Emerging Technologies*, *11*(4), 697–702.

Ghasemlou, M., Khodaiyan, F., & Oromiehie, A. (2011a). Physical, mechanical, barrier, and thermal properties of polyol-plasticized biodegradable edible film made from kefiran. *Carbohydrate Polymers*, *84*, 477–483.

Ghasemlou, M., Khodaiyan, F., & Oromiehie, A. (2011b). Rheological and structural characterisation of film-forming solutions and biodegradable edible film made from kefiran as affected by various plasticizer types. *International Journal of Biological Macromolecules*, *49*, 814–821.

Ghasemlou, M., Khodaiyan, F., Oromiehie, A., & Yarmand, M. S. (2011). Development and characterization of a new biodegradable edible film made from kefiran, an exopolysaccharide obtained from kefir grains. *Food Chemistry*, *127*, 1496–1502.

Gontard, N., Duchez, C., Cuq, B., & Guilbert, S. (1994). Edible composite films of wheat gluten and lipids: Water vapour permeability and other physical properties. *Food Science and Technology*, *29*, 39–50.

Hosseini, S. F., Rezaei, M., Zandi, M., & Ghavi, F. F. (2013). Preparation and functional properties of fish gelatin–chitosan blend edible films. *Food Chemistry*, *136*, 1490–1495.

Janjarasskul, T., & Krochta, J. M. (2010). Edible packaging materials. *Annual Review of Food Science and Technology*, *1*, 415–448.

Lourdin, D., Coignard, L., Bizot, H., & Colonna, P. (1997). Influence of equilibrium relative humidity and plasticizer concentration on the water content and glass transition of starch materials. *Polymer*, *38*, 5401–5406.

Maeda, H. (2000). Soluble soybean polysaccharide. In G. O. Phillips, & P. A. Williams (Eds.), *Handbook of hydrocolloids* (pp. 309–319). Boca Raton, Fla: CRC Press.

Mali, S., Sakanaka, L. S., Yamashita, F., & Grossmann, M. V. E. (2005). Water sorption and mechanical properties of cassava starch films and their relation to plasticizing effect. *Carbohydrate Polymers*, *60*, 283–289.

Nakamura, A., Furuta, H., Maeda, H., Takao, T., & Nagamatsu, N. (2002). Structural studies by stepwise enzymatic degradation of the main backbone

of soybean soluble polysaccharides consisting of galacturonan and rhamnogalacturonan. *Bioscience, Biotechnology, and Biochemistry*, *66*, 1301–1313.

Shellhammer, T. H., & Krochta, J. M. (1997). Whey protein emulsion film performance as affected by lipid type amount. *Journal of Food Science*, *62*(2), 390–394.

Shojaee-Aliabadi, S., Hosseini, H., Mohammadifar, M. A., Mohammadi, A., Ghasemlou, M., Ojagh, S. M., et al. (2013). Characterization of antioxidant–antimicrobial κ-carrageenan films containing *Satureja hortensis* essential oil. *International Journal of Biological Macromolecules*, *52*(1), 116–124.

Shorey, R. L., Willis, R. A., Lo, G. S., & Steinke, F. H. (1985). Effect of soybean polysaccharides on plasma lipids. *Journal of the American Dietetic Association*, *85*, 1460–1465.

Smith, S. A. (1986). Polyethylene, low density. In M. Bakker (Ed.), *The Wiley encyclopedia of packaging technology* (pp. 514–523). New York: John Wiley and Sons.

Sobral, P. J. A., Menegalli, F. C., Hubinger, M. D., & Roques, M. A. (2001). Mechanical, water vapor barrier and thermal properties of gelatin based edible films. *Food Hydrocolloids*, *15*, 423–432.

Sothornvit, R., & Krochta, J. M. (2001). Plasticizer effect on mechanical properties of β lactoglobulin films. *Journal of Food Engineering*, *50*, 149–155.

Stuchell, Y. M., & Krochta, J. M. (1994). Enzymatic treatments and thermal effects on edible soy protein films. *Journal of Food Science*, *59*(6), 1332–1337.

Tsai, A. C., Vinik, A. I., Lasichak, A., & Lo, G. S. (1987). Effects of soy polysaccharides on postprandial plasma glucose, insulin, glucagon, pancreatic polypeptide, somatostatin, and triglyceride in obese diabetic patients. *The American Journal of Clinical Nutrition*, *45*, 596–601.

Xiao, Q., Lim, L.-T., & Tong, Q. (2012). Properties of pullulan-based blend films as affected by alginate content and relative humidity. *Carbohydrate Polymers*, *87*, 227–234.