Thermal Properties, Heat Sealability and Seal Attributes of Whey Protein Isolate/Lipid Emulsion Edible Films

S-J. Kim and Z. Ustunol

ABSTRACT: From 5% w/v whey protein isolate (WPI), whey protein/lipid emulsion edible films were produced that were sorbitol- or glycerol-plasticized, containing butterfat (0.2% w/v) or candelilla wax (0.8% w/v). Thermal properties of the films determined by Differential Scanning Calorimetry (DSC) showed onset temperatures ($T_o$) of 126 to 127 °C for sorbitol- and 108 to 122 °C for glycerol-plasticized films. $T_o$ values were used as the basis for heat sealing temperatures. Temperature (110, 120, 130 °C), pressure (296, 445 kPa), and dwell time (1, 3 s) affected seal strength. Optimum heat sealing temperature was 130 °C for sorbitol- and 110 °C for glycerol-plasticized films. All films were heat sealable with an impulse heat-sealer. Electron Spectroscopy for Chemical Analysis (ESCA) of the surfaces of both sealed and unsealed films showed increase in hydrogen and covalent bonds involving C–O–H and N–C, which may be the main forces responsible for the sealed joint formation of the films.

Keywords: whey, protein, films, heat, sealability

Introduction

Edible packaging materials provide new and unique opportunities for food processing and product development. Over the years, various applications of edible films have been proposed, but none have been investigated extensively. Some proposed applications of edible films include pouches or sachets to package dry ingredients, such as beverage mixes (Debeaufort and others 1998), or ‘ingredient delivery systems’ to deliver premeasured ingredients during processing to prevent human error in weighing and handling. In the manufacturing of pouches, sachets, or ‘ingredient delivery systems’, sealability of the material and the formation of an adequate seal are important. The seal must have sufficient strength to hold the product in the package and not release its contents during handling or storage.

In the packaging industry, heat sealing is widely used to join polymer films (Dodin 1981; Theller 1989; Meka and Stehling 1994; Mueller and others 1998). During the heat sealing process, 2 films are pressed together between heated plates or dies. The surface of the crystalline polymer melts, due to heat. The application of pressure results in the interfacial interactions across the joint surfaces, which require time. This is a necessary step to give sufficient seal strength to the sealed film. Upon cooling, a heat-sealed joint is produced due to recrystallization of the polymer. The joint formation on the polymer surface is dependent on the surface chemistry of the material (Allen 1997). Temperature, pressure, and dwell time are considered important process variables which affect seal strength (Theller 1989). Measurement of seal strength is typically used as an indicator of seal quality (ASTM 1997). Heat-sealing process variables, and testing for seal properties, have been reviewed extensively by Dodin (1981) and Theller (1989).

Electron Spectroscopy for Chemical Analysis (ESCA) is useful for qualitative and quantitative characterization of materials’ surfaces (or near-surfaces) (Briggs and Seah 1990; Cayless 1991). Lee (1994) used ESCA to study the surfaces of polyimide films and reported functional groups such as hydroxyl (OH), aldehyde (CHO), and carboxylic acid (COOH) to be present on the surface. Modifications of these polar functional groups were responsible for the adhesion strength differences of the films. Wu and others (1995) reported enhanced seal strength of low-density polyethylene and high-density polyethylene films upon ammonia plasma treatment. ESCA revealed that the enhancement in seal strength was due to enhanced interactions between nitrogen- and oxygen–Containing functional groups. Possart and Dieckhoff (1999) employed ESCA to study surfaces of polycyanurates to determine the groups capable of interfacial interaction, and reported hydrogen bonds involving OH groups to be responsible for the interactions at the interfacial region. To our knowledge, however, the heat-sealing mechanism of protein-based edible films is not known, and thus far ESCA has not been used to study surfaces of biodegradable and/or edible biopolymer materials.

Until now, research on protein-based edible films has focused on their formation, testing for their barrier and mechanical properties, and improving on their properties (Banerjee and Chen 1995; Gennadios and others 1996; Rayas and others 1997; Frinault and others 1997; Miller and Krochta 1997; Lim and others 1999; Perez-Gago and Krochta 1999). Very little information is available on these films’ thermal properties (Cherian and others 1995; Cuq and others 1997; Frinault and others 1997; Miller and Krochta 1997; Lim and others 1999; Perez-Gago and Krochta 1999). But there is no information available on heat sealability, seal properties, and mechanism of seal formation in whey protein-based edible film. We hypothesize that the whey protein isolate/lipid emulsion edible films are in fact heat sealable, and thus similar to other polymers, if the sealing temperature and conditions are optimized. We further hypothesize that the sealed joint formation is not merely due to the melting and solidification of the lipid in the film. Therefore, the purpose of this research was to determine the thermal properties of whey protein isolate/lipid emulsion edible films in order to optimize their sealing conditions. Effects of temperature, pres-
sure, and dwell time on seal properties was determined, and mechanism of sealed joint formation was explored.

Materials and Methods

Materials

WPI, or whey protein isolate (ALACEN 895) was obtained from New Zealand Milk Products (North America) Inc., Santa Rosa, Calif., U.S.A.). Glycerol was purchased from J.T. Baker Co. of Phillipsburg, N.J., U.S.A.). Candelilla wax was purchased from Strahl and Pittsch Inc. (West Babylon, N.Y., U.S.A.), and unsalted butter was obtained from Land O'Lakes Inc. (Arden Hills, Minn., U.S.A.). D-sorbitol was obtained from Sigma Chemical Co. (St. Louis, Mo., U.S.A.), and the NaOH came from Mallinckrodt Specialty Chemical Co. (Par- ky, U.S.A.).

Film preparation

WPI (5% w/v) and sorbitol (5.0, 4.8, or 4.2 % w/v) or glycerol (3.5, 3.3, or 2.7 % w/v) were mixed in distilled water, and the pH adjusted to 8 with 2N NaOH. These concentrations of film-forming components were selected because they provided for the optimum freestanding films. Solutions were heated at 90 ± 2 °C for 15 min, while being stirred continuously. BF; or butterfat (0.2% w/v based on the solids content of the butter), or candelilla wax (CW at 0.8% w/v) were added during heating and allowed to melt into the solutions, so as to provide a solids content of 10% w/v for sorbitol- and 8.5% w/v for glycerol-plasticized film-forming solutions. The solutions were homogenized for 2 min using a Polytron PT 10/35 homogenizer with a PTA- 20 TS homogenizing head at a setting of 5 (Tekmar Co., Cincinnati, Ohio, U.S.A.). The solutions were filtered through a layer of cheese cloth (to ensure the complete mixing of the lipid components), and vacuum-degassed for 30 min, then cast on 18.5-cm circular Teflon™ surfaces.

The films were dried at room temperature 23 ± 2 °C and 30 ± 5% RH for 18 ± 3 h. Dried films were peeled and stored at 23 ± 2 °C and 50 ± 5% RH until tested.

Film thickness

Film thickness was determined using a TMI Model 549M micrometer (Testing Machines Inc., Amitville, N.Y., U.S.A.). Measurements were taken at 5 locations. Mean thickness of sorbitol-plasticized films was 140 ± 19 μm and glycerol-plasticized films was 120 ± 15 μm.

DSC analysis

Diffuse Scattering Calorimetry (DSC) was used to determine the thermal transition temperatures of the film-forming components and the films. The instrument was calibrated using pure indium (melting point 156.4 °C). Film-forming components were conditioned in similar manner to the films as described above (23 ± 2 °C and 50 ± 5% RH). Ten mg of sample was weighed and sealed in an aluminum sample pan (TA Instruments, Newcastle, Del., U.S.A.), using an encapsulating press. Samples were heated from 0 to 250 °C at a rate of 20 °C/min. An empty sample pan was used as a reference. During data collection, the DSC cell was flushed with nitrogen at 20 ml/min to maintain an inert environment. A Du Pont 2920 DSC unit, equipped with General V4.1 C software program (Wilmington, Del., U.S.A.) was used to measure the differential temperature and enthalpy change (ΔH). Onset (T₀) and peak (Tₚ) temperatures were assigned according to Standard D-3418 (ASTM 1997).

Seal strength determination

Film samples were cut into strips of 7.62 x 2.54 cm, using a Precision Sample Cutter (Thawing-Albert Instrument Co., Philadelphia, Penn., U.S.A.). Two film strips were placed on top of one another, and an area of 2.54 x 1.5 cm (at the edge of the film) was heat-sealed at 110, 120, or 130 °C for 1 or 3 s of dwell time at 296 or 445 kPa pressure, using a thermal heat-sealer Model-12ASL (Sencorp System Inc., Hyannis, Mass., U.S.A.). All sealed film samples were conditioned for 48 h under the test conditions prior to determining seal strength. Seal strength of the heat-sealed films was determined according to Standard ASTM F-88 (ASTM 1997), using an Instron Universal Testing Machine Model 2401 (Instron Corp., Canton, Mass., U.S.A.), at 23 ± 2 °C and 50 ± 5% RH. Each leg of the sealed film was clamped to the machine, with each end of the sealed film held perpendicularly to the direction of the pull. The distance between the clamps was 5.08 cm. A 1-kN static load cell and crosshead speed of 50.8 cm/min were used. Seal strength was calculated as follows: Seal strength—peak force/film width. The maximum force required to cause seal failure was reported as seal strength in newtons/meter (N/m).

Surface analysis by ESCA

Whey protein isolate/lipid emulsion edible films were heat sealed, as described above (110 °C, 296 kPa, 1s). ESCA (PHI 5400 ESCA lab workstation, Physical Electronic, Eden Prairie, Minn., U.S.A.) was used for surface analysis, surface component determination, and bonding distributions of sealed and unsealed. A circular film of 15 mm dia was placed in the sample holder. Monochromatic x-rays were used as the radiation source. All spectra were collected using an Mg anode operated at a power of 300 W, with an analyzer pass energy of 33 eV. The optimum spot size for the conditions used was 1 mm dia aperture. Intensity of the emitted photoelectrons (according to their binding energies) was plotted with an electron kinetic energy analyzer (Physical Electronic, Eden Prairie, Minn.). No compensation for differential surface charging was needed, due to the shape of the spectra. The bonding scale was calibrated to 284.6 eV for the main carbon 1s (C–H) feature. Both low-resolution (survey scan) and high-resolution modes for the carbon 1s’ (C1s), oxygen 1s’ (O1s), and nitrogen 1s’ (N1s) regions were run. Chemical changes on the surface of the polymers were elucidated by curve-fitting the C1s, N1s and O1s spectra. (Curve-fitting defines and interprets carbon chemistry as detected at the sample surface by allowing the user to distinguish overlapping features within the spectral envelope). The spectra were fit with a Lorentzian-Gaussian mix Voigt profile function, using a nonlinear, least-square curve-fitting program, PHI PC Explorer Software from Physical Electronic, (Eden Prairie, Minn., U.S.A.).

Statistical analysis

Seal strength experiments were replicated 3 times in a randomized, complete block experiment. Days were blocked. A new film-forming solution and new set of films were prepared for each replicate. Statistical analysis was conducted using Sigma Stat 2.0 (Jandel Corp., San Rafael, Ca- lif., U.S.A.). Treatment means were compared using the Student-Newman-Keuls comparison. Comparisons were made only within the same film. Significance of differences was defined at p ≤ 0.05. ESCA and DSC analyses were conducted in duplicate on 1 replicate set of films. Representative data were presented.
Thermal properties
Whey protein isolate contained 93.5% protein and <1% fat and lactose (data provided by New Zealand Milk Products (North America) Inc.). Composition of WPI was confirmed using Association of Official Analytical Chemists (AOAC 1990) procedures.

Figure 1 shows the DSC thermograms of the individual film-forming components used in the production of whey protein/lipid emulsion edible films. WPI powder exhibited a broad endothermic peak of first-order transition between 125 to 173 °C similar to the distinctive melting transition characteristic of semicrystalline polymers (Rosen 1982), suggesting that WPI may be a partially crystalline amorphous (semicrystalline) polymer. WPI had an additional peak at 241 °C, due to the degradation of the protein at this temperature. Sorbitol showed a narrow endothermic peak (96 to 106 °C) and a Tp at 101 °C, corresponding to the melting temperature of sorbitol (Budavari and others 1989). Glycerol showed a broad endothermic peak at 165 to 220 °C, with a Tp of 178 °C, which was consistent with the degradation temperature of 182.2 °C reported by Budavari and others (1989). Butterfat showed 2 low Tps at 10 and 30 °C, corresponding to the low and high melting triacylglycerol fractions of butterfat (Kaylegian and others 1993). A third distinctive peak at 101 °C probably was due to decomposition of the triacylglycerol. On the other hand, candelilla wax showed a Tp at 68 °C, consistent with its melting temperature (Bennett 1975).

Figure 2 and 3 show the DSC thermograms of WPI/lipid emulsion edible films plasticized with sorbitol and glycerol, respectively. Table 1 summarizes the transition temperatures (To and Tp) of these films. All films showed broad endothermic peaks at 108 to 221 °C. The thermograms of the films were distinctly different from the individual components. This is due to the influences of each component on the thermal properties of other components, and thus the film as a whole. The To values of glycerol-plasticized films (108 to 122 °C) were lower than those of the sorbitol-plasticized films (126 to 127 °C), probably due to the differences in the plasticizing effects of the 2 plasticizers. Functional efficacy of a plasticizer is often estimated by its reduction in the thermal transition temperature of a polymer (Karlsson and Singh 1998; Wu and McGinity 1999). The transition temperature of a polymer decreases with decreasing molecular size (Gutierrez-Rocca and McGinity 1994; Galietta and others 1998). These reports are consistent with our study's results on WPI films. Overall, glycerol (92.02 daltons) was more effective than sorbitol (182.17 daltons) in lowering the transition temperature of the films. However, these observed differences in transition temperatures may also be due to the plasticizing effect of water (Fennema 1996). Glycerol-plasticized films had higher moisture contents than sorbitol-plasticized films (data not shown).

WPI films containing butterfat or candelilla wax showed similar trends to WPI films without lipids, except that candelilla wax-containing films also had a more definite narrow endothermic peak at 66 °C, which corresponded to the melting temperature of candelilla wax (Figure 1). All films showed multiple peaks at 175 to 212 °C, due to the degradation of the films. Thermal degradation of polymers results in breaking

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Transition temperature (°C)</th>
<th>Heat flow (J/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>WPI-S</td>
<td>126 143</td>
<td>15.6</td>
</tr>
<tr>
<td>WPI-G</td>
<td>108 145</td>
<td>169.9</td>
</tr>
<tr>
<td>WPI-S-BF</td>
<td>127 160</td>
<td>202.9</td>
</tr>
<tr>
<td>WPI-G-BF</td>
<td>122 132</td>
<td>31.8</td>
</tr>
<tr>
<td>WPI-S-CW</td>
<td>127 135</td>
<td>84.0</td>
</tr>
<tr>
<td>WPI-G-CW</td>
<td>116 142</td>
<td>208.0</td>
</tr>
</tbody>
</table>

1WPI = whey protein isolate; S = sorbitol; G = glycerol; BF = butterfat; CW = candelilla wax
2To = onset transition temperature; Tp = peak transition temperature

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of bonds by heat in the absence of oxygen. As temperature increases, chemical bonds with low-energy values will be broken first, and the more thermally stable bonds will resist thermal degradation and require higher energies to dissociate them. Thus, degradation of multicomponent materials results in multiple peaks (Throne 1986; Hernandez 1997).

**Seal strength**

Seal sealing of the films was conducted near the T_g of the films as determined above, because thermal transition temperatures are typically used in determining sealing temperatures of other polymers (Hernandez 1997). Films were heat-sealed on the nonlipid-oriented side, because the lipid-oriented side did not seal, or formed seals that easily delaminated. All films were heat-sealable. Table 2 and 3 show the seal strength measurements of sorbitol- and glycerol-plasticized films, respectively. Seal strength of sorbitol-plasticized films ranged from 105 to 301 N/m and glycerol-plasticized films ranged from 141 to 323 N/m. Heat sealing temperature had a significant influence (p < 0.05) on seal strength, whereas pressure variation did not affect seal strength significantly. Increase in dwell time increased seal strength; however, these increases were not significant in all cases. Highest (p < 0.05) seal strength was observed at 130 °C for sorbitol-plasticized films and 110 °C for the glycerol-plasticized, which corresponds with the T_g of these films, as determined by DSC. Similar to other polymers, thermal transition temperature may be useful in determining thermal processing temperatures of protein-based films. Lower (p < 0.05) seal strength of glycerol-plasticized films at 130 °C was due to excessive heat treatment, which resulted in distorted and weakened seals. If the heat required to produce a seal exceeds the heat-sealing temperature range for that material, it induces a distorted or nonfunctional seal (Martin 1986). Deformation of the seal structure was visible with glycerol-plasticized films heat sealed at 130 °C.

The optimum seal strengths obtained in this study, 301 and 323 N/m for sorbitol- and glycerol-plasticized films, respectively, were lower than seal strengths of heat-sealed synthetic polymers (> 730 N/m) (Martin 1986). However, our films were comparable in seal strength to carrageenan- (Ninomiya and others 1990) and lactic acid casein- (Chick 1998) based edible films. Ninomiya and others (1990) and lactic acid casein-based edible films were comparable in seal strength to carrageenan- (Ninomiya and others 1990) and lactic acid casein-based edible films. Ninomiya and others (1990) reported the seal strength of heat-sealed carrageenan-based edible films plasticized with glycerol and sorbitol to be 137 and 130 N/m, respectively. Chick (1998) reported that the seal strength of lactic-acid-casein-based films, plasticized with sorbitol, sealed at 107 to 120 °C to range 153 to 247 N/m. However, these are very general comparisons that should be made with caution, since compositions of the films, sealing, and testing conditions vary significantly among these studies.

**Surface analysis of heat-sealed and unsealed films**

Figure 4 shows the surface components of whey protein isolate/lipid emulsion edible films before and after heat sealing, as determined by ESCA. The C1s' spectra of the films were composed of 4 components: C–H (at 284.6 eV), C–O (286.3 eV), O=C (287.6 eV), and O–C=O (289.5 eV), the primary components being C–H and C–O. However, comparisons of the C1s' spectra showed no differences in the relative intensities of different carbon components between the un-
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Table 4—Surface components (%) of whey protein isolate/lipid emulsion edible films before and after heat sealing as determined by Electron Spectroscopy for Chemical Analysis

<table>
<thead>
<tr>
<th>Binding energy</th>
<th>O=C 532.3eV</th>
<th>O=C–O–H 534.3 eV</th>
<th>N=C 401.8eV</th>
<th>N–C 399.7 eV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Treatments</td>
<td>Unsealed</td>
<td>Sealed</td>
<td>Unsealed</td>
<td>Sealed</td>
</tr>
<tr>
<td>WPI-S</td>
<td>48.7</td>
<td>44.9</td>
<td>44.1</td>
<td>47.8</td>
</tr>
<tr>
<td>WPI-G</td>
<td>47.8</td>
<td>47.1</td>
<td>41.3</td>
<td>44.2</td>
</tr>
<tr>
<td>WPI-S-BF</td>
<td>47.9</td>
<td>46.4</td>
<td>40.7</td>
<td>49.5</td>
</tr>
<tr>
<td>WPI-G-BF</td>
<td>46.3</td>
<td>46.2</td>
<td>41.2</td>
<td>45.7</td>
</tr>
<tr>
<td>WPI-S-CW</td>
<td>44.8</td>
<td>44.6</td>
<td>43.8</td>
<td>47.4</td>
</tr>
<tr>
<td>WPI-G-CW</td>
<td>48.5</td>
<td>46.8</td>
<td>38.8</td>
<td>43.2</td>
</tr>
</tbody>
</table>

1 WPI = whey protein isolate; S = sorbitol; G = glycerol; BF = butterfat; CW = candelilla wax
2 nd = not detectable

Although not conclusive, ESCA results suggest that formation of O=C–O–H and N=C bonds upon heat sealing may be responsible for the mechanism of seal formation. Figure 5 shows a proposed model for C–O–H and N=C bonding upon heat sealing of whey protein isolate/lipid emulsion edible films. Polar polymers (such as proteins) associate by high degree of hydrogen bonding. Hydrogen bonding could also occur between protein and plasticizer, as well as between the plasticizer molecules, (that is, glycerol as in Figure 5a, or sorbitol). Plasticizers are typically added to films to reduce hydrogen bonding between polymer chains by competing for polymer interactions, thus enhancing flexibility. Plasticizers such as glycerol also have been reported as heat-sealing promoters (Georgevits 1967). Hydrogen bonding could also occur between the plasticizer (for instance, glycerol) and protein, such as COOH side-groups of amino acids like aspartic acid (Figure 5b), or glutamic acid. β-lactoglobulin and α-lactalbumin are the 2 most abundant whey proteins. Aspartic and glutamic acids are the most abundant amino acids in β-lactoglobulin (11 and 16%, respectively) and α-lactalbumin (9 and 8%, respectively) (Swaisgood 1996). Thus, these were selected for the proposed model. An example for hydrogen bonding among proteins is illustrated in Figure 5c. Covalent bond formation due to heat sealing could occur between, for example, ε-NH₂ group of lysine and carboxyl side-group of asparagine (Figure 5d) or glutamine. Lysine is also fairly abundant in whey proteins, making up 15% of the amino acids in β-lactoglobulin and 12% of α-lactalbumin (Swaisgood 1996). Thus, it was selected for illustration in our proposed models. This is a simple model, and although not illustrated in this study, other interactions are likely.

Conclusions

In heat processing of protein–based edible films, an important limitation is the lack of information on these mate-

Figure 5—A proposed model for C–O–H and N=C bonding upon heat sealing of whey protein isolate/lipid emulsion edible films; (a) plasticizer-plasticizer, (b) plasticizer-protein, (c) protein-protein, (d) protein-protein (that is, lysine-asparagine)
rials’ thermal properties. Thus, exploring the thermal properties of protein-based edible films is important in the commercial development of edible packaging materials. DSC is a useful tool in determining heat-sealing temperatures and degradations of whey protein isolate lipid/emulsion films, and may be useful in obtaining other information on their heat-processing parameters. ESCA is also useful in studying the mechanism of heat-seal formation. However, further studies are needed with other protein-based materials.

References


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Authors S-J. Kim and Z. Ustunol are with Michigan State University’s Department of Food Science and Human Nutrition, East Lansing, MI 48824-1224. Direct inquiries to author Ustunol (E-mail: ustunol@msu.edu).