Water Vapor Permeability and Mechanical Properties of Grain Protein-Based Films as Effected by Mixtures of Polyethylene Glycol amd Glycerin Plasticizers

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Abstract. *Grain protein-based films containing mixtures of glycerin and polyethylene glycol (PEG) as plasticizers were prepared and evaluated for water vapor permeability (WVP), tensile strength (TS), and elongation (E). Changes in mechanical properties during storage were also studied. The PEG produced opposite trends in E of wheat gluten and corn-zein films. The TS of wheat gluten films increased and E of the films decreased as the ratio of glycerin/PEG decreased. Corn-zein films containing only glycerin were very brittle (E of 4%), and E of the films improved to 94% when the ratio of mL PEG/g protein was 0.39. The WVP of both films decreased as the ratio of glycerin/PEG was decreased. Also, WVP of both films increased as the total amount of plasticizer added to the films increased. Mixtures of glycerin and PEG as plasticizer are less fugitive than glycerin alone in grain protein-based films and can reduce the deterioration of mechanical properties during storage.* ***Keywords.*** *Plasticizers, Corn-zein, Wheat gluten.*

Protein-based films have been investigated for potential applications as wrapping and coating materials for food products. Protein-based films have been made from grain proteins (cereals and oilseeds) and animal proteins (casein, gelatin, keratin, etc.) (Guilbert, 1986). The development history, property evaluations, and application trials of protein-based films have been reviewed by Kester and Fennema (1986), Guilbert (1986), Gennadios and Weller (1990) and Park (1991).

Protein-based films and coatings have been used as gas and grease barriers and for mechanical protection in several food systems as a means of extending shelf life and minimizing quality changes during marketing (Aydt et al., 1991; Gennadios et al., 1993b; Guilbert, 1986; Park and Chinnan, 1993; Park et al., 1992). Soybean films such as "Yuba" in Japan, "Tou-Fu-Pi" in China, and "Fu Chok" in Malaysia have been used traditionally as edible wraps for containing meat and vegetable mixes which are fried in oil (Snyder and Kwon, 1987). Cosler (1958) used corn-zein as an oxygen barrier coating to prevent oxidation in confectionery products. Park et al. (1993a) reported that corn-zein provides a good oxygen barrier on tomatoes, resulting in doubling of their shelf life. Trezza and Vergano (1993) found zein-coated paper to have a high potential as a wrapping material in quick service restaurants. Krochta et al. (1988) developed casein coatings which can be used for precut fruits and vegetables to extend the marketing period.

Barrier and mechanical properties of protein-based films are affected by factors such as concentration and selection of plasticizer, pH of solvent, salts, and other additives. Gennadios et al. (1993a) found that the addition of mineral oil and keratin to film-forming solutions reduced water vapor permeability in protein-based films. Gennadios et al. (1992) also reported that the tensile strength of corn zein and wheat gluten films varied significantly with conditioning at different combinations of relative humidity (23 to 75%) and temperature (5 to 45° C). Gontard et al. (1992) reported that pH and ethanol concentration had strong interactive effects on film opacity, water solubility, and water vapor permeability. The concentration of plasticizer greatly affects mechanical properties of grain protein-based films. Flexibility is improved, but puncture strength and water vapor and oxygen barrier properties are decreased as plasticizer concentration is increased in wheat gluten films (Gontard et al., 1993; Park et al., 1992).

In these studies, only one plasticizer, glycerin, was used in the protein-based films. Even glycerin that is well dispersed in protein film solutions migrates from the bulk to the surface of the film matrix because of binding limitations between protein molecules and glycerin. Excess glycerin can migrate through the film matrix. The migration speed of glycerin depends on the types of functional groups, polarity, and structure of the film matrix. For example, the surface of new corn zein films is transparent, but it has a greasy appearance within a few hours because of glycerin migration to the surface. The loss of flexibility of the film is an another indicator of glycerin migration to the surface (Park et al., 1992). Also, glycerin could migrate slowly from the film matrix of wheat gluten during storage if the protein molecules of wheat gluten

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have a limited capability for binding with glycerin. The lowering of elongation values of wheat gluten film during storage is an indicator of glycerin migration. The structure and molecular size of glycerin (MW, 92) allows more rapid diffusion than polyethylene glycol (PEG) (MW, 400) because PEG as a plasticizer has longer carbon chains (number of carbons in chain ≈ 16) than glycerin (number of carbons in chain = 3). Loss of flexibility in protein-based films during storage is a major limitation for their use as a wrap or coating for food products. However, no evidence has been found of attempts to use other plasticizers to overcome loss of elongation properties in protein-based films.

The objectives of this study were to:

- Determine the effect of glycerin and PEG mixtures as plasticizers on water vapor permeability and mechanical properties (tensile strength and elongation) of grain protein-based films.
- Evaluate the effectiveness of PEG and glycerin mixtures in reducing elongation during storage.

Materials and Methods

Materials

The following materials were used to prepare the grain protein-based films: wheat gluten protein (ADM Arkady, Olathe, Kans.); corn-zein (Freeman, Inc., Tuckahoe, N.Y.); ammonium hydroxide (5N), glycerin (USP grade) (Aldrich Chemical Company, Inc., Milwaukee, Wis.); polyethylene glycol, 400, (NF grade), ethanol, 95% (Fisher Scientific, Pittsburgh, Pa.).

Preparation of Grain Protein-based Films

Wheat gluten-based film solutions were prepared by dissolving 75 g of wheat gluten in a mixed solvent of 360 mL of ethanol (95%), 228 mL of distilled water and 72 mL of ammonium hydroxide (5N). A mixture of glycerin and PEG was then added as plasticizer. Wheat gluten-based film solutions were homogenized at 6,000 rpm for 5 min (Virtis Co., Inc., Gardiner, N.Y.) and were filtered through cheese cloth to remove insoluble carbohydrates (Park et al., 1992). Solutions were conditioned at 85° C for 15 min in a water bath (Versa-Bath S, model 236, Fisher Scientific, Pittsburgh, Pa.) before pouring 70 mL onto a glass plate (28 cm × 28 cm) and then drying at 55° C.

Corn-zein based film solutions were prepared by dissolving 54 g of corn-zein in a solvent of 325 mL of ethanol (95%). A mixture of glycerin and PEG was then added as plasticizer. These solutions were also conditioned at 85° C for 15 min. Then 60 mL was poured onto a glass plate (28 cm × 28 cm) and dried at 55° C.

Both film types were detached from the glass plates and used to prepare samples for determining water vapor permeability and mechanical properties.

Mixture of Glycerin and PEG

Three total amounts of plasticizer for each film type (0.20, 0.28, and 0.36 mL plasticizer/g protein for wheat gluten film solutions; 0.17, 0.28, and 0.39 mL plasticizer/g protein for corn zein film solutions) were studied. These total amounts were used with ratios of glycerin/PEG of 100:0, 75:25, 50:50, 25:75, and 0:100 (v/v).

Thickness Measurement

A hand-held micrometer (B. C. Ames Co., Waltham, Mass.) was used to measure film thickness to an accuracy of 0.000127 cm. Five measurements were made on each test sample, and a mean thickness was calculated.

Mechanical Properties

Twenty specimen samples, 10 × 2.54 cm, were cut from film samples prepared on three glass plates (28 × 28 cm). Samples were conditioned for 48 hr at 25° C and 50% relative humidity (RH) in an environmental chamber (model 317332, Hotpack, Corp., Philadelphia, Pa.) before measuring tensile strength (TS) and elongation (E). Both TS and E of samples were measured as quickly as possible. An Instron (model 4210, Instron Engineering Corp., Canton, Mass.) was used to measure TS and E at break, according to the ASTM standard method D 882-88 (ASTM, 1989). Initial grip separation and crosshead speed were set at 50 mm and 500 mm/min, respectively.

Measurement of Water Vapor Permeability (WVP)

Cups and sealing rings made from poly methylmethacrylate (Piedmont Plastics, Inc., Greenville, S.C.) were used to determine WVP. These cups and rings were originally designed by Dr. J. M. Krochta (Dept. of Food Science and Technology, University of California, Davis) and manufactured in the Department of Mechanical Engineering at Clemson University. Cups consisted of cylindrical bottoms (ID = 4.6 cm, OD = 8.7 cm and depth = 2.1 cm) and sealing rings (ID = 4.6 cm, OD = 8.7 cm and thickness = 0.5 cm). Cups were filled to a depth of 0.9 cm with distilled water and covered with a film to be tested. Cups and sealing rings were tightened with four screws and placed in a chamber conditioned at 25° C, 50% RH (model 317332, Hotpack, Corp., Philadelphia, Pa.). An air flow meter (Datatrak™, model No. 635, Sierra Instrument Co., Carmel Valley, Calif.) was used to measure air flow rate inside the chamber. The air flow rate determined was 177.7 ft/min. Weight change of the cups versus time was measured and plotted. Linear regression was used to calculate the slope of a fitted straight line. The WVP was then calculated as described by Park and Chinnan (1993).

Storage Test

Wheat gluten films and corn-zein films containing 0.36 and 0.39 mL plasticizer/g protein, respectively, were prepared as described above. Ratios of glycerin to PEG of 100:0, 50:50, and 0:100 were used in both types of film. Forty specimens, 10 × 2.54 cm, from each film type were cut and stored in a chamber (25° C and 50% RH). Mechanical properties (TS and E) of 20 specimens from each film type were measured after 10 and 20 days.

Analysis of Data

Duncan's multiple range test was applied to compare means for water vapor permeability and mechanical properties. Tables 1, 2, and 3 show the resulting comparisons with a level of significance of $\alpha = 0.05$ (SAS Institute, Inc., 1985).

Table 1. Effect of plasticizer (P) ratio and concentration on tensile strength (TS) and elongation (E) of grain protein-based films*

	Wheat Gluten Film			Corn-zein Film		
Ratio† (GLY: PEG)	Concentration (mL P/ g protein)	TS (MPa)	E (%)	Concentration (mL P/ g protein)	TS (MPa)	E (%)
100: 0	0.36	4.4a	142a	0.39	8.4a	3a
75: 25	0.36	4.4a	102b	0.39	9.6b	7a
50: 50	0.36	3.5b	103b	0.39	13.4c	76b
25: 75	0.36	5.5c	43c	0.39	8.4a	89c
0: 00	0.36	5.7c	7d	0.39	9.5b	94c
100: 0	0.28	6.4a	68a	0.28	18.1a	3a
75: 20	0.28	7.3b	59b	0.28	20.2b	6b
50: 50	0.28	8.9c	42c	0.28	22.8c	18c
25: 75	0.28	8.9c	16d	0.28	21.8bc	19c
0: 00	0.28	9.1c	5e	0.28	22.1c	23d
100: 0	0.20	13.4a	5a	0.17	26.2a	2a
75: 25	0.20	14.9a	4a	0.17	32.7a	3a
50: 50	0.20	15.1a	4a	0.17	35.5a	7b
25: 75	0.20	15.1a	3b	0.17	30.9a	9c
0: 00	0.20	-	-	0.17	33.2a	10c

* Units are MPa for TS and percent (%) for E; values refer to mean of 20 samples; values at the same concentration in a column with different letter superscripts are significantly different as determined by Duncan's multiple range test; mechanical measurement of wheat gluten films containing 0.20 mL PEG / g protein as plasticizer was not possible because of brittleness.

† Ratio (v/v); GLY (Glycerin) to PEG (polyethylene glycol).

Table 3. Storage time effect on tensile strength (TS) and elongation (E) of grain protein-based edible films

		Mechanical Property*			
		Wheat Gluten Film‡		Corn-zein film‡	
Storage Time (d)	Ratio† (GLY:PEG)	TS (MPa)	E (%)	TS (MPa)	E (%)
0	100: 0	4.4a	142a	8. 4a	3a
		(0.5)	(22)	(1.8)	(1)
	50: 50	3. 5a	103a	13.4a	76a
		(0.4)	(10)	(1.0)	(14)
	0: 100	5.7a	7a	9.5a	94a
		(0.6)	(1)	(1.6)	(27)
10	100: 0	4.9a	104b	8 .6a	3a
		(0.3)	(31)	(2.7)	(1)
	50: 50	3.9b	83b	24.1b	18b
		(0.4)	(18)	(3.7)	(11)
	0: 100	5.9a	5b	15.8b	71b
		(2.5)	(2)	(1.4)	(30)
20	100: 0	5.9b	55c	10.9b	3a
		(2.0)	(36)	(2.9)	(0)
	50: 50	4.3b	79b	25.8b	12b
		(0.8)	(22)	(3.7)	(4)
	0: 100	-	-	16. 6b	66b
		-	-	(2.4)	(37)

* Units are MPa for tensile strength and percent (%) for E; values refer to mean and standard deviation (in parentheses) of 20 samples; TS and E at the same ratio of GLY/PEG in a column with different letter superscripts are significantly different as determined by Duncan's multiple range test.

† Ratio (v/v); GLY (Glycerin) to PEG (polyethylene glycol).

‡ Wheat gluten and corn-zein films contained 0.36 and 0.39 mL of plasticizer/g protein, respectively; mechanical measurement of wheat gluten films containing 100% PEG as plasticizer was not possible after 20 days storage because of brittleness.

Results and Discussion

Mechanical Properties

The effects of the amount of plasticizer added in wheat gluten and corn zein films on mechanical properties are shown in table 1. The TS decreased and E increased as the amount of plasticizer was increased in these grain protein-based films. For example, TS of a wheat gluten film containing 0.36 mL glycerin/g protein was 4.4 MPa and increased to 13.4 MPa as the ratio of glycerin/g protein decreased to 0.20 in the films. These results are similar to results reported by Gontard et al. (1993); puncture strength, one of their mechanical tests, increased as glycerin was decreased in wheat gluten films. For corn-zein films, TS increased and E decreased as the amount of plasticizer was decreased, except for films which contained only glycerin as a plasticizer. The E of corn-zein films which contained only glycerin showed no change as the amount of plasticizer was changed in the film. Corn-zein films containing only glycerin as plasticizer were brittle for all concentrations of glycerin studied, and all exhibited around 3% elongation.

Table 2. Water vapor permeabilities of grain protein-based edible films as affected by plasticizer treatment

	Water Vapor Permeabililty*			
Ratio†	Wheat Gluten Film‡		Corn-zein Film‡	
(GLY:PEG)	A	B	A	B
100: 0	1.11a	0.62a	0.59a	0.42a
	(0.04)	(0.05)	(0.07)	(0.01)
50: 50	0.91a	0.57a	0.53ab	0.41a
	(0.06)	(0.03)	(0.06)	(0.03)
0: 100	0.70b	0.56a	0.41b	0.38a
	(0.12)	(0.01)	(0.00)	(0.02)

* Unit of permeability is in ng·m/m^2·s·Pa; values refer to means (n - 3) and standard deviation (in parentheses); permeabilities in a column with different letter superscripts are significantly different as determined by Duncan's multiple range test.

† Ratio (v/v); GLY (Glycerin) to PEG (polyethylene glycol).

‡ A and B represent 0.36 and 0.28 mL plasticizer/g protein, respectively, in wheat gluten films, and 0.39 and 0.28 mL plasticizer/g protein, respectively, in corn-zein films.

The effects of mixtures of glycerin and PEG as plasticizers on mechanical properties of wheat gluten and corn-zein films are also shown in table 1. The statistically significant differences in mean values, determined by Duncan's multiple range tests, are noted in this table. The TS of wheat gluten films increased and E of the films decreased as the ratio of glycerin/PEG was decreased. Wheat gluten films containing 100% PEG were brittle. The mechanical properties of wheat gluten films with 0.20 mL PEG/g protein could not be measured because of their brittleness. The TS of corn-zein films was highest at the 50:50 ratio of glycerin and PEG, and E increased as the ratio of glycerin/PEG was decreased. The E of corn-zein films containing only glycerin was about 3%, but this increased to 94% for films containing 0.39 mL PEG/p protein.

The PEG produced opposite trends in E of wheat gluten and corn-zein films. The PEG in corn-zein film increased elongation values, whereas PEG in wheat gluten film decreased E values as the ratio of glycerin/PEG decreased. This could be explained by differences in hydrophobicity of the two protein molecules. Average hydrophobicity of a protein can be calculated from its amino acid composition (Fennema, 1985). The average hydrophobicity of corn-zein

protein was calculated to be about 25% higher than that of wheat gluten protein. Based on calculated hydrophobicities of the two proteins, PEG could provide better linkages, through hydrophobic interaction, between corn-zein protein molecules than between wheat gluten molecules.

Water Vapor Barrier

Effects of plasticizer mixtures of glycerin and PEG on water vapor permeabilities (WVP) of wheat gluten and corn-zein films are shown in table 2. The WVP of both wheat gluten and corn-zein films decreased as the ratio of glycerin to PEG was decreased. The PEG is less hydrophilic than glycerin because of its functional groups; therefore, WVPs of films containing only glycerin were expected to be higher than those for 50:50 and 0:100 ratios of glycerin/PEG.

Effects of plasticizer concentration on WVP of wheat gluten and corn-zein films are also shown in table 1. The WVP increased in both films as the concentration of plasticizer increased. The effect of plasticizer concentration on WVP was higher for films containing only glycerin than for films containing 50:50 and 0:100 ratio of glycerin/PEG, for both wheat gluten and corn-zein films. For example, WVP of wheat gluten films containing only glycerin increased by 79%, whereas WVP of the films containing only PEG increased by 25%, as the ratio of mL plasticizer/g protein increased from 0.28 to 0.36 in the films. Similar results were reported by Gontard et al. (1993); water vapor transmission rates of wheat gluten films increased as glycerin concentration increased.

The concentration of plasticizer also affected oxygen permeability (OP) in grain protein-based films. Park et al. (1992) found an increase in OP for protein-based films (wheat gluten and corn-zein) when the concentration of glycerin in the films was increased. In cellulose-based edible films, plasticizer concentration can enhance or retard WVP and OP (Banker et al., 1966; Park et al., 1993b).

Mechanical Properties During Storage

Storage effects of plasticizer mixtures of glycerin and PEG on mechanical properties of wheat gluten and corn-zein films are shown in table 3. In general, TS increased and E decreased in both wheat gluten and corn-zein films during storage. The degree of decrease of E in wheat gluten films containing only glycerin was higher than that of films containing a 50:50 ratio of glycerin/PEG after 20 days storage. For example, E of wheat gluten films containing only glycerin decreased by 61%, whereas E of the films containing a 50:50 ratio of glycerin/PEG decreased only 23% after 20 days in storage. Wheat gluten films containing 100% PEG were brittle and their mechanical properties could not be measured after 20 days storage. The degree of decrease of E in corn-zein films containing a 50:50 ratio of glycerin/PEG was lower than that of films containing only PEG after 20 days storage. The E of corn-zein film containing only glycerin as a plasticizer was too low to allow comparison of storage effects. These results support the general idea that mixtures of glycerin and PEG as plasticizers are less fugitive than glycerin alone in grain protein-based films and can reduce the deterioration of E during storage.

Overall, for optimum physical properties, a 50:50 mixture of PEG and glycerin plasticizers is preferred for use in protein films. For concentrations of plasticizer up to the 36 or 39% levels used in this study, flexibility increased with increasing concentration. This improvement in flexibility is a tradeoff with TS. For both corn-zein and wheat gluten, even though the dependency of elongation on glycerin/PEG ratio differs, the 50:50 mixture provides the best combination of flexibility and strength. In comparison to polymer films currently utilized in food packaging, these films have properties within the commercial range except for their stability with used storage time. Storage tests beyond the 20 days used in this study would certainly be of interest.

Summary and Conclusions

Controlling mechanical properties is critical for commercial use of grain protein-based films. Glycerin has been commonly used in protein-based films as a plasticizer, but it tends to migrate to the film surface. Corn-zein film containing only glycerin is extremely brittle, and its limited flexibility is one of the major restrictions for its commercial application. Improved E properties of corn-zein films would allow their use as individual packaging films for small food products.

The E of corn-zein films containing only glycerin was about 3%, but this increased to 94% for films containing 0.39 mL PEG/g protein. However, E of wheat gluten films decreased as the ratio of glycerin/PEG was decreased, and E of the films containing 100% PEG became brittle. The TS of wheat gluten films increased and E of the films decreased as the ratio of glycerin/PEG decreased. In both films, WVP increased as the concentration of plasticizer was increased. The increases of WVP in both films containing only glycerin were higher than those of films containing 50:50 and 0:100 ratios of glycerin/PEG. The PEG in the mixture of plasticizer had the effect of decreasing reduction of E for both films during storage. For the plasticizer compositions evaluated in this study, the 50:50 glycerin/PEG at the highest concentration (36% in wheat gluten and 39% in corn-zein films) produced the best combination of properties. Although these properties are within the range of commercial interest, the lack of stability over time limits utilization of these films.

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