Plasticization of Soy Protein Polymer by Polyol-Based Plasticizers

Xiaoqun Mo and Xiuzhi Sun*

Department of Grain Science and Industry, Kansas State University, Manhattan, Kansas 66506

ABSTRACT: Soy protein isolate (SPI) was mixed with four polyol-based plasticizers and molded into plastics using a hot press. The plasticized SPI powder was evaluated for denaturation temperatures and denaturation enthalpies. The SPI plastics were studied for mechanical properties, glass transition temperatures, storage modulus, morphology, and water absorption. Thermal properties of the SPI plastics with propylene glycol were depressed to the largest degree, and the plastics with glycerol showed the largest strain at break, whereas plastics with 1,3-butanediol gave the highest tensile strength. The morphology of the fractured surface of the SPI plastics changed from brittle fracture for the unplasticized SPI to ductile fracture for the plasticized SPI. Water absorption of all the plasticized SPI plastics was lower than that of the unplasticized SPI plastics.

Paper no. J9967 in *JAOCS 79,* 197–202 (February 2002).

KEY WORDS: Mechanical properties, polyol plasticizers, soy protein, thermal properties.

Biodegradable polymers have recently attracted much public and industrial interest. The replacement of some petroleum-based plastics with agricultural materials would help preserve fossil-based raw materials, reduce the volume of garbage, reduce release of carbon dioxide into the atmosphere, and add value to agricultural biopolymers (1). Soybean protein-based plastics have been prepared by compression molding (2,3), injection molding (4), and lamination (5). Plastics made solely from soybean proteins, however, are rigid and brittle. Polyols have often been used as plasticizers to improve their processibility, flexibility, and stretchability (6).

Three theories have been proposed to explain the mechanism of the plasticizer effect (7). These theories are: (i) lubricity theory—a plasticizer is considered as a lubricant to facilitate the movements of the macromolecules over each other; (ii) gel theory—a plasticizer disrupts the polymer–polymer interactions including hydrogen bonds and van der Waals and ionic forces; and (iii) free volume theory—a plasticizer may depress the glass transition temperature by increasing polymer free volume. The fundamental concept underlying these theories is that a plasticizer can interpose itself between the polymer chains and decrease the forces holding the chains together (8).

Soybeans consist of a discrete group of proteins that span a broad range of molecular sizes. The predominant structural proteins are the two major globulins, β-conglycinin (7S) and glycinin (11S). 7S Globulin includes a heterogeneous class of glycoproteins composed of varying combinations of three possible subunits termed α′ (58,000 Da), α (57,000 Da), and β (42,000 Da) (9). 11S Globulin is a large oligomeric protein of *ca.* 360,000 Da (10). Soybean proteins are composed of about 63% polar amino acids (11). Their conformation structures are mainly stabilized by hydrogen bonds, electrostatic forces, and hydrophobic interactions. A plasticizer that contains polar groups should be compatible with soybean proteins. The plasticization effect of a plasticizer could be influenced by its ease of insertion and position within a three-dimensional protein network (12). Molecular weight, numbers, and positions of hydroxy groups of a plasticizer are all variables affecting its ability to plasticize soybean protein-based polymer. Wang *et al.* (6) investigated the plasticization effects of glycerol and polyethylene glycol (PEG) on soy protein isolate (SPI) plastics at various concentrations. The tensile strength of the SPI plastics decreased as glycerol or PEG content increased, and the elongation of the plastics increased as the glycerol content increased but showed little improvement as PEG content increased.

In this study, glycerol (GLY), propylene glycol (PRO), 1,2-butanediol (BUT12), and 1,3-butanediol (BUT13) were chosen as plasticizers. GLY and PRO have three carbons in common, but GLY has three hydroxy groups, and PRO has two hydroxy groups; BUT12 and BUT13 have four carbons in common and two hydroxy groups attached to 1,2 and 1,3 carbons, respectively. The objective of this study was to characterize the thermal and mechanical properties, morphology, and water absorption of plasticized soybean protein polymer as affected by polyols with different molecular sizes and positions of hydroxy groups.

MATERIALS AND METHODS

Materials and SPI preparation. Defatted soy flour was obtained from Cargill (Cedar Rapids, IA) and used for the preparation of SPI. GLY, PRO, BUT12, and BUT13 were purchased from Sigma Chemical Co. (St. Louis, MO).

SPI was extracted from defatted soy flour following the procedure described by Mo and Sun (13) by isoelectric point precipitation at pH 4.2. The precipitate was freeze-dried and then milled into powder. The SPI had an average protein content of 93% (dry basis) and moisture content of 5.8%.

Specimen preparation. Plasticizers were added to the SPI powder in drops and mixed in a mixer (National Manufacturing Co., Lincoln, NE) for 45 min at room temperature. The final content of plasticizer was 25% (wt%) to ensure an effective plasticization (14) and uniform distribution of plasticizers in SPI. The mixture was equilibrated for 24 h and then ground to break up lumps.

---

*To whom correspondence should be addressed. E-mail: xss@ksu.edu

The plasticized SPI powder was molded into a tensile bar specimen (Type IV) according to standard method ASTM D638-91 (15). SPI powder (5 g) was placed into a dog-bone-shaped molder and compression-molded in a hot press (Model 3890 Auto "M"; Carver Inc., Wabash, IN) at 20 MPa and 140°C for 5 min. The specimen was then cooled to 50°C at 30°C/min before removal from the mold. Flash was removed by sanding the edges of the specimen with grade 180 abrasive sandpaper. Unplasticized SPI was used as a control.

DSC measurements. Thermal denaturation of the plasticized SPI was studied using a DSC (DSC 7; PerkinElmer, Norwalk, CT), which was calibrated with indium and zinc. About 10 mg plasticized SPI or ground plastic powder was weighed into a DSC pan and the pan was sealed. The samples were measured at a temperature range of 50 to 200°C and a heating rate of 10°C/min. Denaturation enthalpy was calculated as the sum of the 7S and 11S denaturation enthalpies. The reported values are averages of two replicates.

Dynamic mechanical measurements. Dynamic mechanical properties of the molded SPI plastics were studied using a dynamic mechanical analyzer (DMA) (DMA 7e; PerkinElmer) in a three-point bending configuration. The sample size of the SPI plastics was about 13 mm × 6 mm × 2 mm. Temperature was increased from −30 to 200°C at a rate of 5°C/min with a static force of 660 mN, dynamic force of 600 mN, and frequency of 1 Hz. The reported values are averages of three replicates.

Tensile testing and fracture morphology. Mechanical properties of the molded SPI plastics were measured using an Instron (Model 4465; Canton, MA) according to ASTM D638-91 (15). The SPI plastics were preconditioned at 23°C and 50% relative humidity for 48 h before testing. The crosshead speed was 5 mm/min. Tensile strength, strain at break, and Young's modulus were calculated. The reported results are averages of five samples. The fracture surface of the broken plastic after tensile testing was examined using a scanning electronic microscope (SEM) (Hitachi S-3500N; Hitachi Science System, Ltd., Tokyo, Japan) with a secondary electron detector at an accelerating voltage of 5 kV, a working distance of 15.6 mm, and a magnification of 300 times. The sample was stuck to an aluminum stub, and the fractured surface was coated with a mixture of 60% gold and 40% palladium particles by a sputter coater (Desk II Sputter/Etch Unit, Moorestown, NJ).

Water absorption measurement. Water absorption was determined following ASTM D570-81 (16). The SPI plastic samples were preconditioned at 50°C for 24 h and then cooled in a desiccator before being weighed. The preconditioned SPI plastics were submerged in distilled water at 25°C for 2 h and 24 h. The extra water on the surface was removed with a paper towel before weighing. Water absorption was calculated as a percentage of initial weight. Soluble material loss of the sample was determined by weighing the specimens after drying them in an oven at 50°C for 24 h, which was included in the total water absorption calculation for the 24 h water soaking.

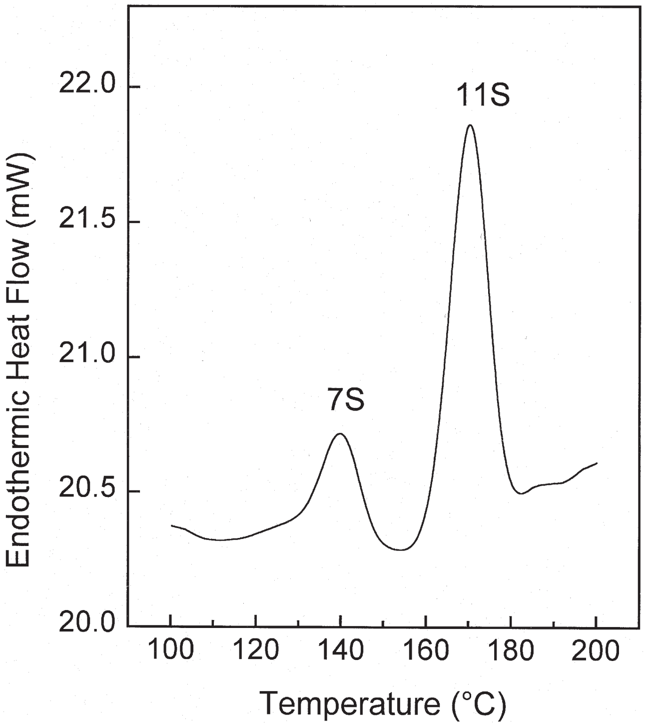

FIG. 1. A typical DSC thermogram of soy protein isolate (SPI) with 10% moisture content.

RESULTS AND DISCUSSION

Thermal properties. Heating converts soybean protein from its native state to a denatured state, accompanied by unfolding and disruption of the intramolecular bonding (17), which is observable as an endothermic peak. A typical denaturation of (Fig. 1) gave two peaks, one at 139.7 and one at 169.9°C, which corresponded to the low molecular weight 7S globulins and high molecular weight 11S globulins, respectively. These results are in agreement with the values reported by Kitabatake *et al.* (17).

Denaturation temperatures of 7S globulins (T_{d1}) and 11S globulins (T_{d2}) for the plasticized SPI powder were lower than those of the unplasticized SPI (Table 1). The effects of plasticization on the denaturation temperatures of the plasti-

TABLE 1
Denaturation Temperatures and Enthalpies of Plasticized Soy Protein Isolate (SPI)^a

Sample designation^b	$T_{d1}{}^c$ (°C)	$T_{d2}{}^c$ (°C)	$\Delta H_d{}^c$ (J/g)
BUT12	89.52d	109.5d	11.25b
BUT13	95.76c	113.8c	11.76a,b
GLY	117.0b	140.9b	11.46b
PRO	82.69e	107.5e	10.65b
CON	139.7a	169.9a	13.48a

^{a}Means within a column with the same roman letter are not significantly different based on the least significant difference (LSD) procedure at an α = 0.05 level.
^{b}BUT12, BUT13, GLY, PRO, and CON represent the sample with 1,2-butanediol, 1,3-butanediol, glycerol, propylene glycol, and the control (without plasticizer), respectively.
^{c}T_{d1}, T_{d2}, and ΔH_d represent denaturation temperatures for 7S, 11S, and denaturation enthalpy, respectively.

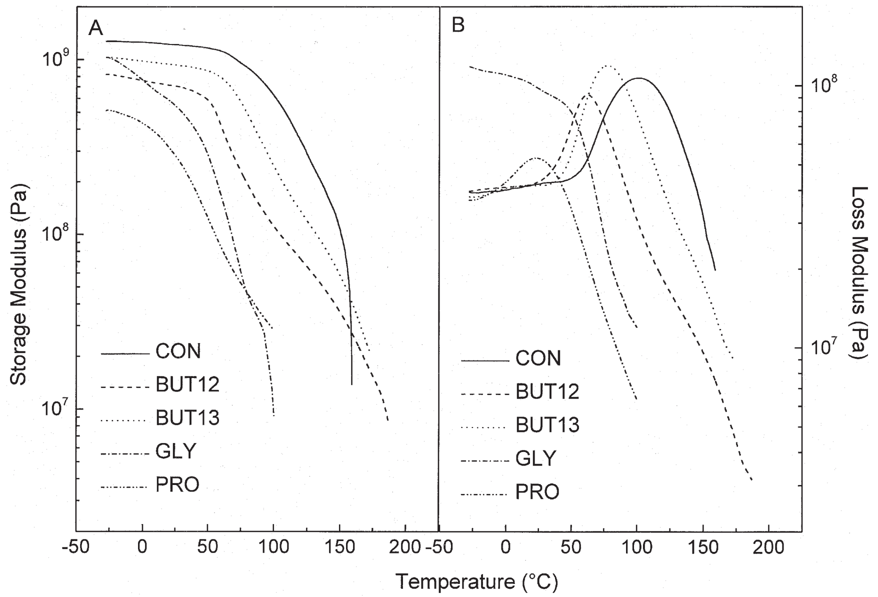

FIG. 2. Dynamic mechanical analysis in three-point bending mode: dependence of storage modulus (A) and loss modulus (B) on temperatures for SPI plastics molded at 140°C, 20 MPa for 5 min. BUT12, BUT13, GLY, PRO, and CON represent the SPI plastics with 1,2-butanediol, 1,3-butanediol, glycerol, propylene glycol, and unplasticized SPI plastics, respectively. See Figure 1 for abbreviation.

cized SPI were in the order: PRO > BUT12 > BUT13 > GLY. The enthalpies of denaturation (ΔH) were reduced by about 2 J/g for all plasticized samples. Soybean proteins have a three-dimensional structure with most hydrophobic groups buried inside and hydrophilic groups facing outside. To denature the proteins, chemical reagents must first disrupt the hydrophilic shell and then penetrate into the hydrophobic region. However, polyols with polar hydroxy groups usually can effectively disrupt only the hydrophilic shell. Therefore, the denaturing ability of polyols on soybean proteins should be enhanced as their hydrophobicity increases (18). For the three-carbon plasticizers (GLY and PRO), PRO has one hydroxy group less than GLY and is more hydrophobic. As a result, the SPI with PRO had lower T_d than that of the SPI with GLY. For the plasticizers with four carbons (BUT12 and BUT13), SPI with BUT12 had a lower T_d, probably because the two hydroxy groups are attached to the 1,2 carbons, which may be more hydrophobic than BUT13.

Dynamic mechanical properties. The storage modulus (E') and loss modulus (E'') curves of the molded plastics as a function of temperature are presented in Figure 2. The E' decreased as temperature increased for all plastics, and the onset temperatures of the E' at which the E' started to drop shifted to lower temperatures for the plasticized SPI plastics. The plastics plasticized by PRO had the lowest E' (Fig. 2A, Table 2).

Glass transition temperature (T_g) is an important property for an amorphous polymer. T_g is the point at which the segmental motion of macromolecular chains is reactivated when a polymer is heated, corresponding to a system change from a brittle or glassy state to a rubber-like or viscous liquid state. The T_g was identified as the peak temperature of E'' (19). The glass transition occurred within a broad temperature range, from 50 to 150°C, for the unplasticized SPI plastics (Fig. 2B). This broad range may be caused by a wide variety of peptide chains in the soybean protein (20). The T_g for 7S globulins and 11S globulins of the unplasticized SPI plastics are 77.1 and 99.6°C, respectively, which agreed with values determined by Morales and Kokini (20). The T_g values for the plasticized plastics are summarized in Table 3.

TABLE 2
Storage Modulus of Molded Plastics Made from Plasticized SPI[a]

Sample	−30°C (MPa)	25°C (MPa)
BUT12	959.4b,c	823.9c
BUT13	1074a,b	955.3b,c
GLY	953.8b,c	511.4d,e
PRO	585.9d	318.5e
CON	1250a	1173a

[a] Means within a column with the same roman letter are not significantly different based on the LSD procedure at an $\alpha = 0.05$ level. See Table 1 for abbreviations.

TABLE 3
Glass Transition Temperatures of Plastics Made from Plasticized SPI[a]

Sample	T_{g1}[b]	T_{g2}[b]
BUT12	51.64c	62.34c
BUT13	59.71b	71.12b
GLY	18.28d	48.96d
PRO	—	21.87e
CON	77.06a	99.56a

[a] Means within a column with the same roman letter are not significantly different based on the LSD procedure at an $\alpha = 0.05$ level. See Table 1 for abbreviations.

[b] T_{g1} and T_{g2} represent the glass transition temperatures for 7S and 11S, respectively.

TABLE 4
Mechanical Properties of Molded Plastics Made from Plasticized SPI[a]

Sample	Maximum stress (MPa)	Strain at break (%)	Young's modulus (MPa)
BUT12	31.96b	4.220c	1082c
BUT13	34.44a	3.040d	1286b
GLY	13.76d	177.5a	250.5d
PPO	4.531e	8.520b	108.4e
CON	21.66c	1.812e	1667a

[a]Means within a column with the same roman letter are not significantly different based on the LSD procedure at an α = 0.05 level. See Table 1 for abbreviations.

The efficiency of a plasticizer is usually measured by its effect on the T_g of a polymer. The depressing T_g effect of plasticizers was in the order: PRO > GLY > BUT12 > BUT13. The degree of T_g drop is an indicator of chemical compatibility of a plasticizer with a protein (22). Both PRO and GLY appear to be more compatible with soybean protein than BUT12 or BUT13. This is because their smaller molecular size fits more easily into the protein chains and establishes hydrogen bonds with polar groups of proteins (23). As a result, the protein–protein interactions decreased owing to the increased plasticizer–protein interactions. Conversely, BUT have a relatively larger molecular size than either PRO or GLY, preventing them from easily inserting into the three-dimensional protein structures, which would inhibit the interactions between BUT and some of the polar groups in the proteins. Thus, the two members of the BUT series were less effective in disrupting the protein–protein interactions. BUT12 depressed the T_g of the SPI plastics more than BUT13 did, indicating that BUT12 was more compatible with soybean proteins. The two adjacent hydroxy groups in the BUT12 could allow it to interact with proteins more easily than BUT13, which has two separated hydroxy groups.

Mechanical properties and fracture morphology. One of the primary functions of a plasticizer is to improve the strain at break and toughness. However, this has to be achieved at the expense of modulus. Mechanical properties of the plasticized SPI plastics are shown in Table 4. As expected, the Young's modulus of all plasticized SPI plastics decreased significantly. However, the plastics with BUT showed higher tensile strength and lower strain at break than those with PRO and GLY. This is because BUT have relatively long carbon chains and a relatively low plasticization effect. In BUT series, the plastics with BUT12 had higher strain at break and lower Young's modulus than those with BUT13. These results are in agreement with the effectiveness of depressing T_g observed in the dynamic mechanical analysis.

The SPI plastics with GLY had the highest strain at break, and the plastics with PRO had the lowest tensile strength and a low strain at break. As discussed before, PRO has smaller molecular size than GLY. Therefore, it would interact easily

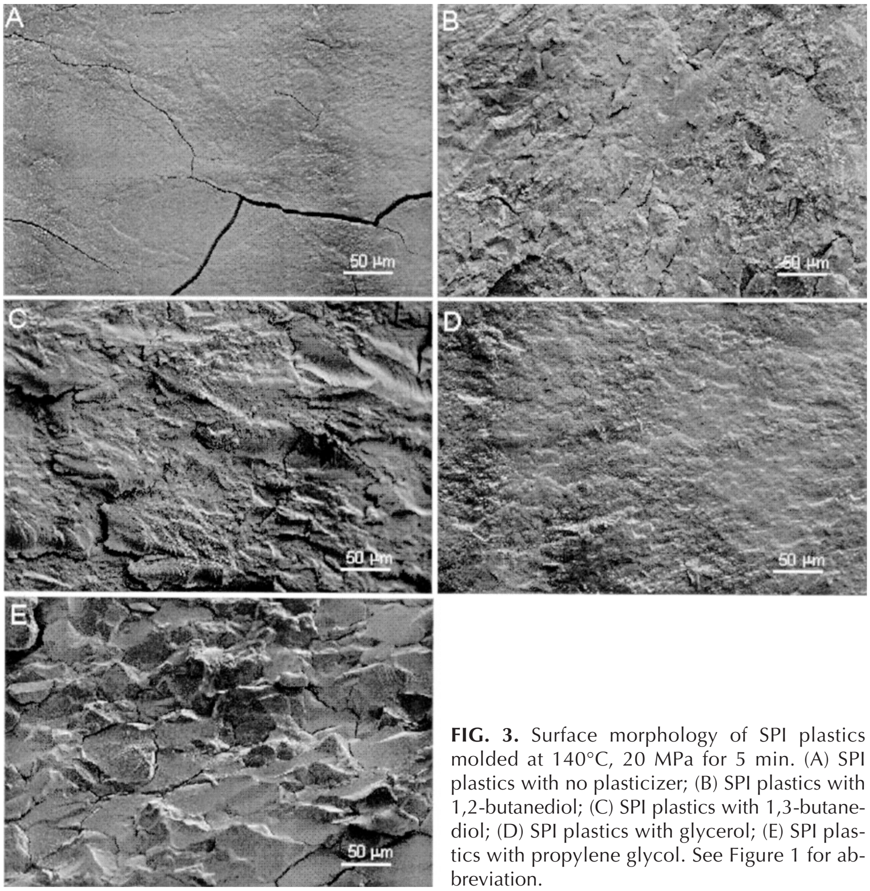

FIG. 3. Surface morphology of SPI plastics molded at 140°C, 20 MPa for 5 min. (A) SPI plastics with no plasticizer; (B) SPI plastics with 1,2-butanediol; (C) SPI plastics with 1,3-butanediol; (D) SPI plastics with glycerol; (E) SPI plastics with propylene glycol. See Figure 1 for abbreviation.

TABLE 5
Water Absorption (%) of Plastics Made from Plasticized SPI[a]

	Plasticizer				
	BUT12	BUT13	GLY	PRO	CON
2 h Water absorption	54.33c	62.29c	78.66b	62.48c	93.42a
24 h Water absorption	267.2d	291.7c	338.2b	230.6e	468.8a
24 h Soluble material loss	15.03b	13.47b	24.82a	22.45a	5.664c
24 h Total water absorption	282.2d	305.2c	363.1b	253.4e	474.5a

[a] Means within a row with the same roman letter are not significantly different based on the LSD procedure at $\alpha = 0.05$ level. See Table 1 for abbreviations.

with the polar groups associated with the peptide chains, largely weakening the protein–protein interactions, and resulting in low cohesiveness in the protein network.

The cross section of the unplasticized SPI plastics appeared to be smooth with sharp cracks (Fig. 3A), suggesting a brittle-type failure. The cross sections of the plasticized SPI plastics appeared to be rough, suggesting a ductile failure (Figs. 3B–D). The plastics plasticized with BUT12 had smaller cracks (Fig. 3B) than the plastics with BUT13 (Fig. 3C), which is in agreement with its thermal and mechanical properties. The cross section of the plastics plasticized with GLY appears to be a fluctuated and continuous matrix (Fig. 3D), whereas plastics plasticized with PRO had relatively smooth and extensive cracks (Fig. 3E), indicating poor mechanical behavior, which was caused by excessive weakening of protein interactions.

Water absorption. The SPI plastics plasticized with all four polyols had lower water absorption than the unplasticized SPI plastics after 2 or 24 h water soaking (Table 5). The reduction in water absorption caused by plasticization was in the order: PRO > BUT12 > BUT13 > GLY > CON, which was the same order as the reduction in the denaturation temperature of the plastics. Proteins with a large degree of aggregation or entanglement usually have low water absorption. Proteins with high denaturation temperatures often have a small degree of aggregation or entanglement after molding. SPI plasticized with GLY had higher denaturation temperatures than those with BUT and would aggregate/entangle to a lower degree after molding; thus, the plastic with GLY had higher water absorption.

The plasticized SPI plastics had higher soluble material loss than the unplasticized plastics (Table 5). The soluble material likely consisted of plasticizers and some low molecular weight proteins. The soluble material loss was lower for the plastics with BUT than those with GLY and PRO. The BUT had relatively large molecular size and may not easily leach out of the protein network.

Both PRO and GLY are small hydrophilic molecules, which effectively depressed T_g and had good compatibility with soybean protein. The BUT series have relatively high molecular weight and showed poor plasticizing effects. The difference in plasticization effects between BUT12 and BUT13 could be caused by the difference in the positions of their hydroxy groups. The efficiency of a plasticizer is significantly related to its chemical structure, including carbon chain length, the number and positions of hydroxy groups, and the effectiveness of the interactions between plasticizer and protein.

ACKNOWLEDGMENT

Contribution No. 01-337-J from the Kansas Agricultural Experimental Station, Manhattan, Kansas.

REFERENCES

1. Mohanty, A.K., M. Misra, and G. Hinrichsen, Biofibres, Biodegradable Polymers and Biocomposites: An Overview, *Macromol. Mater. Eng. 276/277*:1–24 (2000).
2. Mo, X.Q., X.S. Sun,, and Y.Q. Wang, Effects of Molding Temperature and Pressure on Properties of Soy Protein Polymer, *J. Appl. Polymer Sci. 73*:2595–2602 (1999).
3. Ogale, A.A., P. Cunningham, P.L. Dawson, and J.C. Action, Viscoelastic, Thermal, and Microstructural Characterization of Soy Protein Isolate Films, *J. Food Sci. 65*:672–679 (2000).
4. Otaigbe, J.U., H. Goel, and T. Babcock, Processability and Properties of Biodegradable Plastics Made from Agricultural Biopolymers, *J. Elastomers Plast. 31*:59–71 (1999).
5. Pol, H.V., and A.A. Ogale, Laminated Protein Films, *Annu. Tech. Conf.–Soc. Plast. Eng. 2*:1751–1755 (2000).
6. Wang, S., H.J. Sue, and J. Jane, Effects of Polyhydric Alcohols on the Mechanical Properties of Soy Protein Plastics, *J. Micromol. Sci. Pure Appl. Chem. A33*:557–569 (1996).
7. Sears, J.K., and J.R. Darby, Mechanism of Plasticizer Action, in *The Technology of Plasticizers,* edited by J.K. Sears and J.R. Darby, Wiley-Interscience, New York, 1982, pp. 35–77 .
8. Entwistle, C.A., and R.C. Rowe, Plasticization of Cellulose Ethers Used in the Film Coating of Tablets, *J. Pharm. Pharmacol. 31*:269–272 (1978).
9. Thanh, V.H., and K. Shibasaki, Major Proteins of Soybean Seeds. A Straightforward Fractionation and Their Characterization, *J. Agric Food Chem. 24*:1117–1121 (1976).
10. Badley, R.A., D. Atkinson, H. Houser, G. Oldani, J.P. Green, and J.M. Stubbs, The Structure, Physical and Chemical Properties of Soybean Protein Glycinin, *Biochim. Biophys. Acta 412*:214–228 (1975).
11. Ly, Y.T.P, Johnson, L.A., and J. Jane, Soy Protein as Biopolymer, in *Biopolymers from Renewable Resource,* edited by D.L. Kaplan, Springer, Berlin, 1998, pp. 144–176.
12. Cuq, B., N. Gontard, J.L. Cuq, and S. Guilbert, Selected Functional Properties of Myofibrillar Protein-Based Films as Affected by Hydrophilic Plasticizers, *J. Agric. Food Chem. 45*: 622–626 (1997).
13. Mo, X.Q., and X.S. Sun, Thermal and Mechanical Properties of Plastics Molded from Urea-Modified Soy Protein Isolates, *J. Am Oil Chem. Soc. 78*:867–872 (2001).
14. Sue, H.J., S. Wang, and J.L. Jane, Morphology and Mechanical Behavior of Engineering Soy Plastics, *Polymer 38*:5035–5040 (1997).
15. *Annual Book of ASTM Standards,* Standard Test Methods for Tensile Properties of Plastics, American Society for Testing and Materials, Philadelphia, 1993, pp. 161–173, ASTM Standard D638-91.

16. *Annual Book of ASTM Standards,* Standard Test Methods for Water Absorption of Plastics, American Society for Testing and Materials, Philadelphia, 1993, pp. 143–145, ASTM Standard D570-81 (Reapproved 1988).
17. Kitabatake, N., M. Tahara, and Etsushiro Doi, Thermal Denaturation of Soybean Protein at Low Water Content, *Agric. Biol. Chem. 54*:2205–2212 (1990).
18. Fukushima, D., Denaturation of Soybean Proteins by Organic Solvents, *Cereal Chem. 46*:156–163 (1969).
19. Aklonis, J.J., W.J. Macknight, and M. Shen, Transitions and Relaxation in Amorphous Polymer, in *Introduction to Polymer Viscoelasticity,* John Wiley & Sons, New York, 1972, pp. 60–87.
20. Morales, A., and J.L. Kokini, Glass Transitions of Soy Globulin Using Differential Scanning Calorimetry and Mechanical Spectrometry, *Biotechnol. Prog. 13*:624–629 (1997).
21. Johari, G.P., and G. Sartor, Thermodynamic and Kinetic Features of Vitrification and Phase Transformation of Proteins and Other Dry Constituents of Dry and Hydrated Soybean, a High Protein Cereal, in *The Properties of Water in Foods –ISOPOW6,* Blackie Academic & Professional, London, 1998, pp. 103–138.
22. Gioia, L.D., and S. Guilbert, Corn Protein-Based Thermoplastics Resins: Effect of Some Polar and Amphiphilic Plasticizers, *J. Agric. Food Chem. 47*:1254–1261 (1999).
23. Galietta, G., L.D. Gioia, S. Guilbert, and B. Cuq, Mechanical and Thermomechanical Properties of Films Based on Whey Proteins as Affected by Plasticizer and Cross-Linking Agent, *J. Dairy Sci. 81*:3123–3130 (1998).

[Received May 4, 2001; accepted November 20, 2001]