Preparation and Functional Properties of Soy Hull Pectin

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ABSTRACT: Soy hulls (400 g), a co-product of the soybean processing industry, were used as a source of pectin which was extracted by 0.05 N hydrochloric acid and precipitated with alcohol. The effect of various ratios of hull to extraction solvent (1:10, 1:15, 1:20, and 1:25) on the yield and purity of soy hull pectin was measured. The soy hull pectin extracts contained 63.07 to 68.72% galacturonic acid at various hull/solvent ratios. The pectin yield increased from 7.68 to 13.73% as the hull/solvent ratio increased from 1:10 to 1:15. Changes in pectin yield for higher hull/solvent ratio were insignificant (16.31–13.28% for 1:20–1:25 ratio, respectively). The hull/extraction solvent ratio did not significantly affect the pectin content and degree of esterification. Diffuse reflectance Fourier transform infrared spectroscopy of soy hull pectin revealed a similar surface structure to commercial- and analytical-grade pectins. The solubility of soy hull pectins was independent of solvent extraction ratios and pH. Rheology studies showed that pectin solution viscosity increased as the ratio of extraction solvent to hull decreased.

Paper no. J9872 in *JAOCS 78*, 709–713 (July 2001).

KEY WORDS: FTIR, functional properties, pectin, processing, rheology, soy hull.

Pectin is a complex carbohydrate consisting of D-galacturonic acid linked by α 1–4 glycosidic linkages (1). The molecular weight of pectin varies from 50,000 to 150,000 Da depending on the extraction procedure and source materials (2). Pectins are used as gelling agents in jams, jellies, and fruit preparations and as stabilizers in confections, dairy products, bakery fillings, and icings. Medical applications include serum cholesterol-lowering agents (3), antidiarrheal, detoxicant, demulcent (4), and emulsion stabilizers for water-in-oil emulsions.

Pectins are commercially extracted from citrus peels and apple pomace with hot acidified water. The extract is then centrifuged, filtered a number of times, and finally precipitated with alcohol. Most pectins used in the food industry are prepared from citrus peel (4). Citrus peel and apple pomace contain about 25 and 12% pectin, respectively, and the commercial extraction yield is about 25% (5,6). Extraction of pectins from sugar beets, sunflower head residues, and dehulled rapeseed has also been reported (7–9).

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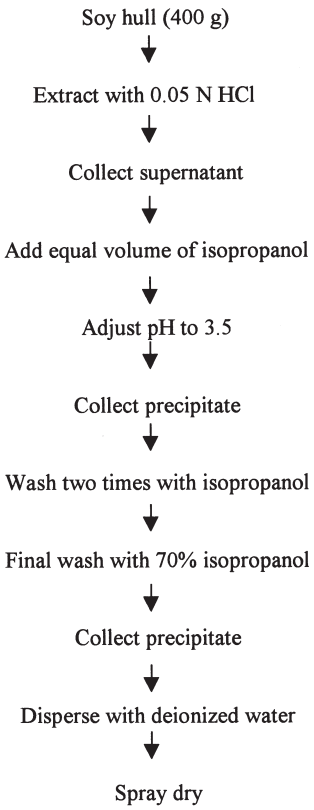
Soy hulls are major by-products of the soybean processing industry, and the insoluble carbohydrate fraction contains 30% pectin, 50% hemicellulose, and 20% cellulose (10). Soy hull is potentially an inexpensive commercial source of pectin. Citrus peel and apple pomace are difficult to process unless they are first dried. In contrast, soy hull can be stored and transported without further processing. Gnanasambandam and Proctor (11) extracted pectin from soy hull by adapting the acid extraction and alcohol precipitation method used for citrus pectin production. Kalapathy and Proctor (12) subsequently optimized the acid extraction and alcohol precipitation conditions on the yield and purity of soy hull pectin.

The objectives of this study were (i) to optimize the hull/solvent (0.05 N HCl) ratio for large-scale soy hull pectin preparation and (ii) to evaluate the solubility and rheological properties of soy hull pectins relative to selected commercial food-grade pectins.

MATERIALS AND METHODS

Pectin samples. The analytical-grade citrus pectin was purchased from the Sigma Chemical Co. (St. Louis, MO). The commercial food-grade pectins were obtained from Danisco Ingredients Inc. (New Century, KS). The degree of esterification (DE) values for commercial I and commercial II pectins were 76.2 and 32, respectively. The DE value for analytical-grade citrus pectin was not provided.

Preparation of soy hull pectins. Soy hulls were obtained from Riceland Foods (Stuttgart, AR) and ground to a particle size of 80 mesh with a centrifugal grinding mill (Model ZM-1; Retsch/Brinkmann, Westbury, NY). The overall scheme for pectin preparation is shown in Scheme 1. Pectin was extracted from ground soy hull (400 g) using either 4.0, 6.0, 8.0, or 10.0 L of 0.05 N HCl at 90°C for 60 min with constant stirring. The extracts were cooled to room temperature in a water bath and centrifuged (CRU 5000; IEC, Needham Heights, MA) at $2700 \times g$ for 15 min. The supernatants were collected and dispersed in an equal volume of isopropanol. Pectin was precipitated by adjusting the pH of the dispersion to 3.5 (12) and allowed to stand for 6 h. The precipitate was collected by centrifugation, dispersed in isopropanol (1.0 L), stirred for 1 h, and centrifuged. This washing process was repeated with isopropanol before a final washing with 70% isopropanol (1.0 L). The precipitates were dispersed in deionized water and spray-dried in a lab S1 spray drier (APV Anhydro Inc., Copenhagen, Denmark). The inlet temperature of the spray



SCHEME 1

drier was maintained between 200 and 220°C, and the outlet temperature was 70–80°C. The spray-dried pectin powder was subjected to the following analysis.

Pectin yield. Pectin yields (w/w) were calculated as the percentage of spray-dried soy hull pectin weight of the total pectin present in soy hull. The pectin content of soy hull as a percentage of galacturonic acid was determined by colorimetry using *m*-hydroxydiphenyl (13).

Pectin content. Pectin content, as a percentage of galacturonic acid, was determined by high-performance liquid chromatography (HPLC) (14). A Spectra System AS 1000 HPLC (Spectra Physics) equipped with a Spherisorb ODS2 250 × 4.6 mm C18 column (particle size, 5 μm), a 15 × 3.2 mm guard column (Water Corporation, Milford, MA), and a forward optical scanning detector (Spectra Focus, Spectra Physics) was used to determine galacturonic acid content in pectin samples. D-Galacturonic acid monohydrate (Sigma) was used as the standard.

DE. A diffuse reflectance Fourier transform infrared spectroscopic method (15) was used to determine the DE of the soy hull pectin samples.

Loss on drying, total ash, water-insoluble ash, and acid-insoluble ash. Loss on drying was determined by the method of Food Chemical Codex (16). Total ash, water-insoluble ash, and acid-insoluble ash were determined by the method of Harbers (17). An Isotemp programmable forced-draft furnace (Fisher Scientific, Fair Lawn, NJ) was used for all ashing.

Surface structure analysis. Diffuse reflectance Fourier transform infrared spectra (DRIFTS) of soy hull pectin samples were collected using an Impact 410 Nicolet Instrument (Nicolet Analytical Instruments, Madison, WI). DRIFTS were obtained by co-adding 100 scans at a resolution of 8.0 cm^{-1}. Fourier transform infrared (FTIR) spectra were analyzed for surface chemical functional groups by a software package (OMNIC FTIR Software, v4.1; Nicolet Analytical Instruments) and compared with the commercial citrus pectins.

Pectin solubility. Pectin water solubility at pH 2, 4, 6, 8, and 10 was determined gravimetrically. Pectin samples (5.0 g) were dispersed in 100 mL of deionized water, and either 0.1 N HCl or 0.1 N NaOH was used to maintain pH in the range of 2 to 10. The insoluble pectin portion was determined after centrifugation (2700 × *g*, 15 min, CRU 5000; IEC) and used to determine pectin solubility.

Rheology of pectin solution. The rheology of 3% pectin solution in deionized water (wt/vol) was determined with a Haake VT 550 Rheometer (Haake MessTechnik Gmbh Co., Karlsruhe, Germany) with an SV-DIN sensor. All rheological experiments were carried out at a constant temperature (23°C) immediately after the pectin solutions had been prepared. The shear rate was increased from 0 to 400 s^{-1} by the software control, and strain measurement was recorded. Each sample was tested three times. Rheology curves were prepared to show shear rate vs. shear stress and shear rate vs. apparent viscosity.

Statistical analysis. Results of three replicates were used, and Student's *t*-test was used to analyze data. Least significance difference (LSD) values were used to differentiate mean values and significance was defined at $P < 0.05$ (18).

RESULTS AND DISCUSSION

Yield, pectin content, and DE. Yield, galacturonic acid content, and DE of soy hull pectin at various hull/solvent ratios are presented in Table 1. Pectin yields for hull to solvent ratios of 1:15, 1:20, and 1:25 were similar, whereas the 1:10 ratio produced significantly lower yield than the others. The pectin yield for commercial citrus pectins was typically 20–26%, (19), and soy pectin yield was 7–16%. The pectin content of the extractions was similar to that from citrus pectin, which contains 70% galacturonic acid. In a laboratory-scale preparation, Gnanasambandam and Proctor (11) found that the pectin content of soy hull extracts, expressed as the percentage galacturonic acid in aqueous extraction (1:20), was 55%, and that 0.1 N HNO_3 extraction (1:20) improved the yield and galacturonic acid content to 15.0, and 76.7%, respectively. For Kalapathy and Proctor (12), the yield and pectin content of soy hull extracts produced with 0.05 N HCl followed by precipitation with isopropanol at pH 3.5 were 26 and 70%, respectively. In the present work, the DE values for soy hull pectins were very low (Table 1) which may be due to ester hydrolysis during extraction.

Loss on drying, total ash, water-insoluble ash, and acid-insoluble ash. Loss on drying, total ash, water-insoluble ash, and acid-insoluble ash of soy hull pectin at different hull/sol-

TABLE 1
Yield (%), Galacturonic Acid Content (%), and Degree of Esterification (DE) of Soy Hull Pectin Extracted at Different Hull/Solvent Ratios[a]

Hull/solvent ratio	Pectin yield (%)[a]	Galacturonic acid content (%)	DE
1:10	7.68 ± 1.17[b]	63.07 ± 1.06[b]	17.09 ± 1.11[a]
1:15	13.73 ± 1.11[a]	66.23 ± 1.06[a]	20.79 ± 0.95[a]
1:20	16.31 ± 2.23[a]	68.72 ± 1.81[a]	18.02 ± 1.70[a]
1:25	13.28 ± 1.80[a]	67.13 ± 0.65[a]	17.45 ± 1.04[a]

[a]Values with different roman superscripts in each column are significantly ($P < 0.05$) different.

TABLE 2
Loss on Drying, Total Ash, Water-Insoluble Ash, and Acid-Insoluble Ash of Soy Hull Pectin Extracted at Different Hull/Solvent Ratios[a]

Hull/solvent ratio	Loss on drying (%)	Total ash (%)	Water-insoluble ash (%)	Acid-insoluble ash (%)
1:10	3.61 ± 0.36[b]	4.57 ± 0.88[a]	3.95 ± 1.02[a]	0.33 ± 0.08[b]
1:15	4.70 ± 0.97[b]	3.54 ± 0.45[a,b]	3.08 ± 0.28[a]	0.47 ± 0.01[a]
1:20	6.39 ± 0.44[a]	3.29 ± 0.07[b]	2.79 ± 0.06[a]	0.29 ± 0.05[b,c]
1:25	6.96 ± 0.56[a]	4.06 ± 0.22[a]	3.50 ± 0.27[a]	0.21 ± 0.02[c]

[a]Values with different roman superscripts in each column are significantly ($P < 0.05$) different.

vent ratios are presented in Table 2. Pectin produced in high hull/solvent ratio loses more weight during the drying process. Loss on drying for all the pectins produced at various hull/solvent ratios was less than 12%, which is a requirement of the Food Chemical Codex (16). Acid-insoluble ash contains silicates. The limit for pectin total ash is 10% and acid-insoluble ash is 1% (16). Pectin produced with various hull/solvent ratios were below this limit.

Surface structure analysis. The DR1FTS (400–4000 cm^{-1}) of soy hull pectin samples, commercial pectins, and analytical-grade pectins are presented in Figure 1. FTIR spectra indicated that there were no major structural differences in pectins produced using different extraction procedures, and the soy pectin structures were comparable to those in commercial- and analytical-grade pectins. FTIR spectra of the region between 1000 and 2000 cm^{-1} include the major chemical functional groups in pectin (20), and those regions are typically used to identify different types of pectin (5). The absorption bands between 1100 and 1200 cm^{-1} were from ether (R-O-R) and cyclic C-C bonds in pectin molecules. The absorption band at 1500 cm^{-1} was due to OH bending vibration. The region between 1600 and 1800 cm^{-1} is of special interest, since it provides structural information that can be used to compare different types of pectin. This spectral region reveals the existence of two bands at 1640 and 1750 cm^{-1} from free and esterified carboxyl groups, respectively. It was observed that the esterified carboxyl groups showed an increase in their intensities and band areas as the DE values increased. The intensity and band area of esterified and free carboxyl groups were similar for all the soy hull pectins produced at various hull/solvent ratios (A–D in Fig. 1). The intensity and band area of esterified carboxyl groups for commercial pectin I (line G in Fig. 1, DE = 76.2) were higher than for commercial pectin II (F, DE = 32). The esterified carboxyl band areas for all the soy hull pectins were less than the commercial- and analytical-grade pectins tested. This indicated the low DE values for soy hull pectins. At 2900 cm^{-1} the band corresponds to the C-H stretching of the CH_3 groups. The broader band from 2400 to 3600 cm^{-1} was from stretching of the hydroxyl groups due to moisture in the pectin samples.

Pectin solubility. Solubility of soy hull pectins extracted at various hull/solvent ratios of different food-grade and analytical-grade pectins are presented in Table 3. The solubility of soy hull pectins at different pH values does not vary significantly, and solubility of samples extracted at various hull/solvent ratios was similar. Commercial pectin I (DE = 76.2) had the lowest solubility among the pectin samples tested in contrast to commercial pectin II (DE = 32) which had the highest

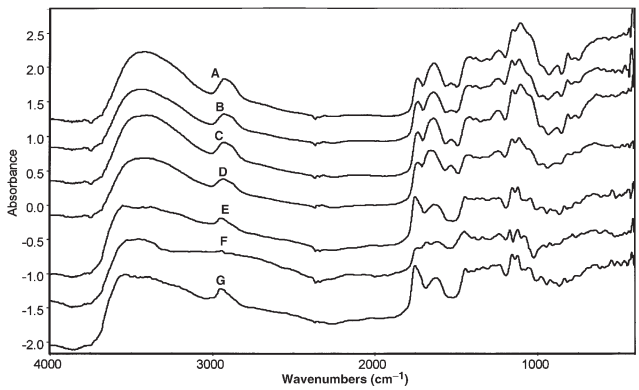


FIG. 1. Diffuse reflectance Fourier transform infrared spectra of the 4000–400 cm^{-1} region of soy hull pectin extracted at different hull/solvent ratios: (A) 1:10, (B) 1:15, (C) 1:20, and (D) 1:25; (E) analytical-grade citrus pectin (Sigma Chemical Co., St. Louis, MO); commercial food-grade pectins (F) Commercial II (DE = 32), and (G) Commercial I (DE = 76.2), where DE is degree of esterification.

TABLE 3
Solubility of Soy Hull Pectin Extracted at Different Hull/Solvent Ratios, of Commercial Food-Grade Pectins I (DE = 76.2) and II (DE = 32), and Analytical-Grade Pectin (citrus pectin; Sigma Chemical Co., St. Louis, MO)[a]

	Solubility (%) of pectins at different pH				
Samples	2.0	4.0	6.0	8.0	10.0
Soy pectin (1:10)	1.98 ± 0.07a[a,b]	1.93 ± 0.24a[b,c]	1.90 ± 0.18a[a,b]	1.44 ± 0.05b[c,d]	1.77 ± 0.14a[b]
Soy pectin (1:15)	1.80 ± 0.35a[b]	1.61 ± 0.07a[c]	1.73 ± 0.14a[b,c]	1.56 ± 0.14a[c,d]	1.62 ± 0.13a[b,c]
Soy pectin (1:20)	1.91 ± 0.09a[b]	1.98 ± 0.12a[b]	1.73 ± 0.03b[b,c]	1.89 ± 0.07a[b]	1.73 ± 0.08b[b]
Soy pectin (1:25)	1.83 ± 0.16a[b]	1.73 ± 0.21a[b,c]	1.73 ± 0.15a[b,c]	1.64 ± 0.13a[c]	1.63 ± 0.12a[b,c]
Commercial pectin I	1.27 ± 0.06a[c]	1.29 ± 0.20a[d]	1.50 ± 0.14a[c]	1.35 ± 0.03a[d]	1.36 ± 0.14a[c]
Commercial pectin II	2.24 ± 0.16b[a]	2.49 ± 0.07a[a]	2.54 ± 0.07a[a]	2.52 ± 0.12a[a]	2.55 ± 0.17a[a]
Citrus pectin (Sigma)	1.50 ± 0.04a[c]	1.72 ± 0.23a[b,c]	1.78 ± 0.14a[b]	1.62 ± 0.07a[c,d]	1.75 ± 0.19b[b]

[a]Values with same roman letter in each row and same roman superscript in each column are not significantly different ($P < 0.05$) from each other. See Table 1 for abbreviation.

solubility. The solubility of soy hull pectins was comparable to that of citrus pectin (Sigma Chemical Co.) and significantly higher than that of commercial pectin I. The lower solubility of commercial pectin I (DE = 76.2) may be due to the presence of high levels of ester groups. Usually a high-speed mixer with superior shearing action is used to prepare pectin solutions, and blending with sugars enhances the solubility of pectins.

Rheology of pectin solution. The shear stress and apparent viscosity as a function of shear rate for the soy hull pectins, commercial pectins, and analytical-grade pectin are presented in Figures 2 and 3, respectively. Shear stress and apparent viscosity for all the soy hull pectins were higher than for the commercial pectin II. Soy hull pectin at hull/solvent ratios 1:10 and 1:15 was comparable to analytical-grade pectin. Commercial pectin I had the highest, and commercial pectin II had the lowest shear stress and viscosity among the pectins tested. Commercial pectin I also had the least solubility (Table 3) and highest DE (76.2) values among the pectins tested. Pectins prepared at higher hull/solvent ratios tended to have lower shear stress and viscosity than pectins prepared at lower hull/solvent ratios. Since all these pectins had similar DE values, differences in rheological properties may be due to higher degree of acid hydrolysis of pectin molecules with more solvent. Hence, flow behavior of soy hull pectin solutions depends on the solubility and chain length of the molecules as well as on their DE values.

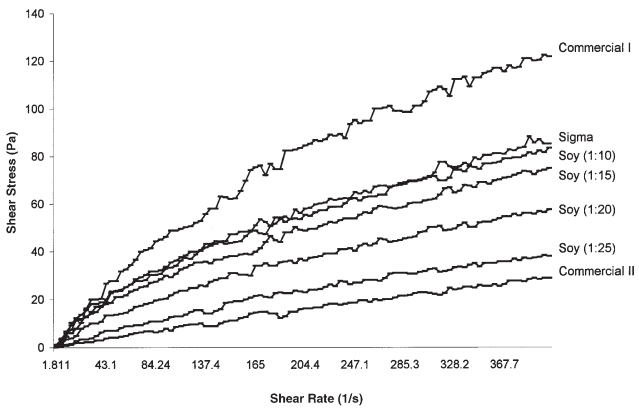


FIG. 2. Changes in shear stress as a function of increasing shear rate [1/s] of pectin solutions: commercial pectin I, Sigma pectin, soy hull pectin at various hull/solvent ratios (1:10, 1:15, 1:20, and 1:25 respectively), and commercial pectin II.

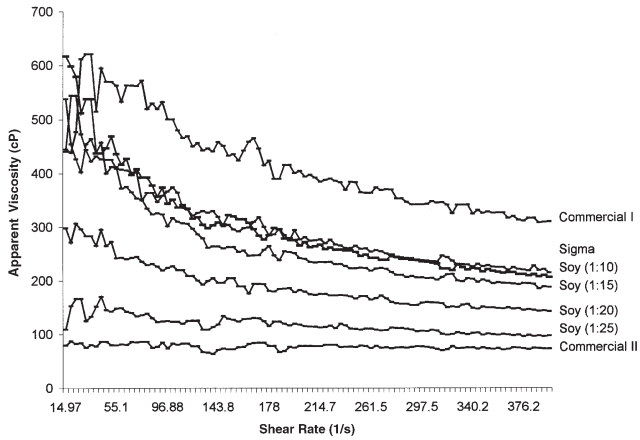


FIG. 3. Changes in apparent viscosity (cP) as a function of increasing shear rate [1/S] of pectin solutions: commercial pectin I, Sigma pectin, soy hull pectin at various hull/solvent ratios (1:10, 1:15, 1:20, and 1:25, respectively), and commercial pectin II.

This study showed that a yield of 16% soy pectin with *ca.* 68% galacturonic acid was produced in pilot plant-scale production. The pectin content, yield, and functional properties of soy hull pectin were within the range of the commercial pectins and analytical-grade pectins.

ACKNOWLEDGMENT

This project was funded by the U.S. Department of Agriculture (NRI Competitive Grants Program, Project # 1822). We wish to thank Riceland Foods (Stuttgart, AR) for providing the soybean hulls.

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[Received January 16, 2001; accepted April 16, 2001]