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Composite Films from Pectin and Fish Skin Gelatin or Soybean Flour Protein

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Composite films were prepared from pectin and fish skin gelatin (FSG) or pectin and soybean flour protein (SFP). The inclusion of protein promoted molecular interactions, resulting in a well-organized homogeneous structure, as revealed by scanning electron microscopy and fracture−acoustic emission analysis. The resultant composite films showed an increase in stiffness and strength and a decrease in water solubility and water vapor transmission rate, in comparison with films cast from pectin alone. The composite films inherited the elastic nature of proteins, thus being more flexible than the pure pectin films. Treating the composite films with glutaraldehyde/methanol induced chemical cross-linking with the proteins and reduced the interstitial spaces among the macromolecules and, consequently, improved their mechanical properties and water resistance. Treating the protein-free pectin films with glutaraldehyde/methanol also improved the Young's modulus and tensile strength, but showed little effect on the water resistance, because the treatment caused only dehydration of the pectin films and the dehydration is reversible. The composite films were biodegradable and possessed moderate mechanical properties and a low water vapor transmission rate. Therefore, the films are considered to have potential applications as packaging or coating materials for food or drug industries.

KEYWORDS: Pectin; fish skin gelatin; soybean flour protein; composite films; cross-linking

INTRODUCTION

The U.S. fruit juice and sugar beet processing industries produce about 10^8 tons of orange peel and sugar beet pulp annually. Residues from these industries could generate $>10^6$ tons of purified pectic polysaccharides. However, only about 0.1% of the potential pectin is produced, and most of it is used in the food industry (*1*). The development of nonfood applications for pectin presents a new strategy to profitably use these underutilized carbohydrates. Recent work in our laboratory has shown that pectin can be used to prepare delivery systems for controlled drug release, for implantable cell carriers in tissue engineering (*2*−*5*), and for prebiotics (*6*, *7*). In earlier work, pectin films made from the blends of high-methoxyl pectin and high-amylose starch or poly(vinyl alcohol) appear to be suitable for some commercial applications where biodegradability is required (*8*−*11*). These applications include drug encapsulation and tablet coating in the pharmaceutical industry (*12*) and disposable packaging materials for food and household products (*13*). Although the pectin blend films showed a higher modulus and tensile properties than a large number of other polymeric films, the pectin−starch films swell upon exposure to moisture and dissolve in contact with water. Studies also showed that pectin-derived films or gels appeared to be effective in food protection with low-moisture foods, but were poor moisture barriers (*14*−*18*). Poor processing endurance and high water susceptibility are two obstacles limiting the expansion of pectin film applications.

We previously proposed the development of alternative pectin composite films, prepared by the substitution of rigid starch fillers with several proteins, such as bovine serum albumin (BSA), chicken egg albumin (CEA), type B bovine skin gelatin (BSG), type A porcine skin gelatin (PSG), fish skin gelatin (FSG), and type I soybean flour protein (SFP) (*19*, *20*). The pectin and protein composite films are expected to possess diverse physical, biological, and chemical properties, which can be tailored to satisfy various applications. In our preliminary studies, the pectin−protein composite films showed appreciable increases in storage modulus and loss modulus and better moisture resistance even at a higher relative humidity in comparison with pectin films or pectin−starch films (*20*). In an attempt to develop pectin as edible films for food and drug coating or packaging, here, we report an extended study performed on pectin−FSG and pectin−SFP composite films. Gelatin and oilseed proteins have demonstrated a good film-forming property and have a long history of safe use in the food and food packaging industries (*17*, *18*). Furthermore, unlike

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bovine-derived gelatin, both FSG and SFP seem to be acceptable as edible food ingredients by large populations with diverse backgrounds.

In this paper, we report the results of the structural and mechanical properties of these new composites, as well as their dissolution behavior and moisture permeability.

MATERIALS AND METHODS

Materials. Citrus pectin (galacturonic acid content, 79%; degree of methyl esterification, 71%) was obtained from Danisco-Cultor (Kansas City, KS) and used as received. FSG (type B, ~95%), SFP (52−56%), glycerol (reagent grade, 99+%), and paraffin wax were purchased from Sigma-Aldrich (St. Louis, MO). Deionized water (DI water) was prepared using a Barnstead E-pure water system (Dubuque, IA) and used to prepare all aqueous solutions.

Film Preparation. Films were prepared as described previously (*8*, *20*). Briefly, pectin solution (pH 8.5) was mixed with protein solutions, FSG (pH 7.5) or SFP (pH 9.0), to produce pectin−protein mixtures with 10 and 20% protein contents (weight percent of total solid). The pH of the mixtures was adjusted to pH 4.0 using 0.01 N HCl, and a small volume of DI water was added to reach a total solid amount of 5%, w/v. To 500 mL of the pectin−protein mixture was added 10.72 g of glycerol, and the mixture was stirred vigorously. The resultant gel was degassed and spread on a polypropylene plate using a film applicator (Paul N. Gardner Co., Pompano Beach, FL) followed by air-drying for 48 h and vacuum-drying for an additional 2 h. The thickness of dry films thus prepared was 0.15 ± 0.01 mm. For film cross-linking, dried films were immersed in 0.1% (w/v) glutaraldehyde/methanol overnight at room temperature. The films were then washed with pure methanol several times, air-dried for 4 h, and stored in a desiccator at room temperature. All experiments were done at room temperature.

Scanning Electron Microscopy (SEM). Specimens of films were mounted with adhesive to specimen stubs, and the edge was painted with colloidal silver adhesive. The specimens were then sputtered with a thin layer of gold and examined in the high-vacuum/secondary electron imaging mode of a Quanta 200 FEG scanning electron microscope (SEM, FEI, Hillsboro, OR) (*21*). Digital images were collected at 500×, 5000×, and 50000×.

Mechanical Testing. Mechanical property measurements performed on the films included tensile strength, tensile modulus, and elongation. These properties were measured using an upgraded Instron mechanical property tester, model 1122, equipped with Testworks 4 data acquisition software (MTS Systems Corp., Minneapolis, MN) (*22*). Samples of pectin−FSG and pectin−SFP films were tested at the following settings: gauge length (clamp distance), 102 mm; strain rate (crosshead speed), 50 mm/min.

Acoustic Emission (AE) Analysis. AE measurements were performed simultaneously during tensile stress−strain tests for all samples. A small piezoelectric transducer was clipped against the sample specimen. This transducer (model R15, Physical Acoustics Corp., Princeton Junction, NJ) resonates at 150 kHz and is 10 mm in diameter, and it was coated with a very thin film of petroleum grease (Dow Corning Corp., Midland, MI) for more efficient acoustic coupling. AE signals emanating from this transducer when the Instron stretched the specimens were processed with a model 1220A preamplifier and an upgraded LOCAN-AT acoustic emission analyzer connected to a PC with enhanced graphing and data acquisition software: SPARTAN-2000, LAU-LOC (Physical Acoustics Corp.) (*23*).

Water Resistance. All specimens (4 × 2 cm) were predried under house vacuum for 48 h prior to test.

Dehydration Behavior. The dehydration of the composite films was tested in water at different pH values. Specimens (4 × 2 cm) were placed in 50 mL of 0.1 M acetate (pH 4.0), phosphate (pH 7.2), or Tris-HCl (pH 8.5) buffer solutions for 48 h. The specimens were removed, and the amounts of pectin and protein released from the films into the water were measured by galacturonic acid assay (*24*) and protein BCA assay (*25*), respectively.

Water Adsorption. The amount of water adsorbed on the films was determined by measuring the percentage of weight gain of a specimen after conditioning at 95% relative humidity (over a saturated solution of KNO_3 in DI water) in a desiccator at 22 °C for 2 weeks (*20*) : $(W_t - W_0)/W_t \times 100\%$, where W_0 was the weight of specimens prior to water adsorption experiment and W_t was the weight after conditioning.

Water Vapor Transmission Rate (WVTR) Determination. WVTR was determined according to ISO 2528 (*26*). Briefly, an aluminum container (diameter, D, = 7.5 cm) with anhydrous $CaCl_2$ desiccant was covered with the films on the top and sealed with paraffin wax. The container was conditioned at 95% relative humidity in a desiccator at 22 °C and weighed at desired time points. After the water transfer equilibrated (the gain in mass between two successive weighings was <5%), the WVTR was calculated from the weight increase of the container over time according to

$$\mathrm{WVTR} = w/(tA) \quad (1)$$

where w is the increase (mg) in mass, t (day) is the duration of the experiment, and A is the permeation area (44.16 cm^2). Differences in film thickness were ignored in this study. An empty aluminum cup covered with the film was used as a control.

Experiments for water resistance, mechanical examination, and acoustic emission were carried out five times for each sample. Data are expressed as the mean ± SD. Significance was determined with the use of a Student's t test.

RESULTS AND DISCUSSION

Structural Analysis. Micrographs of pectin film and pectin−SFP and pectin−FSG composite films are shown in **Figure 1**. SEM revealed that the frozen-fractured faces of pectin films containing no proteins had relatively smooth morphology (**Figure 1A**). With the inclusion of proteins, the films were rough, dense, and brittle in appearance. Some irregular particles were uniformly distributed within the pectin phase (**Figure 1B**,**C**). This was more obvious for the composite films with SFP inclusion than with FSG inclusion and could be attributed to the minor components in soybean flour, such as insoluble polysaccharides and fat (**Figure 1B**). For the composite films containing FSG, the protein appeared to be fairly distributed, although some dense rods in micron scales could be identified (**Figure 1C**). This was also seen for the blends of pectin with other proteins, including BSA, CEA, BSG, or PSG (data not shown). Similar results were revealed in a previous study by confocal laser scanning microscopy using fluorescence red 646 labeled BSA and measured at 640/666 nm (excitation/emission) for the protein and at 425/475 nm for the autofluorescence of pectin in two channels (*19*, *20*). In general, in comparison with the films prepared from the mixture of pectin and starch, where the starch is acting more as a filler than as a secondary polymeric component (*9*, *11*), the substitution of starch with FSG or SFP seems to produce a more compatible composite film.

The organization and microstructure of glutaraldehyde-treated pectin−FSG films were examined (**Figure 1D**). The films displayed a well-oriented characteristic with some structural heterogeneity. The polymers were organized into small packs in regular shape of 100−300 nm scale. This can be seen more clearly on a higher magnification (**Figure 2**). For glutaraldehyde-mediated protein−polysaccharide interaction, it has been demonstrated that the cross-linking, under current experimental conditions, occurred between the active aldehyde and primary amines of the proteins and could not be referred to the hydroxyl groups of polysaccharides (*27*). The cross-linked protein networks appeared to dominate the structure of the composite films. It could be deduced that the glutaraldehyde treatment introduced protein interactions via bridging of their $-NH_2$ groups; resultant protein packs were arranged into fibers, and the fibers were then parallel to each other to form a tightly

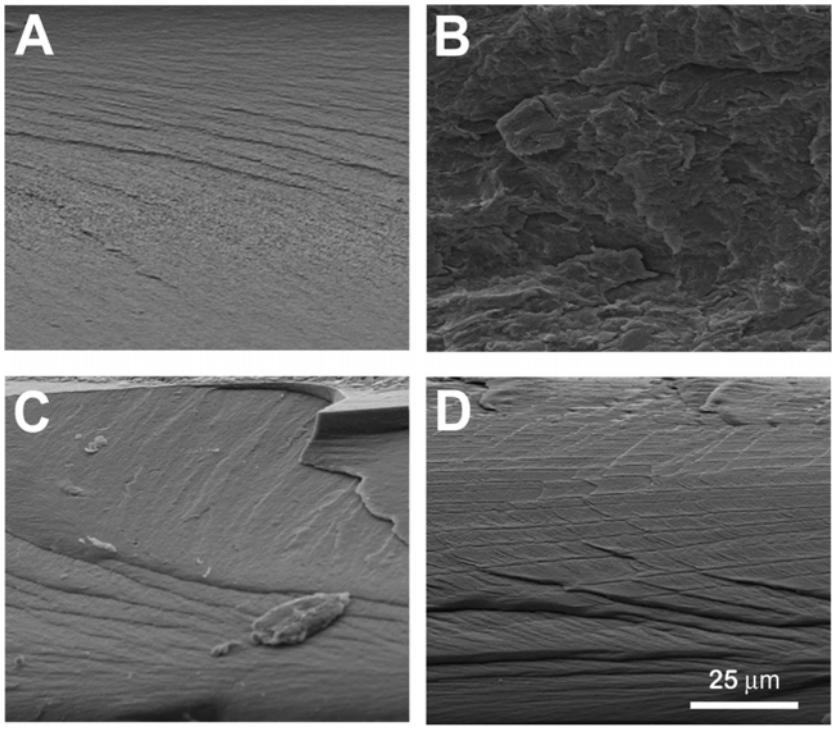

Figure 1. SEM photograph of frozen-fractured pectin films containing no protein (**A**), 10% SFP (**B**), 10% FSG (**C**), and glutaraldehyde-treated pectin/FSG film (**D**).

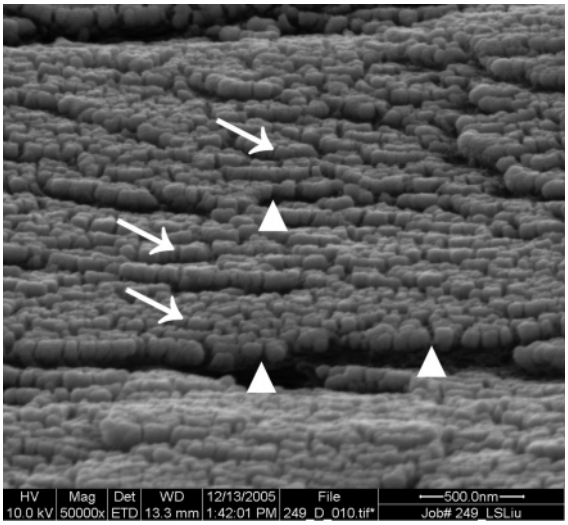

Figure 2. SEM photograph of frozen-fractured pectin/FSG film pretreated with 0.1% glutaraldehyde in methanol, showing gaps (arrow) between adjacent packs with the size of <5 nm, and crevices (triangle) with the size ranging from 50 to 100 nm between parallel fibers.

Table 1. Mechanical Properties of Pectin/Proteins Composite Films[a]

natérial	tensile modulus (MPa)	tensile strength (MPa)	elongation (max) (%)
pectin●,#,▲,§	1082 ± 168	17.0 ± 3.4	2.5 ± 0.6
pectin-gt*[b]●	4139 ± 766	59.2 ± 11.1	1.7 ± 0.5
FSG‡	1906 ± 34	71.8 ± 0.9	6.4 ± 2.4
FSG-gt‡	3132 ± 319	99.2 ± 6.1	3.6 ± 0.6
pectin–FSG (0.1)[c,#,∧]	1825 ± 43	43.5 ± 7.6	3.0 ± 1.5
pectin–FSG (0.1)-gt†,∧	3306 ± 86	54.2 ± 6.9	2.1 ± 0.4
pectin–SFP (0.1)§,⊗	1213 ± 286	24.0 ± 3.0	2.9 ± 0.9
pectin–SFP (0.1)-gt†,⊗	2158 ± 113	33.0 ± 1.7	1.7 ± 0.4
pectin–FSG (0.2)▲	2178 ± 224	59.1 ± 12.4	3.2 ± 1.1
pectin–FSG (0.2)-gt	3016 ± 58	58.9 ± 1.8	2.6 ± 0.3

[a] The following symbols indicate statistical significance ($p < 0.01$): #, †, ‡, ▲, ●, ∧, §, ⊗. [b] -gt* indicates the treatment of film with 0.1% glutaraldehyde/methanol. [c] Data in parentheses indicate the weight percent of protein in composite.

packed, nonwoven structure. Supposedly, the "free" pectin aggregates were penetrated through the protein packs (*28*). The micrographs also showed there were gaps between adjacent packs with the size of <5 nm (**Figure 2**, indicated by arrow), forming closer connections, and crevices ranging from 50 to 100 nm between parallel fibers (**Figure 2**, indicated by triangle), forming relatively loose connections.

Mechanical Properties. The mechanical properties of the composite films, such as stiffness, strength, and flexibility, are given in **Table 1**. Tensile strength is defined as the tensile stress at which the film fractures. Elongation refers to the ability of a material to lengthen or stretch, which is defined as the percent strain at fracture, when a tensile stress is applied to it. The inclusion of FSG or SFP remarkably enhanced both the tensile strength and the elongation of pectin films. In this study, the pectin and pectin−protein films were also treated with glutaraldehyde/methanol. The chemical treatment induced chemical cross-linking to the composites (*27*), which further strengthened the films. The chemical treatment also has a tendency to produce a stiffer composite as indicated by the increase in tensile modulus and the decrease in elongation. The chemical treatment improved the stiffness and tensile strength of pectin films, but these films were unable to retain their flexibility (**Table 1**). It is worth noting that methanol is a dehydration reagent. Although aldehyde-initiated cross-linking could be omitted for pectin films, the methanol could reduce the free spaces between pectin macromolecules and, consequently, enhance the polysaccharide chain−chain interactions, such as hydrogen bonding and hydrophobic interactions, resulting in stiffer pectin films.

Adequate tensile strength is very important in manufacturing polymeric films, where the material is often subjected to a force

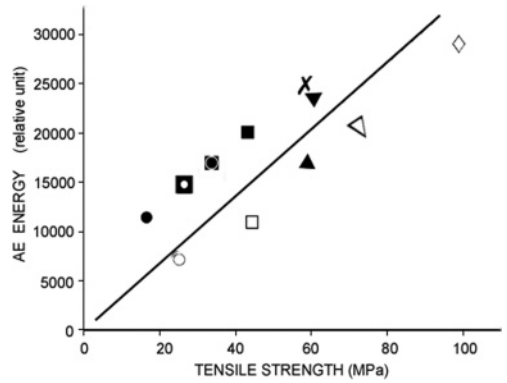

Figure 3. Acoustic energy versus tensile strength of various pectin/protein films: pectin (closed circle), pectin-gt (X), FSG (open triangle), SFG-gt (diamond), pectin–FSG (0.1) (open square); pectin–SFP (0.1) (open circle), pectin–SFP (0.1)-gt (closed circle in a square); pectin-FSG (0.2) (up closed triangle); pectin–FSG (0.2)-gt (closed down triangle); pectin–SFP (0.2) (open circle in square); pectin–SFP (0.2)-gt (closed square).

during mechanical stretching. In a variety of end uses, products must be capable of resisting considerable stress without fracture. The pectin−protein composite films exhibit tensile strengths as high as 24−59 MPa (**Table 1**). By comparison with the tensile strength of 29 MPa for biodegradable blends from SFP and carboxymethylated corncob (*29*), 35 MPa for nonbiodegradable polyvinyl chloride, and 55 MPa for polystyrene (*30*), the composite materials presented in the current study appear to be promising candidates for biodegradable wrapping and packaging materials.

Acoustic Emission Studies. Besides tensile strength, it is also important to know the fracture mechanism of a film when it is stretched or compressed. The fractural characteristics indicate the limitation of end use when the films function as packaging materials and also reflect the mouth feeling and texturizing properties when the film is an element of foods. We used AE techniques to probe the deformation and fracture mechanisms of composite films caused by an external force. During a tensile test, composite deformation and fracture are accompanied by a rapid movement, relocation, or breaking of structural elements such as fillers, fibers, matrices, and their interfacial areas. As a result, sound waves are produced that can be detected by an acoustic transducer and converted into electronic signals by an AE analyzer as a "hit" (*23*). We examined the acoustic emission for all samples simultaneously with tensile strength measurement. First, we measured the total elastic energy released by an acoustic event in response to the maximal stress that specimens were subjected to. **Figure 3** demonstrates the correlation between acoustic energy and tensile strength. In general, stronger films require greater mechanical force to fracture than weaker films, thus producing more mechanical energy and generating more transient elastic waves by detected acoustic emission (*31*). There is a clear correlation between the tensile strength and AE energy released at fracture. However, as shown in **Figure 3**, some spreading is also observed for this plot. For instance, the AE energy released for composite films with 10% proteins was detected lower than those of protein-free pectin films [comparing pectin film (●) with the composite films of 10% SFP (○) or FSG (□) shown in **Figure 3**]. The AE energy measurement is not only a function of the acoustic emissions alone but is also affected by structural factors (*31*); examples include signal attenuation due to scattering or absorption losses during sound wave propagation from the AE source to the transducer and internal energy dissipation by the friction and toughening mechanisms. Some preliminary results suggested that proteins might function as a lubricant in the blend structure (*23*, *32*). Observations indicated that the composite films, after blending with an adequate amount of protein, might be able to abate wave propagation. Moreover, data also showed AE energy increases with protein content or with the protein cross-link density. These studies are intended to elucidate the relationships between microstructural events and macroscopic behavior of the blend materials.

We further studied the correlation between the stress−strain curve and AE hit rate pattern. The stress−strain curve indicates property changes under an external force, whereas the hit−strain curve indicates the structural changes. By referring one to another, we are able to better understand the fracture mechanisms and the structure−property relationship of a sample. In the current experiment, five specimens were tested for each sample. The results from each test were similar, and the differences in shape and position in the stress−strain−AE hit plots were negligible. The typical curves are shown in **Figure 4**. For films consisting of a single component, either pectin or FSG, the specimens behaved as a uniform material regardless of whether or not they were chemically treated. There were no acoustic events before the peak stress; the AE activities occurred exclusively at the peak stress when the specimens completely fractured. This behavior is due to their homogeneous structure, in which the single component specimens were able to transfer the stress evenly. In contrast, the composite films emitted sound at an earlier strain due to the microstructural movement of individual components, which was correlated to the increase in the initial slope of the stress−strain curves (**Figure 5A**,**C**). Observation also showed that a sudden increase in AE hits occurred at the peak stress. Furthermore, AE hits are more frequent and are more evident for cross-linked films than for those not cross-linked. Composite cross-linked films produced a wide band of acoustic waves at a much earlier strain (**Figure 5B**,**D**). This behavior may be ascribable to structural defects such as crevices found between fibers in cross-linked blend films (**Figure 2**). If the direction of those crevices and the direction of film elongation coincide, AE events would be further enhanced. More experiments are required to confirm it.

Water Resistant Properties. Results of dissolution/swelling experiments showed that protein-free pectin films dissolved in water. Film dissolution is pH-dependent; the dissolution rate increased in the sequence of pH 4.0 < pH 7.2 < pH 8.5. The pretreatment of the pectin films with glutaraldehyde/methanol did not stop, but did slow, the dissolution process. The pectin−FSG and pectin−SFP films did not dissolve at any pH tested, but did display a pH-dependent swelling behavior. Both composite films swelled least at pH 4.0. As the solution pH increased, the composite films were more swelled. Measurements at the same pH revealed that pectin films containing SFP swelled into a larger size than pectin films containing an equal amount of FSG. The pH influences on the dissolution of polysaccharide−protein films have been well studied. The effect of pH on the dissolution of the paired biopolymers has been referred to the electrostatic interactions between these biopolymers. These electrostatic interactions in turn are responsible for the mechanism of the formation−deformation of the electrostatic complex (*28*, *33*, *34*). Accordingly, the values of pK_a or p*I*, respectively, are around 4 for pectin (*28*), 4.8−5.2 for FSG (*35*), and 4.5−5.1 for SFP (*36*); electrostatic complexes between pectin and proteins could be formed at pH 4 solution. This may stabilize the films and suppress their swelling.

Because the mechanical properties and other physical properties of the films are the function of their compositions, we are

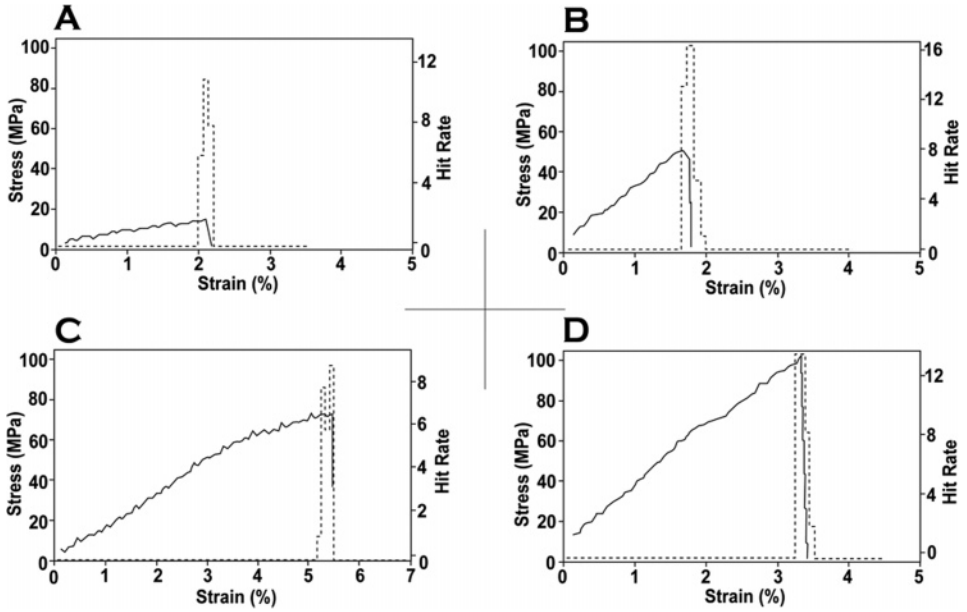

Figure 4. Relationship of strain–stress curve (solid line) with the acoustic emission hits (dotted line): pectin (**A**) and FSG (**C**) films; glutaraldehyde/methanol-treated pectin (**B**) and FSG (**D**) films.

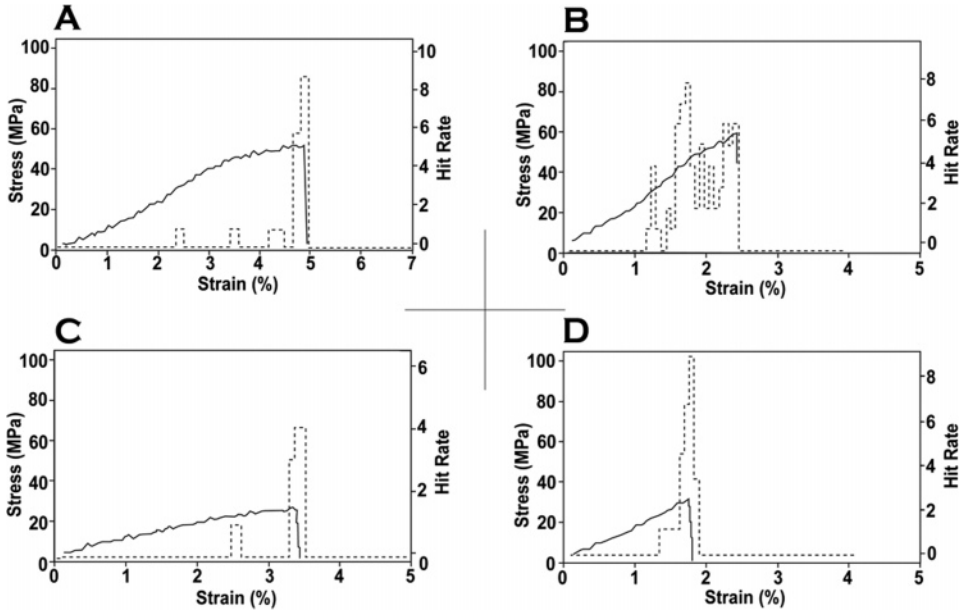

Figure 5. Relationship of strain–stress curve (solid line) with the acoustic emission hits (dotted line): pectin–FSG (**A**) and pectin–SFP (**C**) films; glutaraldehyde/methanol-treated pectin–FSG (**B**) and pectin–SFP (**D**) films.

interested in knowing the change in compositions during dissolution. We measured the release of pectin and protein from the swelling composites (**Figure 6**). For the composite films without chemical treatment, about 50% of pectin or proteins were released into the dissolution buffer at pH 4.0 in 48 h (**Figure 6A**), whereas the values increased as the solution pH increased and reached the highest decomposition around 80−90% at pH 8.5 (**Figure 6B**,**C**). The chemical treatment did not influence the release of pectin from composite films, but did suppress the release of protein. This was more pronounced for the films containing FSG than SFP. As shown in **Figure 6**, <20% of the incorporated FSP released into the three dissolution buffers, whereas the SFP release seemed to be more pH-dependent even after chemical treatment. This could be attributed to more primary amine being available in FSG than in SFP, which generated more cross-linked bonds with FSG films than with SFP films (*35*, *37*). As shown in **Figures 1D** and **2**, the cross-linked proteins formed a tightly packaged network, which, in turn, limited the mobility of the un-cross-linked polysaccharides.

Table 2 shows the water adsorption and water vapor transmission properties of the films. The protein-free pectin films showed the highest values in both water adsorption and water vapor permeability. The chemical treatment showed little impact on water adsorption of pectin films but dramatically reduced the water vapor permeability of the films. As discussed in the above section, film dehydration in anhydrous methanol may enhance the level of chain−chain packing and reduce interstitial spaces among the pectin molecules. The interstitial space is a measure of film free volume, which determines the mass permeability (*28*, *38*). After conditioning in a more humid environment for a longer period, that is, at 95% relative humidity

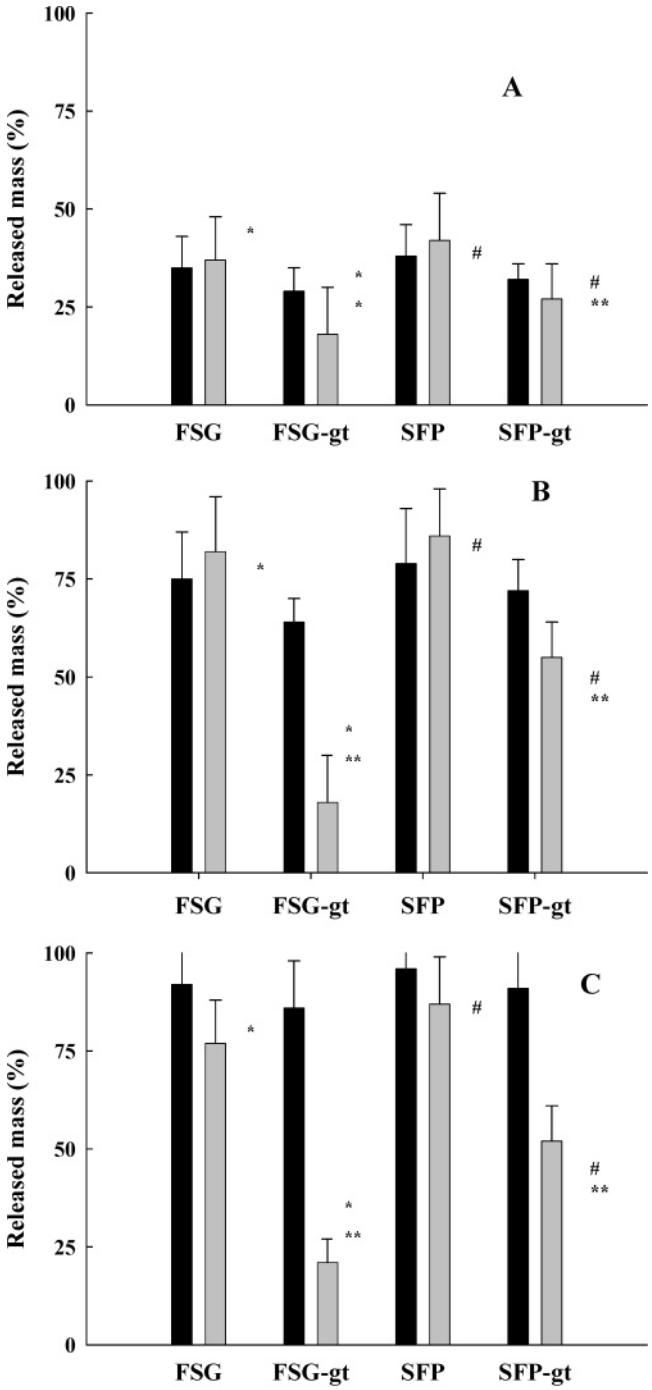

Figure 6. Pectin (black column) and protein (gray column) released from composite films containing 20% FSG or SFP after incubation at room temperature for 48 h at pH 4.0 (**A**), pH 7.2 (**B**), and pH 8.5 (**C**). The suffix -gt indicates treatment with glutaraldehyde/methanol solution. Data are expressed as the mean ± SD ($n = 5$): $*$, $^{\#}$, proteins released from chemically treated films versus relative nontreated films, $p < 0.01$; $**$, proteins released from chemically treated pectin–FSG film versus pectin–SFP film, $p < 0.01$.

for 2 weeks as in this study, the dehydrated films could gradually be rehydrated; thus, their water adsorption capability could be partially recovered.

For pectin−FSG composite films, both water adsorption and water vapor permeability were suppressed. The smaller values of water adsorption and WVTR were obtained at higher FSG content (**Table 2**). The chemical cross-linking further suppressed water adsorption and water vapor transmission (**Table 2**); these values were even lower than those obtained for transglutaminase-modified protein and pectin−protein films (*28*, *39*). The blends of SFP with pectin and the subsequent chemical cross-linking showed an impact on water adsorption and penetration, which had the same trend as the inclusion of FSG but smaller (**Table 2**). It is consistent with the results from the dissolution studies.

Table 2. Water Resistant Property of Pectin/Protein Films

material	water adsorption[b] (%)	WVTR[c] ($g \times m^{-2} \times day^{-1}$)
pectin$^{\otimes,\S,\perp,\wedge,\blacklozenge}$	47 ± 6	226 ± 11
pectin–gt$^{d*,\otimes}$	42 ± 7	178 ± 23
FSG$^{\blacktriangle}$	28 ± 4	98 ± 11
FSG-gt$^{\blacktriangle}$	6 ± 2	112 ± 15
pectin–FSG (0.1)$^{e,\#,\perp,\S}$	32 ± 4	147 ± 7
pectin–FSG (0.1)-gt§,†	12 ± 2	103 ± 12
pectin–SFP (0.1)$^{\perp,\ddagger}$	37 ± 5	196 ± 16
pectin–SFP (0.1)-gt$^{\perp,\dagger,\ddagger}$	17 ± 6	158 ± 17
pectin–FSG (0.2)$^{\blacklozenge,\S}$	26 ± 4	121 ± 10
pectin–FSG (0.2)-gt§	8 ± 3	114 ± 9
pectin–SFP (0.2)$^{\wedge,\bullet}$	29 ± 6	184 ± 15
pectin–SFP (0.2)-gt$^{\bullet}$	11 ± 2	161 ± 18

[a] The following symbols indicate statistical significance ($p < 0.01$): ⊗, §, ⊥, †, ♦, ‡, ●, ▲, ^. [b] Measured by weight gain after conditioning at 95% relative humidity at room temperature. [c] Determined by ISO 2528 (1995E). [d] -gt* indicates the treatment of films with 0.1% glutaraldehyde/methanol solution. [e] Data in parentheses indicate the weight percent of protein in composite.

Conclusions. The results presented in this study indicate that inclusion of proteins into pectin films improved both mechanical strength and flexibility. The treatments of resultant composite films with glutaraldehyde/methanol further enhanced film strength and reduced water vapor permeability, while retaining the flexibility of the original pectin films to some degree. However, it appears that only chemical cross-linking can suppress the films' solubility in water, because methanol treatment is a dehydration process that reduces only the interstitial spaces between macromolecular chains and is reversible. The results suggest the potential of pectin and protein composite films in the applications of wrapping or packaging materials compared to other commercial films, which requires moderate mechanical strength and low water vapor transmission.

Acoustic emission reflects structural changes of a material during and at fracture. AE analysis records the kinetics of defect formation as the materials are stretched to failure, serving as a complement to the microscopic study for structural investigation. AE analysis correlated well with the SEM results and suggested that an improved cross-linking method or procedure is required to eliminate the crevices and gaps formed during cross-linking in order to enhance the mechanical properties of the resultant composite films.

Although it was not the goal of this study, we found that the composite films from pectin and a small fraction of FSG or SFP are able to abate sound propagation, comparing the fracture energy with elastic energy released at fracture. This finding suggests the potential to develop sound protection materials from such kinds of biomass.

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