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Acetylation of Chicken Feathers for Thermoplastic Applications

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ABSTRACT: Poultry feathers are renewable resources, inexpensive and abundantly available, but have limited applications. Although keratin extracted from feathers has been chemically modified, there are no reports on the chemical modification or development of thermoplastics from poultry feathers. Acetylation is an inexpensive and environmentally friendly approach to make biopolymers thermoplastic. Several biopolymers have been acetylated and used to produce fibers, films, and extrudates. In this research, chicken feathers were acetylated, and the structure and properties of the acetylated feathers were studied. Acetylation conditions such as concentration of chemicals and catalyst and time and temperature of acetylation were optimized. Acetylation of feathers was confirmed using Fourier transform infrared (FTIR) and pyrolysis—gas chromatography—mass spectrometry (P-GC-MS). The acetylated feathers were analyzed using thermogravimetric analysis (TGA) and differential scanning calorimetry (DSC) to understand their thermal behavior. Acetylated feathers were thermoplastic and could be compression molded to form transparent films despite the relatively low percentage of acetyl content.

KEYWORDS: chicken feather, acetylation, thermoplastics

INTRODUCTION

Poultry feathers are inevitably generated as byproducts and are available in large quantities but have limited industrial applications. Feathers are renewable and biodegradable, and utilizing the feathers for industrial applications would help to add value to feathers and benefit the poultry industry. However, the >4 billion pounds of poultry feathers generated in the United States every year are mostly disposed of in landfills. This not only leads to the discarding of a potentially valuable polymer but also causes environmental concerns.[1−3] In addition, this valuable raw material is >90% protein (keratin). Technologies have been developed to clean poultry feathers and separate them as feather fibers (barbs) and quills on a commercial scale as a raw material for various applications.

Efforts have been made to utilize feathers (feather fibers and/or quill) as reinforcement for composites with natural and or synthetic matrix materials. Feathers have low density and a unique hierarchical structure, which makes them preferable for composite applications, especially lightweight composites for automotive applications.[1,2] Commercially available feather fibers and powdered quill have been used as reinforcements for lightweight composites.[1,2] It was found that feathers provided similar flexural and tensile properties but better acoustic properties than common reinforcing materials such as natural cellulose fibers. It has also been reported that using whole feathers in their native form (with barbs and quill) provides better flexural properties compared to using feather fiber and powdered quill as reinforcement for lightweight composites.[4]

Studies are also available on the modification of keratin extracted from feathers for various applications. Feather keratin was graft polymerized using 2-hydroxyethyl methacrylate and was studied as part of the composition for fertilizers.[5] In another paper, feathers were graft polymerized with methyl methacrylate for potential use as reinforcement for composites.[6] It has recently been demonstrated that grafting with methyl methacrylate made feathers thermoplastic and suitable to be compression molded into films.[7]

Acetylation is a common approach to make biopolymers thermoplastic; it is relatively inexpensive and environmentally friendly. Acetylated feathers will also be more biodegradable than feathers grafted with acrylates.[7] Cellulose acetates are produced on an industrial scale and used for fibers, films, and other applications. Acetylated starch has been reported to be thermoplastic and used to produce various types of injection and compression-molded products.[8,9] We have recently demonstrated that acidic conditions provide better acetylation of distillers dried grains (DDG) and therefore good thermoplasticity than acetylation under alkaline conditions.[10,11] It would be preferable to use acidic conditions for acetylating feathers because alkaline conditions could hydrolyze the feathers. In addition to acetylation, etherification using acrylonitrile has also been shown to provide good thermoplasticity.[12] Thermoplastic films developed from DDG modified by etherification were compression-molded into films with good tensile strength and elongation without the need for plasticizers.[12]

In this research, we have acetylated chicken feathers and optimized the acetylation conditions. The acetylated feathers were characterized for their structure and properties, and the potential of developing thermoplastics from the acetylated feathers was studied.

EXPERIMENTAL PROCEDURES

Materials. Chicken feathers (whole feathers with quill and barbs) were obtained from Feather Fiber Corp., Nixa, MO. The feathers were

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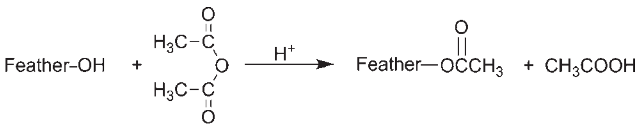
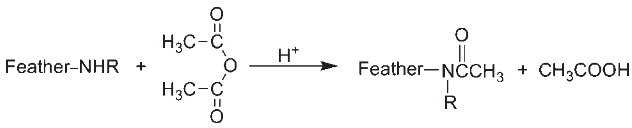
Scheme 1. Possible Reaction between Acetic Anhydride and the Hydroxyl Groups in Feather Proteins under Acidic Conditions[a]



[a] Feather—OH represents the hydroxyl groups in the proteins in chicken feather.

Scheme 2. Possible Reaction between Acetic Anhydride and the Amine Groups in Feather Proteins under Acidic Conditions[a]



[a] Feather—NHR represents primary and secondary amine groups in the proteins in chicken feathers.

washed, cleaned, and mechanically processed to cut the feathers. Acetic acid, acetic anhydride (98% ACS grade), and other chemicals (reagent grade) used for acetylating the feathers were purchased from VWR International, Bristol, CT. All chemicals were used as received without further purification.

Methods. *Acetylation of Chicken Feather.* Chicken feathers were finely ground in a laboratory-scale Wiley mill to pass through a 20 mesh dispenser. The powdered feathers were acetylated using acetic anhydride as the acylation agent, acetic acid as solvent, and sulfuric acid as catalyst. Initially, glacial acetic acid was added to the chicken feather at a weight ratio of 10:1 at room temperature under constant stirring. Acetic anhydride (1:1 to 5:1 acetic anhydride to feather weight ratio) was added to the acetic acid feather mixture. Later, sulfuric acid was added (3−20% on the basis of the weight of the feather), and the mixture was stirred at a temperature below 30 °C. The acetylation was completed by heating the mixture containing feather, acetic acid, acetic anhydride, and sulfuric acid for a specified time (10−120 min) at a specified temperature (50−90 °C). After completion of the reaction, 10% (w/w) aqueous sodium hydroxide was added to neutralize the acid remaining after reaction. The acetylated feathers obtained were thoroughly washed in distilled water at 50 °C for 30 min under constant stirring five times to ensure complete removal of the unreacted chemicals. The feathers were later dried at 40−50 °C for 12 h for further analysis.

The possible reactions between acetic anhydride and the proteins are shown in Schemes 1 and 2. Acetylation occurs on both the hydroxyl and amine groups in feather proteins. Scheme 1 represents the reaction between the hydroxyl groups in the feather proteins and acetic anhydride.[13] Scheme 2 depicts the reaction between the primary and secondary amines in the proteins and acetic anhydride.[14] The reaction between the acetic anhydride and the hydroxyl and amine groups results in the formation of the acetylated feathers.

Acetyl Content. The extent of acetylation of the feathers was quantitatively determined in terms of the percent acetyl content on the basis of the number of acetyl groups on the feathers. The acetyl content is defined as the weight percentage of acetyl (CH_3CO-) groups on the initial weight of feathers used. Determination of acetyl groups was based on the fact that *O*-acetyl can be hydrolyzed by cold dilute NaOH, whereas the *N*-acetyl groups can be removed only by boiling in dilute acid solution.[15] The method used to analyze the total acetyl was similar to that reported by Blackburn for acetylation of wool.[16] Approximately 0.3 g of the acetylated feather was boiled under reflux for 4 h with 10 mL of 2.5 mol/L H_2SO_4. The hydrolysate obtained was distilled, and water was added as necessary until 200 mL of the distillate had been collected. The distillate obtained was titrated using 0.02 mol/L NaOH, and the values obtained were subtracted from the values for the blank titration obtained by the similar hydrolysis and distillation of the unacetylated chicken feathers.

The percent acetyl content was calculated using eq 1

$$\% \text{ acetyl content} = (A - B) \times M \times (F/W) \tag{1}$$

where A is the amount (mL) of NaOH solution required for titration of the sample, B is the amount (mL) of NaOH solution required for titration of the blank, M is 0.02 (the molar concentration of NaOH used for titration), W is the weight (g) of feathers obtained after acetylation, and F is 4.305 for acetyl, which is related to the molecular weight of the acetyl group (CH_3CO-), the unit conversion from liters to milliliters, and fraction to percentage:

$$F = \frac{\text{molwt}}{1000 \text{ mL/L}} \times 100 = \frac{43.05}{10} = 4.305 \tag{2}$$

Percent Weight Gain. Percent weight gain values, which describe the percent increase in the weight of acetylated chicken feather compared to the weight of unmodified chicken feather used for the reaction, were obtained to quantitatively determine the efficiency of acetylating the chicken feather. The acetylated feather was thoroughly washed as described earlier to remove chemicals and soluble impurities and later dried in an oven at 50 °C until constant weight was obtained. The percent weight gain values were calculated according to the formula

$$\% \text{ wt gain} = ((W_{\text{mod}} - W_{\text{unmod}})/W_{\text{unmod}}) \times 100$$

where W_{unmod} is the initial oven-dried weight of the chicken feather before chemical modification and W_{mod} is the oven-dried weight of the acetylated chicken feathers.

Fourier Transform Infrared (FTIR) Spectrum Analysis. FTIR spectra of unmodified and acetylated chicken feather were measured on a Nicolet NEXUS 670 (Thermo-Nicolet, Waltham, MA) FTIR spectrometer using KBr powder at room temperature. The samples were thoroughly washed in distilled water to remove the solvent and catalysts prior to mixing with KBr. Samples in the form of thin films were placed in the cell and measured from 400 to 4000 cm^{-1} with a resolution of 4 cm^{-1}, and 64 scans were collected. The FTIR spectra obtained were analyzed using OMNIC software (Thermo Electron Corp.).

Pyrolysis−Gas Chromatography−Mass Spectrometry Analysis. Pyrolysis was performed in a Chemical Data Systems Pyroprobe 120 pyrolyzer equipped with a platinum coil and quartz sample tube interfaced to a Shimadzu QP 2010 (Japan) GC-MS device. To carry out the analysis, samples of 10−15 mg were pyrolyzed at 200−300 °C for 10 s. A helium carrier gas at a 48.2 mL/min flow rate purged the pyrolysis chamber into a fused silica capillary gas chromatographic column (25 m × 0.2 mm) coated with a bonded methyl silicone phase (0.33 μm). The temperature was 40 °C for 3 min with a temperature ramp of 10 °C/min. The carrier gas was helium, and the split ratio was 50:1. The injector and mass spectrometer interface temperatures were 280 and 300 °C, respectively. The mass spectrometer was operated in electron impact (EI) mode at 70 eV, scanning in the mass range from 33 to 400 atomic mass units (amu). The temperature of the GC-MS interface was held at 300 °C. The acceleration voltage was turned on after a solvent delay of 80 s. The detector voltage was 1100 V. Mass spectral similarity searches were performed using NIST MS Search 2.0 (NIST/EPA/NIH Mass Spectral Library).

Thermal Analysis. Thermogravimetric analysis (TGA) was performed on the unmodified and acetylated chicken feather with an instrument (Perkin-Elmer STA 6000, Norwalk, CT) calibrated with nickel. Samples

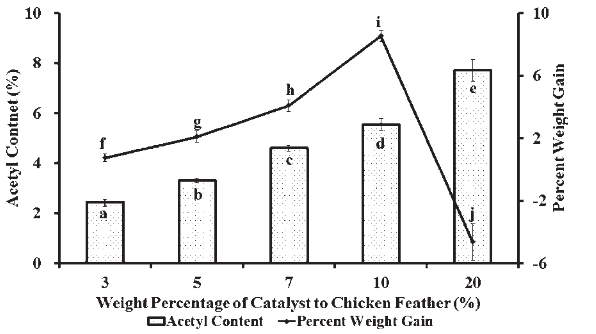


Figure 1. Effects of catalyst to chicken feather ratio (% w/w) on the percent acetyl content and percent weight gain of acetylated chicken feathers. Acetylation was carried out at 70 °C for 60 min with an acetic anhydride to chicken feather ratio of 3:1. Data points with the same letter are not statistically different from each other.

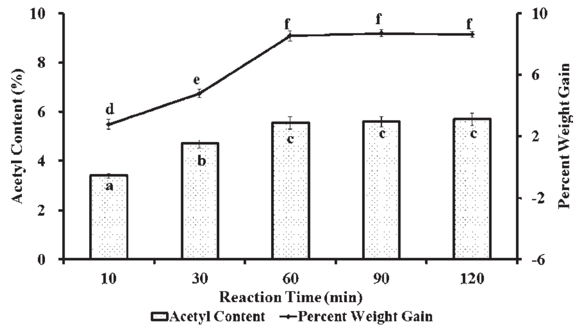


Figure 2. Effects of reaction time on percent acetyl content and percent weight gain of the acetylated chicken feathers. The acetylation was carried out at a temperature of 70 °C with an acetic anhydride to chicken feather ratio of 3:1 and a catalyst concentration of 10%. Data points with the same letter are not statistically different from each other.

(18−26 mg) were placed under nitrogen atmosphere and heated from 50 to 650 °C at a heating rate of 20 °C min^{-1}. A Mettler Toledo (model DSC822[e]) DSC was also used to study the thermal behavior of the unmodified and acetylated feathers. Samples (about 10 mg) oven-dried at 105 °C for 5 h were placed in the DSC and heated at a rate of 20 °C min^{-1} after holding at 50 °C for 10 min to remove moisture in the samples. The samples were then heated to 180 °C at a rate of 20 °C min^{-1}.

Development of Thermoplastics. The unmodified and acetylated chicken feathers were compression-molded in a Carver (Wabash, IN) press to evaluate their thermoplasticity and potential for various thermoplastic applications. Glycerol (20%, w/w) on weight of feathers was used as a plasticizer to improve the thermoplasticity of the feathers. Samples of about 10 g were compressed at 170 °C for 15 min under a pressure of 138 MPa. The press was cooled by running cold water, and the films formed were collected.

Statistics. All of the experiments were repeated three times unless specified. The data reported are the mean ± one standard deviation. Fisher's least significant difference (LSD) was used to test the effect of various conditions on the properties of products using SAS (SAS Institute Inc., Cary, NC). Statistical significance was considered at $p < 0.05$. Any two data points with the same letter are not statistically different.

RESULTS AND DISCUSSION

Effects of Catalyst Concentration on Percent Acetyl Content and Percent Weight Gain of Acetylated Chicken Feathers. Figure 1 depicts the effect of increasing the percent of catalyst (sulfuric acid) on the acetyl content and percent weight gain of acetylated chicken feathers. As seen from Figure 1, increasing catalyst concentration had a considerable effect on the percent acetyl content and percent weight gain of feathers. The highest percent acetyl content of 7.7% was obtained at a catalyst concentration of 20%. The percent weight gain also increased progressively when the catalyst concentration was increased from 3 to 10% but decreased substantially at 20% catalyst concentration. The highest weight gain obtained was about 8.6% with a catalyst concentration of 10%. Increasing the catalyst concentration above 10% increased the percent acetyl content but decreased the percent weight gain due to hydrolysis of the proteins at low pH and high temperatures. Hydrolysis of the feathers leads to small molecules that are easily removed during washing. The feathers remaining after washing have molecules with higher degrees of acetylation and. therefore, there was an increase in the percent acetyl content but a weight loss of 4.6% for the feathers acetylated using a catalyst concentration of 20%.

Effects of Reaction Time on Percent Acetyl Content and Percent Weight Gain of Acetylated Chicken Feathers. The changes in the percent acetyl content and percent weight gain of the acetylated chicken feathers with increasing reaction time are shown in Figure 2. Both the percent acetyl content and percent weight gain showed similar trends with increasing time. Increasing time from 10 to 60 min increased the acetyl content and weight gain, but reaction time above 60 min did not show any statistically significant increase in weight gain or percent acetyl content. The optimum percent acetyl content of 5.6% and percent weight gain of 8.5% were obtained at 60 min. However, increase in reaction time above 60 min did not increase either the percent acetyl content or percent weight gain, indicating that the reaction had reached equilibrium under the conditions studied.

Effects of Reaction Temperature on Percent Acetyl Content and Percent Weight Gain of Acetylated Chicken Feathers. Figure 3 shows the effect of increasing reaction temperature on the percent acetyl content and percent weight gain of the acetylated chicken feather. Increasing reaction temperature from 50 to 60 °C and from 60 to 70 °C increased the acetyl content by about 22 and 30%, respectively. The corresponding increases in percent weight gain were 35 and 39%. However, further increase in temperature above 60 °C did not increase the percent acetyl content or the percent weight gain. An optimum acetyl content of 5.6% was obtained when the reaction was carried out at 70 °C, and the highest percent weight gain of 8.6% was also obtained at 70 °C. Increasing reaction temperature increased the accessibility of the proteins to chemicals and increased the acetyl content and the weight percent. However, most of the available hydroxyl and amine groups have been reacted, and the reaction reaches equilibrium at about 70 °C; we therefore did not see any further increase in the percent weight gain or percent acetyl content above 70 °C.

Effects of Concentration of Acetic Anhydride on Percent Acetyl Content and Percent Weight Gain of Acetylated Chicken Feathers. The effect of increasing the weight ratio of acetic anhydride to chicken feather on the acetyl content and percent weight gain of acetylated chicken feathers is shown in Figure 4. The acetyl content increased continually when the ratio of acetic anhydride was increased from 1:1 to 4:1 but did not increase above an acetic anhydride to feather ratio of 5:1. At a ratio of 1:1, there is insufficient anhydride to react with the hydroxyl and amine groups in the proteins, and hence there was a low level of acetylation and weight gain. Most of the accessible

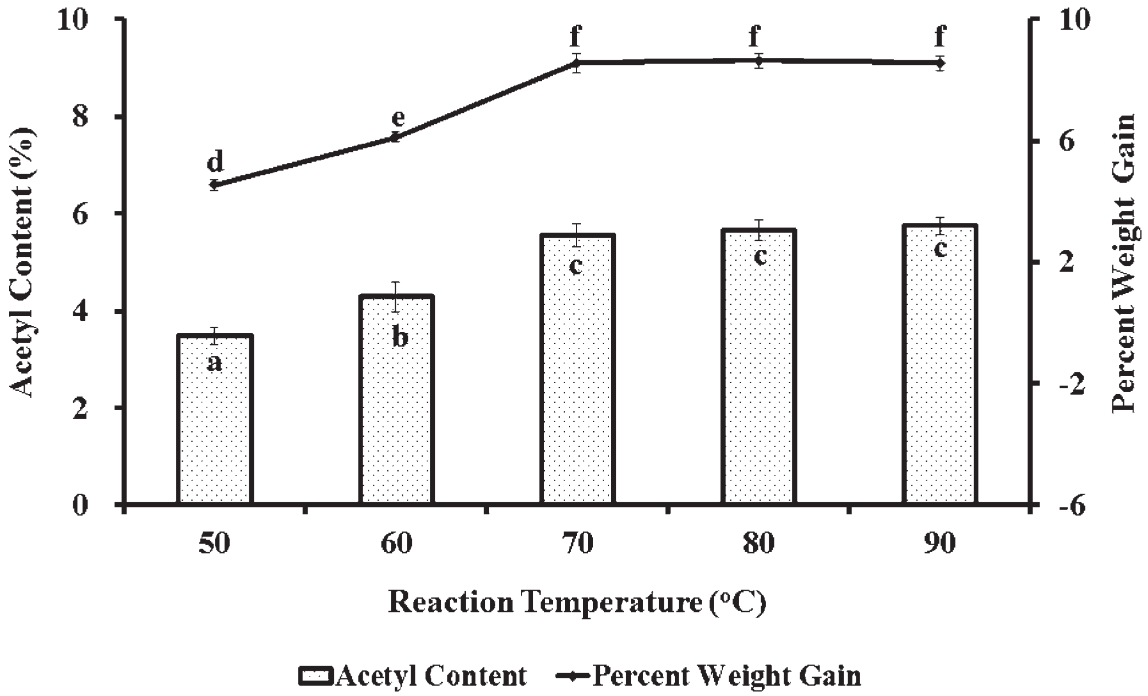


Figure 3. Effect of reaction temperature on percent acetyl content and percent weight gain of acetylated chicken feathers. The acetylation was carried out for 60 min with an acetic anhydride to chicken ratio of 3:1 and a catalyst concentration of 10%. Data points with the same lettes are not statistically different from each other.

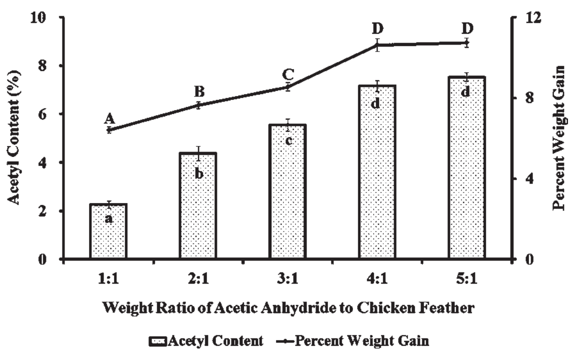


Figure 4. Effect of weight ratio of acetic anhydride to chicken feather on the percent acetyl content and percent weight gain of acetylated chicken feathers. The acetylation was carried out at 70 °C for 60 min with a catalyst concentration of 10%. Data points with the same letter are not statistically different from each other.

hydroxyl and amine groups in the proteins should have been acetylated at an anhydride to feather ratio of 4:1, and hence there was no increase in acetyl content upon further increase in anhydride ratio. The highest acetyl content obtained was 7.5%, and the highest percent weight gain obtained was 10.8% at an acetic anhydride to feather ratio of 5:1.

The percent acetyl content of 7.5% obtained in this research was close to the theoretically possible acetylation of the hydroxyl and amine groups in feathers. The molar ratio of hydroxyl and amine groups on the side chains of the major amino acids (serine, threonine, and arginine) was 219 mmol per 100 g of feathers. We have calculated the percent acetylation on the basis of the moles of the hydroxyl and amine groups in the major amino acids in feathers and the moles of acetyl groups on the acetylated feathers on the basis of an acetyl content of 7.5% as shown in Table 1.

At a maximum acetyl content of 7.5%, before any substantial hydrolysis, the molar ratio of acetyl groups was 188 mmol per 100 g of acetylated feathers. However, some of the hydroxyl and amine groups could be in the crystalline regions and not accessible to acetylation, and therefore the highest percent acetyl content obtained was lower than the maximum possible acetylation of 219 mmol per 100 g of feathers.

P-GC-MS Analysis. The mass spectrometry spectra in Figure 5 showed that the acetylated chicken feather had a sharp peak at about 4.255 min, and there was no apparent peak at this position for the unmodified feathers. Spectral match with the NIST library produced a match of 96% for acetic acid formed during the pyrolysis of the acetyl group.[17] This indicates that the peak in the spectrum for the modified feathers should be from the acetylation of the feathers. In addition, acetylation of the amide groups introduces an extra peak at 3.950 min due to pyrolysis of the amide groups and removes a peak at 3.205 min in the acetylated feathers due to the acetylation of the imine and amine groups.[18,19]

FTIR Measurements. Figure 6 shows the FTIR spectra of the unmodified and acetylated chicken feathers. The presence of an absorbance peak at 1732 cm^{-1} belonging to the stretching of the ester carbonyl C=O group is seen in the acetylated chicken feathers but not in the unmodified feather. Similarly, the appearance of the peak at 3307 cm^{-1} is mainly due to the unfolding of the proteins after acetylation. In addition, the groups may be partially acetylated, and the unacetylated parts cause vibrations of the hydrogen and N−H bonds.[20,21] The increase in the intensity of the amide III peak at 1238 cm^{-1} should be due to the stretching of the C−N group in the secondary amides and the C−C−O stretch of the acetates around 1240 cm^{-1}.[20] Variations in the peak intensities between the unmodified and modified feathers at 2970 and 2882 cm^{-1} should be due to the asymmetric

Table 1. Calculation of the Percent Acetylation Based on the Moles of Hydroxyl and Amine Groups

amino acid	% w/w in feather	mol weight (MW)	MW − water molecule	mol of OH/NH_2 per 100 g of feather	mol of acetyl ($COCH_3$) per 100 g of feather
serine	11.44	105	87	11.44/87 = 0.131	acetylation mol (%)
threonine	4.66	119	101	4.66/101 = 0.046	7.5/43 = 0.174
arginine	6.58	174	156	6.58/156 = 0.042	mol of acetyl per g of feather[a]
				total = 0.219	0.174 × 1.081 = 0.188

[a] Considering a weight gain of 7.5% due to acetylation.

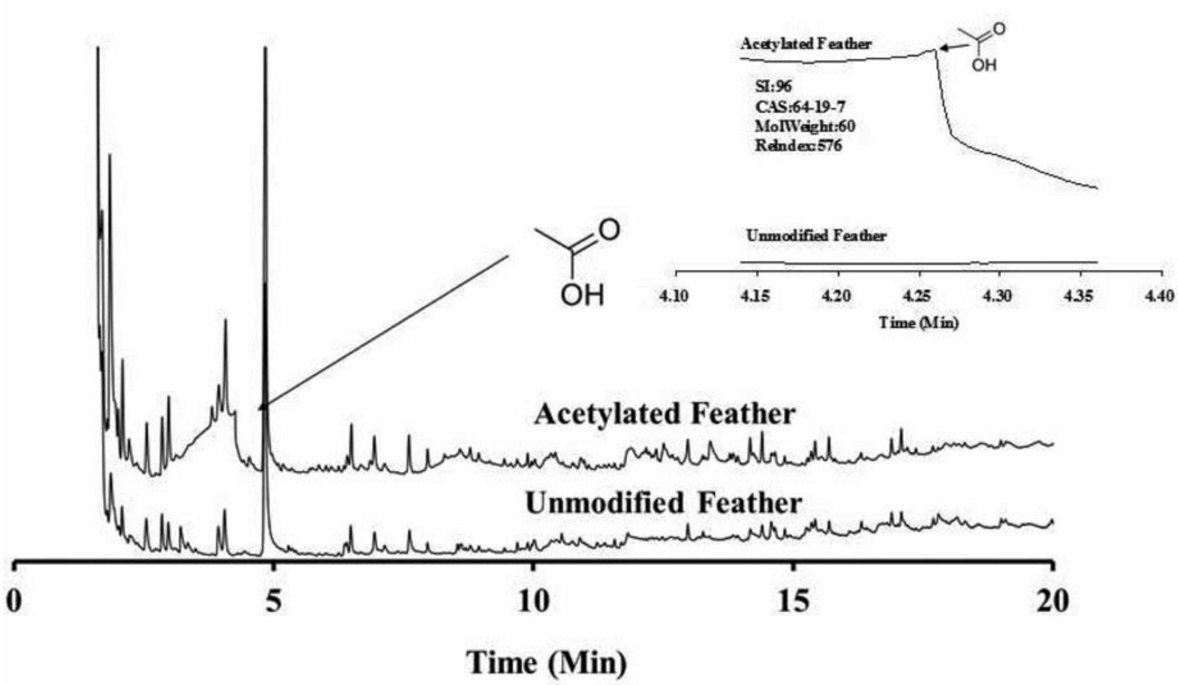


Figure 5. Pyrolysis−gas chromatography−mass spectrometry of the unmodified and acetylated feathers.

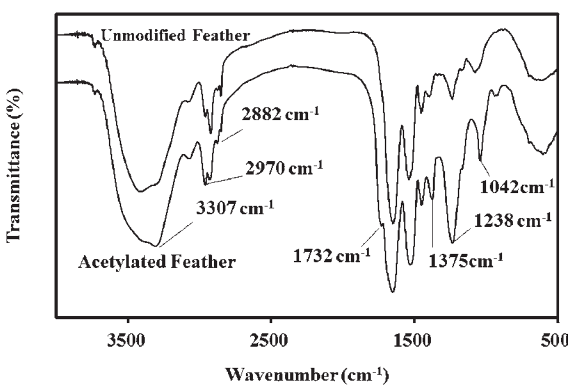


Figure 6. Infrared spectra of unmodified and acetylated chicken feathers.

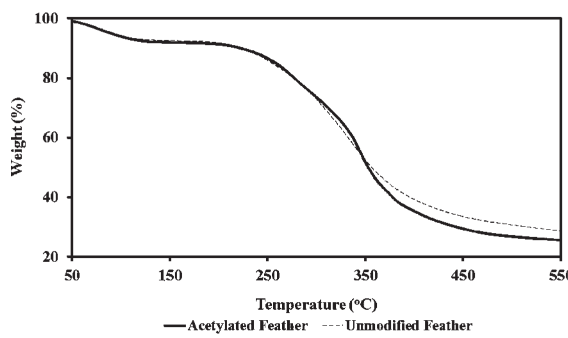


Figure 7. Comparison of the thermogravimetric curves for unmodified and acetylated chicken feathers.

and symmetric vibration of the CH_3 group, respectively.[22] In addition, the presence of the three characteristic ester peaks close to 1100, 1200, and 1700 cm^{-1} (1042, 1238, and 1732 cm^{-1}) confirmed acetylation of the feathers.[22]

Thermal Analysis. The thermal behavior of the acetylated chicken feather is compared to that of the unmodified chicken feather in Figures 7−9. The unmodified and acetylated chicken feathers have similar thermal degradations up to about 250 °C. However, the acetylated chicken feather showed slightly higher overall weight loss than the unmodified chicken feather. The overall weight loss of the acetylated chicken feather was about 75% compared to 68% for the unmodified chicken feather. The higher percent weight loss for the acetylated chicken feather compared to the unmodified chicken feather should mainly be due to the relatively poor thermal instability of the acetyl groups.

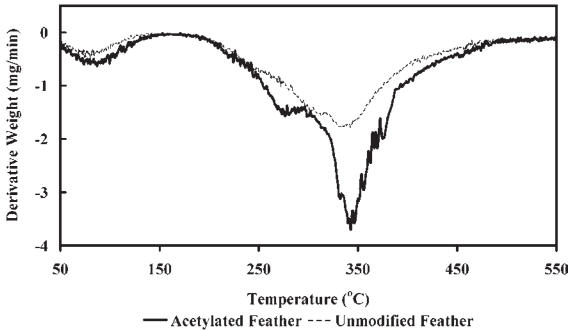


Figure 8. DTG curves of the unmodified and acetylated feathers.

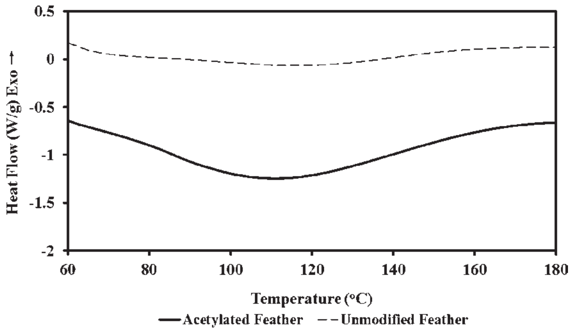


Figure 9. DSC curves of unmodified and acetylated chicken feathers.

Figure 8 shows the DTG curve of the unmodified and acetylated feathers. Both the modified and unmodified feathers show a peak at 70 °C most likely due to the evaporation of water. The peak at about 270 °C, especially in the acetylated feathers, should be due to the substitution on the amino group that decreases the thermal stability of the parent polymer.[23] Also, a prominent peak is seen at 330 °C for the unmodified feather but at a slightly higher temperature (340 °C) for the acetylated feather due to the degradation of the proteins in the feathers.[7] The acetylated feathers have a faster degradation than the unmodified feathers due to the thermal instability of the acetyl groups.

DSC thermograms in Figure 9 show that the acetylated chicken feathers had a thermal behavior different from that of the unmodified chicken feathers. The DSC curve for the acetylated chicken feathers had a broad endothermic melting peak at around 115 °C, indicating that the acetylated feathers were thermoplastic. The unmodified chicken feathers did not show any melting peak. It should also be noted that the melting temperature of the acetylated chicken feathers at about 115 °C is much lower than those of starch acetates (220−270 °C) and cellulose acetates (230−300 °C).[24,25] The lower melting temperature of acetylated chicken feather is beneficial because high temperatures would damage the proteins and result in thermoplastic products with poor properties. It has been shown that feathers are thermally damaged when compression-molded above 180 °C.[1,2] Therefore, lower melting temperatures are desirable to process the feathers into various products. However, the low melting temperature would be a constraint if feathers are mixed with polymers that have high melting temperatures to develop blend products. Similarly, feathers would not be suitable for applications in which products are exposed to temperatures higher than or close to 115 °C.

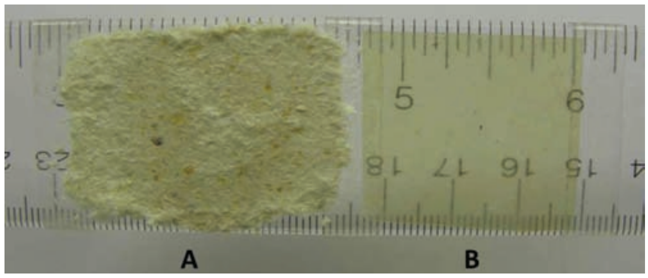


Figure 10. Unmodified chicken feather (A) is not affected by the thermal treatment and transparent thermoplastics developed from acetylated chicken feather (B) after compression molding at 170 °C for 15 min.

Biothermoplastics from Acetylated Chicken Feather. The unmodified and acetylated feathers were compression-molded to verify the possibility of developing thermoplastics from the acetylated chicken feathers. Figure 10 shows the digital image of the modified and unmodified feathers after compression molding. As seen from Figure 10, the unmodified chicken feathers (Figure 10A) did not melt under the pressing conditions (20% glycerol, 170 °C for 15 min) used. However, the acetylated chicken feather melted and formed a transparent film, indicating that the acetylated chicken feathers could be converted to various thermoplastic products (Figure 10B).

This research showed that chicken feathers can be used to develop thermoplastic products after acetylation, which is a green and relatively inexpensive process. Acetylation was performed under acidic conditions, and under the optimized acetylation conditions the percent acetyl content obtained was 7.2% after acetylation using a 4:1 ratio of acetic anhydride to feathers, 10% catalyst, a reaction temperature of 70 °C, and a reaction time of 60 min. The corresponding increase in weight of feathers was 10.6%. Pyrolysis−MS and FTIR confirmed acetylation of feathers. Acetylated feathers had a melting peak at about 115 °C and a slightly higher overall weight loss after thermal degradation. Acetylated feathers were compression-molded to form transparent thermoplastic films. The low melting temperature of acetylated feathers will provide an opportunity to develop feather thermoplastics without damaging the proteins. Acetylated poultry feathers could be used to develop inexpensive, biodegradable, and environmentally friendly films, extrudates, and other thermoplastic products.

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