

TREATMENT OF LOW-QUALITY HIDES WITH FILLERS PRODUCED FROM SUSTAINABLE RESOURCES. EFFECT ON PROPERTIES OF LEATHER

by

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ABSTRACT

Prior research from this laboratory reported on the use of gelatin, alone or in combination with dairy byproducts (casein or whey), as a filler for leather. It was found that all these treatments had fully penetrated the blue stock, were not removed during washing, and had no significant effect on mechanical properties when compared to untreated controls, but did show improvements in the subjective evaluations over the controls with respect to handle, break, dye uptake, and fullness. In this present study we applied these treatments to hides that had grain properties that were characterized as being loose, more commonly known as having spring break, to see if a reduction in these undesirable properties could be realized. The treatments were applied to the butt, belly and neck areas of the hide, and these samples were subsequently retanned, colored and fatliquored (RCF). There were no significant differences between the untreated controls and treated samples with respect to mechanical properties. Importantly, however, it was determined from subjective evaluations, that those commonly inferior areas, such as belly and neck, showed improved cutting area when treated. At the same time, Scanning Electron Microscopy (SEM) was used to compare the blue stock of both poor quality hides and hides evaluated to be of better quality before and after RCF; distinct differences in fiber structure were observed, most dramatically in the belly area. Applying these treatments to low quality hides makes economic sense. Firstly, leathers are produced that present more quality cutting area, and, secondly, these renewable resources have the potential to replace petroleum feedstuffs that are increasingly becoming scarce as well as expensive.

RESUMEN

Investigación previa por este laboratorio informó sobre el uso de la gelatina, sola o en combinación con derivados lácteos (caseína o suero), como un relleno para el cuero. Se encontró que todos estos tratamientos habían penetrado totalmente el cuero wet blue, no se removieron durante el lavado, y que no se obtuvo efecto significativo sobre las propiedades mecánicas en comparación con los controles sin tratar, pero mostraron mejoras en las evaluaciones subjetivas superiores a los controles con respecto al tacto, quiebre, la absorción del colorante, y plenitud. En el presente estudio se aplican estos tratamientos a las pieles que tienen propiedades de flor que fueron caracterizadas como suelta, más comúnmente denominada como “quiebre típico primaveral”, para ver si una reducción de estas propiedades no deseadas podrían realizarse. Los tratamientos se aplicaron a las áreas de culata, flancos y cabeza de la piel, y esas muestras fueron posteriormente recurtidas, teñidas y engrasadas (RCF). No hubo diferencias significativas entre los controles sin tratar y las muestras tratadas con respecto a las propiedades mecánicas. Lo importante, sin embargo, se determinó a partir de evaluaciones subjetivas, por la cuál las zonas comúnmente inferiores, como el flanco y la cabeza, mostraron una mayor área de corte útil cuando tratadas. Al mismo tiempo, Microscopía Electrónica de Barrido (SEM) se utilizó para comparar el cuero wet-blue de ambos casos, los de peor calidad y los evaluados como de mejor calidad antes y luego del RCF; diferencias distintivas en estructura de fibra se observaron, la mayoría de manera espectacular en la zona del flanco. La aplicación de estos tratamientos a las pieles de baja calidad tiene sentido desde el punto de vista económico. En primer lugar, se producen pieles que presentan mayor área utilizable, y en segundo lugar, estos recursos renovables tienen potencial para sustituir productos de petróleo que son cada vez más escasos y caros.

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INTRODUCTION

Poor quality loose grain hides are a continuing problem to the tanning industry. How hides are processed during beaming and tanning may be a contributing factor to this problem. However, a more perplexing dilemma is hides with loose grain that sometimes appear after the winter kill; these latter hides have been commonly referred to as having “spring break”. The leather produced from these hides exhibits a loose or poor break on the grain surface. This poor break manifests itself to a lesser degree in the butt and neck areas but more predominantly in the belly area. Many products, such as the addition or combination of anionic resins, anionic acrylic polymers, amphoteric polymers, polymeric softeners, protein fillers and lastly vegetable extracts, were offered in an attempt to fill the leather. Out of all these products, protein fillers showed a modest improvement in the cutting yield of the finished products.

In prior research from our laboratory we had demonstrated that chemical (glutaraldehyde and genipin) and enzymatic (microbial transglutaminase) modified protein products could successfully be used as fillers in leather processing.¹⁻³ We applied these modified protein products to blue stock and, using epifluorescent microscopy, verified that these products were uniformly distributed through the hide and were not removed during washing.^{4,5} Furthermore, when these treated hides were brought to crust, it was found that the treatments had no effect on mechanical properties when compared to controls, and subjective evaluations showed that the treatments gave superior products.³

Based on industry’s moderate success treating loose grain hides with protein products, we decided to examine the above-mentioned enzymatically-modified proteins and determine if these would not only be applicable as fillers in hides identified as having “spring break”, but it also could possibly reduce the bad break.^{4,9} We prepared, characterized and applied enzymatically-modified products from gelatin/sodium caseinate and gelatin/whey protein concentrate (WPC). After treatment with these products, the blue stock was retanned, colored, and fatliquored (RCF), dried either by toggling or vacuum, and subjected to mechanical testing. Percent extractables, subjective evaluation, and yellowing tests were performed. These data along with Scanning Electron Microscopy (SEM), X-Ray microtomography (Micro-CT), and Stereo Microscopy images will be presented.

EXPERIMENTAL

Materials

Activa TG-TI, a microbial transglutaminase (mTgase) (approximately 100 units/g) containing maltodextrin as a

carrier, with activity from pH 4.0 to 9.0, at 0 to 70°C, was obtained from Ajinomoto USA, Inc. (Paramus, NJ), stored at 4°C in a sealed package, and used without further preparation. Commercial Type B gelatin from bovine skin, characterized in this laboratory as 175 grams Bloom, was obtained from Fisher (Fairlawn, NJ). Sodium caseinate (Alanate® 180) was generously supplied by NZMP (formerly New Zealand Milk Products) (Lemoyne, PA). Whey protein concentrate (WPC) (Hilmar™ 8000) was generously supplied by Hilmar Ingredients (Hilmar, CA). Tannigan PAKN was obtained from Bayer (Pittsburgh, PA); Trutan PA-65, PRP-77, and mimosa were obtained from the former Pilar River Plate Corp. (Newark, NJ); Havana Dye (Derma Havana Brown R Powder) and Melioderm Brown DV were obtained from Clariant Corporation (Charlotte, NC); Magnapol TGR, was obtained from TFL (The Woodlands, TX); Atlasol-CAM, Eureka 400R, and Eureka 950R were obtained from Atlas Refinery, Inc. (Newark, NJ); and Leukotan 1084 was obtained from Rohm and Haas (Philadelphia, PA). Chrome tanned stock (upholstery weight) was purchased from several area tanneries. All other chemicals were analytical grade and used as received.

METHODS

Preparation of biopolymer products

Gelatin samples (175 Bloom) in combination with sodium caseinate or WPC, were suspended in water and allowed to swell for about 2 h at RT; they were stored overnight at 4°C (Figure 1).

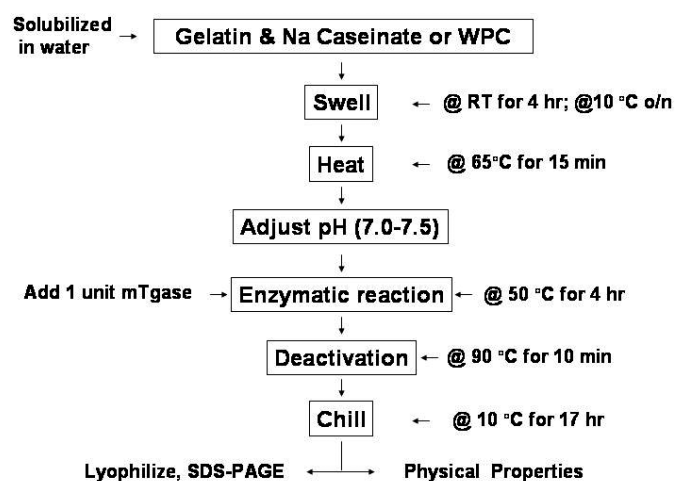


Figure 1. – Schematic for preparation of either gelatin/sodium caseinate or gelatin/WPC biopolymers using 1 unit (per g protein) of mTgase.

They were placed in a bath at 65°C until dissolved. Control samples to which no enzyme was added, were run to monitor changes in physical properties. The pH was adjusted to 7.0-7.5 with 1 N NaOH. To the samples that contained WPC,

0.5% (w/v) DTT was added, and the resulting mixtures were heated at 38°C for 1 h. Microbial transglutaminase (calculated to be 1 unit/g of total protein for biopolymer reactions) was prepared in water and the solution was added with stirring to the protein solution to give a final protein concentration of 10% w/w for gelatin and 2% w/v for sodium caseinate or WPC. Aliquots (10 ml) of the reaction mixture were added to test tubes for melting point determination and 30 ml aliquots were poured into appropriate containers for determining gel strength. The samples were warmed to 50°C in a shaker bath and the reaction was carried out for 4 h. The enzyme was inactivated by heating the reaction products at 90°C for 10 min. The samples were cooled to room temperature and then chilled for 17 h at 10°C in a constant temperature bath. Physical analyses (gel strength, melting point and viscosity) were run on these samples. Aliquots of the samples were lyophilized and molecular weight distribution was determined. Sodium azide (70 µl of 1% solution) was added as a preservative to the biopolymer solutions and the samples were stored at 4°C until use.

Evaluation of wet blue

Wet blue hides from two different tanneries, were evaluated with respect to looseness of grain. The hides from tannery A were labeled H-1 and H-2, and the hide from tannery B was labeled H-3; those hides that showed poor quality were subjected to filler treatment.

Application of filler to wet blue leather

Six pieces of blue stock (~100g each), two pieces each from the butt, belly and neck area, were divided into tests and controls, placed in two Dose drums (Model PFI 300-34, Dose Maschinenbau GmbH, Lichtenau, Germany), washed (400% float based on wet blue weight) by tumbling for 30 min at 50°C, drained and refloatated in sodium bicarbonate (1% on wet blue weight in 400% float) (Figure 2). The samples were tumbled at ambient temperature (25-28°C) until the pH stabilized.

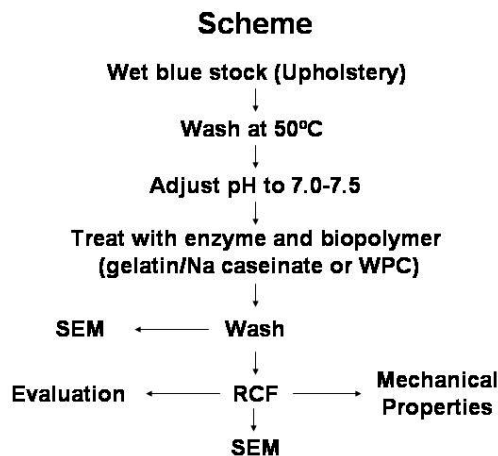


Figure 2. – Schematic for treatment of wet blue with either gelatin/sodium caseinate or gelatin/WPC biopolymer products.

The floats were drained, mTgase (5% based on wet blue weight in 400% float) was added to the test samples, while water (400% float) was added to the controls and all samples were tumbled for 1 h at ambient temperature. The floats were drained and the prepared biopolymer solutions (diluted to give a 400% float based on wet blue weight) were added to the test drums; water (400% float) was added to the control samples. The fillers were applied, based on wet blue weight, using either a 5% gelatin/1% sodium caseinate (Trials A, B, and C) or a 5% gelatin/1% WPC (Trials D, E, and F) loading. The samples were then tumbled for 1 h at ambient temperature and then for 4 h at 50°C. The floats were drained and the samples were washed twice for 10 min at 50°C (400% float), drained, patted dry, and stored at 4°C.

Retan/color/fatliquor (RCF) and drying

The filled samples from the gelatin/caseinate and their respective controls were RCF using the upholstery formulas as seen in Figure 3 (Scheme I) and those from the gelatin/WPC treatment as seen in Figure 4 (Scheme II).

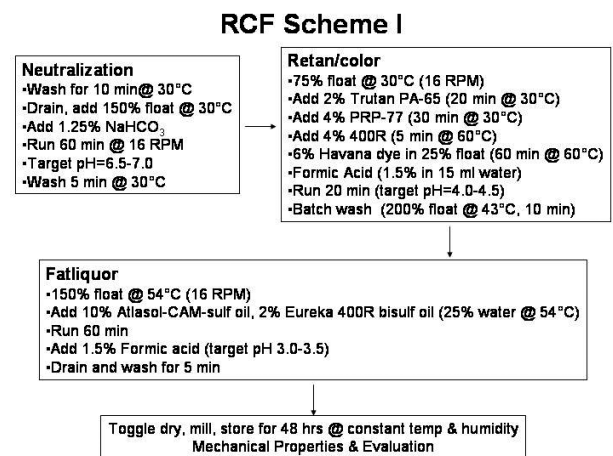


Figure 3. – Schematic (Scheme I) for RCF of treated (gelatin/sodium caseinate) and untreated blue stock (samples toggled dried).

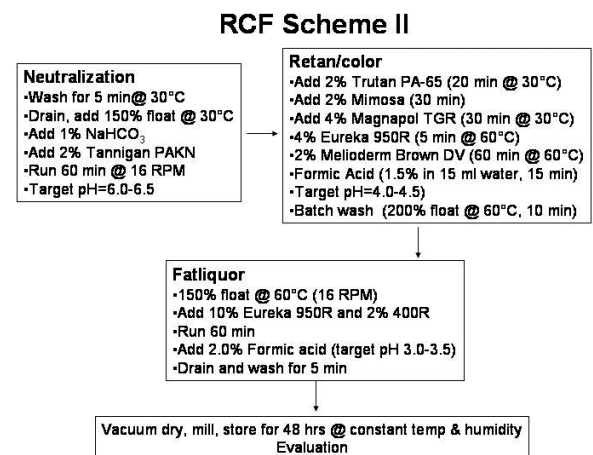


Figure 4. – Schematic (Scheme II) for RCF of treated (gelatin/WPC) and untreated blue stock (samples vacuum dried).

When completed, all samples from the gelatin/sodium caseinate treatment were toggled and left to dry at ambient temperature and humidity and those from the gelatin/WPC treatments were vacuum dried. The samples from both treatments were rewet, put into plastic bags for one day, then staked twice, and milled for approximately 16-18 h. No finishing operations were done to the hides and they were kept on a shelf in the conditioned room, at 20°C and 65% relative humidity (RH) for at least 3 days.

Analyses

Physical properties, molecular weight distribution and extractables

Gel strength, melting point, viscosity, and molecular weight distribution (by SDS-PAGE) of the enzyme-treated proteins were determined as described in previous publications.¹⁰⁻¹¹ Percent extractables in RCF samples was determined as described in ASTM D 3495-83.

Protein concentration determination

Protein concentrations in the float, at different stages of the treatment, were determined using the bicinchoninic acid (BCA) assay¹² according to the directions supplied with the kit and with modification in which gelatin was used as standard as opposed to bovine serum albumin (BSA). Samples were centrifuged at 13,400 rpm for 30 min in a microcentrifuge (Eppendorf MiniSpin plus, Westbury, NY). One ml of protein supernatant was removed and typically a 1:25 (v/v) dilution was prepared in order to fall within the linear concentration range for the assay (200 to 1000 µg/ml protein). A 50 µl aliquot of the diluted solution was mixed with 1.0 ml of BCA reagent and incubated at 37°C for 30 minutes. The absorbance of a sample solution at 562 nm minus a reagent blank was compared with a standard curve using known concentrations of gelatin.

Subjective evaluation RCF leather

Each treated and untreated sample was evaluated with respect to handle, fullness, grain (break) and color. A value from 1 to 5 was assigned for each parameter, with 1 being the worst and 5 being the best. From these ratings, an overall evaluation was determined and this value (from 1 to 5) was reported.

Yellowing test

Two three-inch (76 mm) square pieces were cut from the each of the treated and untreated samples. One square of each sample was placed in an oven, at 120°C, for 72 h. After this time period, the heated samples were then compared to the unheated samples and evaluated with respect to color change. They were rated on a scale of 1 to 5, with 1 being the worst (highest color change) and 5 being the best (least affect on color).

Mechanical properties

The samples were stored in a conditioned room at 20°C and 65% RH according to ASTM D1610-01. Mechanical property measurements were performed parallel to the backbone with a strain rate of 10 in/min and a gage length of 4 inches. The mechanical property measurements included: tear strength, tensile strength, elongation, and Young's Modulus. The tear strength, defined as the load at which the initial tear occurs in the sample, was determined according to ASTM D4704 and was normalized by dividing the tear load by the thickness of the sample and is presented with the units of N/mm. Dog-bone shaped samples were cut out and the tensile strength, defined as the stress required to rupture the leather, was determined according to ASTM D2209. Elongation is defined as the maximum strain at rupture. Young's modulus is a physical quantity representing the stiffness of the material. It is determined by measuring the slope of a line tangent to the initial stress-strain curve.¹³ An upgraded Instron mechanical property tester, model 1122, and Testworks 4 data acquisition software (MTS Systems Corp., Minneapolis, MN) were used throughout this work. Each test was conducted on five samples of untreated and filled RCF blue stock; the average was calculated, and from the mean and standard deviation (STD), error bars were determined.

Scanning electron microscopy (SEM) and X-Ray Tomography (Micro-CT)

Rectangular strips (~ 1 × 2.5 cm) of wet blue hides (spring break and normal) and RCF spring break hides, oriented with the long axes of the hair shafts on the grain surface running parallel to the long axis, were excised with surgical scissors, and multiple cross-sections, 1-2 mm thick, were cut manually from the flesh surface to the grain, with a stainless steel razor blade. Cross-sections were mounted to specimen stubs with Duco cement (ITW Performance Polymers, Riviera Beach, FL) and sputter coated with a thin layer of gold before imaging with a Quanta 200 FEG scanning electron microscope (FEI Co., Inc., Hillsboro, OR) operated in the high vacuum, secondary electron imaging mode.

Samples of loose (spring break) and normal blue stock from the belly areas of the hides were submitted to Microphotonics (Allentown, PA) for X-Ray tomography studies. Samples were analyzed using the SkyScan 1172 Desktop X-ray Microtomograph (Micro-CT).¹⁴

Area fraction analysis

Three SEM images from each sample were selected. Image analysis was performed with Fovea 4.0 plug-ins (Reindeer Graphics, Inc., Asheville, NC) to PhotoShop CS2 (Adobe Systems, Inc., San Jose, CA). The cropped, secondary electron images from the scanning electron microscope were calibrated, flattened and a Gaussian blur was applied to reduce noise. Gray-level thresholds were set to segment the

dark areas between the fibril bundles visually. An area fraction (void) of the segmented dark areas was integrated and an average of the three images was calculated.

Stereo microscope

Samples of treated and untreated spring break hides that were RCF, were examined using a Nikon SMZ-2T stereo microscope (Melville, NY). It has magnification capability from 1.0 X to 6.3 X and is a trinocular stereoscopic model that was designed to accept a digital camera.

RESULTS AND DISCUSSION

Evaluation of Wet Blue

Three wet blue sides were received from two different tanneries and were evaluated for quality, and the results of the evaluation can be found in Table I.

TABLE I

Evaluation of hide quality

Hide ^a	Loose grain location
H-1	butt, belly & down backbone
H-2	butt, belly, neck & down backbone
H-3	None

^aH-1 and H-2 are from tannery A; H-3 is from tannery B.

In hide H-1, loose break could be found in the butt, belly and down the backbone; in hide H-2 looseness could be found also in the butt, belly, and down the backbone as well as in the neck area. Hide H-3 showed no loose break. Samples of the hides, evaluated as normal or having loose break, were examined using SEM. The poor quality hides (H-1 and H-2) were further subjected to filler treatment and then RCF with subsequent evaluation.

SEM Analysis of Spring Break and Normal Hides

In an effort to understand why hides with loose grain, when converted to leather, give such a poor quality product, we examined SEM images of spring break hides and compared their structure to “normal” hides. Samples from the butt, belly and neck areas were taken from both types of blue stock. Representative images of spring break and normal hides can be seen in Figure 5.

When one compares the structure of the spring break blue stock (Figure 5a) from the different areas to the corresponding areas of a normal hide (Figure 5b), one can observe that there are distinctions. A brief description of our observations can be seen in Table II.

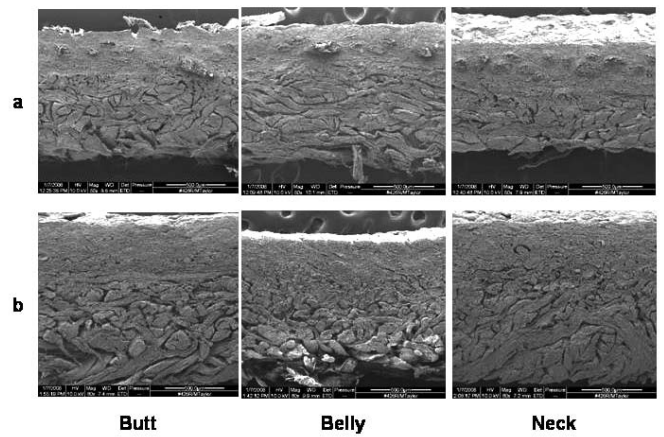


Figure 5. – Scanning Electron Micrograph (SEM) images of blue stock from butt, belly and neck areas of spring break (a) and normal (b) hides, 80X (— = 500 μm).

TABLE II

SEM examination of normal and spring break hides

Area	Normal	Spring break
Butt	Open structure, fibers have weave	Structure maybe less open; otherwise similar
Belly	Open structure, fiber weave	Less open structure, fibers oriented horizontally
Neck	Fibers not as open as in butt and belly, can see weave	Hard to see fibers, appear to be cemented over a large area

It appears that in the three areas from the normal hide, the structure is more open. In the spring break hides, the structure is less open and the fibers do not have the weave pattern as seen in the normal hide. Moreover, in the neck area of the spring break hides, the fibers appear to be cemented over a large area.

The findings from these SEM studies were further corroborated when samples from the belly areas of spring break and normal hides were analyzed using X-ray microtomography. This instrument provides non-destructive 3D imaging of the internal structures of small composite objects with high spatial resolution and unprecedented speed.¹⁴ The instrument moves the source and camera closer together, according to the field of view, to increase the intensity of X-rays that the camera magnifies by up to 16 times.

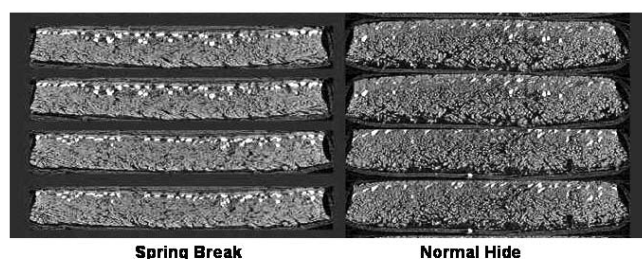


Figure 6. – X-Ray microtomography images of blue stock from belly areas of spring break and normal hides.

Approximately 1000 virtual slices or images were collected for each sample in about five minutes. Representative selections of these images are shown in Figure 6 and again one can see that in the spring break hide sample, the areas between the fibers are more compact than those seen in the normal hide. To further substantiate these findings, when one measures the area (void) between the fibers (Figure 7), one can see that there is a noticeable difference in this area in all three sections of the hides, with the normal hides' area between the fibers being greater.

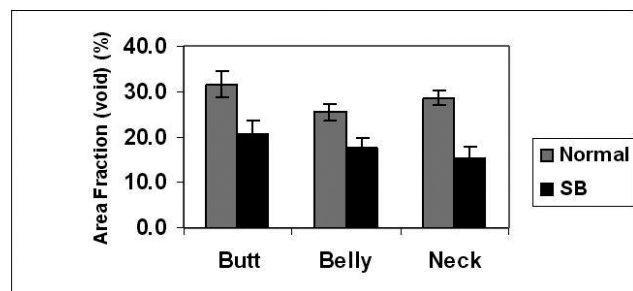


Figure 7. – Percent area fraction (void) between fibers of butt, belly and neck areas of normal and spring break hides.

Treatment of Hides with Gelatin/Sodium Caseinate Biopolymer

Product preparation and characterization

In prior research⁴ we had defined the conditions for preparing a viable gelatin/sodium caseinate filler and found that this product enhanced the subjective properties of what could be described as good quality blue stock. In this present study, we applied this product to hides of poor quality, specifically those identified as having spring break, to determine if it would have a positive effect with respect to providing more cutting area. Gelatin and sodium caseinate were modified with microbial transglutaminase (Figure 1) and the physical properties of the products are shown in Table III.

TABLE III
Physical properties:
gelatin/Na caseinate or WPC

	Gel strength (grams)	MP (°C)	Viscosity @ 60°C (cP)
Gelatin/casein ^a (1u) ^b	393.1	35.4	7.70
Gelatin/casein ^a (0u)	235.3	33.0	7.46
Gelatin/WPC ^b (1u)	380.5	36.4	7.56
Gelatin/WPC ^b (0u)	382.7	35.8	6.82

^aConcentration in solution: Gelatin = 10% w/w; Sodium caseinate = 2% w/v. ^bGelatin = 10%; whey protein concentrate (WPC) = 2% w/v.

If one compares the enzyme modified products to that which has not been modified one can see that the gel strength has increased significantly from 235g to 393g an observation that we have made previously,⁶ the melting point has increased from 33.0°C to 35.4°C and the viscosity has increased from 7.46 to 7.70 cP. The product that we were making should have melting point and viscosity amenable to being used at temperatures between 28 and 50°C, a range typical of post tanning processes. Our previous finding have shown that addition of increasing amounts of enzyme in preparation of biopolymer would give products that would be outside of this range for as gelatin reacts well with mTgase, sodium caseinate is even more reactive.⁶ The SDS-PAGE gel (Figure 8) indicates the band that does not enter the gel (>200,000 Da) in the treated sample has increased in density when one compares this gel to an untreated gelatin/caseinate gel.

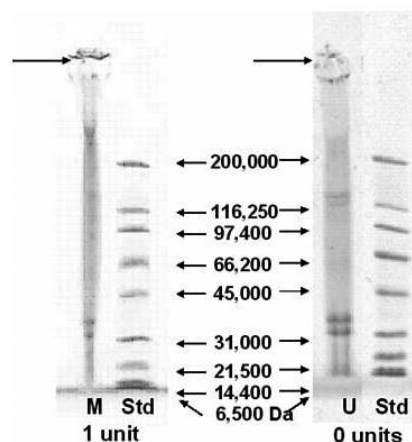


Figure 8. – SDS-PAGE of 175 Bloom gelatin, 10% w/w concentration and 2% sodium caseinate, untreated (U) and modified with 1 unit (per g of protein) microbial transglutaminase (M); molecular weights are shown in Da.

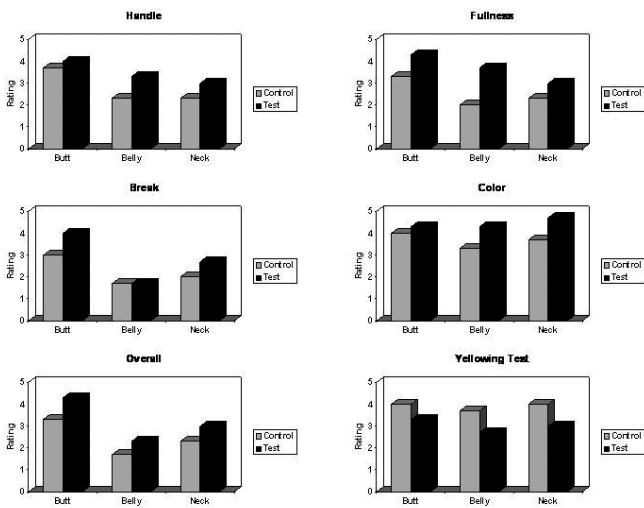


Figure 9. – Subjective evaluation (handle, fullness, break, color and overall rating) using rating scale of 1 = worst to 5 = best, of spring break blue stock, treated with pH-adjusting agents alone (controls) and with mTgase-modified gelatin/sodium caseinate (tests), then RCF; data are from three trials (Trials A, B, and C).

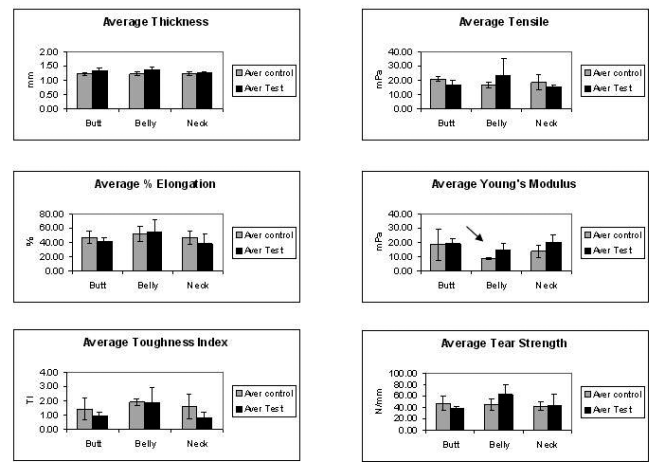


Figure 10. – Mechanical properties (with STD Dev) of spring break blue stock, treated with pH-adjusting agents alone (controls) and with mTgase-modified gelatin/sodium caseinate (tests), then RCF; data are from three trials (Trials A, B, and C).

TABLE IV

Evaluation of treated, RCF blue stock

	Butt	Belly	Neck
Handle			
A ^a	0 ^b	3	2
B	0	0	1
C	1	0	0
Fullness			
A	2	3	2
B	1	2	1
C	1	0	-1
Break			
A	1	1	2
B	2	0	1
C	1	-1	0
Color			
A	1	2	2
B	1	1	2
C	0	0	0
Overall			
A	2	2	2
B	1	1	1
C	1	-1	-1

^aTrial.

^bRating of test minus rating of control.

Blue stock treatment and evaluation

Samples of blue stock identified as having spring break were treated with gelatin/caseinate product. Controls to which no filler was added were also run. The samples were then RCF as described in Figure 3 (Scheme I).

The samples were evaluated with respect to subjective properties; the values from three trials were averaged (Figure 9) and the spring break hides that were filled with gelatin/caseinate fared better than the untreated samples in handle, fullness, break and color. The overall rating shows an improvement in all test areas over the control samples.

When the data for the individual trials was analyzed (Table IV), it was found that 31 of the 45 pairs (or 69%) were superior to controls, 10 pairs (22%) were equal to controls, and 4 pairs (9%) were worse than the controls. In the yellowing test (Figure 9), all of the treated samples fared worse than the controls, an observation that was seen in our previous studies³ and attributed to the fact that when protein is added, the samples customarily give a poorer yellowing result.

Mechanical properties were determined on samples which were filled with gelatin/caseinate as well as corresponding control samples.

The averaged data from three trials is summarized in Figure 10 and shows that the only significant difference can be seen is in the Young's Modulus of belly area with it being greater than the control, indicating a less soft leather which one would expect from a filled sample. When data from the individual trials was analyzed (Table V), those pairs in which there was a significant difference (as indicated by the error bars) between test and controls are indicated by +.

TABLE V

Summary of mechanical property data

Mechanical Property	Control	Test
Thickness		++++ ^a
Tensile	++	+
Elongation	++	
Young's modulus		++++ ^b
Toughness	+++	+
Tear strength	+	+

^aSamples (from 9 pairs analyzed) that are significantly different.

^bIndicating stiffer than control.

These data are showing that of the nine pairs analyzed, four of the tests were thicker, two of the controls in tensile and elongation were higher than tests, three controls had a higher toughness index (TI), control in one tear set is higher, one test in tensile and toughness sets is higher, four of the Young's Modulus determination of the test were stiffer than the controls, and all other parameters were not significantly different.

With respect to percent extractables (Table VI), when the average and standard deviation of the samples from the three runs were calculated, there was no significant difference between the tests and the controls from each area; thus no trend was indicated.

SEM analysis of RCF samples

Samples of RCF-spring break hides from the butt, belly and neck areas, both controls and tests, were further examined by SEM. In most cases, the images were similar to those we have seen for normal RCF samples in that it is difficult at this stage of processing to differentiate between the test and control.

However, representative SEM images of the belly area of the untreated spring break samples (Figure 11a) showed a structure that was open and loose and this possibly could explain poor quality upon evaluation. When this area was treated with fillers, representative SEM images (Figure 11b) showed an improvement in the fiber structure and was typical of that ordinarily seen in RCF leather. Evaluation for break supported this.

TABLE VI

% Extractables

	Butt	Belly	Neck
Control ^a			
Extractable (%)	10.5	13.1	12.1
Std. Dev.	1.2	1.4	3.7
Test ^a			
Extractables (%)	10.8	11.9	13.8
Std. Dev.	2.4	2.1	2.4

^aN=3.

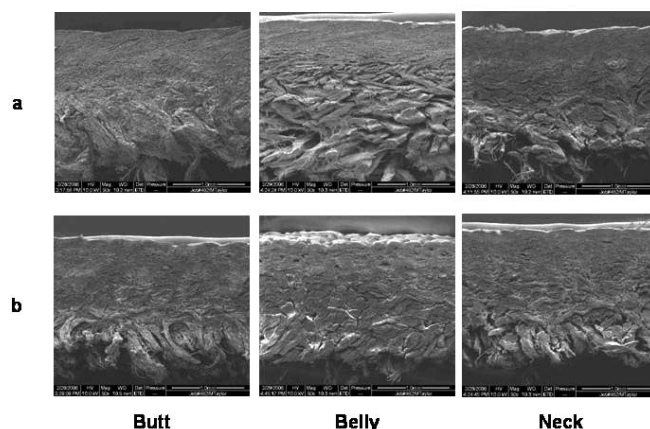


Figure 11. – Scanning Electron Microscope (SEM) images of RCF spring break blue stock from butt, belly and neck areas: controls (a), treated with pH-adjusting agents alone, and filled blue stock (b), treated with gelatin/sodium caseinate biopolymer), 50X (— = 1 mm).

Treatment of Hides with Gelatin/WPC Biopolymer Product preparation and characterization

Our initial experiments, as described above, used gelatin and sodium caseinate, and though quite effective, it was found during the course of the study, that the latter protein (casein) was becoming increasingly more expensive.¹⁵ In previous studies, we had carried out similar successful experiments using modified gelatin/whey protein isolate (WPI),^{5,7-9} but, because of increasing demand, the cost for isolate has increased. It was suggested and we concurred, that whey protein concentrate (WPC), at approximately a fifth of the cost, would be a more economical alternative.¹⁵ WPC contains approximately ~80% protein as opposed to ~91% protein found in WPI.¹⁶ The gelatin/WPC (10%/2% concentration) was treated with 1 unit of enzyme. The product was characterized with respect to physical properties (Table III); the gel strength of unmodified and modified gelatin/WPC is basically the same, whereas the melting point and viscosity of the modified product have increased.

Molecular weight distribution can be seen in Figure 12; the band that does not enter the gel (>200,000 Da) in the sample treated with 1 unit of enzyme is a little denser than this corresponding band in the control sample, and the bands, indicative of whey, as seen between 16,000 and 21,000 Da and between 31,000 and 45,000 Da, are lighter in the modified sample, all indicating that polymerization has taken place.

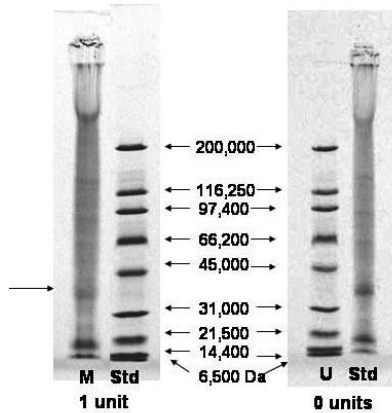


Figure 12. – SDS-PAGE of 175 Bloom gelatin, 10% w/w concentration and 2% WPC, untreated (U) and modified with 1 unit (per gm of protein) microbial transglutaminase (M); molecular weights are shown in Da.

Treatment of blue stock and evaluation

Blue stock characterized as having spring break was treated with gelatin/WPC biopolymer (5%/1% offered on weight of blue stock, Trial D). Two further runs (Trials E and F) were carried out using large panels. During the protein treatment of these latter runs, aliquots were taken from the floats so that protein uptake could be determined. The results of the protein analysis can be seen in Figure 13.

The rate of product uptake was quite reproducible with a 67% uptake after 5 h. It appears that the uptake after two hours is approximately 61% and it is postulated that in an industrial trial with much more mechanical action, the time for treatment could be reduced considerably.

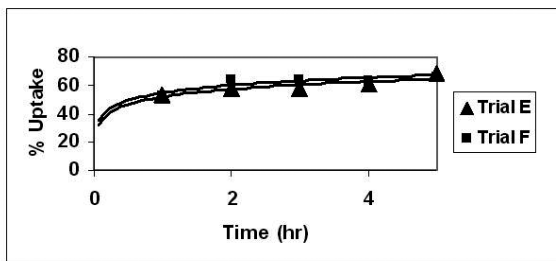


Figure 13. – Protein uptake profiles (Trials E and F) of wet blue pretreated with 5% mTgase and then treated with 5% gelatin and 1% WPC. All percentages were based on the wet blue weight and added in a 400% float.

The blue stock was subsequently RCF according to Scheme II, outlined in Figure 4. Subjective evaluation (handle, fullness, break, color and overall) of the gelatin/WPC treated samples from the butt, belly, and neck areas can be seen in Figure 14.

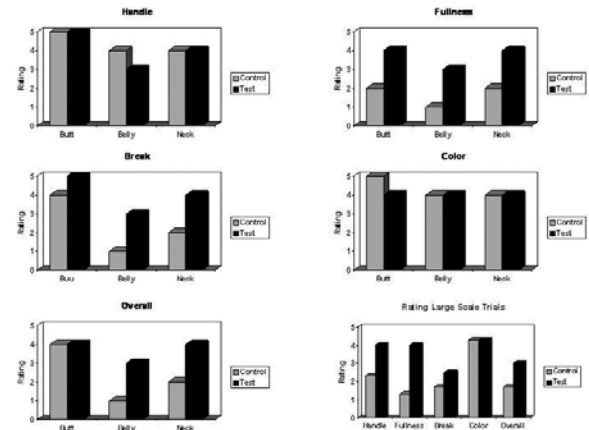


Figure 14. – Subjective evaluation (handle, fullness, break, color and overall rating) of spring break blue stock (butt, belly, and neck areas, Trial D and large panels, average ratings, Trials E and F), treated with pH-adjusting agents alone (controls) and with mTgase-modified gelatin/WPC (tests), then RCF and vacuum dried; rating scale of 1 = worst to 5 = best.

Of the 15 evaluations, eight of the test samples were superior to the control samples, five were equal to the control samples and two did not do as well as the controls. In the two further trials, using large panels mainly from the back-bone area, toggle drying was replaced by vacuum drying and the average evaluation ratings can also be seen in Figure 14. Of the five evaluations (average of two trials) carried out on these larger sections, four of the five were superior to the control sections and one (color) was equal to the control.

Stereo microscope analyses

Samples from belly area of RCF spring break hides, both control and test (gelatin/WPC treatment) were examined using the Stereo Microscope and these images can be seen in Figures 15a and b, along with digital photographs of the hide samples (Figures 15c and d).

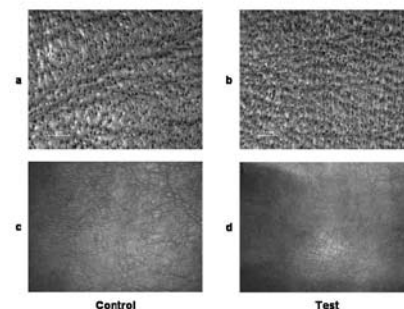


Figure 15. – Stereo Microscope images (— = 1 mm) of hides treated with pH-adjusting agents alone (control, a) and with mTgase-modified gelatin/WPC (test, b), then RCF and vacuum dried; images c (control) and d (test), were taken with digital camera.

When the images were examined using the stereo microscope, one can see the rougher surface of the control (Figure 15a) as opposed to the almost smooth surface of the test (Figure 15b). When photographed using a digital camera, the untreated belly area of the hide (Figure 15c) has a poor grain appearance whereas the treated belly area (Figure 15d) exhibits a flatter grain. Treatment with the filler thus had a positive effect on the grain appearance of hides that had been evaluated initially as being of poor quality, having loose break (spring break).

CONCLUSIONS

Wet blue hides were obtained from two local tanneries, one tannery of which was experiencing problems with hides that they identified as having spring break characteristics. Upon receipt the two batches of wet blue hides were evaluated as to whether they exhibited loose grain or whether they were normal. Samples of the hides, from the butt, belly and neck areas were further examined using SEM and X-ray microtomography (Micro-CT). It was found that in the spring break hides, the fiber structure was considerably different than the normal hides. Subsequently, those hides were treated with biopolymers produced from the enzymatic treatment of gelatin/ sodium caseinate. After treatment, the hides were RCF using conventional upholstery formulas. Mechanical properties were determined and there were almost no significant differences in these properties. These hides were evaluated with respect to handle, fullness, break, color and overall appearance and given a rating from 1 to 5 with one being the worst and 5 being the best. It was found in all parameters the treated spring break hides were superior. They were further examined by SEM and the images were similar to those we have seen for normal RCF samples in that it is difficult at this stage of processing to differentiate between the test and controls. However, in the belly area one could see a significant difference in the fiber structure (much improved) between the treated samples and the untreated controls. Additional improvements in these treatments were realized by the introduction of a more economical treatment, gelatin/WPC; vacuum drying also appeared to show a significant improvement in evaluation. Stereo microscopic images of the belly areas showed a dramatic enhancement in grain surface characteristics. Thus, applying these treatments to low quality spring break hides makes economic sense. To begin with, leathers are produced that have increased cutting area, and, secondly, these renewable resources have the potential to replace petroleum feedstuffs that are increasingly becoming scarce as well as expensive.

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