Research Article
Evaluation of Novel Pre-Slaughter Cattle Wash Formulations for Meat and Byproduct Safety and Quality

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Abstract: The aim of this study is to ensure safety and quality of meat and leather, innovative new carcass washing formulations need to be developed and tested. This study investigated six novel spray wash solutions for their effectiveness on reducing microbial concentrations from the carcass while concurrently examining their effects on leather quality produced subsequent to treatment. The combination of surfactants and anti-bacterial agents was the basis of developing effective carcass wash formulations. Cleansing with water or only surfactant dissolved aqueous solution was found to be ineffective in reducing bacterial concentration including aerobic, *Enterobacter iaceae*, *Salmonella* and *E. coli*. In comparison to spray water treatment, the effective formulations had average reductions of 4.19 to 5.59 log CFU of aerobic bacteria and up to 7 log CFU of *Enterobacter iaceae*, *Salmonella* and *E. coli* concentrations per selected area. Microscopic analysis of the leather produced from treated hides revealed insignificant/no adverse impact from some of the developed formulations on finishing byproduct. Additionally, mechanical properties of finished leather produced from the hides treated with the formulations and water were found comparable. From this research, several effective spray wash formulations were developed for carcass decontamination keeping the integrity of hide to produce quality leather, a valuable byproduct.

Keywords: Decontamination, *E. coli*, *Enterobacter iaceae*, meat byproduct quality, meat safety, *salmonella*

INTRODUCTION

Cattle products such as beef and hides for leather production are a colossal universal commodity, particularly in the United States of America. In 2017, over 995.2 million heads of cattle were reported worldwide, with the United States have one of the most abundant quantities at over 93.6 million heads of cattle (United States Department of Agriculture, Foreign Agricultural Service, 2017; United States Department of Agriculture, National Agriculture Statistic Services, 2017). Moreover, in the United States the leading product of the cattle industry is beef valued at $105 billion dollarsin 2015 (United States Department of Agriculture, Economic Research Service, 2017) and one leading byproduct is hide which is used to make leather valued at $2.2 billion dollars annually (Ramos *et al.*, 2012).

Prior to processing, surfaces of cattle harvested for such products are vulnerable to contamination from pathogens, to include *Enterobacter iacea* bacteria (e.g., *Salmonella*, *E. coli*) from environmental sources (e.g.,

soil and manure) which are prone to become firmly attached to the haired surface of cattle. Through hide removal and meat procurement, cross-contamination can occur from firmly attached pathogens on the haired outer regions of cattle to the inner meat and additionally to the processing equipment (Anderson *et al.*, 1977; Dickson and Anderson, 1992; Conner *et al.*, 1997; Johnston *et al.*, 1982; McEwen *et al.*, 1988). This serves as a hazard to public health and a challenge for the beef industry. Moreover, firmly attached bacteria on the outer grain surface of cattle may lead to putrefaction of hides which may reduce the quality of leather produced as it has been noted that postmortem and during the hide procurement from a carcass, a hide is vulnerable to bacteriological damage (Mohamed *et al.*, 2016).

Enterobacter iaceae, a large family of gram negative bacteria which include well-known pathogenic bacteria such as *Salmonella* and some *E. coli* have been noted in meat related outbreaks. Diarrhea, fever and abdominal cramps are the most common symptoms

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when people are infected with *Salmonella* bacteria (Center for Disease Control and Prevention, 2012; *Enteritidis* infections linked to ground beef: Signs and symptoms). Such bacterial infections have been reported in multistate associating meat related outbreaks. For instance, in Arizona, Illinois, Iowa, Michigan, Pennsylvania and Wisconsin, *Salmonella typhimurium* was linked to an outbreak in 2013 where, 22 illnesses were reported, which were tracked back to two potential companies (Centers for Disease Control and Prevention, 2013: Multistate outbreak of *Salmonella typhimurium* infections linked to ground beef). The outbreaks caused a recall of ~1,050 pounds of ground beef. Similarly, although many *E. coli* are benign and are commonly found in the digestive tracts of mammals, some *E. coli* causes major health issues, including diarrhea, urinary tract infections, respiratory illness and bloodstream infections (Centers for Disease Control and Prevention, 2017: Shiga toxin-producing *E. coli* & food safety). In 2016, veal, beef and cattle products contaminated with *E. coli* from a slaughterhouse in Massachusetts caused a multistate food related outbreak in Connecticut, Massachusetts, Pennsylvania and West Virginia and a recall was enacted on meat products from the specific vendor (Emmert, 2016). In 2014, over 1.8 million pounds of ground beef from a packing facility in Michigan was recalled due to its association with an outbreak with cases in Massachusetts, Michigan, Missouri and Ohio(Centers for Disease Control and Prevention, 2014: Multistate outbreak of shiga toxin-producing *Escherichia coli* o157:h7 infections linked to ground beef). Investigations of such widespread outbreaks often concluded that, contamination likely occurred on the farm or during packing.

Bacteria from grained surfaces may infiltrate and multiply on exposed flesh surfaces. Some aerobic bacteria produce proteolytic enzymes that are capable of penetrating the hide structure and damage to structural proteins (e.g., collagen), reducing hide and leather quality (Mohamed *et al*., 2016). Moreover, acidic, alkaline and other chemical hide wash treatments may reduce bacterial counts, but may also damage the grain surface of the leather made from the cattle hide (Anadan *et al*., 2008; Auer *et al*., 1999; Ramos *et al*., 2013; Marmer and Dudley, 2004; Ramos *et al*., 2012).

To reduce detrimental bacterial damage to meat, cattle may be washed on the grain surface of cattle during pre- and post- harvest operations with surfactants and/or antimicrobials washes during processing to reduce pathogens. However, limited research has investigated solutions' ability to reduce pathogenic concentrations which may cross-contaminate to meat while concurrently investigating their effect on the quality of animal byproducts (e.g., hides used for leather). In this study, six novel solutions of chemicals were investigated for their ability not only

to reduce aerobic bacteria, *Enterobacter iaceae*, *Salmonella* and *E. coli*, on the grained surfaces of cattle hides, but to evaluate the quality of leather produced after treatment. To evaluate leather quality, microscopic grain (stereo microscopic) and surface (Scanning Electron microscopic, SEM) analyses were performed on leather made from hides post-antimicrobial treatments and subsequently compared to leather quality made from untreated hides. Tannery subjective tests (break, handle, fullness and color) are indicators of both leather and hide quality when compared to controls and were performed by an expert in-house tanner. Both chemical treatment and bacterial contaminants and enzymes may degrade the grain and internal structure of the hide and the mechanical properties of the leather may be affected. Thus, mechanical properties (tensile strength, elongation, Young's Modulus and fracture energy) were conducted on crust leather products created from the hides to identify any adverse effects on leather quality compared to their respective controls.

MATERIALS AND METHODS

Hide preparation: Fresh de-fleshed bovine hides were acquired from a local meat packing facility, courtesy of JBS Packerland (Souderton, PA). For bacterial recovery, swabbed samples were collected from randomly selected 10in x 5in surfaces of the hides' backbone area after treating with the respective formulation. For leather quality assessment, one hide was cut into panels with the dimension of 12in x12in for subsequent spray wash treatment with individual formulations.

Anti-microbial formulation preparation: All chemicals used in testing formulations were of commercial grade. *Dioctyl sulfosuccinate sodium salt* (*DOSS*), *Lactic acid solution ≥85%*, *Alkyltrimethylammonium bromide* (*ATMAB*), *Chlorhexidine Di-Gluconate* (*CDG*) were purchased from Aldrich Chemical (Milwaukee, WI). All other reagents used for the formulations were of the highest purity available from commercial suppliers. The preparation of all formulations was carried out as detailed in Table 1, where dissolved in tap water at room temperature (~21°C). All formulations were prepared ~24 h prior to experimental spray treatments of hides.

Spray treatment: For antimicrobial testing the following solutions (Solution A)tap water (control), (Solution B) 0.4% *DOSS* (w/v) + 1% *Lactic acid* (w/v), (Solution C) 0.4% *DOSS* (w/v), (Solution D) 0.6% *ATMAB* (w/v) + 2% *Lactic acid* (w/v), (Solution E) 0.6% *ATMAB* (w/v) + 0.06% *CDG* (v/v), (Solution F) 0.6% *ATMAB* (w/v +0.065 *CDG* (v/v) +H_2O_2 (135 ppm)

+ *Peracetic Acid* (80 ppm) and (Solution G) 0.6% *ATMAB* (w/v) + 0.06% *CDG* (v/v) + 0.043% *NaOCl* (v/v) + 2% *Latic Acid* (w/v) were applied using a handheld 1000 mL polyethylene spray bottle for 10 puffs (12 mL) to cover 10in x 5in surface area. The solutions were allowed to sit for ~1 min before the samples were collected for microbial testing. For hide/leather evaluation testing, solutions were applied using a 1-gallon handheld sprayer at a rate of 515 mL per min. The hides were sprayed for one minute on each 12in x 12in panel of the hide independently to allow for completely covering the panel. After treatment, all hide panels were washed separately in a 6-in-1 drum set-up (Dose Maschinenbau GmbH, Lichtenau, Germany) for 2 h using the USDA hide washing protocol (100% water, 0.15% *Boron TS* and 0.1% *Proxel*).

Microbial testing: A 10in x 5in surface of spray-washed treated areas were aseptically and independently swabbed with a sterile sponge and placed into a bag with 25 mL of buffered peptone water from a sampling kit for analysis (Nasco Meat and Turkey Carcass Sampling Kit, Salida, California).Sample bags containing the buffered peptone and the swabbed sponges were hand massaged for ~2 min. Samples were subsequently diluted and spread-plated on *Tryptic Soy Agar* (*TSA*), *MaConkey Agar* (*MAC*), *Xylose-Lysine-Tergitol 4* (*XLT-4*) *Agar*, *Sorbitol MacConkey Agar*, with *Cefixime* and *Tellurite* (*CT- SMAC*) (all agar was obtained from Fisher Scientific, Pittsburg, PA) for aerobic bacteria, E*nterobacter iaceae bacteria*, *Salmonella* and *E. coli* counts, respectively. Samples were incubated between 24-48 h at 37°C and bacterial colonies were enumerated for bacterial recovery with the lowest detection level at 1 CFU per 10in x 5in area.

Leather preparation: After antimicrobial sample washing, the 12in x 12in hide sample panels were placed in one dehairing drum and the control panel was placed in another dehairing drum and de-haired per the USDA tanning protocol (Cabeza *et al*., 1998). All hide panels were combined into one drum for the pickle, tanning, re-tanning, coloring and fat liquoring steps. The samples were tanned into crust upper shoe leather (Crust L-A, Crust L-B, Crust L-C, Crust L-D, Crust L-E, Crust L-F, Crust L-G which were treated with solution A, solution B, solution C, solution D, solution E, solution F and solution G, respectively) following the standard USDA tanning procedures (Cabeza *et al*., 1998). The resulting leather samples were kept in a temperature (21°C) and humidity (50% relative humidity) controlled environmental chamber (Caron Environmental Chamber, Marietta, OH) until subjective, mechanical and stereo microscopy analyses were performed.

Evaluation of leather quality: To assess the effects of the solutions on leather quality produced from treated hides, the mechanical properties (Young's modulus, tensile strength, fracture energy, elongation) were measured. Dog bone shaped leather samples (1- x 10-cm) were cut along the dimension parallel to the backbone of the cattle hide following the protocol in ASTM D2813-03. The range of average thickness of the leather samples were observed from 2.0 mm to 2.7 mm. An MTS Insight tester and Testworks-4 data acquisition software (MTS Systems Corp., Minneapolis, MN) were used to evaluate the mechanical properties of the leather samples. The strain rate and the grip distance for this study were set to 24.5 cm/min 10.16 cm respectively. Samples were tested in a room set at 73±3°C and 50±5% relative humidity. Tannery subjective tests (break, handle, fullness and color) were conducted by an expert in-house USDA tanner.

Microscopic imagining: Representative crust leather samples produced from the hides subjected to spray treatment with individual formulation were inspected under a stereo microscope (Nikon Digital Microscope SMZ-2T, Melville, NY) to determine any detectible changes in the hide grain structure from the treatment. Additionally, Scanning Electron Microscope (SEM) images were taken to identify potential finer structural changes in the surface of the leather. For SEM images, samples were mounted on stubs and sputter gold coated for 1 min (EMS 150R ES, EM Sciences, Hatfield, PA). Samples were viewed with aFEI Quanta 200 F Scanning Electron Microscope (SEM), (Hillsboro, OR, USA) with an accelerating voltage of 10KV in high vacuum mode.

Statistical analysis: Based on a minimum of three replications per treatment, log-values of microbial populations were analyzed by One-way analysis of variance, using SPSS software (version 14.0, SPSS Inc., Chicago, IL). To compare treatment group differences against the control group (water treatment alone), Dunnett's post-hoc analyses were conducted. For Mechanical properties, five samples were tested. Microbial counts were converted to logarithmic values for calculating means, Standard Deviation (SD) and/or reductions. Differences were considered significant at $p \leq 0.05$.

RESULTS

Reduction of aerobic bacteria: Total aerobic bacterial recovery of 7.92±0.20 log CFU/10in x 5in was found when the selected areas were spray-washed with water (solution A). When the hides were treated with solution B, D, E, F and G, average significant reduction($p \leq 0.05$) of aerobic bacteria was seen of 5.59, 4.19,
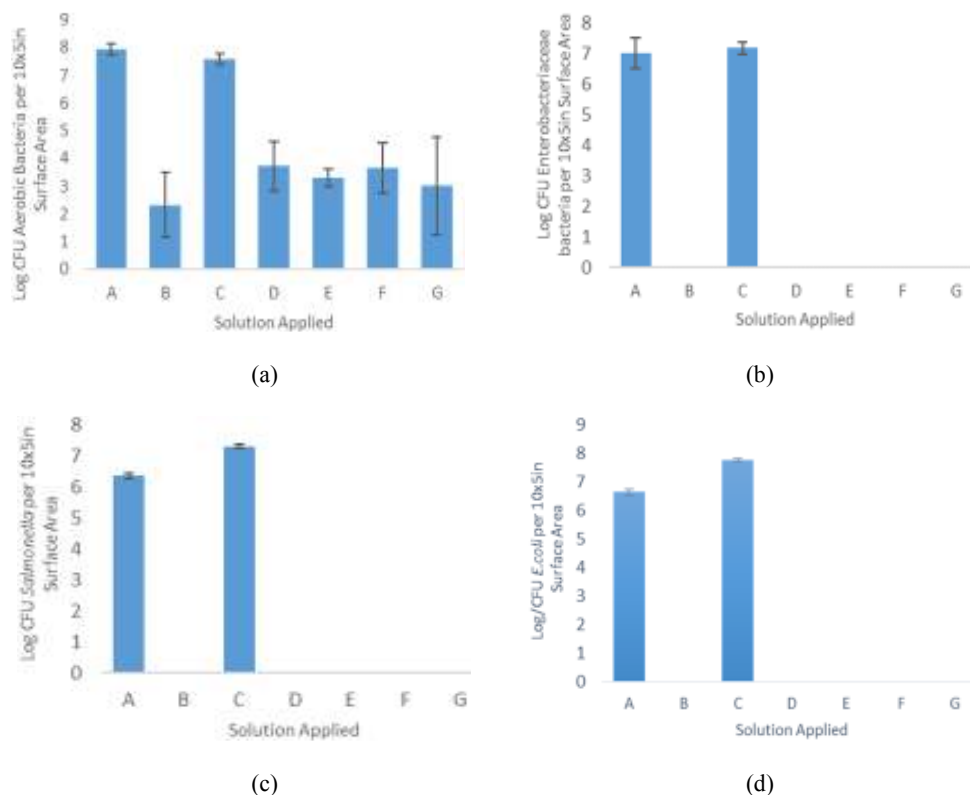
(a)

(b)

(c)

(d)

Fig. 1: Reduction of aerobic bacteria; (a): *Enterobacter iaceae*; (b): *Salmonella*; (c): and *E.coli*; (d): after spray wash treatments with solutions A-G

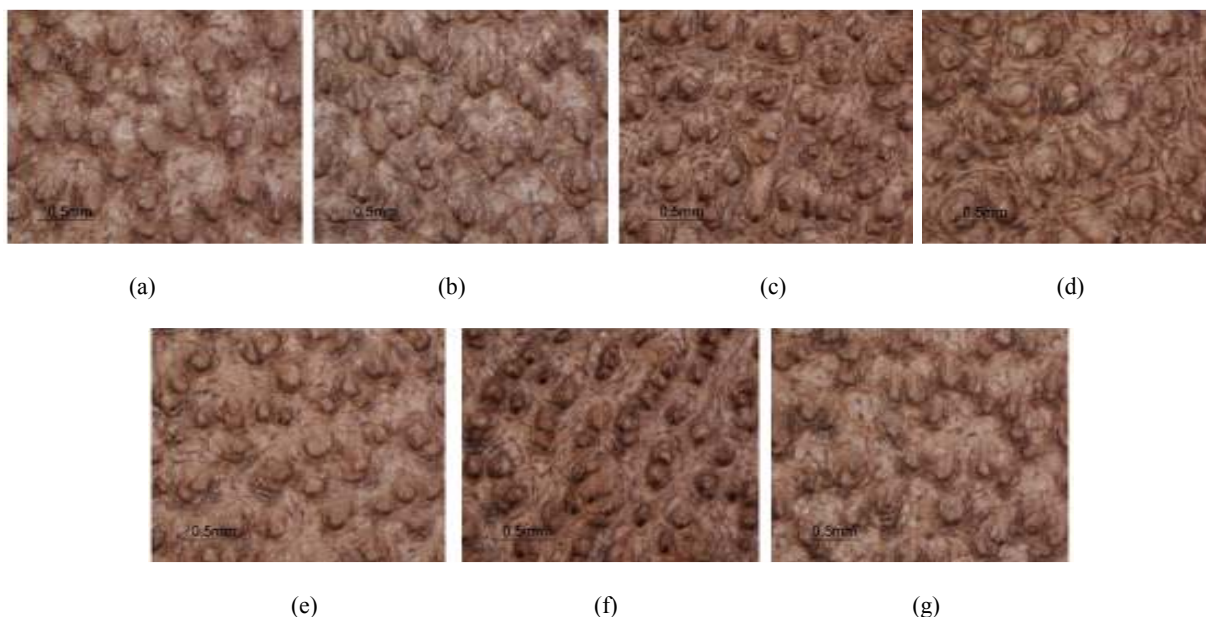(a)

(b)

(c)

(d)

(e)

(f)

(g)

Fig. 2: Stereo microscopic image of crust leather made from hides; (a): Crust L-A (Control); (b): Crust L-B; (c): Crust L-C; (d): Crust L-D; (e): Crust L-E; (f): Crust L-F and; (g): Crust L-G; Bars Represent 0.5 MM

4.62, 4.25 and 4.89 log CFU, respectively (Fig. 1a). No significant reduction between Solution A and C was observed. Furthermore, there was no significant difference in total aerobic bacterial recovery between solution B, D, E, F and G (Fig. 1a).

Reduction of *Enterobacter iaceae*. After spray wash treatment, *Enterobacter iaceae bacteria* counts recovered from the grain surface of panel treated with water (solution A) was 7.01±0.49 log CFU. Hide areas treated with solutions B, D, E, F and G revealed

significant average reductions of *Enterobacter iaceae* bacterial recovery of an average of 7 log CFU (p≤0.05) (Fig. 1b). Furthermore, there were no discernible differences between *Enterobacter iacea* recovery from hides treated with solutions B, D, E, F and G. Spray washing with solution C revealed no significant reduction of *Enterobacter iaceae* counts on hides in comparison to treatment with water alone (Solution A) (Fig. 1b).

Reduction of *Salmonella*: Spray wash treatment with (Solution A) had *Salmonella* recovery of an average of 6.36 log CFU per 10in x 5in area. Similar to *Enterobacter iacea* treated hides, hides treated with Solution B, D, E, F and G had significant reductions (p≤0.05) compared to treatment with water alone (Solution A) (Fig. 1c). In addition, there were no observable differences between hides treated with solutions B, D, E, F and G. Again solution C had no significant reduction in *Salmonella* recovery in comparison to treatment with water alone (Solution A) (Fig. 1c).

Reduction of *E. coli*: Spray washing with water (solution A, Fig. 4) had a recovery of an average of 6.65 log CFU. Similar to *Salmonella* results, hides treated with solutions B, D, E, F and G had significant reductions (p≤0.05) compared the control (solution A; Fig. 1d). There were no observable differences between solutions B, D, E, F and G. Solution C had no

significant reduction in *E. coli* recovery in comparison to the control (Solution A) (Fig. 1d).

Grain analysis: The grain structure of the crust leather produced from hides treated with water Crust L-A (Fig. 2a) and crust leather produced from hides spray washed with solutions B-G (Crust L-B-Crust L-G) (Fig. 2b through g) were analyzed under a stereo microscope. There were no significant differences between the grain structures of leather produced from water (Fig. 2a) and the developed solutions except little abrasion on Crust L-E and Crust L-G (Fig. 2e and g). The panels were folded and a stereo microscopic image was taken at the crease to enhance the surface features (Fig. 3a to f). Again, there were no observable (loosen fiber) differences between leather treated with water (Fig. 3a) and those treated with solutions b-g (Fig. 3b-g).

Microscopic analysis: SEM images of the surface of crust leather produced from hides treated with water (Fig. 4a) and spray solutions B- G (Fig. 4b-g) were analyzed. There were no discernable differences among the surfaces of the leather produced from treated hides except Crust L-E and Crust L-G (Fig. 4e and g) where slight abrasion of the surfaces were observed.

Subjective analysis: A rating value from 1 to 5 was allocated for each parameter, with 1 being the worst and 5 being the best. From these ratings, an overall evaluation was determined from 1 to 5. Crust leather samples treated with water (Crust L-A) had the highest
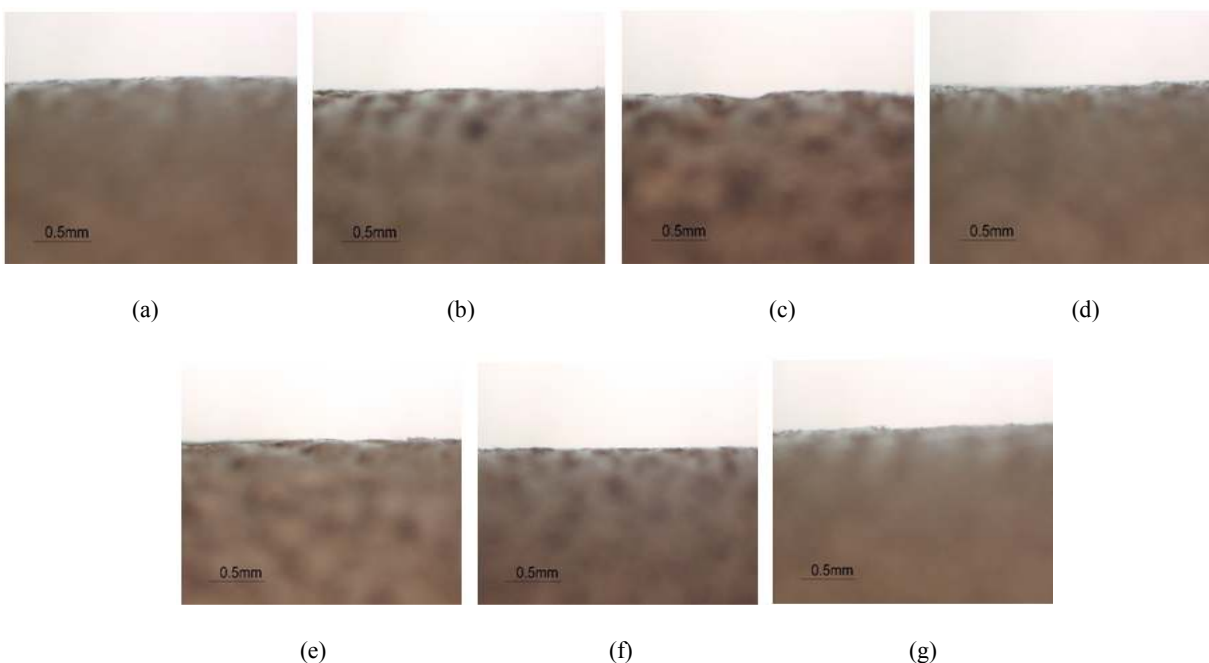

(a) (b) (c) (d)

(e) (f) (g)

Fig. 3: Stereo microscopic images of creased edges of the crust leather made from hides for examination of grain; (a): Crust L-A (Control); (b): Crust L-B; (c): Crust L-C; (d): Crust L-D; (e): Crust L-E; (f): Crust L-F and; (g): Crust L-G; Bars Represent 0.5 MM
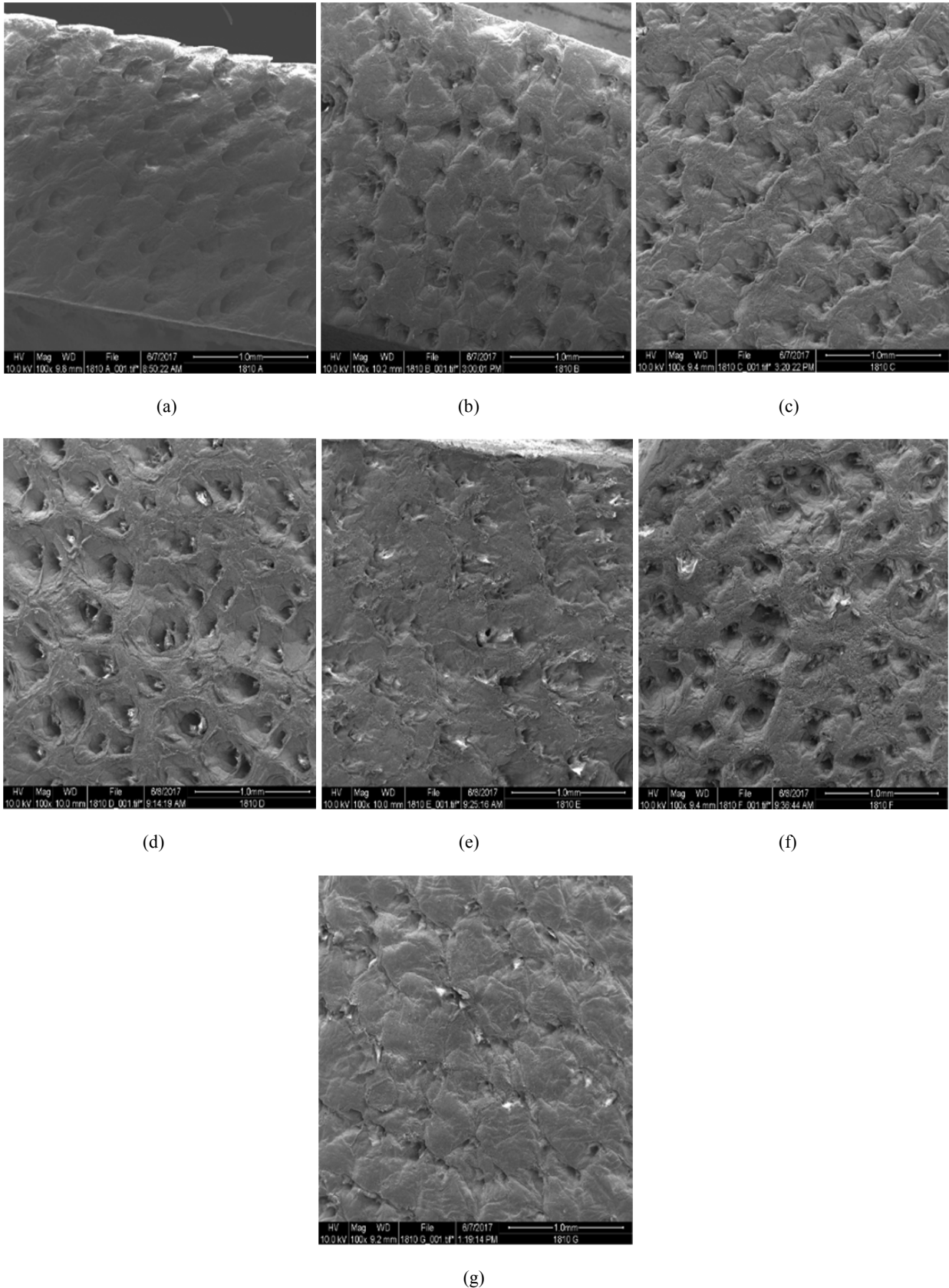
Fig. 4: Scanning electron micrographs of grain surface of Crust Leather 100x; (a): Crust L-A (Control); (b): Crust L-B; (c): Crust L-C; (d): Crust L-D; (e): Crust L-E; (f): Crust L-F and; (g): Crust L-G

Table 1: Subjective properties of the crust leather made from hides.

Treatment	Break	Handle	Fullness	Color	Overall
Crust L-A (control)	5	5	5	5	5
Crust L-B	4	4	5	5	4.5
Crust L-C	3	3	3	4	3
Crust L-D	5	5	5	5	5
Crust L-E	5	5	5	5	5
Crust L-F	3	3	4	5	3
Crust L-G	5	5	5	5	5

Table 2: Mechanical properties of the crust leather made from spray treated hides

Treatment	Tensile strength (MPa)	Elongation (%)	Young's Modulus (MPa)	Fracture energy (J/cm^3)
Crust L -A	16.01±2.75	40.46±4.19	19.30±5.47	2.21±0.37
Crust L -B	17.53±2.17	39.75±3.61	21.53±3.78	2.42±0.48
Crust L -C	12.14±4.41	34.83±7.05	18.69±6.26	1.55±0.76
Crust L -D	13.32±2.27	33.20±8.55	22.48±8.55	1.53±0.50
Crust L -E	13.26±2.12	36.05±2.47	18.17±3.97	1.64±0.38
Crust L -F	8.09±2.12	33.62±5.71	40.12±11.91	0.86±0.26
Crust L -G	15.21±4.27	34.62±5.42	25.78±7.21	1.92±0.73

score of 5 for all testing same as for Crust L -D, -E and G (Table 1).

Mechanical property analysis: Mechanical properties (tensile strength, elongation, Young's Modulus and fractur energy) were measured on test hide panels cut out (ASTM Method 2813) from the final leather product of Crust L-A through -G, as shown in Table 2. In the crust leather produced, there were significant group differences between Tensile Strength (p<0.001), Young's Module (p = 0.015) and Fracture Energy (p<0.001). However, when post-hoc analyses were conducted, there were significant differences between the Crust L-A and Crust L-F in Tensile Strength, Fracture Energy and in softness (Young's Modulus). All others (Crust L -B, -C, -D, -E and -G) did not reveal significant group differences against water (Crust L -A) (Table 2). This indicates that the application of the solutions did not have any detrimental effect on the mechanical properties of the final leather product except solution F.

DISCUSSION

In the cattle industry, there are millions of heads of cattle which are processed regularly for meat and their byproducts. During cattle's growth and lifespan they are commonly exposed to bacterial contamination, pathogenic and nonpathogenic, on the outer grained surface of their hide. In recent years meat contamination has been known to be a major issue throughout the world and the United States. Such contaminations can cause illnesses and also death. In addition to health issues, recalls of the marketed products can greatly influence an industry's reputation and income. Therefore, there is an enormous need to develop antimicrobial solutions to reduce such contaminates. The current study concurrently investigated the ability of developed formulations to reduce selected bacteria and these solution's effects on the leather produced from the cattle hide subsequent to spray washing.

To develop the reported six formulations, two types of surfactant (*ATMB*, *DOSS*) and two antimicrobial agents (*Lactic acid*, *CDG*) have been used. Also, two combinations of spore killing agents (*hydrogen peroxide* with *peracetic acid* and *sodium hypochlorite*) have been added to solutions F and G to increase antimicrobial properties. To provide strong cleansing capabilities, all solutions included surfactants which possess *amphiphilic* characteristic having both *hydrophilic* and *lipophilic* properties and which assist cleaning through the formation of micelles. Among the surfactants used, *ATMB* is a quaternary ammonium compound which in addition to possess antibacterial properties (Laemmli, 1970; Ito *et al*., 2009). *ATMB* is able to damage the cell membranes and destroy the cellular structure of various microorganisms that causes disease, including fungi, bacteria and other single cell organisms. *ATMB* is non-toxic to be applied directly to the skin at reported concentration. On the other hand, *DOSS* is a biodegradable surfactant which degrades quickly in water and soil. It can be removed from the atmosphere through a photochemical reaction with an estimated half-life of 18 h (TEXNET (Toxicology Data Network), year). Among the antibacterial chemicals used, *chlorohexidine salts* (*CDG*) dissociate in water and releases *chlorhexidine* cation, which results bactericidal effect through the binding between positively charged *chlorhexidine cation* and negatively charged bacterial cell walls (Leikin and Paloucek, 2008). *CDG* is active against both Gram-positive and Gram-negative organisms and also active for facultative anaerobes, aerobes and yeasts (Leikin and Paloucek, 2008). *Lactic acid* is a well-known antimicrobial (Mies *et al*., 2004) which was combined with surfactants to enhance its' effectiveness in decontaminating the carcass. It is also one of the most commonly used organic acids for treatment of pre-evisceration beef carcasses. In a study by Castillo *et al*. (1998), revealed that spray washing with 2% lactic acid with water reduced *Salmonella typhimurium*, *E. coli* O157:H7, aerobic plate counts and *Enterobacter iaceae* by >4.9, 4.6, 4.6 and 4.3 mean logs, respectively. Further, in a

study by Bosilevac *et al.* (2006), 2% spray washing with lactic acid and water reduced aerobic plate counts by 1.6 logs and *Enterobacter iaceae* counts by 1.0 log on beef carcasses. The antimicrobial ability of lactic acid has further been seen in a study by Mies *et al.* (2004) where 2% lactic acid resulted in least square means of log reduction of *Salmonella* counts of 1.3.

In our study, results of spray wash treatment with solutions B, D, E, F and G showed (Fig. 1a) the formulations had the ability to significantly reduce the aerobic bacteria (p≤0.05). Whereas, spray washing with solution C did not significantly reduce aerobic bacteria concentrations in comparison to washing with water alone (solution A). Similarly, *Enterobacter iaceae* concentrations were not reduced significantly either by spray washing with water or solution C. Solutions B, D, E, F and G showed significant reductions where, no observed counts were observed (Fig. 1b). Similar to *Enterobacter iaceae*, treatments with Solutions B, D, E F and G revealed significant reductions of *Salmonella* and *E. coli* (Fig. 1c and d) where, solution C was not significantly different than wishing with water alone.

According to these results, solution C, which was consisted of only surfactant (*DOSS*) had no ability to significantly reduce selected bacteria on the outer grain surface of bovine hides. While solutions B, D, E, F and G made of the combination of surfactants and antimicrobials showed promise. This suggests that proper cleaning even with surfactant/detergent is not enough to decontaminate a carcass, where pathogenic bacterial cross-contamination is concerned.

Additionally, while investigating the quality of the hide with a stereo microscope (Fig. 2 and 3) and SEM (Fig. 4), consistent results were observed. Minimal abrasion of grained surface of Crust L-E and Crust L-G was identified in both cases. This problem can be mitigated by optimizing the concentration of the formulation which is believed will have a similar antimicrobial effect. However, subjective analyses found that Crust L-C and Crust L -F had the lowest ratings in comparison to the other crust leathers which were closer to the quality of leather produced from the spray-treated hide with water alone. Furthermore, when investigating the mechanical properties (Table 2), all crust leathers were not significantly different from crust leather produced after water spray washing except Crust L-F. In this, Crust L-F produced from spray washing with solution F showed a significant reduction in tensile strength and fracture energy. However, considering the close composition of solution F to other solutions (solution E and G), the resulted impact on leather quality is unlikely to be produced by the influence of the particular solution, instead, this adverse impact may be attributed to the naturally occurred uneven thickness of the bovine hide. The overall results of this study showed that at least some of the reported antimicrobial solutions have the potential for application in the industry.

CONCLUSION

The combination of surfactant and antibacterial agent works effectively to clean the carcass. The efficacy of the developed formulations on reduction of aerobic, *Enterobacter iaceae*, *Salmonella* and *E. coli* is encouraging. The water solubility, bio-degradability and low toxicity of the used chemicals would potentially make these aqueous based formulations ecofriendly. In addition, low concentration of active chemicals in these solutions would be cost effective and finally, with less/no detrimental impact on leather can conceivably make these formulations viable for industrial application.

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