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Effect of Lactic Acid on Quality of Buffalo Offals

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Abstract: A study was carried out to determine the influence of different concentration and contact time combinations of lactic acid solutions on microbial, sensory and physico-chemical characteristics of buffalo offals viz., head meat, heart, liver and rumen. The following concentration and contact time combinations were used: 1% lactic acid for 20 sec, 1.5% lactic acid for 15 sec and 2% lactic acid for 10 sec. A total of 80 buffalo offal samples (20 numbers of each kind) were collected from a buffalo offal market and subjected to immersion treatments. Water washed offal pieces were used as controls. Sensory evaluations were conducted using a sensory panel comprising postgraduate students and scientists of Livestock Products Technology division, Indian Veterinary Research Institute (India). The data obtained were subjected to statistical analysis using the Analysis of Variance (ANOVA). Mean $\log_{10}$ reductions (CFU g^{-1}) achieved, based on the different treatments and kinds of buffalo offal were between 0.22 and 1.05 for total viable counts; 0.22 and 1.19 for coliforms counts and 0.25 and 0.98 for staphylococcal counts. Immersion in 2% lactic acid solution for 10 sec gave the best overall reduction effect. Sensory evaluations recorded minimal effects of treatments on buffalo offals. These findings show that immersion in 2% lactic acid for 10 sec is suitable for decontamination of Buffalo Offals.

Key words: Buffalo offals, lactic acid, microbial quality, immersion treatments, lactic acid

INTRODUCTION

The shelf life of meat is mainly determined by nature and degree of initial contamination of carcass surfaces. Numerous studies have described the numbers and types of bacteria on fresh meats. Most of which deal with the microbial flora of carcasses whereas only few reports relate to the microbial flora of organ meats. Gardner (1971) reported initial Total Colony Counts (TCC) of $\log_{10}$ of 4 to 5 CFU cm^{-2} on porcine liver, resulting in spoilage when count had increased to $\log_{10}$ of 8 to 9 CFU cm^{-2} after 7 day of storage at 5°C. According to Shelef (1994) initial bacterial counts of beef livers from supermarkets were approximately 10^5 g^{-1}. The microbial flora consisted of gram positive cocci, spore formers, coliforms and gram negative rods. Hanna *et al.* (1982) evaluated Aerobic Plate Counts (APCs) and also determined the presence of specific genera (eg. Stapylococcus and Streptococcus) on beef liver, kidney and heart obtained from commercial packaging plant. Similarly, Sinell determined the frequency of occurrence of *Salmonella* sp. on beef heart, lung and rumen at a central market in Germany. Sheridan and Lynch (1988) observed the influence of processing and refrigeration on the bacterial numbers of beef offals. These studies indicate that offals are generally meat products with poor microbial quality. Control and reduction of such contamination is of great concern. Generally, decontamination procedures substantially reduce the initial microbial load; as a result, fewer microorganisms are present, which are then more easily inhibited in subsequent processing steps. Dickson and Anderson (1992) also concluded that a decontamination step during the slaughtering process can reduce contamination and possibly contribute to improvement of shelf life and safety and should be an essential part of the slaughtering/dressing process.

Several methods have been developed for bacterial reduction on carcasses which include trimming (Prasai *et al.*, 1995a, b) water washing (Hardin *et al.*, 1995; Reagan *et al.*, 1996) hot water spraying (Smith and Graham, 1978) steam pasteurizing (Phebus *et al.*, 1997) as well as sanitizing by chemicals such as organic acids (Hardin *et al.*, 1995; Dorsa *et al.*, 1997b) chlorine compounds (Kotula *et al.*, 1974) polyphosphates (Dickson *et al.*, 1994) and disinfectants.

Although there are several studies on decontamination of carcasses, only limited reports are available on offals (Patterson and Gibbs, 1979;

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Woolthuis *et al.*, 1984; Delmore *et al.*, 2000). These studies on carcasses and offals indicate that an efficient method of surface decontamination provides an additional barrier protection beyond low temperature control and, thereby, offers substantial advantages in terms of food safety, spoilage and economics.

Organic acids are the most frequently used chemical decontaminants (Belk, 2001) and are one of the Generally Recognized As Safe (GRAS) compounds. Several authors have studied the effect of organic acids (especially lactic acid) on bacterial populations as well as on certain specific pathogenic organisms (Ockerman *et al.*, 1974; Osthold *et al.*, 1983; Smulders and Woolthius, 1985; Anderson and Marshall, 1990; Shelef, 1994; Bajaj *et al.*, 2003) and reported that lactic acid treatment, when applied early postmortem, significantly reduced these microorganisms. Hence, the present study has been undertaken to identify the optimum concentration and contact time combination of Lactic Acid (LA) for the decontamination of buffalo offals.

MATERIALS AND METHODS

A study to assess the effect of lactic acid on quality of buffalo offals was carried out in the Division of Livestock Products Technology, IVRI, Bareilly during the period from November 2004 to January 2006.

Collection of buffalo offal samples: Buffalo offals *viz.*, head meat, heart, liver and rumen were collected from Bareilly (India) offal market within 3 to 4 h of slaughter and were packed individually in clean polyethylene bags. Then, they were transported in insulated, iced containers to microbiology laboratory of Livestock Products Technology (LPT) division, Indian Veterinary Research Institute, under hygienic condition for treatment and analysis.

Standardizing contact time for lactic acid solutions (Preliminary trials): Each kind of offal was hygienically cut into pieces of 100 g. one piece was washed with sterile tap water and was maintained as control. The remaining pieces were separately dipped in glass beakers containing solutions of 1% lactic acid (pH 2.6) for 5, 10, 15, 20, 25 and 30 sec. After the specified periods of immersion, the offal pieces were removed, drained and placed individually in clean plates. Sensory analysis was conducted 45 min after the treatments by a sensory panel comprising postgraduate students and scientists of Livestock Products Technology Division, Indian Veterinary Research Institute. Sensory evaluation scale as described by Anna (2001) was suitably modified and used for the present study. The maximum contact time at which the solution of 1% lactic acid had minimal/no effect on colour and odour of offal piece was selected for the further study. The description of scale used in the study is given below.

Score	Effect on odour	Effect on colour
6	Odour improved	Colour improved
5	No chemical odour	No bleaching
4	Traces of chemical odour	Mild bleaching
3	Slight chemical odour	Moderate bleaching
2	Moderate chemical odour	Severe bleaching
1	Strong chemical odour	Very severe bleaching

Similar procedure was repeated using solutions of 1.5 (pH 2.5) and 2% (pH 2.4) lactic acid to identify a suitable contact time for each. Sterile tap water was used for the preparation of different lactic acid solutions (vol/vol) and the lactic acid (Hi media, Bombay, India) used was of analytical grade.

Treatment of buffalo offals with lactic acid solutions: Each kind of offal was hygienically cut into pieces of 100 g and the pieces were divided into four groups. Offal pieces from three groups were individually dipped in 1, 1.5 and 2% lactic acid solutions at ambient temperature for standardized contact times, respectively. The fourth group was washed with sterile tap water and was maintained as a control. Then, the control and treated offal pieces were analyzed 45 min after the treatments for various quality characteristics *viz.*, microbial, sensory and physico-chemical parameters.

Microbial quality: Microbiological quality of control and treated offal pieces were determined based on total viable counts, coliform counts and staphylococcal counts. All microbial groups were assessed by pour plate method following the procedures of American Public Health Association (APHA, 1984). Five grams from each offal piece was aseptically blended with 45 mL of 0.1% sterile peptone water in a pre-sterilized mortar. Decimal dilutions were prepared from the blended samples using sterile 0.1% peptone water. For the counts, one ml of each of the serially diluted homogenate was inoculated in duplicate, to the appropriate growth media in sterile petri dishes using pour plate method. Inocula on plate count agar were incubated at 37±1°C for 48 h under aerobic conditions to assess the total viable counts. Enumeration of *coliforms* was carried out on Violet Red Bile Agar (VRBA) incubated at 37±1°C for 24 h aerobically. *Staphylococci* were enumerated on Baird Parker Agar incubated at 37±1°C for 48 h under aerobic conditions. The average numbers of colonies were expressed as $\log_{10}$ cfu g^{-1} of head meat. All the work was carried out in a clean UV sterilized laminar flow.

Table 1: Buffalo offal samples processed to study the effect of lactic acid solutions on microbial, sensory and physicochemical characteristics

Concentration and type of decontaminant used	Contact time	No. of samples processed				
		Head meat	Heart	Liver	Rumen	Total
Lactic acid treatment						
1. Control	-	5	5	5	5	20
2. 1% Lactic acid	20 sec	5	5	5	5	20
3. 1.5% Lactic acid	15 sec	5	5	5	5	20
4. 2% Lactic acid	10 sec	5	5	5	5	20
Total		20	20	20	20	80

Sensory characteristics: The effect of treatments on colour and odour of offal pieces were assessed 45 min after the treatments by a sensory evaluation panel comprising post graduate students and scientists of LPT division. The six point sensory scale as described by Anna (2001) was used for scoring colour and odour of offal pieces with suitable modifications.

Physicochemical characteristics: The control and treated offal pieces were evaluated for pH and titratable acidity.

pH: pH of the offal pieces were determined by homogenizing 10 g of sample from each offal piece (control and treated) with 50 mL distilled water in Ultra Turrex (IKA, Model T-25, Germany) homogenizer for one min at 3000 rpm. pH of the suspension was recorded by immersing the combined glass electrode of digital pH meter (Model CP-901, Century Instruments Ltd., India).

Titratable acidity: Method described by Konecko (1979) was used with some modifications for estimation of titratable acidity. Five g of sample from each offal piece (control and treated) was homogenized in distilled water and volume was made upto 50 mL. The homogenate was filtered through No. 1 Whatman filter paper to prepare the aliquot. Ten mL of aliquot was titrated against 0.01N NaOH in the burette using 0.1% phenolphthalein solution as indicator. The volume of 0.01N NaOH per g of sample utilized was expressed as titratable acidity.The number of samples processed for each lactic acid treatment is given in the Table 1.

Statistical analysis: For statistical analysis, average counts of colonies on duplicate plates were transformed into log CFU g^{-1}. Then the data were analysed using Analysis of Variance (ANOVA).

RESULTS

Standardizing contact time for lactic acid solutions: Based on the results of preliminary trials conducted to standardize the contact time for lactic acid solutions, 20, 15 and 10 sec were chosen for 1, 1.5 and 2% lactic acid solutions, respectively. Effects of these combinations are compared in the following paragraphs.

Effect of lactic acid solutions on microbial quality of buffalo offals: Effects of tap water washing (control) as well as immersion in lactic acid solutions on total viable count, coliforms count and staphylococcal count of buffalo offals have been presented in Table 2.

Head meat: Overall mean values of total viable count for control and head meat treated with 1, 1.5 and 2% LA were 6.00, 5.74, 5.31 and 4.95, (log cfu g^{-1}), respectively. 1% LA treatment did not result in significant reduction of TVC when compare to control whereas, 1.5 and 2% LA treatment resulted in significant reduction ($p<0.01$) of TVC when compare to control. The level of reduction was 0.69 and 1.05 (log cfu g^{-1}) for 1.5 and 2% LA treatments, respectively. Among the treatment groups (1, 1.5 and 2% LA treated samples), there was a significant difference ($p<0.01$) in level of reduction of TVC. 2% LA treatment resulted in significantly higher ($p<0.01$) level of reduction in TVC when compare to other treatment groups.

Overall mean values of coliform count for control and head meat samples treated with 1, 1.5 and 2% LA were 5.26, 5.04, 4.65 and 4.18, (log cfu g^{-1}), respectively. 1% LA treatment did not result in significant reduction of coliforms count when compare to control whereas, 1.5 and 2% LA treatment resulted in significant ($p<0.01$) reduction when compare to control. The levels of reduction in coliform count were 0.61 and 1.08 (log cfu g^{-1}) for 1.5 and 2% LA treatment. There was a significant difference ($p<0.01$) among the treatment groups. Of the treatment groups, 2% LA treatment resulted in significantly ($p<0.01$) higher reduction of coliforms when compare to other treatment groups.

The mean staphylococcal counts of control and 1, 1.5 and 2% LA treated head meat samples were 5.62, 5.37, 5.11 and 4.73, (log $cfu^{-1}g$), respectively. 1% LA treated samples did not differ significantly from control 1.5 and 2% LA treated samples have shown significantly ($p<0.01$) lower staphylococcal count when compare to control. The levels of reduction in staphylococcal count were 0.51 and 0.89 (log cfu g^{-1}) for 1.5 and 2% LA treatments, respectively. Significant difference ($p<0.01$) was observed among the treatment groups. Of the treatment groups, 2% LA treatment resulted in significantly higher ($p<0.01$) reduction in staphylococcal count while comparing with others.

Table 2: Effect of Lactic Acid (LA) solutions on microbial quality of buffalo offals

Kind of buffalo offal	Treatments	Average microbial count (log cfu g^{-1}) Total viable count	Coliforms count	Staphylococcal count
Head meat	Control	6.00±0.08^{a}	5.26±0.10^{a}	5.62±0.08^{a}
	1% LA/20 sec	5.74±0.09^{a}	5.04±0.13^{a}	5.37±0.08^{a}
	1.5%LA/15 sec	5.31±0.07^{b}	4.65±0.13^{b}	5.11±0.08^{b}
	2%LA/10 sec	4.95±0.12^{c}	4.18±0.10^{c}	4.73±0.09^{c}
Heart	Control	5.46±0.13^{a}	4.98±0.16^{a}	5.24±0.12^{a}
	1% LA/20 sec	5.22±0.14^{ab}	4.60±0.18^{ab}	5.02±0.13^{ab}
	1.5%LA/15 sec	4.91±0.18^{b}	4.26±0.14^{b}	4.72±0.15^{b}
	2%LA/10 sec	4.43±0.16^{c}	3.75±0.11^{c}	4.26±0.11^{c}
Liver	Control	5.53±0.10^{a}	5.04±0.15^{a}	5.26±0.08^{a}
	1% LA/20 sec	5.27±0.09^{ab}	4.70±0.15^{ab}	5.02±0.09^{a}
	1.5%LA/15 sec	5.00±0.14^{b}	4.38±0.15^{b}	4.69±0.11^{b}
	2%LA/10 sec	4.59±0.08^{c}	3.87±0.19^{c}	4.36±0.09^{c}
Rumen	Control	6.26±0.07^{a}	5.23±0.12^{a}	5.69±0.08^{a}
	1% LA/20 sec	6.04±0.09^{a}	4.91±0.11^{a}	5.45±0.09^{a}
	1.5%LA/15 sec	5.74±0.12^{b}	4.46±0.12^{b}	5.11±0.09^{b}
	2%LA/10 sec	5.31±0.10^{c}	4.04±0.10^{c}	4.81±0.09^{c}

Means within a column for given offal sharing the same letter did not differ significantly ($p<0.01$)

Heart: The mean values of TVC for control and 1, 1.5 and 2% LA treated heart samples were 5.46, 5.22, 4.91 and 4.43, (log cfu g^{-1}), respectively. 1% LA treatment did not result in significant reduction of TVC when compare to control whereas, 1.5 and 2% LA treatment resulted in significant reduction ($p<0.01$) of TVC when compare to control. The level of reduction was 0.55 and 1.03 (log cfu g^{-1}) for 1.5 and 2% LA treatments, respectively. Among the treatment groups, no significant difference in level of reduction of TVC was observed between 1 LA and 1.5% LA treatment. 2% LA treatment resulted in significant reduction ($p<0.01$) of TVC when compare to 1 as well as 1.5% LA treatment.

The mean coliform counts of control and heart samples treated with 1, 1.5 and 2% LA were 4.98, 4.60, 4.26 and 3.75, (log cfu g^{-1}), respectively. No statistical difference were observed between 1% LA treated samples and control. However, 1.5 and 2% LA treated samples have shown significant ($p<0.01$) reduction in coliform count when compare to control. The levels of reduction in coliform count were 0.72 and 1.23 (log cfu g^{-1}) for 1.5 and 2% LA treatment. Among the treatment groups, 2% LA treated samples have shown significant ($p<0.01$) reduction in coliform count when compare to 1 and 1.5% LA treated samples whereas, there was no significant difference between 1 and 1.5% LA treated samples.

Overall mean values of staphylococcal count for control and 1, 1.5 and 2% LA treated heart samples were 5.24, 5.02, 4.72 and 4.26 (log cfu g^{-1}), respectively. No significant difference was observed between 1% LA treated samples and control, whereas 1.5 and 2% LA treated samples have shown significantly ($p<0.01$) lower staphylococcal count while comparing with control. The levels of reduction were 0.52 and 0.98 (log cfu g^{-1}) for 1.5 and 2% LA treatments, respectively. Among the treatment groups, 2% LA treated samples have shown significantly ($p<0.01$) lower staphylococcal count while comparing with 1 as well as 1.5% LA treated samples whereas no significant difference was observed between 1 and 1.5% LA treated samples.

Liver: Overall mean values of TVC for control and 1%, 1.5 and 2% LA treated liver samples were 5.53, 5.27, 5.00 and 4.59, (log cfu g^{-1}), respectively. No significant difference in level of reduction of TVC was perceived between 1% LA treated samples and control whereas, 1.5 and 2% LA treated samples showed significantly ($p<0.01$) lower TVC while comparing to control. The level of reduction was 0.53 and 0.94 (log cfu g^{-1}) for 1.5 and 2% LA treatments, respectively. Among the treatment groups, 2% LA treated samples have shown significantly ($p<0.01$) lower TVC when compare to 1 and 1.5% LA treated samples. There was no statistical difference between 1 and 1.5% LA treated samples.

Overall mean values of coliform count for control and liver samples treated with 1, 1.5 and 2% LA were 5.04, 4.70, 4.38 and 3.87, (log cfu g^{-1}), respectively. No statistical difference was observed between 1% LA treated samples and control with respect to coliform count, whereas 1.5% as well as 2% LA treated samples has shown significant ($p<0.01$) reduction in coliform count when compare to control. The levels of reduction in coliform count were 0.66 and 1.17 (log cfu g^{-1}) for 1.5 and 2% LA treatment. Among the treatment groups, 1 and 1.5% LA treated samples did not differ significantly. However, 2% LA treated samples have shown significantly ($p<0.01$) lower coliform count when compare to 1 as well as 1.5% LA treated samples.

Mean staphylococcal count of control and 1, 1.5% and 2% LA treated liver samples were 5.26, 5.02, 4.69 and 4.36, (log cfu g^{-1}), respectively. No statistical difference was observed between 1% LA treated samples and

control whereas 1.5 and 2% LA treated samples showed significantly ($p<0.01$) lower staphylococcal counts while comparing with control. The levels of reduction in staphylococcal count were 0.52 and 0.90 (log cfu g^{-1}) for 1.5 and 2% LA treatments, respectively. Among the treatment groups, significant difference ($p<0.01$) was observed in the level of reduction of staphylococcal count. Of the treatment groups, 2% LA treatment resulted in significantly ($p<0.01$) higher reduction in staphylococcal count while comparing with others.

Rumen: Mean values of TVC for control and 1, 1.5 and 2% LA treated rumen samples were 6.26, 6.04, 5.74 and 5.31, (log cfu g^{-1}), respectively. No significant difference was observed between 1% LA treated samples and control whereas, 1.5 and 2% LA treated samples had significantly ($p<0.01$) lower TVC when compare to control. The level of reduction was 0.52 and 0.95 (log cfu g^{-1}) for 1.5 and 2% LA treatments, respectively. Among the treatment groups, 2% LA treatment resulted in significantly ($p<0.01$) higher reduction in TVC while comparing with other treatment groups.

Overall mean values of coliform count for control and rumen samples treated with 1, 1.5 and 2% LA were 5.23, 4.91, 4.46 and 4.04, (log cfu g^{-1}), respectively. No statistical difference was observed between 1% LA treated samples and control but, 1.5 as well as 2% LA treated samples has shown significantly ($p<0.01$) higher reduction in coliform count when compare to control. The levels of reduction in coliform count were 0.77 and 1.19 (log cfu g^{-1}) for 1.5 and 2% LA treatment. Among the treatment groups, 2% LA treatment has resulted in significantly ($p<0.01$) higher reduction in coliform count when compare to other treatment groups.

Mean staphylococcal counts for control and 1, 1.5 and 2% LA treated rumen samples were 5.69, 5.45, 5.11 and 4.81 (log cfu g^{-1}), respectively. 1% LA treated samples did not differ significantly from control whereas 1.5 as well as 2% LA treated samples have shown significantly ($p<0.01$) lower staphylococcal count while comparing with control. The levels of reduction in staphylococcal count were 0.58 and 0.88 (log cfu g^{-1}) for 1.5 and 2% LA treatments. Significant difference ($p<0.01$) was perceived among the treatment groups. Of the treatment groups, 2% LA treatment resulted in significantly ($p<0.01$) higher reduction in staphylococcal count while comparing with others.

Effect of lactic acid solutions on sensory characteristics of buffalo offals: Effect of Lactic Acid (LA) treatments (1%/20, 1.5%/15 and 2%/10 sec) on odour and colour of buffalo offals have been presented in Table 3. Tap water washed buffalo offals were used as control.

Table 3: Effect of Lactic Acid (LA) solutions on sensory quality of buffalo offals

		Sensory scores	
Kind of buffalo offal	Treatments	Odour	Colour
Head meat	Control	5.00±0.00^{a}	5.00±0.00^{a}
	1% LA/20 sec	4.48±0.11^{b}	4.43±0.16^{b}
	1.5%LA/15 sec	4.24±0.12^{b}	4.48±0.11^{b}
	2%LA/10 sec	4.19±0.11^{b}	4.24±0.17^{b}
Heart	Control	5.00±0.00^{a}	5.00±0.00^{a}
	1% LA/20 sec	4.29±0.14^{b}	4.14±0.10^{b}
	1.5%LA/15 sec	4.00±0.15^{bc}	4.19±0.15^{b}
	2%LA/10 sec	3.86±0.16^{c}	4.10±0.14^{b}
Liver	Control	5.00±0.00^{a}	5.00±0.00^{a}
	1% LA/20 sec	4.33±0.14^{b}	4.71±0.20^{a}
	1.5%LA/15 sec	3.90±0.17^{c}	4.19±0.19^{b}
	2%LA/10 sec	3.76±0.18^{c}	4.05±0.16^{b}
Rumen	Control	5.00±0.00^{a}	5.00±0.00^{a}
	1% LA/20 sec	4.24±0.15^{b}	4.57±0.16^{ab}
	1.5%LA/15 sec	3.81±0.15^{c}	4.19±0.15^{bc}
	2%LA/10 sec	3.62±0.18^{c}	4.05±0.22^{c}

Means within a column for given offal sharing the same letter did not differ significantly ($p<0.01$)

Head meat: Overall mean odour scores for control and 1, 1.5 and 2% LA treated head meat samples were 5.00, 4.48, 4.24 and 4.19, respectively. All the treatment groups have differed significantly ($p<0.01$) from control whereas, no significant difference was observed among the treatment groups.

Mean colour scores of control and 1, 1.5 and 2% LA treated head meat were 5.00, 4.43, 4.48 and 4.24, respectively. All the treatment groups have differed significantly ($p<0.01$) from control. Among the treatment groups, no significant difference was noticed.

Heart: The mean odour scores for control and 1, 1.5 and 2% LA treated heart samples were 5.00, 4.29, 4.00 and 3.86, respectively. All the treatment groups have differed significantly ($p<0.01$) from control. Among the treatment groups, 2% LA treated samples differed significantly ($p<0.01$) from 1% LA treated samples. No statistical difference was observed between 1 and 1.5% LA treated samples as well as between 1.5 and 2% LA treated samples.

Mean colour scores of control and 1, 1.5 and 2% LA treated heart samples were 5.00, 4.14, 4.19 and 4.10, respectively. Control has differed significantly ($p<0.01$) from all the treatment groups whereas, no statistical difference was observed among the treatment groups.

Liver: Mean odour scores of control and 1, 1.5 and 2% LA treated liver samples were 5.00, 4.33, 3.90 and 3.76, respectively. All the treatment groups have differed significantly ($p<0.01$) from control. Similarly, 1% LA treated samples differed significantly ($p<0.01$) from 1.5 as well as 2% LA treated samples. No statistical difference was observed between 1.5 and 2% LA treated samples.

Mean colour scores of control and 1, 1.5 and 2% LA treated liver samples were 5.00, 4.71, 4.19 and 4.05, respectively. 1% LA treated samples did not differ significantly from control, whereas 1.5 and 2% LA treated samples differed significantly ($p<0.01$) from control. Among treatment groups, 1% LA treated samples differed significantly ($p<0.01$) from 1.5% as well as 2% LA treated samples. No significant difference was observed between 1.5 and 2% LA treated samples.

Rumen: Mean odour scores of control and 1, 1.5 and 2% LA treated rumen samples were 5.00, 4.24, 3.81 and 3.62, respectively. All the treatment groups have differed significantly ($p<0.01$) from control. 1% LA treated samples differed significantly ($p<0.01$) from 1.5 as well as 2% LA treated samples. No statistical difference was observed between 1.5 and 2% LA treated samples.

Mean colour scores of control and 1, 1.5 and 2% LA treated rumen samples were 5.00, 4.57, 4.19 and 4.05, respectively. 1% LA treated samples did not differ significantly from control whereas, 1.5 and 2% LA treated samples differed significantly ($p<0.01$) from control. No significant difference was observed between 1 and 1.5% LA treated samples as well as between 1.5 and 2% LA treated samples. 1% LA treated samples differed significantly ($p<0.01$) from 2% LA treated samples.

Effect of lactic acid solutions on physicochemical characteristics of buffalo offals: Effect of lactic acid treatments (1%/20, 1.5%/15 and 2%/10 sec) on pH and titratable acidity of buffalo offals have been presented in Table 4. Tap water washed buffalo offals were used as control.

Head meat: Overall mean pH values for control and 1, 1.5 and 2% LA treated head meat samples were 6.31, 5.42, 5.27 and 4.98, respectively. All the treatment groups differed significantly ($p<0.01$) from control. There was no significant difference between 1 and 1.5% LA treated samples. One and 1.5% LA treated samples differed significantly ($p<0.01$) from 2% LA treated samples.

Overall mean values of titratable acidity for control and 1, 1.5 and 2% LA treated head meat samples were 0.81, 0.96, 1.09 and 1.21, respectively. Titratable acidity of all the treatment groups was significantly ($p<0.01$) higher than that of control. Similarly, among the treatment groups, 2% LA treatment resulted in significantly ($p<0.01$) higher titratable acidity.

Heart: Mean pH values of control and 1, 1.5 and 2% LA treated heart samples were 5.81, 5.16, 5.06 and 4.74, respectively. All the treatment groups differed significantly ($p<0.01$) from control. There was no significant difference between 1 and 1.5% LA treated samples. One and 1.5% LA treated samples differed significantly ($p<0.01$) from 2% LA treated samples.

Table 4: Effect of Lactic Acid (LA) solutions on certain physicochemical characteristics of buffalo offals

		Physicochemical characteristics	
Kind of offals	Treatments	pH	Titratable acidity
Head meat	Control	6.31±0.08^a	0.81±0.04^d
	1%LA/20 sec	5.42±0.03^b	0.96±0.03^c
	1.5%LA/15 sec	5.27±0.05^b	1.09±0.03^b
	2%LA/10 sec	4.98±0.06^c	1.21±0.03^a
Heart	Control	5.81±0.02^a	0.86±0.02^d
	1%LA/20 sec	5.16±0.04^b	1.10±0.03^c
	1.5%LA/15 sec	5.06±0.05^b	1.19±0.02^b
	2%LA/10 sec	4.74±0.07^c	1.26±0.02^a
Liver	Control	6.35±0.04^a	5.26±0.05^c
	1%LA/20 sec	5.75±0.05^b	5.38±0.04^c
	1.5%LA/15 sec	5.50±0.07^c	5.59±0.04^b
	2%LA/10 sec	5.27±0.10^d	5.82±0.03^a
Rumen	Control	6.50±0.02^a	0.62±0.01^c
	1%LA/20 sec	5.75±0.04^b	0.63±0.02^c
	1.5%LA/15 sec	5.52±0.06^c	0.71±0.02^b
	2%LA/10 sec	5.30±0.07^d	0.89±0.02^a

Means within a column for given offal sharing the same letter did not differ significantly ($p<0.01$)

Mean values of titratable acidity for control and 1, 1.5 and 2% LA treated heart samples were 0.86, 1.10, 1.19 and 1.26, respectively. Titratable acidity of all the treatment groups was significantly ($p<0.01$) higher than that of control. Among the treatment groups, 2% LA treatment resulted in significantly ($p<0.01$) higher titratable acidity.

Liver: Overall mean pH values for control and 1, 1.5 and 2% LA treated liver samples were 6.35, 5.75, 5.50 and 5.27, respectively. All the treatment groups differed significantly ($p<0.01$) from control. There was significant difference ($p<0.01$) among the treatment groups.

Overall mean values of titratable acidity for control and 1, 1.5 and 2% LA treated liver samples were 5.26, 5.38, 5.59 and 5.82, respectively. Titratable acidity of all the treatment groups was significantly ($p<0.01$) higher than that of control except 1% LA treatment. Among the treatment groups, 2% LA treatment resulted in significantly ($p<0.01$) higher titratable acidity.

Rumen: Mean pH values of control and 1, 1.5 and 2% LA treated rumen samples were 6.50, 5.75, 5.52 and 5.30, respectively. All the treatment groups differed significantly ($p<0.01$) from control. Treatment groups also differed significantly ($p<0.01$) among them.

Overall mean values of titratable acidity for control and 1, 1.5 and 2% LA treated rumen samples were 0.62, 0.63, 0.71 and 0.89, respectively. Titratable acidity of 1.5% and 2% LA treated samples was significantly ($p<0.01$) higher than that of control whereas 1% LA treatment did

not differ significantly. Among the treatment groups, 2% LA treatment resulted in significantly ($p<0.01$) higher titratable acidity.

DISCUSSION

From the above results, it has been found that 2% lactic acid appears to be more effective when compared to 1 and 1.5% may be due to the reason that acid exhibits effective antimicrobial action, only when appropriate amounts of undissociated molecules of that acid penetrate bacterial cell by means of diffusion and interfere with intra cellular enzymes (Smulders *et al*., 1986). Increasing the amount of acid applied or lowering the pH will increase the amount of undissociated acid molecules, thereby, antimicrobial action. The results of present study are in confirmation with the work done by Saoji *et al*. (1990) who observed similar results in buffalo meat streaks treated with progressively higher concentration of lactic acid solutions. Delmore *et al*. (2000) also reported that immersion of 6 beef variety meat (cheek meat, large intestine, lips, liver, oxtail and tongue) in 2% lactic acid solution for 10 sec resulted in reduction of aerobic plate count (= 0.7 log CFU g^{-1}) and total coliforms count (= 0.5 log CFU g^{-1}). Patterson and Gibbs (1979) observed a 2 log CFU cm^{-2} reduction of aerobic plate counts on beef liver following immersion in 1% (vol/vol) lactic acid (7°C) for 15 min with constant agitation. Similarly, Woolthuis *et al*. (1984) also reported that immersion of pork livers for 5 min in 0.2% (vol/vol) solution of lactic acid and the resulting product had a total colony count that was 2.2 log CFU g^{-1} lower than that for control.

Woolthuis and Smulders (1985) used lactic acid (2%) to decontaminate calf carcasses and reported that 3 was no effect on the flavour of treated carcasses whereas slight to traces of chemical odour was observed in the treatment groups of all kind of buffalo offals in the present study when compared to their respective controls. The sensory evaluation was conducted 24 h after the treatment by those researchers whereas 45 min after the treatment in the present study which might be attributed to the difference in these results. However, Saoji *et al*. (1990) observed a mild acidic odour in buffalo meat streaks treated with 4% lactic acid solution. Mild bleaching in colour was observed in buffalo offals of all the treatment groups when compared to their respective controls. Delmore *et al*. (2000) found that immersion treatments of variety meats had greatest effect on their colour and also reported that the colour of beef liver was lighter after immersion in 2% lactic acid for 10 sec. Saoji *et al*. (1990) also observed similar discolouration in buffalo meat streaks treated with 4% lactic acid for 10-15 sec.

Compare to control as well as other treatment groups, 2% LA treated buffalo offals had significantly lower pH which might be contributed by the higher concentration of acid molecules in 2% lactic acid solution. Similarly, 2% LA treated buffalo offals had significantly higher titratable acidity when compared with control as well as other treatment groups. Factors that were responsible for lowering the pH might be contributed to this higher titratable acidity.

CONCLUSION

Overall, immersion of buffalo offals in 2% lactic acid for 10 sec significantly reduced the levels of total viable count, coliforms count and staphylococcal count. Hence, this treatment would result in buffalo offals of improved quality and safety.

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REFERENCES

American Public Health Association, 1984. Compendium of Methods for the Microbiological Examination of Foods. Speck, M.L. (Ed.), American Public Health Association, Washington, D.C.

Anderson, M.E. and H.T. Marshall, 1990. Reducing microbial populations of beef tissues. Concentration and temperature of an acid mixture. J. Food Sci., 55: 903-905.

Anna Anandh, M., 2001. Studies on development of extruded tripe snack food from buffalo rumen meat. M.V.Sc. Thesis. Deemed University. Indian Veterinary Research Institute, Izatnagar, India.

Bajaj, B.K., Vibhor Sharma, Sanjana Koul and R.L. Thakur, 2003. Incidence of *Salmonella* in poultry and meats and growth inhibition of *Salmonella enteritidis* by organic acids. J. Food Sci. Tech., 40: 556-558.

Belk, K.E., 2001. Beef decontamination technologies. Beef facts. Centennial. CO: National Cattlemen's Beef Association. Research and Technical Services. (Ed.), by R.D. Huffman, 2002.

Delmore, R.J., J.N. Sofos, G.R. Schmidt, K.E. Belk, W.R. Lloyd and G.C. Smith, 2000. Interventions to reduce microbiological contamination of beef variety meats. J. Food Protect., 63: 44-50.

Dickson, J.S. and M.E. Anderson, 1992. Microbial decontamination of food animal carcasses by washing and sanitizing systems: A Review. J. Food Protect., 55: 133-140.

Dickson, J.S., C.G. Nettles-cutter and G.R. Siragusa, 1994. Antimicrobial effects of trisodium phosphate against bacteria attached to beef tissue. J. Food Protect., 57: 952-955.

Dorsa, W.J., C.N. Cutter and G.R. Siragusa, 1997b. Bacterial profile of ground beef made from carcass tissue experimentally contaminated with pathogenic and spoilage bacteria before being washed with hot water, alkaline solution, or organic acid and then stored at 4°C or 12°C. J. Food Protect., 61: 1109-1118.

Gardner, G.A., 1971. A note on the aerobic microflora of fresh and frozen porcine liver stored at 5°C. J. Food Tech., 6: 225.

Hanna, M.O., G.C. Smith, J.W. Savell, F.K. McKeith and C. Vanderzant, 1982. Microbial flora of livers, kidneys and hearts from beef, pork and lamb: Effects of refrigeration, freezing and thawing. J. Food Protect., 45: 63-73.

Hardin, M.D., G.R. Acuff, L.M. Lucia, J.S. Oman and J.W. Savell, 1995. Comparison of methods for contamination removal from beef carcass surfaces. J. Food Protect., 58: 368-374.

Konecko, E.S., 1979. Handbook for meat chemists. Avery publishing Group, Wayne, pp: 52.

Kotula, A.W., W.R Lusby, J.D. Crouse and Bert de Vries, 1974. Beef carcass washing to reduce bacterial contamination. J. Anim. Sci., 39: 674-679.

Ockerman, H.W., R.J. Borton, V.R. Cahill, N.A. Parett and H.D. Hoffman, 1974. Use of acetic and lactic acid to control the quantity of microorganisms on lamb carcasses. J. Milk Food Tech., 37: 203-204.

Osthold, W., H.W. Shin, J. Dressel and L. Leistner, 1983. (Verbesserung der Haltbarkeit von slachttierkorpern her beige fuhrt durch ober flachenbehandlung mit einen saurespray). Improving the storage life of carcass by treating their surfaces with an acid spray. Fleischwirtschaft, 64: 828-830.

Patterson, J.T. and P.A. Gibbs, 1979. Vacuum-packaging of bovine edible offals. Meat Sci., 3: 209.

Phebus, R.K., A.L. Nutsch, D.E. Schafer, R.C. Wilson, M.J. Riemann, J.D. Leising, C.L. Kastner, J.R. Wolf, and Prasai, 1997. Comparison of steam pasteurization and other methods for reduction of pathogens on surfaces of freshly slaughtered beef. J. Food Protect., 60: 476-484.

Prasai, R.K., L.R. Campbell, L.R. Vogt, C.L. Kastner and D.Y.C. Fung, 1995a. Effectiveness of trimming and /or washing on microbiological quality of beef carcasses. J. Food Protect., 58: 1114-1117.

Prasai, R.K., R.K. Phebus, Garcia C.M. Zepeda, C.L. Kastner, A.E. Boyle and D.Y.C. Fung, 1995b. Effectiveness of trimming and or washing on microbiological quality of beef carcasses. J. Food Protect., 58: 1114-1117.

Reagan, J.O., G.R. Acuff, D.R. Buege, M.J. Buyck, J.S. Dickson, C.L. Kastner, J.L. Marsden, J.B. Morgan, R. Nickelson, C.G. Smith and J.N. Sofos, 1996. Trimming and washing of beef carcasses as a method of improving the microbiological quality of beef carcasses. J. Food Protect., 59: 751-756.

Saoji, S.S., A.T. Sherikar, K.N. Bhilegaonkarand U.D. Karkare, 1990. Preservative effect of acetic and lactic acids on buffalo meat stored at refrigeration temperature. J. Bombay Vet. Coll, 2: 37-45.

Shelef, L.A., 1994. Antimicrobial effects of lactates: A review. J. Food Protect., 57: 445-450.

Sheridan, J.J. and B. Lynch, 1988. The influence of processing and refrigeration on the bacterial numbers on beef and sheep offals. Meat Sci., 24: 143-150.

Smith, M.G. and A. Graham, 1978. Destruction of *Escherichia coli* and salmonellae on mutton carcasses by treatment with hot water. Meat Sci., 2: 119-128.

Smulders, F.J.M. and C.H.J. Woolthuis, 1985. Immediate and delayed microbial effects of lactic acid decontamination of calf carcasses-influence on conventionally boned versus hot-boned and vacuum packaged cuts. J. Food Protect., 48: 838-847.

Smulders, F.J.M., P. Barendsen, Van J.G. Logtestijn, D.A.A. Mossel and G.M. Van Der Marel, 1986. Lactic acid: considerations in favour of its acceptance as meat decontaminant. J. Food Tech., 21: 419-436.

Woolthius, C.H.J., D.A.A. Mossel, J.G. Van Logtestijn, J.M. de Kruijf and F.J.M. Smulders, 1984. Microbial decontamination of porcine liver with lactic acid or hot water. J. Food Protect., 47: 220-226.

Woolthius, C.H.J., D.A.A Mossel, Van J.G. Logtestijn, J.M. de Kruijf and F.J.M. Smulders, 1984. Microbial decontamination of porcine liver with lactic acid or hot water. J. Food Protect., 47: 220-226.