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The Effect of Pre-chilling with Acetic and Lactic Acid on Shelf-life of Broiler Carcasses

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Abstract: The effect of pre-chilling in acetic and lactic acid solutions on shelf-life of broiler carcass was investigated. Broiler carcasses were subjected to a 10-min pre-chill treatment with acetic acid, lactic acid and combination of them and examined for their sensorial properties, microbiological quality, pH values and ammonia levels. Treating with organic acids decreased initial microbial load of the carcasses, but not changed their colour, odour and appearance. Controls were spoiled on the 4th day of the storage. The shelf-life of carcasses treated with organic acids were 2-4 days longer. Especially, treatments with 0.6 % lactic acid and 1.0 % acetic acid enhanced the shelf-life twice more. Microbial counts (especially *pseudomonas*), NH_3 amount and pH values of carcass were increased parallel to sensorial alterations. The data from the present study suggest that the treatment of broiler carcasses with pre-chill water containing acetic or lactic acid can help to decontaminate and to increase the shelf-life of carcasses without altering the colour and appearance of the skin.

Key words: Acetic acid, lactic acid, broiler, decontamination, shelf-life

Introduction

Carcasses are contaminated with various spoilage and pathogenic microorganisms at many stages of poultry processing, even though good manufacturing practices are observed during slaughtering (Lillard, 1990). Since the microorganisms are firmly attached on poultry skin and are localized in the capillary spaces, they are not easily rinsed off and are not completely affected by bactericides (Lillard, 1989). For this reason, foodborne diseases associated with poultry meat consumption are frequently reported and the shelf-life of poultry meat is shorter than red meat (Bryan, 1988).

Different methods have been studied for controlling the cross-contamination and decontamination in poultry processing plants. Several chemicals such as organic acids (Dickens and Whittemore, 1994; Ellerbroek *et al*., 1998), chlorine and chlorine derivates (Lillard, 1980), hydrogen peroxide (Mulder *et al*., 1987), ozone (Sheldon and Brown, 1986), and other compounds (Mulder *et al*., 1987) have been applied by dip or spray methods. However, most of them altered the visual appearance of skin and meat. Some of them are even suspicious with regards to public health. In addition to these chemicals, electrical stimulation (Slavik *et al*., 1991), irradiation (Kampelmacher, 1983), sonication (Sams and Feria, 1991) and heat treatment (Davidson *et al*, 1984) have also been used as decontamination methods.

Organic acids such as acetic and lactic acid, and their derivatives were reported as natural alternatives to increase the shelf-life and microbial safety of food products. Acetic acid and lactic acid have been used by adding to scald or chill water during poultry processing and have given relatively reasonable results (Dickens and Whittemore, 1994). Potential disadvantages of these acids are associated with colour changes on the skin, acidic taste and odour imparted to the meat.

The lower the initial contamination, the longer the shelf life at the lowest temperatures (Mulder, 1995). Treatment of carcasses with lactic and acetic acids, reduces the initial microbial load due to their antimicrobial effects. The inhibitory effects of organic acids are mainly related to the amount of undissociated acids; inside the cell they diffuse through the cell membrane and dissociate. This leads to an acidification of the cytoplasm, disruption of the proton-motive force, inhibition of substrate transport, energy-yielding processes and molecular synthesis (Baird-Parker, 1980). Undissociated weak acids are 10 - 600 times more effective in growth inhibition and death of microorganisms in dissociated form. When a strong inorganic acid such as HCl was used to acidify the medium, the inhibition at a given pH was lower than acetic or lactic acids (Guerrero and Taylor, 1994). Most meat spoilage bacteria, except *Lactobacilli*, are inhibited by organic acids (Ouattara *et al*., 1997). This study was performed to investigate the effectiveness of acetic and lactic acids in improving microbiological quality and enhancing shelf-life of broiler carcasses.

Materials and Methods

Broilers: 126 carcasses from a local processor were removed from the evisceration line immediately at different times after the final wash.

Treatment: Broiler carcasses were subjected to a 10 min pre-chill treatment with different concentration of organic acids in two distinct experiments. 54 carcasses were used for first and 72 were for second. As pre-chilling solution, tap water (control), 0.3% lactic acid (A), 0.6% lactic acid (B) for the first experiment and tap water (control), 0.3% lactic acid (C), 1.0% acetic acid (D), equal mix of 0.3% lactic acid and 1.0% acetic acid (E) for the second experiment, were used. For each treatment, six carcasses were immersed in 20 L pre-chill solutions at 10 °C. After 10 min pre-chilling period, all carcasses were removed and chilled for 30 min in (non-agitated) slush ice and drained for 5 min. This procedure was repeated three times per sampling days for a consecutive week.

Treated and untreated carcasses were overlaid with polythene film in a straphore plate and were stored in a refrigerator at 4 °C.

Analyses: After chilling and during cold storage sensorial properties, amount of ammonia (mg/100 g skin), pH value and microbial load (total aerobes, *Enterobacteriaceae*, *pseudomonans*, yeast and mould) of carcasses were determined.

Changes in colour, odour, appearance and texture of treated and untreated carcasses were visually observed and compared for differences. Off-odour and slime formations were evaluated as spoiling indications and detection of these marked the end of shelf-life.

pH of white meat was measured by pH Meter (CG 710 Schott

Mainz). Ammonia amounts (mg/100 g skin) were determined by spectrophotometric method suggested by AOAC (1984).

For microbiological analysis, the treated and untreated carcasses were sampled individually using the whole carcass rinse technique by Cox *et al.* (1981). The carcasses rinsed for 1 min in the plastic bags containing 100 ml sterile diluent. After rinsing, the carcasses were drained for 30 s in bags. The volume of the resulting rinse diluent was measured and 10-fold serial dilutions were made into culture tubes containing 9 ml of sterile 0.1% peptone water. Several dilutions were plated out general and selective growth mediums. Aerobic plate counts, *Enterobacteriaceae* counts, *pseudomonas* count and yeast-mould counts were determined on Plate Count Agar (Oxoid), Violet Red Bile Agar (Oxoid) with 1% glucose, *Pseudomonas* Selective Agar (Oxoid), and Yeast Glucose Chloramphenicol Agar (Oxoid) respectively. Plates were incubated at appropriate temperature and were examined for colony counts after incubation periods.

Statistical analysis: Colony counts (aerobic plate, Enterobacteriaceae, *pseudomonas*, yeast and mould) were transported to logrithmic values. Microbial log counts, ammonia amounts and pH values were analyzed by analysis of variance and treatment means were separated by least significant differences (SAS, 1985).

Results and Discussion

Treatment of broiler carcasses in pre-chill water containing acetic or lactic acid, resulted in a significant reduction for initial microbial load but no alterations on appearance and odour were determined (Table 1,2,3,4). In our previous studies (Bostan *et al.*, 1995) broiler carcasses were subjected to a 10-min pre-chill treatment with 0.1, 0.3 and 0.6% of both acetic acid and lactic acid. Aerobic plate counts and *Enterobacteriaceae* counts were significantly ($P<0.05$) affected by 0.3 and 0.6% of acetic acid treatment and all of the lactic acid treatments. Aerobic plate count values for 0.3 and 0.6% acetic acid and 0.1, 0.3 and 0.6% lactic acid samples were lower than control by 0.54, 0.80, 0.40, 0.91 and 1.43 logs, respectively. Similarly, *Enterobacteriaceae* counts for samples were lower than control by 0.61, 1.02, 0.53, 1.19 and 1.62 logs, respectively. There was also microbial reduction in the pre-chill water with acetic acid and lactic acid. Similar results were already reported on decontamination with acetic acid and lactic acid of poultry carcasses (Mulder *et al.*, 1987; Dickens and Whittemore, 1994). However, some of them observed that carcasses treated with 1.0% and over lactic acid exhibited mild skin discolouration (Mulder *et al.*, 1987) and that feather follicles of carcasses treated with 0.3 and 0.6% acetic acid were protruded or puckered (Dickens and Whittemore, 1994) but no off odour was detected. The feather follicles of both treated and untreated carcasses were also puckered but this situation possibly resulted in the effect of low temperature of chilling solutions. Morrison and Fleet (1985) reported that the microbiological effectiveness of a carcass immersion in 0.25% lactic acid at 18 °C for 10 min gave little microbiological improvement over that of a water immersion control was considered to be of no practical value. However, data showed that initial microbial counts of carcasses treated witn 0.3% LA, except pseudomonas and yeast-mould counts at second experiments were significantly lower than controls.

Shelf life of poultry can be defined as the (storage) period after production that products can be consumed without safety risks for consumers. Treatments with organic acid increased the shelf-life of carcasses by 2-4 days depending on concentration of acids. The end of the shelf life period can be observed or detected because of off-odour formation (Mulder, 1995). In first experiment, the carcasses pre-chilled with tap water, 0.3% (A) and 0.6% lactic acid (B), spoiled respectively on 4^{th}, 6^{th} and 8^{th} day of cold storage at 4 °C. In the second experiment, controls showed typical spoilage indications at 6^{th} day. The same period were eight days for 0.3% lactic acid (C) and ten days for 1.0% acetic acid (D) and the combination of acetic and lactic acid (E). Marel *et al.* (1983) also reported that lactic acid improved the microbial quality and increased the shelf-life of treated carcasses. Cuojoe (1988) reported that spraying the meat surface of skinned cow heads with 1% lactic acid resulted in significant reduction in total viable count of bacteria and the shelf lives of all sprayed heads extended for about three days at 4 °C.

The onset of spoilage in the first experiment was shorter than in the second experiment. This situation was resulted from their high initial microbial load. The shelf-life of freshly processed carcasses depends on the number and types of microorganisms present, the time and temperature of storage, and the methods of packaging. When counts of psycrotrophic gram negative bacteria on chilled poultry reached to 10^7 - 10^8 per cm^2, off-odours often followed by slime formation occured. As shelf life has to do with microbial growth there are several microbiological standards indicating the end of shelf-life, which correlate with the formation of off-odour or even slime on the products. Such a standard could be 5.000.000 cfu's per gram of skin estimated on Plate Count Agar incubated at 10 °C (Mulder, 1995).

PH values and ammonia amounts of broiler carcasses were changed parallel with spoiling periods. At the beginning of storage, pH values of treated and untreated carcasses were between 5.98 and 6.13, and ammonia amounts were 7.10-8.17 mg/100 g. At the days when spoilage was observed sensorially, the pH values and ammonia amounts were high from levels assumed as critic points (pH 6.4; NH_3 30 mg/100 g skin) for meat (Table 5 and 6). Schmitt and Schmidt-Lorenz (1992a) reported that the concentrations of ammonia in the skin increased in pre-spoilage stage but increased rapidly at the onset of spoilage. In present study, there was a constant

Table 1: Aerobic plate counts of broiler carcasses treated at 4°C with lactic acid and acetic acid (Log_{10} cfu/carcass)

Treatment	after chilling	Storage day				
		2^{nd} day	4^{th} day	6^{th} day	8^{th} day	10^{th} day
First experiment						
Control	9.434±0.538 [a]	10.086±0.341[a]	11.662±0.377 [a]	11.895±0.147 [a]	12.076±0.237[a]	
0.3 % LA (A)	7.127±0.314 [b]	8.307±0.269 [b]	9.414±0.433 [b]	11.134±0.242 [b]	11.741±0.137 [a]	
0.6 % LA (B)	6.409±0.239 [c]	7.239±0.412 [c]	8.329±0.532 [b]	9.156±0.368 [c]	11.124±0.304 [b]	
Second experiment						
Control	8.518±0.110 [a]	9.144±0.229 [a]	9.917±0.382 [a]	10.911±0.277 [a]		
0.3 % LA (C)	7.133±0.200 [b]	7.186±0.165 [b]	9.013±0.176 [b]	10.227±0.428[a b]	11.502±0.328 [a]	
1.0 % AA (D)	7.078±0.401 [b]	7.746±0.219 [b]	8.492±0.402 [b]	9.463±0.540 [b]	10.638±0.299 [b]	11.423±0.326[a]
AA+LA (E)	6.945±0.201 [b]	7.592±0.285 [b]	8.147±0.520 [b]	9.284±0.564 [b]	10.471±0.289 [b]	11.220±0.261[a]

Means(±SD) of 3 replication, each with 6 carcasses per treatment

Means in a column with different letters are significantly ($P<0.05$) different from one another.

Table 2: Enterobacteriaceae counts of broiler carcasses treated at 4 °C with lactic acid and acetic acid (Log_{10} cfu/carcass)

Treatment	after chilling	Storage day				
		2nd day	4th day	6th day	8th day	10th day
First experiment						
Control	8.346±0.369 a	8.884±0.120 a	9.245±0.420 a	10.340±0.884 a	11.209±0.265 a	
0.3 % LA (A)	6.783±0.153 b	7.691±0.151 b	8.680±0.284 a	9.580±0.154 a	10.943±0.220 a	
0.6 % LA (B)	5.840±0.210 c	6.549±0.218 c	7.295±0.291 b	8.240±0.621 b	9.709±0.251 b	
Second experiment						
Control	8.212±0.438 a	8.587±0.291 a	9.369±0.452 a	9.873±0.306 a		
0.3 % LA (C)	6.426±0.163 b	7.634±0.183 b	8.401±0.422 ab	9.182±0.525 ab	9.607±0.388 a	
1.0 % AA (D)	6.227±0.282 b	6.968±0.282 c	7.810±0.387 b	8.354±0.579 b	8.763±0.317 b	9.256±0.396 a
AA+LA (E)	6.206±0.303 b	6.667±0.339 c	7.511±0.457 b	8.334±0.421 b	8.626±0.419 b	9.032±0.249 a

Means(±SD) of 3 replication, each with 6 carcasses per treatment
Means in a column with different letters are significantly (P<0.05) different from one another.

Table 3: Pseudomonads counts of broiler carcasses treated at 4 °C with lactic acid and acetic acid (Log_{10} cfu/carcass)

Treatment	After chilling	Storage day				
		2nd day	4th day	6th day	8th day	10th day
First experiment						
Control	6.434±0.344 a	8.416±0.182 a	10.593±0.271 a	11.403±0.337 a	11.980±0.228 a	
0.3 % LA (A)	5.683±0.167 b	7.507±0.362 b	8.761±0.254 b	11.007±0.263 a	11.551±0.235 a	
0.6 % LA (B)	5.127±0.328 b	6.239±0.336 c	7.578±0.456 c	8.986±0.292 b	10.899±0.292 b	
Second experiment						
Control	6.141±0.293 a	7.781±0.450 a	9.670±0.286 a	10.834±0.285 a		
0.3 % LA (C)	5.809±0.413 ab	7.311±0.433 ab	8.648±0.494 b	9.797±0.561 b	11.292±0.345 a	
1.0 % AA (D)	5.679±0.534 ab	6.623±0.257 b	8.074±0.552 bc	9.184±0.515 b	10.319±0.362 b	11.072±0.157 a
AA+LA (E)	5.358±0.165 b	6.437±0.334 b	7.616±0.296 c	8.991±0.550 b	10.073±0.324 b	10.981±0.260 a

Means(±SD) of 3 replication, each with 6 carcasses per treatment
Means in a column with different letters are significantly (P<0.05) different from one another.

Table 4: Yeast and molds counts of broiler carcasses treated at 4 °C with lactic acid and acetic acid (Log_{10} cfu/carcass)

Treatment	After chilling	Storage day				
		2nd day	4th day	6th day	8th day	10th day
First experiment						
Control	6.338±0.402 a	7.823±0.134 a	9.649±0.303 a	10.303±0.376 a	11.974±0.316 a	
0.3 % LA (A)	5.342±0.247 b	7.311±0.372 a	7.814±0.321 b	9.609±0.351 a	10.255±0.324 ab	
0.6 % LA (B)	5.011±0.262 b	5.720±0.277 b	6.714±0.316 c	7.886±0.300 b	9.845±0.420 b	
Second experiment						
Control	6.690±0.395 a	7.917±0.431 a	9.170±0.506 a	10.086±0.414 a		
0.3 % LA (C)	6.510±0.375 a	7.319±0.498 ab	7.985±0.435 b	9.038±0.380 b	10.647±0.259 a	
1.0 % AA (D)	6.384±0.463 a	7.043±0.604 ab	7.602±0.522 b	8.468±0.488 b	9.698±0.308 b	11.630±0.291 a
AA+LA (E)	6.279±0.379 a	6.760±0.415 b	7.344±0.530 b	8.258±0.362 b	9.494±0.349 b	10.505±0.347 a

Means(±SD) of 3 replication, each with 6 carcasses per treatment
Means in a column with different letters are significantly (P<0.05) different from one another.

Table 5: Ammonia amounts of broiler carcasses treated at 4 °C with lactic acid and acetic acid (mg/100 g skin)

Treatment	After chilling	Storage day				
		2nd day	4th day	6th day	8th day	10th day
First experiment						
Control	7.57±0.75 a	17.60±2.03 a	36.67±2.10 a	57.33±3.13 a	89.60±6.90 a	
0.3 % LA (A)	7.53±0.71 a	12.90±1.64 b	21.97±2.04 b	35.00±2.78 b	65.83±4.25 b	
0.6 % LA (B)	7.10±0.76 a	9.47±1.26 c	14.10±1.21 c	24.53±2.01 c	34.60±1.80 c	
Second experiment						
Control	8.17±0.49 a	22.13±3.63 a	31.57±3.50 a	62.67±5.81 a	84.43±5.85 a	
0.3 % LA (C)	7.97±0.50 a	18.37±2.01 ab	21.87±2.72 b	33.83±3.11 b	64.17±4.04 b	
1.0 % AA (D)	7.63±0.67 a	15.43±1.62 bc	18.63±1.88 b	28.17±2.20 bc	32.87±1.68 c	46.60±1.96 a
AA+LA (E)	7.47±0.72 a	13.30±1.65 c	17.10±1.85 b	26.47±2.46 c	30.93±2.14 c	43.93±2.22 a

Means(±SD) of 3 replication, each with 6 carcasses per treatment
Means in a column with different letters are significantly (P<0.05) different from one another.

rise for ammonia amounts.

During the cold storage, the microbial counts (aerobic plate, *Enterobacteriaceae, pseudomonas* and yeast-mould) of the treated and untreated broiler increased continuously, but at all the periods, controls contained more than other groups. Increasing acid concentration decreased the microbial counts (Table 1,2,3 and 4). Ingham (1989) also determined that in catfish fillets dipped in chilled solutions of 1.70 and 2.55% lactic acid, aerobic plate count was significantly lower than controls during 3 days of storage.

The fastest growth was observed for *pseudomonas*. With the beginning of spoilage this group microorganisms were increasingly predominant. The colony counts of yeasts and moulds were increased similar to *pseudomonas*. The colony count of *Enterobacteriaceae* increased slowly during storage. Schmitt and Schmidt-Lorenz (1992b) determined that the onset of spoilage was noticeable in packed carcasses after 8 days and pseudomonas were predominant after 4 days of storage. Regez *et al.* (1988) also reported that the spoilage flora of poultry carcasses consisted of 60 to 80% of

Table 6: pH values of broiler carcasses treated at 4 °C with lactic acid and acetic acid

Treatment	After chilling	Storage day				
		2nd day	4th day	6th day	8th day	10th day
First experiment						
Control	6.13±0.04 [a]	6.25±0.02 [a]	6.42±0.05 [a]	6.61±0.08 [a]	6.75±0.07 [a]	
0.3 % LA (A)	6.09±0.04 [a]	6.16±0.04 [b]	6.26±0.02 [b]	6.41±0.06 [b]	6.59±0.04 [b]	
0.6 % LA (B)	6.05±0.03 [a]	6.13±0.04 [b]	6.20±0.06 [b]	6.27±0.06 [c]	6.39±0.04 [c]	
Second experiment						
Control	6.03±0.03 [a]	6.15±0.05 [a]	6.29±0.06 [a]	6.47±0.06 [a]	6.63±0.05 [a]	
0.3 % LA (C)	6.02±0.05 [a]	6.07±0.06 [a]	6.20±0.02 [a]	6.34±0.04 [b]	6.49±0.05 [b]	
1.0 % AA (D)	5.99±0.05 [a]	6.04±0.06 [a]	6.12±0.04 [b]	6.23±0.04 [c]	6.31±0.08 [c]	6.49±0.05 [a]
AA+LA (E)	5.98±0.08 [a]	6.04±0.06 [a]	6.11±0.03 [b]	6.21±0.05 [c]	6.28±0.04 [c]	6.48±0.05[a]

Means(±SD) of 3 replication, each with 6 carcasses per treatment
Means in a column with different letters are significantly (P<0.05) different from one another

pseudomonas at storage temperatures of 0 and 4 °C.

Lactic acid has a very strong retarding effect on bacteria, however, yeast are better retarded by acetic acid. The combination of acetic and lactic acid has a much stronger retarding effect on microorganisms (Koos, 1992). In our study, the combination of lactic acid (0.3%) and acetic acid (1%) showed higher antimicrobial effect than 0.3% lactic acid alone, but not than 1% acetic acid alone.

In conclusion, the data from the present study suggested that the treatment of broiler carcasses with pre-chill water containing

acetic or lactic acid, particularly 1% acetic acid, and 0.6% lactic acid, can help to decontaminate and to prevent the cross contamination of carcasses without altering the colour and appearance of the skin, if used at a low concentration, and thus enhance the shelf-life of poultry carcasses.

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