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Effect of Cetylpyridinium Chloride (Cecure[®] CPC Antimicrobial¹) on the Refrigerated Shelf Life of Fresh Boneless, Skinless Broiler Thigh Meat

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Abstract: Two similar trials were conducted to evaluate the effects of treating boneless, skinless thigh meat with Cetylpyridinium Chloride (CPC), trade named Cecure[®], on refrigerated (2.5°C) shelf life. In Trial 1, 0.0 (tap water control), 0.5, 1.0 and 1.5% Cecure[®] treatments were evaluated. In Trial 2, a non-sprayed control, a tap water control and 0.4, 0.5 and 1.0% Cecure[®] were evaluated. Cecure[®] was applied using a hand-held spray bottle and applying 7 mL of solution per thigh (3.5 mL/side). In Trial 1, the 0.5% Cecure[®] solution resulted in a 1 day extension in shelflife; both the 1.0 and the 1.5% Cecure[®] treatments resulted in a 2 day extension in shelf life. In Trial 2, both the 0.4 and 0.5% Cecure[®] treatments resulted in a 1 day extension in shelflife; the 1.0% Cecure[®] treatment resulted in a 2 day extension in shelf life. The combined data from the two trials suggest that the extension in shelf life observed in these trials was due to the initial reduction (1 to 2 logs) in Aerobic Plate Count (APC) on the day of treatment. The Cecure[®] treatments did not result in any adverse sensory characteristics (color or odor).

Key words: Broilers, shelf life, cetylpyridinium chloride, Cecure[®]

Introduction

Due to the convenience of preparation, many consumers worldwide prefer boneless, skinless poultry meat instead of whole carcasses. Many of these products (primarily breasts and thighs) are skinned and de-boned by hand, often up to several days post-slaughter. Thus, maintaining an adequate retail and post-purchase shelf life for the finished product is critical. According to Jay (1996), the essential feature of poultry spoilage is related to sliminess at the outer surface of the carcass or parts. If left intact, the inner muscles are generally sterile. The surfaces of raw poultry, stored in an environment of high humidity, are susceptible to the growth of aerobic organisms such as *Pseudomonas* and other spoilage-type organisms. These organisms can reproduce quickly on poultry surfaces where they form minute colonies that later coalesce to produce the sliminess and off-odor characteristic of spoiled poultry. Raw poultry is typically considered spoiled when the total number of surface bacteria reaches 7 in terms of log₁₀ CFU/cm² or per mL (Ayres *et al.*, 1950; Mielnik *et al.*, 1990). The shelf life of raw poultry is primarily dependent on initial levels of total organisms and storage temperature. Certain other intrinsic factors including pH and A_w (water activity) of the product are also important. Various technologies have been investigated in an effort to reduce spoilage and pathogenic organisms on fresh poultry. Some of these technologies include the use of chlorine, acidified sodium chlorite, chlorine dioxide, ozone, organic acids, hydrogen peroxide, trisodium phosphate, electrical conductivity, and irradiation. Many of these technologies are microbiologically effective but

are limited to a great extent by adverse sensory effects, environmental issues, or consumer concerns.

In early 2004, CPC (as the commercial product formulation, Cecure[®]) was approved by the United States Food and Drug Administration (2004) for use on raw, pre-chill poultry (21 Code of Federal Regulations, Section 173.375). In addition, an amendment to this regulation has recently been filed to allow for treatment of raw poultry (whole carcasses) post-chill (United States Food Additive Petition No. 6A4767). The pre-chill application of Cecure[®] is the subject of a recently published article describing successful commercial use of the product in three commercial broiler processing plants in the United States (Beers *et al.*, 2006). These authors focused on control of total aerobic plate count (APC), *E. coli*, coliforms, *Campylobacter*, and *Salmonella* during on-line reprocessing of ingesta-contaminated pre-chill broiler carcasses. CPC is a positively charged cationic quaternary ammonium compound commonly found in many over-the-counter brands of mouthwash, toothpaste, throat lozenges, nasal sprays and topical antiseptic ointments. The molecular formula of CPC is C₂₁H₃₆NCl; the molecular weight is 340. The Cecure[®] product is a concentrate of 40% (by weight) U.S. Pharmacopeia-grade CPC in food grade propylene glycol and water. The product has a neutral pH, is diluted in water to the appropriate concentration for use and is stable under normal storage conditions.

Numerous studies have cited the antimicrobial properties of CPC for control of microorganisms on raw poultry (Breen *et al.*, 1995; Kim and Slavik, 1996; Breen

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Table 1: Aerobic Plate Count¹ on boneless, skinless broiler thighs sprayed with Cecure[®]%

Days of Storage at 2.5°C (Trial 1) log ₁₀ CFU/mL (n = 4) ³											
Cecure [®] , ²	0	1	2	3	4	5	6	7	8	9	10
0.0 ³	2.3 ^{b-s}	2.1 ^{q-s}	2.6 ^{p-r}	3.1 ^{m-p}	3.8 ^{j-m}	4.6 ^{b-j}	5.2 ^{gh}	6.2 ^{ef}	7.1 ^{b-d}	7.5 ^{ab}	8.0 ^a
0.5	1.5 st	1.9 ^{qs}	2.3 ^{p-s}	3.1 ^{m-p}	3.5 ^{k-n}	4.2 ^{jk}	5.1 ^{g-i}	5.7 ^{eg}	6.5 ^{ef}	7.1 ^{b-d}	7.6 ^{ab}
1.0	1.1 ^t	1.6 st	2.2 ^{q-s}	2.5 ^{p-r}	3.4 ^{l-o}	3.9 ^{il}	4.3 ^{ik}	5.6 ^{fg}	6.1 ^{ef}	6.5 ^{c-e}	7.2 ^{ac}
1.5	1.0 ^t	1.5 st	1.9 ^{qs}	2.9 ^{n-q}	3.1 ^{m-p}	4.6 ^{b-i}	4.6 ^{b-j}	5.7 ^{eg}	6.5 ^{c-e}	6.3 ^{df}	7.1 ^{b-d}

¹Least Squares means values followed by different letters are significantly different ($p \leq 0.05$), ²Room temperature Cecure[®] spray (7mL/thigh); control sprayed with room temperature tap water (7 mL/thigh), ³Raw poultry is considered spoiled at 7.0 (log₁₀ CFU/mL)

Table 2: Aerobic Plate Count¹ on boneless, skinless broiler thighs sprayed with Cecure[®]%

Days of Storage at 2.5°C (Trial 2) log ₁₀ CFU/mL (n = 4) ³											
Cecure [®] , ²	0	1	2	3	4	5	6	7	8	9	10
No water spray ⁴	2.5 ^{t-v,3,4}	2.8 ^{p-u}	3.2 ^{n-s}	3.7 ^{m-p}	3.7 ^{m-p}	5.3 ^{h-k}	6.5 ^{d-g}	6.6 ^{ef}	7.2 ^{ae}	7.9 ^a	8.1 ^a
0.0	2.9 ^{p-u}	2.6 ^{q-u}	3.3 ^{n-r}	3.6 ^{m-q}	4.5 ^{k-m}	4.6 ^{km}	6.3 ^{e-h}	6.7 ^{cf}	7.1 ^{ae}	8.0 ^a	7.9 ^a
0.4	1.9 ^{uv}	2.6 ^{q-u}	2.7 ^{p-u}	3.2 ^{n-s}	4.1 ^{hn}	4.8 ^{il}	6.0 ^{f-h}	6.3 ^{d-g}	6.8 ^{bf}	7.6 ^{ac}	7.8 ^a
0.5	1.5 ^w	2.1 ^{t-v}	2.8 ^{p-u}	3.3 ^{n-r}	3.9 ^o	4.4 ^{km}	6.0 ^{f-h}	6.4 ^{d-g}	6.8 ^{c-f}	7.3 ^{ad}	7.8 ^a
1.0	0.7 ^w	1.9 ^{uv}	2.2 ^{s-v}	3.1 ^{o-t}	4.0 ^o	4.9 ^l	5.6 ^{gj}	5.8 ^{li}	6.6 ^{c-f}	6.7 ^{cf}	7.6 ^{ac}

¹Least Squares means values followed by different letters are significantly different ($p \leq 0.05$), ²Room temperature Cecure[®] spray (7mL/thigh); control sprayed with room temperature tap water (7 mL/thigh), ³Raw poultry is considered spoiled at 7.0 (log₁₀ CFU/mL). ⁴Thighs were not sprayed with water or Cecure[®]

et al., 1997; Xiong, 1998; Waldroup *et al.*, 1999; 2000). However, none of the above mentioned studies investigated the use of CPC for treating boneless, skinless product or evaluated refrigerated shelf life of CPC-treated poultry products.

The objective of the following study was to investigate the effects of using Cecure[®] to treat post-chill boneless, skinless broiler thighs on the subsequent refrigerated shelf life.

Materials and Methods

Treatment conditions: Two similar trials were conducted over a two month period. For each trial, post-chill boneless, skinless chicken thighs (n = 240) were obtained from a local commercial processing facility immediately after the skinning and boning operations (“Day 0”). It should be noted that Day 0 in the tables and figures represents the day which the thighs were skinned and boned (as well as the day they were treated with Cecure[®]), but does not represent the day of slaughter. Thus, the thighs may possibly have been one or two days post-slaughter on Day 0. The thighs were bagged and placed in ice for transport to the laboratory (less than a 30-min transport time). Upon arrival at the laboratory³, the thighs were randomly divided into four treatment groups of 60 thighs each. In each trial, within each treatment group, the thighs were further divided into groups of four and were placed onto Styrofoam[™] trays. Thus, for each treatment group there were 15 trays, each holding four thighs.

On day 0, all thighs were sprayed with either 7 mL of sterile tap water (control) or 7 mL of a Cecure[®] treatment group (0.5, 1.0 and 1.5% CPC in Trial 1; 0.4, 0.5 and 1.0% CPC in Trial 2). In Trial 2 there was also a group of thighs that were not sprayed with water or Cecure[®]. The

spraying operation was accomplished using a small plastic hand-held spray bottle. The operator sprayed 3.5 mL of solution on one side of each thigh and then turned the thighs over with gloved hands and sprayed the other side of each thigh with the remaining 3.5 mL. All spray solutions were at ambient temperature. All thighs remained on the trays during the spray application. No attempt was made to remove any residual spray solution from the trays. After spraying, each individual tray of four thighs was placed into a gallon-size plastic Ziploc[®] storage bag (no attempt was made to evacuate the air). For each trial, one tray of thighs per treatment was microbiologically sampled on Day 0; all other trays of thighs were placed in a walk-in refrigeration unit held at 2.5°C.

Microbiological sampling procedures: In each trial, one tray (n = 4) of thighs was microbiologically sampled each day beginning on Day 0 and ending on Day 10. In addition, on each sampling day the thighs were also evaluated subjectively, by the authors, for any uncharacteristic odor or discoloration. For microbiological sampling, each individual thigh was placed inside a quart-size Ziploc[®] bag and was rinsed (and gently massaged) by hand for one minute in 100 mL 0.1% buffered phosphate. All rinse samples were serologically plated on Petrifilm[™] according to the manufacturer’s directions. The Petrifilm[™] was incubated at 30°C for 48 h for APC. The lower detection level for the APC was 1.0 CFU/mL of rinse fluid.

Statistical analyses: All microbiological data were transformed to log₁₀ CFU/mL prior to statistical analysis. For any negative samples the lower detection level was utilized in the analysis. Data were analyzed using the Tukey-Kramer HSD (Honesty Significant Difference) and

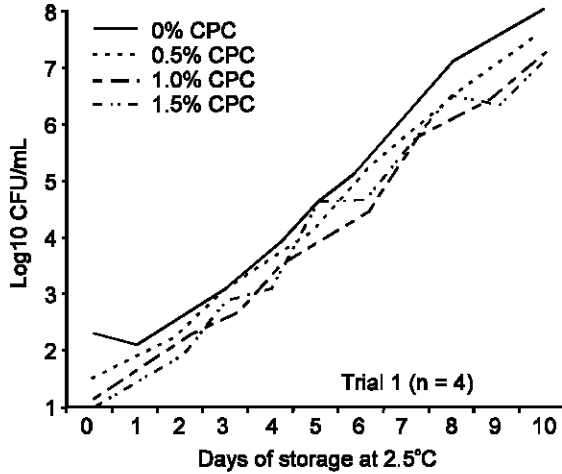


Fig. 1: Aerobic Plate Count on boneless, skinless broiler thighs sprayed with Cecure® (Trial 1)

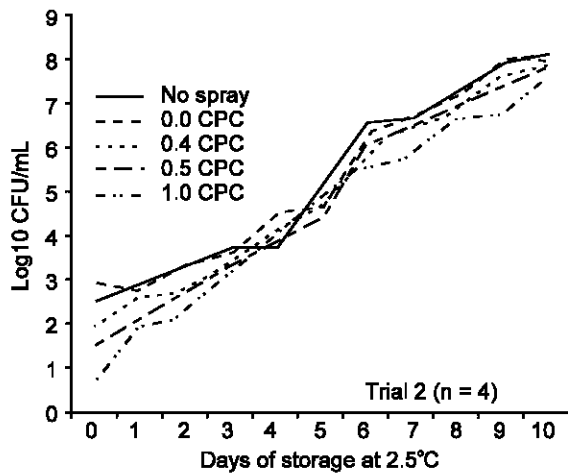


Fig. 2: Aerobic Plate Count on boneless, skinless broiler thighs sprayed with Cecure® (Trial 2)

Abs (Dif)-LSD (Least Squares Difference) procedures⁵ as well as the GLM (General Linear Model) procedure of SAS⁶.

Results and Discussion

There was a significant sampling day by treatment (concentration of CPC) interaction in both trials. Thus, the Least Squares APC mean values (\log_{10} CFU/mL) are presented in Table 1 and 2, respectively. In Trial 1, both the 1.0 and 1.5% Cecure® spray treatments resulted in significant reductions in the APC on Day 0 (1.2 to 1.3 logs) (Table 1). In a study conducted by Waldroup *et al.* in 2000, post-chill broiler carcasses were sprayed with either 0.25% or 0.4% CPC (as Cecure®) and reductions in APC ranged from 0.8 to 2 logs on the day of application. Beers *et al.* (2006) reported initial reductions

in APC on pre-chill broilers treated with 0.5 to 0.7% Cecure® ranging from 2 to 2.5 \log_{10} CFU/mL.

On days 1 through 5 the bacterial counts between treatments were not statistically different; however, in general, numerical counts were lower for all Cecure® treatments on each of these days in comparison to control counts. By Day 6, the 1.0% Cecure® treatment group had a significantly lower count than did the control group and this trend was consistent through Day 9 for this treatment group. By Day 8, the control had reached the spoilage level of \log_{10} 7 CFU/mL (Ayres *et al.*, 1950; Mielnik *et al.*, 1999). The 0.5% Cecure® treatment group spoiled on Day 9 and the 1.0 and 1.5% Cecure® groups spoiled on Day 10. At spoilage, all groups of carcasses were slimy and exhibited the characteristic spoilage odor associated with raw poultry (Jay, 2006). There was no observable color difference between treatment groups on any of the days of sampling. Fig. 1 is a visual representation of the bacterial growth curves for the treatment groups in Trial 1. It appears that the extension in product shelf life was related to the initial bacterial reduction on the day of treatment (Day 0) since the slope and shape of all growth curves were very similar throughout the 10-day storage period.

In Trial 2, only the 1% Cecure® treatment resulted in a significant reduction in APC on Day 0. This agrees with the findings in Trial 1; however, the reduction with 1% Cecure® in Trial 2 was much greater than in Trial 1 (1.2 logs in Trial 1; 2.2 logs in Trial 2). In a study conducted by Cutter *et al.* (2000) on beef carcass shortplates, a 1% CPC spray resulted in a $> 2 \log_{10}$ reduction in APC on the day of treatment. In the present study, it should be noted that on Day 0 the level of APC on the control group was much greater in Trial 2 than in Trial 1. This was most likely due to the amount of time that had elapsed between slaughter and boning and skinning in the two separate trials. On Day 0, the water spray did not produce a significant reduction in APC compared to the control that was not sprayed. In the beef study conducted by Cutter *et al.* (2000), a water spray at 35°C did significantly reduce \log_{10} APC. It should be noted that the water spray in the current experiment was at ambient temperature. The 1% Cecure® treatment produced a significantly greater reduction in APC than did the 0.4% Cecure® treatment, but was not different from the 0.5% Cecure® treatment on Day 0. Breen *et al.* (1997) clearly demonstrated that the antimicrobial effects of CPC are directly dependent upon concentration and exposure time.

On days 1 through 8, APC were not significantly affected by treatment; however, counts were consistently numerically lower for the 0.5 and 1.0% Cecure® treatment groups. In fact, by Day 8, the “no water spray” group and the control group had reached spoilage levels

(Ayres *et al.*, 1950; Mielnik *et al.*, 1990). By Day 9, a significant decrease was noted in the APC for the 1.0% Cecure[®] treatment; in addition, the 0.4 and 0.5% Cecure[®] treatment groups were spoiled. By Day 10, the 1.0% Cecure[®] treatment group had spoiled and all counts were equal regardless of treatment. As in Trial 1, it appears that the extension in shelf life (1 day for the 0.4 and 0.5% Cecure[®] treatments; 2 days for the 1.0 and 1.5% Cecure[®] treatments) was due to the initial reduction on Day 0, even though reductions on the initial day of the trial were not always statistically significant. As noted in Trial 1, the slope and shape of the growth curves were similar regardless of treatment. There were no observable differences between treatment groups on any of the sampling days in regards to uncharacteristic odor or alterations in color.

Data from these trials demonstrate that Cecure[®] can be utilized to extend the shelf life of fresh refrigerated boneless, skinless chicken muscle without producing any adverse sensory effects. This extension in shelf life is due to an initial reduction in total organisms on the day of treatment. Application of 0.4% or 0.5% Cecure[®] resulted in a 1 day extension in shelf life and application of 1.0% or 1.5% Cecure[®] resulted in a two day extension in shelf life when the product was held refrigerated at 2.5°C.

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⁶SAS Institute, Inc., 1999. SAS, Version 8. Cary NC 27513, USA.