Extension of Poultry Shelf-Life by Poly (Hexamethylenebiguanide Hydrochloride)

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ABSTRACT

Immersion of freshly processed poultry carcasses in solutions of poly (hexamethylenebiguanide hydrochloride), PHMB, retarded bacterial growth and markedly improved the shelf-life during storage at 2°C. Birds treated with 200, 300, and 400 ppm PHMB had average shelf-lives of 22.9, 25.9, and 26.0 days, respectively, compared to the 10.5 days of shelf-life for water-treated controls. Duncan's multiple range test revealed that the shelf-life differences among PHMB-treated birds were not statistically significant.

The limited shelf-life of fresh poultry has been a serious problem to the poultry industry. Methods for prolonging the freshness of poultry have constantly been sought. These included irradiation, pasteurization, storage in germicidal ice, use of edible coating, and exposure to antibiotics and other chemicals (7, 13). Mountney and O'Malley (11) extended shelf-life of cut-up poultry parts 6 days beyond that of controls by dipping them in solutions of succinic and adipic acids at pH 2.5. However, as the pH was raised the preservative effect diminished and at pH 4.0 no inhibition of spoilage was observed. Heat treatment in combination with succinic acid was shown by Cox et al. (3) to retard poultry spoilage, but it resulted in bleached skin and a "cooked" appearance. In-plant chlorination is effective in reducing slime and odor on processing equipment, but it has very little effect on poultry shelf-life when added to the chill-immersion tanks (10). The combination of chlorine, succinic acid and heat, extended shelf-life, but caused discoloration of the skin due to partial cooking (6). Islam et al. (7) screened 53 chemicals and found that chloroacetamide and iodoacetamide extended poultry shelf-life for 3 and 4 days, respectively, at a storage temperature of 5°C. Thomson et al. (17) found that 0.5% glutaraldehyde extends poultry shelf-life about 6 days beyond that of controls at 2°C. Arafa and Chen (1) obtained similar shelf-life improvement for cut-up broiler parts by dipping in 1% ascorbic acid solution of pH 2.75.

Poly (hexamethylenebiguanide hydrochloride) or PHMB has been used successfully as a biocide for several years, particularly in the European countries. Under the trade name "Vantocil IB" it has been marketed for industrial disinfection (2) and for short-term preservation of cattle hides and sheepskins (4, 5). In addition under the trade name of "Bacquacil SB" it has been marketed for several years as a swimming pool sanitizer (6). PHMB alone or in combination with other chemicals also retards development of dental plaque, calculus and caries in animals (12, 14, 18). Strandskov and Bockleman (15) in 1975 obtained a U.S. patent for preservation of beverages such as beer by incorporating PHMB up to a level of 50 ppm.

In view of the above demonstrated applications, PHMB seemed to have a good potential for use as a poultry preservative. Hence, this study was designed to evaluate the effectiveness of poly (hexamethylenebiguanide hydrochloride) in extending the shelf-life of fresh poultry.

MATERIALS AND METHODS

Preparation of PHMB solutions

Distilled water in 14-liter lots was poured into four double-layered 30-gal. size plastic bags. Appropriate amounts of 20% PHMB solution ("Vantocil IB") obtained from the ICI-Americas, Wilmington, Delaware, were added to the water to reach the final concentrations of 0, 200, 300, and 400 ppm, respectively. The plastic bags containing these solutions were placed in rigid containers for easier handling, and then stored at 2 ± 0.5°C for at least 12 h before use. Portions (about 25 ml) from each of these solutions were withdrawn with sterile pipettes and their pH values determined by a Beckman digital pH meter.

Sample preparation

Twenty-eight fresh broiler carcasses were brought to the laboratory ice-packed in foamed plastic containers from a nearby processing plant. The carcasses were randomly divided into four groups of seven for treatment with the PHMB solutions prepared above. The birds were submerged in respective solutions for 2 h at 2 ± 0.5°C and then drained for 10 min on alcohol-swabbed metal racks. They were then packaged in gallon-size clear polyethylene bags and stored at 2 ± 0.2°C. Three carcasses from each group were designated for sensory evaluation and the remaining four for bacterial count during storage. Bacterial count was limited to the total aerobic psychrotrophs because of their predominance in spoiled chicken (3, 7).

Sensory evaluation

Since two recent studies (1, 17) using similar storage conditions have shown that the control chickens remain fresh for at least 10-11 days, the sensory evaluation in this study began after a week of storage at 2 ± 0.2°C. Commencing on the 8th day of storage, and every alternate day thereafter, a five-membered panel consisting of departmental faculty and graduate students evaluated the carcasses for development of off-odor. The following statement appeared on the evaluation forms, "please open each bag, smell the carcass and then rate the off-odor on a 5-point hedonic scale where 1, 2, 3, 4, and 5 represent none, slight, moderate, strong, and very strong, respectively. A score of 3 or less would indicate that the bird is acceptable to you as a potential consumer". This method of evaluation is somewhat similar to that described by Thomson et al. (17). When a bird was rated 5 by at least three of the five panels it was removed from the observation.

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Shelf-life for each bird was calculated based on the number of days it maintained a sensory score of 3 or less. The shelf-life data were then analyzed as a 4 x 5 factorial in a completely randomized design with the treatments and the panelists as the main effects. This was followed by Duncan’s multiple range test to find if there was any statistically significant difference between treatments (9). A separate but similar analysis was carried out in a 12 x 5 factorial arrangement with the birds and the panelists as the main effects, to find if there was any difference between the birds within the and between each treatment group.

Total aerobic psychrotroph count

The bacterial densities on the right and left breasts of the broiler carcasses were assumed to be the same based on the earlier studies by Cox et al. (9) and Kotula (8).

Two of the four carcasses designated for bacterial count were swabbed on the right breast before and after dipping on day 0, and on the left breast on day 5 and 10. The other two carcasses from each group were swabbed subsequently on days 15, 20, 25, and 30. This type of split-sampling within a group was necessary because of the limitation of swabbing space on the same bird. Also, if the same bird was swabbed eight times over a 30-day period, it would have been subjected to a greater degree of contamination. The birds which scored 5 because of their off-odor were swabbed only once beyond their final evaluation day. A 4-cm² area at different locations on the chicken breast was swabbed on each sampling day. Appropriate dilutions were prepared in 0.1% peptone and spread-plated in duplicate on tryptic soy agar (TSA, Difco). The plates were incubated at 7°C for 10 days before enumeration of total aerobic psychrotrophs. Average number of colonies from the four plates in each group were divided by four and reported as log bacterial count per cm².

RESULTS AND DISCUSSION

Total aerobic psychrotroph count

PHMB, at the levels used in this study, had an unusually strong bactericidal effect on the microbial population of the chicken carcasses compared to that of the other potential poultry preservatives reported in the literature (1,3,7). This strong bactericidal effect resulted in marked improvement in the shelf-life of PHMB-treated carcasses. The average initial psychrotroph count on the untreated birds was about 10^5 organisms/cm² of skin. These same birds, when swabbed immediately after dipping in PHMB solutions, had so few organisms that no bacterial count could be obtained even at the lowest dilution (10-fold) used. For the control samples there were slight decreases in bacterial count possibly due to the mechanical rinsing effect; however, within the 5th day of storage the log count exceeded its original level (Fig. 1). At the 5th day, the PHMB-treated samples had a very small increase in bacterial growth, but the number of colonies was too few to count except for the 200-ppm group. Hence, to plot growth curves, the unobtainable counts were estimated and linked with dotted lines (Fig. 1).

After the initial reduction in bacterial count, growth curves for all of the groups exhibited a similar lag phase which lasted about 5 days before entering the exponential or the log phase. During this phase the rate of microbial growth for the control group was considerably greater than the growth rates of the flora on PHMB-treated groups. The difference in the rate of growth was partly responsible for the longer shelf-life of PHMB-treated birds. But the main extension of shelf-life seems to have originated from the initial bactericidal effect of PHMB.

At each of the sampling intervals, the average bacterial count on the birds dipped in 200 ppm of PHMB was found to be higher than those on the birds dipped in 300 or 400 ppm PHMB. This difference, although very small, was consistent throughout the study. However, there was no such consistent trend in the bacterial counts between the 300- and 400-ppm groups. Overall, the average bacterial counts on all the PHMB-treated birds were very similar despite the large difference in chemical concentration. This suggests that the bactericidal effect of PHMB reached a plateau at about 200 ppm. Perhaps another study with lower concentrations of PHMB would help identify the optimum concentration.

Sensory evaluation

Data on development of off-odor during storage are presented in Fig. 2. The five judgements on each bird were averaged and then the mean score of the three birds per treatment was plotted for each evaluation day. Some of the panel members detected slight off-odor in a few carcasses on the first day of evaluation which indeed was the 8th day of storage at 2°C. This off-odor, particularly on some PHMB-treated birds, was likened to that of “fresh wheat flour” by two of the panel members. While most of the control samples exhibited strong off-odor on the 10th day of storage, the PHMB-treated samples maintained slight to moderate off-odor until the
20th day. Table 1 lists the shelf-lives of all the 12 birds that were designated for sensory evaluation. These shelf-life data were tabulated based on the number of days the birds maintained a score of 3 or less. Analysis of variance on the 12 x 5 factorial arrangement showed a highly significant difference (p < 0.01) in the shelf-lives of the 12 carcasses. Within the control group, the three birds had shelf-lives of 10.6, 9.6, and 11.4 days, respectively, and these values were considerably lower than the lowest value (19.2 days) with PHMB treatment. Duncan’s multiple range test showed that statistically there was no difference (p > 0.01) in the shelf-life values of the three control samples. Significant differences (p < 0.01) existed among the nine birds within and between the PHMB treatments although there was considerable overlapping (Table 1). Analysis of variance and Duncan’s multiple range test on the 4 x 5 factorial design clearly revealed that there was no significant difference (p > 0.01) between the three PHMB treatment in regard to the shelf-life extension. In other words, statistically the shelf-life improvement with 200 ppm was the same as with the 300 or 400 ppm PHMB. This observation on shelf-life seems to be consistent with the effect of PHMB on bacterial count. The effect of 200 ppm PHMB reached a plateau for both bacterial inhibition as well as shelf-life improvement. In view of these findings, another study with lower concentrations of PHMB is being planned.

pH of PHMB solutions

At about 2 C the pH values of the 200, 300, and

![Figure 2. Off-odor development in poultry carcasses treated with various concentrations of poly(hexamethylenebiguanide hydrochloride) and stored at 2 C.](image)

### Table 1. Shelf-life of poly(hexamethylenebiguanide hydrochloride) treated chicken carcasses at storage temperature of 2 C.

<table>
<thead>
<tr>
<th>Treatment PHMB (ppm)</th>
<th>Individual carcasses Mean ± S.D.</th>
<th>Overall mean for each treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>10.6 ± 6a</td>
<td>10.5x</td>
</tr>
<tr>
<td>200</td>
<td>19.2 ± 1.6b</td>
<td>22.9y</td>
</tr>
<tr>
<td>300</td>
<td>22.4 ± 3.0bc</td>
<td>25.9y</td>
</tr>
<tr>
<td>400</td>
<td>29.2 ± 2.5e</td>
<td>26.0y</td>
</tr>
</tbody>
</table>

*Means followed by different letters within each column are significantly different (p < 0.01) according to Duncan's multiple range test.*

**Overall efficacy of PHMB**

The mean shelf-lives of poultry using 200, 300 and 400 ppm PHMB were 22.9, 25.9, and 26.0 days, respectively, compared to only 10.5 days shelf-life for the control samples (Table 1). Thus, the PHMB, at levels used in this study, extended poultry shelf-life from 12 to 14 days beyond that of the controls. This degree of shelf-life extension is indeed encouraging since the most recent attempts with other chemicals (7,17) improved poultry shelf-life a maximum of only 6-7 days. However, one ought to realize that this preservation or the absence of spoilage is based on only the low microbial counts and the lack of off-odor development. No attempt was made in this study to assess the extent of tissue breakdown due to enzymatic action; also there was no evaluation of the cooking quality of PHMB- preserved chicken. Organoleptic evaluation involving taste, however, cannot be done till there are sufficient toxicological data available on PHMB.

The safety of PHMB is suggested because of its use as a sanitizing agent in swimming pools and for food processing and brewery fermentation equipment. Specific pharmacological and toxicological investigations will be necessary before it can be used as a poultry preservative.

**ACKNOWLEDGMENTS**

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