Summary

Catalase present in the suspending medium during irradiation significantly reduced the lethal action of gamma radiation from cobalt-60 for anaerobic bacterial spores. This finding supports the theory that the lethal action of ionizing radiations is at least partially due to secondary effects of the irradiation. Furthermore, such protection of anaerobic bacterial spores is important when sterilization of foods containing catalase is considered inasmuch as it will likely increase the dosage required.

REFERENCES

Reed, J. M., Bohrer, C. W., and Cameron, E. J. 1951 Spore destruction rate studies on organisms of significance in the processing of canned foods. Food Research, 16, 383-408.

Antibiotic Treatment of Crab and Oyster Meats¹

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Laboratory studies of broad-spectrum antibiotics for food preservation are well documented. Primarily, these studies attempted to extend the edible life of highly perishable commodities.

Economic loss accruing to both processor and consumer from spoilage of such commodities as poultry, beef and comminuted meats, leafy vegetables, and fin and shellfish, approaches the astronomical. Of these, shellfish have received comparatively little study. As a group, however, they are probably the most highly perishable of all food items; bacterial decomposition proceeds with extreme rapidity shortly after landing.

Surprisingly enough, however, the industry’s annual loss through spoilage is lower than would be anticipated. This is due to the fact that the processor is forced to place his product into commercial distribution channels shortly after the catch is brought in. Thus, he creates an unstable economy for himself, and is unable to seek additional markets in areas beyond the present transportation-storage life of his product.

Present cold storage conditions normally obtained in processing plants can preserve the typically fresh flavor and appearance of crabmeat from 3 to 10 days and up to 15 days for oysters depending upon the degree of bacterial contamination during picking and/or shucking and packing. Apparently, cold storage alone is not sufficient to prevent deterioration caused by the rapid proliferation of psychrophilic bacteria commonly found in crab and oyster meats. Supplementary treatment appears necessary.

Although no definitive bacterial study of crabmeat spoilage has been made, between five and ten genera have been isolated from spoiled meat (Fieger et al., 1956; Gardner and Watts, 1956). For this reason, it was believed that the broad-spectrum antibiotics, particularly the tetracycline group, supplementing refrigeration, might secure additional storage life. The investigation to be reported was undertaken in an attempt to test this hypothesis.

Although shellfish, including both mollusks (oysters, clams, snails, mussels, and scallops) and crustaceans (crabs, lobsters, shrimp, and crayfish) have great commercial value, comparatively few reports concerning their preservation have been published.

Farber (1954) reported oxytetracycline and chlorotetracycline to be ineffective for shrimp preservation. This has been supported by recent findings of Tomiyama (1955) and Fieger et al. (1956).

Wrenshall (1956) reported a 5-fold increase in storage life of lobsters and clams treated with oxytetracycline.

The data to be reported here were obtained from the treatment of fresh crab and oyster meats with oxy- and chlorotetracycline.

Material and Methods

For tests with crabmeat, food-grade formulations of oxy- and chlortetracycline\(^2\) were dissolved in 5 gallons of tap water to yield concentrations of 5, 10, 15, 20, 30, 40, and 60 \(\mu g\) per ml.

Half-pound quantities of “regular”\(^3\), meat of the Atlantic Blue Crab (Callinectes sapidus Rathbun) were obtained from commercial crab houses and our laboratory. These were placed onto squares of no. 10 cheesecloth and fashioned into sacks tied with rubber bands. The sacks were immersed in appropriate antibiotic solutions for 2 min. During a portion of this time the sacks were agitated to insure adequate bathing of all meat. At 2 min, the sacks were drained of excess liquid and the meat placed in half-pound snap-lock crab cans for storage at 1 C.

Control samples were dipped in untreated tap water or packed without dipping.

At intervals, samples were removed to determine bacterial counts, pH, antibiotic residues, and sensory characteristics. A 50 g sample of meat was placed into a chilled food blender containing 450 ml of sterile, chilled, distilled water. After 3 min of blending, additional 10-fold serial dilutions were made. Triplicate 1-ml portions of the appropriate dilutions were plated in nutrient agar (BBL) pH 6.8 and incubated at 24 ± 0.5 C for 72 hr. Bacterial plate counts were made at that time. Sensory evaluation was performed by the laboratory personnel.

Food and Drug Administration cup-plate tests for residues of oxy- and chlortetracycline were performed by Chas. Pfizer & Co. and American Cyanamid, respectively. In certain instances, the Antibiotics Division of the Food and Drug Administration assayed both products.

The procedures for oysters varied somewhat. Fifty gallons of tap water were drawn into 200 gallon “blow” tanks. Sufficient antibiotic was added to obtain concentrations of 5 or 15 \(\mu g\) per ml.

Five gallons of “select” and “extra-select”\(^4\) oysters (Crassostrea virginica) were added to each 50 gallon solution to be “blown”\(^5\) for 3 min. Controls were “blown” in untreated tap water. After “blowing,” the oysters were spraywashed, packed in pint cans and sealed. These procedures were performed in a commercial oyster house. The cans were stored in the laboratory at 1 C.

As the moisture content of oysters was sufficiently high to yield a smooth homogenate, no additional water was needed for blending.

All remaining procedures were similar to that used with crabmeat.

Results and Discussion

Laboratory processed meat was used for the initial trial. Only sensory judgments were made on these samples.

Four days after dipping the meat, all samples were considered “typical.” On the 11th day, all samples were “off,” and when opened on the 14th day, they were all “foul.” This storage life was similar to that generally obtained with laboratory picked meat. Apparently, antibiotic residues of 2.0 and 2.2 for the 5 ppm dip of oxy- and chlortetracycline respectively, were unable to prevent the normal bacterial growth. Similarly ineffective was the 3.6 and 2.2 residues obtained with the 10 ppm dip.

This test was repeated using commercially processed meat. The figures in table 1 show the inadequacy of 5 and 10 ppm dips. All treated meat was putrid by the 6th day. In fairness, however, it must be pointed out that this result was completely unexpected. The very high count of the untreated sample at 24 hr was unusual. When the facts were ascertained, it was learned that the meat purchased as “fresh,” was not so. However, it served to indicate that poor quality meat could not be rehabilitated by antibiotic treatment.

<table>
<thead>
<tr>
<th>Treatment: 2 Min Dip Conc. Total Viable Aerobic Bacteria (\times 10^6) g per Days of Storage</th>
<th>0</th>
<th>1</th>
<th>4</th>
<th>6</th>
<th>8</th>
</tr>
</thead>
<tbody>
<tr>
<td>ppm</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>OTC(^*)</td>
<td>5</td>
<td>0.20</td>
<td>0.081</td>
<td>3.70</td>
<td>650 F(^†)</td>
</tr>
<tr>
<td>CTC(‡)</td>
<td>5</td>
<td>0.27</td>
<td>0.014</td>
<td>3.10</td>
<td>700 F</td>
</tr>
<tr>
<td>OTC</td>
<td>10</td>
<td>0.15</td>
<td>0.078</td>
<td>2.10</td>
<td>650 F</td>
</tr>
<tr>
<td>CTC</td>
<td>10</td>
<td>0.047</td>
<td>0.32</td>
<td>3.10</td>
<td>31 F</td>
</tr>
<tr>
<td>Untreated (not dipped)</td>
<td>0</td>
<td>0.090</td>
<td>1.70</td>
<td>22.0 F</td>
<td>100 F</td>
</tr>
</tbody>
</table>

\(^*\) Oxytetracycline as contained in Biostat, product of Chas. Pfizer & Co., New York.

\(^†\) F = foul.

\(‡\) Chlortetracycline as contained in Acrizon, product of American Cyanamid Chemical Corp., New York City.

Storage temperature, 1 C.

All data are the mean values of triplicates showing variations, normally encountered in plate counts.
TABLE 2

Effect of various antibiotic treatments on the spoilage of fresh crabmeat (laboratory picked)

<table>
<thead>
<tr>
<th>Treatment: 2 Min. Dip</th>
<th>Conc ppm</th>
<th>Total Viable Aerobic Bacteria × 10⁵/g per Days of Storage</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>0</td>
</tr>
<tr>
<td>OTC*</td>
<td>15</td>
<td>0.17</td>
</tr>
<tr>
<td>CTC†</td>
<td>15</td>
<td>0.0024</td>
</tr>
<tr>
<td>OTC</td>
<td>20</td>
<td>0.0039</td>
</tr>
<tr>
<td>CTC</td>
<td>20</td>
<td>0.013</td>
</tr>
<tr>
<td>Tap water dip</td>
<td>0</td>
<td>0.025</td>
</tr>
<tr>
<td>Untreated (not dipped)</td>
<td>0</td>
<td>0.0062</td>
</tr>
</tbody>
</table>

* Oxytetracycline as contained in Biostat, product of Chas. Pfizer & Co., Brooklyn, New York.
† B = "off" odor; F = foul.
‡ Chlortetracycline as contained in Acronize, product of American Cyanamid Chemical Corp., New York City.
Storage temperature 1°C.
All data are the mean values of triplicates showing variations, normally encountered in plate counts.

TABLE 3

The effect of various antibiotic treatments on the spoilage of fresh commercial crabmeat

<table>
<thead>
<tr>
<th>Treatment: 2 Min Dip</th>
<th>Conc ppm</th>
<th>Total Viable Aerobic Bacteria × 10⁵/g per Days of Storage</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>1</td>
</tr>
<tr>
<td>OTC*</td>
<td>30</td>
<td>0.014</td>
</tr>
<tr>
<td>CTC†</td>
<td>30</td>
<td>0.072</td>
</tr>
<tr>
<td>OTC</td>
<td>40</td>
<td>0.03</td>
</tr>
<tr>
<td>CTC</td>
<td>40</td>
<td>0.0012</td>
</tr>
<tr>
<td>Untreated control (not dipped)</td>
<td>0</td>
<td>0.0012</td>
</tr>
</tbody>
</table>

* Oxytetracycline as contained in Biostat, product of Chas. Pfizer & Co., Brooklyn, New York.
† F = foul.
‡ Chlortetracycline as contained in Acronize—product of American Cyanamid Chemical Corp., New York City.
Storage temperature 1°C.
All data are the mean values of triplicates showing variations, normally encountered in plate counts.

The seeming extension of storage time obtained in the first trial was doubtlessly due to the initial high sanitary quality of the laboratory meat rather than an effect of the chemical treatment.

Increasing the solution concentrations to 15 and 20 μg per ml was similarly unavailing. Table 2 summarizes this test. At 12 days, meat dipped in 20 ppm chlortetracycline was judged acceptable by the staff although a slightly higher count was present. At the 14th day, this treatment appeared better than the others, but it too was "off." Although a bacterial suppressing action appeared to be operating, it was not sufficient to prevent customary spoilage. Here the presence of residues of 2.52 and 3.60 μg per g of oxy- and chlortetracycline, respectively, for the 15 ppm dip, and 2.40 and 2.88 μg per g for the 20 ppm after 15 days, was not protective.

The results of dipping crabmeat in solutions containing 30 and 40 μg per ml are noted in table 3.

By the 8th day of storage, the counts were lower than expected. That the control had less than one million at that time, indicated a good to high quality product to begin with. And this was substantiated by the 24 hr counts, all low.

By the 15th day, however, all the meat showed the spoilage normal for that storage period. Here again, treatment was ineffective. Oxytetracycline residues present in the meat at the end of the storage were 2.8 and 3.7 μg per g for the 30 and 40 ppm dips, respectively, while chlortetracycline assayed at 5.2 and 6.7 μg per g.

To try to break-through the 15 day storage period, dips containing 60 ppm were employed. Storage life in this instance was judged solely by organoleptic evaluation. Residue analysis showed that the meats contained 35.0 and 37.0 μg per g of oxy- and chlortetracycline respectively at the beginning of the storage period. Again, 15 days was the storage limit. At that time, the meats contained 13.5 and 22.0 μg per g of the antibiotics. In addition, the chlortetracycline treated meat had a green cast. This would, of course, preclude its use.

The data reported above appear to indicate that although antibiotic treatment could briefly suppress bacterial numbers, it was ineffective in extending the presently attainable storage life of crabmeat.

In general, a rising pH was seen as the storage period increased. Although this is in accord with the findings of others (Alford et al., 1941; Harris, 1932) that fresh crabmeat pH's between 7.4 to 7.6 and rises to 8.0 to 8.5 on spoiling, it is not generally used as a guide to quality or indicator of storage life.

Table 4 compares two concentrations of each of the antibiotics against an untreated control for ability to extend storage life of shucked oysters.

In general, the treated group, irrespective of antibiotic or concentration, had lower bacterial counts than the untreated control. The lower counts of the treated samples became evident at the 10th day and persisted through the 50th day.
TABLE 4  
Effect of various antibiotic treatments on the spoilage of commercially shucked oyster meats

<table>
<thead>
<tr>
<th>Treatment: Blow Tank 3 Min.</th>
<th>Conc</th>
<th>Total Viable Aerobic Bacteria X 10^6 /g per Days of Storage</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>ppm</td>
<td>0</td>
</tr>
<tr>
<td>OTC*</td>
<td>5</td>
<td>0.35</td>
</tr>
<tr>
<td>CTC†</td>
<td>5</td>
<td>0.52</td>
</tr>
<tr>
<td>CTC</td>
<td>15</td>
<td>0.43</td>
</tr>
<tr>
<td>OTC</td>
<td>15</td>
<td>0.75</td>
</tr>
<tr>
<td>Untreated (not dipped)</td>
<td>0</td>
<td>0.52</td>
</tr>
</tbody>
</table>

* Oxytetracycline as contained in Biostat, product of Chas. Pfizer & Co., Brooklyn, New York.
† Chlortetracycline as contained in Aeronize, product of American Cyanamid Chem. Corp., New York City.

Storage temperature 1 C.

All data are the mean values of triplicates showing variations, normally encountered in plate counts.

TABLE 5  
Residual antibiotic concentrations in commercially shucked oyster meats

<table>
<thead>
<tr>
<th>Treatment: Blow Tank 3 Min</th>
<th>Conc</th>
<th>Antibiotic Residues, mg/g</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>ppm</td>
<td>Days in storage at 1 to 4 C</td>
</tr>
<tr>
<td></td>
<td></td>
<td>0</td>
</tr>
<tr>
<td>OTC*</td>
<td>5</td>
<td>4.9</td>
</tr>
<tr>
<td>CTC†</td>
<td>5</td>
<td>2.9</td>
</tr>
<tr>
<td>CTC</td>
<td>15</td>
<td>5.35</td>
</tr>
<tr>
<td>OTC</td>
<td>15</td>
<td>9.0</td>
</tr>
<tr>
<td>Untreated (not dipped)</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

* Internal temperature, 212 F, 12 min cooking time.
† OTC Assay performed by Chas. Pfizer & Co., Brooklyn, New York, F & DA Cup-plate.
† CTC Assay performed by American Cyanamid Chemical Corp., New York City, F & DA Cup-plate.

seen at that time. Because, however, a similar high count was present at the 39th day without the lower pH, it does not appear tenable to do so.

The general shape of these pH curves appears to be similar to that obtained by others (Baldwin et al., 1941; Gardner and Watts, 1956; Hunter and Linden, 1923) but without the organoleptic qualities proposed for segments of the curve. It has been suggested that pH 6.3 to 5.9 indicates high quality oysters while stale to sour oysters are indicated at pH 5.8 to 5.6; below this are the foul to putrid meats. Sensory evaluations by our staff and commercial processors failed to bear this out with the oysters used in this test.

It must be pointed out that at the 17th day, and thereafter, discrepancies as to the sensory qualities of the oyster meats existed between the laboratory staff and the commercial processors. Although no differences, as noted earlier, could be detected between the treated and untreated samples, the staff indicated all oyster samples to be "off" (having a sour tomato juice-like odor) while the processors agreed that they were all "definitely" acceptable.
This discrepancy raises the suspicion that the reported differences in edible storage life and general acceptability of various products may well be due to different standards of evaluation.

It may appear that an unusually long storage period was obtained for the untreated oysters when in the introduction we stated that up to 15 days was their normal storage life. The statement in the introduction refers to "The typically fresh flavor and appearance" of oysters. Many oysters are sold, which people buy, that are not typically fresh, by far. These may have been in storage 30 days. Although this study indicated that both untreated and treated oysters were satisfactory after 50 days, it should be kept in mind that there was a discrepancy between the organoleptic evaluation rendered by the laboratory personnel and the processors. Our people found these same oysters, both treated and untreated, to be "off" by the 17th day, more in line with that normally encountered.

In addition, the unusually long storage period attained may have been due to the type of refrigeration. In our laboratory, mechanical refrigeration maintains the walk-in "box" at approximately constant temperature. A variation of 6 degrees (33 to 39 F) may occur over 7 days. This is quite different from the 40 to 50 F condition generally seen in commercial plants.

Although it has not been reported, it may be that in the lower temperature ranges oysters can maintain an acceptable, though not a typically fresh, condition for an extended period.

Table 5 shows the antibiotic residues present during the storage period and after frying for 12 min to an internal temperature of 100 C.

It is uncertain whether the widely different figures for oxy- and chlorotetacycline at zero days represents true differences in actual tissue penetration or a characteristic attributable to the assay procedures or both.

In either case, the concentrations present at all sampling periods are substantial. The fried oysters, on the other hand, contain substantially smaller amounts.

During the 3 min "blow" period using a chlorotetacycline solution of 15 mcg per ml, the foam, generated by the bubbling action of air-in-water, became green. This suggested that a lesser amount of antibiotic would be available for treatment. Assay, however, showed no untoward loss; in fact, that treatment contained the highest residue.

ACKNOWLEDGMENT

It is a pleasure to thank Mrs. Dorothy Collins for her technical assistance in this study.

* This was as far as our supplies lasted. Indications were that the upper limit was not reached.

SUMMARY

The results herein reported tend to support observations made in our laboratory during the past 2 years that crabmeat of high quality loses this quality during the first 6 to 8 days storage. Crabmeat kept beyond this time lacks the strong typical flavor; a flatness becomes evident though the meat is still quite edible. Spoilage occurs shortly after the "flat" stage, from 10 to 15 days after storage at 1 C.

Chemical treatment with antibiotics as reported here have not been able to extend the storage life presently attainable with untreated meat.

Decreased bacterial counts have been obtained with antibiotic treatments in some instances, but this treatment does not extend the length of fresh storage.

Similarly with oysters, antibiotic treatment has been able to reduce the total viable aerobic population, but the storage life was not increased beyond that of the untreated samples.

From the data obtained, the conclusion seems unavoidable that antibiotics as employed in this study were unable to extend the useful storage life of crab and oyster meats.

ADDITIONAL

Since this work was completed, dips of 120 mcg per ml of oxytetracycline were employed and no increase in storage life was obtained.

REFERENCES


Wrenshall, C. L. 1956 Advances in food technology made possible through the use of antibiotics. Paper Presented at Fourth Annual Antibiotic Symposium, Washington, D. C.