Growth and Survival of *Clostridium perfringens* in Rare Beef Prepared in a Water Bath

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**ABSTRACT**

The low-temperature, long-time cooking of beef, using either a water bath or a conventional oven, resulted in partial inactivation of *Clostridium perfringens* vegetative cells. Beef roasts were cooked in a water bath for process times calculated to inactivate low and high levels of *C. perfringens* vegetative cells. Cooking beef in a water bath to an internal temperature of 60 °C and holding for at least 12 min, as required by the USDA, reduced a population of *C. perfringens* by approximately 3 log cycles. To decrease the risk of subsequent outgrowth of *C. perfringens*, roasts (≤ 1.5 kg) may be subjected to a process calculated for a 12-log reduction in population, which would include holding times of 2.3 h or longer at an internal temperature of 60 °C. Recommendations are given for cooking and cooling roasts to minimize microbiological problems.

Time-temperature relationships during cooking have been recognized as factors influencing the ultimate tenderness and juiciness of meat. Many workers have investigated the effects of low-temperature with long-time (LTLT) cooking processes on the quality of cooked meat. They have reported that, generally, roasts are more tender and cooking losses are decreased when the LTLT cooking process is used. Tender and cooking losses are decreased when the LTLT is exposed to temperatures above 64 °C. Further, they stated that minimal exposure to temperatures near 60 °C may not be sufficient to inactivate a vegetative cell population of *C. perfringens*. Significant increases in yield, tenderness and overall acceptability were observed, as compared to beef cooked in a conventional oven, using a LTLT cooking process.

Since the surface heat transfer coefficient between the cooking medium and the meat is roughly 20 times greater when the product is cooked in water as opposed to air, a water bath method of cooking meats provides the advantage of faster rates of heat penetration and offers the potential for greater microbiological safety. Recent outbreaks of salmonellosis related to consumption of precooked rare roast beef sold in delicatessens (4,5,11) clearly demonstrate the potential microbiological problems associated with meat cookery, especially LTLT cooking processes. Meat is an ideal culture medium for many organisms, including pathogenic bacteria. *Salmonella*, staphylococci, and *Clostridium perfringens* have been responsible for most reported foodborne disease outbreaks. Meats and other products of animal origin are the primary foods that have been associated with these outbreaks (19).

When considering a low-temperature cooking process for beef, *C. perfringens*, an etiologic agent of enteric intoxication, is an organism of primary concern for the following reasons. *C. perfringens* is a common contaminant of raw beef and unlike staphylococci and salmonella, is an obligate anaerobe. The organism grows rapidly at relatively high temperatures and it multiplies exponentially in large masses of foods, particularly after heating has reduced the oxygen content and decreased the number of competing organisms. Heat may also cause germination activation of *C. perfringens* spores at temperatures of 60 - 80 °C (20). Willardsen et al. (21) found that inactivation of *C. perfringens* began at approximately 55 °C. These authors demonstrated, however, that minimal exposure to temperatures near 60 °C may not be sufficient to inactivate a vegetative cell population of *C. perfringens*. Further, they stated that the potential exists for growth during certain LTLT cooking practices. In view of this, *C. perfringens* appeared to be an appropriate organism for assessing the microbiological safety of roasts cooked by a LTLT water bath process.

The objectives of this research were threefold: (a) to study the survival of *C. perfringens* in beef cooked in a water bath vs. beef cooked in a low-temperature air oven, (b) to develop process times necessary for 3- and 10-log reductions of *C. perfringens* in beef, based on integrated

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lethality concepts and (c) to determine the actual growth and survival of *C. perfringens* in beef processed for the times developed in (b) above.

**MATERIALS AND METHODS**

**Source of experimental material**

Forty samples were prepared from contralateral semimembranosus, semitendinosus, biceps femoris, quadriceps femoris and longissimus dorsi muscles removed from both sides of three steer carcasses prepared in the University of Massachusetts Meats Laboratory. Following slaughter, carcasses weighing 220-252 kg, and judged to be equivalent to USDA Good grade in quality, were aged 10-14 days at 2 °C before removal of muscle samples. Although samples used in the various trials varied in shape and weight (0.7-2.0 kg), care was taken to ensure that the roasts within a pair were removed from the same relative locations on contralateral muscles and that they were approximately equal in shape and weight. Paired roasts were placed in polyethylene bags, vacuumized, frozen and stored at -29 °C for 3-5 months until required for experimentation. Two days before initiation of a trial, the roasts were thawed in a refrigerator at 2-4 °C. At the initiation of each trial all roasts had an internal temperature of 2.4 °C.

**Cooking procedures**

To compare the survival of *C. perfringens* in beef cooked in a water bath and beef cooked in a low-temperature air oven, four pairs of samples were used for four replications of the two cooking methods. Right and left roasts from a pair were randomly assigned to each of the two methods. Cooking procedures designed by Buck and coworkers (3) were used. Water bath-cooked roasts were first browned on all sides by rotation for a total of 1 min on the surface of a Teflon-coated griddle with thermocouples, one located in the center of the oven and the other located at the approximate center point of the roast. The bag was sealed under vacuum, and the bagged meat was completely submerged in a water bath (blue M Model MW-1140E-1) operating at 61.1 °C. Water in the bath was constantly agitated by a mechanical stirrer and bath temperature was monitored by a thermocouple. Bagged muscles were removed when their internal temperatures reached 60 °C.

Samples for oven roasting were placed on a wire rack in an open shallow roasting pan, and cooked in a conventional household-type dry air oven set at 94 °C and cycling from 88 to 99 °C. Oven temperature and roast temperature were monitored with thermocouples, one located in the center of the oven, and the other located at the approximate center point of the roast. The roast was removed from the oven when its internal temperature reached 59.5 °C. After removal, roasts reached a final temperature of 60-60.5 °C.

To determine the actual growth and survival of *C. perfringens* in beef processed for predetermined process times, 10 pairs of samples were used in 10 trials. One pair of samples was cooked in the water bath during each trial. During each trial, one bagged roast served as a control. This was removed from the water bath when its internal temperature reached 60.9 °C, the maximum temperature for growth of *C. perfringens*. The other bagged roast was removed after cooking for a calculated process time. When the internal temperature of this roast reached 60 °C, the bath temperature was lowered to 60 °C to maintain that temperature in the meat. Process times were calculated based on the heat penetration characteristics of the roasts and the heat resistance of the experimental contaminant, *C. perfringens* strain S-45.

**Heat penetration determination**

Time-temperature data were plotted on semilogarithmic paper to obtain heating curves for cooking roasts. A semilogarithmic heat penetration curve was plotted for each individual roast. The function, $\ln t$, taken as the time in minutes required for the straight-line portion of the semilogarithmic heat penetration curve to traverse one log cycle, was determined for each roast.

**Process time determination**

Employing the heat penetration parameters and heat resistance data, the mathematical procedure of Stumbo (17) for integrating the sum of all lethal effects in conduction-heating a food during processing was used to evaluate the efficacy of the process by computer analysis. Process times were determined for the following two levels of inactivation: (a) under process = 3-log reduction, and (b) over process = 10-log reduction.

**Experimental contaminant**

*C. perfringens* strain S-45 was obtained from the Department of Health, Education and Welfare, FDA, Cincinnati, Ohio. *C. perfringens* S-45 was originally isolated from dried beef (8), and was chosen as an experimental contaminant because of its characteristic heat resistance.

A stock culture of *C. perfringens* was maintained in cooked meat medium (Difco) stored at 2-4 °C. Vegetative cells used for inoculum to contaminate roasts were grown in fluid thioglycollate medium (FTM) (Difco) at 37 °C for 24 h (maximum stationary phase). The initial population of the inoculum was estimated with a Bausch & Lomb Spectronic 20 colorimeter at a wavelength of 420 nm (from a pre-calibrated optical density-concentration curve) and was in the range 3 x 10^8 to 2 x 10^10 cells per ml.

Roasts were experimentally contaminated with *C. perfringens* by injecting 1.0 ml of inoculum into the roast near the center point. After injection, roasts were immediately cooked.

Four uninoculated roasts were also examined for inherent *C. perfringens* contamination. The roasts were cut in half and one half was sampled raw and the other portion was cooked to 60 °C in the 61.1 °C water bath before sampling.

**Thermal resistance**

Thermal resistance of viable cells of *C. perfringens* S-45 was determined at 60 and 65.5 °C in FTM, using the three-necked flask method of Levine et al. (13). Cells to be tested were grown in FTM at 37 °C for 8 h (maximum stationary phase). After sterilization, the assembly was placed in a hot water bath operating at 60.6 °C to determine $D_{60}$ values and at 61.1 °C to determine $D_{61.1}$ values. A 4.0-ml portion of the 8-h-old culture was pipetted into the flask containing 400 ml of preheated FTM that was continuously stirred. Ten-ml samples were removed from the flask at selected time intervals and 1.0 ml was pipetted into each of 10 screw capped test tubes containing 10.0 ml of FTM. Tubes were then incubated for 24 h at 37 °C and the Most Probable Number (MPN) value for survivors was determined, using the modified Halvorsen-Ziegler equation described by Stumbo (17).

Also, the thermal resistance of *C. perfringens* S-45 was determined in beef roasts, using integrated lethality of heat. Seven roasts were each injected with a known number of organisms and cooked to an internal temperature of 60 °C in 61.1 °C water bath. Using heat penetration data from each roast and assuming the logarithmic order of death, $D_{60}$ values were calculated from the initial number of *C. perfringens* cells, plus a growth factor of 1 log cycle, minus the number of *C. perfringens* cells surviving after heating to 60 °C.

**Sampling procedures**

Core samples were taken with a sterile 5.1-cm diameter cylindrical stainless steel coring tool. A center core sample was taken from each roast from the area where inoculum injections were made, parallel to the muscle fibers, and from one side of the roast to the other. The efficiency of the sampling procedure was ascertained by injecting two roasts from the semimembranosus with crystal violet. One roast was injected with 1.0 ml and the other with 2.0 ml of crystal violet, using the same procedure used for injecting *C. perfringens*. After injection, roasts were bagged and cooked in a water bath (61.1 °C). When their internal temperature reached 60 °C, they were removed from the bath and were then sliced transversely down the center lines both longitudinally and laterally, through the area where the injection was made. The distance travelled by the stain was measured both parallel and perpendicular to the muscle fibers.

The distance travelled by crystal violet injected into the roasts was primarily dependent on the volume of stain injected and on the orientation of the muscle fibers. The 2-ml portion of stain travelled 3.5 cm parallel with the fibers and 1.5 cm across the fibers. The 1-ml
portion of stain migrated 1.8 cm parallel with the fibers and 0.3 cm across the fibers. These results suggest that the larger the volume injected into the roast, the farther the inoculum will migrate along the length of the muscle fibers and across the fibers.

Assuming that a bacterial suspension injected into a roast will behave in a manner similar to crystal violet, the sampling procedure employed in this study was considered efficient. The 5.1-cm diameter core, taken parallel to the muscle fibers from the area where inoculum injections were made, would recover either a 2 ml or a 1 ml of inoculum. In this study, a 1 ml inoculum was used for inoculations.

Microbiological analyses

Cores, ranging in weight from 70 to 160 g, were placed in sterile blender jars and homogenized for 2 min in 300 ml of 0.1% sterile peptone solution. A 3-tube MPN technique was used to enumerate the surviving vegetative cells of C. perfringens. Serial dilutions of the homogenized cores and of the drippings recovered from the cooking bag and roasting pan were made in 0.1% peptone solution. One-ml portions of the appropriate dilutions were pipetted into screw capped tubes containing 10 ml of FTM. The tubes were incubated at 37 C for at least 24 h. One-ml portions of all positive tubes were transferred to another tube of FTM. After a 24-h incubation period at 37 C, plates of laboratory-prepared Tryptose Sulfite Cycloserine (TSC) agar (U5) were streaked for isolation of typical black colonies. The plates were incubated at 37 C for 24 h in anaerobic jars (BBL GasPak System). The TSC agar was freshly prepared before each experiment to avoid hydrogen peroxide production (9). Positive tubes were also confirmed by stabbing buffered motility nitrate and lactose gelatin media (6) with portions of isolated colonies from the TSC agar and incubating 24 h at 37 C.

Statistical analyses

The paired t-test was employed with analysis of variance to determine the effects of cooking methods on the variables studied.

RESULTS AND DISCUSSION

Thermal resistance

The D-values determined in FTM for C. perfringens S-45 at 60 and 65.6 C were 5.4 and 0.65 min, respectively. The z-value was calculated to be 6.09 C degrees. The D60 value determined in the beef roasts was 14.5 min (formulæ and definitions are included in the Appendix). The thermal resistance of C. perfringens S-45 in beef roasts was increased nearly three times over the resistance in FTM. It is well established that environmental factors active during heating of bacterial suspension exert widely variable influences on the thermal resistance of the bacteria (16). A similar effect was noticed by Willardsen and coworkers (21) when growth curves of C. perfringens in FTM and autoclaved ground beef were compared; autoclaved ground beef had a definite effect in reducing lag and generation time. The marked effect of chemical environment of bacteria on their thermal resistance at the time heat is applied makes it imperative that whenever possible in making thermal resistance determinations, the bacteria should be suspended in the food for which a sterilization process is to be established. In this study the D60 value determined in the beef roasts and the z-value calculated from the flask method were used to calculate process times for the roasts.

Inherent Clostridium perfringens contamination

In the uninoculated meat sampled, both raw and cooked to 60 C, C. perfringens vegetative cells were not detected down to the 0.01 per gram level. Assuming the roasts used in this study were relatively homogeneous as to their microbial flora, these results suggest that inherent C. perfringens contamination was negligible, if present at all.

Survival of C. perfringens in oven-vs. bath-cooked beef

The log reductions of C. perfringens vegetative cells in roasts cooked to 60 C in a 94-C conventional oven and in a 61.1-C water bath are shown in Table 1. The log reductions in the oven-roasted beef averaged 3.24. Log reductions in the bath-cooked beef averaged slightly higher (3.31), but the difference was not significant.

<table>
<thead>
<tr>
<th>Source</th>
<th>Bath</th>
<th>Oven</th>
<th>Log reduction</th>
<th>Time</th>
<th>Log reduction</th>
<th>Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>biceps femoris</td>
<td>2.40</td>
<td>45</td>
<td>2.80</td>
<td>75</td>
<td></td>
<td></td>
</tr>
<tr>
<td>biceps femoris</td>
<td>2.71</td>
<td>45</td>
<td>2.02</td>
<td>67</td>
<td></td>
<td></td>
</tr>
<tr>
<td>quadriceps femoris</td>
<td>3.04</td>
<td>75</td>
<td>3.23</td>
<td>120</td>
<td></td>
<td></td>
</tr>
<tr>
<td>seminembranosus</td>
<td>5.10</td>
<td>75</td>
<td>4.89</td>
<td>105</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Although there was no significant difference in the log reduction of C. perfringens vegetative cells in beef cooked using the two methods, a greater reduction of cells in the water bath-cooked beef might be expected. Figure 1, typical cooking-temperature profiles for contralateral 1.5-kg roasts from the quadriceps femoris cooked in a 94-C oven and in a 61.1-C water bath, clearly shows the initial increased rate of heat penetration obtained with the water bath. Because of the higher rates of heat penetration, the bath-cooked beef had significantly less time (P < 0.01) in the temperature range of 20 - 50 C, the growth zone for C. perfringens, than did the oven-roasted beef. The time, in minutes, that the interior of the beef samples were in this temperature range is shown in Table 1. The interior of bath-cooked roasts required an average of 60 min to traverse this zone, compared to an average of 92 min for the oven-roasted beef. Since the bath-cooked beef had less time in this temperature range, there was less time for multiplication of C. perfringens cells. Based on this fact, fewer viable cells might be expected to survive in the bath-cooked beef than in the oven roasts when their internal temperatures reached 50 C.

The greater rate of heat penetration in the bath-cooked beef also resulted in a decrease in the time that its internal temperature was in the range of 50 - 60 C, the lethal zone for C. perfringens vegetative cells. This decrease in time in the lethal zone, along with the decrease in time in the growth zone would account for the similar log reductions of C. perfringens cells observed in the bath-cooked and oven-roasted beef.

Another explanation for the lower than expected lethal effect of the bath process is offered in the findings of Willardsen et al. (21). When ground beef was heated at
constantly rising temperatures, they found that higher
temperatures were necessary to observe the cessation of
growth and initiation of inactivation of \( C.\ perfringens \)
cells at more rapid rising rates. Since the internal
temperature of the bath-cooked beef rose more rapidly
than that of the oven-cooked beef, cessation of growth
and subsequent inactivation may have occurred at
a higher temperature in the bath-cooked beef. Regardless
of the cooking method, the results suggest that
minimal exposure to temperatures near 60°C is sufficient
to reduce a vegetative cell population of \( C.\ perfringens \)
S-45 by an average of 3 log cycles when using roasts of
the size used in this study.

\( C.\ perfringens \) vegetative cells were not detected down
to the 0.01 per gram level in the drippings recovered from
the cooking bag and roasting pan. These results suggest
that any inoculum which may have migrated along the
site of injection to the surface could not survive the
higher temperature at the surface of the oven roast and
the moist environment at the surface of the bath roast.

Survival of \( C.\ perfringens \) in beef processed in water bath

Figure 2 illustrates the effect of time and temperature
on the growth and survival of \( C.\ perfringens \) in a typical
roast from the semitendinosus cooked in a 61.1°C water
bath. The log count of cells per sample was plotted at
initiation of heating, after the internal temperature of
the sample reached 48.9°C and when the sample was
removed from the bath. Sixty minutes was the average
time for roasts used in this study to be in the growth zone
for \( C.\ perfringens \). Growth of \( C.\ perfringens \) during this
period was observed in roasts removed from the bath
when their internal temperature reached 48.9°C; the
vegetative cell count increased by an average of 1 log
cycle. The increase in cell count calculated from the
control roast of each pair was added to the initial cell
count when calculating log reductions in the bath-cooked
roasts.

Values for \( t_{50} \) varied with each roast, and ranged from
46.5 to 104 min for roasts ranging from 0.68 to 2.04 kg.
The initial temperature, \( T_{50} \), used in process time
determinations was 48.9°C, the temperature at which
lethality begins for \( C.\ perfringens \).

The process times calculated for a 3-log reduction in
\( C.\ perfringens \) S-45 population and the actual log
reductions observed in roasts processed for these times
are shown in Table 2. The average log reduction in
population was 2.86. These results suggest that for this
particular organism the process times calculated for a
process with a low margin of safety (3-log reduction) were
reliable and accurate. The average length of time
samples were held at 60°C during this process was
9.5 min. This time is similar to the 12 min at 60°C
required by the USDA regulations for cooked beef (1).
These results suggest that cooking beef in the manner
required by the USDA (to protect consumers from
salmonellae food poisoning) would also cause a 3-log
reduction of \( C.\ perfringens \) vegetative cells.

The process times calculated for a 10-log reduction in
\( C.\ perfringens \) S-45 vegetative cells are shown in Table 3.
The log reduction in population actually observed
averaged 7.9. The observed log reduction, therefore, was
approximately 2 log cycles less than the predicted
reduction of 10 log cycles.

Holding times at 60°C for these roasts averaged 96 min
and the average number of survivors in the roast after
processing was \( 4 \times 10^{-3} \) per gram. This number of
survivors would not present a problem from a public
health standpoint; however, more complete inactivation
of a population is desirable to prevent subsequent
outgrowth. Roasts with an initial contamination level of
\( 10^6 \) per gram and subjected to a process time calculated
for a 12-log reduction, should be rendered virtually free.
of *C. perfringens* viable cells (<10^6 per gram). Using heat penetration data from roasts processed for a 10-log reduction, process times were calculated for a 12-log reduction. These times are shown in Table 3. Holding times at 60°C averaged 126 min. These results suggest that for roasts of this size (<1.5 kg) and with a *C. perfringens* contamination level of 10^6 per gram, cooking in a 61.1°C water bath and subsequent holding of the roasts at 60°C for at least 2.3 h (140 min) would provide for a survival probability of not greater than 10^{-6}, which would give a high margin of safety beyond the minimal treatment needed for inactivating the cells. It should be kept in mind that the process time equivalent to 2.3 h at 60°C would apply only for the initial concentration of 10^6 per gram or less, and the probability of survivors from contamination above that initial level would increase logarithmically with log increases in initial population. Since Toumy and Lechnir (18) found that the color of cooked beef depended on the temperature rather that time of exposure and that beef samples still appeared rare in color after 7 h at 60°C, holding for 2.3 h or longer at 60°C should not affect the rare color of beef.

**CONCLUSIONS**

The recently developed water bath process is microbiologically as safe as a low-temperature over-roasting method which employs conventional equipment. Roasts heated for a process time calculated for low level inactivation of *C. perfringens* vegetative cells, had holding times at 60°C similar to the processing time of 12 min required by the USDA regulations for cooked beef (1). Therefore cooking beef in water bath to an internal temperature of 60°C and holding for at least 12 min will eliminate the public health hazard presented by salmonellae and will reduce a population of *C. perfringens* by approximately 3 log cycles.

This study illustrates a very important point when developing thermal process times; that is, regardless of the method used for the calculation of the thermal process times, these times always should be verified by actual tests with food. After being subjected to a process time calculated for a 10-log reduction, roasts contained a number of survivors that was far less than that which would be considered a public health hazard. To decrease the risk of subsequent outgrowth, water bath-cooked roasts may be subjected to a process calculated for a 12-log reduction in population without apparent loss of quality.

If food service administrators attempt to minimize costs by holding roasts at 60°C for only the required 12 min (I), they must be prepared to monitor proper handling and cooling. Because of the possible *C. perfringens* outgrowth during improper cooling and handling, especially from heat-activated spores, the following recommendations are made to foodservice operators employing the water bath process for cooking rare beef.

(a) The internal temperature of beef should be held at 60°C in the bath for at least 12 min in accordance with USDA regulations.

(b) After the minimum 12 min of processing time, the roasts should be held at 60°C in the bath until ready to be served.

(c) If cooked beef is to be kept or served cold, rapid cooling should take place so that the internal temperature of the beef traverses the 'danger zone', 20 - 50°C, quickly. The water bath process lends itself to rapid cooling since the bagged beef may be transferred

**TABLE 2. Process times calculated for a 3-log reduction in *C. perfringens* S-45 inoculated into beef cooked in a water bath (61.1°C).**

<table>
<thead>
<tr>
<th>Source</th>
<th>Weight (kg)</th>
<th>f_h (min)</th>
<th>Process time (B)³</th>
<th>Holding time at 60°C (min)</th>
<th>Actual log reduction</th>
</tr>
</thead>
<tbody>
<tr>
<td>longissimus dorsi</td>
<td>0.79</td>
<td>57.0</td>
<td>85</td>
<td>15</td>
<td>4.07</td>
</tr>
<tr>
<td>longissimus dorsi</td>
<td>1.13</td>
<td>50.0</td>
<td>81</td>
<td>10</td>
<td>3.21</td>
</tr>
<tr>
<td>semimembranosus</td>
<td>0.68</td>
<td>51.5</td>
<td>80</td>
<td>8</td>
<td>1.56</td>
</tr>
<tr>
<td>semimembranosus</td>
<td>2.04</td>
<td>104.0</td>
<td>120</td>
<td>5</td>
<td>3.18</td>
</tr>
<tr>
<td>biceps femoris</td>
<td>0.90</td>
<td>63.0</td>
<td>90</td>
<td>7</td>
<td>2.40</td>
</tr>
<tr>
<td>biceps femoris</td>
<td>0.77</td>
<td>46.5</td>
<td>78</td>
<td>12</td>
<td>2.71</td>
</tr>
</tbody>
</table>

³Time beef spent in bath after reaching internal temperature of 48.9°C until removed from bath.

**TABLE 3. Process times calculated for a 10- and 12-log reduction in *C. perfringens* S-45 inoculated into beef cooked in a water bath (61.1°C).**

<table>
<thead>
<tr>
<th>Source</th>
<th>Weight (kg)</th>
<th>f_h (min)</th>
<th>Process time (B)³</th>
<th>Holding time at 60°C (min)</th>
<th>Actual log reduction</th>
</tr>
</thead>
<tbody>
<tr>
<td>semitendinosus</td>
<td>0.84</td>
<td>54</td>
<td>191</td>
<td>220</td>
<td>105</td>
</tr>
<tr>
<td>semitendinosus</td>
<td>0.79</td>
<td>57</td>
<td>194</td>
<td>224</td>
<td>90</td>
</tr>
<tr>
<td>quadriceps femoris</td>
<td>1.45</td>
<td>84</td>
<td>217</td>
<td>248</td>
<td>98</td>
</tr>
<tr>
<td>semimembranosus</td>
<td>0.90</td>
<td>63</td>
<td>195</td>
<td>228</td>
<td>90</td>
</tr>
</tbody>
</table>

³Time beef spent in bath after reaching internal temperature of 48.9°C until removed from bath.
directly to an ice bath.

(d) Adequate monitoring of procedures for refrigerating should be carried out so that a temperature no higher that 7 C is maintained in the beef.

Serious consideration should be given to the use of a water bath for preparing rare beef. The microbiological safety of the process is as great as that of a low-temperature oven-roast and the bath process has the added advantages of increased yield, tenderness and overall acceptability of product. If a water bath process is developed for home use, similar precautions should be taken regarding cooking and handling.

ACKNOWLEDGMENTS

This work was supported in part by the Massachusetts Agricultural Experiment Station.

APPENDIX

Formulae Used for D-Value Calculation

From Integrated Lethality of Heat

\[ B = \frac{f_h}{U} \left( \log \frac{l_p}{l_f} - \log g_c \right) \]

\[ f_h/U : g_c \ [\text{from table (7)}] \]

\[ U = F \times F_p \]

\[ F = D \left( \log a + 1 - \log b \right) \]

\[ D = \left( \log a + 1 - \log b \right) / F \]

Definitions and Symbols

a. The initial and final number, respectively, of viable cells of an organism per unit volume of an inoculated medium just prior to applying lethal heat and after heat treatment at a constant temperature for some designated period of time.

B. The thermal processing time without correcting for the time required to bring the sample to processing temperature.

D. The time, at any specified lethal temperature, required to inactivate a population of viable cells of an organism by one log cycle.

F. The time, in minutes, of heating at a constant lethal temperature.

F_p. The time at any other temperature equivalent to 1 min of processing time at 60 C.

f_h. The time, in minutes, required for the straight line portion of the semilog heating curve to traverse one log cycle.

g_c. The difference between the water bath temperature and the maximum sample temperature at the point of concern.

I_p. The difference between the water bath temperature and the temperature at which lethality began with regard to C. perfringens (48.9 C).

I_0. A heating or cooling lag factor (2.00).

U. The time, in minutes, required at water bath temperature to accomplish the same amount of bacterial inactivation equivalent to the F value of the process.

REFERENCES


