CHANGES IN BACTERIAL COUNTS DURING STORAGE OF HOT BONED BOXED BEEF TRIMMINGS

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CHAPTER I

INTRODUCTION

In the traditional way of processing, sides of beef are chilled for 24 - 48 hours after slaughter and then fabricated into primals and/or subprimal cuts. In a later modification of processing, wholesale cuts are vacuum packaged, and boxed for further distribution. This modification has been called "boxed beef" (Henrickson and Ferguson, 1981).

In recent years, boning of the beef carcass while the meat is still warm, or hot boning, has been shown to have several practical and economical advantages. Removal of excess fat and bone results in considerable conservation of energy represented by savings over 50% in refrigeration energy and nearly 80% in cooler space requirements, as well as, a reduction in transportation and labor costs (Henrickson, 1975; Henrickson and Ferguson, 1981).

Vacuum packaging of hot boned beef, especially boxed, has been found to have problems such as short lasting vacuums, high leaker rate and poor color quality. In addition, the lack of a mechanism to quality or yield grade hot beef carcasses along with economic reasons concerning refitting and/or construction of proper plants seem to be among the main technical disadvantages preventing industry from moving to hot boning (Anonymous, 1981).

On the other hand, several factors such as high temperature, high oxidation-reduction potential (Eh), and high pH of pre-rigor muscle

(Forrest et al., 1975; Rey et al., 1978) along with the high surface moisture of warm tissues favor the growth of microorganisms on hot boned meat.

The advent of a relatively new technology, electrical stimulation of carcasses, makes hot boning more feasible for commercial adoption. The risk of cold and thaw shortening is overcome since rigor mortis develops sooner, when an electrical current is passed through the carcass thus permitting earlier chilling of the hot boned meat (Davey et al., 1976; Gilbert et al., 1977). Electrical stimulation has been expected to discourage bacterial growth on meat since it increases the rate of glycogen and ATP depletion, lowering the pH to about 5.9; causes changes in Eh; and triggers release of some proteolytic enzymes in the sarcoplasm (Sorinmade et al., 1978; Kotula, 1980; Mrigadat et al., 1980; Dutson et al., 1980).

The microbiology of electrically stimulated hot boned beef has been studied by several researchers mostly in the United States, Great Britain, and New Zealand. However, these studies have dealt mainly with bacterial growth in hot meat under vacuum-package storage.

The objective of this study was to ascertain changes in total aerobic, anaerobic, and psychrotrophic bacterial counts at three different positions inside boxes of electically stimulated hot boned beef, chilled aerobically for different periods of time.

CHAPTER II

REVIEW OF LITERATURE

Microbiology of Hot Boned Beef

The economic advantage offered by hot boning of meat due to more effective energy utilization and shorter processing time is probably the major factor responsible for stimulating recent interest in this processing method. However, the high temperature and surface moisture of hot boned meat has been associated with microbial problems which formed the objective of recent studies.

Schmidt and Gilbert (1970) used six Angus steers and bulls of different maturities to determine if organoleptically and microbiologically acceptable beef could be produced by pre-rigor excision followed by rigor setting and short term aging of bovine muscle. Pre-rigor boneless wholesale cuts were removed from one side of each carcass within two hours of slaughter, placed into gas impermeable bags and stored at 15°C for 48 hours. The other side, used as control, was chilled at 9°C for 24 hours, after which the same wholesale cuts were fabricated. Total bacterial numbers were determined on the pre-rigor wholesale cuts immediately after excision (0 hours), and after 24 and 48 hours of storage; on the control sides before and after chilling; and on the control wholesale cuts. Since mean bacterial numbers on muscle surface were within the range of 10^2 to $10^5/cm^2$, it was concluded that microbiological spoilage was satisfactorily controlled during the prolonged storage at

15°C, and that meat of an acceptable microbiological standard can be produced by pre-rigor excision of muscle from beef carcasses.

As part of a study made to evaluate the feasibility of hot boning beef carcasses and to determine the minimum conditioning time before boning could be initiated, Falk (1974) compared psychrotrophic and mesophilic bacterial counts on ground lean trim obtained from both hot boned and cold boned (1.1°C for 48 hours) beef sides conditioned for three, five, and seven hours at 16°C. Psychrotrophic counts were less than 10^3/g, with no significant difference ($P>0.05$) between the mean log numbers of bacteria per gram of hot versus cold boned trim at any of the three conditioning periods. Mesophilic counts ranged from 10^3 to 10^4/g and were not significantly different at the three-hour and seven-hour postmortem periods. However, they were significantly different ($P<0.05$) after five hours of conditioning at 16°C.

Kastner et al. (1976) compared total aerobic mesophilic and psychrotrophic bacterial counts from beef sides held at 16°C for six, eight, and ten hours postmortem with the corresponding bacterial counts from sides held at 2°C for the same periods. Samples were taken from the flank and plate regions of each side by removing the portion of muscle adjacent to the interior perimeter of sanitized waxed paper templates previously affixed to the sampling zones. Low bacterial numbers, ranging from 10^2 to $10^3/cm^2$ were detected and no statistical differences ($P>0.10$) were observed in either mesophilic or psychrotrophic mean log counts between the hot boning (16°C) and conventional (2°C) treatments at any postmortem sampling time.

Cuthbertson (1977) reported that total viable counts from hot boned vacuum packaged roasts initially held for 24 hours at 10°C were up to

1000 times higher than cold boned roasts after storage of both (hot and cold boned roasts) at 1°C for three weeks. However, counts were similar after eight weeks of storage, when maximum numbers were reached.

Emswiler and Kotula (1979) compared the bacteriological quality and shelf life of ground beef prepared from hot (two hours postmortem) and cold (24 hours at 3°C) boned beef sides at the time of preparation (0 hours) and at three-day intervals up to 45 days of storage at 0°C. The hot ground beef, which was chilled with CO_2 snow during preparation, as well as the cold ground beef were packaged in oxygen impermeable polyethylene bags to make five-lb chub packs. Aerobic plate counts at 5, 20 and 35°C, and most probable numbers of coliforms and Escherichia coli were determined. Based on the fact that coliforms and E. coli numbers were very low, and that aerobic plate counts at 5, 20 and 35°C in ground beef from hot boned sides were either significantly lower or not significantly different from the corresponding counts in ground beef from cold boned sides, these authors claimed that preparation of ground beef from hot boned carcasses as a method for energy conservation in the meat industry was feasible.

Mesophilic and psychrotrophic bacterial populations and occurrence of indicator organisms and potential pathogens were monitored on hot boned and conventionally processed beef by Fung et al. (1980). Meat samples aseptically removed from the plate region of beef sides at two hours postmortem (hot boned) or after chilling for 48 hours at 2.2°C were bacteriologically analyzed immediately after their removal and after 14 days in a vacuum bag at the center of a box filled with meat masses from other parts of the carcasses. Low initial bacterial numbers were observed in samples from both hot boned and conventionally processed

sides. However, after 14 days of vacuum packaged boxed storage, hot boned samples showed higher mesophilic and psychrotrophic counts than the conventionally treated ones. Also, mesophilic counts were higher than psychrotrophic counts in either hot or conventionally processed sides. No Salmonella were recovered and some hot boned samples had coliforms, Clostridium perfringens, coagulase positive Staphylococcus aureus and fecal streptococci; however, they were found in numbers low enough not to be considered a health hazard. These authors also observed that the hot boned meat chilled more slowly than meat handled in the conventional manner. This was attributed to vacuum packaging and boxing soon after cutting.

In further experiments, Fung et al. (1981) studied the effect of chilling to 21°C in 3, 5, 9, or 12 hours on the development of spoilage, indicator, and pathogenic organisms in vacuum packaged hot boned boxed meat samples stored for 14 to 21 days at 2.2°C and then displayed at the same temperature for three days under natural fluorescent lighting. They found that when the temperature was lowered to 21°C in 3 to 9 hours, hot boned meat was acceptable in color, odor and bacterial quality after 14 days of storage and three days of display and recommended adoption of this rate of chilling combined with additional continuous chilling to below 10°C in 24 hours.

McMillin et al. (1981) determined bacterial numbers in frozen patties prepared from ground hot boned beef held at 10°C for one, two, four, or eight hours after slaughter. No statistical difference ($P>0.05$) was found in numbers of coliforms, presumptive coagulase positive staphylococci, psychrotrophs, or mesophiles, at any of the holding times between the hot boned patties and control patties prepared from ground

conventionally chilled beef. Changes in temperature and pH of the ground hot boned meat during the holding periods, or a lethal effect from freezing were assumed by these authors to be responsible for the absence of differences between the microbial quality of patties prepared from either hot or chilled beef.

Hot boned meat containing 0, 3, or 5 percent added salt and stored in polyethylene bags at either 2°C for 7, 14, or 21 days or -10°C for 7, 14, or 28 days was utilized by Reagan et al. (1981) in weiner preparation. Bacterial enumerations were carried out after completion of storage, before weiner manufacture. Neither salt level nor time of storage were found to exert a significant effect on microbial numbers during storage at 2°C. Log_{10} of bacterial counts/g ranged from 4.8 in meat with 3% salt to 4.5 in meat with 5% salt, and from 4.0 in meat stored 14 days to 5.2 in meat stored 21 days, with 4.7 in meat stored for 7 days. Unfortunately bacterial counts on meat without any salt added (0%) were not estimated because of undesirable physico-chemical characteristics in the meat after seven days storage. Microbial levels were low for all storage periods in the hot boned meat stored at -10°C.

Microbiology of Electrically Stimulated Hot Boned Beef

One of the earlier studies reporting some microbial aspects of electrically stimulated hot boned meat was done by Gilbert and Davey (1976). The right sides of six Angus steer carcasses were stimulated for two minutes with an electrical current of 3600 V while the left sides acted as unstimulated controls. Stimulated sides were hot boned after five hours and hot cuts were halved, wrapped in heat shrinkable plastic bags and

frozen at -18°C or stored at 10°C for 72 hours before freezing at -18°C. The same treatment was applied to the unstimulated control sides after 24 hours storage at 2°C. Bacterial analysis of samples taken at different stages during processing of either the stimulated or unstimulated cuts showed low total bacterial counts ($<10^2/cm^2$) before boning. The numbers increased to $10^4/cm^2$ through contamination from hands, equipment and air during the boning process. A similar trend was observed in counts of psychrotrophs and Microbacterium thermosphactum, which were in smaller numbers. However, since the counts did not increase further during the 72-hour aging period it was concluded that bacteriological condition of the meat was not changed by electrical stimulation followed by early boning.

As a continuation of the work done by Gilbert and Davey (1976), Gilbert et al. (1977) compared total bacterial counts and numbers of psychrotrophs and M. thermosphactum on hot boned cuts from electrically stimulated beef sides with the similar cuts from conventionally chilled unstimulated sides. The cuts, individually packaged in gas impermeable films, were placed in boxes and stored at 5°C for 46 hours or at 10°C for 65 hours. The $\log_{10}$ of bacterial counts/cm^2 on the stimulated and unstimulated cuts at boning were low; however, they increased appreciably (to about 4.0) on the unstimulated cuts but only slightly (to 2.0-3.0) on the stimulated meat. This meant that bacterial growth was limited by the shorter processing time in the hot boned meat, which made a more wholesome product than the conventional procedure.

Raccach and Henrickson (1978) studied the effect of electrical stimulation (300 V for 15 minutes) and hot boning of beef sides on the storage stability and bacteriological quality of ground beef stored at

5±1°C in polystyrene foam trays wrapped with polyvinyl chloride film. Ground meat from the unstimulated opposite side in each carcass was used as control. Spoilage was found to occur after 4 to 5 days in the control samples while samples from the electrically stimulated sides spoiled after 7 to 8 days storage, when bacterial numbers reached about 8 X 10^8/g in both cases. Non-pigmented Pseudomonas species predominated as part of the spoilage flora and no pathogenic bacteria were found, except Staphylococcus aureus which was detected in very low numbers (10/g).

Kotula (1980) investigated the microbiological condition of a) primal cuts from electrically stimulated hot boned beef sides, b) primal cuts from electrically stimulated conventionally chilled sides, and c) ground meat from hot boned beef sides. Aerobic plate counts (APC) at 5, 20, and 35°C determined immediately after boning and after 20 days of storage at 2°C showed that electrical stimulation alone did not influence microbial counts on the beef primals, but hot boning in conjunction with electrical stimulation resulted in significant and important higher levels of bacteria in some primals. Nevertheless, the APC at 5°C (psychrotropic bacteria) was lower in the hot boned than in the cold boned primals after storage. Ground hot boned beef had greater but not significantly higher bacterial numbers than ground conventionally chilled beef.

In order to assess the general level of contamination during boning and after storage in carton boxes for 5 or 21 days at 1°C, Taylor et al. (1981) determined total viable bacterial counts on surface samples of primal cuts obtained from beef sides that were either electrically stimulated (700 V) and hot boned; hot boned without previous electrical

stimulation; or held at 15°C for seven hours and chilled at 0-1°C before boning at 48 hours post mortem. In addition, presumptive coliforms, E. coli, Enterobacteriaceae and fecal streptococci were enumerated and internal muscle samples were examined for C. perfringens. Statistical analysis showed no significant differences in initial numbers of any bacterial group among the different boning procedures. These results agree with those obtained by Gilbert and Davey (1976) and Gilbert et al. (1977). Also, there was no difference in bacterial counts after storage, except for fecal streptococci which were slightly more numerous in some of the hot boned cuts. C. perfringens was found more in hot than in cold boned cuts, but in very low numbers that do not represent a health hazard.

Kotula (1981), in a review on the microbiology of hot boned and electrostimulated meat, stated that the rapid decrease in pH and other yet to be characterized biochemical changes that might occur as a result of electrical stimulation do not exert an economically important impact on microbial populations on meat. Despite that he concluded that hot boning of carcasses of any species need not cause inordinate increases of any group of microorganisms on or in the resultant meat or meat products, and that the reported microbiological data do not preclude use of electrical stimulation coupled to hot boning.

CHAPTER III

MATERIALS AND METHODS

Electrical Stimulation

Fifteen commercial Hereford steers of approximately 900 lb (410 kg) live weight, slaughtered at weekly intervals, were used. Each animal was electrically stimulated immediately after exsanguination (within five minutes after stunning) by using an "Electro-Stim" electrical stimulator (Double J. Products, Wichita, Kansas) which supplied an electrical alternating current of 48 volts and less than 0.5 amperes, with a pulsation time of one second, during a 90 second period, through a spring loaded clamp attached to the nostril and positive ground probes inserted into the hocks of the animal (Figure 1).

Preparation of Hot Boned Meat Samples for Boxed Storage

After dressing, splitting, and washing of each carcass, one of the sides was randomly designated to be hot boned. Hot boning was done within three hours after slaughtering. The semitendinosus muscle was excised and placed on a sterilized tray. Using a sterile knife the muscle was sectioned transversally so that 12 slices approximately two cm in thickness was obtained (Figure 2). Three of the meat slices were randomly designated for analysis to determine the initial bacterial load on the muscle surface. The remaining nine meat slices were randomly

Figure 1. Electrical Stimulation

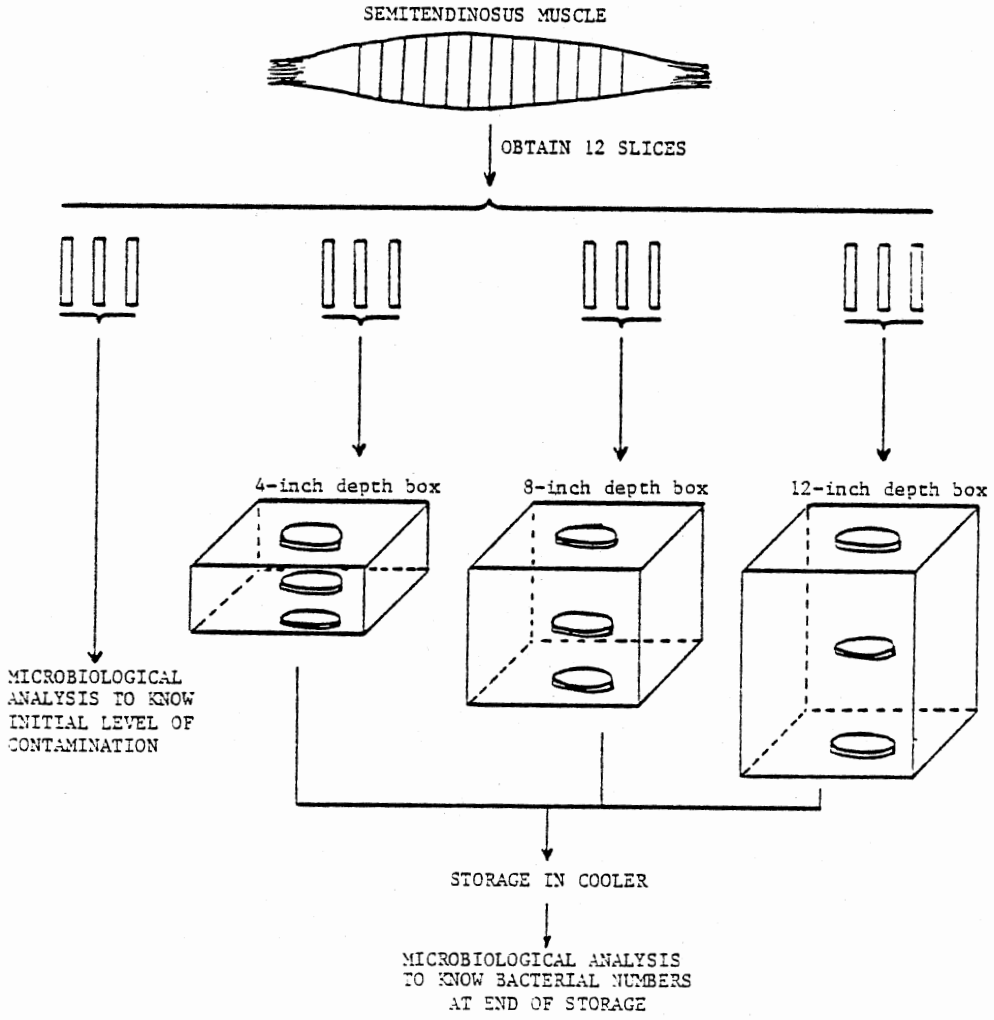

Figure 2. Schematic Drawing of Meat Slices Utilized for Microbiological Analysis and Their Position Within the Boxes During Storage

assigned to three different positions (top, middle, and bottom) within each of three single-wall cardboard boxes (Container Service Corporation, Oklahoma City, Oklahoma) differing only in depth (12X12X4, 12X12X8, and 12X12x12 inches, respectively).

The boxes were filled with 20 (4-inch depth box), 40 (8-inch depth box), and 60 (12-inch depth box) pounds of meat trimmings obtained from the side forequarter. Before filling, the interior of each box was covered with Cryovac clear film in order to avoid adherence of the meat to the interior walls of the boxes. The "bottom," "middle," and "top" positions were located when the boxes were filled with 0, 10, and 20 pounds of meat trimmings, respectively, for the 4-inch depth boxes; 0, 20, and 40 pounds of meat trimmings for the 8-inch depth boxes; and 0, 30, and 60 pounds, respectively, for the 12-inch depth boxes.

Storage of Boxed Hot Meat

The boxes containing the meat were sealed and stored in a refrigerated cooler (36°F, 2.2°C) at the Oklahoma State University Meat Laboratory. The boxes were sufficiently separated from each other, on a stainless steel rack, in order to obtain uniform distribution of air around them (Figure 3).

Boxes of hot boned beef prepared from groups of three randomly selected beef sides were assigned to each of the following storage periods: 0, 24, 48, 72, and 96 hours; i.e., boxes from three sides of beef were stored for 0 hours, boxes from three other beef sides were stored during 24 hours, and so on.

The temperature at each position within each box was determined at different intervals of time (0, 6, 12, 24, 48, 72, and 96 hours) during

Figure 3. Cooler Storage and Temperature Recording of Hot Boned Boxed Beef Trimmings

each corresponding period of storage. This was accomplished by inserting a thermocouple close to each meat slice. The thermocouples were attached to a Honeywell multipoint temperature recorder. A free termocouple, in contact with the air, was used to record the environmental temperature.

Preparation of Meat Samples for Microbiological Analysis

After each respective storage period, the boxes were removed from the cooler, and the meat slices were aseptically removed from the boxes. For the zero-hour storage period the meat slices in each box were left in contact with the rest of the meat trimmings for five minutes before being removed from the boxes for analyses. This contact procedure provided a means for determining if the contact with the meat trimmings in the boxes influenced the initial bacterial load on the individual samples. Sterile medical gloves (Bard-Parker) were worn in handling each meat slice during its removal from the boxes. The slices were individually placed in sterile wide-mouth Mason jars (one-quart capacity) and their weight was determined. The jars were then placed in ice to prevent or delay additional bacterial growth on the meat slices, and transported to the laboratory, where microbiological analysis was immediately initiated.

Microbiological Analysis

A pre-measured volume of sterile 0.1% peptone (Difco Laboratories) water equal to twice the weight of the meat cut was delivered into each jar. The jar was then shaken, making 25 back-and-forth movements of

about one foot in seven seconds to permit removal of bacteria from the meat surface (Speck, 1976). Each ml of "rinse" thus prepared represented 0.5 g of sample.

Serial dilutions, as needed, were prepared from each jar using sterile 99-ml dilution bottles containing 0.1% peptone water as diluent. Preparation of the dilutions and further platting were made following the specifications indicated in the Compendium of Methods for the Microbiological Examination of Foods (Speck, 1976).

The total aerobic, psychrotrophic, and anaerobic bacterial counts were determined for each of the meat slices, in duplicate 100X15 mm sterile disposable petri dishes (Curtin Matheson Scientific, Inc.) by the pour plate technique using Plate Count Agar (Difco Laboratories).

Total aerobic and psychrotrophic counts were obtained after incubation of the plates at 32°C for 48 hours in a Freas 815 low temperature incubator (GCA/Precision Scientific), and at 5°C for seven days in a cooler at the Meat Laboratory, respectively. The anaerobic count was determined by incubating the plates in anaerobic Gas-Pak jars at 32°C for 48 hours. After the respective incubation time the colonies were counted using a Spencer colony counter and recorded as indicated in the Compendium of Methods for the Microbiological Examination of Foods (Speck, 1976). The average number of colonies on the selected duplicate plates was multiplied by the appropriate dilution factor, multiplied by two, and referred to as count per gram (count/g). The bacterial counts thus obtained were converted to common logarithms (Log_{10}) before statistical analysis was carried out. Bacterial counts on the meat slices utilized for determination of the initial level of contamination were not considered further in this study since the counts were similar to

those obtained at the zero-hour storage period.

Statistical Analysis

The experiment was conducted as a split-split plot experiment, with whole unit treatments (five different storage periods) in a completely randomized design. Each carcass is considered a whole unit with three box depths (split unit treatments) within each carcass and three positions (split-split treatments) within each box. Duncan's (1955) multiple range test was employed to evaluate differences among means.

CHAPTER IV

RESULTS

Tables I, II, and III in Appendix A show the actual numbers of total aerobic, anaerobic, and psychrotrophic bacteria per gram of meat, respectively, determined throughout the experiment. Tables IV, V, and VI in Appendix B show the analysis of variance carried out on the common logarithms (Log_{10}) of the respective bacterial numbers. Appendix C contains tables with the mean Log_{10} bacterial numbers derived from the statistical analyses, as well as mean temperatures at different intervals of time during boxed storage of the meat.

Change in Total Aerobic Bacterial Numbers

As shown in Table IV, a significant difference in mean Log_{10} total aerobic bacteria was detected among storage periods ($P = 0.02$), box depths ($P = 0.00$), and positions ($P = 0.00$). Mean Log_{10} bacterial counts at 0, 24, 48, 72, and 96 hours storage periods were 3.04, 4.25, 5.33, 5.64, and 5.97, respectively (Table VII). Duncan's mean separation test showed no statistical difference ($P>0.05$) among bacterial counts at 48, 72, and 96 hours storage periods, but they were significantly greater ($P<0.05$) than counts at 0 hours; while no separation from either grouping could be exerted on counts at the 24 hour storage period.

Mean Log_{10} total aerobic bacteria in the meat at 4, 8, and 12-inch depth boxes were 4.23, 4.83, and 5.48, respectively (Table VII).

Duncan's test showed these means were significantly different ($P<0.05$) from each other. Although the variation among storage periods and among box depths were significantly different ($P = 0.02$ and $P = 0.00$, respectively), the interaction between storage period and box depth was not significant ($P = 0.14$), which meant that the total aerobic bacterial count increased in a similar manner within the boxes as time passed.

At the top, middle, and bottom positions within the box the mean Log_{10} total aerobic counts were 5.27, 4.90, and 4.37, respectively (Tables X and XI), and the Duncan's test showed that they were significantly different ($P<0.05$) from each other. In addition, mean Log_{10} total aerobic bacterial numbers at each position were significantly affected by the depth of the box ($P = 0.00$) and by the storage period ($P = 0.00$). These means were higher at the top position and lower at the bottom position in either the 4, 8, or 12-inch depth boxes (Figure 4, Table X). Similar results were obtained at all storage periods except at 0 hours, where the greater mean count was obtained at the bottom position while the smaller mean was obtained at the top position (Figure 5, Table XI). The interaction of position, box depth, and storage period did not have a significant influence ($P = 0.24$) on total aerobic counts.

Change in Anaerobic Bacterial Numbers

Table V reveals a significant difference in mean Log_{10} anaerobic bacterial count at the different storage periods ($P = 0.01$), box depths ($P = 0.00$), and positions ($P = 0.00$).

Numbers of anaerobic bacteria increased with increasing storage period. Mean Log_{10} bacterial counts at 0, 24, 48, 72, and 96 hours

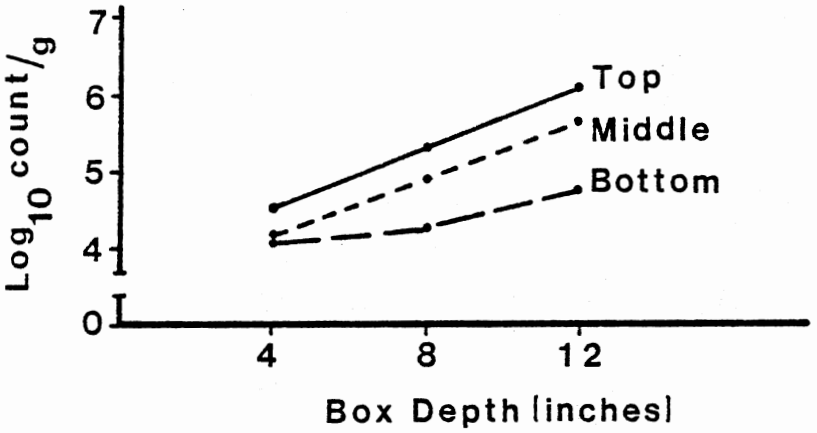

Figure 4. Influence of Box Depth on Total Aerobic Bacterial Counts at Different Positions in Boxes of Hot Boned Beef Trimmings (Each Point Represents the Average Log_{10} Count/g of Fifteen Determinations)

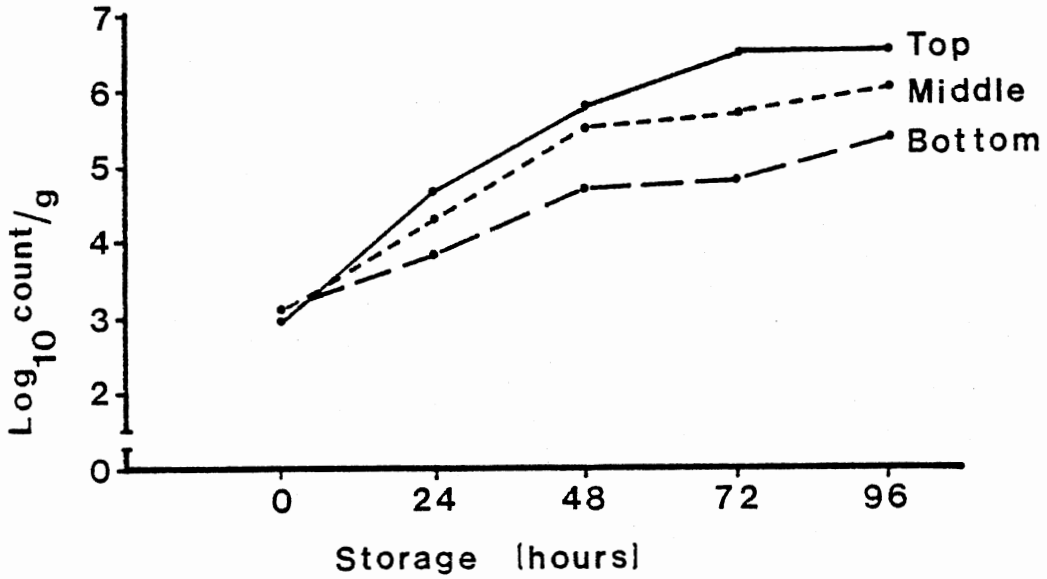

Figure 5. Influence of Storage on Total Aerobic Bacterial Counts at Different Positions in Boxes of Hot Boned Beef Trimmings (Each Point Represents the Average Log_{10} Count/g of Nine Determinations)

storage periods were 2.43, 3.93, 4.82, 4.93, and 5.11, respectively (Table VIII). Duncan's test showed that means at 0 hours were significantly lower ($P<0.05$) than means at 24, 48, 72, and 96 hours, which showed no significant difference ($P>0.05$) among them.

Increasingly box depth was related to an increase in numbers of anaerobic bacteria. Mean Log_{10} anaerobic counts were 3.46, 4.31, and 4.97 in the 4, 8, and 12-inch depth boxes, respectively (Table VIII); and they were significantly different ($P<0.05$) from each other according to Duncan's test for mean separation. The interaction of box depth and storage period did not have a significant influence ($P = 0.23$) on anaerobic counts.

The number of anaerobic bacteria showed significant differences among positions in the boxes ($P = 0.00$). The middle position had the higher mean Log_{10} bacterial count (4.50), followed by the top and bottom positions (4.22 and 4.01, respectively) (Tables XII and XIII). Duncan's test showed that these means were significantly different from each other ($P<0.05$). The interaction between position and box depth produced a significant influence ($P = 0.00$) in anaerobic counts. Mean Log_{10} anaerobic counts in the 8 and 12-inch depth boxes were higher at the middle than at the top or bottom positions; however, an opposite effect existed in the 4-inch depth boxes, in which the mean bacterial numbers were higher at the bottom, followed by the top and middle positions (Figure 6, Table XII). Also significant was the effect produced on anaerobic counts by the interaction between position and storage period ($P = 0.00$). As shown in Figure 7 and Table XIII, at 0 hours the top and middle positions had higher mean Log_{10} anaerobic bacterial numbers than the bottom position, but the difference among them was relatively small.

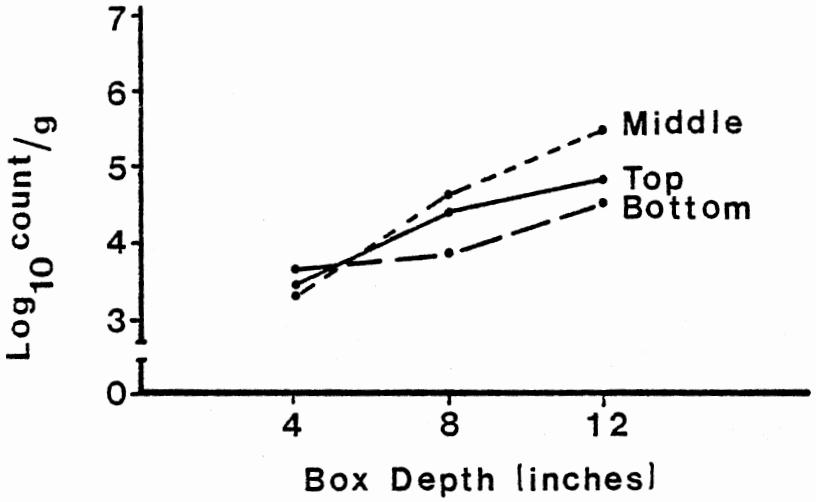

Figure 6. Influence of Box Depth on Anaerobic Bacterial Counts at Different Positions in Boxes of Hot Boned Beef Trimmings (Each Point Represents the Average Log_{10} Count/g of Fifteen Determinations)

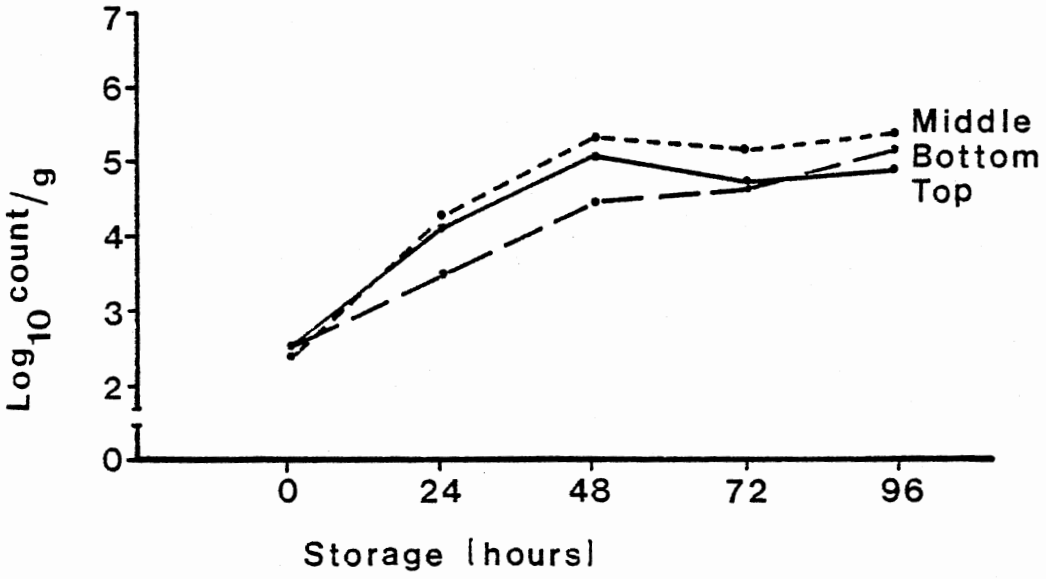

Figure 7. Influence of Storage on Anaerobic Bacterial Counts at Different Positions in Boxes of Hot Boned Beef Trimmings (Each Point Represents the Average Log_{10} Count/g of Nine Determinations)

At 24, 48, and 72 hours the counts were higher at the middle than at the bottom or top positions, being the counts at the top higher than at the bottom. At 96 hours the anaerobic counts were still higher at the middle position, but now the counts at the bottom were higher than counts at the top. The three-way interaction between position, box depth, and storage period did not significantly affect (P = 0.46) the anaerobic counts.

Change in Psychrotrophic Bacterial Numbers

Psychrotrophic bacterial counts were significantly different among storage periods (P = 0.01), box depths (P = 0.00), and positions in the box (P = 0.00) (Table VI).

Mean Log_{10} psychrotrophic counts were 1.97, 3.98, 5.17, 5.60, and 5.94 at 0, 24, 48, 72, and 96 hours storage periods, respectively (Table IX). Mean counts at 0 hour were significantly lower ($P<0.05$) than means at 48, 72, and 96 hours, which were nonsignificantly different ($P>0.05$) among them. No separation could be established by Duncan's test in the mean count at 24 hour storage period.

As with total aerobic and anaerobic counts, the number of psychrotrophic bacteria increased with increasing box depth. Mean Log_{10} psychrotrophic counts were significantly different ($P<0.05$) in the 4, 8, and 12-inch depth boxes, with mean values of 3.93, 4.57, and 5.10, respectively (Table IX). The two-way interaction of box depth and storage period did not have a significant influence on the psychrotrophic counts (P = 0.28).

Growth of psychrotrophic bacteria was higher at the top than at the middle or bottom positions in the boxes. Significant differences

($P<0.05$) were shown by Duncan's test among the mean Log_{10} psychrotrophic counts, which had values of 5.16, 4.48, and 3.96, respectively (Tables XIV and XV). There was a significant two-way interaction of position and box depth ($P = 0.00$) on the number of psychrotrophs. The top position had higher number of psychrotrophic bacteria, followed by the middle and bottom positions in either the 4, 8, and 12-inch depth boxes, respectively (Figure 8, Table XIV). The interaction between position and storage period also had a significant effect on the number of psychrotrophic bacteria ($P = 0.00$). At the 0 hour storage period higher psychrotrophic bacterial numbers were obtained in meat at the bottom position, followed by the top and middle positions, but they were not considerably different. However, during all the subsequent storage periods the top position showed higher bacterial numbers, while lower counts were detected at the bottom position (Figure 9, Table XV). The three-way interaction of position, box depth, and storage period, as occurred with total aerobic and anaerobic counts, did not significantly influence psychrotrophic bacterial numbers ($P = 0.05$).

Temperature Variations

The mean air and meat temperature change at different positions within boxes of hot boned meat are presented in Figures 10, 11, and 12, and in Table XVI. It can be seen that, as expected, meat in the 4-inch depth boxes had a faster decline in temperature than meat in the 8-inch or 12-inch depth boxes. The average initial temperature of the meat (at 0 hours) ranged between 78.2 and 83.8°F. After 24 hours of storage the meat temperature in the 4-inch depth boxes was close to the air temperature (43.5°F). At 24 hours, meat in the 8-inch and 12-inch depth boxes

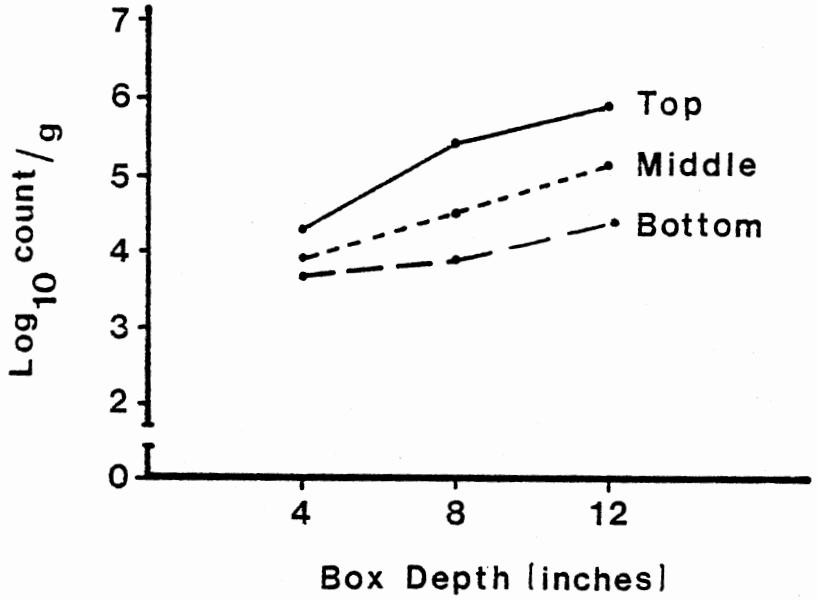

Figure 8. Influence of Box Depth on Psychrotrophic Bacterial Counts at Different Positions in Boxes of Hot Boned Beef Trimmings (Each Point Represents the Average Log_{10} Count/g of Fifteen Determinations)

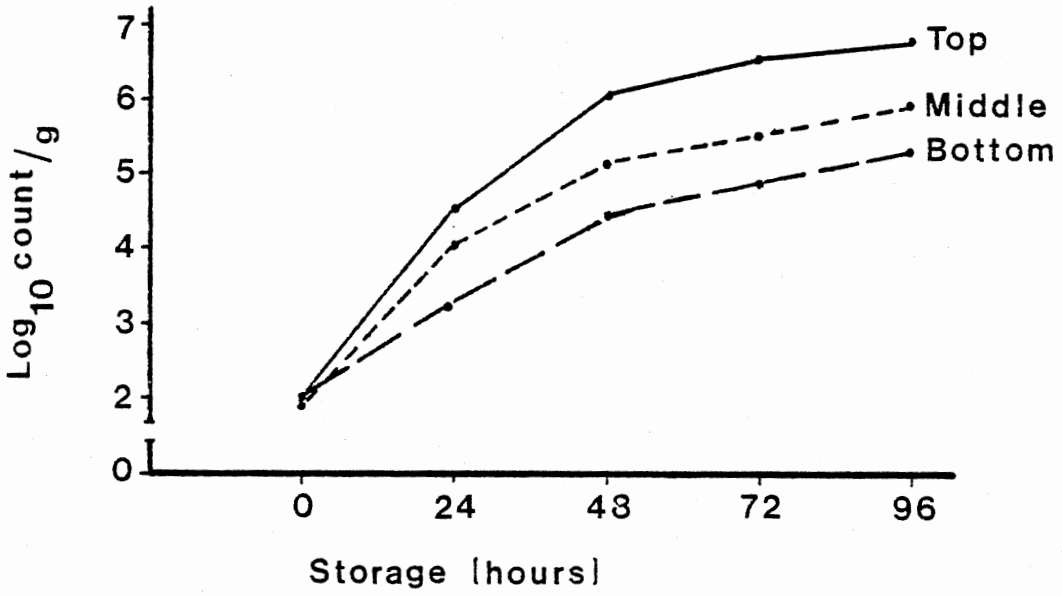

Figure 9. Influence of Storage on Psychrotrophic Bacterial Counts at Different Positions in Boxes of Hot Boned Beef Trimmings (Each Point Represents the Average Log_{10} Count/g of Nine Determinations)

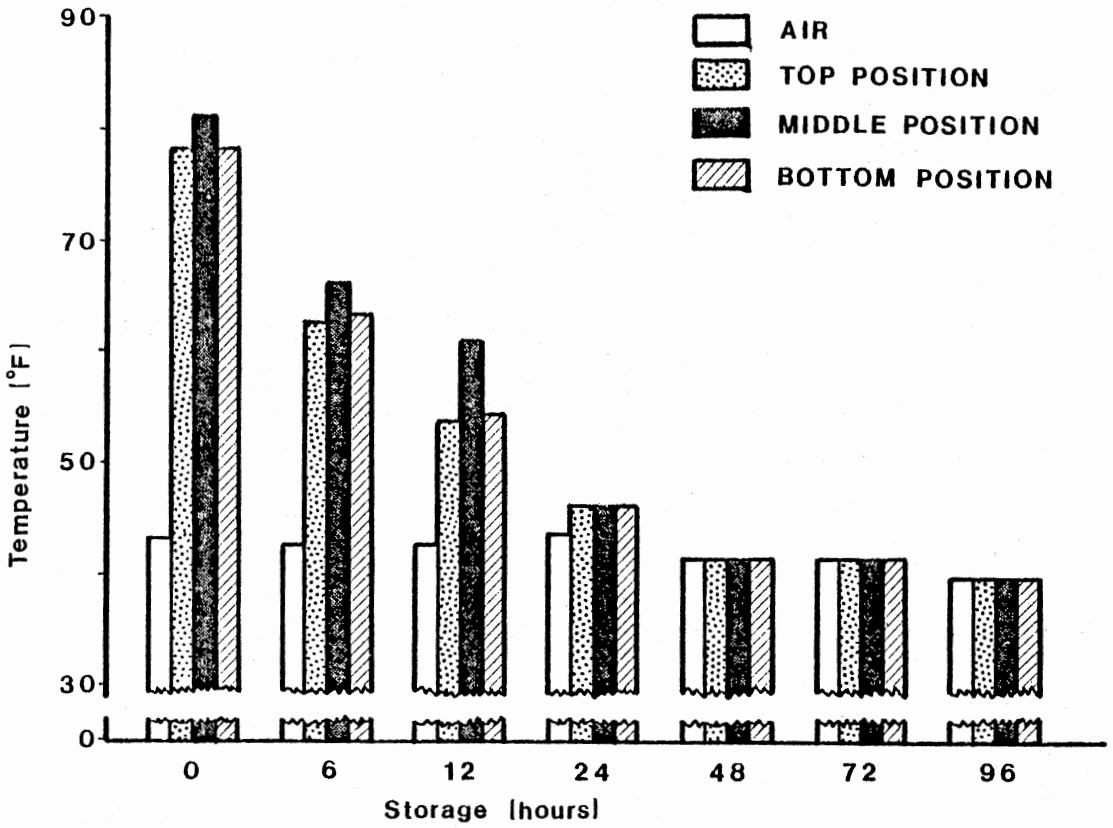

Figure 10. Average Temperature Change During Storage of Hot Boned Beef Trimmings in 4-inch Depth Boxes

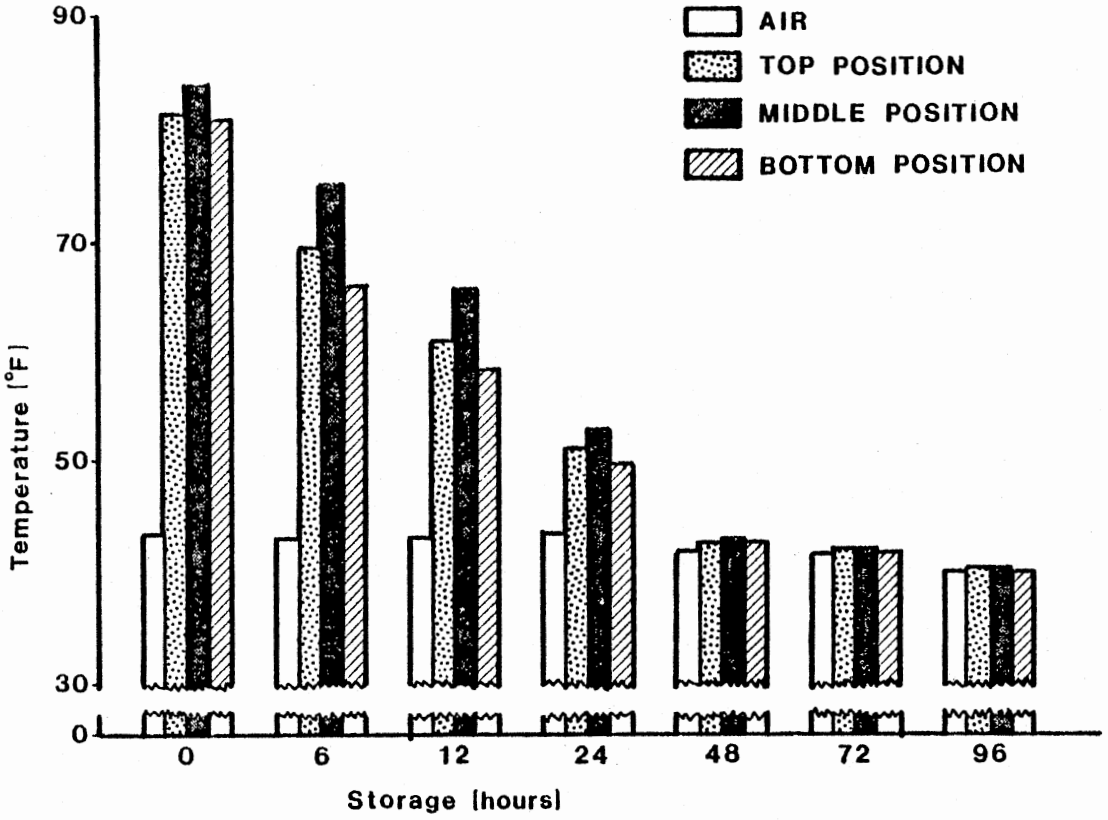

Figure 11. Average Temperature Change During Storage of Hot Boned Beef Trimmings in 8-inch Depth Boxes

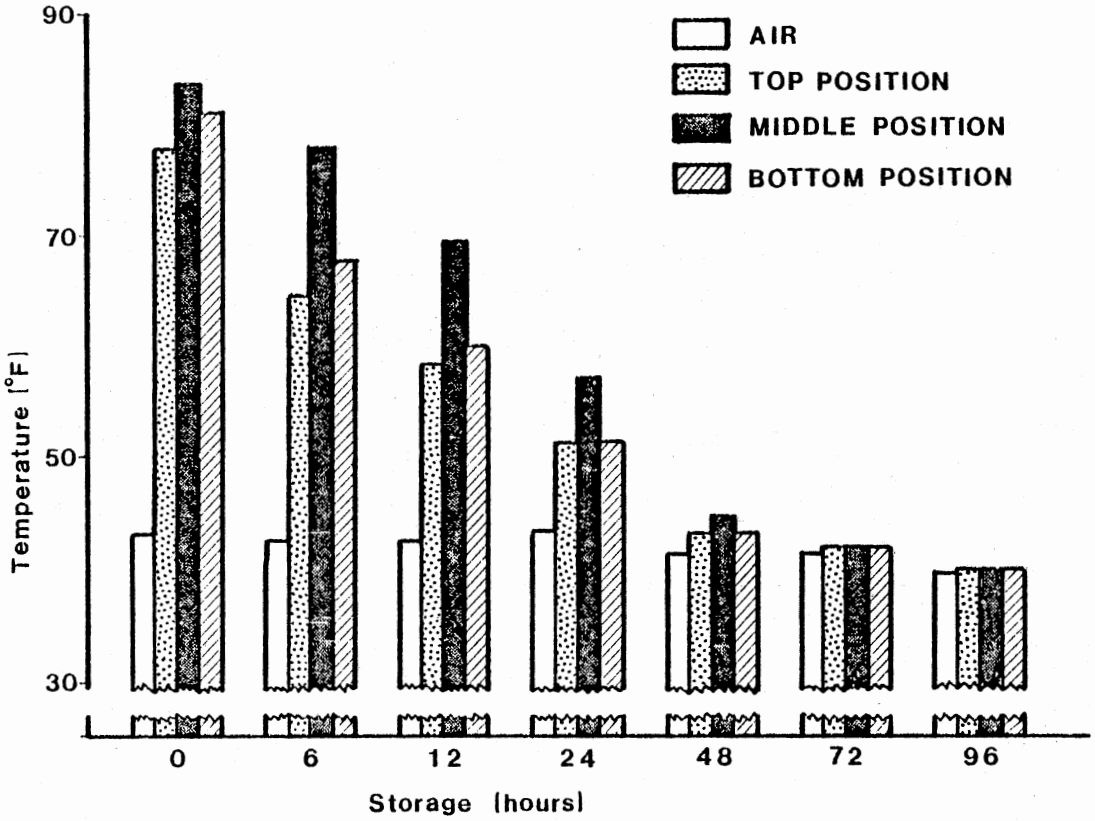

Figure 12. Average Temperature Change During Storage of Hot Boned Beef Trimmings in 12-inch Depth Boxes

still had temperatures between 50 and 60°F. At 48 hours of storage, the meat temperature in the 4-inch depth boxes had already equalized the air temperature. By that time the 8-inch and 12-inch depth boxes had meat temperatures close to the air temperature, the 8-inch depth boxes showing lower temperature values than the 12-inch depth boxes. Regardless of box depth, the middle position temperature declined slower than either the top or bottom position temperatures, and this condition was more noticeable with increasing box depth.

CHAPTER V

DISCUSSION

Raw beef is increasingly being distributed in a box. Although currently about 50% of all choice beef is marketed in the form of boxed, vacuum packaged primal cuts (Ayres et al., 1980), beef trimmings used for preparation of ground beef or hamburger are also being distributed boxed.

The feasibility of boxed beef has been enhanced through a combination of electrical stimulation followed by hot boning of the beef carcass. While electrical stimulation speeds up the onset of rigor mortis (Carse, 1973; Chrystall and Hagyard, 1976; Davey et al., 1976) permitting hot boning and rapid chilling with reduced risk of cold shortening (Gilbert and Davey, 1976; Gilbert et al., 1977); hot boning favors savings in energy (Henrickson, 1975). Electrical stimulation was included in the present study since it will likely be used when hot boning is adopted by industry.

Total aerobic, as well as anaerobic and psychrotrophic bacteria increased in numbers as time passed. This result seems to be obvious for at least two reasons: 1) conditions within the boxes were suitable for bacterial growth, and 2) meat temperatures at the beginning of storage were high, and remained relatively high during the first 12 hours of storage.

Temperature is critical in determining both the rate and the total

amount of bacterial growth that can take place on meat substrates (Price and Schweigert, 1978). This explains the finding of significantly increased growth in total aerobic, anaerobic and psychrotrophic bacteria with increased box depth. In the 4-inch depth boxes bacterial numbers were lower than in the 8- or 12-inch depth boxes because the rate of temperature decline was faster, thus allowing a more effective control of bacterial growth.

Since the rate of chilling was slower at the center of the boxes (middle position), it seemed logical that at this point more opportunity for bacterial multiplication existed (Taylor et al., 1981). Nevertheless, total aerobic and psychrotrophic bacteria were more abundant at the top position in all box depths and at all storage periods beyond 0 hours. Probably at 0 hours bacteria had not yet grown and increased in numbers. Packaging alters the bacterial metabolism in different ways (Ingram, 1962), and even though the most important single factor governing microbial growth is temperature, other factors are interrelated and their individual importance varies with the particular circumstances being considered (Lawrie, 1979). Frazier and Westhoff (1978) indicated that the oxygen or partial pressure of oxygen and the oxidation-reduction potential (Eh) about a food influences the type of organisms which will grow and hence the changes produced in the food. Moreover, presence of oxygen contributes to maintaining Eh at a high level, thus determining the growth of surface spoilage organisms in meat (Lawrie, 1979); and the maximum cell density of a bacterial population may be determined by the rate at which oxygen becomes available to the cells (Gill and Newton, 1977). Therefore, it is likely that under the conditions of the present study the aerobic environment favored a higher

oxygen tension at the top position, and was responsible for such a significant effect upon bacterial growth. These results are different from those of Fung et al. (1981), who showed higher numbers of mesophilic and psychrotrophic bacteria on meat at the center of hot boned beef boxes, where the rate of chilling had been slower. However, these authors dealt with meat samples packaged under vacuum, which creates a different environment which in turn influences the development of bacteria in a different manner (Gill, 1980).

The internal atmosphere within a box of meat aerobically stored would be such that, as previously indicated, the availability of oxygen at the top, although reduced, is suitable to maintain growth of aerobic bacteria. Deeper in the box such oxygen availability may still be more reduced and may provide partial anaerobic conditions that would favor the growth of facultative anaerobes and/or microaerophilic bacteria. This may explain the higher anaerobic bacterial counts found at the middle position in the 8-inch and 12-inch depth boxes and at all storage periods, except at 0 hours. However, this consideration does not rule out the effect of temperature, which causes important modifications in bacterial flora under anaerobic conditions (Gill, 1980). Thus, the lower anaerobic count determined at the middle position in the 4-inch depth boxes may have been a consequence of the faster meat chilling rate.

Fung et al. (1981) recommended chilling hot boned meat to 21^{o}C (69.8^{o}F) within 3-9 hours in order to obtain a microbiologically acceptable product after 14 days of storage. However, this recommendation is valid for vacuum packaged boxed beef but not necessarily for hot boned meat boxed aerobically as in the present study. The shelf life of meat is increased in vacuum packages because carbon dioxide produced from

muscle respiration accumulates and inhibits some bacterial growth (Ingram, 1962). This phenomenon does not occur in hot boned meat stored aerobically, where aerobic bacteria have more opportunity to thrive.

Even though the average $\log_{10}$ of bacterial numbers was as high as 6.57/g at the top position of the 12-inch depth boxes after 96 hours of storage, and the $\log_{10}$ of some individual samples was as high as 8.23/g, no apparent spoilage was observed in the meat throughout this experiment. While some authors have indicated that bacterial spoilage in meat occurs when the $\log_{10}$ of bacteria/g or cm^2 is 7.00 (Ayres et al., 1980; Fung et al., 1981), others have reported spoilage when the $\log_{10}$ of bacterial numbers is 8.48-8.60/g or cm^2 (Dainty et al., 1975; Forrest et al., 1975). In addition, it is important to note that Hansen (1960), cited by Lawrie (1979), is of the opinion that the interaction of atmosphere and microorganisms within packs tends to upset the usual correlation between spoilage and bacterial count, and that spoilage becomes evident only after the number of microorganisms has been maximal for some time.

There is no doubt that having lower and more constant temperature a better control over bacterial growth can be achieved in boxed beef. However, the increasing cost of energy makes it necessary to determine the most appropriate temperature for storage and distribution of this product, taking into consideration that a constant temperature is not normally maintained under commercial conditions. In addition, the present study suggests that the effect of factors other than temperature (i.e., oxygen availability) may be playing an important role influencing the types and amount of bacteria when hot boned meat is boxed aerobically. Hence, a better understanding of the action of these factors is advisable.

CHAPTER VI

SUMMARY AND CONCLUSIONS

Fifteen commercial Hereford steers, slaughtered at weekly intervals, were used to determine bacterial changes in hot boned boxed beef. Each carcass was electrically stimulated and one side randomly assigned to be hot boned within three hours postmortem. The semitendinosus muscle was removed and transversally cut in slices that were assigned to the top, middle and bottom positions in three boxes differing only in depth. Box dimensions were 12X12X12, 12X12X8, and 12X12X4 inches, respectively. After being filled with hot boned trimmings from the side forequarter, the boxes were closed and stored in a cooler at 36°F (2.2°C). Boxes from groups of three beef sides were assigned at random to each of the following storage periods: 0, 24, 48, 72, and 96 hours. Changes in the meat temperature during storage were determined by inserting a thermocouple close to the slice of meat at each position within the boxes. After the corresponding storage period, total aerobic, anaerobic, and psychrotrophic bacterial numbers on each meat slice were determined and converted to $\log_{10}$ count/g for statistical analyses.

Total aerobic, as well as anaerobic and psychrotrophic bacteria increased significantly as both storage period and box depth increased. These results may have been produced by the warm temperature of the meat during the first hours of storage, the favorable conditions for bacterial growth within the boxes, and the slower decline in meat temperature

associated with increased box depth.

Higher numbers of total aerobic and psychrotrophic bacteria were found at the top position than at the other positions in all box sizes and at all storage periods, except at 0 hours; whereas anaerobic bacteria were in higher numbers at the middle position in the 8-inch and 12-inch depth boxes and at all storage periods, except at 0 hours. These findings seemed to be the effect of higher oxygen availability at the top position within the boxes. In the 4-inch depth boxes, anaerobic bacteria were in lower numbers at the middle position, probably because of the faster rate of chilling. Bacterial counts at the 0-hour storage period were inconsistent with counts at subsequent storage periods apparently because bacteria had not yet grown and thus increased in number.

Spoilage of the hot boned meat was not observed in any of the boxes after 96 hours of storage, despite the fact that some bacterial counts were relatively high after 48 hours. Therefore, before any recommendation is given, determination of the most appropriate temperature for storage and a better understanding of the influence of other factors, like oxygen availability, on the type and growth of bacteria associated with hot boned aerobically stored boxed beef is suggested. This may represent considerable energy savings for the industry, and a better quality product for the consumer.

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APPENDIXES

APPENDIX A

BACTERIAL COUNTS

TABLE I

TOTAL AEROBIC BACTERIAL COUNT PER GRAM OF MEAT DETERMINED AT THREE POSITIONS IN BOXES OF HOT BONED BEEF TRIMMINGS DURING STORAGE

Box Depth (inches)	Position	Storage (hours)				
		0	24	48	72	96
4	Top	6.4×10^{2}	2.0×10^{3}	2.6×10^{6}	2.8×10^{4}	1.4×10^{4}
		2.8×10^{2}	2.4×10^{4}	2.3×10^{3}	1.1×10^{7}	3.0×10^{7}
		9.1×10^{2}	2.0×10^{3}	2.6×10^{4}	1.6×10^{4}	1.7×10^{6}
	Middle	1.6×10^{3}	9.2×10^{2}	3.0×10^{6}	1.1×10^{4}	7.2×10^{3}
		2.1×10^{2}	4.3×10^{4}	1.9×10^{3}	4.3×10^{5}	5.8×10^{5}
		1.5×10^{3}	4.2×10^{3}	1.8×10^{4}	1.8×10^{4}	1.7×10^{6}
	Bottom	8.6×10^{2}	7.6×10^{2}	7.8×10^{5}	2.0×10^{4}	3.4×10^{3}
		6.4×10^{2}	2.3×10^{4}	2.2×10^{3}	3.8×10^{5}	5.5×10^{5}
		1.8×10^{3}	3.3×10^{3}	2.0×10^{4}	4.5×10^{3}	1.4×10^{5}
8	Top	7.6×10^{3}	1.0×10^{4}	4.6×10^{7}	7.4×10^{5}	1.3×10^{4}
		2.8×10^{2}	4.6×10^{5}	1.1×10^{5}	8.0×10^{7}	3.5×10^{6}
		1.7×10^{3}	2.8×10^{4}	8.3×10^{5}	4.5×10^{6}	1.3×10^{8}
	Middle	9.0×10^{2}	2.2×10^{6}	5.8×10^{6}	5.8×10^{4}	2.8×10^{4}
		9.3×10^{2}	8.2×10^{3}	5.8×10^{3}	1.6×10^{7}	1.2×10^{7}
		1.1×10^{3}	1.4×10^{5}	3.1×10^{5}	1.6×10^{5}	1.5×10^{7}
	Bottom	5.4×10^{2}	2.4×10^{3}	7.3×10^{5}	6.8×10^{3}	2.2×10^{4}
		7.5×10^{2}	3.0×10^{4}	1.7×10^{3}	8.2×10^{5}	2.3×10^{6}
		1.6×10^{3}	2.8×10^{3}	1.8×10^{4}	1.0×10^{4}	5.5×10^{5}
12	Top	1.3×10^{3}	1.6×10^{5}	4.7×10^{7}	9.8×10^{5}	2.8×10^{5}
		2.8×10^{2}	1.4×10^{6}	9.5×10^{5}	7.3×10^{7}	1.6×10^{8}
		1.8×10^{3}	6.1×10^{5}	4.2×10^{7}	8.7×10^{7}	1.7×10^{8}
	Middle	9.4×10^{2}	1.4×10^{5}	3.4×10^{7}	3.0×10^{5}	1.1×10^{6}
		4.3×10^{2}	2.2×10^{5}	1.5×10^{5}	9.2×10^{6}	3.3×10^{6}
		6.0×10^{3}	1.5×10^{5}	5.0×10^{6}	4.0×10^{7}	1.1×10^{7}
	Bottom	1.7×10^{3}	1.0×10^{4}	9.9×10^{5}	3.2×10^{4}	1.7×10^{5}
		1.1×10^{3}	2.0×10^{4}	7.2×10^{3}	9.6×10^{5}	3.2×10^{6}
		6.7×10^{3}	9.9×10^{3}	5.1×10^{5}	3.3×10^{5}	8.3×10^{5}

TABLE II

ANAEROBIC BACTERIAL COUNT PER GRAM OF MEAT DETERMINED AT THREE POSITIONS IN BOXES OF HOT BONED BEEF TRIMMINGS DURING STORAGE

Box Depth (inches)	Position	Storage (hours) 0	24	48	72	96
4	Top	1.1×10^{2}	5.8×10^{2}	6.6×10^{5}	6.8×10^{3}	2.2×10^{2}
		2.8×10^{2}	2.5×10^{3}	7.7×10^{2}	4.0×10^{4}	2.2×10^{5}
		2.1×10^{2}	7.8×10^{2}	1.3×10^{3}	1.1×10^{3}	3.0×10^{4}
	Middle	1.0×10^{2}	4.6×10^{2}	9.8×10^{5}	3.4×10^{3}	3.0×10^{2}
		2.6×10^{2}	1.1×10^{4}	6.1×10^{2}	9.5×10^{3}	2.1×10^{4}
		2.7×10^{2}	1.2×10^{3}	1.7×10^{3}	1.1×10^{4}	1.5×10^{5}
	Bottom	1.9×10^{2}	3.2×10^{2}	5.0×10^{5}	7.8×10^{3}	3.0×10^{2}
		2.8×10^{2}	8.4×10^{2}	9.3×10^{2}	1.1×10^{5}	3.3×10^{5}
		5.9×10^{2}	1.4×10^{3}	2.3×10^{3}	9.9×10^{3}	1.7×10^{5}
8	Top	5.8×10^{2}	3.0×10^{3}	3.1×10^{6}	3.4×10^{3}	4.8×10^{3}
		2.6×10^{2}	7.9×10^{4}	5.9×10^{4}	3.5×10^{5}	5.9×10^{5}
		7.9×10^{2}	6.7×10^{3}	5.6×10^{4}	2.5×10^{5}	9.3×10^{5}
	Middle	1.7×10^{2}	4.8×10^{4}	5.8×10^{6}	2.0×10^{4}	1.6×10^{4}
		2.6×10^{2}	1.2×10^{4}	8.4×10^{3}	9.2×10^{5}	3.1×10^{6}
		4.9×10^{2}	2.1×10^{4}	3.6×10^{5}	1.2×10^{5}	4.9×10^{6}
	Bottom	7.0×10^{1}	2.8×10^{3}	2.2×10^{5}	2.6×10^{3}	1.7×10^{4}
		3.1×10^{2}	2.0×10^{3}	1.2×10^{3}	2.6×10^{5}	1.1×10^{6}
		3.2×10^{2}	1.5×10^{3}	1.3×10^{4}	3.0×10^{4}	2.1×10^{5}
12	Top	2.2×10^{2}	1.2×10^{5}	4.0×10^{6}	2.6×10^{3}	1.8×10^{4}
		2.0×10^{2}	2.6×10^{5}	6.5×10^{5}	1.5×10^{6}	5.8×10^{5}
		4.9×10^{2}	8.6×10^{4}	2.0×10^{5}	3.6×10^{6}	6.1×10^{5}
	Middle	1.6×10^{2}	6.6×10^{5}	2.7×10^{7}	2.4×10^{5}	9.0×10^{5}
		2.1×10^{2}	7.0×10^{5}	2.1×10^{5}	5.1×10^{6}	1.9×10^{6}
		4.7×10^{2}	6.8×10^{5}	6.7×10^{6}	3.8×10^{7}	6.7×10^{6}
	Bottom	3.4×10^{2}	3.2×10^{4}	4.3×10^{5}	1.3×10^{4}	1.9×10^{5}
		4.6×10^{2}	1.3×10^{4}	9.9×10^{3}	5.2×10^{5}	1.4×10^{6}
		3.2×10^{2}	1.1×10^{4}	4.3×10^{5}	2.7×10^{5}	6.8×10^{5}

TABLE III

PSYCHROTROPHIC BACTERIAL COUNT PER GRAM OF MEAT
DETERMINED AT THREE POSITIONS IN BOXES OF
HOT BONED BEEF TRIMMINGS DURING STORAGE

Box Depth (inches)	Position	Storage (hours) 0	24	48	72	96
4	Top	2.2×10^{2}	1.1×10^{3}	3.0×10^{6}	3.2×10^{4}	1.7×10^{4}
		2.0×10^{1}	2.1×10^{4}	9.0×10^{2}	1.2×10^{7}	4.4×10^{7}
		2.0×10^{1}	1.5×10^{3}	2.5×10^{4}	1.6×10^{4}	2.5×10^{6}
	Middle	5.0×10^{2}	2.4×10^{2}	1.8×10^{6}	1.1×10^{4}	1.0×10^{4}
		1.0×10^{1}	2.0×10^{4}	4.2×10^{2}	5.0×10^{5}	1.2×10^{6}
		4.0×10^{1}	3.6×10^{3}	1.0×10^{4}	6.9×10^{3}	2.2×10^{6}
	Bottom	1.0×10^{2}	8.0×10^{1}	8.7×10^{5}	2.2×10^{4}	2.2×10^{3}
		2.0×10^{1}	3.7×10^{3}	8.3×10^{2}	4.9×10^{5}	6.7×10^{5}
		6.0×10^{1}	2.3×10^{3}	9.7×10^{3}	6.2×10^{3}	1.8×10^{5}
8	Top	4.8×10^{3}	5.6×10^{3}	7.3×10^{7}	8.2×10^{5}	7.4×10^{3}
		3.0×10^{1}	3.6×10^{5}	9.0×10^{4}	1.0×10^{8}	2.8×10^{8}
		8.0×10^{1}	3.5×10^{4}	1.0×10^{6}	6.0×10^{6}	1.3×10^{8}
	Middle	1.3×10^{2}	1.5×10^{3}	4.9×10^{6}	4.0×10^{4}	9.4×10^{3}
		1.6×10^{2}	3.9×10^{4}	2.9×10^{3}	1.9×10^{7}	1.6×10^{7}
		1.0×10^{1}	1.1×10^{4}	3.4×10^{4}	9.0×10^{4}	1.4×10^{7}
	Bottom	8.0×10^{1}	4.4×10^{2}	8.5×10^{5}	6.4×10^{3}	6.0×10^{3}
		1.5×10^{2}	5.5×10^{3}	5.0×10^{2}	7.5×10^{5}	3.2×10^{6}
		5.0×10^{1}	2.3×10^{3}	6.3×10^{3}	1.0×10^{4}	7.5×10^{5}
12	Top	7.0×10^{2}	6.0×10^{4}	5.2×10^{7}	1.1×10^{6}	3.2×10^{5}
		5.0×10^{1}	1.5×10^{6}	1.2×10^{6}	8.5×10^{7}	1.1×10^{8}
		2.0×10^{1}	8.7×10^{5}	4.9×10^{7}	8.2×10^{7}	1.4×10^{8}
	Middle	3.8×10^{2}	1.6×10^{4}	2.0×10^{7}	1.1×10^{5}	5.6×10^{4}
		4.0×10^{1}	1.0×10^{5}	5.4×10^{4}	6.3×10^{6}	4.1×10^{6}
		3.2×10^{2}	1.4×10^{5}	1.6×10^{6}	1.1×10^{7}	6.7×10^{6}
	Bottom	7.0×10^{2}	1.9×10^{3}	1.1×10^{6}	4.2×10^{4}	5.6×10^{3}
		7.0×10^{1}	1.2×10^{4}	1.9×10^{3}	6.8×10^{5}	3.3×10^{6}
		2.8×10^{2}	8.3×10^{3}	1.5×10^{5}	3.1×10^{5}	1.2×10^{6}

APPENDIX B

ANALYSES OF VARIANCE

TABLE IV

ANALYSIS OF VARIANCE: TOTAL AEROBIC BACTERIAL COUNTS (LOG_{10}) ON HOT BONED BOXED BEEF TRIMMINGS

Source of Variation	df	Sum of Squares	F Value	Pr>F
Total Corrected	134	332.48		
Among Carcasses	14			
Storage Period (S)	4	155.12	5.01	0.02
Error a	10	77.33		
Within Carcasses Among Box Depths	30			
Box Depth (B)	2	35.11	34.22	0.00
B X S	8	7.36	1.79	0.14
Error b	20	10.26		
Within Box Depths-Among Positions	90			
Position (P)	2	18.70	44.83	0.00
P X B	4	3.96	4.75	0.00
P X S	8	7.84	4.70	0.00
P X B X S	16	4.29	1.29	0.24
Error c	60	12.52		

TABLE V

ANALYSIS OF VARIANCE: ANAEROBIC BACTERIAL COUNTS (LOG_{10}) ON HOT BONED BOXED BEEF TRIMMINGS

Source of Variation	df	Sum of Squares	F Value	Pr>F
Total Corrected	134	293.36		
Among Carcasses	14			
Storage Period (S)	4	133.26	5.94	0.01
Error a	10	56.13		
Within Carcasses - Among Box Depths	30			
Box Depth (B)	2	51.37	48.11	0.00
B X S	8	12.66	1.48	0.23
Error b	20	10.68		
Within Box Depths - Among Positions	90			
Position (P)	2	5.40	17.50	0.00
P X B	4	7.42	12.04	0.00
P X S	8	4.69	3.80	0.00
P X B X S	16	2.50	1.01	0.46
Error c	60	9.25		

TABLE VI

ANALYSIS OF VARIANCE: PSYCHROTROPHIC BACTERIAL COUNTS (LOG_{10}) ON HOT BONED BOXED BEEF TRIMMINGS

Source of Variation	df	Sum of Squares	F Value	Pr>F
Total Corrected	134	498.62		
Among Carcasses	14			
Storage Period (S)	4	280.49	6.15	0.01
Error a	10	114.11		
Within Carcasses - Among Box Depths	30			
Box Depth (B)	2	30.78	32.83	0.00
B X S	8	5.06	1.35	0.28
Error b	20	9.38		
Within Box Depths - Among Positions	90			
Position (P)	2	32.59	115.63	0.00
P X B	4	4.76	8.45	0.00
P X S	8	8.96	7.95	0.00
P X B X S	16	4.03	1.79	0.05
Error c	60	8.46		

APPENDIX C

MEANS

TABLE VII

TOTAL AEROBIC BACTERIA IN BOXES OF HOT BONED BEEF TRIMMINGS STORED FOR DIFFERENT PERIODS

Box Depth (inches)	Storage (hours)					Overall Means[2]
	0	24	48	72	96	
4	2.94[1]	3.44	4.64	4.80	5.35	4.23
8	3.04	4.31	5.11	5.70	5.99	4.83
12	3.14	5.01	6.25	6.43	6.57	5.48
Overall Means[3]	3.04	4.25	5.33	5.64	5.97	

[1]Each value is the average $\log_{10}$ count/g of 9 determinations.
[2]Each value is the average $\log_{10}$ count/g of 45 determinations.
[3]Each value is the average $\log_{10}$ count/g of 27 determinations.

TABLE VIII

ANAEROBIC BACTERIA IN BOXES OF HOT BONED BEEF TRIMMINGS STORED FOR DIFFERENT PERIODS

Box Depth (inches	Storage (hours)					Overall Means[2]
	0	24	48	72	96	
4	2.35[1]	2.82	3.98	4.00	4.15	3.46
8	2.47	3.91	4.98	4.79	5.38	4.31
12	2.47	5.07	5.84	5.68	5.79	4.97
Overall Means[3]	2.43	3.93	4.82	4.93	5.11	

[1]Each value is the average $\log_{10}$ count/g of 9 determinations.
[2]Each value is the average $\log_{10}$ count/g of 45 determinations.
[3]Each value is the average $\log_{10}$ count/g of 27 determinations.

TABLE IX

PSYCHROTROPHIC BACTERIA IN BOXES OF HOT BONED BEEF TRIMMINGS STORED FOR DIFFERENT PERIODS

Box Depth (inches)	Storage (hours)					Overall Means[2]
	0	24	48	72	96	
4	1.70[1]	3.29	4.39	4.80	5.46	3.93
8	2.02	3.93	5.10	5.69	6.10	4.57
12	2.19	4.72	6.02	6.30	6.25	5.10
Overall Means[3]	1.97	3.98	5.17	5.60	5.94	

[1]Each value is the average $\log_{10}$ count/g of 9 determinations.
[2]Each value is the average $\log_{10}$ count/g of 45 determinations.
[3]Each value is the average $\log_{10}$ count/g of 27 determinations.

TABLE X

TOTAL AEROBIC BACTERIA ON HOT BONED BEEF TRIMMINGS AT THREE POSITIONS IN BOXES OF DIFFERENT DEPTH

Position	Box Depth (inches)			Overall Means[2]
	0	8	12	
Top	4.46[1]	5.30	6.06	5.27
Middle	4.15	4.95	5.61	4.90
Bottom	4.08	4.24	4.77	4.37
Overall Means[3]	4.23	4.83	5.48	

[1]Each value is the average $\log_{10}$ count/g of 15 determinations.
[2]Each value is the average $\log_{10}$ count/g of 45 determinations.
[3]Each value is the average $\log_{10}$ count/g of 45 determinations.

TABLE XI

TOTAL AEROBIC BACTERIA AT THREE POSITIONS IN BOXES OF HOT BONED BEEF TRIMMINGS STORED FOR DIFFERENT PERIODS

Position	Storage (hours) 0	24	48	72	96	Overall Means[2]
Top	2.95[1]	4.69	5.79	6.43	6.50	5.27
Middle	3.06	4.25	5.49	5.68	6.01	4.90
Bottom	3.10	3.82	4.72	4.81	5.39	4.37
Overall Means[3]	3.04	4.25	5.33	5.64	5.97	

[1]Each value is the average $\log_{10}$ count/g of 9 determinations.
[2]Each value is the average $\log_{10}$ count/g of 45 determinations.
[3]Each value is the average $\log_{10}$ count/g of 27 determinations.

TABLE XII

ANAEROBIC BACTERIA ON HOT BONED BEEF TRIMMINGS AT THREE POSITIONS IN BOXES OF DIFFERENT DEPTH

Position	Box Depth (inches) 0	8	12	Overall Means[2]
Top	3.42[1]	4.41	4.83	4.22
Middle	3.34	4.63	5.53	4.50
Bottom	3.62	3.88	4.54	4.01
Overall Means[3]	3.46	4.31	4.97	

[1]Each value is the average $\log_{10}$ count/g of 15 determinations.
[2]Each value is the average $\log_{10}$ count/g of 45 determinations.
[3]Each value is the average $\log_{10}$ count/g of 45 determinations.

TABLE XIII

ANAEROBIC BACTERIA AT THREE POSITIONS IN BOXES OF HOT BONED BEEF TRIMMINGS STORED FOR DIFFERENT PERIODS

Position	Storage (hours) 0	24	48	72	96	Overall Means[2]
Top	2.47[1]	4.08	5.06	4.68	4.82	4.22
Middle	2.38	4.26	5.31	5.17	5.38	4.50
Bottom	2.45	3.46	4.43	4.61	5.12	4.01
Overall Means[3]	2.43	3.93	4.82	4.93	5.11	

[1]Each value is the average $\log_{10}$ count/g of 9 determinations.
[2]Each value is the average $\log_{10}$ count/g of 45 determinations.
[3]Each value is the average $\log_{10}$ count/g of 27 determinations.

TABLE XIV

PSYCHROTROPHIC BACTERIA ON HOT BONED BEEF TRIMMINGS AT THREE POSITIONS IN BOXES OF DIFFERENT DEPTH

Position	Box Depth (inches) 0	8	12	Overall Means[2]
Top	4.22[1]	5.39	5.85	5.16
Middle	3.89	4.46	5.09	4.48
Bottom	3.68	3.85	4.35	3.96
Overall Means[3]	3.93	4.57	5.10	

[1]Each value is the average $\log_{10}$ count/g of 15 determinations.
[2]Each value is the average $\log_{10}$ count/g of 45 determinations.
[3]Each value is the average $\log_{10}$ count/g of 45 determinations.

TABLE XV

PSYCHROTROPHIC BACTERIA AT THREE POSITIONS
IN BOXES OF HOT BONED BEEF TRIMMINGS
STORED FOR DIFFERENT PERIODS

Position	Storage (hours) 0	24	48	72	96	Overall Means[2]
Top	1.98[1]	4.59	6.01	6.49	6.71	5.16
Middle	1.92	4.04	5.09	5.48	5.88	4.48
Bottom	2.00	3.32	4.42	4.83	5.22	3.96
Overall Means[3]	1.97	3.98	5.17	5.60	5.94	

[1]Each value is the average $\log_{10}$ count/g of 9 determinations.
[2]Each value is the average $\log_{10}$ count/g of 45 determinations.
[3]Each value is the average $\log_{10}$ count/g of 27 determinations.

TABLE XVI

AVERAGE AIR AND MEAT TEMPERATURE (°F) AT DIFFERENT POSITIONS DURING BOXED STORAGE OF HOT BONED BEEF TRIMMINGS

Storage (hours)	Air Temperature	Box Depth (inches)								
		4			8			12		
		T	M	B	T	M	B	T	M	B
0	43.8	78.2	81.3	78.3	80.9	83.7	80.3	77.9	83.8	81.1
6	42.7	62.4	66.2	63.3	69.0	74.8	65.6	64.7	78.1	67.8
12	42.8	53.8	56.0	54.2	60.5	65.2	57.8	58.5	69.6	60.1
24	43.5	46.4	46.5	46.3	50.9	52.6	49.5	51.3	57.2	51.5
48	41.5	41.5	41.5	41.5	42.5	42.6	42.3	43.3	44.7	43.1
72	41.5	41.5	41.5	41.5	41.8	41.8	41.7	42.0	42.0	42.0
96	39.7	39.7	39.7	39.7	40.0	40.0	39.7	40.0	40.0	40.0

T = Top position; M = Middle position; B = Bottom position.

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