THE DEVELOPMENT OF MICROBIAL DECONTAMINATION AND MOISTURE LOSS CONTROL PROCEDURES FOR BEEF, PORK AND LAMB CARCASSES

By

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Abstract of Dissertation Presented to the Graduate Council
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THE DEVELOPMENT OF MICROBIAL DECONTAMINATION AND MOISTURE LOSS CONTROL PROCEDURES FOR BEEF, PORK AND LAMB CARCASSES

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Surface microbial decontamination and moisture loss control procedures were evaluated on lamb, beef and pork carcasses.

Decontamination of beef carcasses, resulting in a significant decrease in surface microbial flora at 24 and 96 hr postmortem was accomplished by combining hypochlorous acid with an acetate-acetic buffer. This antimicrobial activity was observed with concentrations between 25 and 200 ppm available chlorine when suspended in 0.01 M, pH 4.5 acetate buffer. Significant reduction in surface microbial flora was observed within 12 hr post-treatment but was non-significant at 1 hr post-treatment. When hypochlorous acid was combined with a sodium alginate solution (Flavor-Tex®) and sprayed onto pork carcasses, no significant reduction in surface microbial flora occurred. However, lamb carcass surface microbial flora was significantly reduced by the Flavor-Tex film.

Application of the edible alginate film coating as a moisture loss

control procedure to freshly slaughtered carcasses significantly reduced chill cooler shrinkage, and maintained this superiority through 72 hours postmortem.

At low alginate concentrations, loosely held water within the gel allowed for a greater reduction in shrinkage loss than when high concentrations of the alginate were sprayed onto beef carcasses. This trapped water permitted the gel to act as the moisture sacrificing agent on the carcass during chill cooler storage, with maximum shrinkage control being obtained within the initial 72 hr period. Oxygen permeability of the Ca-alginate films was not impeded; increasing as moisture content of the film decreased.

Application of either or both control procedures could effectively reduce shrinkage and surface microbial flora on lamb, beef and pork carcasses.

INTRODUCTION

The loss of moisture associated with beef, lamb and pork carcasses during the post-slaughter chill and distribution periods is of major concern to the meat packing industry. Following an initial 1.0-1.5% moisture loss, an estimated 0.5% daily reduction in weight occurs. Continued moisture loss results in discoloration of lean, desiccation of surfaces, increased trim losses and ultimately decreased shelf-life and consumer acceptability.

Control of the heat and mass transfer during cooling can be maintained by increasing the relative humidity in the chill cooler and/or reducing air circulation to a minimum. However, elevated relative humidity in the chill cooler increases the water activity (a_w) on the carcass surface. This increased a_w promotes surface microbial growth even though good processing and sanitation procedures are observed. Techniques commonly used to reduce the initial moisture loss from beef carcasses involve the moist cotton shroud which is tightly wrapped around the carcass. Pork and lamb carcasses remain unprotected throughout the initial chill and storage period.

Reduction of the surface microbial flora remains a difficult procedure for the meat processor. Techniques employing elevated CO_2 atmospheres, high pressure hot, warm and cold waters, drying of the carcasses, ultra-violet light, spraying of antibiotics, use of organic sprays and bactericidal agents have at one time or another been used to reduce this flora.

Development of procedures which effectively reduce and maintain negligible surface microbial levels could be beneficial to meat when it is aged to enhance tenderness. The use of moderate chill temperatures of 14-19 C have been shown to enhance tenderness, however processors are hesitant to use this procedure due to increased surface microbial growth.

From these observations, the objectives of this study were

(1) To develop procedures to effectively reduce and maintain low levels of microbial growth on the surface of these carcasses.

(2) To evaluate various moisture loss control procedures on beef, lamb and pork carcasses.

LITERATURE REVIEW

Carcass Chilling

The movement of heat from carcass tissues to the surrounding cold air results in a rapid decrease in carcass temperature. This chilling process, according to Locker et al. (1975) can be separated into two phases: (1) cooling and (2) storage. During the cooling phase the carcass surface heat (38-40 C) initially interacts with the chill cooler air (1-3 C). The rate of heat transfer from the hot carcass to the air will be proportional to the air velocity in the chill cooler. Except when there is little air velocity, the resulting high vapor pressure between the hot carcass surface and surrounding air produces a rapid flow of heat and moisture from the carcass. It is in this phase of the cooling process that the greatest shrinkage, or moisture loss, occurs.

In the storage phase, the vapor pressure between the carcass surface and surrounding air has decreased, therefore the regulation of the relative humidity in the chill cooler becomes critical for the control of shrinkage and microbial growth. When the convectional heat has been removed from the carcass, the air and carcass temperature become equilibrated and the vapor pressure difference between the carcass surface and the surrounding air is reduced. At this point, increases in the relative humidity reduces the drying power of the air. This increased relative humidity can be employed effectively to reduce further moisture loss. However, elevation of the relative humidity increases the water

activity (a_w) on the carcass surface and may result in enhanced microbial growth. Because moisture loss from the carcass is relatively slight in the storage phase, a reduced relative humidity is generally employed so that increased microbial growth can be prevented.

Development of an alternative chill cooling procedure to reduce beef carcass shrinkage has been reported by Watt and Herring (1974). Their study indicated that chilling beef carcasses in an ammonia-mechanical blast cooler at -29 C for four hours followed by a 14 hr equilibration period at 2 C significantly reduced shrinkage; however cold shortening, which results in a toughening of the muscle tissue, developed. This toughening of the muscle tissue is an important factor which must be considered when attempting to chill carcasses rapidly.

In addition to refrigeration and environmental factors, physiological characteristics of the animal may influence the degree of shrinkage. Some of the more important physiological factors are as follows: ante-mortem stress of the animal (Davidson et al., 1968), subcutaneous fat thickness of the carcass (Smith and Carpenter, 1973), and carcass weight and sex of animal (Fredeen et al., 1971).

Beef Carcass Shrinkage

A standard procedure used for the control of beef carcass shrinkage during the initial chill period is the shrouding of hot carcasses in a cotton shroud. The wet cloth, which is soaked in hot salt water prior to being tightly wrapped around the carcass, reduces carcass moisture loss during the cooling phase. The removal of the moisture from the shroud instead of the carcass during the transfer of heat to the surrounding air allows the wet cloth to act as a moisture sacrificing agent.

Following a 18-24 hr chill, the shroud is removed from the carcass.

Data collected by Fredeen et al. (1971) relating bovine animal and carcass characteristics to carcass shrinkage, revealed that moisture loss was inversely related to carcass weight and that the animal's sex influenced shrinkage with heifers having the lowest loss (0.4%) and bull carcasses the greatest (0.91%). Overall fat thickness, which is inversely related to carcass shrinkage (Smith and Carpenter, 1973) was greater in the heifer carcasses than bull carcasses.

Control and reduction of shrinkage for beef carcasses and wholesale cuts during shipment to central distribution centers or retail outlets is important to the meat industry. In a study by Rea et al. (1972), beef wholesale cuts were wrapped in paper, polyethylene bags or polyvinyl chloride films prior to long distance shipment. Essentially all treatments had significantly lower intransit shrinkage when compared with the unprotected cut. Muscle color scores for the individual treatments were similar, however subcutaneous fat color was significantly fresher looking in appearance (whiter) when the cuts were packaged in polyethylene bag containers.

Lamb Carcass Shrinkage

Control of lamb carcass shrinkage has centered around the interrelationship of temperature and relative humidity to control surface desiccation (Fleming and Earle, 1968; Smith and Carpenter, 1973). Because no artificial covering is placed about the carcass, the fell membrane, a connective tissue lying on the surface, can provide only limited protection from moisture loss. Smith and Carpenter (1973) reported that 92% of the 72 hr weight loss in lamb carcasses occurred

during the first 36 hr period. Subcutaneous fat thickness and carcass weight significantly affected shrinkage loss with reduced losses being attributed to either decreases in total surface area per unit weight and/or increased fat covering. Fat thickness between 2.5 and 9.1 mm was associated with decreased weight loss over the time periods examined. Lamb carcass shrinkage, as affected by relative humidity, was shown by Fleming and Earle (1968) to be lowest at 90-97% relative humidity and highest at 51-57%. Actual and estimated weight losses indicated that by 6 hr post-slaughter, a 25 kg ewe had an accumulated weight loss of 1.8%. Nottingham (1971) reported that when lamb carcasses were stored at 100% relative humidity and little air velocity, shrinkage amounted to 0.6% while at 85% relative humidity and 0.1 m/sec air velocity, shrinkage amounted to 2.3%. Carpenter et al. (1975) reported that shrinkage losses for lamb carcasses wrapped in PVC film were substantially reduced and external fat covering was more attractive than when carcasses were left uncovered. Recently, Smith et al. (1976) reported that high velocity air drying increased lamb carcass shrinkage significantly while polyvinyl chloride film wrap decreased moisture loss significantly, especially when placed about the carcass in a mummy-type manner.

Pork Carcass Shrinkage

Very little published information is available concerning pork carcass shrinkage. In a study by Davidson et al. (1968), the effects of ante-mortem stress induced by fasting littermate pigs for 68-70 hr were evaluated for slaughter and carcass characteristics. Following a 24 hr chill, shrinkage was 2.42 and 2.83% for the fed and fasted groups,

respectively. With swine carcasses being fabricated into wholesale cuts, wrapped, boxed and shipped within 24 hr after slaughter, the need to reduce this large amount of shrinkage during the cooling phase becomes economically important.

Edible and Non-Edible Coatings

Early research by Pearce and Lavers (1949) indicated that poultry carcasses dipped in a melted (65.5 C) solution of carrageenin (a gelatinous polysaccharide extracted from Irish moss) and sodium chloride prior to freezing delayed off-odors on eviscerated surfaces of defrosted carcasses. Meyer et al. (1959), in evaluating an agar and carrageenin gel on poultry parts, observed little improvement in retarding microbial spoilage when coated samples were stored at 24 or 13 C. Shelf-life improved slightly when the coated poultry was stored at 2 C. Use of these gels as a carrier for water soluble antibiotics proved successful in inhibiting microbial growth. Ayres (1959) found that when a hot melt (65.5 C) diacetin fat agar solution with plasticizer was coated onto fresh meats, microbial growth was retarded and desiccation prevented; however, these coated meats had an undesireable color. Zabik and Dawson (1963) reported off-flavors but increased percent press fluids and less cooking losses than controls when poultry pieces were coated with an edible acetylated mono-glyceride solution (Myvacet 7-00, Myvacet 7-15) and stored for 1 and 2 weeks at 4 C. Ayres (1964), using an inedible hot melt coating material (Lepak), observed that pork chops stored for 5 months at -30 C in an air-blast freezer had a 2% moisture loss, while controls had 2.4% loss. Off-flavors in the coated samples were observed prior to cooking and may have been due to the high temperature (180-220 C)

of the hot melt solution.

Naturally occurring film components were evaluated by Allen et al. (1963). These films, consisting of either sodium alginate or alginate-cornstarch solutions, were heated to 87.7 C with beef steaks, pork chops and poultry pieces being dipped into them. After dipping, the sample was dipped into 5 M $CaCl_2$ to form a semisolid film. In general the alginate-cornstarch mixture retarded moisture loss more than the plain alginate, while both coating solutions effectively reduced moisture loss when compared to untreated controls. Earle (1968a) described a calcium alginate gel coating not requiring elevated temperatures for solubility, to be used for the protection of raw fish, meat and poultry. This edible coating, known commercially as Flavor-Tex®, involves the formation of a film around the food product by gelling the maltodextran sodium alginate coating with a calcium chloride-carboxymethylcellulose solution. Earle (1968b) reported that Flavor-Tex treated samples had less moisture loss than untreated controls and exhibited no bitter off-flavors as had been reported by Allen et al. (1963).

Microbial Aspects of Meat Carcasses

Microbial growth and spoilage of meat present many problems to the meat industry. Dockerty et al. (1970) indicated three general factors that can influence ultimate spoilage of fresh meat: (1) dressing; (2) wholesale cutting and shipping; and (3) retail cutting and storage shelf-life. The sources from which spoilage organisms originate are numerous; hides, equipment, water and man are the primary sources (Empey and Scott, 1939; Ayres, 1955; Patterson, 1967, 1968). The bacterial flora on the surface of carcasses prior to entering the chill cooler

and their subsequent growth becomes a source for cross-contamination and distribution onto cut surfaces.

Besides physical conditions which contribute to the surface microbial flora, environmental conditions determine the eventual growth of these microorganisms (Locker et al., 1975). Rapid chilling is one effective means of controlling microbial growth; but the effects of cold shortening as mentioned earlier must be considered so that a balance is established between palatability and sanitation.

Normal chill room temperatures exert a lethal effect on bacteria, particularly when combined with decreased water activity and nutrient availability (Locker et al., 1975). Although the a_w for most bacteria is in the range of 0.90-0.99 (Lamana and Mallette, 1965), psychrotrophic bacteria become the predominant flora following the initial 24-48 hr chill (Thatcher and Clark, 1968). These surviving bacteria, and the presence of yeasts and filamentous fungi which survive low a_w's, contribute to the eventual spoilage of meat tissue (Lawrie, 1974).

Reducing the relative humidity in the chill cooler, although effective in inhibiting bacterial growth, exerts a negative effect on carcasses by increasing shrinkage, desiccation, and, when frozen, freezer burn, which reduces the quality of the meat. These effects are especially important for the lamb carcass which has a large surface area to volume ratio (Locker et al., 1975).

Beef Surface Microbial Flora

Beef slaughtering is a highly industrialized process involving modern machinery to remove the hide and shanks automatically. This mechanization eliminates a large source of microbial contaminants by

reducing the direct contact of the carcass with those less clean parts. Use of wash-water tunnels with optimum pressure nozzles for washing carcass surfaces has helped to further remove many contaminating materials (bone, hair, fecal matter). Following washing procedures, no further treatments are normally performed to reduce the remaining microbial flora. An exception to this is the Clor-Chil® process, implemented by Swift & Company (Heitter, 1975). In this process, carcasses are intermittently sprayed in the chill cooler with a mild chlorine solution to reduce surface microflora and moisture loss.

Lamb Surface Microbial Flora

The lamb slaughter process involves extensive human contact with the carcass surface. The removal of the pelt by the fisting technique and the presence of soiled fleece increases the level of surface microbial flora. Washing thoroughly helps to reduce the surface flora, however ample microbial growth has been demonstrated on the carcass (Patterson, 1968). An attempt to control microbial growth on lamb carcasses was conducted by Carpenter *et al.* (1975). Treatment of carcasses with 200 ppm available chlorine reduced bacterial counts substantially without impairing subsequent meat flavor. It was also observed that decontaminating agents were most effective in reducing bacterial growth when applied to carcasses immediately post-slaughter. Treatment of carcasses with chlorine after 7 days storage also resulted in a substantial reduction in microbial growth.

Pork Surface Microbial Flora

The manner in which swine are slaughtered presents many opportunities

for cross-contamination as well as reduction of the microbial flora. The scalding of hogs at 58-62 C in a common vat following exsanguination provides a medium for the dispersal of pathogenic and saprophytic bacteria from one carcass to another. However, in the singeing and surface cleaning process, reduction of the microbial load has been observed (Dockerty et al., 1970).

Control of Carcass Microorganisms

To effectively control carcass surface microorganisms, the kinds of bacteria, yeasts and filamentous fungi normally present must be identified. Early investigations (Empey and Scott, 1939; Haines, 1933) revealed the presence of *Achromobacter* species. Taxonomic revisions now include this genus in the genus *Pseudomonas* which was then commonly reported to be present most frequently. Stringer et al. (1969) indicated that in addition to *Pseudomonas*, *Micrococcus* and *Bacillus* constituted the majority of carcass surface microorganisms. More recently, Locker et al. (1975) reported that the initial microflora on the surface of beef carcasses consisted of *Micrococcus* (43%), *Staphylococcus* (27%) and smaller amounts of *Acinetobacter*, *Pseudomonas*, *Corynebacteria* and bacteria of the *Enterobacteriaceae* family.

The combination of chill cooler temperature and reduced a_w can selectively inhibit some bacteria (e.g. *Enterobacteriaceae*) and enhance the growth of others (*Micrococcus*, *Staphylococcus*, *Pseudomonas*) (Ingram, 1951). These selected bacteria then, become the important microflora to inhibit or reduce.

A review of the literature reveals that four basic processes have been investigated as possible means for controlling and reducing meat

surface microorganisms: (1) Alteration of the atmosphere in which the carcasses are stored; (2) antibiotics; (3) chlorine; and (4) organic acid sprays. Besides these four chemical treatments, Patterson (1972) indicated that the most commonly practiced method of reducing initial microbial flora is a final wash with either cold or warm water under pressure. When different water pressures were evaluated, he observed that water at 68 kg/6.45 cm^2 of pressure was significantly more effective in reducing surface microflora than either 113 or 159 kg/cm^2. Increased pressures were thought to drive the bacteria into the tissues, however the effects of water temperature (4-18 C), and chill cooler variation (3-10 C), could also have influenced this response.

Atmospheric Changes. As reported by Lugg and Woodruff (1973), Kolbe, in 1882, stored meat in an elevated carbon dioxide environment for 4-5 weeks without deteriorative changes. Killefer (1930), in comparing carbon dioxide, nitrogen and air atmospheres for the storage of pork and lamb carcasses, observed that carbon dioxide was superior to air or nitrogen after 10 days of storage while Brooks (1933) found that carbon dioxide atmospheres greater than 20% resulted in loss of acceptable beef muscle color. Recently, Marriott et al. (1976a) reported that trans-oceanic shipment of beef quarters and subprimal cuts, shipped in either air or modified atmosphere (60% CO_2, 25% O_2 and 15% N_2) vans resulted in no significant differences in weight loss, appearance or microbial flora following 20-21 days shipment. In a similar study, Marriott et al. (1976b), using similar air and modified atmosphere vans as before, noted that beef quarters had significantly less microbial flora from the modified van transport than the normal atmosphere van following 7-11 days of intransit storage.

Antibiotics and Radiation. Permission to use certain antibiotics (chlortetracycline, oxytetracycline) to control the microbial flora on raw poultry was granted by the Food and Drug Administration in November, 1955. By 1959, chlortetracycline was permitted in or on uncooked vertebrate fish; permission was not granted for red meats (Firman et al., 1959). Further studies on residual antibiotic compounds, development of resistant bacteria and the concept of additives to natural foods, ultimately forced all antibiotics to be prohibited in fish and red meat products (Desrosier, 1970).

Experiments in the application of beta or gamma radiation to fresh meat suggest this procedure results in a sterilization of the surface tissue (Phillips et al., 1961). This sterilization, however requires 2-4 megarads and can result in undesireable off-flavors. To prevent off-odors, which develop more rapidly in beef than pork (Kirn et al., 1956), lower radiation dosages (45,500 rads) in combination with the infusion of 30-50 ppm oxytetracycline into the meat tissues have been successfully used (Wilson et al., 1960).

Organic Acids. The initial use of organic acids on surface microbial flora centered on the treatment of chicken carcasses and sought to reduce or destroy salmonellae. Because chicken carcasses share a common chill water bath, the reduction of salmonellae as well as saprophytic bacteria would reduce cross-contamination. Data reported by Mountney and O'Malley (1965) indicated that acetic, adipic and succinic acids at 6.0, 3.5 and 1.0% concentration (all at pH 2.5) respectively, were more effective in reducing microbial numbers than were citric, fumaric, malonic, sorbic, hydrochloric, phosphoric and lactic acids. Thomson et al. (1967) reported that citric (0.3%) and succinic (1.0%) acids

reduced the growth of *Salmonella enteritidis* on inoculated fryer chickens. For the control of lamb carcass microflora, Ockerman et al. (1974) indicated that acetic acid at a concentration of 18% was significantly more effective than 6 or 12%, but that a bleaching effect occurred at 12%. Use of lactic acid, varying between 12 and 18% concentration over the 12 day storage period, was not as effective and a bleaching effect was again observed at 18%. Varnadore (1972) reported that propionic acid (4%) was more effective in reducing the microbial flora on lamb carcasses stored at 16 C for the first 24 hr postmortem and then moved to a 0 C cooler, than it was for carcasses stored at 0 C initially. However, carcasses treated with the propionic acid were dry on the surface and the subcutaneous fat was tannish-yellow in color.

For reduction of the microbial flora on pork carcasses, Biemuller et al. (1973) noted that, regardless of a 30 or 60 second spraying time, pH 2.0 acetic acid (0.1 N) significantly reduced surface flora but caused some surface discoloration. Five percent hydrogen peroxide, although effective in antibacterial activity, also caused a marked skin discoloration. Stannous chloride (5%), a reducing agent, caused the least amount of surface discoloration and significantly reduced surface microflora.

Chlorine Compounds. The first use of hypochlorites as a disinfectant occurred in the 1780's, followed by development of calcium hypochlorite as a sewage deodorant in England in the 1850's (Rudolph and Levine, 1941). As reported by Charlton and Levine (1937) the bactericidal activity of hypochlorites was first reported in 1881 by Koch, but it was not until World War I that chlorine compounds (chloramine, chloramine T, azochloramide) were widely used as tissue disinfectants.

The use of hypochlorites in water systems was first described by Johnson (1911), who reported that as little as 0.2 ppm might render water safe.

As early as 1904, the germicidal action of hypochlorites and chlorinated waters was thought to be due to the same substance; namely hypochlorous acid (HClO) (Charlton and Levine, 1937). Both the reaction of gaseous chlorine in the chlorination of water ($Cl_2 + H_2O \rightarrow HCl + HClO$) and the reaction of calcium hypochlorite ($Ca(OCl)_2 + HCl \rightarrow Ca(OH)_2 + HClO$) were shown to be greatly increased in the presence of various acids (Charlton and Levine, 1937). This pH dependence coincided with the oxidative nature of hypochlorous acid, wherein the 'nascent oxygen' oxidizes proteins following contact. The bactericidal property of hypochlorite has been shown to be proportional to the ratio of hypochlorous acid/hypochlorite ion in solution. At pH 7.5, half of the available chlorine is present as hypochlorous acid and half as hypochlroite ion. At pH 10, only 0.3% of the available chlorine is present as hypochlorous acid (Anonymous, 1964). To maximize bactericidal activity, the pH can be lowered (4.0-6.0), however the acid becomes less stable and inactivity develops. Other factors which contribute to deterioration of HClO are temperature and amount of organic matter present. High concentrations of proteinaceous materials are acted upon by the hypochlorous acid, while carbohydrates are relatively inert to oxidation.

Besides the treatment of municipal water systems with hypochlorous acid via Cl_2 gas, considerable data have been collected on the spraying and dipping of poultry and meat carcasses with an available chlorine compound to reduce *Salmonella*, other pathogens and saprophytic bacteria.

Dixon and Pooley (1961) reported that treatment of chicken carcasses with 200 ppm of chlorine solution for 10 min effectively reduced the presence of salmonellae when the carcasses had less than 1×10^3 total organisms. Thomson et al. (1967) indicated that if chicken carcasses were sprayed with water or with chlorine, *Salmonella typhimurium* counts were lower than unsprayed controls and that chlorine treatments resulted in significantly lower *S. typhimurium* counts than did water washing. However, no significant differences occurred between 100 and 200 ppm chlorine treatments.

For the control of surface microbial flora on lamb carcasses, Patterson (1968) recommended the use of 20 ppm free residual chlorine in the spray wash. Kotula et al. (1974) reported that a high pressure 24.6 kg/cm^2 wash water containing 200 ppm available chlorine was more effective than a 4.2 kg/cm^2 pressure and that the chlorine wash water at 51 C resulted in a larger decrease in viable bacteria than chlorine water at 18 C. Heitter (1975) reported that the Clor-Chil process, which involves intermittent spraying of carcasses with chlorine solutions during the initial chill phase, reduced viable bacterial counts on pork carcasses by 97-99%, on beef by 94-98% and by 94-99% on lamb carcasses. Marriott et al. (1976a,1976b) however, indicated that the use of 200 ppm sodium hypochlorite on shipments of beef quarters and subprimals resulted in no significant change in the microbial flora.

MATERIALS AND METHODS

Experiments were designed on an animal group and number available basis as they were received at the Meats Laboratory for slaughter. Due to availability, all groups of animals could not be evaluated for the same specific treatment. This was especially true for lamb carcasses which consisted of one experiment involving 90 animals. Beef carcasses were more readily available and were the most frequent species evaluated.

Processing of Carcasses

Lamb. A total of 90 lambs were slaughtered on three consecutive days (30/day). Each lamb was randomly assigned to a day of slaughter and to one of three shrinkage treatments: calcium alginate edible film coating, plastic wrap or control (no covering). The edible Ca-alginate film coating, Flavor-Tex® (U.S. patent No. 3,395,024, Food Research Inc., Tampa, Fla.) consists of two solutions: (1) sodium alginate-maltodextran, (142 g/liter water); and (2) $CaCl_2$-carboxymethylcellulose. Solution 1 was sprayed directly onto all surfaces of the carcass, followed immediately by the spraying of solution 2 over solution 1. Interaction of these two solutions by cooperative association (Morris, 1973) causes formation of a clear homogeneous film over the entire carcass. Both solutions were applied using a Binks Model 33-112 compressed air system fitted with dual spray guns (Binks Mfg. Co., Chicago, Ill.)

Carcasses which received the plastic wrap (Borden Resinite-90, a low moisture, high oxygen transfer wrap) were wrapped while hanging from the overhead rail. At the end of 24 hr, the plastic wrap was removed.

Control carcasses received no external covering. Immediately following slaughter and washing, all carcasses were moved by overhead rail into an adjacent room (10 C) where an initial microbial count, internal temperature from the thick area of the legs, hot carcass weight and shrinkage treatment were conducted. All carcasses were placed in a 2 C cooler with a relative humidity of 80% and a wind velocity of approximately 24 km/hr.

Carcass weights were determined at 0, 1, 2, 3, 5 and 7 days postmortem using a Toledo Model 2071 scale with 0.1 lb gradations. Shrinkage at each day postmortem was based on the initial hot carcass weight taken after washing the carcass.

Carcass temperature was determined by averaging the internal temperatures from both hind legs. Temperatures were collected at 0, 6, 24 and 48 hr postmortem using a pyrometer equipped with a 7.6 cm probe (PYRO Surface Pyrometer, Pyrometer Instrument Co.).

Beef. Eight experiments involving 172 beef carcasses were evaluated for moisture loss control procedures and/or surface microbial decontamination. For shrinkage treatments, following slaughter and washing, one side was randomly assigned to either the control (shroud), treatment (Flavor-Tex) or naked (no shroud) group. Flavor-Tex treated sides were sprayed with various concentrations of the sodium alginate-maltodextran solution. At the end of 24 hr the shroud cloth was removed from the control side.

Carcass weights were determined at 24 hr intervals using an on the rail scale with 0.5 lb gradations. Percent shrinkage at each day postmortem was calculated using the initial hot carcass weight after washing.

Pork. Following slaughter and washing, carcasses were sprayed with various concentrations of sodium alginate-maltodextran. Percent shrinkage at each day postmortem was calculated using the initial hot carcass weight after washing.

Decontamination of Carcass Surfaces

Carcass surface areas were sprayed with antimicrobial agents using a Binks Model 33-112 compressed air system fitted with dual spray guns. Application pressure was maintained at 1.0 kg/cm^2.

The primary antimicrobial agent employed in this study consisted of hypochlorous acid. From a filtered stock solution of calcium hypochlorite (1%), working concentrations up to and including 200 ppm were prepared (Anonymous, 1968). Final concentration of available chlorine was determined using a Hach High Range Chlorine Test Kit Model CN 21-P (Hach Chemical Co., Ames, Iowa).

The effectiveness of hypochlorous acid, as produced by electrolysis of chloride ions was evaluated by spraying concentrations up to 200 ppm available chlorine prepared by a Morton Biocidal Flow-Thru Design Unit, Model 110-415 1D (Morton Salt Co., Chicago, Ill.).

Organic buffer systems were prepared at various concentrations and pH using the Henderson-Hasselbach equation. Following buffer preparation, a given amount of stock calcium hypochlorite solution was added to the buffer system. This solution was then sprayed onto the carcasses.

Microbial Determinations

Lamb surface microbial samples were collected from the sirloin and belly (flank-plate juncture) areas of the carcass. A 6.46 cm^2 area was swabbed using the standard moist-swab technique (APHA, 1972). Serial dilutions were prepared using Butterfield's phosphate diluent and plates were poured with Standard Plate Count Agar (Difco) for aerobic counts. Incubation of the plates were for 5 days at 20 C. Samples were collected at 0, 2, 5 and 7 days postmortem.

Preliminary research by Lazarus and West (1975), as well as the work of Stringer (1975), using beef carcasses, indicated that the neck area, as opposed to the flank, outside round, inside round or rib region provided the highest concentration of microbial flora. This area has both lean and fat surfaces, is conveniently accessible, is not easily cross-contaminated and can be readily treated by decontaminating solutions. Swab samples were collected, serially diluted, plated and incubated as previously outlined. Surface microbial flora were identified according to the procedures of Vanderzant and Nickelson (1969) and Breed et al. (1975).

Pork surface microbial flora was monitored by swabbing the shoulder area of the split carcass at various time periods. Serial dilutions, agar medium and incubation of plates were similar to procedures previously outlined.

Analytical Procedures

Oxygen permeability of the Ca-alginate, Borden Resinite-90 and a high oxygen low moisture transmission film (Goodyear Prime Wrap) was

determined using a Model 777 Beckman Oxygen Analyzer. Figure 1 illustrates the apparatus constructed to determine the permeability of the films. Analyses were conducted at 2 C to simulate carcass storage conditions. Following removal of oxygen from the T-chamber by perfusion with nitrogen, the outlet tube was clamped (in addition to remaining in water). Oxygen in normal atmospheric air which entered through the film was monitored until saturation (160 mm O_2) of the chamber was obtained.

Calcium alginate gel properties, employing various concentrations of sodium alginate-maltodextran and calcium chloride were determined by reacting known weights and volumes of the two components in tared beakers. The resultant gel material as well as remaining liquid was weighed on a wet and dry basis (83 C, 24 hr) using a Mettler H10 balance (Mettler Instrument Corp., Princeton, N.J.). Percentage liquid and solid was determined from the total weights of the two solutions.

Loss of moisture from the Ca-alginate film was determined by reacting the two solutions on tared glass plates. The coated plates were stored at 2 and 83 C for a total of 72 hr with weights being collected at various time periods on a Mettler H10 balance. Percentage gel remaining was determined using the initial weight of the film.

Statistical Analysis

Data were analyzed using the Statistical Analysis System (SAS) designed and implemented by Barr and Goodnight (1972), analysis of variance (Snedecor and Cochran, 1967) and the mean separation technique of Duncan (1955).

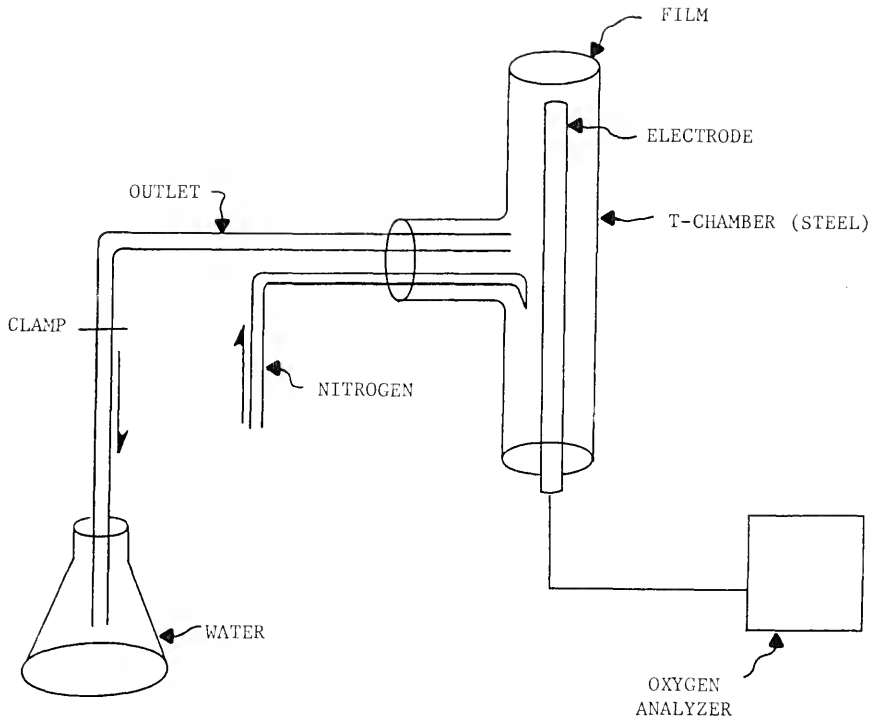

Fig. 1. Apparatus for measuring oxygen permeability of Ca-alginate and plastic wrapping films.

RESULTS AND DISCUSSION

Lamb Carcass Shrinkage Experiment

Overall, lamb carcass weights (n = 90) were from 17.8 - 28.8 kg with fat thickness measurements at the 13th rib ranging between 3.8 and 8.9 mm. This variation in level of fatness should not influence carcass shrinkage because it is within the 2.5 to 9.1 mm range described by Smith and Carpenter (1973) as being non-significant with respect to moisture loss.

Mean values for postmortem shrinkage of lamb carcasses analyzed by shrinkage treatment and by day of slaughter are presented in Table 1. At all time periods postmortem, the carcasses coated with Ca-alginate (142 g/l) or wrapped in plastic film had significantly ($P < 0.05$) lower shrinkage values than the control carcasses. In addition, the plastic wrapped carcasses had significantly less shrinkage than the Ca-alginate treated carcasses.

Smith and Carpenter (1973) reported that 75% of the 72 hr weight loss of lamb carcasses was incurred during the initial 24 hr post-slaughter chill period. These workers attributed this large initial shrinkage to loss of water added during the washing procedure. Subsequent weight loss was attributed to moisture loss, in the form of evaporation from the carcass tissue.

In this experiment, the plastic wrap impeded both evaporation of moisture and heat transfer (Table 1) from the carcass during the first

Table 1. Mean values[a] for postmortem shrinkage (%) and internal leg temperature (C) of lamb carcasses by shrinkage treatment and day of slaughter.

Day postmortem	Shrinkage treatment[b]			Day of slaughter		
	Control	Ca-alginate	Plastic wrap	1	2	3
	Shrinkage loss (%)					
1	2.77a	1.55b	1.20c	2.22a	1.59b	1.67b
2	3.25a	2.22b	1.88c	2.93a	2.04b	2.35b
3	3.80a	2.96b	2.49c	3.49a	2.68b	3.05b
5	4.71a	4.01b	3.43c	4.72a	3.64b	3.75b
7	5.36a	4.81b	4.19c	5.59a	4.34b	4.40b
Hour postmortem	Internal temperature (C)					
0	38.7a	38.2a	38.4a	36.4a	39.3b	39.8b
6	10.3a	10.0a	14.3b	10.3a	11.5ab	12.7b
24	4.1a	4.4a	5.0a	3.3a	4.7b	5.4b
48	5.3a	5.4a	5.4a	4.7a	6.0b	5.5b

[a]Means on the same horizontal line bearing different letters differ significantly ($P < 0.05$)

[b]n = 30 carcasses/treatment

24 hr. The 1.20% shrinkage recorded for this treatment group should primarily reflect loss of accumulated wash water. Accumulated moisture on the inside portion of the wrap and very moist carcass surfaces were noted for this group. These conditions would be expected to influence both microbial growth and initial chill rate.

In order to measure the effect of shrinkage treatment on chill rate, internal leg temperatures were collected (Table 1). At 6 hr postmortem, the plastic wrap had prevented the dissipation ($P < 0.05$) of carcass heat, but after 24 hr, no difference was observed. Addition of 30 hot carcasses/day to the cooler reduced ($P < 0.05$) (Table 1) the chilling rate of the carcasses; however no interaction between shrinkage treatment and day slaughtered occurred, indicating the loss of carcass heat was uniform across the treatments regardless of the effects of day slaughtered.

Visual evaluation of carcass appearance revealed treatment differences. Those carcasses receiving the plastic wrap treatment had moist surfaces and a softer, whiter subcutaneous fat covering than did the other carcasses. Smith et al. (1976) reported that carcasses stored in polyvinyl chloride film wrap throughout a 5 day storage period had an accumulation of moisture on the surfaces and exhibited an extremely attractive white fat. The alginate coated carcasses had a glossy pseudo-moist appearance and the surface fat was slightly darker than the fat on the control carcasses. Little variation in lean color was observed between the treatment groups.

The carcasses from the first day's slaughter group had greater ($P < 0.05$) shrinkage values on all subsequent days than did those carcasses slaughtered on following days (Table 1). The addition of 30 hot

carcasses/day to the chill cooler reduced the efficiency of the cooler, however a significant interaction between shrinkage treatment and day of slaughter was not observed.

Beef Carcass Shrinkage Experiments

Because the cotton shroud is commonly used in the meat industry to retard moisture loss, smooth and whiten subcutaneous fat, and improve carcass conformation, an experiment was conducted to determine what influence the shroud cloth had on moisture loss. As can be seen in Table 2, there were no differences ($P > 0.05$), using paired sides, between the two treatments. The overall small percentage moisture loss for these carcasses, regardless of the treatment, probably contributed to this response.

There is a need for an alternative process to replace the shroud cloth, which must be placed about the carcass manually, has a limited longevity and is costly both in terms of energy required for laundry and handling. With the beef carcass side being quite large, use of a polyvinyl chloride wrap becomes impractical. A reasonable alternative to the shroud is an edible film which could be sprayed on automatically, reducing labor costs while at the same time achieving improved moisture-control in the beef carcass.

Initial experiments in this study employed Flavor-Tex at a concentration of 127.5 g/l (sodium alginate-maltodextran, Solution 1). When this treatment concentration was compared with the shroud cloth to evaluate moisture loss control, the alginate coated carcasses had a lower ($P < 0.05$) moisture loss through 96 hr storage (Table 3) when compared with 24 hr shroud covered controls. However, in a later

Table 2. Mean values[a] for postmortem shrinkage[b] (%) of beef carcasses as affected by shroud cloth.

Treatment[c]	Hour postmortem		
	24	48	72
No shroud	1.31	1.38	1.48
Shroud[d]	1.23	1.35	1.39

[a]No significant differences ($P > 0.05$) between treatments

[b]Based on pre-washed hot carcass weight; paired sides

[c]n = 15 carcass sides/treatment

[d]Removed 24 hours postmortem

Table 3. Mean values[a] for postmortem shrinkage (%) of beef carcasses following shrinkage treatment.

Hour postmortem	Shrinkage treatment[b]	
	Control	Ca-alginate[c]
24	1.99a	1.51b
48	2.32a	1.81b
72	2.65a	2.04b
96	3.17a	2.67b

[a]Means on the same horizontal line bearing different letters differ significantly ($P < 0.05$)

[b]n = 12 carcass sides/treatment

[c]127.5 grams sodium alginate-maltodextran/liter water

experiment, employing three sodium alginate-maltodextran concentrations (Table 4), no differences ($P > 0.05$) were observed between the alginate coated carcasses and the paired side controls. The reason for the lack of agreement is probably due to the total number of carcasses being placed in the cooler. Where significance occurred (Table 3), only 12 carcasses were placed in the chill cooler. With the low number of carcasses in the cooler, low humidity (80% R.H.) and high vapor pressure between the carcass and cooler air resulted in a clear distinction between the two treatments. In the second experiment (Table 4), 78 hot carcasses were introduced into the chill cooler (60 were used in the experiment). This would increase the humidity, reduce the vapor pressure differential and tend to equalize all treatments being evaluated. This effect can be seen in the differences between the shrinkage control carcasses in the two experiments (1.99% for the 12 carcass experiment and 1.51% for the 60 carcass experiment).

Pork Carcass Shrinkage Experiments

Since pork carcasses are cut and fabricated following a 24 hr chill, the reduction of moisture loss from the carcass could be economically important to the processor. With an increased carcass surface area to volume ratio, considerable moisture is removed during the initial chill period. Data presented in Table 5 summarizes the affect of Ca-alginate (131 g/l) on controlling the initial shrinkage. Although only 24 hr weights could be collected, a reduction ($P < 0.01$) in shrinkage occurred. When carcasses became available, so that weights could be collected up to 72 hr postmortem, a similar response using 98 g/l sodium alginate-maltodextran was observed (Table 6). Similarly, when various concentra-

Table 4. Mean values[a] for postmortem shrinkage (%) of beef carcasses[b] with different concentrations of alginate-maltodextran.

Hour postmortem	Alginate-maltodextran (g/l)					
	Control	98	Control	120	Control	142
24	1.45	1.46	1.64	1.50	1.48	1.44
48	1.70	1.75	1.91	1.81	1.67	1.93
72	2.07	N.D.[c]	2.24	N.D.	2.04	N.D.

[a]No significant ($P > 0.05$) differences between alginate concentrations and their respective controls or between alginate treatments

[b]n = 10 carcass sides/treatment

[c]N.D. = not determined

Table 5. Mean values[a] for postmortem shrinkage (%) of pork carcasses with Ca-alginate film.

Hour postmortem	Shrinkage treatment[b]	
	Control	Ca-alginate[c]
24	2.02a	1.53b

[a]Means bearing different letters differ significantly ($P < 0.01$)

[b]n = 10 carcasses (control), 15 carcasses (treatment)

[c]131 grams sodium alginate-maltodextran/liter water

Table 6. Mean values[a] for postmortem shrinkage (%) of naked vs Ca-alginate coated pork carcasses.

Hour postmortem	Shrinkage treatment[b]	
	Control	Ca-alginate[c]
24	2.76a	2.17b
48	3.40a	2.83b
72	3.58a	2.95b

[a]Means on the same horizontal line with different letters differ significantly ($P < 0.05$)

[b]n = 5 carcasses/treatment

[c]98 grams sodium alginate-maltodextran/liter water

tions of alginate-maltodextran were evaluated simultaneously (Table 7), no differences ($P > 0.01$) between the three concentrations were observed in the initial 24 hr chill; however all showed significantly ($P < 0.01$) less moisture loss than the control group.

Data from the beef carcass shrinkage experiments indicated that the 98 g/l alginate-maltodextran allowed the lowest numerical percent shrinkage, however this was not true for pork carcasses. One possible reason was that these carcasses appeared to be exceptionally prone to increased shrinkage, with nearly 4% being lost within 24 hours.

Properties of Ca-alginate Films Relating to Shrinkage

Commercial alginates are largely copolymers of polymannuronic and/or polyguluronic acid (Anonymous, 1973). Recent studies by Morris (1973) using computer model building and X-ray diffraction of alginate fibers revealed that polyguluronic acid chains can adopt a 'zig-zag' shape with hydrophilic 'nests' which accomodate a calcium ion. These nests combine with other chains to form a 'microcrystalline bundle'. Overall, the calcium polyguronate becomes linked by segments of polymannuronate and heteropolymeric acids in normal random solution conformation holding approximately a hundred times its own weight of water.

For the effect of alginate concentration on controlling moisture loss in beef and pork carcasses, there appears to be some differences between the three concentrations with respect to reducing moisture loss, although not always significant. From initial observations, it appeared that the 142 g/l alginate would have provided a better reduction of moisture loss from the carcass. However, the amount of water held in the gel influences the total moisture lost from the carcass.

Table 7. Mean values[a] for postmortem shrinkage (%) of pork carcasses with different concentrations of alginate-maltodextran

Hour postmortem	Shrinkage treatment[b]			
	Alginate-maltodextran[c]			
	Control	98	120	142
24	3.87a	3.20b	3.15b	2.93b
48	3.93a	3.36b	3.19bc	2.93c

[a]Means on the same horizontal line with different letters differ significantly ($P < 0.01$)

[b]n = 8 carcasses/treatment

[c]Grams sodium alginate-maltodextran/liter water

Laboratory experiments evaluating the gelling characteristics of these concentrations are presented in Table 8. Data indicate that more water was held within the gel when the alginate concentration was 98 g/l than when it was 142 g/l. The trapping of water in the gel, especially at low calcium chloride concentrations, permits the gel to act as the sacrificing agent instead of the carcass tissue.

The manner in which Ca-alginate gel may act as the moisture sacrificing agent on the carcass, especially during the initial chilling phase, can be seen in Figure 2. Using glass plates coated with Ca-alginate (142 g/l), stored at 2 C in the chill cooler, trapped water was rapidly released from the gel during the first 24 hr (55%). This rapid release of moisture from the gel accounts for the significant decrease in moisture loss which occurred in the lamb, beef and pork carcasses when they were coated with the Flavor-Tex film. Continued release of moisture through 72 hr storage results in essentially moisture-free Ca-alginate.

Oxygen Permeability of Ca-alginate and Plastic Wrap Films

Using the apparatus (Figure 1) constructed for measuring oxygen permeability through the Ca-alginate and plastic wrap films, data presented in Figure 3 indicate that the permeability of the Ca-alginate gel to oxygen was greatest at the lower (98 g/l) concentration and decreased with increasing alginate-maltodextran (142 g/l). The alginates, in general, were more permeable to oxygen than the plastic Borden Resinite-90 and Goodyear Prime-Wrap films, both high oxygen low moisture transfer wraps.

Table 8. Percent water held in various concentrations of sodium alginate-maltodextran and calcium chloride gels.

Alginate[a]	Calcium chloride (M)			
	0.1	0.3	0.5	0.8
98	92.33[b]	91.01	90.58	86.43
120	91.87	90.44	89.18	84.79
142	88.70	87.66	86.37	84.34

[a]Grams sodium alginate-maltodextran/liter water

[b]Based on gel weight less dry gel weight/gel weight x 100

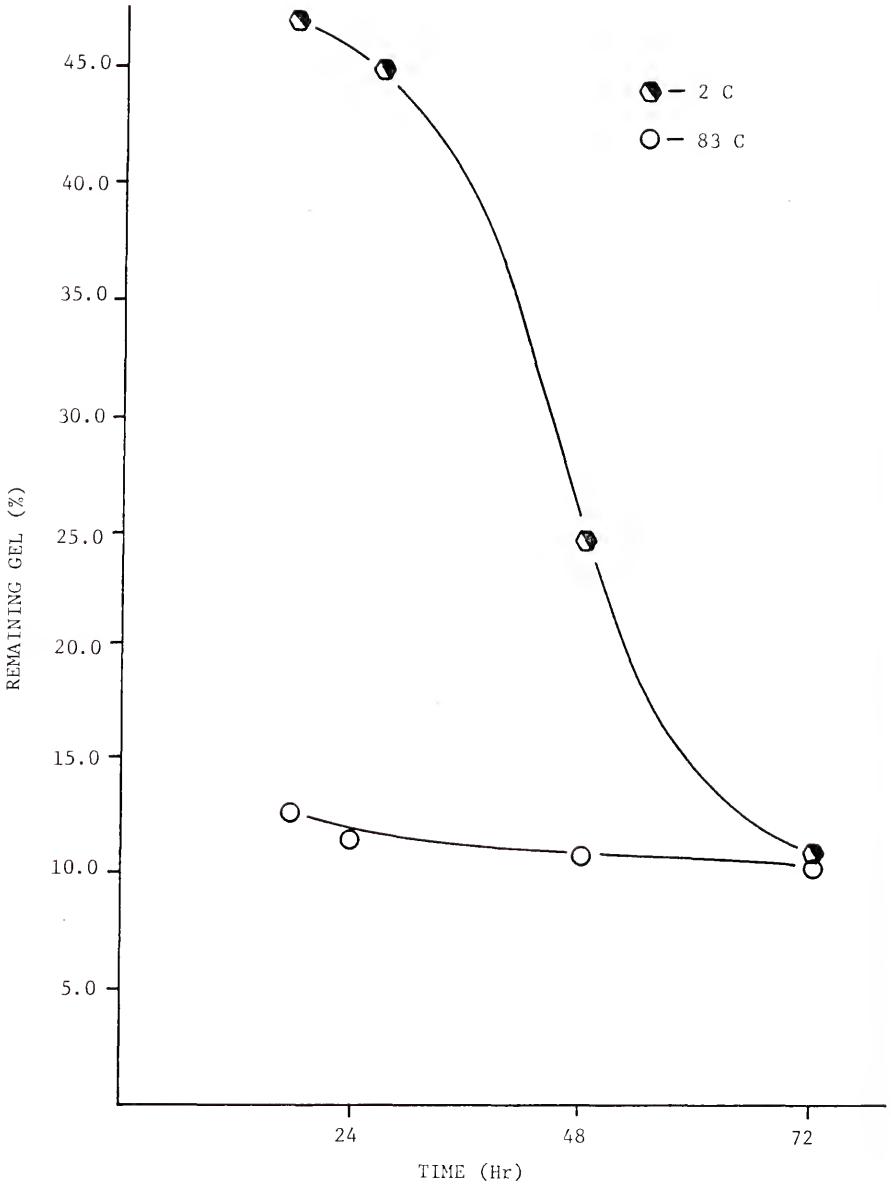

Fig. 2. Release of moisture from Flavor-Tex alginate film at 2 and 83 C.

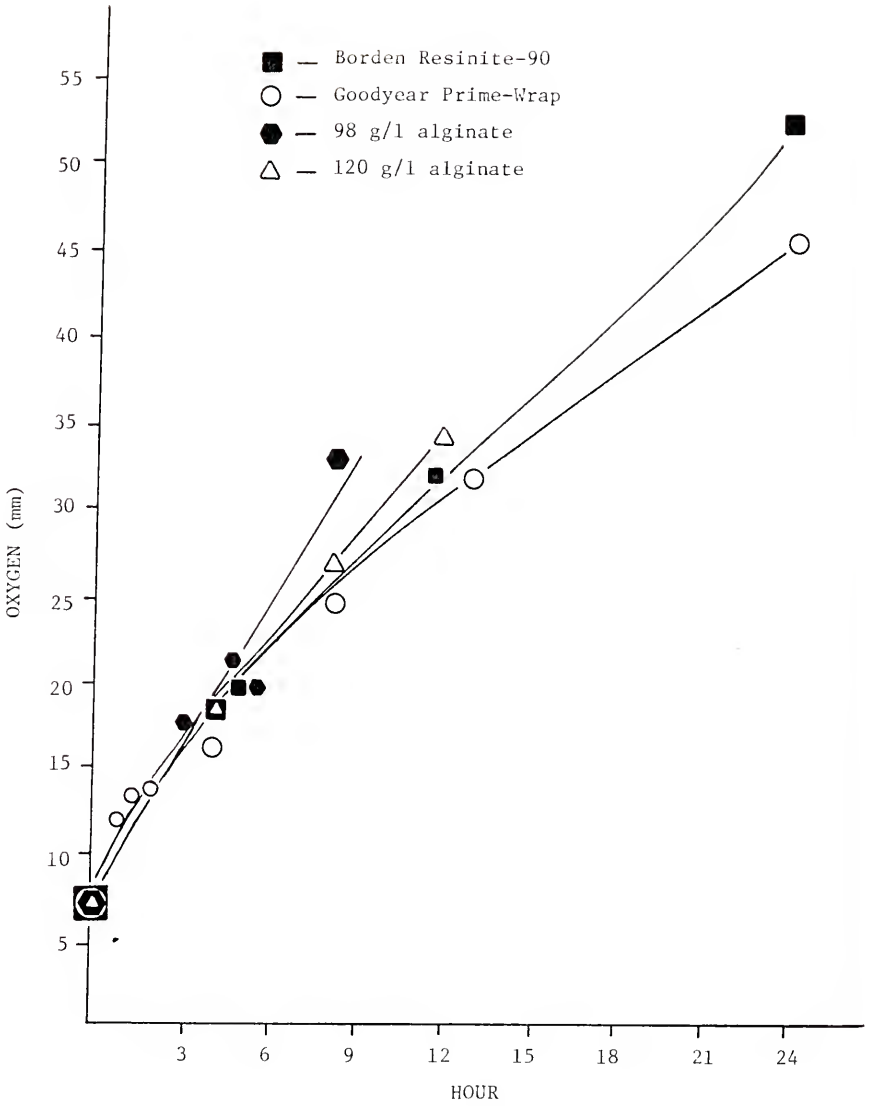

Fig. 3. Oxygen permeability of Ca-alginate films and plastic wraps at 2 C.

The increasing permeability of the Ca-alginate films can be attributed to the release of moisture from the film, thereby permitting the oxygen to pass through more readily. This increased permeability reduces the concern that lean tissue would appear too dark in color due to the reduced state of myoglobin or that anaerobic bacteria could grow if provided additional favorable conditions of increased temperatures.

Decontamination of Meat Carcass Surfaces

In addition to the control and reduction of carcass shrinkage, the meat processor and consumer have become more aware of the presence of undesireable bacteria which can cause a rapid alteration of the acceptable quality of the meat.

In order to reduce and control surface microbial flora, treatment of carcasses with antimicrobial agents in conjunction with good processing procedures should be used as the initial step toward reducing this surface flora.

As presented earlier, the Flavor-Tex alginate coating was effective in reducing shrinkage losses from lamb, beef and pork carcasses. Additionally, Flavor-Tex as well as true antimicrobial agents were evaluated for their ability to reduce and maintain low surface microbial flora.

Lamb. Flavor-Tex and plastic wrap shrinkage treatment effects on lamb surface microbial growth were monitored by swabbing the sirloin and belly (flank-plate juncture) areas of each carcass at 0, 2, 5 and 7 days postmortem. These areas were selected to represent areas which are covered predominantly with fat (sirloin) or with lean (belly). Mean values for total microbial count/6.46 cm^2 from both sampling areas are

presented in Table 9. In the sirloin area, no significant differences were observed between treatment groups immediately postmortem. At 2 days postmortem, those carcasses which had been wrapped with the plastic wrap had higher ($P < 0.05$) microbial counts ($\log_{10} = 3.65$) than did the control ($\log_{10} = 3.04$) or alginate treated ($\log_{10} = 2.87$) carcasses. This difference was maintained through day 5 and day 7 with the plastic wrapped carcasses at day 5 having a higher ($P < 0.05$) microbial count than either control or alginate-treated carcasses. Elevated microbial counts from the plastic wrapped carcasses probably were due to the reduction in surface evaporation thereby maintaining a more favorable water activity (a_w) for growth. The calcium alginate-coated carcasses tended to have lower surface microbial counts from the sirloin area at all time periods, although not always significant. Inhibition of microbial growth by the Flavor-Tex coating may be due partially to the ionic effect of $CaCl_2$ (0.8 M) which was used for the gelling of the alginate on the carcass surface.

Microbial counts from the belly area were not significantly different among treatments at all intervals evaluated (Table 9). The high counts and lack of significant differences were probably due to cross-contamination that occurred during the weighing of the carcasses.

Beef. Although there is wide variation in the bovine animal with respect to type of feed consumed, geographical region in which it is raised and the slaughter processing technique employed, the initial surface microbial flora and chill cooler flora appear to be rather constant. Data presented in Table 10 confirm the report by Locker et al. (1975) that the predominant genera of bacteria on the beef carcass after washing are *Micrococcus*, *Flavobacterium* and *Acinetobacter*. The presence

Table 9. Mean $\log_{10}$ values[a] for total microbial count/6.46 cm^2 from sirloin and belly areas of control and treated lamb carcasses.

Postmortem	Shrinkage treatment[b]		
	Control	Ca-alginate[c]	Plastic wrap
		Sirloin area	
0	3.64a	3.75a	3.86a
2	3.04a	2.87a	3.64b
5	3.47a	2.80b	3.82c
7	3.45ab	3.11b	4.14a
		Belly area	
0	3.90a	3.90a	4.03a
2	3.84a	3.99a	4.09a
5	4.24a	4.34a	4.31a
7	4.47a	4.46a	4.01a

[a]Means on the same horizontal line bearing different letters differ significantly ($P < 0.05$)

[b]n = 30 carcasses/treatment

[c]142 g sodium alginate-maltodextran/liter water

Table 10. Beef carcass surface microbial flora and their relative percentages at various time periods post-slaughter.

Hour postmortem					
0	%[a]	24	%	96	%
Micrococcus	35.1	*Micrococcus*	33.3	*Micrococcus*	83.3
Flavobacterium	18.9	*Acinetobacter*	22.2	*Acinetobacter*	8.3
Acinetobacter	10.8	*Staphylococcus*	22.2	Yeasts	8.3
Staphylococcus	5.4	*Flavobacterium*	11.1		
Pseudomonas	5.4	*Enterobacteriaceae*	11.2		
Lactobacillus	5.4				
Streptococcus	5.4				
Enterobacteriaceae	2.7				
Yeasts	10.8				

[a]Percent of total microorganisms identified

of yeasts, which may vary with the type of diet, constituted 11% of the total initial microbial flora. Following a 24 hr chill, little change occurred with respect to the relative percentages of bacteria present. However, after 96 hr postmortem, *Micrococcus* was most prevalent with *Acinetobacter* and yeasts being the only other organisms isolated. The yeasts present were visually and morphologically similar to the genera *Rhodotorula* and *Candida*.

The control and reduction of these microorganisms which have been enhanced by the reduction of a_w and temperature environments becomes one of the important criteria in the overall decontamination of beef carcass surfaces. In order to effectively apply decontamination processes which would be acceptable to governmental agencies and the meat industry the process must meet basic criteria. Some of these criteria are: nontoxic to humans, no affect on taste of meat, inexpensive, easy to apply, stable and effective in reducing and maintaining low surface microbial levels.

A review of the literature revealed that one of the most practical and inexpensive antimicrobial agents was hypochlorous acid (HClO). Enhanced antimicrobial activity from low levels (5-200 ppm) of hypochlorous acid can be obtained when the pH of the solution is maintained between 4-6 (Charlton and Levine, 1937). However, the stability of the acid decreases rapidly, resulting in a reduction of antimicrobial activity, as the hydrogen ion concentration increases.

The development of an effective decontamination process for meat carcasses which can be applied in the meat industry is described in this series of experiments. The basic process to be presented involves the buffering of hypochlorous acid in an organic solution so that stability of the active component (HClO) can be maintained, thereby permitting it to react effectively with, and reduce, the surface microbial flora.

The buffer system found to be most effective in maintaining HClO stability was acetate-acetic. Other buffers evaluated were Citric-Citrate, Sorbic-Sorbate and Tris(Hydroxymethyl-aminomethane). Data presented in Table 11 indicate that 0.01 M, pH 4.5 acetate-acetic acid buffer in conjunction with 150 ppm available chlorine as HClO (Treatment 1) reduced ($P < 0.01$) and maintained low surface microbial flora through 96 hr postmortem. Addition of 0.001% Tween-80 (polyoxyethylene sorbitan monooleate) (Treatment 2), a surface tension reducer, did not increase the overall antimicrobial activity, however, the mean microbial count at 96 hr for this treatment was lower than the 24 hr count, whereas all other treatments had increased microbial counts at this time period. Spraying of the buffer-HClO solution through the shroud cloth (Treatment 3) appears to have impeded the solution from reaching the carcass surface even though the solution was applied at a pressure of 1 kg/cm^2. When 24 hr microbial counts from all treatments were analyzed for significance (column), the control, Tween-80 and shroud cloth treatments were higher ($P < 0.01$) than the acetate buffer-HClO treatment (2), indicating that even though the effects of chill temperature and water activity reduced the total surface flora, treatment with acetate buffer-HClO further reduced the number of viable organisms.

Because the addition of Tween-80 reduced the flora at 96 hr, the treatment was repeated and incorporated into a second experiment designed to determine whether increased molarity of the acetate-acetic buffer could influence the surface microbial flora. Additionally, sorbate-sorbic acid at 0.1 M, pH 5.5, was evaluated for its effectiveness in reducing yeast microbial flora which was observed to be frequently present following hypochlorous acid treatment. The results

Table 11. Mean $\log_{10}$ values[a] for total microbial count/6.46 cm^2 from the neck area of control and acetate buffer-HClO treated beef carcasses.

Treatment[c,d]	Hour postmortem		
	0[b]	24	96
Control	3.30a	2.80b	3.14ab
1		2.09b	2.89c
2		2.96ab	2.72b
3		2.53b	2.98a

[a]Means on the same horizontal line with different letters differ significantly ($P < 0.01$)

[b]Pooled response; collected prior to treatment application

[c]Treatments denoted as:
Control = untreated
1 = 0.01 M acetate-acetic, pH 4.5 + 150 ppm available chlorine
2 = same as treatment 1 + 0.001% Tween-80
3 = same as treatment 1 except sprayed through the shrouded carcass

[d]n = 20 carcass sides/treatment

from this experiment are presented in Table 12. The presence of the surfactant (Tween-80) in the acetate buffer-HClO solution reduced ($P < 0.01$) the surface microbial flora again but was not found to be superior to other treatments at the 96 hr storage period. Use of 0.1 M acetate-acetic-HClO did not result in greater surface microbial flora reduction than 0.01 M acetate-acetic-HClO nor did the sorbate-sorbic buffer provide an improved response. Evaluation of microbial colonies from the agar plates indicated no noticeable reduction in the number of yeasts. In addition to its apparent failure to reduce the yeast flora, the sorbate-sorbic buffer had a precipitate due to insolubility of sorbic acid at this pH. Consequently, when sprayed onto the carcasses, flecks of the chemical could be seen on the surface. Analysis of the mean values from each treatment at 24 hr postmortem (column) indicated that the 0.01 M acetate buffer-HClO-Tween-80 treatment and the 0.1 M acetate buffer-HClO treatment were lower ($P < 0.01$) in total surface microbial flora than the control or sorbate-sorbic buffer treated carcasses. Although effective, the 0.1 M acetate-acetic buffer treatment does not appear to be superior to the 0.01 M acetate-acetic buffer. At this concentration, the buffer had a pungent odor which persisted on the carcass for 4-6 hours.

Interim USDA guidelines suggest the use of hypochlorous acid, with a maximum of 200 ppm available chlorine, as a surface rinse or spray; thus an experiment was conducted to evaluate various concentrations of hypochlorous acid. The buffer system to which the hypochlorite solution was added was 0.01 M acetate-acetic, pH 4.5. Results presented in Table 13 indicate that a reduction ($P < 0.01$) in surface microbial flora occurred equally between 25 and 200 ppm available chlorine at both 24 and 96 hr postmortem.

Table 12. Mean $\log_{10}$ values[a] for total microbial count/6.46 cm^2 from the neck area of control and treated beef carcasses.

Treatment[c,d]	Hour postmortem		
	0[b]	24	96
Control	3.52a	2.57b	3.10ab
1		1.70b	1.80b
2		1.85b	1.55b
3		2.22b	1.90b

[a]Means on the same horizontal line with different letters differ significantly ($P < 0.01$)

[b]Pooled response; collected prior to treatment application

[c]Treatments denoted as:
Control = untreated
1 = 0.01 M acetate-acetic, pH 4.5 + 150 ppm available chlorine + 0.001% Tween-80
2 = 0.1 M acetate-acetic, pH 4.5 + 150 ppm available chlorine
3 = 0.1 M sorbate-sorbic, pH 5.5

[d]n = 15 carcass sides/treatment

Table 13. Mean $\log_{10}$ values[a] for total microbial count/6.46 cm^2 from the neck area of control and acetate buffer-HClO treated beef carcasses.[b]

Hour postmortem	Control	Available chlorine[d]		
		25	100	200
0[c]	3.73			
24	3.13a	2.62b	2.31b	2.21b
96	3.53a	2.72b	2.33b	2.52b

[a]Means on the same horizontal line with different letters differ significantly ($P < 0.01$)

[b]n = 15 carcass sides/treatment

[c]Pooled response; collected prior to treatment application

[d]Parts per million available chlorine suspended in 0.01 M acetate-acetic buffer, pH 4.5

The reaction of hypochlorous acid with microorganisms involves the oxidation of proteinaceous material (Anonymous, 1964). Experiments conducted by Johns (1934) revealed that bacteria (*Staphylococcus aureus*, 1×10^6 cells/ml) were destroyed within 5 minutes by 10 ppm available chlorine in nutrient broth. With adequate washing of the beef carcass surface, the remaining microbial flora should be effectively inhibited by small concentrations of the highly active HClO. Therefore, the concentration which should be applied to carcass surfaces will depend partially upon the thoroughness of the final wash water treatment. Use of any antimicrobial agent is not meant to replace sanitation and hygiene treatments of the carcass, but rather to aid in reducing the remaining surface flora so that ultimately an increased shelf-life of the fresh meat can be obtained. The use of the lowest effective concentration will help to ensure that adequate wash water treatment will be conducted and that the chance for secondary HClO reactions involving the oxidation of organic matter, which may ultimately be consumed, will be essentially non-existent.

As a comparison to the calcium hypochlorite method of forming hypochlorous acid, the Morton Biocidal System (Model 110-415 1D) using chloride ion electrolysis to form chlorine (Cl_2), followed by reaction in acidic water to form hypochlorous acid was evaluated. Additionally, the relative time required to observe a significant decrease in the surface microbial count was determined. These data are presented in Table 14. At 1 hr post-treatment, no differences ($P > 0.05$) were observed between the control and treated carcasses. However, by 12 hr post-treatment, the 50 ppm available chlorine treatment carcasses had lower ($P < 0.05$) microbial counts than the control carcasses. The 170

Table 14. Mean $\log_{10}$ values[a] for total microbial count/6.46 cm^2 from the neck area of control and treated[b] beef carcasses.[c]

Hour post-treatment	Available chlorine (ppm)		
	Control	50	170
0[d]	3.05		
1	2.82a	2.26a	2.39a
12	2.38a	1.69b	2.07ab

[a]Means on the same horizontal line with different letters differ significantly ($P < 0.05$)

[b]Morton Biocidal Flow-Thru Design Unit Model 110-415 1D

[c]n = 10 carcass sides/treatment

[d]Pooled response; collected prior to treatment application

ppm available chlorine treatment carcass counts were similar to the control and 50 ppm treatment carcasses. Ram Ayar (1930) observed that a concentration of 20 ppm available chlorine was more effective than 100 or 200 ppm against spores of *Bacillus subtilis*. Johns (1934) also observed increased efficiency on dilution of sodium hypochlorite solutions and attributed the results to the increase in hydrogen ion concentration. Kotula (1975) reported that 30 ppm chlorinated water was equally as effective in reducing surface microbial flora on beef carcasses as was 95 ppm.

Pork. One experiment was conducted on the decontamination of pork carcasses. This experiment involved the addition of hypochlorous acid directly to the alginate-maltodextran or calcium chloride solution. Early interests were to mix the hypochlorous acid and alginate together so that they could be sprayed simultaneously, however a basic problem due to the pH of the two components existed. Because alginate had a pH of 7.1 and calcium chloride a pH of 6.7, addition of the hypochlorite ion would not permit the active hypochlorous acid to completely form. When the alginate pH was decreased with organic or inorganic acids, a gel formed. However, by adjusting the water and hypochlorite to pH 6.1-6.3 with acetic acid, followed by the addition of the alginate, a relatively soluble mixture could be obtained. No difficulties were encountered with the acidification of the calcium chloride solution.

When these treatment preparations were sprayed onto pork carcasses and analyzed for surface microbial reduction (Table 15), no differences ($P > 0.05$) among the five treatments occurred. The lack of a significant decrease in microbial counts may have been due to the initial low numbers of surface flora. Also, there was no significant increase in

Table 15. Mean $\log_{10}$ values[a] for total microbial count/6.46 cm^2 from the shoulder area of control and treated pork carcasses.

Treatment[b,c]	Hour postmortem			
	0[d]	24	48	96
Control	2.82	2.39	2.64	2.87
1		2.36	2.52	2.49
2		2.76	2.61	2.55
3		2.23	2.39	2.39
4		2.76	2.32	2.57

[a]Means on the same horizontal line are not significantly ($P > 0.05$) different

[b]Treatments denoted as:
Control = untreated
1 = 50 ppm available chlorine rinse, pH 6.3
2 = 0 ppm alginate film coating
3 = 50 ppm available chlorine suspended in sodium alginate (Solution 1)
4 = 50 ppm available chlorine suspended in calcium chloride (Solution 2)

[c]n = 5 carcasses/treatment

[d]Pooled response; collected prior to treatment application

the flora after 96 hr postmortem as was observed in all previous experiments. With pork carcass skin surfaces being dry, smooth, firm and rather free of available nutrients, the opportunities for increasing surface microbial growth appear remote, especially at chill cooler temperatures.

The surface microbial flora, which was identified from the control (0 hr) and Ca-alginate (48 hr) treated carcasses is presented in Table 16. The flora was essentially the same as the beef carcass (Table 10) at similar time periods. The somewhat increased staphylococci percentage may be an indication of the greater human handling and manual processing that occurs for these carcasses during the slaughter process.

Reduction of lamb surface microbial flora has been demonstrated through application of the Flavor-Tex alginate coating. The application of this film coating in addition to reducing carcass moisture loss can provide some antimicrobial activity. This reduction in surface flora is thought to be due to the ionic effect of calcium chloride. Use of a plastic wrap allowed for an increase in surface flora by retaining moisture and heat from the carcass. These changes resulted in an increase in the surface microbial flora through 7 days postmortem.

The data presented in the beef carcass decontamination experiments demonstrate that a practical process employing low concentrations of available chlorine (25-100 ppm) in combination with 0.01 M, pH 4.5 acetate-acetic acid buffer can effectively reduce and maintain low levels of surface microbial flora. This process incorporates the fundamental criteria outlined previously concerning the requirements for an antimicrobial agent that can be acceptable to governmental agencies and the meat industry. Application of this process not only to the

Table 16. Pork carcass surface microbial flora and their relative percentages following treatment with a Ca-alginate coating.

Hour postmortem			
0		48	
Control	%[a]	Ca-alginate	%[a]
Micrococcus	20	*Staphylococcus*	38
Staphylococcus	15	*Flavobacterium*	23
Moraxella	10	*Pseudomonas*	23
Acinetobacter	10	*Micrococcus*	8
Flavobacterium	10	Yeasts	8
Enterobacteriaceae	10		
Pseudomonas	5		
Streptococcus	5		
Yeasts	15		

[a]Percent of total microorganisms identified

carcass immediately post-slaughter, but to wholesale and retail cuts could help to increase the shelf-life and overall acceptance of the particular meat product.

Addition of hypochlorous acid to the alginate film solutions prior to spraying onto pork carcasses did not reduce surface microbial flora; nor did it influence the type of flora remaining on the carcass when compared with untreated controls. Although carbohydrates normally do not interact with hypochlorous acid, the use of 50 ppm HClO may have interacted with the alginate sufficiently to become inactive.

The application of low concentrations of hypochlorous acid, suspended in the acetate-acetic acid buffer, followed by the application of an alginate film at a concentration of approximately 100 g/l could result in a significant reduction in both surface microbial flora and moisture loss from the meat carcass.

SUMMARY

Lamb, beef and pork carcasses were evaluated for moisture loss (shrinkage) procedures by employing Ca-alginate (Flavor-Tex) edible films and/or plastic wrap (Borden Resinite-90). For lamb carcasses, shrinkage was best controlled by wrapping the carcass in plastic wrap, however increased microbial growth was observed through 7 days postmortem. The Ca-alginate film significantly reduced shrinkage and surface microbial flora through 5 days postmortem.

Beef carcass shrinkage, using the cotton shroud cloth, was not significantly reduced when compared with naked carcasses, however the shroud provided the carcass with a smooth and more uniform appearing surface.

Use of Ca-alginate films significantly reduced beef carcass shrinkage through 96 hr storage when compared with shrouded control carcasses. Application of various concentrations of the sodium alginate-maltodextran solution resulted in a similar response in the reduction of beef carcass shrinkage. Data indicated that when low concentrations of the alginate solution were employed, water molecules were more easily held within the Ca-alginate gel than when higher concentrations were used. The trapping of water within the gel allows the alginate film to act as the moisture sacrificing agent during the initial 72 hr chill cooler storage period.

Oxygen permeability studies of the Ca-alginate and plastic wrap

films indicate that the Ca-alginate film impeded the flow of oxygen through the film less than the plastic wrap, Resinite-90.

Pork carcass shrinkage, which approached 4% within 24 hr portmortem, was significantly reduced through 72 hr storage by application of the Ca-alginate film. Use of various concentrations of sodium alginate-maltodextran resulted in no significant differences between treatments, but all concentrations significantly reduced moisture loss when compared with untreated controls.

Surface microbial flora on beef carcasses was significantly reduced through 96 hr chill cooler storage by the application of low concentrations (25 ppm) of chlorine as hypochlorous acid. The optimum buffer system in which to suspend the hypochlorous acid was observed to be 0.01 M, pH 4.5 acetate-acetic acid. No significant differences in surface microbial growth were observed between the 25 and 200 ppm available chlorine treatments.

The addition of 50 ppm available chlorine to either Flavor-Tex solutions resulted in no significant reduction of pork carcass surface microbial flora nor did the alginate influence the selection of microorganisms on the carcass after 48 hr postmortem storage.

The application of low (25-100 ppm available chlorine) concentrations of hypochlorous acid, suspended in a weak acetate-acetic acid buffer can significantly reduce and maintain low surface microbial growth. The application of this decontamination process followed by the application of an alginate film at a concentration of approximately 100 g/l and 0.3 M calcium chloride could result in a significant reduction in both surface microbial flora and moisture loss from the meat carcass.

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BIOGRAPHICAL SKETCH

Charles Raphael Lazarus was born in Elkins, West Virginia on January 28, 1940, son of Maurice J. and Jeanette A. Lazarus. He received his primary education from Kruger Street and Bridge Street School, and graduated from Triadelphia High School, Wheeling, West Virginia in May 1957. He entered the United States Air Force in March 1960, and trained as a Medical Laboratory Technician. He was honorably discharged in January 1964. He entered West Virginia University in January 1964, and received his Bachelor of Arts degree in Biology in June 1966. In September 1966 he began studying for the Master of Science degree in Bacteriology at the same university and obtained this degree in May 1969. Following employment as a microbiologist, he entered the University of Florida Graduate School in June 1974 and is a candidate for the degree of Doctor of Philosophy.

The author is a member of Alpha Zeta, Sigma Nu, American Society for Microbiology, American Society of Animal Science, Institute of Food Technologists, American Association for the Advancement of Science and American Meat Science Association. He has a daughter Amy by a previous marriage.

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.

A.Z. Palmer, Chairman
Professor of Animal Science

I certify that I have read this study and that in my opinion it conforms to acceptable standards of scholarly presentation and is fully adequate, in scope and quality, as a dissertation for the degree of Doctor of Philosophy.

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August, 1976

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