

DISTRIBUTION PACKAGING METHOD AND STORAGE TIME EFFECTS ON THE MICROBIOLOGICAL CHARACTERISTICS AND INCIDENCE OF THE PATHOGENS *LISTERIA MONOCYTOGENES* AND *SALMONELLA* IN PORK¹

M. F. MILLER^{2,6}, M. A. CARR², A. R. SCHLUTER³, D. K. JONES⁴,
M. K. MEADE⁵ and C. B. RAMSEY²

²*Department of Animal Science and Food Technology
Texas Tech University
Lubbock, Texas 79409-2162*

Accepted for Publication November 11, 1995

ABSTRACT

*Microbiological and yield characteristics were determined on bone-in pork loins and Boston butts (n = 65 each) that were selected from a commercial facility and subjected to one of three packaging treatments: (1) paper wrapped, (2) modified atmosphere packaging (66% O₂, 2.26% CO₂, and 8% N₂), and (3) vacuum packaging. Cuts were stored up to 21 days at 0 ± 2C for yield characteristics and an added 28 and 35 days for microbiological characteristics. Treatment and storage effects on the incidence of the pathogens *Listeria monocytogenes*, *Salmonella* and numbers of aerobic bacteria, lactic acid bacteria and coliforms were determined. The amount of purge was variable (100 to 500 g) among packaging treatments. The vacuum packaged and modified atmosphere packed pork loins and butts had lower aerobic plate counts (P < .05) compared with the paper wrapped loins and butts. The numbers of *Listeria* species decreased at a greater rate for the vacuum packaged and modified atmospheric packaged pork loins compared with the paper wrapped loins. No *Salmonella* were found on meat from any packaging treatment or storage time. The microbial quality of pork loins and butts can be improved by using vacuum packaging compared with paper wrapping or modified atmosphere packaging.*

¹Mention of trade name, propriety product or specific equipment does not constitute a guarantee or warranty of the product by Texas Tech University and does not imply approval to the exclusion of other products that also may be suitable.

³Schluter Farms, Rt.1, Box 40, Rhome, TX 76078

⁴Monfort Inc., P. O. Box G, Greeley, CO 80623

⁵Texas Beef Industry Council, 8310 Capita Hwy. North, Suite 440, Austin, TX 78731

⁶Please direct inquiries to: Dr. Miller. Tel. No.: 806-742-2804, FAX: 806-742-0169, E-mail: mfmraider@aol.com

INTRODUCTION

The beef industry has utilized vacuum-packaging for many years to ensure maximum shelf life of products. The pork industry, however, has traditionally used other packaging methods. Three packaging methods [fresh paper wrap, modified atmosphere packaging (MAP), and vacuum packaging] for fresh pork loins are available for use in industry.

Almost 60% of the fresh pork distributed in the United States was overwrapped with paper (Moore 1989). Vacuum-packaging requires special packaging materials and equipment and the creation of a partial vacuum within the package before it is sealed.

Less than 20% of the fresh pork distributed in the United States was vacuum packaged in 1989 (Moore 1989), although this percentage increases annually. Today, approximately 40% of fresh pork is vacuum packaged (Jones 1995). Gas-flush packaging requires a modified atmosphere containing various levels of oxygen (O₂), carbon dioxide (CO₂) and nitrogen (N₂). Gas mixtures for meat packaging consist of CO₂, which inhibits spoilage by aerobic bacteria, O₂, which helps keep meat surface pigments oxygenated, and N₂, which is used primarily to dilute the concentration of CO₂ and O (Seideman *et al.* 1979). Many studies have focused on the use of modified gaseous atmospheres to limit microbial growth (Seideman and Durland 1984). To increase the shelf life of fresh pork, most packers are trying to adopt vacuum or MAP techniques.

Because of increased media coverage and consumer awareness, food safety issues have become important in the meat industry. Todd (1989) estimated 12.6 million cases of foodborne illness/year costing \$8.4 billion/year. Microbiological diseases represent 84% of the United States cost, with salmonellosis and *Staphylococcus* intoxication being the most economically important diseases (Todd 1989). Estimates of human foodborne salmonellosis in the United States vary from 740,000 to 5,300,000 cases annually (Flowers *et al.* 1992).

Muscles of animals are sterile (Bell *et al.* 1986). As an unavoidable result of processing, microbiological contamination occurs during the conversion of live animals to meat for consumption (Dickson 1992). Although the majority of the microflora transferred to tissue surfaces are nonpathogenic (Dickson 1992), pathogens such as *Salmonella* (Currier *et al.* 1986) may be transferred infrequently to carcasses during slaughter and packing house operations. *Salmonella*, *Escherichia coli* 0157:H7, *Listeria monocytogenes*, *Staphylococcus aureus*, *Colstridium perfringens*, *Campylobacter jejuni*, and *Yersinia enterocolitica* are pathogens that have been associated with meat and meat products (Meat and Poultry Inspection 1985).

The purpose of this study was to evaluate the effects of packaging method (paper wrapping, MAP, and vacuum packaging) on purge yield, microbiological characteristics, and the percentage incidence of the pathogens *Listeria monocytogenes* and *Salmonella* in pork loins and butts.

MATERIALS AND METHODS

Selection, Fabrication and Packaging

Sixty-five bone-in pork loins and 65 Boston butts were selected from carcasses in the same contemporary group from a commercial pork processor (same producer and sex of pig and similar age of pig) to decrease the effects of genetic variation in the pigs. The pigs were slaughtered under normal industry conditions in a large midwestern packing plant. The loins and Boston butts were fabricated to current Institutional Meat Purchasing Specifications (IMPS #127 and #130) after chilling the carcasses for about 24 h at 0C. Each loin and butt was randomly assigned to one of three packaging treatments: (1) paper overwrap, (2) vacuum packaging, or (3) MAP. Paper overwrap was applied by accepted industry techniques using one sheet of G-8 Deli paper (Papercon, Omaha, NE). The vacuum-packaging was done with a CVP Fresh-Pac Model 8600 machine (Cryovac, Inc., Duncan, SC). The gas-flush packaging was done with a CVP Fresh-Pac Model A-200 machine (Cryovac, Inc., Duncan, SC) producing an atmosphere of 66% O₂, 26% CO₂ and 8% N₂. All loins (MAP) were boxed (N = 5/box) and placed in heat sealable bags with the following characteristics: oxygen transmission rate = 32 cc m²/24 h/24C/50% RH, moisture vapor transmission rate = 0.8 - 1.9 g/m²/24 h/37.7C/70% RH and carbon dioxide transmission rate = 47 cc/m²/24 h/24C/50% RH. Loins in the MAP treatment were injected with 500 cc of gas mixture (head space to meat volume ratio of 1:1). Specific percentages were chosen by compiling results of Seideman *et al.* (1979) that indicated this composition was optimal for maintaining desirable color and low microbial counts. After the loins and butts were packaged and boxed at the slaughter and fabrication plant, they were trucked immediately after fabrication for 36 h at 3±1C to the Texas Tech University Meat Science Laboratory where they were stored at 2±1C until 7, 10, 14, 21, 28 or 35 days postmortem storage time (including the 36 h trucking time) elapsed, depending on the treatment.

Microbiological Analyses

Surface tissue (36 cm²) samples (40 g) were aseptically collected, using sterile forceps and a scalpel, from each treatment group on one of two days after 0, 7, 10, or 14 days of storage depending on the treatment. The duplicate samples were removed from the surface of each loin and butt and placed in sterile plastic bags.

The meat samples were homogenized in 180 mL of Butterfield's phosphate

buffer (BBL, Cockeysville, MD) for 2 min with an Osterizer blender. The blender was sanitized with 77C hot water between each sample. The samples were evaluated for the presence of *Salmonella* (Poelma *et al.* 1984), aerobic plate counts (APC), lactic acid bacteria, *Listeria* spp., *Listeria monocytogenes* and total coliforms. A 25-g sample was pre-enriched in 225 mL lactose broth (Difco, Detroit, MI) for 24 ± 2 h at 35C (FDA, 1992). Biocontrol™ 1-2 tests (Biocontrol/Systems, Botheal, WA) were used to detect the presence or absence of *Salmonella*. *Listeria* spp and *Listeria monocytogenes* were enumerated according to the USDA method for detection of *L. monocytogenes* using MX agar (Modified Oxford medium; Difco, Detroit, MI) and incubated at 35C for 24 and 40 h (Poelma *et al.* 1984). APC were determined by placing 10 mL of the original sample homogenate in 90 mL of Butterfield's phosphate buffer. After blending, 0.1 mL from the appropriate 10-fold dilutions was plated on duplicate pre-poured plates of nutrient agar. The samples were evenly spread on the surface of the plates with a sterile bent glass rod. The plates were incubated at 35C for 48 h to produce clearly defined colonies. Plates with 25 to 250 colonies were counted and reported as the log₁₀ CFU/cm² for each sample. Total coliforms were enumerated by placing 1 mL of each dilution on *Escherichia coli* Petrifilm™ (3M Health Care, St. Paul, MN) and incubating at 37C for 24 h (3M Health Care, St. Paul, MN). Petrifilm™ containing 15 to 150 colonies was counted. Lactic acid bacteria were enumerated using MRS agar (BBL, Cockeysville, MD) incubated at 25C for 48 h. Plates containing 25 to 250 colonies were counted.

Statistical Analyses

Data were analyzed using a split plot arrangement for a 3 (packaging treatment) × 4 (storage time) × 5 (replication) factorial arrangement. Data for percentage purge was collected from five storage times. Main effects and interactions were tested within loin and within Boston butt cuts (Montgomery 1984). When a significant ($P < .05$) F-test was observed, the least squares means were separated by the PDIFF option of SAS (1990).

RESULTS AND DISCUSSION

Purge

As a result of the wrapping paper absorbing much of the purge, the paper wrapped loins and butts did not have measurable purge. The vacuum packaged loins had more purge ($P < .05$) at all four storage times compared with the MAP loins (Fig. 1). The vacuum packaged butts had less purge ($P < .05$) at all four storage times in contrast to the MAP butts. Loins have more surface area per unit of weight than do butts and possibly the difference in surface area produced these

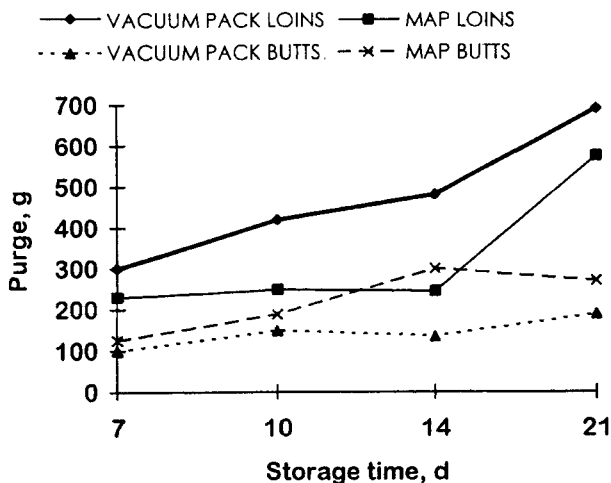


FIG. 1. AMOUNT OF PURGE FROM VACUUM PACKAGED AND MAP PORK LOINS AND BUTTS

results. The higher purge losses would result in greater shrink and value losses for the retailer. Many factors affect the quantity of purge loss. Some reported factors include amount of time between slaughter and fabrication of the animal, degree of vacuity in the package, rigidity of the packaging material, rate of postmortem chilling, extent of temperature fluctuation, and amount of handling and stacking (Cantoni and Bolther 1974; Johnson 1974). Our results indicate that purge will be lower in butts than loins regardless of packaging method and is due to the slower movement of water through the greater number of fat layers on the butts.

Microbiological Analyses

All three packaging treatments decreased ($P < .05$) the incidence of *Listeria* species after 10 days of storage (Table 1). At 10 days of storage, a 57% reduction in incidence of *Listeria* species for paper wrapped, 100% reduction for vacuum packaged, and 84% reduction for MAP pork loins was found. *Listeria monocytogenes* and *Salmonella* were not found on pork loins from any packaging treatment except for the 0 day vacuum packaged loins (5% incidence of *Listeria monocytogenes*).

The percentage incidence of *Listeria* species decreased ($P < .05$) for both paper wrapped and MAP pork butts as storage time increased (Table 1). Vacuum-packaged butts had a lower incidence of *Listeria* species at 0 and 7 days storage

than paper wrapped or MAP butts. The incidence of *Listeria* species was low for all treatments and the lack of *Listeria monocytogenes* in all treatments through 14 days storage shows a low incidence of this pathogen on the pork butts. Manu-Tawiah *et al.* (1993) reported that vacuum packaging was no more effective than gas mixtures in retarding the growth of *Listeria monocytogenes*. The only positive *Salmonella* samples were removed from butts that were vacuum packaged and stored 10 days.

Through 10 days of storage, storage time and packaging method did not affect counts of lactic acid bacteria. The paper wrapped loins had higher ($P < .05$) counts of lactic acid bacteria after 14 days of storage than loins that were subjected to MAP and vacuum packaging (Table 2). A significant increase in counts of lactic acid bacteria occurred between days 10 and 14 of storage. After 21 and 35 days of storage, the MAP and vacuum packaged loins had increased further ($P < .05$) in counts of lactic acid bacteria (data not shown). The increase in lactic acid bacteria could be a result of the selective inhibition of normal spoilage bacteria that is produced by a vacuum or MAP atmosphere (Pierson *et al.* 1970). Vacuum packaging or MAP would decrease the number of lactic acid bacteria competitors because lactic acid bacteria can grow in low oxygen concentrations while other spoilage bacteria cannot. Pierson *et al.* (1990) reported an increase in lactic acid bacteria from 10^3 to $10^8/\text{cm}^2$ when beef was packaged anaerobically for 15 days. Meade *et al.* (1993) obtained lower ($P < .05$) lactic acid bacteria counts after 21 days of storage of MAP pork loins (75% N_2 , 25% CO_2) stored at $0\pm 2\text{C}$ when compared with paper overwrapped loins.

A treatment x storage interaction ($P < .05$) affected APC of pork loins. APC of loins did not differ among packaging methods during the first 7 days of storage. After 10 days of storage, vacuum packaged loins had higher APC than the other loins. However, after 14 days of storage, the vacuum packaged loins had lower ($P < .05$) APC than the other loins, and MAP loins had lower APC than paper wrapped loins. Ordal (1962) determined that increases in total plate counts after 12 days probably were due to increased lactic acid bacteria. The removal of most of the oxygen from the packages and the production of an environment not conducive to aerobic bacteria could have caused the decrease in aerobic bacteria in the vacuum packaged pork loins from 10 to 14 days of storage. Meade *et al.* (1993) obtained reductions ($P < .05$) in aerobic bacteria for MAP pork loins (75% N_2 , 25% CO_2) after 21 days of storage at $0\pm 2\text{C}$.

A significant treatment x storage interaction affected total coliform counts of pork loins. Total coliform counts did not change for the paper wrapped pork loins during 14 days of storage. However, coliform counts were higher ($P < .05$) for vacuum packaged and MAP pork loins after 14 days than after 7 or 10 days of storage.

All lactic acid bacteria counts for pork butts were low and indicate that the fabrication process added few spoilage bacteria. Vacuum packaged butts had the highest lactic acid bacteria counts after 10 days of storage, but by the 14th day the

counts among packaging methods were not different.

A significant treatment x storage interaction affected APC of pork butts in a pattern similar to that found on the loins. The highest counts were found after 10 days of storage for the paper wrapped and MAP butts, but the highest count for the vacuum packaged butts was after 10 days of storage. After 14 days storage, all three packaging treatments for butts had higher ($P < .05$) APC compared with 0 days storage. APC were higher ($P < .05$) for paper wrapped butts compared with vacuum and MAP butts after 14 days storage.

TABLE 1.
INFLUENCE OF PACKAGING METHOD ON THE INDICENCE (%) OF *LISTERIA* SPP.,
LISTERIA MONOCYTOGENES, AND *SALMONELLA* ON PORK LOINS AND BUTTS
DURING STORAGE AT 2C (N=65)

Cut and trait	Treatment	Storage time, d			
		0	7	10	14
Pork loin					
<i>Listeria</i> species	Paper wrap	40 ^{bx}	30 ^{bx}	17 ^{by}	<1 ^z
	Vacuum pack	26 ^{cx}	18 ^{cx}	<1 ^{cy}	<1 ^y
	MAP ^a	31 ^{cx}	<1 ^{dy}	5 ^{cy}	<1 ^y
<i>Listeria</i> <i>monocytogenes</i>	Paper wrap	<1	<1	<1	<1
	Vacuum pack	5	<1	<1	<1
	MAP	<1	<1	<1	<1
<i>Salmonella</i>	Paper wrap	0	0	0	0
	Vacuum pack	0	0	0	0
	MAP	0	0	0	0
Pork butt					
<i>Listeria</i> species	Paper wrap	20 ^{bx}	10 ^{by}	8 ^y	<1 ^z
	Vacuum pack	<1 ^{cy}	<1 ^{cy}	9 ^x	<1 ^y
	MAP	25 ^{bx}	12 ^{by}	7 ^y	<1 ^z
<i>Listeria</i> <i>monocytogenes</i>	Paper wrap	<1	<1	<1	<1
	Vacuum pack	<1	<1	<1	<1
	MAP	<1	<1	<1	<1
<i>Salmonella</i>	Paper wrap	<1	<1	<1	<1
	Vacuum pack	<1	<1	2	<1
	MAP	<1	<1	<1	<1

^aMAP = modified atmosphere packaging.

^{b,c,d}Means in a column within bacterial type with the same or no superscript do not differ ($P > .05$).

^{x,y,z}Means in a row with the same or no superscript do not differ ($P > .05$).

A significant ($P < .05$) treatment x storage interaction also was noted for total coliform counts. Total coliforms were higher before storage than after storage for 10 days if the butts were paper wrapped. Counts were not different from 7 to 14 days of storage. However, for vacuum packaged butts, counts were much higher after 14 days of storage than earlier. Higher counts were found on MAP butts after 10 days of storage than after 0 and 7 days.

TABLE 2.
INFLUENCE OF PACKAGING METHOD AND STORAGE TIME ON LACTIC ACID BACTERIA, AEROBIC BACTERIA AND TOTAL COLIFORMS ON PORK LOINS AND BUTTS DURING STORAGE AT 2C (LOG_{10} CFU/cm², n=65)

Pork cut and trait	Treatment	Storage time			
		0	7	10	14
Pork loin					
Lactic acid bacteria	Paper wrap	3.3 ^b ±0.00	3.3 ^b ±0.00	3.3 ^b ±0.00	3.7 ^d ±0.45
	Vacuum pack	3.3 ^b ±0.00	3.3 ^b ±0.00	3.3 ^b ±0.00	3.5 ^c ±0.34
	MAP ^a	3.3 ^b ±0.00	3.3 ^b ±0.00	3.3 ^b ±0.00	3.5 ^c ±0.34
APC	Paper wrap	3.5 ^b ±0.33	4.1 ^b ±0.23	3.5 ^b ±0.24	7.3 ^d ±0.00
	Vacuum pack	3.9 ^b ±0.60	4.1 ^b ±0.46	6.3 ^c ±0.52	4.0 ^b ±0.71
	MAP	3.8 ^b ±0.55	3.6 ^b ±0.53	3.7 ^b ±0.51	6.4 ^c ±0.61
Total coliforms	Paper wrap	2.4 ^{de} ±0.24	1.9 ^d ±0.02	1.8 ^d ±0.11	1.7 ^{cd} ±0.66
	Vacuum pack	2.3 ^{de} ±0.08	1.1 ^{bc} ±0.27	1.0 ^b ±0.00	2.9 ^e ±0.84
	MAP	2.2 ^{de} ±0.39	2.0 ^d ±0.15	1.8 ^d ±0.23	3.7 ^f ±0.30
Pork butt					
Lactic acid bacteria	Paper wrap	3.45 ^a ±0.21	3.60 ^{ab} ±0.42	3.30 ^a ±0.00	3.45 ^a ±0.21
	Vacuum pack	3.30 ^a ±0.00	3.30 ^a ±0.00	3.99 ^b ±0.29	3.30 ^a ±0.00
	MAP ^a	3.30 ^a ±0.00	3.30 ^a ±0.00	3.45 ^a ±0.21	3.45 ^a ±0.21
APC	Paper wrap	4.3 ^{cd} ±0.12	4.9 ^{de} ±0.77	3.6 ^{bc} ±0.42	7.3 ⁱ ±0.00
	Vacuum pack	3.4 ^b ±0.21	3.9 ^{bc} ±0.07	7.0 ^{hi} ±0.31	5.9 ^g ±0.00
	MAP	4.2 ^{bcd} ±0.36	3.6 ^{bc} ±0.49	5.1 ^{ef} ±0.62	6.2 ^{hg} ±0.41
Total coliforms	Paper wrap	2.9 ^{cd} ±0.05	2.5 ^{bc} ±0.05	2.0 ^b ±0.07	2.5 ^{bc} ±0.11
	Vacuum pack	2.5 ^{bc} ±0.34	2.4 ^{bc} ±0.05	2.1 ^b ±0.36	3.6 ^{def} ±0.02
	MAP	2.9 ^{cd} ±0.17	2.5 ^{bc} ±0.28	4.2 ^f ±0.79	3.6 ^{def} ±0.44

^aMAP = Modified atmosphere packaging.

^{b,c,d,e,f,g,h,i}Means within a bacterial type with the same superscript do not differ ($P < .05$).

CONCLUSIONS

Purge loss was greater for loins if they were vacuum packaged. However, vacuum packaged butts lost less purge if they were vacuum packaged rather than using MAP. Thus, different pork cuts react differently to packaging method. The vacuum packaged loins and butts had higher APC after 10 days of storage but lower counts after 14 days than paper wrapped or MAP loins and butts. Paper wrapped loins and butts had the fewest coliforms after 14 days of storage.

Although *Listeria* species incidence decreased over time with all packaging methods, quicker reduction was shown in the vacuum packaged and MAP pork loins compared with the paper wrapped loins. The results from this study indicate that the microbial quality of pork loins and butts can be maintained by using vacuum packaging in contrast to paper wrapping and modified atmosphere packaging. The type of bacteria, pathogen and packaging atmosphere used will play an important role in the degree of microbial quality.

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