Levels Of Microbial Contamination in United States Pork Retail Products

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SUMMARY

To determine the extent of microbiological contamination in pork, samples of retail pork were collected from 24 stores across the United States. Additional samples of freshly-ground pork and/or pork sausage were collected from two hot-boning sow/boar sausage plants, two slaughter/fabrication plants and two further processing plants. Microbiological plate counts and pathogen incidences were higher ($P < 0.05$) for more extensively handled/processed products. *Listeria monocytogenes* was present more frequently in ground product and *Yersinia enterocolitica* was detected more often in store-handled product (whole-muscle, store-packaged and store-ground product). Pork products exposed to more handling and processing appeared to have lower microbiological quality.

INTRODUCTION

Most research involving pork contamination has concentrated on the carcass; however, pork can be contaminated with bacteria during fabrication, packaging, distribution and retail preparation and, therefore, more information on contamination levels and pathogen incidence must be gathered. This information can be helpful in the development and revision of HACCP plans in order to improve process control.

The purpose of this study was to develop a microbial baseline to be used to facilitate risk assessment of pork retail products and decision-making at various levels of the food safety chain. By sampling the final, products, which are closest to the consumer and are affected by all processes and handling before the purchase by consumers, the hazards associated with pork products can be evaluated. Therefore, the potential to prevent, eliminate or reduce (to an acceptable level) biological hazards in the food chain can be determined. The objectives of this study were to examine the incidence (presence/absence) of *Salmonella* spp., *Yersinia* spp., *Yersinia enterocolitica*, *Campylobacter jejuni/coli*, *Listeria* spp., and *Listeria monocytogenes*, and determine microbiological plate counts (Aerobic Plate Counts, APC; Total Coliform Counts, TCC; *Escherichia coli* Counts, ECC) for pork products at the wholesale and retail levels. An additional objective of this study was to characterize and detect *E. coli* non-O157 VTEC (verotoxigenic) bacteria in retail pork products to ascertain if a potential foodborne illness problem exists. Verocytotoxin-producing *E. coli* (VTEC) of serotype O157:H7 have been implicated in several foodborne outbreaks; however, it is becoming more evident that other serotypes of VTEC also can be associated with human foodborne illness *(1).*

MATERIALS AND METHODS

Sample collection. In Phase I of this study, six pork processing facilities located in six different cities of the continental United States allowed Colorado State University scientists to collect twenty samples of freshly-ground pork and/or pork sausage from each facility during a two-day period. The pork processing facilities included two hot-boning sow/boar sausage plants, two fed pig slaughter/fabrication plants and two further- processing plants. Plant samples, consisting of 300g of ground pork and/or pork sausage, were collected (aseptically after final grinding) with sterile gloves and placed into sterile bags (Whirl-Pak ®, Nasco, Ft Atkinson, WI).

In Phase II of the study 384 samples of pork products were purchased from four retail stores in each of six cities (Los Angeles, Denver, Dallas, Memphis, Sioux Falls and Baltimore. Products that were sampled included: (a) whole-muscle, store-packaged (tray and polyvinyl chloride film overwrap) retail cuts, (b) fresh, store-ground pork and/or pork sausage (paper-wrapped or tray and polyvinyl chloride film overwrap), (c) pre-packaged (at the processing plant) ground pork and/or pork sausage and (d) whole-muscle, enhanced (injected with a brine solution); pre-packaged under vacuum or store-packaged in trays overwrapped with polyvinyl chloride film) pork cuts. Four samples of each type of product were collected from the four stores in each city over a two-day period. These samples were transported with plant samples, within five hours of purchase, in their intact retail packaging and in pre-chilled containers containing frozen commercial ice substitutes and a temperature-recording device, by overnight delivery to IDEXX Food Safety Net Laboratory (San Antonio, TX) for microbiological testing.

Laboratory procedures.
Both processing plant and retail samples were analyzed for the presence of *Salmonella* spp., *Listeria* spp., *L. monocytogenes* and *C. jejuni/coli* using procedures recommended in the USDA-FSIS Microbiology Laboratory Guidebook, Volume 1 and 2 *(4)* and the presence of *Yersinia* spp. and *Y. enterocolitica* was determined using procedures recommended in the Compendium of Methods for the Microbiological Examination of Foods *(3)).* Additionally, Aerobic Plate Counts (APC), Total Coliform Counts (TCC) and *E. coli* Counts (ECC) were determined using Petrifilm™ (3M™ Microbiology Products, St. Paul, MN) and Butterfield's phosphate buffer (Difco Laboratories, Detroit, MI) to make serial dilutions before plating. Petrifilm™ Aerobic Count Plates for quantifying APC were incubated for 48 h at 35°C, and colonies were counted manually. Following incubation for 48 h at 35°C, coliform colonies both non- *E. coli* and *E. coli* (red and blue colonies associated with a gas bubble) growing on Petrifilm™

E. coli Count Plates were counted manually as well as *E. coli* colonies (dark blue colonies associated with a gas bubble).

Fifty *E. coli* isolates from fifty different pork samples including both processing plant and retail pork samples across cities and plant types were tested for the production of verotoxins (VTs) – also known as Shiga-like toxins (SLTs) – using a commercial microwell enzyme immunoassay (Premier EHEC, Meridian Diagnostics, Inc., Cincinnati, OH).

Statistical analysis. Pathogen incidence data were reported as a percentage of samples that tested positive for each pathogen. Microbiological plate count data (colony forming units – CFU) were transformed into base-ten logarithms ($\log_{10}$ CFU/g) before computing means and performing statistical analyses. All microbiological counts were reported as log CFU/g of product. Minimum detection limits for APC, TCC, and ECC were 1 log CFU/g for each of the types of microbiological plate counts, based on the maximum sensitivity of the tests. The APC, TCC, and ECC values falling below the minimum detection limit were entered into the data as 0.95 log CFU/g. Microbiological plate count data were analyzed using the general linear model procedures of SAS® *(2)*. Microbiological plate count data for pork retail samples were analyzed using a model that included product type as a fixed effect and city as a block effect, while data for plant samples were analyzed using a model that included plant type as a fixed effect. Least square means were separated using a pairwise t-test of SAS® *(2)*. All statistically significant differences were reported at the $P < 0.05$ level of Type I error.

RESULTS AND DISCUSSION

Microbiological plate counts. Mean APC, TCC and ECC (log CFU/g) in freshly-ground pork and/or pork sausage samples produced in three types of plants (hot-boned sow/boar sausage plants, slaughter/fabrication plants, further processing plants) are provided in Table 1. Mean APC and TCC were higher ($P < 0.05$) for samples collected from the slaughter/fabrication plants than for samples collected from the other types of plants. Mean ECC were similar among the three types of plants (1.0 – 1.1 CFU/g), but further-processing plants had the lowest levels of ECC ($P < 0.05$).

For samples collected at retail supermarket stores, mean APC and TCC (log CFU/g) were highest ($P < 0.05$) in ground pork that was processed and packaged in the store, followed by whole-muscle product packaged in the store (Table 1). Whole-muscle enhanced (injected/marinated) products and pre-packaged (at the processing plant, before distribution to the retail store) ground pork and/or pork sausage had the lowest ($P < 0.05$) APC, but did not differ from each other. In addition to having the highest mean APC and TCC of the samples collected from retail stores, store-ground pork and/or pork sausage also generated mean ECC (log CFU/g) values that were higher than mean ECC for the other samples collected from retail stores even though differences were small.

Differences reported in this study between the four retail products may have resulted from a combination of factors. Store-ground pork and/or pork sausage and whole muscle pork products cut and packaged in the stores had higher levels of microbial contamination in comparison to products packaged before distribution to the store. Store-packaged pork samples were possibly exposed to greater amounts of handling and to more equipment, thus contributing to increased exposure to environmental contamination and cross-contamination. Of the whole-muscle enhanced pork samples, 60 percent were store-packaged and 40 percent were pre-packaged at the processing plant of origin. In contrast, the non-injected whole muscle cuts were all store-packaged and, thus were exposed to more handling and contamination. Lower mean APC, TCC and ECC for whole-muscle enhanced products compared to counts for the non-enhanced products also may have been due to possible anti-microbial effects of the enhancing solutions.

Significant differences in APC, TCC, and ECC were apparent between store-ground, store-packaged pork and/or pork sausage and pre-packaged ground pork and/or pork sausage. The mean values for store-ground pork APC exceeded the mean values for pre-packaged product by over 2 log CFU/g. In addition, there was greater than one log CFU/g difference in TCC mean values between these products. Clearly, high levels of contamination occurred in the retail stores involved in this study. Some possible explanations for this include improper cleaning and sanitizing of equipment and poor employee hygiene within the store. It appeared that many stores lacked appropriate good manufacturing practices (e.g., keeping the walls, floors and work areas clean during processing).

Pathogen incidence. The most common pathogen detected in samples collected from processing plants was *L. monocytogenes*, with an overall incidence of 26.7 % (Table 2). The overall incidence of *Y. enterocolitica* in processing plant samples (3.3%) was much lower than that detected in whole-muscle and ground products prepared and packaged in retail grocery stores (incidences of 19.8% and 11.5 %, respectively; Table 3). Contamination with *Y. enterocolitica* appeared to occur after the product left the plant, during further processing and in retail stores.

Only minimal differences existed in the low incidence of *Salmonella* spp. and *C. jejuni/coli* between product types. Overall, *C. jejuni/coli*, with an incidence of 1.3 % across all types of retail products sampled, was the least frequent pathogen found on retail pork samples (Table 3).

As shown in Table 3, there was a high incidence of *Listeria* spp. (pathogenic and non-pathogenic species) in retail pork samples with an overall incidence of 41.9 % across all retail products sampled. *L. monocytogenes* was the most prevalent pathogen found in retail samples (19.8%) and was present more frequently in ground products than in whole-muscle products.

Yersina spp. (non-pathogenic plus pathogenic) were detected more often in products that had been handled and further processed in the retail store. *Yersinia enterocolitica* was detected most often in whole-muscle, store-packaged cuts (19.8%) and second most often in store-ground product (11.5%) in comparison to whole-muscle enhanced products (5.2%) or pre-packaged ground product (1.0%). In all cases, whole or ground, the products that were packaged at the processing facility before distribution had a lower incidence of *Yersinia* spp. and *Y. enterocolitica*.

Verotoxin production. None of the fifty *E. coli* colonies, derived from all regions and from each of the products, tested positive for verotoxins. Even though VTEC was not detected in this study, continuing efforts need to be made to detect and characterize non-0157:H7 *E. coli* in order to prevent potential outbreaks of foodborne illness.

Practical implications. Pork products exposed to more extensive handling and processing (e.g., products processed at the retail store or products that are ground) appeared to have lower microbiological quality. The microbiological plate counts and pathogen incidences were higher in more extensively handled/processed products. Good Manufacturing Practices and Sanitation Standard Operating Procedures are keys to reducing microbiological contamination in pork products. Most retail stores do not have functional quality assurance programs, as evident in the higher microbiological contamination in store processed and packaged products. It is likely that development of retail Hazard Analysis Critical Control Point plans would be beneficial to ensure the safety of pork products. Furthermore, supermarket operations should take a closer look at how completely they are cleaning and sanitizing grinding equipment. Further studies are warranted to identify efficient means for removing bacterial contamination from equipment in both processing plants and retail stores.

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TABLE 1. Bacteria counts (log CFU/g) for pork samples collected from three types of pork production plants and retail supermarkert stores.

Product	n	APC[x]	TCC[y]	ECC[z]
Plant Samples:				
Hot-Boning Sow/Boar Sausage Plants	40	2.92[b]	1.32[b]	1.09[a]
Slaughtering and Fabricating Plants	40	3.29[a]	1.52[a]	1.04[a]
Further-Processing Plants	40	2.98[b]	1.15[b]	0.96[b]
Retail Samples:				
Whole-Muscle, Store-Packaged	96	4.36[b]	1.54[b]	0.97[b]
Whole-Muscle, Enhanced	96	3.80[c]	1.31[c]	0.97[b]
Store-Ground Fresh Pork and/or Sausage	96	5.61[a]	2.19[a]	1.22[a]
Pre-Packaged Ground Pork and/or Sausage	96	3.77[c]	1.28[c]	0.98[b]

[abc]Means in a column, within a type of plate count and type of sample collected (retail or plant), bearing a different superscript letter differ ($P < 0.05$).
[x]Aerobic Plate Counts
[y] Total Coliform Counts
[z] *Escherichia coli* Counts

TABLE 2. Incidence of pathogens in ground pork and/or pork sausage produced in three types of plants.

Plant-Type	n	*Salmonella* spp. Positive Samples	*Salmonella* spp. Percent Positive	*Listeria spp.*[a] Positive Samples	*Listeria spp.*[a] Percent Positive	*Listeria monocytogenes* Positive Samples	*Listeria monocytogenes* Percent Positive	*Yersinia* spp.[a] Positive Samples	*Yersinia* spp.[a] Percent Positive	*Yerinia enterocolitica* Positive Samples	*Yerinia enterocolitica* Percent Positive	*Campylobacter jejuni/coli* Positive Samples	*Campylobacter jejuni/coli* Percent Positive
Hot-Boning, Sow/Boar Plant	40	4	10.0	16	40.0	5	12.5	4	10.0	0	0.0	5	12.5
Slaughtering/Fabricating Plant	40	3	7.5	23	57.5	13	32.5	4	10.0	3	7.5	0	0.0
Further Processing Plant	40	0	0.0	18	45.0	14	35.0	1	2.5	1	2.5	3	7.5
Across all plant-types	120	7	5.8	57	47.5	32	26.7	9	7.5	4	3.3	8	6.7

[a] Includes all positive samples for both pathogenic and non-pathogenic species

TABLE 3. Incidence of pathogens in four different types of pork retail product collected from stores located in six U.S. cities (n=384).

		Salmonella spp.		*Listeria spp.*[a]		*Listeria monocytogenes*		*Yersinia* spp.[a]		*Yerinia enterocolitica*		*Campylobacter jejuni/coli*	
Product	n	Positive Samples	Percent Positive	Positive Samples	Percent Positive	Positive Samples	Percent Positive	Positive Samples	Percent Positive	Positive Samples	Percent Positive	Positive Samples	Percent Positive
Whole-Muscle, Store-Packaged	96	8	8.3	27	28.1	14	14.6	47	49.0	19	19.8	1	1.0
Whole-Muscle, Enhanced	96	10	10.4	24	25.0	14	14.6	19	19.8	5	5.2	1	1.0
Store-Ground Fresh Pork and/or Sausage	96	7	7.3	59	61.5	22	22.9	22	22.9	11	11.5	0	0.0
Pre-Packaged Ground Pork and/or Sausage	96	12	12.5	51	53.1	26	27.1	11	11.5	1	1.0	3	3.1
Across all product-types		37	9.6	161	41.9	76	19.8	99	25.8	36	9.4	5	1.3

[a] Includes all positive samples for both pathogenic and non-pathogenic species