

Title: Microbial Profile of Overhead Surfaces and Condensate in Pork Processing Plants - **NPB #02-139**

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Abstract: Temperature and humidity differences within pork slaughter plants lead to condensate formation and dripping, especially in the carcass cooler. The purpose of this research was to enumerate selected types of microorganisms on rail and overhead surfaces and in condensate on those surfaces in pork coolers where carcasses are chilled and held prior to fabrication. Swab samples were collected from rails and overhead structures in three areas in the cooler before and during operations. Condensate droplets were also collected, when present, from rail or overhead structures. Enumeration of psychrotrophic aerobic plate count (APC), mesophilic APC and coliform bacteria were completed. Samples were also enriched and evaluated for presence of *Listeria*, *Salmonella*, and *Campylobacter* species. For rail and overhead structures, 5.8% of the samples had psychrotrophic APC counts greater than the $1.3 \log_{10}$ colony forming units/cm² (CFU/cm²) enumeration threshold. For both mesophilic APC and coliform samples, 1.4% of samples had greater than $1.3 \log_{10}$ CFU/cm². For the condensate samples, 32.7% of psychrotrophic APC counts and 4.1% of mesophilic APC counts were greater than the $4.2 \log_{10}$ CFU/ml enumeration threshold of the sampling technique. There were no coliforms detected in any of the condensate samples at a detection threshold of about $2.8 \log_{10}$ CFU/ml. The high detection limits for condensate samples resulted from the very low volume of condensate recovered. Rail and overhead samples produced positive results in 3.2% of samples enriched for *Salmonella sp.* and 4.1% for *Listeria sp.* Condensate samples had 4.1% positive for *Salmonella*, 4.2% positive for *Listeria*. None of the samples gave a positive result upon enrichment for *Campylobacter*. The results of this study indicate that in most cases, rail and overhead structures and condensate droplets are free of pathogens. However, when pathogens are present, they may be carried in droplets of condensate that accumulate on overhead surfaces.

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Introduction: Temperature and relative humidity differences among rooms within a pork processing plant often lead to condensation of water vapor on cold structures where warm moist air enters. This is often a severe problem in carcass chill coolers where warm moist air from the abattoir enters along with warm carcasses. Other areas of the plant may experience moisture condensation when warm air is released from smokehouses, cookers or hot products newly removed from the cooking process.

Accumulation of condensation on overhead structures is particularly troublesome when droplets enlarge to the point of falling onto surfaces or products below. USDA inspection policies mandate that condensation be controlled to prevent such droplets from falling. Immediate control measures often involve use of a sponge or other absorbent material attached to a long handle to remove the drops of condensation before they fall. Plants failing to achieve such control receive Noncompliance Reports (NR's) and are forced to cease operations in the area until the condensation is removed. The reasoning behind this policy is that moisture falling from overhead structures may carry pathogenic bacteria thus adulterating products or surfaces below.

An examination of published research related to this policy reveals that very little is known about the microbial status of condensation forming on or falling from overhead structures. One might speculate that, since condensate forms from water vapor, its microbial status would be determined by the condition of the surface where it forms.

Various researchers have documented the occurrence of spoilage or pathogenic bacteria on floors, drains, walls, equipment, workers, carcasses and products in meat processing plants (Grau, 1986 and Lahr, 1996). Nevertheless only one report was found dealing with microbial counts of overhead structures and condensate on those structures. Those researchers (Ockerman et al., 1977) found significant differences between two plants for both mesophilic and psychrophilic counts on overhead supports and rails in pork carcass chill coolers. During normal operations the rails of the chill cooler are exposed repeatedly to the passage of trolleys that may carry contaminants from the abattoir. These workers reported a 1-1.5 log increase in mesophilic and no change in psychrophilic counts on rails when comparing pre-op samples with those collected after operations had commenced. Mesophilic counts on supporting structures above the rail increased about 0.4 log during the same period. Condensate that accumulated on rails and overhead structures was combined and found to carry about 5 log of both mesophilic and psychrophilic bacteria per ml. As expected the mesophilic and psychrophilic counts found on rail and overhead structures correlated well with counts in the combined condensate ($r=0.73 - 0.91$). These researchers found no *coliform* or *staphylococcus* bacteria in any of the swab or condensate samples collected from overhead structures.

The findings reported by Ockerman et al. (1977) demonstrate a strong relationship between the microbial condition of overhead structures and the microbial condition of condensate that forms on those structures. However, this work was completed nearly 25 years ago in plants using much less stringent sanitation and facility design standards than today. While no pathogens were found, the researchers did not attempt to enumerate *Listeria* species. Thus, the current project is intended to revisit the question of microbial counts on overhead structures and in condensate forming on those structures.

Project Objectives:

1. To assess the number of selected types of microorganisms present on overhead structures in rooms where exposed carcasses or meat products are handled or prepared.

2. To observe any change in microbial counts on overhead structures during operations.
3. To assess the number of selected types of microorganisms present in condensate collected from overhead structures.
4. To evaluate the association between surface microbial counts on overhead structures and microbial counts in condensation forming on those surfaces.

Materials and Methods: Surface and condensate samples were collected in 7 pork processing plants in eastern and mid-western states. The plants included three very small establishments (<1000 head/day), two medium establishments (1000-5000 head/day) and two large establishments (> 5000 head/day). Table 1 gives some descriptive information about the cooperating plants.

Sample Collection. Swab samples were collected from overhead structures and rails at three locations in the carcass cooler. The first location was just inside the cooler where the hot carcasses enter. The other two locations were selected to be distant from the entry point and from each other. An area of 12 cm² was swabbed at each site guided by a sterilized metal ring. If condensate was present, droplets were collected by gently touching the droplet with a moist sample sponge. The droplet was absorbed in to the sponge without disturbing the surface itself. One sponge was used to collect as many droplets as possible at each location. Sample collection was completed at two times. The initial samples were collected before the start of operations (after completion of cleaning and sanitation) and then, after about 2 hours of operation. Additional samples were collected from three warm pork carcasses in the cooler during operation and again after the completion of chilling. Pork carcasses were sampled using the three site USDA sponge sampling protocol.

Microbial Detection and Enumeration Procedures. All samples, swabs and condensate, were processed for enumeration of mesophilic and psychrophilic aerobic plate counts (APC) and generic *E. coli*. *Listeria*, *Salmonella* and *Campylobacter* species were evaluated for presence or absence following appropriate enrichment procedures. APC samples were serially diluted in buffered peptone water (BPW) and enumerated on APC Petri Film (3M, Minneapolis, MN). Plates were incubated at 25°C and 37°C respectively for psychrophilic and mesophilic counts. Samples for generic *E. coli* were enumerated using *E. coli*/coliform Petri Film (3M, Minneapolis, MN). For detection of *Listeria* species, samples were enriched in University of Vermont broth (UVM, Difco) followed by enrichment in Fraser broth (Difco). Subsequent samples were plated onto MOX. Resulting black colonies from MOX following enrichment procedures were confirmed using the TECRA *Listeria* kit. *Salmonella* enrichments were completed first in Lactose broth then in Tetrathionate broth and Selenite Systeine broth. Samples were plated on XLD agar and presumptive colonies were confirmed using a *Salmonella* latex agglutination test. *Campylobacter* species were enriched in Bolton broth and plated on mCCDA agar. Presumptive colonies were confirmed using a *Campylobacter* test kit.

Microbial counts from enumeration techniques were transformed to Log₁₀ numbers before conducting Analysis of Variance using the PROC GLM of SAS (SAS Institute, Cary NC). Main effects were sampling time and season of year (warm versus cold). Results for presence or absence of specific pathogens are reported as raw frequencies and percent positive.

Results:

Objective 1.

Plants surveyed in this study included all sizes from very small (less than 10 head per day) to very large (over 28,000) head per day (Table 1). Surface swab samples of rail and overhead structures in pork carcass coolers showed very low numbers (log values less than -5) of mesophilic, psychrotropic and coliform bacteria (Table 2). Most of the swab samples collected from these surfaces gave zero or too few colonies to count (TFTC). For rail and overhead swabs zero or TFTC counts were found for 98.6%, 94.2% and 99.3% of mesophilic APC, psychrophilic APC and coliforms respectively. A limited number (49) of condensate samples were collected since droplets were frequently absent from the structures. Mesophilic APC in condensate was \log_{10} -2.65 while psychrophilic APC was \log_{10} 0.62 cfu/cm². No coliform bacteria were detected in any condensate samples.

Objectives 2& 4.

There were no significant differences between preoperational samples and those taken while hot carcasses were entering the cooler for mesophilic APC, psychrophilic, APC or coliform counts for rail, overhead structures and condensate (Table 3). This suggests that carcasses and trolleys along with air entering the cooler from the slaughter floor do not contribute much to the population of organisms in areas above the carcasses where condensate may form. The hot carcasses had significant populations of mesophilic APC and psychrophilic APC but few coliform bacteria (Table 3). After 18-24 hr chilling psychrophilic APC and coliform counts on carcass surfaces were further reduced ($p < 0.05$). Correlations calculated among counts for swab samples and condensate samples collected from rail and overhead surfaces showed no statistical relationship ($P > 0.05$) between the microbial count on a surface and the count in the condensate collected from that surface (Table 6).

Samples collected throughout the year were sorted into warm and cold season samples. There were few significant differences in microbial counts in the carcass cooler associated with season (Table 4). Mesophilic counts from rails and coliform counts on carcasses were significantly ($p < 0.05$) greater in warm season samples.

Objective 3.

Pathogen testing of all samples collected was done to determine the presence or absence of *Salmonella* sp., *Listeria* sp. and *Campylobacter* sp. Positive *Salmonella* results were found for 3.6%, 2.9% and 4.1% of rail, overhead and condensate samples, respectively (Table 5). Positive *Listeria* results were found for 3.6%, 4.5% and 4.2% of rail, overhead and condensate samples, respectively. No *Campylobacter* were found in any of the rail, overhead or condensate samples collected.

Discussion: Condensation on overhead structures in pork carcass chilling rooms and processing areas requires constant attention to prevent dripping and possible closure of rooms or areas in order to avoid the possibility of product adulteration. The current study was designed to gain some insight into the types and level of microbial contamination on surfaces and in condensate in pork carcass coolers. The total number of bacteria detected on rail and overhead surfaces was quite low with 94-99% of samples having too few bacteria to count. The source(s) of bacteria on the rail and overhead surfaces could not be determined in this study. Air, trolleys or carcasses entering from the slaughter floor were considered as a source of contamination but no increase in APC or coliform counts was observed as operations commenced. The occurrence of pathogens on rail and overhead structures and in condensate was low.

But, with 3-4% of samples positive for *Salmonella* and *Listeria* there is plenty of room for improvement. Based on the results of the current study USDA's zero tolerance policy for falling condensate in carcass coolers would not be justified in pork carcass coolers where regular sanitation and monitoring keep pathogens under control.

Lay Interpretation: Condensation in pork carcass coolers is a problem because it is often assumed that condensate contains pathogens and thus may contaminate products below. The current study was designed to gather some facts about the number and type of bacteria present in condensate and on the surfaces where condensate forms. Most of samples collected gave no detectable counts or had to few colonies for reliable counting. Nevertheless, 3-4% of samples were found to be positive for *Salmonella* or *Listeria*. The presence of these pathogens on the overhead surfaces and in the condensate from those surfaces represents a real concern. Regular sanitation procedures must be applied to rail and overhead structures to eliminate bacteria that might reside there. It was encouraging to note that bacterial numbers on rail and overhead surfaces did not increase during the slaughter operation. Thus, surfaces properly cleaned beforehand stay clean as carcasses are moved into the cooler. The current practice by USDA of zero tolerance for falling condensate should be modified to focus on proper sanitation procedures and bacterial monitoring to assure that surfaces and condensate are free of pathogens. The lead author of this study, Dr. Edward Mills, Penn State University, may be contacted by phone, 814-865-2394, or email ewm3@psu.edu.

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Table 1. Description of Selected Operations in Cooperating Pork Plants

	Very Small Plants (<1000 head/day)	Medium Plants (1000-5000 per day)	Large Plants (>5000 per day)
Plant Capacity			
Production (head/day)	70(10-200)	2350(1200-3500)	18,750 (9000-28,500)
Chain Speed (head/hr)	31(6-80)	360(141-580)	1103(1100-1106)
Slaughter Floor Air Temperature (°F)	72(68-78)	69(65-73)	80(80-80)
Carcass Holding Cooler Temperature (°F)	38.7(37.7-40.4)	34.5(33.9-35)	35(34-36)
Carcass Residence time in cooler (days)	3.5(1-8)	.5(.5-.5)	.9(.75-1)
Frequency of cleaning and sanitizing rail and overhead structures(#/year)	8.3(1-12)	28.0(4-52)	194(24-365)

Table 2. Total Plate and Coliform Counts^a for Surfaces and Condensate in Pork Carcass Coolers.

	<u>Observations</u>	<u>APC Mesophilic^b</u>	<u>APC Psychrotropic^c</u>	<u>Coliform</u>
Rail	138	-6.30(0.28)	-5.19(0.36)	-7.81(0.11)
Overhead	138	-7.13(0.22)	-6.04(0.32)	-7.92(0.08)
Condensate	49	-2.65(0.79)	0.62(0.82)	none detected
Carcasses	140	-2.11(0.20)	1.98(0.36)	-4.42(0.31)

^a Counts are log₁₀ of colony forming units per cm² or per ml for condensate for all sampling times and seasons. Average log values are reported with standard error of mean in parenthesis. Negative log values represent counts less than one per cm² or ml. Many individual samples gave no detectable colonies. Thus the average value may approach zero.

^b Incubated at 37°C.

^c Incubated at 25°C.

Table 3. Total Plate and Coliform Counts^a for Surfaces and Condensate in Pork Carcass Coolers Before and During Operations.

	<u>Preoperation Samples</u>			<u>Samples During Operation</u>		
	<u>APC Meso^b</u>	<u>APC Psy^c</u>	<u>Coliform</u>	<u>APC Meso.</u>	<u>APC Psy.</u>	<u>Coliform</u>
Rail	-6.14(0.39)	-5.12(0.51)	-7.88(0.16)	-6.46(0.39)	-5.27(0.51)	-7.74(0.16)
Overhead	-6.87(0.32)	-5.63(0.45)	-7.85(0.11)	-7.38(0.32)	-6.45(0.45)	nd ^d
Condensate	-3.32(1.54)	-0.63(1.59)	nd	-2.41(0.92)	1.07(0.95)	nd
Hot Carcasses	no carcasses present			2.47(0.29)	2.33(0.24)*	-3.73(0.45)*
Chilled Carcasses	no carcasses present			1.76(0.28)	1.65(0.23)*	-5.09(0.44)*

^a Counts are log₁₀ of colony forming units per cm² or per ml for condensate for all sampling times and seasons. Average log values are reported with standard error of mean in parenthesis. Negative log values represent counts less than one per cm² or ml. Many individual samples gave no detectable colonies. Thus the average value may approach zero.

^b Incubated at 37°C.

^c Incubated at 25°C.

^d None detected.

* Significant time effect, (P<0.05).

Table 4. Total Plate and Coliform Counts^a for Surfaces and Condensate in Pork Carcass Coolers During Warm or Cold Weather.

	<u>Warm Season</u>			<u>Cold Season</u>		
	<u>APC Meso^b</u>	<u>APC Psy^c</u>	<u>Coliform</u>	<u>APC Meso.</u>	<u>APC Psy.</u>	<u>Coliform</u>
Rail	-5.15(0.40)*	-5.30(0.53)	-7.60(0.16)	-6.99(0.38)*	-5.10(0.50)	nd ^d
Overhead	-7.28(0.32)	-5.86(0.46)	-7.84(0.11)	-6.99(0.31)	-6.21(0.44)	nd
Condensate	-2.89(1.27)	2.06(1.31)	nd	-2.50(1.01)	-0.30(1.04)	nd
Carcasses	2.36(0.29)	2.23(0.24)	-2.96(0.46)*	1.89(0.28)	1.75(0.23)	-5.74(0.44)*

^a Counts are log₁₀ of colony forming units per cm² or per ml for condensate for all sampling times and seasons. Average log values are reported with standard error of mean in parenthesis. Negative log values represent counts less than one per cm² or ml. Many individual samples gave no detectable colonies. Thus the average value may approach zero.

^b Incubated at 37°C.

^c Incubated at 25°C.

^d None detected.

* Significant season effect (P<0.05).

Table 5. *Salmonella* sp., *Listeria* sp. and *Campylobacter* sp. Detection^a for Surfaces and Condensate in Pork Carcass Coolers.

	<u>Salmonella sp.</u>	<u>Listeria sp.</u>	<u>Campylobacter sp.</u>
Rail	5/138 (3.62%)	5/132 (3.62%)	0/96 (0.0%)
Overhead	4/138 (2.90%)	6/132 (4.55%)	0/96 (0.0%)
Condensate	2/49 (4.08%)	2/48 (4.17%)	0/32 (0.0%)
Carcasses	2/138 (1.45%)	1/132 (0.76%)	2/96 (2.08%)

^a Values are reported as “number positive/number of samples tested”. Positives were determined following enrichment in appropriate media and plating on selective agar.

Table 6. Correlations Among Microbial Counts for Rail, Overhead and Condensate Samples Collected in Pork Carcass Coolers.

	<u>Condensate Mesophilic</u>	<u>Condensate Psychrophilic</u>
Rail Mesophilic	-0.062	-0.067
Rail Psychrophilic	-0.048	-0.078
Overhead Mesophilic	-0.034	-0.049
Overhead Psychrophilic	0.066	0.037
Condensate Psychrophilic	0.69*	1.00

* Correlation is significant (P<0.01).