

Microbiological Baseline Studies of Raw Pork, Beef and Chicken Carcasses in Ontario Abattoirs – An Overview

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AICF Conference

Edmonton, Alberta, November
2003



Outline

- Definition & purpose of baseline studies
- Study design
- Sampling methodology/sample submission
- Target microorganisms
- Laboratory analytical methods
- Summaries of results
- Practical implications and next steps

Baseline Risk Studies

- Studies of targeted microbial pathogens and indicator organisms and/or studies of chemical and biological contaminants across a segment of a population
- Meat Baseline studies:
 - Hogs 1999-00 (complete)
 - Beef 2000-01 (complete)
 - Chicken 2001-02 (complete)
 - Chemicals in raw meat (2002-05)
 - Ready-to-Eat Meat (2004-06)

Purposes of Baseline Studies

- Determine prevalence of pathogens & levels of indicator organisms - quantitative risk assessment
- Provide baseline to measure impact of intervention programs like HACCP
- Data used to set science-based, objective & achievable standards for industry performance (outcome-based system)
- Target and prioritize resources

Study Designs

- Stratification to account for production volume of high, medium & low volume plants (or large and small plants)
- Sampling frequency based on probabilities proportional to volume within stratum
- Random sampling of plants and carcasses
- Studies conducted over at least 12 months to account for seasonal variation
- Full geographic representation to account for regional variations

Hog Study Design

- Target population:
 - market hogs (> 45 kg)
 - and BBQ hogs (≤ 45 kg)
- Sample size based on variances reported in previous studies of pathogens & indicator organisms
 - 535 samples per stratum
 - 1605 samples in total

Stratification of Hog Study

STRATUM	VOLUME (head/yr)	# PLANTS	% TOTAL PLANTS	% TOTAL VOLUME
High	>5199	6	8.4	77.8
Medium	520-5199	68	35.8	17.7
Low	4-519	106	55.8	4.5

Hog Sampling Methodology

- Carcass selection done by meat inspector using random number table
- Hog sampling 6-12 hrs post-slaughter
- Sponge sampling technique (non-destructive) using BPW as diluent
- Three sampling sites per carcass (belly, ham & jowl) using 100 cm² template
- Inspectors received hands-on training in sampling techniques
- Samples shipped by courier and received by laboratory within 24 hrs of sample collection

Beef Study Design

- Target population:
 - Steers, Heifers, Cows & Bulls
- Sample size based on variances reported in previous studies
 - 522 samples per stratum
 - 1566 samples in total
- Information collected:
 - type of beef (fed vs. culled)
 - beef class (steer, heifer, cow or bull)
 - dressing method (bed or rail)
 - dehiding method (manual or mechanical)
 - shrouding practices

Stratification of Beef Study

STRATUM	VOLUME (head/yr)	# PLANTS	% TOTAL PLANTS	% TOTAL VOLUME
High	> 1039	10	5.6	53.1
Medium	520 - 1039	26	14.7	16.8
Low	1 - 519	141	79.7	30.1

Beef Sampling Methodology

- Carcass selection done by meat inspector using random number table
- Beef sampling 12 - 36 hrs post-slaughter
- Sponge sampling technique (non-destructive) using BPW as diluent
- Three sampling sites per carcass (brisket, flank & rump) using 100 cm² template
- Inspectors received hands-on training in sampling techniques
- Samples shipped by courier and received by laboratory within 24 hrs of sample collection

Chicken Study Design

- **Target Population:**
 - Broilers (≤ 2.2 Kg)
 - Roasters (> 2.2 Kg)
- **Sample size based on variances reported in previous studies**
 - 783 samples per stratum
 - 1566 samples in total

Chicken Study Design (Cont'd)

- **Information collected**

- scalding method
- chilling method
- dressing procedure
- evisceration method
- temperature of chill tank
- chicken weight

Stratification of Chicken Study

STRATUM	VOLUME (head/yr)	# PLANTS	% TOTAL PLANTS	% TOTAL VOLUME
Large	> 100,000	10	20.4	94.3
Small	< 99,999	39	79.6	5.8

Chicken Study Methodology

- Carcass selection done by meat inspector using random number table
- Carcass wash technique (non-destructive)
- Carcass shaken for 1 minute in 3500 mL sterile bag with 400 mL BPW
- Inspectors received hands-on training in sampling techniques
- Samples shipped by courier and received by laboratory within 24 hrs of sample collection

Sample Acceptance / Rejection Criteria

- Samples properly identified & submitted
- Sample completeness (3 sponges/ sample; 3 sites/sample)
- Samples properly & individually packaged
- Sample integrity (no leakage, no visible contamination)
- Age of sample (<24 hrs post-collection)
- Temperature of sample (not >8°C or <0°C)
(10°C for chicken)

Target Organisms

Indicators:

- Aerobic Colony Count (ACC)
- Total Coliform Count (TCC)
- *E. coli* Count (ECC)

Pathogens:

- *Salmonella* spp. (+ enumeration for chicken)
- *Campylobacter jejuni/ coli* (+ enumeration for chicken)
- Verotoxigenic *E. coli*
- *Listeria monocytogenes*

Laboratory Analysis

- Laboratory analysis provided by microbiology laboratory, Laboratory Services Division, University of Guelph (Dr. Joseph Odumeru)

Analytical Methods

Analysis	Methods	HPB References
ACC	Petrifilm	MFHPB-33
TCC/ECC	Petrifilm	MFHPB-34
L. Monocytogenes	LEB Enrichment	MFHPB-30
Salmonella	BPW Enrichment	MFHPB-20
Campylobacter	Rosef's Enrichment	MFLP-46
VTEC	Verocell Assay	MFLP-89

Results of Hog Baseline Study

Table 1. Prevalence of microorganisms on pork carcasses

MICROORGANISMS ¹	ONTARIO (Aggregate)		ONTARIO (BBQ Hogs)		ONTARIO (MKT Hogs)	
	# Samples	% Positive	# Samples	% Positive	# Samples	% Positive
ACC	1553	100.0 +/- NA	168	100.0 +/- N/A	1385	100.0 +/- N/A
TCC ^{ns}	1557	61.3 +/- .01	168	67.9 +/- .04	1389	60.6 +/- .01
ECC ^{**}	1557	39.5 +/- .01	168	49.4 +/- .04	1389	38.3 +/- .01
<i>L. monocytogenes</i> ^{**}	1556	10.7 +/- .01	168	4.8 +/- .02	1388	11.4 +/- .01
<i>Salmonella</i> ^{***}	1540	4.8 +/- .01	168	17.5 +/- .03	1374	3.3 +/- .005
<i>Campylobacter jejuni/coli</i> [*]	1556	26.7 +/- .01	168	33.3 +/- .04	1388	25.9 +/- .01
VTEC ^{ns}	1556	2.1 +/- .004	168	1.2 +/- .01	1388	2.2 +/- .004

¹Statistical Probabilities of Significance: ^{ns} = not significant; * = P<.05; ** = P<.01, *** = P<.001

Summary of Hog Results

- Market hogs were significantly lower in *E. coli*, *Salmonella* spp. and *C. jejuni/coli* incidence than BBQ hogs, but significantly higher in *L.monocytogenes* incidence
- Linear trend showing an inverse relationship between production volume and counts of indicator organisms
- Prevalence of pathogens was significantly affected by season



Results of Beef Baseline Study

Table 5. Prevalence of microorganisms on raw beef carcass surface samples

MICROORGANISMS ¹	Aggregate		Culled Beef		Fed Beef	
	# Samples	% Positive	# Samples	% Positive	# Samples	% Positive
TCC ^{**}	1557	27.8 +/- .01	189	36.51 +/- .04	1239	26.72 +/- .01
ECC [*]	1557	18.6 +/- .01	189	24.87 +/- .04	1239	17.92 +/- .01
<i>L. monocytogenes</i> ^{ns}	1556	9.9 +/- .01	189	7.94 +/- .02	1239	10.17 +/- .01
Salmonella ^{**}	1540	1.6 +/- .01	189	4.23 +/- .03	1239	1.29 +/- .005
<i>C. jejuni/coli</i> ^{**}	1556	1.5 +/- .01	186	4.30 +/- .04	1227	1.22 +/- .01
VTEC ^{ns}	1556	0.3 +/- .004	189	0.00 +/- .01	1238	0.32 +/- .004

Summary of Beef Results

- Fed beef significantly lower in *Salmonella* spp and *Campylobacter jejuni/coli* incidence than culled beef
- Prevalence of total coliforms, *Campylobacter jejuni/coli* and *Salmonella* spp. were influenced by production volume
- Prevalence of total coliforms and *E.coli* was significantly affected by season and geographic location



Results of Chicken Baseline Study

Table 5. Prevalence of microorganisms on raw chicken carcass surface samples

MICROORGANISMS		# SAMPLES	# POSITIVE	% POSITIVE
INDICATORS	ACC	1480	1480	100.0
	TCC	1480	1478	99.9
	ECC	1480	1465	99.0
PATHOGENS	L. monocytogenes	1469	440	30.0
	Salmonella spp.	1480	467	31.6
	C. jejuni/coli	1469	938	63.9
	VTEC	1468	0	0.0

Summary of Chicken Results

- Small plants had a significantly lower incidence of *L. monocytogenes* and *Salmonella* spp. and *E. coli* counts but significantly higher *C. jejuni/coli* than large plants
- Incidence of *C. jejuni/coli* was significantly higher in hot/dry seasons, but no other seasonal effects were evident
- Region had a significant impact on some pathogens
- Processing methods significantly influenced incidence of some pathogens, and this may be related to plant size

Practical Implications & Next Steps

- Data will be used to develop microbiological performance standards for pork, beef and chicken processing in Ontario provincial abattoirs
- Data will be used to measure the impact of intervention programs on microbial risk
- Variations in strata & regions will be used to examine influence of operational variables - e.g. processing rate
- Bigger may not be better ?