

# final report

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## Process control of sheep processing and possible effects on product shelf-life – MLA public

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## **Executive Summary**

Establishment A are seeking to gain information on process control on the sheep slaughter floor and in the boning room of their operation with the overall aim to improve the shelf life of their meat products. To this end the company has implemented a Plant Initiated Project (PIP) to provide information, based on the following objectives:

- 1. Evaluate the processes to determine the potential high microbial contamination or growth sites via investigations on the plant's slaughter floor, boning room and chillers.
- 2. Train laboratory staff in sampling and testing.
- 3. Provide a proposal on possible interventions and their likely effects on shelf life of final product.

To this end, a work program was undertaken comprising the following phases:

- 1. Determination of high-contamination sites on the lamb carcase Discussions with staff determined that nine carcases sites would be sampled.
- 2. Determination of the effect of final carcase wash and chilling on carcase contamination Sampling of carcases along the processing line was undertaken after evisceration and trimming, after the final wash and after overnight chilling.
- 3. Determination of working surface contamination during boning Transfer belts and selected surfaces were sampled throughout the working day
- 4. Determination of microbiological levels on meat cuts and, in discussion with staff, a range of cuts were selected
- 5. A pilot study on the effect of peroxyacetic acid on bacterial levels of primals
- 6. Laboratory staff were involved in all stages of designing, sampling, testing and analysis during the project

As a result of the above work program, Establishment A has acquired an in-depth appreciation of process control on the slaughter floor – of how much contamination is deposited on carcases during removal of the pelt and gut, and of how it is influenced by carcase washing and active chilling. Similarly, a great deal of information has been established on microbial levels of primal cuts and of the working surfaces over which they pass during boning and packing.

Establishment A can now compare their performance with data for ovine shoulders and legs obtained in the 2011 national baseline study by Phillips *et al.* (2012). This comparison indicates that the mean microbiological counts of products were generally lower those in the baseline (mean log<sub>10</sub> TVC 2.1 cfu/cm<sup>2</sup> *versus* mean log<sub>10</sub> TVC 2.8 cfu/cm<sup>2</sup>). It should be noted however, that product boned after 3-4 days (weekend) chilling was the equivalent of the average TVC of the national baseline study.

In summary, the present project achieved its objectives of training laboratory staff and providing key information on process control on the slaughter floor and in the boning room.

Additional to its Stage 1 objectives, the project set up a pilot intervention study on a selected primal cut (entire forequarter), which was then progressed to a shelf life study.

When the results of the shelf life study become known, in January 2016, Establishment A will make a decision on whether to proceed with Stage 2 of this project, which will focus on types of intervention in the boning room.

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## 1 Background

In June 2015, a study was undertaken in conjunction with Establishment A to discuss ways of improving process hygiene, with the primary objective of extending shelf life of chilled products. The outcomes of this meeting are contained in a report "Process control of sheep processing and possible effects on product shelf life (June 15, 2015)" and led to further studies via a Plant Initiated Project (P.PIP.0487: STAGE 1 – Process control of sheep processing and product shelf life improvement).

Work on this project at Establishment A occurred in the weeks of September 14, 2015 and October 13, 2015 and the results obtained are presented in this report.

## 2 **Projective Objectives**

The objectives of the project were to:

- 1. Evaluate the processes to determine the potential high microbial contamination or growth sites via investigations on the plant's slaughter floor, boning room and chillers.
- 2. Train staff in sampling and testing.
- 3. Provide a Stage 2 project proposal on the required interventions and their effects on shelf life of final product.

## 3 Methodology

The work was completed via the following phases:

- 1. Determination of high-contamination sites on the lamb carcase Discussions with staff determined that nine carcases sites would be sampled.
- 2. Determination of the effect of final carcase wash and chilling on carcase contamination

Sampling of carcases along the processing line was undertaken:

- After evisceration and trimming
- After the final wash
- After overnight chilling
- 3. Determination of working surface contamination during boning Transfer belts and selected surfaces were sampled throughout the working day
- Determination of microbiological levels on meat cuts and, in discussion with staff, a range of cuts were selected
- 5. A pilot study on the effect of peroxyacetic acid on bacterial levels of primals

For training purposes, staff were involved in the planning of the studies and throughout sample collection and testing.

For all trials, sponge samples were kept chilled until sample collection and testing were complete. Testing was undertaken immediately after transport to the laboratory.

The contents of each Whirlpak bag were "squished" to release bacteria from the sponge. Serial dilutions were made using Buffered Peptone Water and aliquots plated onto Aerobic Plate Count (APC), *Enterobacteriacae* and *E. coli* Petrifilm.

Cultures were incubated at 35°C for 48 hours when colonies were counted according to manufacturer's instructions and data entered on a spreadsheet. The count/cm<sup>2</sup> was calculated based on the area sponged and expressed as cfu/cm<sup>2</sup>.

#### 3.1 Contamination levels on the finished carcase

To assess the degree of contamination at various locations on the carcase, samples were gathered by sponging areas (~100cm<sup>2</sup>) after final inspection with a Whirlpak sponge, using both sides of the sponge. Sites were selected in discussion with staff. Five carcases were sampled on one occasion at the following sites:

- Neck
- Inside foreleg
- Outside foreleg
- Shoulder
- Brisket
- Flank
- Back
- Hind leg
- Bung

#### 3.2 Effect of final carcase washing and chilling

To determine microbiological levels on the carcase after slaughter and dressing, large-area sponging was undertaken (n=25) at three stages over four working days:

- 1. Final trim stand
  - Left hind leg and half the bung area (up to centre line)
  - Left flank, back and brisket
  - Left shoulder and fore leg
- 2. After carcase wash
  - Left hind leg and half the bung area (up to centre line)
  - Left flank, back and brisket
  - Left shoulder and fore leg
- 3. After overnight chilling
  - Right hind leg and half the bung area (up to centre line)
  - Right flank, back and brisket
  - Right shoulder and fore leg

At stages 1 and 2, the left and right sides of the same carcase, respectively, were sampled, while at stage 3 the carcase next in sequence was sampled. On day 1 five pairs of carcases, one pair at a time, were removed from the dressing chain after the final trim stand and sampled on the retain rail. This process was repeated on days 2 and 3, with five pairs of carcases sampled before and after lunch.

The area of each site was estimated by considering that each sample area consisted of a series of triangular regions. For each triangular region the dimensions were measured (or estimated) and the area calculated. The area estimates were subsequently added to give an estimate of the sample area, namely 475 cm<sup>2</sup>, 1200 cm<sup>2</sup> and 485 cm<sup>2</sup> for the hind leg area, back/flank and fore leg areas of a lamb carcase, respectively. For sheep carcases the area of each site was increased by 20%.

#### 3.3 Hygiene status of food contact surfaces

To assess the microbiological condition of meat contact surfaces in the boning room sponging was undertaken on one day during the breakfast and lunch work breaks and immediately after processing had ceased for the day. Surfaces on the following belts and equipment were sponged and the area calculated.

- Bandsaw table
- Table at shoulders belt
- Main belt 2
- Racks belt
- Shoulders belt
- Legs belt
- Loin plough
- Loin plough board before
- Loin plough board after
- Loin plough button
- Kevlar gloves
- Tub used for racks
- Belt after sanitising

#### 3.4 Hygiene status of primal cuts

The microbiological loading on primal cuts (n=25) in the boning room was undertaking by sponging two areas ( $\sim$ 100cm<sup>2</sup> each) over four working days as follows:

- Legs bone-in
- Legs boneless
- Shoulder bone-in
- Shoulders boneless
- Racks frenched
- Hind shanks
- Breast/flap
- Short loin

## 3.5 Effectiveness of peroxyacetic acid as an intervention

A pilot study was undertaken in which a primal cut (complete forequarter) was dipped in peroxyacetic acid (Inspexx, Ecolab Pty Ltd) at a concentration of 220 mg/kg for 30 seconds. Treated cuts and controls (dipped in water for 30 seconds), were allowed to drain for about 5 minutes before two areas (~100cm<sup>2</sup> each) were sponged. After sponging, cuts were vacuum packed and stored in the chilled product load-out chiller for assessment of shelf life (to be undertaken by staff after appropriate storage periods).

## 4 Results

## 4.1 Contaminations levels on the finished carcase

Five carcases were sponged immediately after final inspection at each of nine sites. The results are presented in Table 1. Bold values indicate high levels of microbial contamination, with neck, flank and bung being the most contaminated areas.

			TVC (lo	g10 cfu/cm2	E. coli	Entero		
Carcase #	1	2	3	4	5	Mean	Det	ections
Neck	2.6	2.3	2.2	1.4	1.5	2.0	1/5	3/5
Inside foreleg	0.0	2.0	1.6	1.5	1.5	1.3	2/5	3/5
Outside foreleg	1.4	1.9	0.5	1.2	0.5	1.1	3/5	1/5
Shoulder	0.7	1.3	1.7	0.0	1.8	1.1	1/5	2/5
Brisket	0.7	1.3	1.5	1.4	1.4	1.3	0/5	0/5
Flank	0.7	1.8	2.4	2.0	2.3	1.9	1/5	4/5
Back	1.4	1.7	2.2	1.7	2.1	1.8	0/5	0/5
Hind leg	0.0	0.5	1.3	1.4	1.6	0.9	0/5	0/5
Bung	2.3	2.1	2.8	2.7	3.2	2.6	5/5	5/5

Table 1: Summary of log<sub>10</sub> TVC/cm<sup>2</sup>, E. coli, Enterobacteriaceae prevalence on carcases after final inspection

## 4.2 Effect of final carcase rinse and chilling

To establish the microbial profile on different carcase sites and at different points along the processing chain a total of 25 pairs of carcases (17 lamb and 8 mutton) were sampled on October 12, 13 and 14. One carcase of each pair was sampled before and after washing, at sites described above, while the other carcase was sampled after chilling (see sites above).

There was little difference in the average  $log_{10}$  TVC between lamb and mutton carcases at each site (Figure 1), though variability in microbial levels between carcases was large (2-3  $log_{10}$  cfu/cm<sup>2</sup> i.e. 100 to 1000 fold).

An analysis of variance model was fitted to the data. The results indicated that there were statistically significant differences between lamb and mutton (P-value < 0.001), the three carcase sites (P-value < 0.001) and the processing stage (P-value < 0.001). The interaction between carcase site and processing stage was not significant (P-value = 0.85).

However, with respect to practical importance:

- The difference between lamb and mutton was 0.42 log<sub>10</sub> cfu/cm<sup>2</sup>, is not of practical importance;
- The difference between before and after wash was 0.10 log<sub>10</sub> cfu/cm<sup>2</sup> (higher after wash), and between after wash and after chilling was 0.38 log<sub>10</sub> cfu/cm<sup>2</sup> (lower after chilling), both are not of practical importance.
- Compared with the hindquarter, the back/flank was 0.03 log<sub>10</sub> cfu/cm<sup>2</sup> lower and the forequarter was 0.57 log<sub>10</sub> cfu/cm<sup>2</sup> higher, which is of practical importance.



Figure 1: Box plots of log TVC (cfu/cm<sup>2</sup>) of lamb and mutton carcases at three stages of processing

In general, average  $log_{10}$  Total Viable Count (TVC) counts were higher (0.5 to 1  $log_{10}$  cfu/cm<sup>2</sup>) on the shoulder and foreleg at each stage of the process (Figure 2). There was little difference in counts before and after washing at each site, and counts were reduced after chilling by about 0.5  $log_{10}$  cfu/cm<sup>2</sup>.



Figure 2: Box plots of log TVC (cfu/cm<sup>2</sup>) of different carcase locations at three stages of processing

In terms of *E. coli*, prevalence was higher on the hindquarters before washing, which led to more even spread over the carcase after washing. After chilling, *E. coli* was more likely on the shoulder and foreleg (Table 2).

Table 2: Summary of E. coli detections, average and maximum counts (cfu/cm<sup>2</sup>) of detections, before and after carcase rinsing and after chilling.

		Before rinse	9		After rinse	Э	After chill			
Carcase Site	EC %	Ave EC	Max EC	EC %	Ave EC	Max EC	EC %	Ave EC	Max EC	
Hind	84.0%	3.7	47.4	92.0%	5.7	42.1	32.0%	0.2	0.7	
Back/Flank	68.0%	0.3	0.9	88.0%	0.2	0.8	36.0%	0.7	6.3	
Fore	68.0%	2.2	25.8	84.0%	1.8	25.8	48.0%	1.1	10.1	

## 4.3 Hygiene status of food contact surfaces

Total bacterial counts on various food contact surfaces in the hot boning room are presented in Table 3.

- In general, counts on meat contact surfaces were low
- The exception is the first sampling occasion on Tuesday. Microbial counts on meat were also highest on this occasion (
- In general, all counts were similar to those determined in the national baseline survey of 2011 (Phillips *et al.* 2012) except for the first sampling occasion on Tuesday, September 15, when TVCs and detection of *E. coli* and *Enterobacteriaceae* were higher than any other sampling time. At that time carcases were boned from animals slaughtered on Thursday and Friday of the previous week.
- As reflected in Table 3, microbial loadings on food contact surfaces were also higher at the first sampling occasion (11.00).
- A run of mutton primals on Wednesday afternoon had similar counts to lamb primals

- In general, *E. coli* and *Enterobacteriaceae* counts, when they occurred, were extremely low, except for one boneless leg, on which *E. coli* counts were 225/cm<sup>2</sup>.
  - Table 4, Table 5 and Table 6).
  - The most highly contaminated surfaces were gloves and bandsaw. Both surfaces are moist and, in the case of gloves, warm from body temperature
  - Belts were generally similar in count to the meat passing over them
  - Microbial counts tended not to increase through the working day contact surfaces are dry and the temperature is low.
  - The racks belt sometimes has higher counts, possibly due to cut surfaces of meat remaining moist
  - Sanitising of belts at breaks was effective in reducing the microbial loading

Day	М	on. 14 Se	р.	Т	ues. 15 Se	əp.	Wed. 1	6 Sep.
Time	11:00	14:00	16:00	11:00	14:00	16:00	11:00	14:00
Bandsaw table	222	64	24	14	26	10	2	60
Table at shoulders belt	5	1	8	3	2	2	3	2
Main belt 2	74	14	0	6	9	0	1	1
Racks belt	24	22	2	2	7	0	44	45
Shoulders belt	5	1	0	1	2	0	1	2
Legs belt	6	6	14	2	5	0	3	1
Loin plough	2	17	6	2	7	1	10	4
Loin plough board								
before	7	11	6	1	4	4	12	5
Loin plough board after	53	4	17	1	4	18	14	1
Loin plough button	-	-	22	22	-	16		1
Gloves	1080	1600	0	3040	1440	112	80000	1760
Tub used for racks	56	7	17	10	12	1	4	126
Belt after sanitising	0	0	-	-	1	-	1	-

Table 3: TVC/cm<sup>2</sup> of contact surfaces in the hot boning room over three working days

#### 4.4 Hygiene status of primal cuts

Selected cuts in their packaging were removed from the final belt on eight occasions over three working days and sampled in the lab. Results for TVC, *E. coli* and *Enterobacteriaceae* are presented in Tables 4, 5 and 6, respectively.

- In general, all counts were similar to those determined in the national baseline survey of 2011 (Phillips *et al.* 2012) except for the first sampling occasion on Tuesday, September 15, when TVCs and detection of *E. coli* and *Enterobacteriaceae* were higher than any other sampling time. At that time carcases were boned from animals slaughtered on Thursday and Friday of the previous week.
- As reflected in Table 3, microbial loadings on food contact surfaces were also higher at the first sampling occasion (11.00).
- A run of mutton primals on Wednesday afternoon had similar counts to lamb primals
- In general, *E. coli* and *Enterobacteriaceae* counts, when they occurred, were extremely low, except for one boneless leg, on which *E. coli* counts were 225/cm<sup>2</sup>.

	Tues. 15 Sep.				V	Ved. 16 S	ep.	Thurs.	Thurs. 17 Sep.		
	1	2	3		1	2	3	1	2		
	Lamb	Lamb	Lamb		Lamb	Lamb	Mutton	Lamb	Lamb	Average	
Legs bone-in	2.4	2.8	1.7		2.0	2.0	1.9	1.5	2.7	2.1	
Legs boneless	2.4	2.7	3.1		2.7	1.4	1.9	2.1	1.6	2.2	
Shoulder bone-in	1.7	1.9	2.1		1.7	1.3	3.4	1.9	1.8	2.0	
Shoulders boneless	3.1	1.8	2.0		1.6	1.9	2.0	1.3	1.6	1.9	
Racks Frenched	1.9	0.9	1.8		1.2	1.0	1.1	1.6	1.3	1.4	
Shanks	3.8	2.6	1.9		2.8	1.8	2.4	2.3	1.7	2.4	
Breast/flap	2.6	3.8	1.9		1.9	2.0	1.9	1.9	2.5	2.3	
Short loin	4.4	2.4	1.6		1.8	2.0	2.7	2.9	2.9	2.6	
Average	2.8	2.3	2.0		2.0	2.0	2.0	2.0	2.0	2.1	

Table 4: log10 TVC/cm<sup>2</sup> of primal cuts sampled over three days

Table 5: E. coli/cm<sup>2</sup> of primal cuts sampled over three days. Cells with no data indicate E. coli was not detected.

	Tues. 15 Sep.			V	Ved. 16 S	Thurs. 17 Sep.		
	1	2	3	1	2	3	1	2
	Lamb	Lamb	Lamb	Lamb	Lamb	Mutton	Lamb	Lamb
Legs bone-in	1.38	0.13	0.13		0.50			
Legs boneless	0.13	0.13	225	0.13				0.88
Shoulder bone-in	0.25							
Shoulders boneless	0.13		0.13	0.75				
Racks Frenched	0.13							
Shanks	0.13					2.25		
Breast/flap	0.13	0.13			0.50			
Short loin	0.13	0.13	0.13					

Table 6: Enterobacteriaceae/cm2 of primal cuts sampled over three days. Cells with no data indicate Enterobacteriaceae were not detected.

	Τι	ues. 15 Se	ep.	V	Ved. 16 S	Thurs.	17 Sep.	
	1	2	3	1	2	3	1	2
	Lamb	Lamb	Lamb	Lamb	Lamb	Mutton	Lamb	Lamb
Legs bone-in	1.38	0.13	0.13		1.00	0.13		
Legs boneless	0.38	0.75	250	0.13			1.00	
Shoulder bone-in	0.63					0.50		
Shoulders boneless	3.75		0.13	0.38	0.13	0.13		
Racks Frenched	0.38							
Shanks	0.50	0.50				2.75		0.13
Breast/flap	0.25		0.38		1.38	0.13		0.38
Short loin	0.13		0.63			0.75		0.13

#### 4.5 Effectiveness of peroxyacetic acid as an intervention

As indicated in Figure 3, immersion of whole forequarters in Inspexx (PAA) solution at 220 mg/kg reduced the mean log TVC by  $0.8 \log_{10} \text{cfu/cm}^2$ , compared to dipping in water. This difference was highly significant (P-value = 0.0002) and large enough to be of practical importance. In addition, the difference is likely to be larger when compared with primals not dipped in water (not undertaken in this pilot trial).



Figure 3: Box plots of log TVC (cfu/cm<sup>2</sup>) of primals dipped in PPA (Inspexx) or water for 30 seconds.

#### 4.6 Staff Training

In terms of experimental design, production and sales staff were involved in pre-site meetings and at each entry meeting, when important variables were determined, *e.g.* sampling sites on the carcase, and identification of primals and food contact surfaces to be sampled.

The company's laboratory staff were fully involved in all sampling and laboratory aspects of the project in which 565 samples were tested for TVC, *E. coli* and *Enterobacteriacae*. At the end of both working weeks the team collated data and presented an overview to senior staff.

The capability of laboratory staff to plan and undertake investigative testing was enhanced by the present project, which extended their current skill level which is based on undertaking routine regulatory (ESAM) or customer (shelf-life) testing.

In addition, both received one-on-one training in spreadsheet design and operation, and this facility was also extended to the Quality Assurance (QA) Manager and Quality Control (QC) staff. During this process, several spreadsheets have been re-designed, or developed, to allow QA and laboratory staff to assess temporal patterns and trends in their data.

## 4.7 Temperature:time relations during transport of meat products to international destinations

The company will insert data loggers in cartons exported to international markets, both by air and sea freight.

## **5** Discussion

As a result of the project, Establishment A now has an in-depth appreciation of process control on the slaughter floor – of how contamination is deposited on carcases during removal of the pelt and gut, and of how it is influenced by carcase washing and active chilling.

Similarly, a great deal of information has been established on microbial levels of primal cuts and of the working surfaces over which they pass during boning and packing.

As a result, Establishment A can compare their performance with data for ovine shoulders and legs obtained in the 2011 national baseline study by Phillips *et al.* (2012). This comparison indicates that the mean microbiological counts of products manufactured were generally lower those in the baseline (mean log<sub>10</sub> TVC of 2.1 cfu/cm<sup>2</sup> *versus* 2.8 log<sub>10</sub> cfu/cm<sup>2</sup>). It should be noted however, that product boned after 3-4 days (weekend) chilling was the equivalent of the average TVC of the national baseline study.

The present project confirmed the relationship between food contact surfaces in the boning room and microbial loading on primal cuts established by previous studies and published in MLA's Processor's Guide to Improving Microbiological Quality (Edition 2, 2015).

During the exit meeting the then-current version of the UTas shelf life predictor was used to apprise staff of the extended shelf life likely to be obtained by improving microbial loading of primals on vacuum packing. The tool was used for various "what-if" scenarios with the result that a 90% reduction in microbial loading might result in an 8-day shelf life extension at just below 0°C.

In summary, the present project achieved its objectives of training laboratory staff and providing key information on process control on the slaughter floor and in the boning room.

Additional to its Stage 1 objectives, the project set up a pilot intervention study on a selected primal cut (entire forequarter), which was then progressed to a shelf life study.

When the results of the shelf life study become known, in January 2016, Establishment A will make a decision on whether to proceed with Stage 2 of this project, which will focus on types of intervention in the boning room.

## Reference

Phillips, D., Bridger, K., Jenson, I. and Sumner J. (2012). Microbiological quality of Australian sheep meat in 2011. Food Control 31:291-294.