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Review of current food safety microbiological sites and the establishment of a more meaningful measure to assess hygienic production

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Abstract

Microbiological testing of beef carcasses occurs, as a regulatory requirement, as a measure of hygienic dressing, and as an indirect measure of food safety. A literature review suggested that changes could occur in the current Australian system of testing sites without increasing food safety risks. Alternative sampling sites were also suggested in the literature. While the alternative sampling sites (neck, shoulder and flank) provide a greater number of unacceptable test results to assess hygienic processing compared to the traditional ESAM sampling sites (rump, flank and brisket), the increase was not statistically significant.

Executive summary

This project aimed to review the current food safety programs (both regulatory and commercial) and assess whether or not it might be more beneficial to undertake microbiological testing at other points in the process and/or other parts of the carcass that allow for evaluation of hygiene process to reflect risk.

The literature review concluded that there is no current regulatory barrier to changing the microbiological testing requirements in Australian beef abattoirs on the basis of risk. Furthermore, the literature proposes alternatives which may yield more meaningful process control outcomes. To assess this we tested carcasses from livestock with graded hide cleanliness during processing and post intervention at varying sites on the carcass, with comparative ESAM site samples, taking into account leading and following carcass side variation.

Samples were taken from four beef establishments down the Eastern seaboard of Australia. The samples were collected from the neck, shoulder, fore arm and brisket on the slaughter floor and chiller. An additional three samples at the rump, loin and flank were taken in the chillers; 308 samples sets in total, with 1546 swabs being tested for *E. coli*, coliforms and Total Viable Counts using the approved ESAM methodology.

Based on the project results, while the alternative sampling sites (neck, shoulder and flank) provide a greater number of unacceptable test results to assess hygienic processing compared to the traditional ESAM sampling sites (rump, flank and brisket), the increase was not statistically significant. Therefore, it could be argued that the alternative sites are just as effective and if a processor wished to use these as an alternative to the traditional ESAM sites due to Work Health and Safety (as in a number of cases these sites can be assessed without a step ladder) this research supports that variation. However, it is recognised that these benefits may not be cost-beneficial when considering the requirement of an equivalence agreement, especially if more important trade barriers or breaches are being negotiated.

A summary of the results based on cattle cleanliness score shows a clear trend to the cattle cleanliness score not being positively correlated to the level of microbiological contamination found on carcasses. In fact, in the post evisceration results the trend is of a negative correlation. However, these results must be interpreted considering the sampling variable of establishment and their individual process hygiene levels. As such with these results, cattle cleanliness score being positively correlated to the level of microbiological contamination found on carcasses cannot be proven or disproven and therefore Jordan et al (2014) stands. Testing at two locations, post evisceration and in the chiller, to measure hygienic process efficiency has demonstrated how meaningful a tool the analysis of samples collected at the two locations can be in understanding the level of hygienic process control rather than the use of a single location. (This concurs with work completed by Blagojevic et al 2011). With regulatory agreement and equivalence from customers and trading partners this process would not only allow for a better understanding of process hygiene by all but could be used as a risk framework with cattle cleanliness for testing frequency should the industry wish to seek equivalence.

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1 Background

The red meat processing industry has a wide-ranging and high level of compliance with food safety testing systems and regulations for different importing countries and markets. As new risk management strategies are understood and adopted, existing testing and reporting regimes need to be evaluated to ensure the industry is responsive to global food safety requirements.

The Australian Government regulates the export of red meat as part of the Australian Meat Export Inspection System (AEMIS). The Department of Agriculture and Water Resources maintains the Product Hygiene Indicator (PHI) in Excel Spreadsheets (previously the *E.Coli* and *Salmonella* Monitoring database {ESAM}) which hold the regulatory and market access required food safety results, both microbiological and macroscopic. The ESAM food safety testing program was developed to meet market access requirements from the United States Department of Agriculture (FSIS) in 1996. The allocation of testing sites was based on limiting the regulatory action to the processors. The FSIS requirements for which this program has equivalence has since been amended, however ESAM still stands. The development of PHI in 2011 was to provide analysis of the objective data captured through ESAM and Meat Hygiene Assessment (MHA) to indicate the level of compliance of processors and to benchmark processors against each other. Industry has expressed concern that the current PHI (and its parts, both ESAM and MHA), for all its costs, does not provide adequate measures of true hygienic process control. This project was the formative stages for a more realistic risk management framework for process control through the review of current sites used for microbiological testing and investigate potential new sites or points in the process that are true indicators of hygienic process control.

Currently regulatory food safety testing protocols are based on historic market access equivalence agreements. This results in expense for testing which provides no real correlation to port of entry testing that when positive (despite acceptable ESAM results) results in loss of market access, loss of customers and increased compliance costs (through retesting, increased testing, reworking, destruction, down grading or return of product).

This project aimed to address these issues by reviewing the current food safety programs (both regulatory and commercial) and assess whether or not it might be more beneficial to undertake microbiological testing at other points in the process and/or other parts of the carcass that allow for evaluation of hygiene process to reflect risk.

2 Project objectives

The objectives of this project were:

1. Review of current food safety systems and work practices, and assessment of current and potential new testing sites to more accurately reflect the level of hygienic processing; and
2. Make recommendations for industry and regulatory adoption
3. Draft paper for publication on the results of the trialled sites and draft market access proposals to vary current equivalence agreements

3 Methodology

3.1 Literature review and Scoping Study

The review of the literature was conducted using a Boolean search methodology (using OR, AND, NOT etc.) combined with a series of preliminary key words (for example, beef, carcass, carton etc.) These search results were refined further by using key word limitation with search terms such as sampling, process hygiene, *E.coli*, etc. Abstracts of sourced articles were assessed for relevance. The reference lists of the resulting journal articles were also reviewed to identify any articles that may be relevant, but had not been detected through the initial search process. Abstracts of these articles were also assessed to determine whether the information was suitable for inclusion.

Searches of MLA and AMPC publications and reports, and internet searches using key terms were also conducted. Government and industry sites, both based in Australia and overseas were also searched using key terms.

A comparison of the cited studies was hampered due to a number of factors. These include differences in sampling techniques (excision, swabbing), sampling sites used, timing of sampling both on the processing line as well as time post chilling, the area of site sampled, and in the case of North American studies, the use of decontamination interventions. However, despite these differences, there did appear to be evidence to warrant investigation of additional or alternative sampling sites.

A copy of the literature review is provided at Appendix 1.

Based on the studies reviewed there is evidence to suggest that investigation of alternative sampling sites is warranted. A number of studies identified the neck as a suitable site (Untermann et al 1997; Zweifel et al 2008). Forearm and shoulder were also identified as suitable microbiological sampling sites (Untermann et al 1997).

Location on the processing chain of sampling is also warranted. It is proposed that sampling be conducted post evisceration i.e. at the point of most contamination. Blagojevic et al (2011) identified the importance of measuring the effectiveness of processing hygiene however in sampling prior to de hidding, they have not captured any potential processing contamination that occurs due to the evisceration process. It is proposed that sampling be conducted post evisceration and again at the end of the slaughter floor post chilling. Comparison of the data obtained at the two points will determine whether moving sampling to post evisceration and adjusting the acceptable microbiological limits is a valid and beneficial change for the microbiological sampling of beef. The ratio between the two samples can also be considered, as per the study conducted by Blagojevic et al (2011). This will determine whether the ratio of microbiological contamination between the two points is a valid measure of process hygiene which could also allow for exploration of the European and United States of America's approach of risk based monitoring over inspection sampling monitoring.

Given the research conducted to date and the Australian microbiological results from the ESAM program and the Australian baseline surveys consideration should be given as to the indicator organisms assessed. The meat industry pathogens such as *Salmonella* and Shiga toxin producing

E.coli strains such as O157:H7 present the opposite qualities occurring too infrequently and with an uneven distribution with very low numbers when present. These organisms are therefore poorly suited as a measure of process control (Blajoveic et al 2011). As such it is proposed that *E.coli* and coliform testing will be conducted using swab sampling during the project.

In summary the conclusion from the literature is that there is no current regulatory barrier to changing the microbiological testing requirements in Australian beef abattoirs on the basis of risk. Furthermore, the literature proposes alternatives which may yield more meaningful process control outcomes. Microbiological process control can be enhanced if considered in conjunction with macroscopic indicators (such as hide cleanliness) which also have the benefit of being determined in real time. To assess this we proposed to test carcasses from livestock with graded hide cleanliness during processing and post intervention at varying sites on the carcase, with comparative ESAM site samples, taking into account leading and following carcase side variation.

3.2 Testing and analysis of potential new sites for sample collection

A draft methodology was developed based on the findings of the literature review conducted in milestone 2. The hypotheses to be tested were:

1. Cattle Cleanliness Score is positively correlated to the level of microbiological contamination found on carcasses i.e. dirtier cattle results in a higher number of unacceptable test results.
2. Following carcase sides will have a higher level of microbiological contamination to leading carcase sides due to the evisceration process i.e. following carcase sides will have a higher number of unacceptable test results.
3. Alternative sampling sites provide a greater number of unacceptable test results to assess hygienic processing compared to the traditional ESAM sampling sites.
4. Hygienic process control can be assessed based on the test results throughout processing.

Based on hypotheses 1, 3 and 4 an assessment can also be made as to whether a risk-based approach can be taken to the microbiological testing component of process hygiene assessment. This would allow hygienic process control monitoring to be modified, to allow companies to design their monitoring programs with periodic slaughter floor, carcase and end of product testing for *E.coli* and coliform counts to be based on a risk-based frequency, rather than uniform whole of industry sampling frequencies.

The initial draft methodology proposed sampling each sample site of interest (i.e. neck or rump), on a single carcase. Sampling would use the standard swabbing method used in ESAM. The sample sites area to be swabbed would be a 10cm by 10cm area. The sampling sites at post evisceration on the slaughter floor would be at the neck, shoulder, fore arm and brisket. The sample sites in the chiller would be the neck, shoulder, fore arm, brisket, rump, loin and flank. This ensured the ability to compare the existing ESAM sites to the new sites identified from the literature review.

During discussions with several company representatives, advice was received that previous commercial research had demonstrated that sampling a single site on a single carcase (i.e. 100cm²) would result in a very high proportion of non-detections or low levels of the target organisms. It was suggested that a composite sample at each sample site be taken across three carcasses to provide

adequate data to allow for comparison of the sample sites. As such, test results were then reported per sample covering three carcasses, not per carcass.

Prior to the commencement of testing, the draft methodology was also discussed with industry representatives and experts in the field, as well as government officials from the Department of Agriculture and Water Resources (DAWR) and Safe Food Production Queensland. No further amendments were recommended.

A trial of the proposed methodology was conducted by the project team at a South-East Queensland establishment. This trial allowed the project team to test the functionality during processing. The findings from this trial resulted in the following changes to the methodology prior to the commencement of sample collection for the project.

- Cattle Cleanliness Scoring - the draft methodology proposed the use of a cattle cleanliness score developed by Jordan (2014) to score cattle in the lairage prior to sampling. Discussions with industry indicated that many establishments already rate cattle cleanliness and have a scoring system in place. These systems rate cattle on a 1 - 3 or 1 – 5 scales for cleanliness. The premise of these systems was that a cleanliness score 1 was applied to clean cattle and a score of 3 (1-3 system) or 5 (1-5 system) to cattle with a build-up of dags. The decision was made to adopt the scoring system already utilised by processors as these were simpler in design and could be more readily adopted by industry. The draft methodology also proposed splitting sampling equally across all cleanliness scores. MLA advised that it was unlikely that this would be achievable, with lower cleanliness scores likely to be more commonly encountered than the higher cleanliness scores. While every effort was made to select cattle from all cleanliness scores, the contingency was to sample from the cleanliness scores available on production days to ensure the total number of tests required could be achieved, given hypothesis 1 had already been proven by Jordan (2014).
- Location of sampling on the slaughter floor – the draft methodology proposed that sampling be conducted on carcasses run onto the retain rail. During the trial, it became evident that this was not operationally feasible. The methodology was amended with testing to take place at chain speed post evisceration and prior to pre-trim. This resulted in a team of four being required to complete the sampling as opposed to the original team of two. The additional two people recruited for sampling were Intern students from Texas Tech University studying meat science. Both were familiar with the processing environment and one had previously conducted carcass swabbing for microbiological testing. The project team leader ensured all team members were aware of the principles of sterility and were trained using an instructional video developed by the University of Nebraska and endorsed by the Food Safety Inspection Services of the United States Department of Agriculture, on how to conduct carcass swabbing. During the sampling process, each student was also paired with a team member from Food and Veterinary Services to ensure oversight of sampling at all times.
- Carcass selection for sampling – the draft methodology proposed sampling one side of a carcass on the slaughter floor and then sampling the second side of the same carcass in the chillers the following day. Following discussion with industry during the trial, it was identified that this would not be operationally possible. Grading and marshalling of carcasses in the chillers would result in carcasses that may have been grouped the previous day (i.e. concurrent bodies in a production run) may no longer be located together. The methodology was amended to assign a portion of the day's production (e.g. bodies 1 – 500) as available to be sampled on the leading side and a different portion of the day's production (e.g. bodies 501 – 1000) as available to be sampled on the following side. Sampling in the chillers on the following day was then reversed (i.e. bodies 1 – 500 available for sampling on the following

side and bodies 501 – 1000 available for sampling on the leading side). This split was to ensure that the same side of a carcass was not sampled on both the slaughter floor and in the chillers.

The methodology proposed conducting testing at four establishments. An initial list of establishments was provided based on high levels of detections in a recent 4000cm² baseline testing survey conducted by MLA and Symbio Alliance. Establishments also varied in process hygiene (i.e. increase in detections, decrease in detections or no change) between slaughter floor and chiller sampling. The proposed methodology was then discussed with establishments that expressed an interest in being involved in the trial. Based on these discussions, a number of establishments were unable to be included in the trial due to their operational inability to support sampling. For example, following discussion and observation of facilities at one establishment, it was evident that the height of the chain meant that post evisceration sampling could not be conducted as it would not be possible to safely reach the sample sites when standing on the slaughter floor prior to pre-trim. Other operational constraints encountered included chain speed and space on the processing line and competing operational priorities for the companies.

3.3 Final Methodology

The methodology used at the four establishments where sampling was conducted was as follows:

3.3.1 Materials

- Sterile specimen sponge in sterile Whirl-pack™
- 25 ml sterile Butterfield's Phosphate Diluent
- Sterile Template 10 cm x 10 cm
- Sterile gloves
- Container for carrying supplies
- Sampling Sheets (Appendix 1)

3.3.2 Sampling Location and Sampling Sites

1. Sampling was conducted at the following locations using the following sample sites.
 - a. Post evisceration Location (Four sample sites: Neck, Shoulder, Fore arm, Brisket)
 - b. Chiller Location (Seven sample sites: Neck, Shoulder, Fore arm, Loin, Brisket, Flank, Rump)

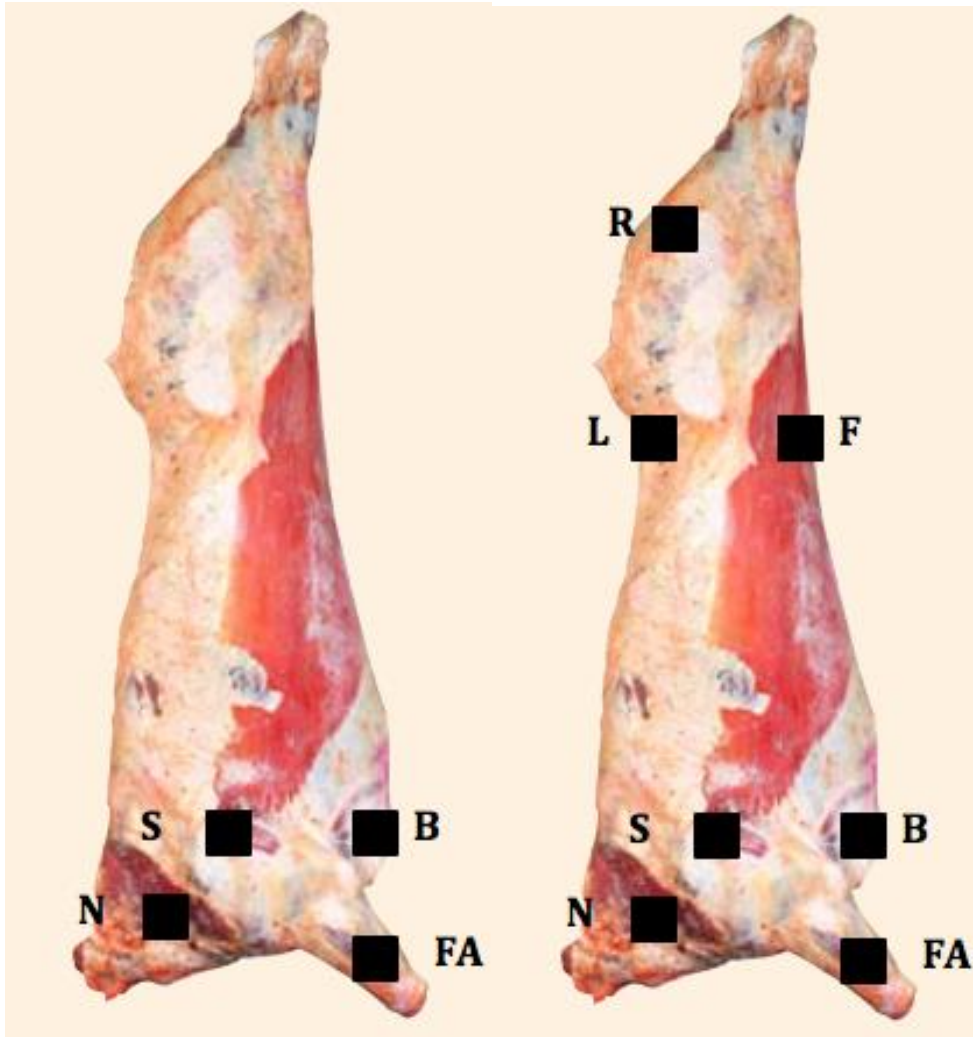


Fig. 1 – Post evisceration sampling sites (left) and Chiller sampling sites (right) annotated with black squares and letters for the sampling sites. N is neck, S is shoulder, B is brisket, FA is Fore Arm, R is rump, L is Loin and F is flank.

- To ensure that each carcass side was sampled only once, sampling was conducted by allocating bodies to either leading or following sampling based on a range of body numbers. For example, if the day's production was 1000 head:

Body number	Leading Side	Following Side
1 - 500	Post evisceration	Chiller
501 - 1000	Chiller	Post evisceration

3.3.3 Sampling Area

Each sample site had a swab area of 10cm x 10cm. Post evisceration and Chiller samples used the same swab across the same site on 3 carcasses to give a composite sampling area of 300cm² for each sample site.

3.3.4 Sampling Procedure

The sampling procedure followed the current ESAM sampling procedure which is the Whirl-pack™ collection method as outlined in the Microbiological Manual for Sampling and Testing of Export Meat and Meat Products (DAWR 2017) and below:

A sampling sponge (which usually comes dehydrated and prepacked in a sterile bag) will be used to sample all the sampling sites as follows:

- 1. Ensure that all bags have been pre-labelled, and all supplies are on hand, including the sampling template.*
- 2. If a reusable template is used, it must be sterilised between the carcasses*
- 3. Locate the sampling sites*
- 4. While holding the sponge bag at the top corner by the wire closure, tear off the clear, perforated strip at the top of the bag.*
- 5. Remove the cap from sterile diluents water.*
- 6. Carefully pour about half the contents of the sterile diluent (approximately 10 ml) into the sponge bag to moisten the sponge. Recap the bottle.*
- 7. Close the top of the bag by pressing the wire closure together. Use hand pressure from outside of the bag and carefully massage the sponge until it is fully hydrated (moistened). Sponges may be pre-moistened prior to entering the plant to sample the carcasses.*
- 8. Prior to collecting the sample, carefully push the moistened sponge to the upper portion of the bag orienting one narrow end of the sponge up toward the opening. DO NOT open the bag or touch the sponge with your fingers.*
- 9. While holding the bag, gently squeeze any excess fluid from the sponge using hand pressure from the outside. The whole sponge should sit in the bag.*
- 10. Open the bag containing the sponge, being careful not to touch the inner surface of the bag with your fingers. The wire closure at the top of the bag should keep the bag open. Set bag aside.*
- 11. Put on a pair of sterile gloves*
- 12. Carefully remove the moistened sponge from the bag with the thumb and fingers (index and middle) of your sampling hand.*
- 13. With your free hand, retrieve the template by the outer edge, taking care not to contaminate the inner edges of the sampling area of the template.*
- 14. Locate the sample site. Place the template over this location.*
- 15. Hold the template in place with one gloved hand. Only the sponge should touch the sampling area. Take care not to contaminate this area with your hands.*
- 16. With the other hand, wipe the sponge over the enclosed sampling area (10 cm x 10 cm) for a total of approximately 10 times in the vertical and 10 times in the horizontal direction. The pressure of swabbing should be as if you were trying to remove a stubborn stain from the carcass. The pressure should not be so hard as to crumble or destroy the sponge. The template may need to be “rolled” from side to side during swabbing since the surface of the carcass is not flat. This will ensure the 100 cm² is enclosed while swabbing.*
- 17. After swabbing the site on all three carcasses, carefully place the sponge back in the sample bag, taking care not to touch the sponge to the outside of the sample bag.*
- 18. Uncap the previously used diluent bottle. Add the additional diluent (about 15 ml) to the sample bag to bring the total volume to approximately 25 ml.*
- 19. Expel excess air from the bag containing the sponge and fold down the top edge of the bag 3 to 4 times to close. Secure the bag by folding the attached wire tieback against the bag.*

3.3.5 Sample Numbers

Target sample numbers were developed based on the allocation of samples to leading or following carcass sides across all sampling sites and equal numbers across the three cleanliness scores (Table 1). The number of samples to be collected at each establishment was determined based on operational factors such as chain speed and number of head processed in a single production day. Each sample was collected at the same site across three carcasses (i.e. composite sample 300cm²). This approach would result in a total of 276 samples sets being taken from 828 carcass sides during this project. (A sample set is defined as the swabs taken across all sampling location at either post evisceration or in the chiller tested for *E.coli*, coliform and TVC. Please note that TVC, APC and SPC have been used interchangeably throughout the report).

Table 1 – Target Sample Numbers

Carcass Side	Following side			Leading side			Grand Total
	1	2	3	1	2	3	
Cleanliness Score							
Sampling Location							
Post Evisceration							
Brisket	23	23	23	23	23	23	138
Fore Arm	23	23	23	23	23	23	138
Neck	23	23	23	23	23	23	138
Shoulder	23	23	23	23	23	23	138
Chiller							
Brisket	23	23	23	23	23	23	138
Flank	23	23	23	23	23	23	138
Fore Arm	23	23	23	23	23	23	138
Loin	23	23	23	23	23	23	138
Neck	23	23	23	23	23	23	138
Rump	23	23	23	23	23	23	138
Shoulder	23	23	23	23	23	23	138
Grand Total	253	253	253	253	253	253	1518

3.3.6 Transport of Samples to the Laboratory

Sponge samples were stored chilled prior to transport to the testing laboratory, either on the same day or overnight. The testing laboratory was NATA (National Association of Testing Authorities) accredited to ISO 17025 and an approved laboratory with the Department of Agriculture & Water Resources for the estimation of Aerobic Plate Count (APC), generic *Escherichia coli* and coliform bacteria in meat and meat products. All samples were analysed no later than on the day following collection at the establishment. Samples arrived at the testing laboratory at <5°C.

3.3.7 Test Methodology

The standard Department of Agriculture and Water Resource (DAWR) approved test methodology for ESAM samples was used to test, analysis and report the results of the swab sampling. Each 300cm² composite sample was tested for *E. coli*, coliforms and Total Viable Count (TVC).

3.3.8 Microbiological Analysis

After manually palpating sponges (~1 min), one mL was removed and serially diluted in PSS (Peptone Salt Solution). Appropriate dilutions were plated onto Aerobic Plate Count (APC) Petrifilm™ and *E. coli*/coliform (E. coli) Petrifilm™. *E. coli* and coliforms were taken directly from the sponges as well as serially diluting. After incubation at 35±1°C for 24-48±3h colonies were counted for the target organism following the manufacturer's instructions and results expressed as CFU/cm². From the colonies counted, a laboratory worksheet was used to convert a result for the original surface area. The limit of detection of this method, based on the sampling area undertaken, was 0.83 CFU/cm² for APC and 0.083CFU/cm² for *E. coli* and coliforms.

3.4 Deviations from the Methodology During Sampling

The final methodology was followed as written for sampling conducted on the slaughter floor at all establishments. Appendix 2 provides photos during sampling. While every effort was made to develop a methodology prior to commencing sampling that would be operationally viable at all establishments, amendments to the methodology for chiller sampling were required at two plants.

Plant B - Sampling of chilled carcasses was only operationally practical if conducted in the chiller corridor at chain speed, due to a lack of available space in the chillers and marshalling area. As chiller sampling required the sampling of seven sites (as opposed to the four sites sampled on the slaughter floor), it was not possible to complete all seven sites at chain speed. The decision was made to conduct fore quarter sampling (Neck, Shoulder, Fore arm, Brisket) across one set of three carcasses and Hind quarter sampling (Loin, Flank, Rump) across the next set of three carcasses. Where practical (i.e. when the chain was stopped on breaks), sampling was conducted as per the methodology (i.e. all seven sampling sites tested on a set of three carcasses).

Plant C – Carcasses from this establishment were loaded out prior to the next day's production. As a result, it was not possible to sample on the slaughter floor one day and sample in the chillers after a minimum of a 12-hour chill, the following day. Chiller samples were conducted at this establishment a minimum of 6 hours after chilling.

4 Results

The total numbers of samples collected is shown in Table 2. In summary, 309 sample sets were taken; 145 full sample sets were taken on the slaughter floor, post evisceration and 105 full sample set and 59 split sample sets (28 on the fore quarter and 31 on the hind quarter) were taken in the chillers.

This resulted in 924 carcase sides being swabbed. A total on 1520 swabs samples were taken, each tested using DAWR approved testing methodologies for coliforms, *E. coli* and Total Viable Count.

The expected total number of samples were collected.

Carcase Side Cleanliness Score	Following side				Leading side				Grand Total
	1	2	3	Not	1	2	3	Not	
Sampling Location									
Post Evisceration									
Brisket	28	19	17	10	25	22	13	11	145
Fore Arm	28	19	17	10	25	22	13	11	145
Neck	28	19	17	10	25	22	13	11	145
Shoulder	28	19	17	10	25	22	13	11	145
Chiller									
Brisket	25	27	5	10	27	18	13	8	133
Flank	25	29	4	10	27	22	11	8	136
Fore Arm	25	27	5	10	27	18	13	8	133
Loin	25	29	4	10	27	22	11	8	136
Neck	25	27	5	10	27	18	13	8	133
Rump	25	29	4	10	27	22	11	8	136
Shoulder	25	27	5	10	27	18	13	8	133
Grand Total	287	271	100	110	289	226	137	100	1520

Table 2 – Sample Numbers Taken

The detection levels set by the DAWR in the Microbiological Manual for Sampling and Testing of Export Meat and Meat Products are as follows:

- *E.coli*: >0cfm/cm² to <20 cfm/cm² is considered marginal and ≥20 cfm/cm² is considered unacceptable
- APC: >1000 cfm/cm² to <31625 cfm/cm² is considered marginal and ≥31625 cfm/cm² is considered unacceptable

As such positive results have been classified as:

- *E.coli*: detection i.e. >0.084 cfm/cm² with results ≥20 cfm/cm² is considered unacceptable (U)
- Coliform: detection i.e. >0.084 cfm/cm² with results ≥20 cfm/cm² is considered unacceptable (U)
- APC: a result >1000 cfm/cm² Broken down to marginal (M) (>1000 cfm/cm² to <31625 cfm/cm²) and unacceptable (U) (<31625 cfm/cm²).

4.1 Results for Establishment A

The complete results for Establishment A are available in Appendix 3. In the appendix all positive results are in a red font. All samples for Establishment A had a cattle cleanliness score of 2. Two unacceptable APC results were reported across the results, one from a post evisceration sample taken from a shoulder site swab and one from a chiller sample taken from a rump site swab, both on following carcass side sets. No Unacceptable *E.coli* or coliform results were reported.

Thirty-six sample sets were taken at the post evisceration sampling location. These were evenly split across leading and following carcass sides. Seventeen sample sets had a minimum of one positive result, 10 of these samples were taken from leading carcass sides and 7 were samples taken from following carcass sides. Table 3 provides a summary of the number of positive sample sets by carcass side sampled and sampling site.

Table 3 – Summary of Number of Positive Sample Sets by Carcass Side Sampled and Sampling Site – Post Evisceration

Carcass side	Sampling Site			
	Brisket	Fore Arm	Neck	Shoulder
Leading (10)	1	2	7	1
Following (7)	3	1	3	3

On every occasion where a positive result was reported for *E.coli*, the corresponding coliform result was also positive. A summary of the positive results by test type is available in Table 4.

Table 4 – Summary of Establishment A Positive Results by Test Type (M denotes marginal results and U denotes Unacceptable results) – Post Evisceration

Test Type	Sampling Site				Carcass Side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
No. Sample Sets: 18						
Coliform	1M	2M	7M	1M	Leading	2
<i>E.coli</i>	0	2M	4M	1M	Leading	2
SPC	0	0	0	0	Leading	2
No. Sample Sets: 18						
Coliform	3M	1M	3M	1M	Following	2
<i>E.coli</i>	1M	0	3M	1M	Following	2
SPC	0	0	0	1M 1U	Following	2

Thirty-three sample sets were taken at the chiller sampling location. These were split with 16 sample sets taken from leading carcass sides and 17 sample sets taken from following carcass sides. Six sample sets had a minimum of one positive result, 1 of these samples was taken from leading

carcase sides and 5 samples were taken from following carcase sides. Table 5 provides a summary of the number of positive sample sets by carcase side sampled and sampling site. A summary of the positive results by test type is available in Table 6.

Table 5 – Summary of Number of Positive Sample Sets by Carcase Side Sampled and Sampling Site - Chiller

Carcase side	Sampling Site						
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder
Leading (1)	1	0	0	0	0	0	0
Following (5)	0	0	1	0	2	1	1

Table 6 – Summary of Establishment A Positive Results by Test Type (M denotes marginal results and U denotes Unacceptable results) - Chiller

Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore	Loin	Neck	Rump	Shoulder		
No. Sample Sets: 16									
Coliform	0	0	0	0	0	0	0	Leading	2
<i>E.coli</i>	0	0	0	0	0	0	0	Leading	2
SPC	1M	0	0	0	0	0	0	Leading	2
No. Sample Sets: 17									
Coliform	0	0	1M	0	1M	0	1M	Following	2
<i>E.coli</i>	0	0	0	0	0	0	0	Following	2
SPC	0	0	1M	0	1M	1U	0	Following	2

4.2 Results for Establishment B

The complete results for Establishment B are available in Appendix 4. All positive results are in a red font. Samples for Establishment B were spread across cattle cleanliness score 2 and 3. Table 7 provides a summary of the number of sample sets by sampling location and cattle cleanliness score. At post evisceration 1 set of carcase sides sampled covered both cattle cleanliness scores and in the chillers 1 fore quarter and 1 hind quarter sampling set covered both cattle cleanliness scores, as such these samples have been categorised as having a 'Not Recorded' cattle cleanliness score. At Establishment B modifications had to be made to the collection of chiller samples (as described in Section 3.3) as such fore quarter, hind quarter and full sample sets are also separated in Table 7.

Table 7 – Summary of Number of Sample Sets by Sampling Location and Cattle Cleanliness Score.

Sampling location Carcase side	Cattle Cleanliness Score		
	2	3	Not Recorded
Post Evisceration			
Leading	4	13	1
Following	1	17	0
Chiller – Full Sample Sets			
Leading	0	2	0
Following	1	0	0
Chiller – Fore Quarter Sample Sets			
Leading	2	11	0
Following	9	5	1
Chiller – Hide Quarter Sample Sets			
Leading	6	9	0
Following	11	4	1

One unacceptable *E.coli* and one coliform result were also reported from the same loin site swab from a hind quarter chiller sample taken from following carcass sides.

Thirty-six sample sets were taken at the post evisceration sampling location. These were evenly split across leading and following carcass sides. Six sample sets had a minimum of one positive result, 2 of these samples were taken from leading carcass sides and 4 were samples taken from following carcass sides. Table 8 provides a summary of the number of positive sample sets by cattle cleanliness score, carcass side sampled and sampling site.

Table 8 – Summary of Number of Positive Sample Sets by Cattle Cleanliness Score, Carcass Side Sampled and Sampling Site – Post Evisceration

Carcass side	Sampling Site			
	Brisket	Fore Arm	Neck	Shoulder
Cattle Cleanliness Score 2				
Leading (0)	0	0	0	0
Following (1)	0	0	1	0
Cattle Cleanliness Score 3				
Leading (2)	0	0	2	0
Following (3)	1	1	2	0
Cattle Cleanliness Score Not Recorded				
Leading (0)	0	0	0	0
Following (0)	0	0	0	0

On the single occasion a positive result was reported for *E.coli*, the corresponding coliform result was also positive. A summary of the positive results by test type is available in Table 9.

Table 9 – Summary of Establishment B Positive Results by Test Type (M denotes marginal results and U denotes Unacceptable results) – Post Evisceration

Test Type	Sampling Site				Carcase Side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
No. Sample Sets: 4						
Coliform	0	0	0	0	Leading	2
<i>E.coli</i>	0	0	0	0	Leading	2
SPC	0	0	0	0	Leading	2
No. Sample Sets: 1						
Coliform	0	0	1M	0	Following	2
<i>E.coli</i>	0	0	0	0	Following	2
SPC	0	0	0	0	Following	2
No. Sample Sets: 13						
Coliform	0	0	2M	0	Leading	3
<i>E.coli</i>	0	0	0	0	Leading	3
SPC	0	0	0	0	Leading	3
No. Sample Sets: 17						
Coliform	1M	1M	2M	0	Following	3
<i>E.coli</i>	0	0	1M	0	Following	3
SPC	0	0	1M	0	Following	3
No. Sample Sets: 1						
Coliform	0	0	0	0	Leading	-
<i>E.coli</i>	0	0	0	0	Leading	-
SPC	0	0	0	0	Leading	-

Three full sample sets and 28 fore quarter and 31 hind quarter split sample sets were taken at the chiller sampling location. These were split across leading carcass sides and following carcass sides as showed in Table 7. One full sample set had a minimum of one positive result, the sample was taken from a neck sampling site of following carcass sides with a cattle cleanliness score of 2. Four fore quarter split sample sets had a minimum of one positive result, these samples were taken from shoulder sampling sites, 2 from leading carcass sides with a cattle cleanliness score of 3 and 2 from following sides with a cattle cleanliness score of 2.

Eleven hind quarter split sample sets had a minimum of one positive result, 2 of these samples were taken from following carcass sides with a cattle cleanliness score of 2, 1 sample at the loin sampling

site and 1 sample at the rump sampling site. The remaining 9 hind quarter split sample sets with a minimum of one positive result were from flank sampling sites of leading carcass sides, 4 samples with a cattle cleanliness score of 2 and 5 samples with a cattle cleanliness score of 3. Tables 10, 11 and 12 provides a summary of the number of positive sample sets by carcass side sampled and sampling site. On every occasion where a positive result was reported for *E.coli*, the corresponding coliform result was also positive. A summary of the positive results by test type is available in Tables 13, 14 and 15.

Table 10 – Summary of Number of Positive Full Sample Sets by Cattle Cleanliness Score, Carcass Side Sampled and Sampling Site - Chiller

Carcass side	Sampling Site						
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder
Cattle Cleanliness Score 2							
Leading (0)	0	0	0	0	0	0	0
Following (1)	0	0	0	0	1	0	0
Cattle Cleanliness Score 3							
Leading (0)	0	0	0	0	0	0	0
Following (0)	0	0	0	0	0	0	0

Table 11 – Summary of Number of Positive Fore Quarter Split Sample Sets by Cattle Cleanliness Score, Carcass Side Sampled and Sampling Site - Chiller

Carcass side	Sampling Site			
	Brisket	Fore Arm	Neck	Shoulder
Cattle Cleanliness Score 2				
Leading (0)	0	0	0	0
Following (2)	0	0	0	2
Cattle Cleanliness Score 3				
Leading (2)	0	0	0	2
Following (0)	0	0	0	0
Cattle Cleanliness Score Not Recorded				
Leading (0)	0	0	0	0
Following (0)	0	0	0	0

Table 12 – Summary of Number of Positive Hind Quarter Split Sample Sets by Cattle Cleanliness Score, Carcase Side Sampled and Sampling Site - Chiller

Carcase side	Sampling Site		
	Flank	Loin	Rump
Cattle Cleanliness Score 2			
Leading (4)	4	0	0
Following (2)	0	1	1
Cattle Cleanliness Score 3			
Leading (5)	5	0	0
Following (0)	0	0	0
Cattle Cleanliness Score Not Recorded			
Leading (0)	0	0	0
Following (0)	0	0	0

Table 13 – Summary of Establishment B Positive Full Sample Set Results by Test Type (M denotes marginal results and U denotes Unacceptable results) - Chiller

Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore	Loin	Neck	Rump	Shoulder		
No. Sample Sets: 1									
Coliform	0	0	0	0	1M	0	0	Following	2
<i>E.coli</i>	0	0	0	0	1M	0	0	Following	2
SPC	0	0	0	0	0	0	0	Following	2
No. Sample Sets: 2									
Coliform	0	0	0	0	0	0	0	Leading	3
<i>E.coli</i>	0	0	0	0	0	0	0	Leading	3
SPC	0	0	0	0	0	0	0	Leading	3

Table 14 – Summary of Establishment B Positive Fore Quarter Split Sample Set Results by Test Type (M denotes marginal results and U denotes Unacceptable results) - Chiller

Test Type	Sampling Site				Carcase Side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
No. Sample Sets: 2						
Coliform	0	0	0	0	Leading	2
<i>E.coli</i>	0	0	0	0	Leading	2
SPC	0	0	0	0	Leading	2
No. Sample Sets: 9						
Coliform	0	0	0	2M	Following	2
<i>E.coli</i>	0	0	0	0	Following	2
SPC	0	0	0	1M	Following	2
No. Sample Sets: 11						
Coliform	0	0	0	2M	Leading	3
<i>E.coli</i>	0	0	0	0	Leading	3
SPC	0	0	0	0	Leading	3
No. Sample Sets: 5						
Coliform	0	0	0	0	Following	3
<i>E.coli</i>	0	0	0	0	Following	3
SPC	0	0	0	0	Following	3
No. Sample Sets: 1						
Coliform	0	0	0	0	Following	-
<i>E.coli</i>	0	0	0	0	Following	-
SPC	0	0	0	0	Following	-

Table 15 – Summary of Establishment B Positive Hind Quarter Split Sample Set Results by Test Type (M denotes marginal results and U denotes Unacceptable results) - Chiller

Test Type	Sampling Site			Carcase Side	Cattle Cleanliness Score
	Flank	Loin	Rump		
No. Sample Sets: 6					
Coliform	1M	0	0	Leading	2
<i>E.coli</i>	0	0	0	Leading	2
SPC	3M	0	0	Leading	2
No. Sample Sets: 11					
Coliform	0	1U	1M	Following	2
<i>E.coli</i>	0	1U	1M	Following	2
SPC	0	0	0	Following	2
No. Sample Sets: 9					
Coliform	5M	0	0	Leading	3
<i>E.coli</i>	4M	0	0	Leading	3
SPC	2M	0	0	Leading	3
No. Sample Sets: 4					
Coliform	0	0	0	Following	3
<i>E.coli</i>	0	0	0	Following	3
SPC	0	0	0	Following	3
No. Sample Sets: 1					
Coliform	0	0	0	Following	-
<i>E.coli</i>	0	0	0	Following	-
SPC	0	0	0	Following	-

4.3 Results for Establishment C

The complete results for Establishment C are available in Appendix 5. All positive results are in a red font. A cattle cleanliness score was not available at Establishment C. Two unacceptable APC results were reported across the results, both from a post evisceration sample taken from the shoulder site swab on following carcass side sets. No Unacceptable *E.coli* or coliform results were reported.

Twenty sample sets were taken at the post evisceration sampling location. These were evenly split across leading and following carcass sides. Seven sample sets had a minimum of one positive result, 2 of these samples were taken from leading carcass sides and 5 were samples taken from following carcass sides. Table 16 provides a summary of the number of positive sample sets by carcass side sampled and sampling site.

Table 16 – Summary of Number of Positive Sample Sets by Carcase Side Sampled and Sampling Site.

Carcase side	Sampling Site			
	Brisket	Fore Arm	Neck	Shoulder
Leading (2)	0	0	2	0
Following (5)	3	0	2	2

On every occasion where a positive result was reported for *E.coli*, the corresponding coliform result was also positive. A summary of the positive results by test type is available in Table 17.

Table 17 – Summary of Establishment C Positive Results by Test Type (M denotes marginal results and U denotes Unacceptable results) – Post Evisceration

Test Type	Sampling Site				Carcase Side
	Brisket	Fore Arm	Neck	Shoulder	
No. Sample Sets: 10					
Coliform	0	0	2M	0	Leading
<i>E.coli</i>	0	0	1M	0	Leading
SPC	0	0	1M	0	Leading
No. Sample Sets: 10					
Coliform	2M	0	0	0	Following
<i>E.coli</i>	2M	0	0	0	Following
SPC	1M	0	2M	2U	Following

Seventeen sample sets were taken at the chiller sampling location. These were split with 8 sample sets taken from leading carcase sides and 9 sample sets taken from following carcase sides. Four sample sets had a minimum of one positive result, 1 of these samples was taken from leading carcase sides and 3 samples were taken from following carcase sides. Table 18 provides a summary of the number of positive sample sets by carcase side sampled and sampling site. A summary of the positive results by test type is available in Table 19.

Table 18 – Summary of Number of Positive Sample Sets by Carcase Side Sampled and Sampling Site - Chiller

Carcase side	Sampling Site						
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder
Leading (1)	1	0	0	0	0	1	0
Following (3)	0	3	0	0	0	0	0

Table 19 – Summary of Establishment C Positive Results by Test Type (M denotes marginal results and U denotes Unacceptable results) - Chiller

Test Type	Sampling Site							Carcase Side
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder	
No. Sample Sets: 8								
Coliform	1M	0	0	0	0	0	0	Leading
<i>E.coli</i>	0	0	0	0	0	0	0	Leading
SPC	0	0	0	0	0	1M	0	Leading
No. Sample Sets: 9								
Coliform	0	1M	0	0	0	0	0	Following
<i>E.coli</i>	0	0	0	0	0	0	0	Following
SPC	0	2M	0	0	0	0	0	Following

4.4 Results for Establishment D

The complete results for Establishment D are available in Appendix 6. All positive results are in a red font. All samples for Establishment D had a cattle cleanliness score of 1. No Unacceptable APC, *E.coli* or coliform results were reported.

Fifty-three sample sets were taken at the post evisceration sampling location. These were split across leading (25) and following (28) carcass sides. Fifty-two sample sets had a minimum of one positive result, 24 of these samples were taken from leading carcass sides and 28 were samples taken from following carcass sides. Table 20 provides a summary of the number of positive sample sets by carcass side sampled and sampling site.

Table 20 – Summary of Number of Positive Sample Sets by Carcass Side Sampled and Sampling Site – Post Evisceration

Carcass side	Sampling Site			
	Brisket	Fore Arm	Neck	Shoulder
Leading (24)	4	5	22	9
Following (28)	11	14	24	24

On every occasion where a positive result was reported for *E.coli*, the corresponding coliform result was also positive. A summary of the positive results by test type is available in Table 21.

Table 21 – Summary of Establishment D Positive Results by Test Type (M denotes marginal results and U denotes Unacceptable results) – Post Evisceration

Test Type	Sampling Site				Carcase Side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
No. Sample Sets: 25						
Coliform	4M	5M	22M	9M	Leading	1
<i>E.coli</i>	2M	4M	14M	2M	Leading	1
SPC	0	0	1M	0	Leading	1
No. Sample Sets: 28						
Coliform	11M	14M	24M	24M	Following	1
<i>E.coli</i>	7M	12M	12M	16M	Following	1
SPC	0	2M	4M	0	Following	1

Fifty-two sample sets were taken at the chiller sampling location. These were split with 27 sample sets taken from leading carcase sides and 25 sample sets taken from following carcase sides. One sample set had a minimum of one positive result, this sample was taken from the rump sample site from following carcase sides. Table 22 provides a summary of the number of positive sample sets by carcase side sampled and sampling site. A summary of the positive results by test type is available in Table 23.

Table 22 – Summary of Number of Positive Sample Sets by Carcase Side Sampled and Sampling Site - Chiller

Carcase side	Sampling Site						
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder
Leading (0)	0	0	0	0	0	0	0
Following (1)	0	0	0	0	0	1	0

Table 23 – Summary of Establishment D Positive Results by Test Type (M denotes marginal results and U denotes Unacceptable results) - Chiller

Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore	Loin	Neck	Rump	Shoulder		
No. Sample Sets: 27									
Coliform	0	0	0	0	0	0	0	Leading	1
<i>E.coli</i>	0	0	0	0	0	0	0	Leading	1
SPC	0	0	0	0	0	0	0	Leading	1
No. Sample Sets: 25									
Coliform	0	0	0	0	0	1M	0	Following	1
<i>E.coli</i>	0	0	0	0	0	0	0	Following	1
SPC	0	0	0	0	0	0	0	Following	1

5 Analysis and Interpretation of Results

5.1 Hypothesis 1: Cattle Cleanliness Score is positively correlated to the level of microbiological contamination found on carcasses

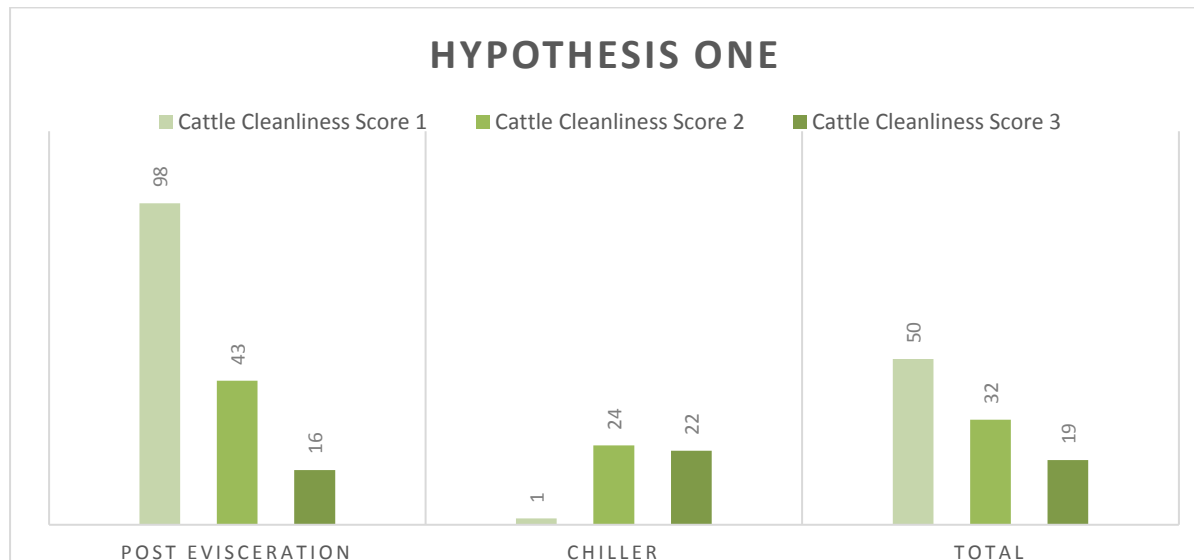
Of the 309 full and split sample sets taken, 269 sets had a cattle cleanliness score allocated to them. Table 24 provides a summary of the total sample sets (including split samples) and the number of positive sample sets. Given the variation in sample set numbers the percentage of positive results has been calculated to allow comparison of the results.

Table 24 – Summary of Results based on Cattle Cleanliness Score

Sampling Location	No. of Sample Sets	Cattle Cleanliness Score		
		1	2	3
Post Evisceration	Total	53	41	30
	Positive	52	18	5
	Percentage of Positive	98.1%	43.9%	16.7%
Chiller	Total	52	62	31
	Positive	1	15	7
	Percentage of Positive	1.9%	24.2%	22.6%
Totals	Total	105	103	61
	Positive	53	33	12
	Percentage of Positive	50.5%	32.0%	19.7%

Figure 1 provides a summary of the results based on cattle cleanliness score, with a clear trend to the null hypothesis, i.e. cattle cleanliness score is not positively correlated to the level of microbiological contamination found on carcasses. In fact, in the post evisceration results the trend is of a negative correlation.

Figure 1 – Summary of Results based on Cattle Cleanliness Score



However, these results must be interpreted considering the sampling variable of establishment and their individual process hygiene levels. Due to the results for Cattle Cleanliness Scores 1 and 3 coming solely from Establishment A and B respectively. As such with these results, hypothesis 1 cannot be proven or disproven.

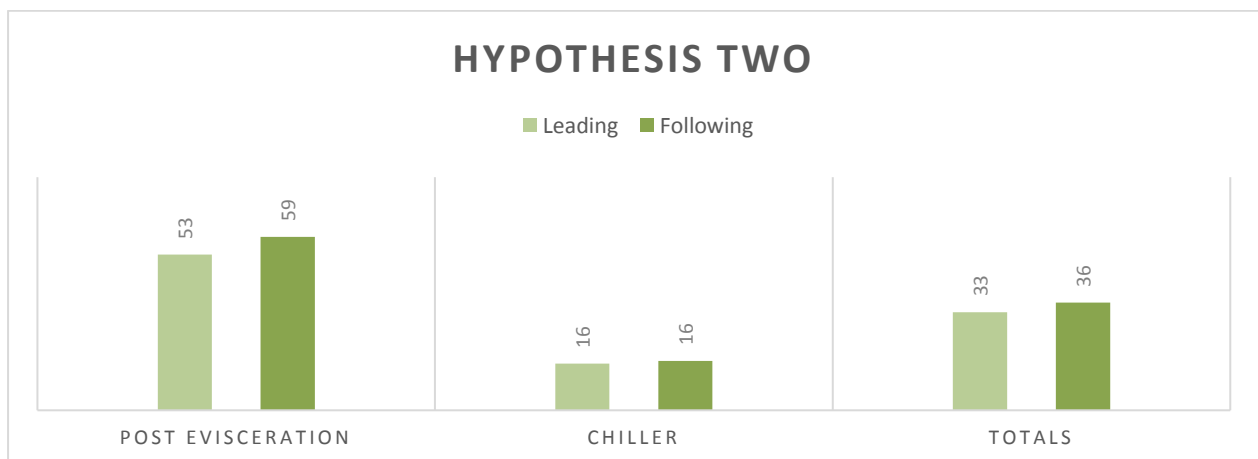
5.2 Hypothesis 2: Following carcass sides will have a higher level of microbiological contamination to leading carcass sides due to the evisceration process

Table 25 and Figure 2 provide a summary of the total sample sets (including split samples) and the number of positive sample sets by carcass side. A two-sample proportional test (z test) was performed to determine whether there was a significant difference between leading and following carcass sides with respect to the percent of positive results. There is no significant difference to a confidence interval of 95%. As such with these results, hypothesis 2 is disproven.

Table 25 – Summary of Results based on Carcase Side

Sampling location	No. of Sample Sets	Carcase Side		Analysis
		Leading	Following	
Post Evisceration	Total	77	74	The Z-Score is -0.7211. The p-value is 0.47152. The result is <i>not</i> significant at $p < 0.05$.
	Positive	38	44	
	Percentage of Positive	53.5%	59.5%	
Chiller	Total	81	83	The Z-Score is -0.1412. The p-value is 0.88866. The result is <i>not</i> significant at $p < 0.05$.
	Positive	13	14	
	Percentage of Positive	16.0%	16.9%	
Totals	Total	152	157	The Z-Score is -0.6235. The p-value is 0.53526. The result is <i>not</i> significant at $p < 0.05$.
	Positive	51	58	
	Percentage of Positive	33.6%	36.9%	

Figure 2 – Summary of Results based on Carcase Side



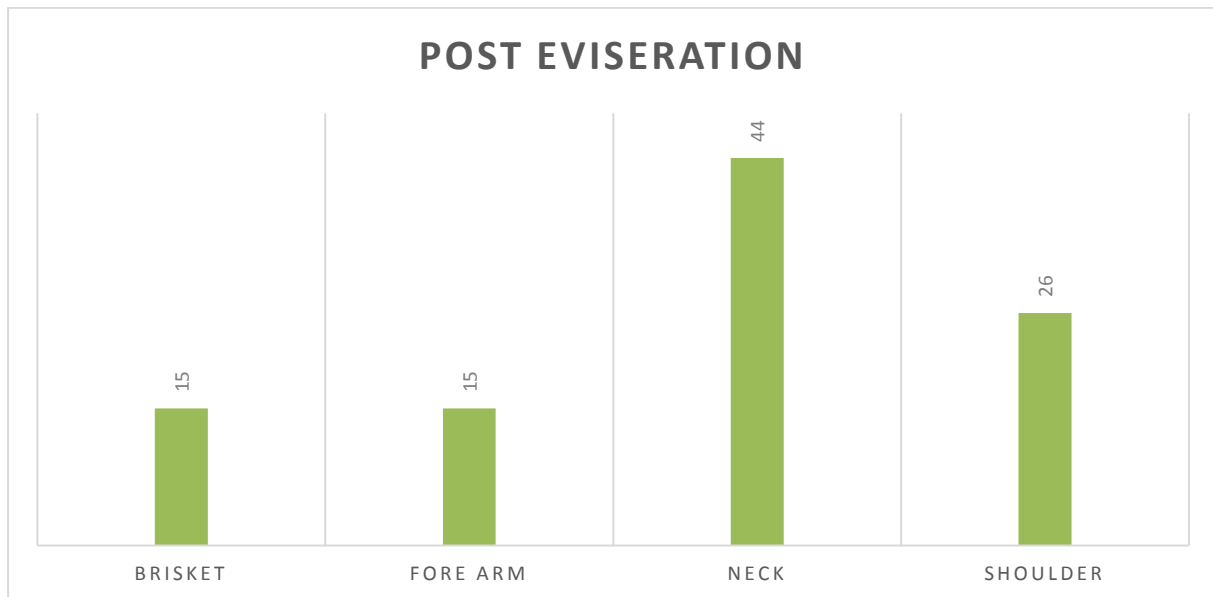
5.3 Hypothesis 3: Alternative sampling sites provide a greater number of unacceptable test results to assess hygienic processing compared to the traditional ESAM sampling sites.

Table 26 and Figure 3 provide a summary of the total sample sets and the number of positive sample sets by sampling site at post evisceration.

Table 26 – Summary of Post Evisceration Results based on Sampling Site

Post Evisceration	Sampling Site			
No. of Sample Sets: 145	Brisket	Fore Arm	Neck	Shoulder
No. of Positive Sample Sets	23	23	65	39
Percentage	15.9%	15.9%	44.8%	26.9%

Figure 3 – Summary of Post Evisceration Results based on Sampling Site



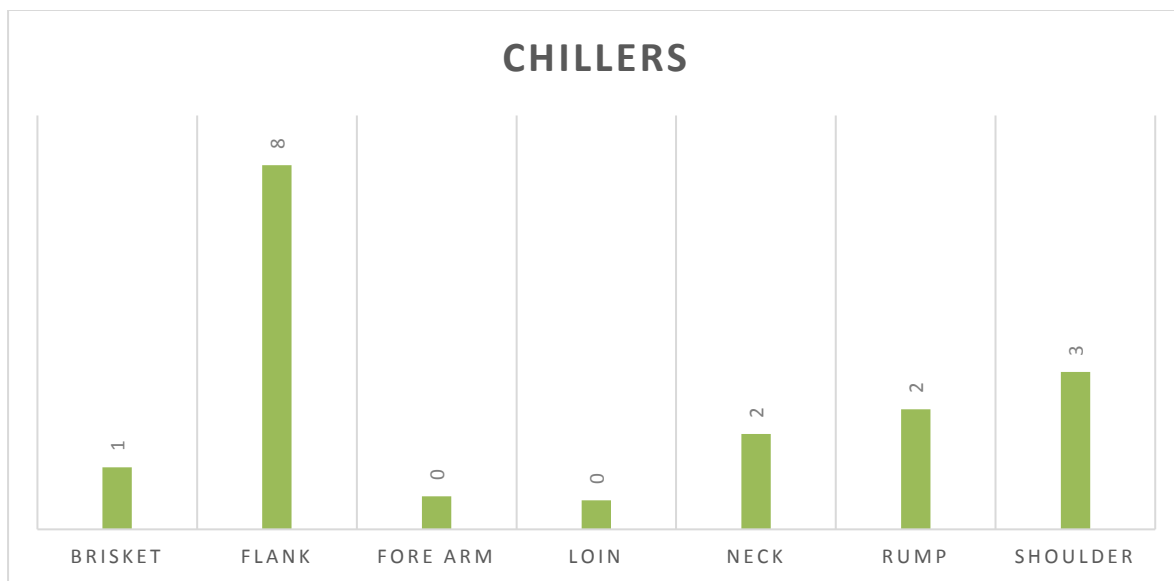
Two-sample proportional tests (z tests) were performed to determine whether there was a significant difference between the conventional brisket sampling site and the alternative sampling sites (fore arm, neck and shoulder) with respect to the percent of positive results. For the comparison of fore arm to brisket the Z-Score is 0 and the p-value is 1. This difference is therefore not significant at a 95% confidence interval. For the comparison of neck to brisket the Z-Score is 5.3645 and the p-value is 0. The difference is therefore significant at a 95% confidence interval. For the comparison of shoulder to brisket the Z-Score is -2.2917 and the p-value is 0.02202. The difference is therefore significant at a 95% confidence interval.

Table 27 and Figure 4 provide a summary of the total sample sets (including split samples) and the number of positive sample sets by sampling site in the chillers.

Table 27 – Summary of Chiller Results based on Sampling Site

Chiller	Sampling Site						
No. of Sample Sets: 133 fore quarter 136 hind quarter	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder
No. of Positive Sample Sets	2	12	1	1	3	4	5
Percentage	1.5%	8.8%	0.8%	0.7%	2.3%	2.9%	3.8%

Figure 4 – Summary of Chiller Results based on Sampling Site



Two-sample proportional tests (z tests) were performed to determine whether there was a significant difference between the conventional sampling sites (brisket, flank and rump) and the alternative sampling sites (fore arm, loin, neck and shoulder) with respect to the percent of positive results. These results are available in Table 28. The only significant differences in the results are between the flank sampling site and either the fore arm, loin or neck sampling sites.

Table 28 – Comparative Analysis of the Conventional Sampling Sites to the Alternative Sampling Sites in the Chillers.

Alternative Sampling Sites	Conventional Sampling Sites		
	Brisket	Flank	Rump
Fore Arm	The Z-Score is 0.5806. The p-value is 0.56192. The result is not significant at p <0.05.	The Z-Score is 3.0863. The p-value is 0.002. The result is significant at p <0.05.	The Z-Score is 1.3292. The p-value is 0.18352. The result is not significant at p <0.05.
Loin	The Z-Score is 0.6001. The p-value is 0.5485. The result is not significant at p <0.05.	The Z-Score is 3.1265. The p-value is 0.00174. The result is significant at p <0.05.	The Z-Score is 1.3541. The p-value is 0.17702. The result is not significant at p <0.05.
Neck	The Z-Score is -0.4515. The p-value is 0.65272. The result is not significant at p <0.05.	The Z-Score is 2.3471. The p-value is 0.01878. The result is significant at p <0.05.	The Z-Score is 0.3531. The p-value is 0.72634. The result is not significant at p <0.05.
Shoulder	The Z-Score is -1.1491. The p-value is 0.25014. The result is not significant at p <0.05.	The Z-Score is 1.7067. The p-value is 0.08726. The result is not significant at p <0.05.	The Z-Score is -0.3731. The p-value is 0.71138. The result is not significant at p <0.05.

The ESAM sampling program uses a single swab to sample the brisket, flank and rump sampling sites. Given the significant difference between the neck and shoulder sampling sites and the brisket sampling site at post evisceration the results have been considered on the basis of at least one positive result in a sample set across the conventional ESAM sampling sites compared to alternative sample sites of the neck, shoulder and flank. These results are summarised in Table 29 with the analysis of comparison.

Table 29 – Summary of the No. of Positive Sample Set Results when considering Conventional Sampling Sites versus Alternative Sampling Sites

	Conventional Sampling Sites	Alternative Sampling Sites	Analysis
Post Evisceration: No. of Sample Sets: 145			
No. of Positive Sample Sets	23	74	The Z-Score is 6.3475. The p-value is 0. The result is significant at $p < 0.05$.
Percentage	15.9%	51.0%	
Chiller: No. of Sample Sets: 164			
No. of Positive Sample Sets	17	20	The Z-Score is 0.5236. The p-value is 0.60306. The result is not significant at $p < 0.05$.
Percentage	10.4%	12.2%	

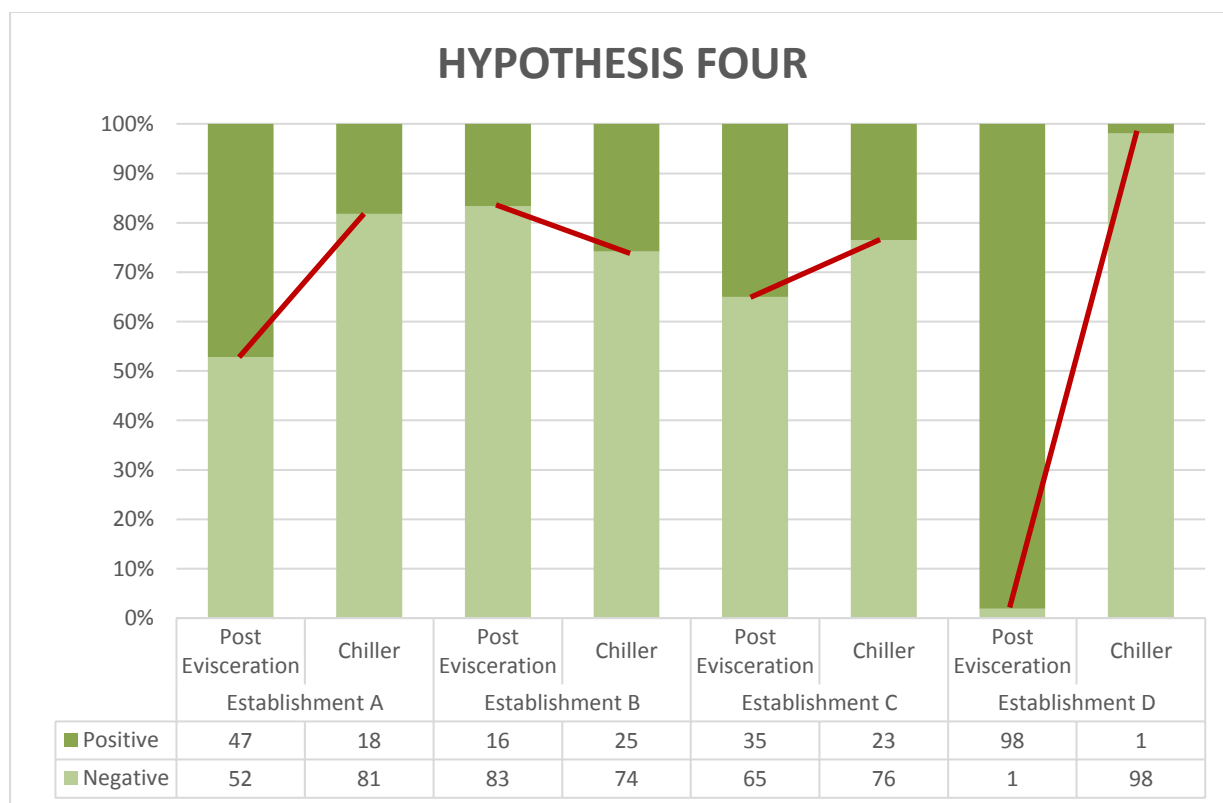
Although the results show that a higher number of sample sets have a least one positive result at the alternative sampling sites compared to the conventional sampling sites at both the post evisceration and in the chillers, there is only a significant difference (to a confidence interval of 95%) in the post evisceration results.

Based on these results while the alternative sampling sites of the neck, shoulder and flank provide a greater number of unacceptable test results to assess hygienic processing compared to the traditional ESAM sampling sites, this increase at the alternative sampling sites has no significant difference either greater or lesser to the conventional sampling sites based on this research and these results.

5.4 Hypothesis 4: Hygienic process control can be assessed based on the test results throughout processing.

Figure 5 provides the percentage of sample sets with positive results through processing i.e. at post evisceration and in the chillers by establishment. This demonstrates that three of the four establishments that participated in the trial had a decrease in positive results from post evisceration to chilling and therefore an improved process hygiene. Figure 5 demonstrates how meaningful a tool the analysis of samples collected at the two locations can be in understanding the level of hygiene process control.

Figure 5 - Percentage of Sample Sets with Positive Results through Processing by Establishment.



For example, if only chiller samples are used to assess process hygiene the chiller results from Establishment B and Establishment C could be assessed as comparable with no significant difference to a confidence interval of 95% (Z-Score = 0.1913 and p-value = 0.8493). Similarly, assessment of only slaughter floor data could lead to the conclusion that Establishment D had significant process hygiene control problems however, the comparison of results throughout processing (at post evisceration and at chilling) demonstrate that the establishment have their process under complete control. Using both slaughter floor post evisceration sampling and chiller sampling provides a better evaluation of process hygiene control.

5.5 Summary of Analysis and Interpretation of Results

Samples were taken from four beef establishments down the Eastern seaboard of Australia. The samples were collected from the neck, shoulder, fore arm and brisket on the slaughter floor and chiller. An additional three samples at the rump, loin and flank were taken in the chillers; 309 sample sets in total, with 1520 swabs being tested for *E. coli*, coliforms and Total Viable Counts using the approved ESAM methodology. The analysis of the data demonstrates that:

- A summary of the results based on cattle cleanliness score shows a clear trend to the null hypothesis, i.e. cattle cleanliness score is not positively correlated to the level of microbiological contamination found on carcasses. In fact, in the post evisceration results the trend is of a negative correlation. However, these results must be interpreted considering the sampling variable of establishment and their individual process hygiene levels. Due to the results for Cattle Cleanliness Scores 1 and 3 coming solely from Establishment A and B respectively. As such with these results, cattle cleanliness score being positively correlated to the level of microbiological contamination found on carcasses cannot be proven or disproven.

- A summary and analysis of the total sample sets (including split samples) and the number of positive sample sets by carcass side proved there is no significant difference to a confidence interval of 95% between following carcass sides having a higher level of microbiological contamination to leading carcass sides due to the evisceration process.
- Based on these results while the alternative sampling sites of the neck, shoulder and flank provide a greater number of unacceptable test results to assess hygienic processing compared to the traditional ESAM sampling sites, this increase at the alternative sampling sites has no significant difference either greater or lesser to the conventional sampling sites based on this research and these results.
- The results demonstrate how meaningful a tool the analysis of samples collected at the two locations can be in understanding the level of hygiene process control rather than the use of a single location.

6 Discussion

6.1 Review of current food safety systems and work practices, and assessment of current and potential new testing sites to more accurately reflect the level of hygienic processing; and

Based on the literature review there was evidence to suggest that investigation of alternative sampling sites was warranted. A number of studies identified the neck as a suitable site (Untermann et al 1997; Zweifal et al 2008). Forearm and shoulder were also identified as suitable microbiological sampling sites (Untermann et al 1997).

Location on the processing chain of sampling was also warranted. It was proposed that sampling be conducted post evisceration i.e. at the point of most contamination. Blagojevic et al (2011) identified the importance of measuring the effectiveness of processing hygiene however in sampling prior to de hidding, they did not capture any potential processing contamination that occurs due to the de hidding and evisceration process. It was proposed that sampling be conducted post evisceration and again at the end of the slaughter floor post chilling. Comparison of the data obtained at the two points would potentially determine whether moving sampling to post evisceration and adjusting the acceptable microbiological limits would be a valid and beneficial change for the microbiological sampling of beef. The ratio between the two samples could also be considered, as per the study conducted by Blagojevic et al (2011). This would determine whether the ratio of microbiological contamination between the two points was a valid measure of process hygiene, which could also allow for exploration of the European and United States of America's approach of risk based monitoring over inspection sampling monitoring.

Given the research conducted to date and the Australian microbiological results from the ESAM program and the Australian baseline surveys consideration was given as to the indicator organisms assessed. As such, it is proposed that *E.coli* and coliform testing will be conducted using swab sampling during the project.

In summary, the conclusion from the literature is that there is no current regulatory barrier to changing the microbiological testing requirements in Australian beef abattoirs on the basis of risk. Furthermore, the literature proposes alternatives which may yield more meaningful process control outcomes. Microbiological process control could be enhanced if considered in conjunction with

macroscopic indicators (such as hide cleanliness) which also have the benefit of being determined in real time. To assess this we proposed to test carcasses from livestock with graded hide cleanliness during processing and post intervention at varying sites on the carcase, with comparative ESAM site samples, taking into account leading and following carcase side variation.

6.2 Make recommendations for industry and regulatory adoption

When considering recommendations it must be remembered that the current ESAM program was design to provide US market access, during the past 21 years the program has also been used to meet the minimum expectations of a number of other customer and trading partners. Whether or not it is of scientific merit is therefore a minimum standard for the industry. The reality therefore is that any changes to this program would need to be seen as beneficial by customer or trading partners irrespective of the financial benefit to Australian processors.

This project aimed to review the current food safety programs (both regulatory and commercial) and assessing whether or not it might be more beneficial to

- a) undertake microbiological testing at other points in the process and/or
- b) other parts of the carcase that allow for evaluation of hygiene process to reflect risk.

Should one or both be the case, the opportunity arises for industry to ask the Department of Agriculture and Water Resources to make an equivalency submission to trading partners (and to customers) to accept these new testing parameters in lieu of the current testing methodology.

Based on the results while the alternative sampling sites of the neck, shoulder and flank provide a greater number of unacceptable test results to assess hygienic processing compared to the traditional ESAM sampling sites of rump flank and brisket, this increase at the alternative sampling sites has no significant difference either greater or lesser than the conventional sampling sites based on this research and these results. Therefore, you could argue that the alternative sites are just as effective and if a processor wished to use these as an alternative to the traditional ESAM sites due to Work Health and Safety (as in a number of cases these sites can be assessed without a step ladder) this research supports that variation. However, it is recognised that these benefits made not be cost-beneficial when considering the requirement of an equivalence agreement, especially if more important trade barriers or breaches are being negotiated.

A summary of the results based on cattle cleanliness score shows a clear trend to the cattle cleanliness score not be being positively correlated to the level of microbiological contamination found on carcasses. In fact, in the post evisceration results the trend is of a negative correlation. However, these results must be interpreted considering the sampling variable of establishment and their individual process hygiene levels. As such with these results, cattle cleanliness score being positively correlated to the level of microbiological contamination found on carcasses cannot be proven or disproven and therefore Jordan et al (2014) stands. Testing at two location, post evisceration and in the chiller, to measure hygienic process efficiency has demonstrate how meaningful a tool the analysis of samples collected at the two locations can be in understanding the level of hygienic process control rather than the use of a single location. (This concurs with work completed by Blagojevic et al 2011). With regulatory agreement and equivalence from customers and trading partners this process would not only allow for a better understanding of process hygiene by all, but could be used as a risk framework with cattle cleanliness for testing frequency. For example, based on this trial Plant A, demonstrating a high level of process hygiene for cattle cleanliness score one and could therefore be on the lowest frequency of testing however

Establishment B, with weak hygienic processing control for cattle cleanliness score 2 and 3 would be on the highest frequency of testing level.

6.3 Draft paper for publication on the results of the trialled sites and draft market access proposals to vary current equivalence agreements

A paper for publication on the results of the trialled sites has been drafted to the Meat Science publication requirements (Appendix 8). A Export Meat Industry Advisory Committee and Food Export Regulatory Steering Committee Paper has been drafted (Appendix 9) as the first step required for Industry to submit to the Department of Agriculture and Water Resources and State Regulatory Authorities if they wish to proceed with a variation and seeking equivalence to the current process hygiene testing requirements in line with the research conducted under this project.

7 Conclusions/recommendations

During the literature review it was identified that in a draft guidance document for comment, United States Department of Agriculture, Food Safety and Inspection Service (2008) noted that high frequency and extensive testing has the potential to be cost prohibitive for a number of small and very small abattoirs. In response, the draft guidelines outlined minimum sampling frequencies for small and very small establishments for the testing of finished ground product for *E.coli* O157:H7. FSIS also recommended increasing sampling rates in the warmer months based on studies that indicated shedding of *E.coli* O157:H7 by cattle is greater during this time. Although *E.coli* O157:H7 is outside of the scope of the project this document does indicate an approach that allows for variation in testing frequency based on risk.

In addition, the United Kingdom's Food Standards Agency (2015) have produced the Meat Industry Guide that outlines the legal obligations of food operators in the meat sector. Chapter 13, Annex 1 of this document outlines the sampling frequency for red meat carcasses. The sampling plan is structured with initial sample frequency based on throughput, however satisfactory results over a given time period results in a reduction in sampling frequency (Food Standards Agency 2015). Such an approach has the potential to further incentivize hygienic production as well as deliver savings to industry in time and resources.

Should the Industry wish to assess process hygiene on a risk based approach rather than standard frequency sampling this research project provides evidence that a more realistic risk management framework for process control can be used for microbiological testing providing a true indicators of hygienic process control.

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8.2 Testing and analysis of potential new sites for sample collection

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9 Appendix 1: Literature Review

Consumers expect that their food is safe. Food safety is dependent on a number of factors up and down the supply chain and is achieved through shared responsibility and cooperation by producers, processors, consumers and regulators. However, foodborne illnesses and recalls do occur. Food Standards Australia New Zealand (FSANZ) classify food recalls under a number of categories, including Microbial contamination. In a 10year period from 1 January 2006 to 31 December 2015, 31% of all recalls were due to microbial contamination (FSANZ 2016). Food borne illness was also one of the drivers for the development and implementation of the Pathogen Reduction: Hazard Analysis and Critical Control Point (HACCP) Systems Final Rule by the United States of America (USDA 1996). In the Final Rule, Food Safety and Inspection Service of the United States Department of Agriculture provided estimates of 4000 deaths and 5 million illnesses annually due to contamination of meat and poultry products with bacteria such as *Salmonella*, *E. coli* O157:H7, *Campylobacter* and *Listeria monocytogenes* (USDA 1996).

The slaughtering and processing of beef involves a number of processes including stunning, sticking, carcass dressing, cooling and carcass breaking (Gill 2005). The muscle of the healthy animal is generally considered to be sterile, however bacteria can be transferred to the meat during the carcass dressing process. Sources of contamination include contamination from the hide, from either hide rolling under at the cut edges, blades of knives cutting through the hide and various other means of transfer (Gill 2005). Airborne contamination has also been identified and investigated as a source of carcass contamination (Chandry 2016). Meat may also be contaminated by ingesta and bacteria through incidences such as outflow of ingesta with manipulation of the oesophagus or rupture of the paunch during the evisceration process (Gill 2005). Plant equipment, walls, floor, fixtures, fitting and personnel are also potential sources of contamination (Gill 2005).

The aim of the project is to review current food safety systems and work practices to identify possible new testing requirements to demonstrate process control. These new testing requirements will be assessed against the current microbiological testing requirements with the aim to provide industry with more accurate, meaningful and cost beneficial testing program that truly reflects the level of hygienic processing.

Australian food safety regulation is a tiered system in which all abattoirs must meet domestic regulatory requirements for state government licensing. Abattoirs seeking to produce export product must comply with additional legislation and guidelines for them to be registered with the federal Department of Agriculture and Water Resources. Dependent on the country of import additional requirements may need to be met despite continued trade negotiation for acceptance of only the Australian Standard. In addition to these regulated requirements, commercial buyers impose their own microbiological testing requirements with the aim to ensure customer safety.

Fundamentally, Australian food safety regulation requires the occupier of an [export-registered] abattoir to take primary responsibility for compliance with food safety objectives while the regulator is responsible for verifying that these objectives have been met (AQIS 2009).

All Australian abattoirs processing meat and meat products are required to comply with the Australian Standard - *Hygienic Production and Transportation of Meat and Meat Products for Human*

Consumption (AS 4696:2007) through state government legislation. This Standard requires that meat is wholesome and defines wholesome as:

When used in relation to meat and meat products meant that the meat and meat products may be passed for human consumption on the basis that they:

- (a) are not likely to cause food borne disease or intoxication when properly stored, handled and prepared for their intended use; and*
- (b) do not contain residues in excess of established limits; and*
- (c) are free of obvious contamination; and*
- (d) are free of defects that are generally recognized as objectionable to consumers; and*
- (e) have been produced and transported under adequate hygiene and temperature controls; and*
- (f) do not contain additives other than those permitted under the Food Standards Code; and*
- (g) have not been irradiated contrary to the Food Standards Code; and*
- (h) have not been treated with a substance contrary to a law of the Commonwealth or a law of the State or Territory in which treatment takes place.*

The Australia New Zealand Food Standards Code, as referred to in AS 4696:2007 contains standards related to the composition, labelling, safe handling and primary production of foods (FSANZ 2016).

State Governments through the Meat Standards Committee (MSC) also mandated the requirement for microbiological testing of the product and contact surfaces at approximately the same time as the introduction of the Federal Government's *E.coli* and *Salmonella* Monitoring (ESAM) Program (AQIS 2000). These testing requirements were implemented and enforced in the domestic sector by state regulators (AQIS 2000). In 2002 the Meat Standards Committee published the Microbiological Testing for Process Monitoring in the Meat Industry Guidelines (MSC 2002). This guideline requires the testing of product and working surfaces (i.e. facilities, equipment and personnel). Cleaned work surfaces are tested for Total Viable Counts of micro-organisms through swabbing and plating, or contact plating methods. Testing is rotated around the plant to provide an overall assessment and is done at a frequency that demonstrates whether the cleaning program is effective (MSC2002). Product can be sampled by either excision or swabbing. The swabbing technique requires samples of 10cm x 10cm at both the flank and brisket area conducted after active chilling has cooled and dried the surface (MSC 2002). Abattoirs processing more than 150 cattle per week are required to establish a baseline through intensive sampling, with the frequency of sampling reducing once a satisfactory baseline has been demonstrated (MSC 2002). Very small premises processing less than 150 cattle per week are required to sample at a frequency that demonstrates hygienic processing (MSC 2002). Samples are tested for *E.coli* and Total Viable Count (MSC 2002).

Export registered abattoirs are required to comply with the *Export Control (Meat and Meat Products) Orders 2005*, which in turn require compliance with AS 4696:2007. Under the legislation requirements have been added through the development of guidelines. These include the testing requirements for equipment and personnel which mirror the Meat Standards Committee's Microbiological Testing for Process Monitoring in the Meat Industry Guidelines (AQIS 2009) as well as the *E.coli* and *Salmonella* Monitoring (ESAM) Program.

The *E.coli* and *Salmonella* Monitoring (ESAM) program

Microbiological testing requirements for generic *E.coli* and *Salmonella* were initially introduced in Australia for export-registered abattoirs with market access to the United States of America, effective from 27 January 1997 (AQIS 1996). This monitoring program was implemented in response to the publication of the 'USA Pathogen Reduction Final Rule' by the United States Department of Agriculture on 25 July 1996 (AQIS 1996).

The United States Department of Agriculture's Food Safety and Inspection Service (FSIS) developed the Pathogen Reduction: Hazard Analysis and Critical Control Point (HACCP) Systems Final Rule to reduce the occurrence and number of organisms on meat as well as reduce the number of foodborne illnesses associated with the consumption of meat (USDA 1996). This Final Rule included requirements to undertake microbiological testing to verify the process controls at the abattoirs and the establishment of pathogen reduction standards for *Salmonella*, which would then need to be met by abattoirs (USDA 1996). This final rule was also intended to make better use of agency resources and provide the framework for the modernization of meat and poultry inspection (USDA 1996) *Salmonella* was selected as the target (indicator) organism for pathogen reduction as it was,

- the most common bacterial cause of foodborne disease,
- colonized a variety of mammals,
- recoverable from meat using the methodologies at the time and
- intervention strategies aimed at reducing sources of *Salmonella* were believed to be effective against other pathogens (USDA 1996).

E.coli was selected as the criteria for process control verification as faecal contamination was considered the primary avenue for contamination of the carcass by pathogens and the scientific community viewed *E.coli* as the best microbial indicator for faecal contamination (USDA 1996).

The first iteration of the ESAM program required:

- Cattle to be sampled after 12 hours of active chilling with a minimum of 8 hours where carcasses were loaded out sooner;
- Hot boned/warm cut carcasses to be drawn for sampling after completion of dressing and immediately prior to exiting the slaughter floor;
- Two sites were to be sampled for cattle – the flank and the brisket (Diagram 1, AQIS 1996);
- Site area to be swabbed was 10cm x 10cm
- All cattle types (i.e. Steers/heifers/cows/bulls) to be sampled at a frequency of 1 test per 300 carcasses for *E.coli*
- A sub-sample from every fifth sample to be tested for *Salmonella* (effective sampling frequency 1 test per 1500 carcasses).

These testing frequencies appear to have been taken from the frequency set by the FSIS which were calculated based on the processing levels and size of abattoirs existing in the United States of America in 1996 to ensure every abattoir conducted a minimum of one test daily (USDA 1996).

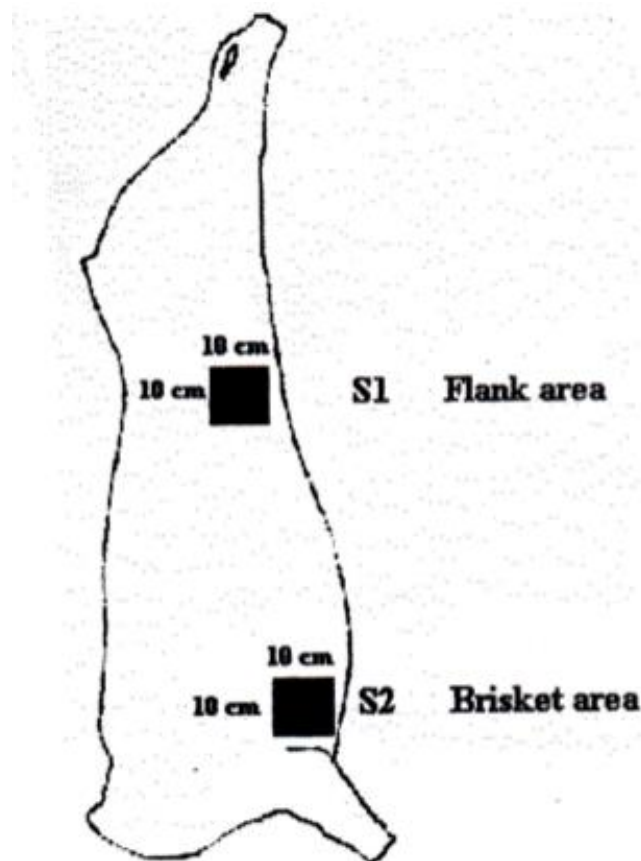


Diagram 1: Sampling location for *E.coli* testing for Steer/Heifer and Cow/Bull carcasses (AQIS 1996).

E.coli results were interpreted using a three class attribute sampling plan applied in a moving window. Cut-offs were denoted by [m] for the lower limit of the marginal range and [M] for the upper limit of the marginal ranges. Results were defined as Acceptable (result less than or equal to [m]), Marginal (result between [m] and less than or equal to [M]) or unacceptable (greater than [M]) (AQIS 1996).

Table 1: Parameters for interpreting *E.coli* testing results (AQIS 1996)

Species	Lower limit of marginal range [m]	Upper limit of marginal range [M]	No. of samples tested [n]	Maximum no. permitted in marginal range [c]
<i>Steers/heifers</i>	Negative*	100cfu/cm ²	13	3
<i>Cows/bulls</i>	Negative*	100cfu/cm ²	13	3

* Negative is defined by the sensitivity of the method used with a limit of sensitivity of at least 5cfu/cm² carcass surface area

The scope of the ESAM program was extended to include all export-registered abattoirs in late 1997 (AQIS 1997). Since its induction nearly 20 years ago, there have been few changes to the ESAM program. Total Viable Count (TVC) testing of carcasses was introduced, initially on a voluntary basis in

1997 (AQIS 1997). This increased the number of tests to 2 tests per sample, as TVC testing was conducted at the same frequency as *E.coli* testing and could be conducted on a sub sample of the test taken for *E.coli*. (AQIS 1997). In 1998, a third sampling site at the rump was introduced for cattle, which resulted in an increased sample area of 300cm². An additional independent sample for the *Salmonella* testing was introduced in 1999 following concerns from microbiologists and importing countries about the sensitivity of the testing (AQIS 1999).

The requirement for TVC testing became a mandatory component of the ESAM program for export-registered abattoirs with market access to the European Union (EU) following the EU audit in 2005 (AQIS 2007).

In 2009, the AQIS introduced the Product Hygiene Index (PHI), with abattoirs required to begin data collection and submission between 1 October 2009 and 1 April 2010 (AQIS 2009). The PHI was intended to allow individual abattoirs to compare their own data against a national baseline of the same data (AQIS 2009). The key performance indicators (KPIs) selected for inclusion in the PHI were those deemed to have a direct bearing on product hygiene and/or the potential for product re-contamination and included *E.coli* on carcasses and Meat Hygiene Assessment of the slaughter floor, offal room and boning room (AQIS 2009). TVC on carcasses continued to be measured, but was not included in the initial PHI (AQIS 2009).

In 2013, TVC and coliform counts on carcass samples became a mandatory part of the PHI (DAFF 2013). Final carton product Aerobic Plate Count and coliform counts also became mandatory and were required to be captured on the PHI data entry sheet (DAFF 2013). Samples were to be taken at the same frequency as *E.coli* for the ESAM carcass samples. For example if an establishment was required to take 3 carcass samples, they would be required to take 3 carton samples. Assuming 1 beef carcass produces 8 cartons of meat, an establishment would need to sample 1 in 2400 cartons to have a sampling frequency equivalent to 1 in 300 carcasses (DAFF 2013).

Table 2 provides an example of the changes in mandatory regulatory testing requirements and the resultant changes to the number of sites swabbed and number of tests conducted on carcasses for two example abattoirs. Abattoir A processes 300 head per day without market access to the United States of America and the European Union and Abattoir B processes 1200 head per day with market access to the United States of America and the European Union.

Table 2: Demonstration of mandatory regulatory test requirements over the last 20 years

Time Period (years)	Abattoir A (300 hd/day, not exporting to EU or USA)	Abattoir B (1000 hd/day, exporting to EU and USA)
1997	Jan 97: Nil	Jan 97: <ul style="list-style-type: none"> • <i>E.coli</i> biotype1: 1 test/300 carcasses • <i>Salmonella</i>: Sub sample of every 5th <i>E.coli</i> test (1 test/1500) • Two swab sites: brisket and flank <p>Total number of sites swabbed/week: 40 Total number of tests/week: 24</p>

	<p>Nov 97 new requirements:</p> <ul style="list-style-type: none"> • <i>E.coli</i> biotype1: 1 test/300 carcasses • <i>Salmonella</i>: Sub sample of every 5th <i>E.coli</i> test (1 test/1500) • Two swab sites: brisket and flank <p>Total number of sites swabbed/week: 10 Total number of tests/week: 6</p>	<p>Nov 97 No new requirements</p> <p>Total number of sites swabbed/week: 40 Total number of tests/week: 24</p>
1998	<ul style="list-style-type: none"> • Rump site added to ESAM <p>Total number of sites swabbed/week: 15 Total number of tests/week: 6</p>	<ul style="list-style-type: none"> • Rump site added to ESAM <p>Total number of sites swabbed/week: 60 Total number of tests/week: 24</p>
1999	<ul style="list-style-type: none"> • <i>Salmonella</i> testing to be conducted with a dedicated sample, NOT a sub sample of sample collected for <i>E.coli</i> <p>Total number of sites swabbed/week: 18 Total number of tests/week: 6</p>	<ul style="list-style-type: none"> • <i>Salmonella</i> testing to be conducted with a dedicated sample, NOT a sub sample of sample collected for <i>E.coli</i> <p>Total number of sites swabbed/week: 72 Total number of tests/week: 24</p>
2007	<p>Nil Change</p> <p>Total number of sites swabbed/week: 18 Total number of tests/week: 6</p>	<ul style="list-style-type: none"> • Mandatory TVC testing EU listed establishments • Sampling and sample rates as per <i>E.coli</i> • Testing can be conducted on same swab sample as <i>E.coli</i> <p>Total number of sites swabbed/week: 72 Total number of tests/week: 44</p>
2013	<ul style="list-style-type: none"> • Mandatory TVC and coliform counts for all export registered establishments • Sampling and sample rates as per <i>E.coli</i> • Testing can be conducted on same swab sample as <i>E.coli</i> • Mandatory carton testing for APC and coliform count 1 test /300 carcasses <p>Total number of sites swabbed/week: 18 Plus 5 carton samples Total number of tests/week: 26</p>	<ul style="list-style-type: none"> • Mandatory coliform counts for all export registered establishments • Sampling and sample rates as per <i>E.coli</i> • Testing can be conducted on same swab sample as <i>E.coli</i> • Mandatory carton testing for APC and coliform count 1 test /300 carcasses <p>Total number of sites swabbed/week: 72 Plus 20 carton samples Total number of tests/week: 104</p>

Microbiological baseline studies have been conducted periodically in Australia. There were three baseline surveys of beef microbiology undertaken between 1993 and 2004: (Phillips et al 2006; Phillips et al 2001; Vanderlinde et al 1998). Improvements in microbiological criteria of beef

carcasses and carton beef were noted, when taking into account differences in sampling and laboratory analysis (Desmarchelier et al 2007). Indicator organism measures generally trended downwards and pathogens were at very low prevalence levels over these studies (Phillips et al 2012). A further baseline study was conducted in 2011 (Phillips et al 2012). Pathogen recovery continued to be very low in the fourth baseline study, however indicator organisms were slightly elevated in this study when compared to previous baseline studies (Phillips et al 2012). It was suggested that this might be attributed to the extreme rain events that occurred during the timeframe of the baseline study (Phillips et al 2012). Additional baseline studies including for salmonella prevalence are currently being undertaken.

The *E.coli* and *Salmonella* Monitoring Report provide by SARDI on National Cow/Bull regulatory testing for the period 1 June 2013 to 31 May 2016 provides a summary of the ESAM program. Abattoirs have reported the national statistics over the 3 year period are a 5.4 % prevalence of *E.coli*, a 13.5 % prevalence of Coliforms and a 0.4% prevalence of Salmonella.

As can be seen through the evolution of the ESAM Program, Export registered abattoirs may also be required to comply with additional importing country requirements for microbial sampling.

A number of markets have no known specific requirements for microbial sampling in addition to the ESAM Program. The Australian Government Department of Agriculture and Water Resources Manual of Importing Country Requirements (MICoR) (2015) indicates the current additional microbial sampling requirements by country. MICoR does not provide advice where the relevant importing country requirement is considered to be addressed through Australian requirements such as ESAM. These importing country requirements may therefore range from nil to equivalent to ESAM notwithstanding that ESAM is required for all markets. It should be noted that in the general principles of meat hygiene under the Code of Hygienic Practice for Meat of Codex Alimentarius (CODEX 2005), importing countries,

'should recognise the equivalence of alternative hygiene measure where appropriate, and promulgate meat hygiene measures that achieve required outcomes in terms of safety and suitability and facilitate fair practices in the trading of meat'

This means that should the recommendation at the end of this project be that the microbiological testing and monitoring program for hygiene controls be changed then a scientific argument of equivalence can be made to importing countries with regard to the change in practice.

Food Safety and Process Control Research in the Meat Industry

Over the last 20 years since the ESAM program was introduced millions of dollars and countless hours have been spent internationally researching food safety and process control when producing meat. To aid in the identification of potential new microbiological testing arrangements to more accurately reflect the level of hygienic processing the key areas of indicator organisms, hide cleanliness, sampling technique, sites, size, frequency, location and timing during processing were investigated.

Indicator Organisms

Indicator organisms are a key part of quality assurance and regulation both in Australia and internationally (Jordan et al 2007). In the case of the meat industry, an indicator organism is usually present in higher numbers with a greater, more uniform distribution on the carcass. (Wang et al 2013). A number of researchers have considered the use of indicator organisms to demonstrate effective and efficient control of process or alternatively discussed the correlation between organisms as a part of their study findings.

The meat industry pathogens such as Salmonella and Shiga toxin producing *E.coli* strains such as O157:H7 present the opposite qualities occurring too infrequently and with an uneven distribution and with very low numbers when present. These organisms are therefore poorly suited as a measure of process control (Blajoveic et al 2011).

Jordan et al (2007) identified that although use of indicator organisms as a measure of process hygiene is common place, there is little information available to objectively describe the relationship between the different indicators under different processing conditions. To address this, a study was conducted by Jordan et al (2007) analysing aerobic plate count, *Enterobacteriaceae*, *E.coli* biotype I and coliforms data from Australian abattoirs to determine the relationship between concentrations of these organisms. The study found that coliform counts could be a useful indicator of how successfully processing prevents faecal contamination. This was because the measurement of coliforms could be determined with minimal additional effort while enumerating *E.coli* biotype I and because coliforms are present in faecal contamination at a higher concentration than *E.coli* biotype I (Jordan et al 2007). However coliforms would not be a suitable indicator for *E.coli* biotype I as testing would result in substantial false positives results (Jordan et al 2007). Presence of coliforms on beef carcasses was found to have a sensitivity of 100% but a specificity of 94% when used as a method of classifying the presence of *E.coli* biotype I (Jordan et al 2007). TVCs were also found to be inferior to both coliforms and *Enterobacteriaceae* in the prediction of *E.coli* biotype I (Jordan et al 2007). Presence of *Enterobacteriaceae* on beef carcasses was found to have a sensitivity of 98% and specificity of 88% when used as a method of classifying the presence of *E.coli* biotype I (Jordan et al 2007). These conclusions reflected the findings of Gill et al (1996) who concluded that while the correlation between *E.coli* and coliform counts is good, there is only a weak correlation between *E.coli* and aerobic counts. Gill et al (1996) suggested that to avoid misinterpretation of hygienic process, *E.coli* and possibly coliforms were better suited for assessment of process than aerobic counts.

In 2006, Tergney and Bolton undertook a study looking to reduce the incidence of faecal contamination and therefore microbiological counts in beef. They swabbed five carcass sites (hock, rump, anus, brisket and flank) at the final inspection stand and undertook TVC, total *E.coli* counts, total coliform counts and total enteric counts (Tergney and Bolton 2006). The study noted that there was no relationship between TVCs and any of the other microbiological counts. There was also no correlation between faecal contamination and TVCs and as such, TVC counts may be a good measure of general hygiene, however may not be suitable as a measure of carcass hygiene or food safety risk (Tergney and Bolton 2006).

Hide Cleanliness

While the classification and identification of macroscopic contamination was outside the scope of this project, macroscopic contamination was considered in terms of ensuring the identification of suitable and appropriate carcass sampling sites. It is generally accepted that the cleanliness of the animals and the procedures employed to decrease transfer of dirt to carcasses during dressing will help to control the transfer of bacteria to meat (Serraino et al. 2012). A number of studies considered the association between hide cleanliness and to what extent it affects the level of microbiological contamination.

Kiermeier et al (2006) conducted a study comparing ESAM data from the calendar year of 2003 to data obtained through a survey of 15 export registered abattoirs in Australia. The veracity of the information obtained through the questionnaire was confirmed through inspection of the sites during on plant visits (Kiermeier et al 2006). A component of this study was the development of a 'problem score' that was calculated based on three elements identified as contributing to hide contamination: how cattle were raised (pasture reared or feedlot), distance transported and a 'tag' score (Kiermeier et al 2006). The 'tag' score was used to determine the proportion of cattle that fit into each of five categories of visible contamination, where Tag 1 was clean cattle and Tag 5 was extremely dirty cattle (Kiermeier et al 2006). The study identified that establishments with problems with incoming stock coupled with poor processes showed higher than average *E.coli* prevalence (Kiermeier et al 2006).

Serraino et al (2012) looked at the correlation between cattle cleanliness and TVCs, *Enterobacteriaceae* counts and *E. coli* counts in an abattoir in Italy. Cattle were visually inspected and categorized on a scale of 1 – 5, where 1 was clean and dry and 5 was wet and filthy (Serraino et al 2012). Fifteen cattle from each cleanliness category were sampled (Serraino et al 2012). Hide samples were collected prior to de-hiding from the brisket, abdominal midline, rump and groin areas while carcass samples for microbiological testing and correlation were collected after carcass splitting (and prior to chilling) from brisket, flank, groin and hock areas (Serraino et al 2012). The brisket and abdominal raphe were identified as the most contaminated areas across all 5 points on the scale of one to five of cleanliness used in the study (Serraino et al 2012). This was attributed to these being the part of an animal that is most likely to be subject to contamination e.g. when lying down (Serraino et al 2012). *E.coli* count and *Enterobacteriaceae* count on hide and carcase was found to be correlated with visibly dirty animals (Serraino et al 2012). It was concluded that for the abattoir evaluated in this study, there was a rise in the amount of bacteria transferred to meat as the dirtiness of cattle and hides increased (Serraino et al 2012).

Burfoot et al (2011) used fluorescence imaging to detect contamination (faecal, hair and other contaminants, such as rail grease) on carcasses. Macroscopic contamination of beef carcasses was found to occur mostly on the legs and ventral cut line (Burfoot et al 2011). Their findings supported the observations of Serraino et al (2012) that contamination is mostly on the ventral aspect, noting that almost all contaminants were present along the legs and ventral cut line (Burfoot et al 2011). Burfoot et al 2011 also stated that faecal contamination was the most common contaminant identified, representing 62% of the contaminants identified.

A study in Norway aimed to evaluate microbiological contamination associated with hide cleanliness and processing (Hauge et al 2012). Norway categorised visual hide cleanliness on a scale of 0 – 2,

where 0 is clean, 1 is moderately dirty and 2 is very dirty (Hauge et al 2012). The study was conducted at 2 abattoirs with a total of 324 swab samples taken from abdomen and brisket areas (Hauge et al 2012). Sample sites were 100 cm² and samples were taken at the brisket and abdomen (1) just after de-hiding and (2) at the end of the slaughter line (Hauge et al 2012). The abattoir used hygienic measures normally used in Australia such as rodding the oesophagus and used the two-knife method, however spraying of carcasses was not undertaken at any time (Hauge et al 2012). Mean *E.coli* values for clean animals (Category 0, including the adjacent carcass class in this study) were lower than the mean *E.coli* values for dirty animals (Category 1 & 2 animals in this study) after de-hiding with mean values of 1.03 [0.78, 1.27] (95% CL) (Hauge et al 2012). However at the end of the slaughter line the mean values 0.37 [0.22, 0.52] (CL 95%) were approximately equal to one another (Hauge et al 2012).

Sampling Techniques, Sites, Location and Timing during Processing

The current ESAM program requires swab samples be taken from the rump, flank and brisket with the addition of carton sampling. Carcasses are sampled after 12 hours active chilling unless loaded out sooner, in which case the minimum time is 8 hours (AQIS 2000). Carton meat samples are collected as close to carton closure as possible (DAFF 2013).

There is extensive literature on the available sampling techniques for recovery of bacteria i.e. swab sampling or excision sampling. The literature provides evidence both for and against each technique however fundamentally the swab sampling is more commercially acceptable than the destructive technique of excision sampling where it can be avoided.

Understandably given the importance of food safety and complexity of process control (in part due to the variation in processing design) researchers have used a number of different sampling sites, locations and times during processing in their work. These have included neck, forearm, shoulder, brisket, back, flank and rump.

There is little uniformity in the time of sampling, from pre-hide removal through to 10-24 hours chilling. Studies conducted sampling at a number of different locations during the slaughter floor process including prior to hide removal, pre and post evisceration, pre and post trimming and during chilling.

Gill et al (1996) conducted a study in Canada sampling three sites (neck, brisket and rump) at four different points along the chain. Neck sites were sampled after de-hiding by down pulling, and after evisceration (Gill et al 1996). Brisket sites were sampled after side pulling of the hide and after evisceration (Gill et al 1996). Rump sites were sampled after de-hiding of the rump and after splitting of the carcass (Gill et al 1996). All sites were again sampled after trimming and after washing (Gill et al 1996).

Aerobic microflora (recovered from swabs following incubation at 25°C) was characterised and numbers of *E.coli* and coliforms determined (Gill et al 1996). It was found that the skinning process resulted in similar levels of contamination at all 3 sites (Gill et al 1996). In terms of final product, brisket was found to be the most contaminated site based on the aerobic count and *E.coli* data (Gill et al 1996).

Untermann et al (1997) conducted a study utilizing 9600 swab samples from multiple sites on 900 beef carcasses, sourced from 10 Swiss abattoirs. All samples were taken from the left half of the carcass only. These abattoirs were all running a mechanized slaughter line and samples were taken within 2 hours of leaving the slaughter floor. Untermann et al (1997) looked at sample sites on both the medial (top round, pelvis canal, pleura, neck medial) and lateral (neck lateral, forearm, shoulder, brisket, abdomen, round, back) surfaces of the carcass. Plated samples were incubated aerobically and anaerobically and colony counts were determined. It was concluded that of the medial sites, only the neck site could be considered as a justifiable sampling site based on the colony counts. On the lateral carcass, the brisket and forearm had the highest contamination rates consistently (Untermann et al. 1997). Carcass neck, forearm, shoulder, brisket and abdomen were all identified as recommended lateral sampling sites (Untermann et al. 1997).

Zweifal, Fischer and Stephan (2008) also identified brisket and neck locations as yielding the highest mean log for TVCs, following excision sampling of neck, brisket, flank and rump of Swiss beef carcasses within 3 hours of chilling, with significant differences noted between neck and flank sites. While neck sites also showed higher *Enterobacteriaceae* count, there was no significant difference between neck and brisket and rump. (Zweifal et al 2008).

Blagojevic et al (2011) provided an explanation of the European process hygiene criterion (PHC), which took a different approach to find an indicator of functioning HACCP and good hygienic practice. Blagojevic (2011) proposed comparing samples taken prior to de-hiding with corresponding samples taken from the final carcass. The rationale was that hides are the greatest source of incoming contamination therefore the ratio between microbiological load from the hide and corresponding dressed carcass would give a more accurate representation of process control. (Blagojevic 2011). Swabs were taken from beef carcasses after sticking but before de-hiding and also at the end of the slaughter line but before chilling. A 2000cm² sampling area comprised of lateral rump-perianal-medial rump-flank-brisket-neck, was sampled on the carcass at both points and the results of the two samples compared. It was concluded that determining a ratio between mean TVC and/or *Enterobacteriaceae* count of the two samples may provide a better assessment of process hygiene as it would take into account the ability to reduce incoming microbial load through process hygiene (Blagojevic et al 2011). It should be noted however, as the values are applied at the end of processing, they are in fact similar to an end product criterion and do not differentiate between more and less hygienic process control during any given processing shift (Blagojevic et al 2011).

Another study conducted at three abattoirs in the United States of America used two sample sites at the top site (inside and outside of the top round) and the bottom site (navel-plate-brisket-foreshank area). Carcasses were sampled at pre-evisceration and before and after pre-evisceration interventions (Wang et al 2013). These interventions consisted of spray delivered in a commercial pre evisceration cabinet, however the type and duration of spray differed between each establishment (Wang et al 2013). The average spray time for each carcass was 5 – 15 seconds and each of the three plants used a different spray (hot water, 5% lactic acid and 220ppm peroxyacetic acid) (Wang et al 2013). Samples were used for the enumeration of TVCs, *E.coli*, coliforms and *Enterobacteriaceae* (Wang et al 2013). The study concluded that there was no significant difference between the top site, bottom site and combined top/bottom site sample in terms of indicator bacteria on pre-evisceration carcasses and as a result, any of the sites would be suitable for monitoring of indicator organisms (Wang et al 2013). This study used swab sample sites of 4000cm²

(8000cm² for the combined sample) (Wang et al 2013). It is understood that Meat and Livestock Australia in conjunction with Symbio are in the process of finalising a research project that supports this finding from Wang et al (2013) in Australian processing.

Swab Site Size

The current requirement for cattle under ESAM is for a composite sample to be taken by swabbing each of three sites (flank, brisket and rump), each site 100cm², giving an overall size of 300cm². The swab sample size needs to be considered for practicality in a commercial setting due to the resources required to conduct the sampling processes and time involved.

There was a large variation in the size of the individual sample sites used in the studies reviewed. Untermann et al (1997) utilized 40cm² sites while Wang et al (2013) used swab sampling on sites of 4000cm². The most commonly selected size for sampling sites observed was 100cm² (Gill et al 1996; Tergeny and Bolton 2006; Hauge et al 2012; Serraino et al 2012).

Variation in the size of individual sites and the number of sites sampled in each study resulted in variation in total sample size up to a maximum area of 8000cm² (Wang et al 2013).

Sampling Frequency

Under the current ESAM program frequency of sampling is based on throughput and class of stock (AQIS 2003). Frequency of sampling also depends on the organism that is being tested for. The current sampling frequencies are:

- *E.coli*:
 - Steers/Heifers: 1 test per 300 carcasses
 - Cows/Bulls: 1 test per 300 carcasses
- *Salmonella*:
 - Steers/Heifers: 1 test per 1500 carcasses
 - Cows/Bulls: 1 test per 1500 carcasses (AQIS 2003).

The ESAM program also requires that *E.coli* and *Salmonella* samples are taken from different carcasses (AQIS 2003) while TVC and coliform counts can be performed on the same sample collected for *E.coli* (DAFF 2013).

Carton meat samples are collected at the same frequency as *E.coli* ESAM carcass samples, at the equivalent of one test per 300 carcasses (DAFF 2013).

Only one study was identified that drew conclusions regarding frequency of sampling. As discussed previously, Untermann et al (1997) conducted a study utilizing 9600 swab samples from multiple sites on 900 beef carcasses, sourced from 10 Swiss abattoirs. Untermann et al (1997) concluded that the observed variation in colony counts identified in their study warranted a sampling plan of five to six swab samples from 10-15 carcasses at least once a month. This sampling plan did not link the number of samples to throughput, but concluded that useful information about hygiene trends could be derived from such a plan (Untermann et al 1997).


Another consideration in the frequency of testing should be the aim to deliver sufficient and viable microbiological data but at a rate that is efficient, both in time and resources for abattoirs. A number of examples of sampling plans were identified during this review.

In a draft guidance document for comment, United States Department of Agriculture, Food Safety and Inspection Service (2008) noted that high frequency and extensive testing has the potential to be cost prohibitive for a number of small and very small abattoirs. In response, the draft guidelines outlined minimum sampling frequencies for small and very small establishments for the testing of finished ground product for *E.coli* O157:H7. FSIS also recommended increasing sampling rates in the warmer months based on studies that indicated shedding of *E.coli* O157:H7 by cattle is greater during this time. Although *E.coli* O157:H7 is outside of the scope of the project this document does indicate an approach that allows for variation in testing frequency based on risk.

The United Kingdom's Food Standards Agency (2015) have produced the Meat Industry Guide that outlines the legal obligations of food operators in the meat sector. Chapter 13, Annex 1 of this document outlines the sampling frequency for red meat carcasses. The sampling plan is structured with initial sample frequency based on throughput, however satisfactory results over a given time period results in a reduction in sampling frequency. For example, an establishment with an annual throughput of greater than 20 000 would initially test for *Enterobacteriaceae* and Total Viable Count by sampling 5 carcasses once a week for 6 week (Food Standards Agency 2015). If the results obtained are satisfactory, the sampling frequency for these organisms decreases to 5 carcasses every 2 weeks (Food Standards Agency 2015). Such an approach has the potential to further incentivize hygienic production as well as deliver savings to industry in time and resources.

While neither of these sampling plans, as written, may be directly transferable to the Australian processing environment, the rationale behind them does warrant investigation.

10 Appendix 2: Example of a Sampling Sheet

 Symbio Laboratories Pty Ltd Delivery Address: Unit 15, 640 – 680 Geelong Rd, Brooklyn, Vic, 3012 Tel: 07-33405700 Fax: 07-32190333																				
Sample Submission Sheet																				
Food & Veterinary Services - MLA/AMPC Alternative Micro Sites Project 2017																				
Date:	/7/2017																			
From Establishment:	B c/o Food and Veterinary Services																			
Address:	15/1 Celestial Court CARINA QLD 4152																			
Contact Name:	Elizabeth Wilcock																			
Contact Email:	elizabeth@foodandvetservices.com																			
Symbio Contacts:	Customer Service Support Person 07-3340 5734 (or Peter Horchner or Elizabeth Chester 0408 320619)					Email : admin@symbiolabs.com.au and c.c. phorchner@symbiolabs.com.au														
Sample Description (Use Body No., L or F & E or C as written on Sponge)	Time sampled	Test(s) Required																		
		TVC	Coliform	E.coli	Cattle cleanines score (1-4)	Sampling Site (e.g. Brisket, Neck etc)	Sampling Site - Post Evisceration		Sampling Point - Chiller											
		M2.5	M8.8.1	M8.8.2			Leading side	Following side	Leading side	Following side										
		X	X	X		Neck	X													
		X	X	X		Shoulder	X													
		X	X	X		Fore Arm	X													
		X	X	X		Brisket	X													
		X	X	X		Neck	X													
		X	X	X		Shoulder	X													
		X	X	X		Fore Arm	X													
		X	X	X		Brisket	X													
		X	X	X		Neck	X													
		X	X	X		Shoulder	X													
		X	X	X		Fore Arm	X													
		X	X	X		Brisket	X													
Special Instructions: LOG IN TEAM - please log in under "SYMBIO MLA", using Sample type "FVS Sponges"																				
Name of Authorised Person (Print):					Sign:															

11 Appendix 3: Photos of Sampling

Photo 11-1: Sampling in Chillers



Photo 11-2: Sampling of the Loin



Photo 11-3: Whirl-pack™ sampling bags being closed after sampling



12 Appendix 4: Results for Establishment A – Slaughter

Table 1: Slaughter floor sample results post evisceration and pre-trim

Body Numbers Test Type	Sampling Site				Carcase Side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
12 13 14						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	<0.83	<0.83	~10	<0.83	Leading	2
25 26 27						
Coliform	<0.083	<0.083	~0.17	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	~0.17	<0.083	Leading	2
SPC	<0.83	57	~17	~1.7	Leading	2
36 37 38						
Coliform	<0.083	<0.083	~0.25	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	~0.17	<0.083	Leading	2
SPC	<0.83	<0.83	~14	<0.83	Leading	2
46 47 48						
Coliform	~0.25	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~17	~4.2	~11	<0.83	Leading	2
57 58 59						
Coliform	<0.083	~0.5	~0.33	<0.083	Leading	2
<i>E.coli</i>	<0.083	~0.42	~0.33	<0.083	Leading	2
SPC	<0.83	48	~18	<0.83	Leading	2
74 75 76						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~2.5	<0.83	~13	<0.83	Leading	2
87 88 89						
Coliform	<0.083	~0.17	~0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	~0.17	~0.083	<0.083	Leading	2
SPC	<0.83	79	~20	~1.7	Leading	2
98 99 100						
Coliform	<0.083	<0.083	~0.42	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	<0.83	<0.83	26	<0.83	Leading	2
109 110 112						
Coliform	<0.083	<0.083	~0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	<0.83	<0.83	39	<0.83	Leading	2
117 118 119						

Body Numbers Test Type	Sampling Site				Carcase Side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
Coliform	<0.083	<0.083	~0.17	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	<0.83	~0.83	~24	<0.83	Leading	2
126 127 128						
Coliform	<0.083	<0.083	~0.33	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	~0.33	<0.083	Leading	2
SPC	~5	<0.83	~13	<0.83	Leading	2
136 137 138						
Coliform	~0.083	<0.083	~0.17	<0.083	Leading	2
<i>E.coli</i>	~0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~1.7	92	~14	<0.83	Leading	2
145 146 147						
Coliform	<0.083	~0.083	~0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	~0.083	<0.083	<0.083	Leading	2
SPC	<0.83	~11	~15	~0.83	Leading	2
152 153 154						
Coliform	<0.083	<0.083	<0.083	~0.17	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	~0.17	Leading	2
SPC	<0.83	~9.2	~8.3	~4.2	Leading	2
161 162 163						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	<0.83	~0.83	<0.83	~0.83	Leading	2
172 173 174						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~3.3	~5.8	~7.5	~0.83	Leading	2
181 182 183						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	<0.83	<0.83	~4.2	<0.83	Leading	2
188 189 190						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~2.5	~1.7	67	<0.83	Leading	2
500 501 502						
Coliform	<0.083	<0.083	~0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	~0.083	<0.083	Following	2
SPC	~14	150	160	~2300	Following	2
508 509 510						
Coliform	2	~0.083	~0.083	<0.083	Following	2

Body Numbers Test Type	Sampling Site				Carcase Side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
<i>E.coli</i>	<0.083	<0.083	~0.083	<0.083	Following	2
SPC	78	76	35	~7.5	Following	2
519 520 521						
Coliform	<0.083	<0.083	~0.17	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	~0.17	<0.083	Following	2
SPC	~0.83	~1.7	180	<0.83	Following	2
545 546 547						
Coliform	~0.083	<0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	~0.83	~5	~5.8	~0.83	Following	2
555 556 557						
Coliform	<0.083	<0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	~1.7	~11	28	<0.83	Following	2
567 568 569						
Coliform	<0.083	<0.083	<0.083	~0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	~0.083	Following	2
SPC	27	<0.83	~20	160	Following	2
575 576 577						
Coliform	<0.083	<0.083	~0.33	~0.42	Following	2
<i>E.coli</i>	<0.083	<0.083	~0.25	~0.33	Following	2
SPC	<0.83	<0.83	120	190	Following	2
584 585 586						
Coliform	<0.25	<0.083	<0.25	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.25	<0.083	Following	2
SPC	<18	<4.2	<27	~0.83	Following	2
593 594 595						
Coliform	~0.083	~0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	~0.083	<0.083	<0.083	<0.083	Following	2
SPC	~2.5	~920	~18	80	Following	2
601 602 603						
Coliform	<0.083	<0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	~0.83	~12	~7.5	<0.83	Following	2
609 610 611						
Coliform	<0.083	~0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	~5	~51	110	~0.83	Following	2
617 618 619						
Coliform	<0.083	<0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	2

Body Numbers Test Type	Sampling Site				Carcase Side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
SPC	~3.3	~8.3	~5.8	~0.83	Following	2
627 628 629						
Coliform	<0.083	<0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	~1.7	~18	110	~7.5	Following	2
641 642 643						
Coliform	<0.083	<0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	~0.83	~14	~11	<0.83	Following	2
649 650 651						
Coliform	~0.17	<0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	~0.17	<0.083	<0.083	<0.083	Following	2
SPC	~14	~1.7	70	<0.83	Following	2
659 660 661						
Coliform	<0.083	~0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	~0.083	<0.083	<0.083	Following	2
SPC	~0.83	~5	<0.83	~4.2	Following	2
670 671 672						
Coliform	~0.083	<1.7	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	~11	~920	~9.2	~15000000	Following	2
679 681 682						
Coliform	<0.083	<0.083	<0.083	~0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	~0.083	Following	2
SPC	<0.83	39	<0.83	~0.83	Following	2

Table 2: Chiller sample results

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
15 17 36									
Coliform	<0.083	<0.083	<0.083	<0.083	~0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	~0.083	<0.083	<0.083	Following	2
SPC	~5.8	~3.3	~8.3	~0.83	~10	<0.83	<0.83	Following	2
20 24 26									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	~13	~18	~6.7	~1.7	~6.7	~3.3	~9.2	Following	2
30 42 43									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	~0.83	~3.3	~3.3	~1.7	<0.83	<0.83	~330	Following	2
39 74 92									
Coliform	<0.083	<0.083	<0.083	<0.083	~0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	~0.083	<0.083	<0.083	Following	2
SPC	~2.5	<0.83	~6.7	<0.83	~4.2	<0.83	~5	Following	2
41 69 72									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	~0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	~0.083	Following	2
SPC	~0.83	<0.83	~5.8	~0.83	25000	<0.83	~0.83	Following	2
44 57 61									

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
Coliform	~0.083	<0.083	<0.083	<0.083	~0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	~0.083	<0.083	<0.083	<0.083	~0.083	<0.083	<0.083	Following	2
SPC	43	~0.83	<0.83	<0.83	~1.7	<0.83	~3.3	Following	2
49 21 34									
Coliform	~0.083	<0.083	~0.17	<0.083	<0.083	<0.083	~0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	~0.083	Following	2
SPC	~7.5	<0.83	~1900	<0.83	~0.83	~250	~17	Following	2
51 91 95									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	~3.3	<0.83	~4.2	<0.83	<0.83	<0.83	~2.5	Following	2
63 62 88									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	~1.7	~0.83	26	<0.83	~0.83	~0.83	<0.83	Following	2
67 73 89									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	~9.2	<0.83	~7.5	<0.83	~1.7	<0.83	~8.3	Following	2
166 163 162									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	210	25	~13	~24	62	29	~12	Following	2
167 168 169									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	110	~4.2	~1.7	~1.7	~20	<0.83	~9.2	Following	2
248 249 250									
Coliform	<0.083	<0.083	<0.083	<0.083	~1	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	~13	~4.2	~4.2	<0.83	220	~6.7	~0.83	Following	2
283 309 310									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	~0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	~0.083	<0.083	Following	2
SPC	~22	~12	50	~3.3	~13	~2.5	~5	Following	2
318 342 367									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	60	~13	~18	33	30	~7000000	~17	Following	2
322 353 356									
Coliform	<0.083	<0.083	<0.083	<0.083	~0.083	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	~7.5	~14	~3.3	<0.83	63	<0.83	~23	Following	2
388 389 390									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	~0.25	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	40	~3.3	~10	~4.2	44	<0.83	67	Following	2
570 605 640									
Coliform	~0.083	<0.083	<0.083	<0.083	<0.083	<0.083	~0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	~0.083	Leading	2

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
SPC	~22	<0.83	~7.5	<0.83	~20	~2.5	~3.3	Leading	2
572 581 592									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~580	~23	<0.83	<0.83	~17	~0.83	~0.83	Leading	2
603 609 610									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~7.5	<0.83	~1.7	<0.83	~1.7	~1.7	~1.7	Leading	2
612 615 616									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~9.2	~1.7	~2.5	<0.83	~2.5	~1.7	~4.2	Leading	2
628 620 631									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~8.3	<0.83	~5	<0.83	<0.83	~2.5	~6.7	Leading	2
639 647 650									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~11	~0.83	25	<0.83	~1.7	~1.7	~2.5	Leading	2
645 649 651									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~1.7	~0.83	~1.7	<0.83	~0.83	~1.7	~0.83	Leading	2

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
653 655 656									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~16	<0.83	~0.83	<0.83	~1.7	<0.83	~14	Leading	2
659 661 665									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~2.5	~6.7	~5.8	~0.83	~4.2	~2.5	~0.83	Leading	2
662 663 669									
Coliform	~0.083	<0.083	~0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	~0.083	<0.083	~0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~0.83	~2.5	~4.2	~0.83	~1.7	<0.83	~0.83	Leading	2
666 687 675									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	<0.83	~12	~3.3	<0.83	~11	<0.83	~1.7	Leading	2
670 694 698									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~2.5	<0.83	~0.83	<0.83	~5	<0.83	~5	Leading	2
672 674 679									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~5.8	~1.7	~4.2	~2.5	~1.7	~10	~3.3	Leading	2
746 627 646									

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	~0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~2.5	~5.8	~2.5	<0.83	~0.83	<0.83	160	Leading	2
792 793 795									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	92	~16	~330	<0.83	~7.5	~2.5	~3.3	Leading	2
796 797 798									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~1000	~0.83	70	<0.83	~2.5	33	~5	Leading	2

13 Appendix 5: Results for Establishment B

Table 1: Slaughter floor sample results post evisceration and pre-trim

Body Numbers Test Type	Sampling Site				Carcase side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
48, 49, 50						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~0.83	~1.7	~1.7	<0.83	Leading	2
56 57 58						
Coliform	<0.083	<0.083	~0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	<0.83	~2.5	~0.83	~1.7	Leading	2
64 65 66						
Coliform	<0.083	<0.083	~0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~0.83	~1.7	~7.5	~1.7	Leading	2
72 73 75						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	2
SPC	~7.5	~12	~8.3	<0.83	Leading	2
84 85 87						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	-
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	-
SPC	~2.5	<0.83	~8.3	~0.83	Leading	-
92 93 94						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	3
SPC	<0.83	~0.83	~0.83	<0.83	Leading	3
102 107 108						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	3
SPC	~3.3	~1.7	~9.2	<0.83	Leading	3
105 117 118						
Coliform	<0.083	<0.083	~0.17	<0.083	Leading	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	3
SPC	~0.83	~6.7	~7.5	~1.7	Leading	3
113 114 115						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	3
SPC	~5.8	<0.83	~5	~4.2	Leading	3
124 125 128						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	3

Body Numbers Test Type	Sampling Site				Carcase side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	3
SPC	~10	~2.5	~5	<0.83	Leading	3
135 136 137						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	3
SPC	~5	<0.83	~4.2	<0.83	Leading	3
143 144 145						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	3
SPC	~0.83	<0.83	~1.7	<0.83	Leading	3
153 154 155						
Coliform	0.083	0.083	0.083	0.083	Leading	3
<i>E.coli</i>	0.083	0.083	0.083	0.083	Leading	3
SPC	1.7	0.83	5	0.83	Leading	3
160 161 162						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	3
SPC	~1.7	~0.83	~2.5	<0.83	Leading	3
169 170 171						
Coliform	~0.083	<0.083	<0.083	<0.083	Leading	3
<i>E.coli</i>	~0.083	<0.083	<0.083	<0.083	Leading	3
SPC	~2.5	~0.83	~4.2	~0.83	Leading	3
177 179 180						
Coliform	<0.083	<0.083	~0.33	<0.083	Leading	3
<i>E.coli</i>	<0.083	<0.083	~0.083	<0.083	Leading	3
SPC	~22	~7.5	~2.5	<0.83	Leading	3
187 188 189						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	3
SPC	~3.3	~0.83	~2.5	~0.83	Leading	3
196 197 199						
Coliform	<0.083	<0.083	<0.083	<0.083	Leading	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	3
SPC	~6.7	~0.83	~5	<0.83	Leading	3
210 212 213						
Coliform	<0.083	<0.083	<0.083	<0.083	Following	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	3
SPC	<0.83	77	<0.83	220	Following	3
218 219 220						
Coliform	<0.083	<0.083	<0.083	<0.083	Following	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	3
SPC	<0.83	<0.83	~2.5	65	Following	3

Body Numbers Test Type	Sampling Site				Carcase side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
225 226 227						
Coliform	<0.083	<0.083	<0.083	<0.083	Following	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	3
SPC	<0.83	~0.83	~2.5	~0.83	Following	3
246 247 248						
Coliform	<0.083	<0.083	<0.083	<0.083	Following	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	3
SPC	<0.83	~12	~7.5	~3.3	Following	3
254 255 256						
Coliform	<0.083	<0.083	<0.083	<0.083	Following	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	3
SPC	~0.83	~0.83	28	~0.83	Following	3
261 262 263						
Coliform	<0.083	~0.083	<0.083	<0.083	Following	3
<i>E.coli</i>	<0.083	~0.083	<0.083	<0.083	Following	3
SPC	~0.83	230	~2.5	<0.83	Following	3
268 269 270						
Coliform	<0.083	<0.083	<0.083	<0.083	Following	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	3
SPC	<0.83	130	~6.7	~0.83	Following	3
275 276 277						
Coliform	~0.17	~0.083	<0.083	<0.083	Following	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	3
SPC	~0.83	~8.3	~18	<0.83	Following	3
282 283 284						
Coliform	<0.083	<0.083	<0.083	<0.083	Following	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	3
SPC	37	~0.83	<0.83	~83	Following	3
288 289 290						
Coliform	<0.083	<0.083	<0.083	<0.083	Following	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	3
SPC	<0.83	40	~3.3	<0.83	Following	3
292 293 294						
Coliform	<0.083	<0.083	<0.083	<0.083	Following	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	3
SPC	~0.83	<0.83	~0.83	~12	Following	3
299 300 301						
Coliform	~0.083	<0.083	<0.083	<0.083	Following	3
<i>E.coli</i>	~0.083	<0.083	<0.083	<0.083	Following	3
SPC	~2.5	~12	~17	200	Following	3
306 307 308						
Coliform	<0.083	<0.083	<0.083	<0.083	Following	3

Body Numbers Test Type	Sampling Site				Carcase side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	3
SPC	<0.83	<0.83	~5.8	<0.83	Following	3
312 313 314						
Coliform	<0.083	<0.083	~0.25	<0.083	Following	3
<i>E.coli</i>	<0.083	<0.083	~0.17	<0.083	Following	3
SPC	<0.83	<0.83	~1500	~1.7	Following	3
320 321 322						
Coliform	<0.083	<0.083	<0.083	<0.083	Following	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	3
SPC	<0.83	~0.83	~4.2	~0.83	Following	3
333 335 336						
Coliform	~0.083	<0.083	<0.083	<0.083	Following	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	3
SPC	<0.83	<0.83	<0.83	~3.3	Following	3
341 342 343						
Coliform	<0.083	~0.17	~0.25	<0.083	Following	3
<i>E.coli</i>	<0.083	~0.083	~0.083	<0.083	Following	3
SPC	~0.83	~500	~750	<0.83	Following	3
346 347 348						
Coliform	<0.083	<0.083	~0.17	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	2
SPC	~5	~7.5	~8.3	~0.83	Following	2

Table 2: Chiller sample results

Body Numbers Test Type	Sampling Site						Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump		
4 5 137								
Coliform		<0.083		<0.083		<0.083		Following -
<i>E.coli</i>		<0.083		<0.083		<0.083		Following -
SPC		37		~6.7		<0.83		Following -
18 19 84								
Coliform	~0.083		<0.083		<0.083		<0.083	Following 3
<i>E.coli</i>	~0.083		<0.083		<0.083		<0.083	Following 3
SPC	~34		57		31		63	Following 3
23 24 25								
Coliform		<0.083		<0.083		<0.083		Following 2
<i>E.coli</i>		<0.083		<0.083		<0.083		Following 2
SPC		~10		~4.2		~1.7		Following 2
42 46 49								
Coliform	<0.083		<0.083		<0.083		<0.083	Following 3
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Following 3
SPC	~9.2		~19		~750		<0.83	Following 3
54 59 104								
Coliform	<0.083		<0.083		~0.083		<0.083	Following -
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Following -
SPC	~5.8		36		~9.2		~23	Following -
71 74 81								
Coliform		<0.083		~0.083		~0.083		Following 3
<i>E.coli</i>		<0.083		<0.083		<0.083		Following 3
SPC		<0.83		<0.83		~180		Following 3

Body Numbers Test Type	Sampling Site						Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump		
79 80 82								
Coliform		<0.083		<0.083		<0.083		Following 3
<i>E.coli</i>		<0.083		<0.083		<0.083		Following 3
SPC		~7.5		<0.83		~8.3		Following 3
86 87 88								
Coliform	<0.083		<0.083		<0.083		<0.083	Following 3
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Following 3
SPC	~5		~12		35		~20	Following 3
96 97 99								
Coliform		<0.083		<0.083		<0.083		Following 3
<i>E.coli</i>		<0.083		<0.083		<0.083		Following 3
SPC		37		~3.3		~0.83		Following 3
112 113 114								
Coliform		<0.083		<0.083		<0.083		Following 3
<i>E.coli</i>		<0.083		<0.083		<0.083		Following 3
SPC		~4.2		~8.3		~5.8		Following 3
127 128 129								
Coliform	<0.083		<0.083		<0.083		~0.083	Following 3
<i>E.coli</i>	<0.083		<0.083		<0.083		~0.083	Following 3
SPC	~11		~19		38		~17	Following 3
141 142 143								
Coliform	<0.083		<0.083		<0.083		<0.083	Following 3
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Following 3
SPC	~3.3		~7.5		~5		~17	Following 3
426 438 447								
Coliform	~0.083		~0.083		<0.083		<0.083	Following 2

Body Numbers Test Type	Sampling Site						Carcase Side	Cattle Cleanliness Score	
	Brisket	Flank	Fore Arm	Loin	Neck	Rump			Shoulder
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Following	2
SPC	~15		~14		38		100	Following	2
433 432 434									
Coliform	~0.083		<0.083		<0.083		<0.083	Following	2
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Following	2
SPC	40		39		~5		~6.7	Following	2
444 503 505									
Coliform	<0.083		<0.083		<0.083		<0.083	Following	2
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Following	2
SPC	~23		40		~3.3		80	Following	2
456 457 462									
Coliform		<0.083		<0.083		<0.083		Following	2
<i>E.coli</i>		<0.083		<0.083		<0.083		Following	2
SPC		~8.3		~2.5		~22		Following	2
461 473 475									
Coliform		<0.083		0.083		~0.58		Following	2
<i>E.coli</i>		<0.083		0.083		~0.58		Following	2
SPC		~11		~23		40		Following	2
471 472 425									
Coliform		<0.083		<0.083		<0.083		Following	2
<i>E.coli</i>		<0.083		<0.083		<0.083		Following	2
SPC		~6.7		<0.83		~2.5		Following	2
484 481 486									
Coliform		<0.083		~23		<0.083		Following	2
<i>E.coli</i>		<0.083		~20		<0.083		Following	2
SPC		38		65		~5.8		Following	2

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
485 495 498									
Coliform		<0.083		<0.083		~0.083		Following	2
<i>E.coli</i>		<0.083		<0.083		~0.083		Following	2
SPC		~8.3		~15		~17		Following	2
493 494 445									
Coliform		<0.083		<0.083		<0.083		Following	2
<i>E.coli</i>		<0.083		<0.083		<0.083		Following	2
SPC		~9.2		~4.2		~5.8		Following	2
496 504 581									
Coliform	<0.083		<0.083		<0.083		<0.083	Following	2
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Following	2
SPC	~9.2		32		~9.2		~12	Following	2
516 517 518									
Coliform	<0.083	<0.083	<0.083	<0.083	~0.17	<0.083	<0.083	Following	2
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	~0.17	<0.083	<0.083	Following	2
SPC	~5	~71	75	~0.83	~11	~17	~5	Following	2
519 520 502									
Coliform		<0.083		<0.083		<0.083		Following	2
<i>E.coli</i>		<0.083		<0.083		<0.083		Following	2
SPC		170		~0.83		~5.8		Following	2
519 520 523									
Coliform	<0.083		<0.083		~0.083		<0.083	Following	2
<i>E.coli</i>	<0.083		<0.083		~0.083		<0.083	Following	2
SPC	42		92		82		37	Following	2
535 619 620									
Coliform	<0.083		~0.083		<0.083		<0.083	Following	2

Body Numbers Test Type	Sampling Site						Carcase Side	Cattle Cleanliness Score	
	Brisket	Flank	Fore Arm	Loin	Neck	Rump			Shoulder
<i>E.coli</i>	<0.083		~0.083		<0.083		<0.083	Following	2
SPC	~2.5		32		~4.2		~12	Following	2
581 587 530									
Coliform		<0.083		<0.083		<0.083		Following	2
<i>E.coli</i>		<0.083		<0.083		<0.083		Following	2
SPC		~0.83		~5		35		Following	2
587 530 591									
Coliform	<0.083		<0.083		<0.083		~0.33	Following	2
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Following	2
SPC	~8.3		~14		~4.2		~2000	Following	2
592 535 612									
Coliform		<0.083		<0.083		<0.083		Following	2
<i>E.coli</i>		<0.083		<0.083		<0.083		Following	2
SPC		28		~13		~5.8		Following	2
604 605 606									
Coliform	<0.083		<0.083		<0.083		<0.083	Following	2
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Following	2
SPC	~5.8		~7.5		~10		~14	Following	2
621 622 623									
Coliform	<0.083		<0.083		<0.083		~0.17	Following	2
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Following	2
SPC	~4.2		~16		~5		~11	Following	2
622 623 624									
Coliform		<0.083		~0.083		<0.083		Following	2
<i>E.coli</i>		<0.083		<0.083		<0.083		Following	2
SPC		~19		~8.3		~15		Following	2

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
201 202 203									
Coliform	<0.083		<0.083		<0.083		~0.5	Leading	3
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Leading	3
SPC	~0.83		35		~7.5		53	Leading	3
206 207 208									
Coliform		~1.1		<0.083		<0.083		Leading	3
<i>E.coli</i>		~1		<0.083		<0.083		Leading	3
SPC		2900		~24		~7.5		Leading	3
214 215 216									
Coliform	<0.083		<0.083		<0.083		<0.083	Leading	3
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Leading	3
SPC	~12		43		~12		31	Leading	3
222 217 271									
Coliform	<0.083		<0.083		<0.083		<0.083	Leading	3
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Leading	3
SPC	~3.3		65		34		110	Leading	3
223 224 225									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	3
SPC	~8.3	~5.8	100	36	~7.5	<0.83	~8.3	Leading	3
226 227 228									
Coliform	<0.083		<0.083		<0.083		<0.083	Leading	3
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Leading	3
SPC	~19		~7.5		<0.83		~3.3	Leading	3
230 236 235									
Coliform	<0.083		<0.083		<0.083		<0.083	Leading	3

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Leading	3
SPC	~23		~420		~5.8		~500	Leading	3
231 233 235									
Coliform		<0.083		<0.083		<0.083		Leading	3
<i>E.coli</i>		<0.083		<0.083		<0.083		Leading	3
SPC		100		~23		~18		Leading	3
248 249 250									
Coliform		~0.17		<0.083		<0.083		Leading	3
<i>E.coli</i>		~0.17		<0.083		<0.083		Leading	3
SPC		~33		~9.2		~23		Leading	3
256 258 260									
Coliform	<0.083		<0.083		<0.083		<0.083	Leading	3
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Leading	3
SPC	<0.83		110		~9.2		~7.5	Leading	3
266 267 268									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	3
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	3
SPC	~6.7	40	58	~16	~2.5	~15	~9.2	Leading	3
269 270 271									
Coliform		<0.083		<0.083		<0.083		Leading	3
<i>E.coli</i>		<0.083		<0.083		<0.083		Leading	3
SPC		~9.2		~9.2		~14		Leading	3
275 276 277									
Coliform	<0.083		<0.083		<0.083		<0.083	Leading	3
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Leading	3
SPC	33		110		~16		~5.8	Leading	3

Body Numbers Test Type	Sampling Site						Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump		
277 278 279								
Coliform		~0.17		<0.083		<0.083		Leading 3
<i>E.coli</i>		<0.083		<0.083		<0.083		Leading 3
SPC		62		~14		<0.83		Leading 3
278 279 280								
Coliform	<0.083		<0.083		<0.083		<0.083	Leading 3
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Leading 3
SPC	<0.83		63		~7.5		~3.3	Leading 3
281 282 283								
Coliform	<0.083		<0.083		<0.083		<0.083	Leading 3
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Leading 3
SPC	~1.7		92		25		53	Leading 3
284 287 334								
Coliform		2.2		<0.083		<0.083		Leading 3
<i>E.coli</i>		1.9		<0.083		<0.083		Leading 3
SPC		110		~0.83		~3.3		Leading 3
287 284 270								
Coliform	<0.083		<0.083		<0.083		~0.25	Leading 3
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Leading 3
SPC	~11		~830		~4.2		31	Leading 3
305 307 308								
Coliform		~0.33		<0.083		<0.083		Leading 3
<i>E.coli</i>		~0.25		<0.083		<0.083		Leading 3
SPC		4800		~8.3		~7.5		Leading 3
322 323 325								
Coliform		<0.083		<0.083		<0.083		Leading 3

Body Numbers Test Type	Sampling Site						Carcase Side	Cattle Cleanliness Score	
	Brisket	Flank	Fore Arm	Loin	Neck	Rump			Shoulder
<i>E.coli</i>		<0.083		<0.083		<0.083		Leading	3
SPC		27		33		100		Leading	3
331 332 333									
Coliform	<0.083		<0.083		<0.083		<0.083	Leading	3
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Leading	3
SPC	~6.7		92		28		~10	Leading	3
340 341 342									
Coliform		~0.083		<0.083		<0.083		Leading	3
<i>E.coli</i>		<0.083		<0.083		<0.083		Leading	3
SPC		32		~4.2		200		Leading	3
529 532 525									
Coliform		<0.083		<0.083		<0.083		Leading	2
<i>E.coli</i>		<0.083		<0.083		<0.083		Leading	2
SPC		4800		~9.2		32		Leading	2
533 534 536									
Coliform		<0.083		<0.083		<0.083		Leading	2
<i>E.coli</i>		<0.083		<0.083		<0.083		Leading	2
SPC		~670		<0.83		~3.3		Leading	2
544 545 546									
Coliform		<0.083		<0.083		<0.083		Leading	2
<i>E.coli</i>		<0.083		<0.083		<0.083		Leading	2
SPC		40		~4.2		~2.5		Leading	2
554 555 556									
Coliform		~0.17		<0.083		<0.083		Leading	2
<i>E.coli</i>		<0.083		<0.083		<0.083		Leading	2
SPC		76		~5.8		~16		Leading	2

Body Numbers Test Type	Sampling Site						Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump		
567 568 569								
Coliform		<0.083		<0.083		~0.083		Leading 2
<i>E.coli</i>		<0.083		<0.083		~0.083		Leading 2
SPC		~2000		25		78		Leading 2
575 576 617								
Coliform	<0.083		<0.083		<0.083		<0.083	Leading 2
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Leading 2
SPC	~1.7		~14		~7.5		~10	Leading 2
583 584 585								
Coliform		<0.083		<0.083		<0.083		Leading 2
<i>E.coli</i>		<0.083		<0.083		<0.083		Leading 2
SPC		~1100		~0.83		~9.2		Leading 2
611 527 548								
Coliform	<0.083		<0.083		<0.083		<0.083	Leading 2
<i>E.coli</i>	<0.083		<0.083		<0.083		<0.083	Leading 2
SPC	~23		~12		~3.3		~12	Leading 2

14 Appendix 6: Results for Establishment C

Table 1: Slaughter floor sample results post evisceration and pre-trim

Body Numbers Test Type	Sampling Site				Carcase Side
	Neck	Shoulder	Brisket	Fore Arm	
Sample 1					
Coliform	<0.083	<0.083	<0.083	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following
SPC	~5	~1.7	~1300	~1.7	Following
Sample 2					
Coliform	<0.083	<0.083	<0.083	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following
SPC	~1100	320000	33	~1.7	Following
Sample 3					
Coliform	<0.083	<0.083	~0.17	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	~0.17	<0.083	Following
SPC	~670	~0.83	26	~0.83	Following
Sample 4					
Coliform	<0.083	<0.083	<0.083	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following
SPC	~3.3	360000	~12	~0.83	Following
Sample 5					
Coliform	<0.083	<0.083	~1.1	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	~0.25	<0.083	Following
SPC	~1000	<0.83	~18	~11	Following
Sample 6					
Coliform	<0.083	<0.083	<0.083	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following
SPC	110	<0.83	~12	~1.7	Following
Sample 7					
Coliform	<0.083	<0.083	<0.083	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following
SPC	~18	<0.83	~14	~23	Following
Sample 8					
Coliform	<0.083	<0.083	<0.083	~0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following
SPC	~4.2	~1.7	~4.2	~3.3	Following
Sample 9					
Coliform	<0.083	<0.083	<0.083	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following
SPC	43	~0.83	40	~14	Following
Sample 10					
Coliform	<0.083	<0.083	<0.083	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following

Body Numbers Test Type	Sampling Site				Carcase Side
	Neck	Shoulder	Brisket	Fore Arm	
SPC	~12	~1.7	~0.83	33	Following
Sample 11					
Coliform	<0.083	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading
SPC	~4.2	<0.83	<0.83	<0.83	Leading
Sample 12					
Coliform	<0.083	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading
SPC	<0.83	<0.83	~1.7	~3.3	Leading
Sample 13					
Coliform	~0.083	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	~0.083	<0.083	<0.083	<0.083	Leading
SPC	180	~5.8	~3.3	<0.83	Leading
Sample 14					
Coliform	~0.17	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	~0.083	<0.083	<0.083	<0.083	Leading
SPC	~19	<0.83	~0.83	25	Leading
Sample 15					
Coliform	~0.083	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading
SPC	37	<0.83	~1.7	~15	Leading
Sample 16					
Coliform	~0.083	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	~0.083	<0.083	<0.083	<0.083	Leading
SPC	41	<0.83	~5.8	<0.83	Leading
Sample 17					
Coliform	<0.083	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading
SPC	~3.3	<0.83	~4.2	~2.5	Leading
Sample 18					
Coliform	<0.083	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading
SPC	36	<0.83	~1.7	<0.83	Leading
Sample 19					
Coliform	~0.083	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading
SPC	~13	~5.8	~15	~1.7	Leading
Sample 20					
Coliform	2	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	2	<0.083	<0.083	<0.083	Leading
SPC	~1800	~21	~1.7	~4.2	Leading

Table 2: Chiller sample results

Body Numbers Test Type	Sampling Site							Carcase Side
	Neck	Shoulder	Brisket	Fore Arm	Flank	Loin	Rump	
1, 4, 3								
Coliform	<0.083	<0.083	~0.92	<0.083	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading
SPC	~420	~0.83	~17	~0.83	~1.7	<0.83	~1100	Leading
2, 14, 24								
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading
SPC	~23	~330	<0.83	~580	82	~3.3	180	Leading
6, 7, 8								
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	~0.083	Leading
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading
SPC	~2.5	~4.2	~8.3	~0.83	~16	26	~92	Leading
11, 36, 38								
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading
SPC	62	~8.3	<0.83	~19	~580	36	~830	Leading
64, 68, 72								
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading
SPC	~0.83	~1.7	~13	34	~4.2	<0.83	28	Leading
132, 142, 144								
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading
SPC	~20	<0.83	~4.2	~13	~0.83	~2.5	110	Leading
136, 147, 148								

Body Numbers Test Type	Sampling Site							Carcase Side
	Neck	Shoulder	Brisket	Fore Arm	Flank	Loin	Rump	
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading
SPC	~2.5	~5	~2.5	~11	~16	~0.83	190	Leading
143, 145, 146								
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading
SPC	~9.2	~0.83	~2.5	~1.7	~13	~4.2	76	Leading
2, 14, 24								
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
SPC	~670	~5	~4.2	53	~750	~9.2	~1.7	Following
13, 17, 20								
Coliform	<0.083	~0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
SPC	~17	~12	<0.83	130	~1600	230	130	Following
15, 16, 18								
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
SPC	~2.5	~4.2	~0.83	~6.7	67	~4.2	190	Following
19, 27, 47								
Coliform	<0.083	<0.083	<0.083	<0.083	~0.25	<0.083	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
SPC	27	~0.83	75	~5	77	~13	76	Following
21, 45, 49								
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
SPC	~2.5	~1.7	~1.7	~4.2	230	~5.8	~170	Following

Body Numbers Test Type	Sampling Site							Carcase Side
	Neck	Shoulder	Brisket	Fore Arm	Flank	Loin	Rump	
25, 30, 33								
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
SPC	<0.83	~8.3	~7.5	~1.7	~670	~3.3	34	Following
29, 46, 48								
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
SPC	~18	~7.5	~1.7	~11	~1300	~7.5	28	Following
64, 68, 72								
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
SPC	~1.7	<0.83	<0.83	~2.5	~5.8	~1.7	110	Following
228, 236, 238								
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following
SPC	<0.83	31	<0.83	~1.7	<0.83	~5.8	~21	Following

15 Appendix 7: Results for Establishment D

Table 1: Slaughter floor sample results post evisceration and pre-trim

Body Numbers Test Type	Sampling Site				Carcase Side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
108 109 110						
Coliform	~0.42	~0.33	~0.75	~0.25	Following	1
<i>E.coli</i>	~0.25	~0.33	<0.083	<0.083	Following	1
SPC	100	~2200	54	<0.83	Following	1
122 123 124						
Coliform	~0.17	~0.25	~0.25	~1.2	Following	1
<i>E.coli</i>	~0.17	~0.25	<0.083	<0.083	Following	1
SPC	50	~10	5300	~13	Following	1
133 134 135						
Coliform	<0.083	<0.083	3.9	~0.5	Following	1
<i>E.coli</i>	<0.083	<0.083	~1.2	~0.5	Following	1
SPC	~0.83	~0.83	6500	~920	Following	1
146 147 148						
Coliform	~0.58	1.8	~0.17	<0.083	Following	1
<i>E.coli</i>	~0.58	1.8	<0.083	<0.083	Following	1
SPC	~920	39	220	~10	Following	1
158 159 160						
Coliform	~0.083	~0.25	<0.083	~0.58	Following	1
<i>E.coli</i>	~0.083	~0.083	<0.083	~0.5	Following	1
SPC	170	~18	150	38	Following	1
171 172 174						
Coliform	~0.25	~0.33	~0.33	3.2	Following	1
<i>E.coli</i>	~0.17	~0.33	<0.083	3.2	Following	1
SPC	~23	~420	220	~830	Following	1
183 184 185						
Coliform	<0.083	3.8	~0.25	<0.083	Following	1
<i>E.coli</i>	<0.083	3.3	<0.083	<0.083	Following	1
SPC	71	~330	220	~320	Following	1
194 195 196						
Coliform	~0.42	<0.083	~0.67	~0.58	Following	1
<i>E.coli</i>	<0.083	<0.083	~0.083	<0.083	Following	1
SPC	180	180	~19	~330	Following	1
205 206 207						
Coliform	<0.083	<0.083	~0.75	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	~0.83	~24	~11	Following	1
214 215 216						
Coliform	<0.083	<0.083	~0.083	~0.17	Following	1

Body Numbers Test Type	Sampling Site				Carcase Side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
<i>E.coli</i>	<0.083	<0.083	~0.083	~0.17	Following	1
SPC	~1.7	~3.3	28	55	Following	1
225 226 227						
Coliform	<0.083	~0.83	<0.083	~0.67	Following	1
<i>E.coli</i>	<0.083	~0.83	<0.083	~0.67	Following	1
SPC	~13	~18	59	~830	Following	1
236 238 239						
Coliform	<0.083	<0.083	1.9	~0.33	Following	1
<i>E.coli</i>	<0.083	<0.083	1.9	~0.25	Following	1
SPC	<0.83	<0.83	~420	~10	Following	1
244 245 246						
Coliform	<0.083	~0.083	~0.33	~0.17	Following	1
<i>E.coli</i>	<0.083	~0.083	<0.083	~0.17	Following	1
SPC	<0.83	~1.7	~5	~7.5	Following	1
254 255 256						
Coliform	2.2	~0.083	~0.58	~0.17	Following	1
<i>E.coli</i>	2.2	~0.083	~0.5	~0.083	Following	1
SPC	~5.8	<0.83	~4.2	<0.83	Following	1
266 267 268						
Coliform	<0.083	~0.083	~17	~0.75	Following	1
<i>E.coli</i>	<0.083	~0.083	~17	~0.75	Following	1
SPC	~7.5	~0.83	~330	~2.5	Following	1
281 282 283						
Coliform	<0.083	~0.33	2.5	~0.25	Following	1
<i>E.coli</i>	<0.083	~0.33	2.5	~0.25	Following	1
SPC	<0.83	~16	150	~20	Following	1
294 295 296						
Coliform	<0.083	~0.58	~0.5	~0.17	Following	1
<i>E.coli</i>	<0.083	~0.58	~0.083	<0.083	Following	1
SPC	~1.7	~2.5	~4.2	~4.2	Following	1
316 317 318						
Coliform	2.7	~0.33	~0.58	3	Following	1
<i>E.coli</i>	2.7	~0.33	~0.25	3	Following	1
SPC	52	~5.8	~420	~500	Following	1
329 331 332						
Coliform	~0.083	<0.083	9.2	3.8	Following	1
<i>E.coli</i>	~0.083	<0.083	9.2	3.6	Following	1
SPC	~3.3	~0.83	~170	220	Following	1
340 341 342						
Coliform	<0.083	2.8	~0.083	5	Following	1
<i>E.coli</i>	<0.083	2.8	<0.083	4.2	Following	1

Body Numbers Test Type	Sampling Site				Carcase Side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
SPC	~5	55	~15	220	Following	1
354 355 357						
Coliform	~0.75	~0.25	~1.2	<0.083	Following	1
<i>E.coli</i>	~0.33	~0.083	~1	<0.083	Following	1
SPC	~5.8	~24	150	54	Following	1
454 455 456						
Coliform	<0.083	~0.083	4.3	~1.1	Following	1
<i>E.coli</i>	<0.083	<0.083	~0.83	~0.33	Following	1
SPC	30	~1.7	~1200	53	Following	1
465 467 468						
Coliform	3	10	1.3	6.3	Following	1
<i>E.coli</i>	<0.083	10	~0.25	<0.083	Following	1
SPC	~170	~27000	92	65	Following	1
508 509 510						
Coliform	~0.083	<0.083	2.7	~0.58	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	0.083	Following	1
SPC	~13	~19	39	25	Following	1
522 523 524						
Coliform	~0.42	~0.67	2.4	~0.42	Following	1
<i>E.coli</i>	<0.083	<0.083	~0.83	~0.17	Following	1
SPC	~6.7	60	~670	130	Following	1
534 535 536						
Coliform	~0.33	<0.083	~0.5	1.6	Following	1
<i>E.coli</i>	<0.083	<0.083	~0.083	<0.083	Following	1
SPC	~16	34	~250	28	Following	1
547 548 549						
Coliform	<0.083	~0.083	4.4	~0.92	Following	1
<i>E.coli</i>	<0.083	<0.083	1.8	~0.75	Following	1
SPC	~0.83	33	4100	~21	Following	1
558 559 560						
Coliform	<0.083	<0.083	~0.33	~0.92	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	~0.33	Following	1
SPC	~3.3	~2.5	~16	40	Following	1
572 573 574						
Coliform	<0.083	<0.083	5	1.3	Leading	1
<i>E.coli</i>	<0.083	<0.083	3.3	1.3	Leading	1
SPC	~8.3	~4.2	~750	27	Leading	1
590 591 592						
Coliform	<0.083	~0.25	6.7	<0.083	Leading	1
<i>E.coli</i>	<0.083	~0.17	3.3	<0.083	Leading	1
SPC	150	~11	~330	~0.83	Leading	1

Body Numbers Test Type	Sampling Site				Carcase Side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
603 604 605						
Coliform	<0.083	<0.083	~15	~0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	5	<0.083	Leading	1
SPC	~2.5	~0.83	~1300	~7.5	Leading	1
614 615 616						
Coliform	~0.33	<0.083	6.7	~0.17	Leading	1
<i>E.coli</i>	~0.33	<0.083	1.7	<0.083	Leading	1
SPC	25	~18	~580	180	Leading	1
626 627 628						
Coliform	<0.083	<0.083	1.4	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	~0.25	<0.083	Leading	1
SPC	~11	~17	72	~7.5	Leading	1
635 636 637						
Coliform	<0.083	~0.17	12	3.1	Leading	1
<i>E.coli</i>	<0.083	~0.17	~0.25	<0.083	Leading	1
SPC	36	~18	~250	~13	Leading	1
643 644 645						
Coliform	<0.083	<0.083	1.8	~0.58	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	83	~5.8	~250	130	Leading	1
651 652 653						
Coliform	~0.083	<0.083	~0.25	~0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	~0.17	<0.083	Leading	1
SPC	34	~18	30	~1.7	Leading	1
664 665 666						
Coliform	~0.25	<0.083	~0.75	~1.1	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	~0.083	Leading	1
SPC	~6.7	~2.5	50	31	Leading	1
674 675 676						
Coliform	<0.083	~0.33	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	~0.33	<0.083	<0.083	Leading	1
SPC	~5	~8.3	~13	~2.5	Leading	1
685 686 687						
Coliform	~0.5	<0.083	~0.25	<0.083	Leading	1
<i>E.coli</i>	~0.5	<0.083	~0.17	<0.083	Leading	1
SPC	25	<0.83	~18	~8.3	Leading	1
696 697 698						
Coliform	<0.083	<0.083	~0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	~0.083	<0.083	Leading	1
SPC	~5.8	<0.83	~18	~1.7	Leading	1
710 711 712						

Body Numbers Test Type	Sampling Site				Carcase Side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
Coliform	<0.083	~1.1	~0.17	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	~0.083	Leading	1
SPC	<0.83	100	~22	~1.7	Leading	1
721 722 723						
Coliform	<0.083	<0.083	~0.67	~0.25	Leading	1
<i>E.coli</i>	<0.083	<0.083	~0.083	<0.083	Leading	1
SPC	<0.83	~0.83	170	~13	Leading	1
731 732 733						
Coliform	<0.083	<0.083	~0.5	1.3	Leading	1
<i>E.coli</i>	<0.083	<0.083	~0.083	<0.083	Leading	1
SPC	~1.7	~12	~170	~670	Leading	1
744 745 746						
Coliform	<0.083	<0.083	6.7	~0.17	Leading	1
<i>E.coli</i>	<0.083	<0.083	5	<0.083	Leading	1
SPC	~2.5	<0.83	~420	~16	Leading	1
756 757 758						
Coliform	<0.083	<0.083	<0.083	~0.67	Leading	1
<i>E.coli</i>	<0.083	<0.083	~0.083	~0.17	Leading	1
SPC	~3.3	~1.7	220	~24	Leading	1
770 771 772						
Coliform	<0.083	1.3	1.8	<0.083	Leading	1
<i>E.coli</i>	<0.083	1.3	~0.5	<0.083	Leading	1
SPC	~0.83	~5.8	~83	<0.83	Leading	1
782 783 784						
Coliform	<0.083	<0.083	~0.25	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	~0.083	<0.083	Leading	1
SPC	~4.2	~0.83	25	~9.2	Leading	1
794 795 796						
Coliform	<0.083	~0.083	~0.67	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	~0.5	<0.083	Leading	1
SPC	~0.83	<0.83	~21	~4.2	Leading	1
806 807 808						
Coliform	~0.25	~0.083	~0.58	~0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	~0.5	<0.083	Leading	1
SPC	~4.2	~5	47	<0.83	Leading	1
820 821 822						
Coliform	<0.083	<0.083	~0.75	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	~0.58	<0.083	Leading	1
SPC	~1.7	~1.7	~420	<0.83	Leading	1
836 837 838						
Coliform	<0.083	<0.083	~0.58	<0.083	Leading	1

Body Numbers Test Type	Sampling Site				Carcase Side	Cattle Cleanliness Score
	Brisket	Fore Arm	Neck	Shoulder		
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	~0.83	<0.83	~20	<0.83	Leading	1
845 846 847						
Coliform	<0.083	<0.083	~0.83	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	~0.083	<0.083	Leading	1
SPC	~0.83	~0.83	39	~2.5	Leading	1
851 852 853						
Coliform	<0.083	~0.083	~0.5	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	~0.083	<0.083	Leading	1
SPC	~3.3	<0.83	28	~6.7	Leading	1

Table 2: Chiller sample results

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
1124 1129 1143									
Coliform	~0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	~0.83	~0.83	<0.83	<0.83	<0.83	~0.83	<0.83	Following	1
1126 1127 1131									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	~0.83	<0.83	<0.83	<0.83	<0.83	<0.83	Following	1
1133 1139 1171									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	<0.83	~1.7	~0.83	<0.83	<0.83	<0.83	Following	1
1141 1155 1179									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	~0.83	~0.83	~0.83	<0.83	<0.83	<0.83	<0.83	Following	1
1144 1146 1147									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	<0.83	<0.83	<0.83	<0.83	<0.83	<0.83	Following	1
1148 1149 1150									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	~13	<0.83	<0.83	~22	<0.83	<0.83	Following	1

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
1151 1153 1154									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	~1.7	<0.83	<0.83	~1.7	~0.83	<0.83	Following	1
1152 1140 1142									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	~1.7	<0.83	~0.83	<0.83	~4.2	~4.2	<0.83	Following	1
1156 1158 1159									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	~9.2	~7.5	<0.83	~3.3	~0.83	~3.3	Following	1
1160 1163 1164									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	<0.83	~3.3	<0.83	~4.2	~0.83	~0.83	Following	1
1161 1172 1176									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	<0.83	<0.83	<0.83	~0.83	<0.83	<0.83	Following	1
1165 1167 1125									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	~0.83	~1.7	<0.83	<0.83	<0.83	<0.83	Following	1
1182 1216 1217									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	~1.7	~0.83	<0.83	<0.83	~0.83	<0.83	Following	1
1186 1188 1189									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	~1.7	~13	<0.83	~67	<0.83	~0.83	Following	1
1190 1192 1193									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	~2.5	<0.83	<0.83	~0.83	~5	<0.83	Following	1
1194 1195 1196									
Coliform	<0.083	<0.083	<0.083	<0.083	~0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	~5.8	~1.7	<0.83	<0.83	<0.83	<0.83	~2.5	Following	1
1197 1198 1200									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	~1.7	~0.83	<0.83	<0.83	~0.83	<0.83	Following	1
1201 1128 1136									
Coliform	<0.083	~0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	~0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	~0.83	~5	~0.83	<0.83	<0.83	<0.83	<0.83	Following	1
1203 1204 1162									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	~0.17	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	~0.83	<0.83	<0.83	~1.7	58	<0.83	Following	1

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
1205 1206 1207									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	~1.7	~5	<0.83	~1.7	~0.83	<0.83	Following	1
1208 1209 1210									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	<0.83	<0.83	<0.83	~0.83	~3.3	<0.83	Following	1
1212 1213 1214									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	<0.83	<0.83	<0.83	<0.83	<0.83	<0.83	Following	1
1218 1219 1177									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	<0.83	~0.83	<0.83	~0.83	<0.83	<0.83	Following	1
1220 1221 1183									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	~4.2	~1.7	0.83	~2.5	~0.83	<0.83	Following	1
1223 1184 1185									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Following	1
SPC	<0.83	<0.83	~0.83	<0.83	~0.83	<0.83	<0.83	Following	1
249 251 264									
Coliform	<0.083	~0.083	<0.083	<0.083	<0.083	~0.083	<0.083	Leading	1

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
<i>E.coli</i>	<0.083	~0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	~3.3	<0.83	<0.83	~0.83	~4.2	<0.83	Leading	1
316 342 343									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	~6.7	<0.83	<0.83	<0.83	<0.83	~5.8	<0.83	Leading	1
324 355 301									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	~0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	~0.083	<0.083	Leading	1
SPC	<0.83	~2.5	<0.83	<0.83	<0.83	~0.83	<0.83	Leading	1
331 332 333									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	~0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	~0.083	<0.083	Leading	1
SPC	<0.83	~0.83	~3.3	<0.83	~0.83	~8.3	<0.83	Leading	1
334 335 336									
Coliform	<0.083	~0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	~1.7	~2.5	~11	<0.83	~0.83	~6.7	~2.5	Leading	1
337 298 339									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	~3.3	~1.7	~2.5	~4.2	~12	~0.83	~0.83	Leading	1
340 313 315									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	~22	~6.7	~13	<0.83	<0.83	~0.83	130	Leading	1

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
346 347 349									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	~1.7	~2.5	<0.83	~0.83	<0.83	<0.83	Leading	1
350 352 353									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	~2.5	<0.83	<0.83	~2.5	~0.83	<0.83	Leading	1
354 318 319									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	~5.8	<0.83	~3.3	~0.83	~1.7	<0.83	Leading	1
1124 1129 1130									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	<0.83	<0.83	<0.83	~3.3	<0.83	<0.83	Leading	1
1133 1139 1171									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	~2.5	~1.7	<0.83	~0.83	<0.83	<0.83	Leading	1
1141 1155 1179									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	<0.83	<0.83	<0.83	~5.8	<0.83	<0.83	Leading	1
1143 1161 1172									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	~5.8	<0.83	<0.83	~4.2	<0.83	<0.83	Leading	1
1162 1206 1207									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	~3.3	~15	<0.83	~4.2	~0.83	~1.7	Leading	1
1176 1187 1166									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	~3.3	<0.83	<0.83	~4.2	<0.83	<0.83	Leading	1
1182 1216 1217									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	~0.83	~8.3	<0.83	~0.83	<0.83	<0.83	<0.83	Leading	1
1191 1199 1215									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	~0.83	~7.5	<0.83	<0.83	<0.83	<0.83	~2.5	Leading	1
1196 1197 1198									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	~0.83	~0.83	<0.83	<0.83	~0.83	<0.83	Leading	1
1200 1203 1204									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	~0.83	~3.3	~0.83	<0.83	~0.83	~0.83	<0.83	Leading	1

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
1201 1128 1136									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	~0.83	~5	~3.3	<0.83	<0.83	~0.83	<0.83	Leading	1
1202 1211 1170									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	<0.83	~0.83	<0.83	~2.5	<0.83	<0.83	Leading	1
1205 1194 1195									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	~0.83	<0.83	<0.83	<0.83	<0.83	~1.7	Leading	1
1208 1209 1210									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	<0.83	~0.83	<0.83	<0.83	~5.8	<0.83	Leading	1
1212 1213 1214									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	~27	<0.83	<0.83	<0.83	<0.83	<0.83	Leading	1
1222 1145 1157									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	<0.83	<0.83	<0.83	<0.83	<0.83	~0.83	Leading	1
1224 1175 1228									
Coliform	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1

Body Numbers Test Type	Sampling Site							Carcase Side	Cattle Cleanliness Score
	Brisket	Flank	Fore Arm	Loin	Neck	Rump	Shoulder		
<i>E.coli</i>	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	<0.083	Leading	1
SPC	<0.83	~1.7	<0.83	<0.83	<0.83	<0.83	~0.83	Leading	1

16 Appendix 8: Draft paper for publication

Investigating Alternative Sampling Sites for Microbiological Testing of Beef Carcasses for Process Hygiene Monitoring

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Abstract

The objective of this paper was to investigate alternative sampling sites for microbiological testings of beef carcasses in Australia. Four sites (neck, shoulder, forearm and brisket) were assessed at post evisceration and seven sites (neck, shoulder, forearm, brisket, flank, loin and rump) were assessed in the chillers. Swabs were tested for *Escherichia coli*, coliforms and Aerobic Plate Count (APC). Results of the sites used in the current National Carcass Microbiology Monitoring Program (formerly ESAM) were compared to the additional sites sampled in this study. The results of this study show that while the alternative sampling sites of the neck, shoulder and flank provide a greater number of unacceptable test results to assess hygienic processing compared to the traditional ESAM sampling sites, this increase at the alternative sampling sites has no significant difference either greater or lesser to the conventional sampling sites at the current location of testing in the chiller.

Keywords: Process hygiene monitoring, Abattoir, *Escherichia coli*, Aerobic Plate Count, Coliform, Beef

1. Introduction

All Australian export registered slaughtering establishments are currently required to take part in the National Carcass Microbiology Monitoring Program (formerly known as the *Escherichia coli* and *Salmonella* monitoring program (ESAM) (DAWR 2017). This program requires establishments to take swab samples from defined sites at the brisket, flank and rump of the carcass. (DAWR 2017). The defined sites are based on the USA Pathogen Reduction Final Rule introduced in 1996. Carcasses are sampled after a minimum of 12 hours active chilling and swabs are tested for Aerobic Plate Count (APC), *Escherichia coli* (*E.coli*) and coliforms as a means of process hygiene verification (DAWR 2017).

The objective of this paper is to investigate alternative sampling sites for microbiological testings of beef carcasses in Australia. Distribution of bacterial counts on a carcass are affected by deposition and redistribution of organisms as well as decontamination (Gill et al 1996). There are multiple points during processing where changes in microbiological load and distribution can occur during processing. Location of sampling in the process has been investigated in previous research including sampling following de-hiding (Gill et al 1996), pre evisceration (Wang et al 2013), post evisceration (Gill et al 1996;), after trimming (Gill et al 1996;) and following carcass washing (Gill et al. 1996) or post intervention (Wang et al 2013).

Previous research has also utilised a number of different sampling sites, locations and times during processing in their studies, in part due to the complexities of sample collection in an operational setting as well as the complexities in process hygiene. Previous studies have considered both forequarter sites such as neck (Gill et al 1996; Untermann et al 1997; Zweifel et al 2008), brisket (Gill et al 1996; Untermann et al 1997; Zweifel et al 2008), shoulder (Untermann et al 1997) and forearm (Untermann et al 1997) as well as hindquarter sites such as flank (Zweifel et al 2008), rump (Gill et al 1996; Zweifel et al 2008) and back (Untermann et al 1997). Medial sites such as pleura and medial neck have also been studied however counts on the medial side are significantly lower than those on the lateral side (Untermann et al 1997). Studies have demonstrated microbiological detections at sites on the carcass (Gill et al 1996; Untermann et al 1997; Zweifel et al 2008) other than those currently defined by DAWR in the “Microbiological Manual for Sampling and Testing of Export Meat and Meat Products”. A number of studies identified neck as a suitable site (Untermann et al 1997; Zweifel et al 2008). Forearm and shoulder were also identified as suitable microbiological sampling sites (Untermann et al 1997). The present study was designed to evaluate alternative sampling sites for microbiological testings of beef carcasses in Australia.

2. Material and Methods

2.1 Sample collection – slaughter floor

A total of 580 swab samples were taken on the slaughter floor of four establishments on the Eastern seaboard of Australia. Samples were taken at the point of post evisceration with sampling conducted at chain speed. To ensure that each carcass side was sampled only once in the study and to ensure sample collection from both sides of the carcass, sampling was conducted by allocating bodies to either leading or following sampling based on a set range of body numbers. For example, if the day's production was 1000 head, body 1 – 500 was eligible for sampling of the leading side while body 501

– 1000 was eligible for sampling of the following sides. Four sites were sampled on the carcass – brisket, neck, shoulder and forearm. Forequarter sites only were sampled on the slaughter floor due to work health safety considerations and to allow collection of samples at chain speed. Each site was sampled on three consecutive carcasses with the same swab to provide a composite sample with an area of 300cm². All four sites were sampled on the same set of three carcasses to allow comparison of the sites. Sampling was conducted using the Whirl-packTM collection method as outlined in the “Microbiological Manual for Sampling and Testing of Export Meat and Meat Products” (DAWR 2017).

2.2 Sample collection – chillers

A total of 940 swab samples were taken in the chillers of four establishments on the Eastern seaboard of Australia. Samples were taken on the carcasses from the same production day as those sampled on the slaughter floor. To ensure that each carcass side was sampled only once in the study and to ensure sample collection from both sides of the carcass, sampling was conducted by allocating bodies to either leading or following sampling based on a set range of body numbers. For example, if body 1 – 500 were eligible for sampling of the leading side on the slaughter floor, these carcasses were eligible to be sampled on the following side in the chiller. Samples were collected at least 12 hours after active chilling at three establishments. At one establishment, samples were collected after at least 6 hours of active chilling as each day’s production was loaded out before the next day commenced. Seven sites were sampled on the carcass – brisket, neck, shoulder, forearm, flank, loin and rump. At three establishments all seven sample sites were sampled on a set of three carcasses. At one establishment, sampling of chilled carcasses was only operationally practical if conducted in the chiller corridor at chain speed, due to a lack of available space in the chillers and marshalling area. As chiller sampling required the sampling of seven sites (as opposed to the four sites sampled on the slaughter floor), it was not possible to complete all seven sites at chain speed on the same set of three carcasses. Sampling for this establishment was split into forequarter sampling (Neck, Shoulder, Forearm, Brisket) across one set of three carcasses and hind quarter sampling (Loin, Flank, Rump) across the next set of three carcasses when sampling at chain speed. Where practical (i.e. when the chain was stopped on breaks), all seven samples were collected on a set of three carcasses as conducted at the other three establishments in the study. Each site was sampled on three consecutive carcasses with the same swab to provide a composite sample with an area of 300cm². Sampling was conducted using the Whirl-packTM collection method as outlined in the “Microbiological Manual for Sampling and Testing of Export Meat and Meat Products” (DAWR 2017).

2.3 Transport of samples

Sponge samples were stored chilled prior to and during transport to the testing laboratory, either the same day or overnight. The testing laboratory was NATA (National Association of Testing Authorities) accredited to ISO 17025 and an approved laboratory with the Department of Agriculture & Water Resources (DAWR) for the estimation of Aerobic Plate Count (APC), generic *E. coli* and coliform bacteria in meat and meat products. All samples were analysed no later than on the day following collection at the establishment. Samples arrived at the testing laboratory at $<5^{\circ}\text{C}$.

2.4 Test methodology

The standard DAWR approved test methodology for ESAM samples was used to test, analysis and report the results of the swab sampling. Each 300cm² composite sample was tested for *E. coli*, coliforms and Aerobic Plate Count (APC).

2.5 Microbiological analysis

After manually palpating sponges (~1 min), one mL was removed and serially diluted in PSS (Peptone Salt Solution). Appropriate dilutions were plated onto Aerobic Plate Count (APC) PetrifilmTM and *E. coli*/coliform (*E. coli*) PetrifilmTM. *E. coli* and coliforms were taken directly from the sponges as well as serially diluting. After incubation at $35\pm 1^{\circ}\text{C}$ for 24-48±3h colonies were counted for the target organism following the manufacturer's instructions and results expressed as CFU/cm². From the colonies counted, a laboratory worksheet was used to convert a result from the original surface area. The limit of detection of this method, based on the sampling area undertaken, was 0.83 CFU/cm² for APC and 0.083CFU/cm² for *E. coli* and coliforms.

2.6 Statistical Analysis

Sample results were classified based on the detection levels set by the DAWR in the "Microbiological Manual for Sampling and Testing of Export Meat and Meat Products" for *E. coli* are defined as $>0\text{cfm}/\text{cm}^2$ to $<20\text{cfm}/\text{cm}^2$ is considered marginal and $\geq 20\text{cfm}/\text{cm}^2$ is considered unacceptable. For APC the detection levels are defined as $>1000\text{cfm}/\text{cm}^2$ to $<31625\text{cfm}/\text{cm}^2$ is considered marginal and $\geq 31625\text{cfm}/\text{cm}^2$ is considered unacceptable. As such this study classified a positive result for *E. coli* detection as $>0.084\text{cfm}/\text{cm}^2$ with results $\geq 20\text{cfm}/\text{cm}^2$ is considered unacceptable (U). Coliform detections were deemed positive when $>0.084\text{cfm}/\text{cm}^2$ with results $\geq 20\text{cfm}/\text{cm}^2$ considered unacceptable (U) and APC were deemed positive when $>1000\text{cfm}/\text{cm}^2$ broken down to marginal (M) ($>1000\text{cfm}/\text{cm}^2$ to $<31625\text{cfm}/\text{cm}^2$) and unacceptable (U) ($<31625\text{cfm}/\text{cm}^2$). Two-sample

proportional tests (z tests) with a 95% confidence interval were performed to determine whether there was a significant difference between sampling sites.

3 Results and discussion

A total on 1520 swabs samples were taken, each tested using DAWR approved testing methodologies for coliforms, *E. coli* and APC.

At post evisceration, 580 swab samples were taken on the slaughter floor across the four sampling sites of brisket, forearm, neck and shoulders resulting in 145 sampling sets. Eighty-two sample sets had a minimum of one positive result, 38 of these samples were taken from leading carcass sides and 44 were samples taken from following carcass sides. Three unacceptable APC results were reported in the post evisceration samples all taken from shoulder site swabs on following carcass side sets. No unacceptable *E.coli* or coliform results were found at post evisceration. Table 1 provides a summary of the total sample sets and the number of positive sample sets by sampling site at post evisceration.

Table 1 – Summary of Post Evisceration Results based on Sampling Site

Post Evisceration	Sampling Site			
No. of Sample Sets: 145	Brisket	Forearm	Neck	Shoulder
No. of Positive Sample Sets	23	23	65	39
Percentage	15.9%	15.9%	44.8%	26.9%

When considering the conventional brisket sampling site and the alternative sampling sites (forearm, neck and shoulder) with respect to the percent of positive results a significant difference was found between the comparison of neck to brisket (Z-Score = 5.3645, p-value = 0) and between the shoulder and brisket (Z-Score = 2.2917, p-value = 0.02202).

In the chiller, 940 swab samples were taken across the four establishments. One hundred and five full sample sets were taken with the addition of 28 forequarter and 31 hind quarter split sample sets taken at the chiller sampling location. The full sample sets were split with 53 sample sets taken from leading carcass sides and 52 sample sets taken from following carcass sides. The forequarter sample sets were split with 13 sample sets taken from leading carcass sides and 15 sample sets taken from following

carcase sides. The hind quarter sample sets were split with 15 sample sets taken from leading carcase sides and 16 sample sets taken from following carcase sides.

Twelve full sample sets had a minimum of one positive result, 2 of these samples were taken from leading carcase sides and 10 samples were taken from following carcase sides.

Four forequarter split sample sets had a minimum of one positive result, these samples were taken from shoulder sampling sites, 2 from leading carcase sides and 2 from following sides.

Eleven hind quarter split sample sets had a minimum of one positive result, 2 of these samples were taken from following carcase sides, 1 sample at the loin sampling site and 1 sample at the rump sampling site. The remaining 9 hind quarter split sample sets with a minimum of one positive result were from flank sampling sites of leading carcase sides.

One unacceptable APC results was reported from the chiller samples from a rump site swab of a following carcase sides. One unacceptable *E.coli* and one coliform result were also reported from the same loin site swab from a hind quarter chiller sample taken from following carcase sides. Table 2 provides a summary of the total sample sets (including split samples) and the number of positive sample sets by sampling site in the chillers.

Table 2 – Summary of Chiller Results based on Sampling Site

Chiller	Sampling Site						
No. of Sample Sets:	Brisket	Flank	Forearm	Loin	Neck	Rump	Shoulder
133 forequarter							
136 hind quarter							
No. of Positive Sample Sets	2	12	1	1	3	4	5
Percentage	1.5%	8.8%	0.8%	0.7%	2.3%	2.9%	3.8%

When considering the conventional sampling sites (brisket, flank and rump) and the alternative sampling sites (forearm, loin, neck and shoulder) with respect to the percent of positive results a significant difference was found between the flank sampling site and either the forearm (Z-Score = 3.863, p-value = 0.002), loin (Z-Score = 3.1265, p-value = 0.00174) or neck (Z-Score = 2.3471, p-value = 0.01878). sampling sites.

The ESAM sampling program uses a single swab to sample the brisket, flank and rump sampling sites. Given the significant difference between the neck and shoulder sampling sites and the brisket sampling site at post evisceration the results have been considered on the basis of at least one positive result in a sample set across the conventional ESAM sampling sites compared to alternative sample sites of the neck, shoulder and flank. These results are summarised in Table 4 with the Z-score and p-value for comparison.

Table 4 – Summary of the No. of Positive Sample Set Results when considering Conventional Sampling Sites versus Alternative Sampling Sites

	Conventional Sampling Sites	Alternative Sampling Sites	Analysis
Post Evisceration: No. of Sample Sets: 145			
No. of Positive Sample Sets	23	74	The Z-Score is 6.3475. The p-value is 0. The result is significant at $p < 0.05$.
Percentage	15.9%	51.0%	
Chiller: No. of Sample Sets: 164			
No. of Positive Sample Sets	17	20	The Z-Score is 0.5236. The p-value is 0.60306. The result is not significant at $p < 0.05$.
Percentage	10.4%	12.2%	

Although the results show that a higher number of sample sets that have at least one positive result at the alternative sampling sites compared to the conventional sampling sites at both the post evisceration and in the chillers, there is only a significant difference (to a confidence interval of 95%) in the post evisceration results. Given the selection of two forequarter sites in the alternative sampling versus the single forequarter site in the conventional sampling and that only forequarter sampling occurred at post evisceration, this could have skewed the data to result in this significance.

Based on these results while the alternative sampling sites of the neck, shoulder and flank provide a greater number of unacceptable test results to assess hygienic processing compared to the traditional

ESAM sampling sites, this increase at the alternative sampling sites has no significant difference either greater or lesser to the conventional sampling sites at the current location of testing in the chiller, based on this research and these results.

4 Conclusions

The purpose of this study was to evaluate alternative sampling sites of microbiological testing of beef carcass in Australia. Based on the results of this study while the alternative sampling sites of the neck, shoulder and flank provide a greater number of unacceptable test results to assess hygienic processing compared to the traditional ESAM sampling sites, this increase at the alternative sampling sites has no significant difference either greater or lesser to the conventional sampling sites at the current location of testing in the chiller, based on this research and these results.

Declaration of interest

None.

Acknowledgements

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17 Appendix 9: Draft Export Meat Industry Advisory Committee and Food Export Regulatory Steering Committee Paper

Agenda Item:

EXPORT MEAT INDUSTRY ADVISORY COMMITTEE /

FOOD EXPORT REGULATORY STEERING COMMITTEE

Agenda Topic:

FOR DECISION

Recommendation:

That the committee consider the option for interested establishments:

1. to utilise a risk based testing frequency for process hygiene monitoring based on cattle cleanliness scoring and two point (post-evisceration and chiller) microbiological sampling to demonstrate process hygiene control, and
2. use alternative sampling sites for process hygiene monitoring. These alternatives being the neck, shoulder and flank.

That for this to occur equivalency submission to trading partners to accept these new testing parameters in lieu of the current testing methodology.

Executive summary

A project has been completed that aimed to review the current food safety programs (both regulatory and commercial) and assess whether or not it might be more beneficial to undertake microbiological testing at other points in the process and/or other parts of the carcass that allow for evaluation of hygiene process to reflect risk.

Testing at two locations, post evisceration and in the chiller, to measure hygienic process efficiency has demonstrated how meaningful the analysis of samples collected at the two locations can be in understanding the level of hygienic process control rather than the use of a single location. With regulatory agreement and equivalence from customers and trading partners, this process would not only allow for a better understanding of process hygiene by all, but could be used as a risk

framework with cattle cleanliness for testing frequency should the industry wish to seek equivalence.

Based on the project results, while the alternative sampling sites of the neck, shoulder and flank provide a greater number of unacceptable test results to assess hygienic processing compared to the traditional ESAM sampling sites of rump flank and brisket, this increase at the alternative sampling sites has no significant difference either greater or lesser than the conventional sampling sites based on this research and these results. Therefore, it could be argued that the alternative sites are just as effective and if a processor wished to use these as an alternative to the traditional ESAM sites due to Work Health and Safety (as in a number of cases these sites can be assessed without a step ladder) this research supports that variation.

Key Issues

When considering recommendations, it must be remembered that the current ESAM program was design to provide US market access, during the past 21 years the program has also been used to meet the minimum expectations of a number of other customer and trading partners. Whether or not it is of scientific merit is therefore a minimum standard for the industry. The reality therefore, is that any changes to this program would need to be seen as beneficial by customer or trading partners irrespective of the financial benefit to Australian processors.

With regard to resources the project team offer support in the drafting of equivalency submission both to trading partners (and customers) given their regulatory background.

Background

The literature review concluded that there is no current regulatory barrier to changing the microbiological testing requirements in Australian beef abattoirs on the basis of risk. Furthermore, the literature proposes alternatives which may yield more meaningful process control outcomes. To assess this, carcasses were tested from livestock with graded hide cleanliness during processing and post intervention at varying sites on the carcass, with comparative ESAM site samples, taking into account leading and following carcass side variation.

Samples were taken from four beef establishments down the Eastern seaboard of Australia. The samples were collected from the neck, shoulder, fore arm and brisket on the slaughter floor and chiller. An additional three samples at the rump, loin and flank were taken in the chillers; 309 sample sets in total, with 1520 swabs being tested for *E. coli*, coliforms and Total Viable Counts using the approved ESAM methodology.

Expected outcomes and benefits

The expected outcomes and benefits to both government and industry are that through adoption of the recommendation establishments' are provided, the option of a sampling protocol which provides a better understanding of their process hygiene and potential benefit of improved work, health and safety which can be cost beneficial.

Full project details are available in MLA Final Report V.MFS.0401: Review of current food safety microbiological sites and the establishment of a more meaningful measure to assess hygienic production. Further information and support is available by contacting the project team on 0410 690819.