# final report

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## Statistical Process Control – Hygiene and Hazards

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# Abstract

Testing requirements for export of beef trim to the United States are continuing to increase in stringency and more emphasis is therefore being placed on improving process control during the slaughter of cattle. This project was undertaken to assess slaughter processes that ensure a very low prevalence of undesirable enteric microorganisms and to propose alternative testing systems to identify significantly increased risk of the detection of pathogenic *E. coli*. A range of investigations were undertaken and important practices were collated in a revision of the MLA “Incoming livestock and slaughter process assessment tool for beef”. Alternative test systems should include larger sampling areas, higher risk carcase sites, microbial indicators that are more frequently detected than *E. coli*, and better alignment between carcase and carton samples to allow better through-chain assessment of microbial contamination.

# Executive Summary

## Where were we in 2013?

In late 2011, the US Food Safety and Inspection Service (FSIS) declared six additional serotypes of shiga toxigenic *E. coli* (STEC) – O26, O45, O103, O111, O121, and O145 – as adulterants. Subsequently, verification testing for these serotypes was introduced for Australian meat processors exporting manufacturing beef to the United States in June 2012. In addition, processors were required to re-assess their HACCP plans for the additional six serotypes.

At about the same time, there has been increasing international focus on improving process control and utilising relevant and suitable data to inform better risk management. The aim of these developments was to reduce reliance on end product testing, *i.e.* robust N-60 testing for *E. coli* O157 and STEC in manufacturing beef to the United States.

In response to these international developments, investigations were undertaken of national microbiological databases (*E. coli* and *Salmonella* Monitoring (ESAM) and Product Hygiene Index (PHI)) in late 2012. However, when process control indicators, such as zero tolerances, MHA scores, and *E. coli* prevalence, were assessed they indicated infrequent loss of process control. In addition, no relationship between these indicators and *E. coli* O157 detections could be ascertained, although higher detection rates were associated with calves and, to a lesser degree, dairy cattle. However, problems with data quality / integrity (*e.g.* due to data entry errors) were noted.

An important aspect of process control is understanding the processing steps which add to or remove microbial contamination from the carcase. The “Incoming livestock and slaughter process assessment tool for beef” had been developed by MLA and made available to QA Managers for this purpose in 2005, but changes in the industry over the last decade were not reflected in this tool. Furthermore, to assist QA Managers with their HACCP re-assessments, MLA ran training workshops on how to undertake investigations that would provide quantitative evidence of the effects of different processing steps and intervention strategies. However, on completion of the training it was apparent that QA staff would need assistance to appropriately analyse the data they collected.

The purpose of this project was to provide the statistical capability to slaughter establishments and the industry to better understand and control microbial hazards during slaughter and dressing. In particular, on both the national and plant level, there was a need to identify risk factors, trends and times of increased risk of microbial hazard contamination, conduct process investigations, design experiments to evaluate alternate process operations, validate interventions and evaluate current process control testing systems.

## Where are we in 2014?

Several establishments provided or collected data throughout this project, with the aim of identifying risk factors or evaluating process changes and interventions. Findings from investigations included:

- Increased risk of STEC detections at one plant possibly due to long haul cattle and processing on shifts with more learner operators.
- Deep trimming of cutting lines is effective at reducing *E. coli* contamination of carcases.
- *E. coli* detections are higher on hot carcases across a range of carcase sites, especially at the rump.
- A hot water pasteuriser is effective at reducing microbial carcase contamination and also results in the apparent redistribution of microorganisms from the top of the carcase (rump) to the bottom (neck).
- The use of ultraviolet light for decontamination of boning room belts was found to be ineffective in reducing TVC and *E. coli* to justify the expenditure.

These analyses have increased the value QA Managers have gained from their investigations by providing them with appropriate data analysis and a better understanding of their process. In addition, some investigations have helped them evaluate the efficacy and hence benefit of potential process interventions, prior to large expenditure of money. Many of these investigations have been written up and collated in a case study booklet and a second edition with additional investigations is currently being prepared.

Investigations that were undertaken with national data, i.e. ESAM, did not identify any trends. This was due to uncertainty in the validity of the data being collected. For example, some plants apparently had no *E. coli* detection from sheep for several months, which was considered to be highly unlikely. However, no reason for these apparently incorrect data could be identified.

Investigation of levels of TVC on carcases and in cartons showed that across all Australian beef export establishments there is a slight correlation between carcase and carton levels, with higher carton levels being observed for higher carcase levels. However, on the plant level, no such relationship is apparent – instead many plants showed considerably more variability on carcases than in carton product. This indicates that incoming contamination into the boning room is distributed and possibly reaches a plant-specific equilibrium point sometime during the day, which results in carton product with similar microbial counts day after day. In addition, this analysis also identified plants which had few TVC detections on the carcase or in the carton. Subsequently, it was found that these plants had inappropriately high limits of detection for TVC. Feedback provided to these plant resulted in a change in dilutions and more appropriate data.

Establishments were also compared according to the variability in their microbial indicators within and between months. This was done in an attempt to ascertain whether high variability, either within months, between months or both, were associated with higher STEC detection rates. Again, no relationship could be found.

As part of this project, a series of establishment visits were undertaken to identify practices that may affect hygienic slaughter performance. These visits involved interviews of QA Managers about livestock, processing, and staffing and observation of plant operations, including pre-slaughter practices, slaughter, dressing and boning room operations. The information that was collected formed the basis for a revision of the 2005 Beef Tool, which was based on:

- Problem – assessing the incoming problem (livestock tag score and time in transport) and
- Process – the ability of the process to cope with the problem.

The revised tool includes additional questions, based on findings in this and previous MLA funded work. Scores for the incoming problem now include processing of calves, feedlot and dairy cattle. Scores for the process include the effects of hide interventions, chain speed, and other mitigation measures. An additional dimension related to People (turnover and training) has also been added in recognition that even a well-designed process may not achieve the desired results if slaughter staff are not adequately skilled to perform the work. The Total Score, which combines the scores awarded to the Problem, Process and People, related only to generic *E. coli*, but not Total Viable Counts or STEC, and considerable variability remains unexplained. This may be because not all the right questions are included, that the scores awarded for each answer are not appropriate, or that day-to-day differences in livestock and operations could not be adequately captured. Feedback on the revised tool is currently being sought via MINTRAC MI&QA networking meetings and work on the tool is continuing outside this project.

## Where to from here?

Given the international developments over the last two years, it is clear that the need to demonstrate effective process control in meat processing will continue to gain importance. However, from the current and previous work it is evident that the majority of Australian processors are achieving low levels of microbial contamination and process performance indicators as measured by existing systems (ESAM, MHA, etc). Because of this, current microbial indicators, such as *E. coli*, occur too infrequently to be useful to analyse national and plant specific trends. In addition, the lack of relationship between carcases and end product and the limited information captured on end product and boning room operations makes through-chain analysis impossible.

Future studies of process control should therefore focus on the following:

- Sampling larger carcase areas will be necessary to increase the frequency and sensitivity with which hygiene indicator organisms, such as generic *E. coli*, are detected, which will lead to better understanding of process control.
- The utility of alternate indicators, which are detected more frequently than *E. coli*, to identify lack of process control or times of increased risk of STEC contamination should be investigated.
- Incoming contamination into boning rooms, in the form of carcases, has been studied extensively, but little information exists with respect to the distribution of microorganisms in boning room. In addition, current data capture systems are inadequate in terms of relating incoming carcase contamination with end product (*i.e.* carton) contamination, which makes comparisons impossible. Consequently, future work should focus on being able to better relate incoming and outgoing contamination.
- Aligning carcase and end product microbiological samples and other hygiene indicators to allow for better comparison and identification of relationships and trends.

In addition, it is recommended that:

- The need for delivering investigation workshops to new QA Managers and other staff should be evaluated. If enough interest is expressed, then these workshops should again be delivered to help the industry to continue to improve their slaughter and dressing processes.
- Support for statistical and data analysis to QA Managers is continued, that is, to assist them in planning, running, analysing and reporting process investigations and process improvement activities.

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# 1 Background

Australian manufacturing beef destined for grinding in the US is required to be tested for *E. coli* O157 using robust N-60 testing since 2007. More recently, the US Food Safety and Inspection Service (FSIS) has focused on six additional serotypes of *E. coli*: O26, O45, O103, O111, O121, and O145. These have now also been declared adulterants and processors are required to re-assess their HACCP plans for the additional six shiga toxigenic *E. coli* (STEC) serotypes. As a result, the Australian industry has been undertaking testing of manufacturing beef for all seven *E. coli* serotypes of interest. There has been a higher detection rate than anticipated.

In addition, MLA has funded work to investigate potential risk factors and interrogate national microbiological databases (ESAM and Product Hygiene Index) for indications of loss of process control, that is, the ability of plants to control microbial hazards. This work found data integrity issues and collection of large amounts of data with limited value for identifying increases in overall probability of detection, but specifically higher detection of *E. coli* O157 associated with calves and to a lesser degree processing of cows and bulls, i.e. mainly dairy cattle.

Furthermore, in the US pathogenic *E. coli* in beef products continue to result in recalls and foodborne illness outbreaks, despite current control measures and testing requirements. Consequently, international focus is being placed on improving process control and utilisation of data to inform better risk management.

The purpose of this project was to provide the statistical capability to slaughter establishments and the red meat industry to better understand and control microbial hazards during slaughter and dressing.

# 2 Project Objectives

1. Investigate slaughter process hygiene to determine approaches, practices and criteria by which processes can be considered to achieve standards suitable to ensure a very low prevalence of undesirable enteric microorganisms on beef carcases.
2. Propose tests and testing systems (frequency, microbiological criteria, and statistical methods) that can alert an establishment or national bodies to a significantly increased risk of the detection of pathogenic *E. coli*.

In addition, the following details were stipulated in the terms of reference.

1. MLA will be involved in the recruitment of a suitable person for this project and recruitment of processing establishments. MLA will also appoint a small steering committee to oversee specific actions being undertaken.
2. Visit ten slaughter establishments and work with QA staff to assist them to better understand their slaughter and dressing processes.
3. Encourage establishments to develop process improvement related projects (possibly PIPs), to secure funding and deliver projects.

4. Analyse existing national data to identify increases in the risk of pathogenic *E. coli* contamination, areas of incomplete data, and a need for additional data collection.
5. Work with establishments to undertake experimental work to assess the effects of processing operations on relevant microbial contamination.
6. Work with five establishments which have had pathogenic *E. coli* detections to undertake a detailed process data analysis (ESAM, PHI and in-house data) to better understand risk factors and identify indicators of loss of process control and increased risk of pathogenic *E. coli* O157 contamination.
7. Contribute to development of case studies that can be used by other establishments to improve their own process control.
8. Interact with MLA's scientific risk management panel as required.

# 3 Project Summary

The following summary provides an overview of achievements of the two project objectives. The additional details (1-8 above) are addressed under the first project objective and these resulted in a revision of the *Incoming livestock and slaughter process assessment tool for beef* (the "Beef Tool") developed in 2005.

## 3.1 Objective 1: Investigate slaughter process hygiene

> *Investigate slaughter process hygiene to determine approaches, practices and criteria by which processes can be considered to achieve standards suitable to ensure a very low prevalence of undesirable enteric microorganisms on beef carcases.*

The methodology followed in this project included recruitment of a suitable candidate, familiarisation with the industry, including plant operations and national data sources – *E. coli* and *Salmonella* Monitoring (ESAM) and Product Hygiene Index (PHI) data. The following sections provide information about the eight *Additional Details* that were included in the project schedule with the aim of providing some guidance in achieving the project objectives.

A range of analyses and investigations were undertaken in this project to assist establishments with gaining a better understanding of their processing hygiene. These analyses, together with the investigation of hygienic slaughter practices at eight establishments, led to a revision of the 2005 *Incoming livestock and slaughter process assessment tool for beef* (see Section 3.1.9).

### 3.1.1 MLA will be involved in the recruitment of a suitable person for this project and recruitment of processing establishments. MLA will also appoint a small steering committee to oversee specific actions being undertaken.

The recruitment panel, consisting of Andreas Kiermeier (SARDI), David Hamilton (SARDI) and Ian Jenson (MLA) interviewed a number of candidates. A recent

statistics graduate from The University of Adelaide, Sam Rogers, was recruited in July 2013.

The steering committee consisted of Ian Jenson (MLA Project Manager), Andreas Kiermeier (formerly SARDI, then MLA Consultant) and John Sumner (MLA Consultant).

### 3.1.2 Visit 10 slaughter establishments and work with QA staff to assist them to better understand their slaughter and dressing processes.

The intention for this part of the project was two-fold. Firstly to provide an introduction to red meat processing and the ESAM data collection, and its limitations, and secondly to introduce Sam Rogers and provide an opportunity for establishments to work with and utilise Sam's data analysis skills for on their process investigations.

A total of 13 slaughter establishments (12 export and one domestic) in South Australia, Victoria, Queensland and New South Wales were visited between August and October 2013. Throughout the project several establishments utilised Sam's skills, many of which are documented in the second edition of the "Processor's Guide to improving Microbiological Quality" (in preparation).

Sam Rogers also attended several MINTRAC MI&QA meetings, which provided an opportunity to meet QA managers and other industry representatives.

### 3.1.3 Encourage Establishments to develop process improvement related projects (possibly PIPs), to secure funding and deliver projects.

Throughout the project, several establishments undertook small investigations, but only one was deemed suitable for a Plant Initiated Project (PIP). In this PIP the microbiological performance of a hot water intervention and chilling were investigated. Sam Rogers was involved in the design of the experimental work and performed the statistical analysis (Rogers, 2014). Samples were regularly collected from carcases prior to the pasteuriser, after the pasteuriser and after chilling.

The three main findings of the project were:

1. The hot water pasteuriser is effective in reducing *E. coli* contamination,
2. There was a redistribution of microorganisms from top to bottom of the carcase, and
3. There is a growth of microorganisms on the lower carcase sites during chilling.

These findings were presented to industry at the 2014 MINTRAC conference and are available on the MLA website. The findings are also included in the second edition of the "Processor's Guide to Improving Microbiological Quality" (in preparation).

### 3.1.4 Analyse existing national data to identify increases in the risk of pathogenic *E. coli* contamination, areas of incomplete data, and a need for additional data collection

From early data familiarisation and analysis attempts, it was apparent that the large amounts of data that are collected in the ESAM and PHI databases are of limited value for identifying trends and increases in overall probability of detection of *E. coli* O157 and STEC. This is due to a range of reasons, including:

1. Generic *E. coli* and other hygiene indicators (TVC and coliforms) on carcases and in cartons are generally low (both prevalence and concentration);
2. *E. coli* O157 and STEC are infrequently detected in cartons of beef trim;
3. Process and Product hygiene monitoring results, *e.g.* Meat Hygiene Assessment (MHA), are generally good;
4. There is no direct relationship between process/carcase monitoring (microbial and non-microbial) and *E. coli* O157/STEC testing;
5. Data errors and reliability issues in ESAM and PHI data.

In light of this, alternative approaches to the analysis of national data were discussed. The following four possible data investigations were proposed during a teleconference of the steering committee on 4th March 2014.

1. Analyse climate effects in relation to ESAM data to investigate if drought, high rainfall or other extreme weather conditions contribute to poor process hygiene. Some work on this was undertaken in 2011 by SARDI, but there is scope for extension and update.
2. Analyse data supplied directly from a chosen establishment, to try and determine risk factors. Work of this type was done in 2005/2006 by MLA vacation student Karl Jackson. This would only be feasible if the chosen plant already has extensive electronic records. This option was not pursued.
3. Investigate variability of box plots from ESAM reports for individual plants to compare plants with high and low variability and explore any possible contributing factors.
4. Investigate box plot outliers from individual plants' ESAM reports to seek a cause for some of these outliers. It was decided that this approach is not likely to be feasible as most plants would not have adequate information about individual animals so it is unlikely that a cause would be found in most cases. Hence, this option was not pursued.

Extreme weather events and the effect of these on microbiological results in plants were investigated. This work formed the basis of a case study in the second edition of the "Processor's Guide to Improving Microbiological Quality" (in preparation).

Variability in monthly ESAM results (TVC, coliforms and *E. coli*) were analysed and plants were categorised according to whether they had low/high variability within and between months. Several plants were selected for further detailed process investigation and identification of potential risk factors for increased *E. coli* O157 contamination, as described in section 3.1.6.

The relationship between ESAM carcase swabs and carton samples were also investigated, but there appears to be little relationship between the two. Many establishments had very little variability in their carton results, but much higher variability in their carcase results. This implies that regardless of the incoming contamination on carcases into the boning room, it is “evened out” and ends up reasonably consistent in cartons at most plants. This is summarised in Appendix 2: Relationship between carcase and carton data.

Since October 2014, data entry into the ESAM database has been completed transitioned to the PHI system (via Excel spreadsheets). Subsequently, the quality of the ESAM data has improved, but substantial delays in data availability is limiting reporting and utility of the data.

### 3.1.5 Work with Establishments to undertake experimental work to assess the effects of processing operations on relevant microbial contamination

Throughout this project, several slaughter establishments were assisted with data analysis and process investigations. Some of these establishments experienced an increase in the number of STEC confirmations and requested assistance in investigating risk factors. Other plants had collected in-house data over several years, and requested help with examining data for indicators of loss of process control. Investigations undertaken include:

- Increased risk of STEC detections at one plant possibly due to long haul cattle and processing on shifts with more learner operators.
- Deep trimming of cutting lines is effective at reducing *E. coli* contamination of carcases.
- *E. coli* detections are higher on hot carcases across a range of carcase sites, especially at the rump.
- A hot water pasteuriser is effective at reducing microbial carcase contamination and also results in the apparent redistribution of microorganisms from the top of the carcase (rump) to the bottom (neck).
- The use of ultraviolet light for decontamination of boning room belts was found to be ineffective in reducing TVC and *E. coli* to justify the expenditure.

Some of these investigations were included in the second edition of the “Processor’s Guide to Improving Microbiological Quality” (in preparation).

### 3.1.6 Work with five establishments which have had pathogenic *E. coli* detections to undertake a detailed process data analysis (ESAM, PHI and in-house data) to better understand risk factors and identify indicators of loss of process control and increased risk of pathogenic *E. coli* O157 contamination.

In November 2014, Sam Rogers (SARDI), Andreas Kiermeier (MLA Consultant) and Clive Richardson (MINTRAC) visited eight beef slaughter establishments, with low and high *E. coli* O157/STEC detection rates, to obtain detailed information about their slaughter practices. Answers to a series of questions were obtained either

directly from QA Managers or from observing slaughter floor and boning room operations (Appendix 1: Process control interviews).

The answers were subsequently assessed during a meeting in Adelaide in January 2015. Together with findings from other investigations undertaken in this project this lead to a proposed revision of the 2005 Beef Tool, which aims to summarise the important practices that affect beef carcases hygiene (see Section 3.1.9).

### 3.1.7 Contribute to the development of case studies that can be used by other establishments to improve their own process control

Prior to this project commencing, MLA ran process investigation training workshops in April-May 2013. During the workshops, participants undertook investigations in their establishments, and each participant was actively involved in designing, running, analysing and reporting their chosen investigation.

As part of this project, the investigation reports were collated and published in the first edition of the "Processor's Guide to Improving Microbiological Quality" (MLA, 2014) in February 2014. The intention was to demonstrate the types of investigations that can be undertaken, and the findings as inspiration for other establishments to continually improve their processes. It also included guidance on how to plan, run, analyse and report an investigation.

Since publication of the first edition, several additional investigations were undertaken and these have been written up as further case studies for the second edition of the "Processor's Guide to Improving Microbiological Quality", which is currently being prepared for publication.

### 3.1.8 Interact with MLA's scientific risk management panel as required

Interaction with MLA's scientific risk management panel was not required.

### 3.1.9 Revision of the "Beef Tool"

In 2005, MLA research resulted in the creation of the "Incoming livestock and slaughter process assessment tool for beef" (the "Beef tool")[1], which aimed to provide QA Managers with information about important processing steps and practices in the hygienic slaughter of cattle (Kiermeier *et al*., 2006). This tool combined the

- Incoming problem (livestock tag score + time in transport) and
- Ability of the process to cope with the problem.

[1] Available at: http://www.mla.com.au/off-farm/Products-and-services/Incoming-livestock-and-slaughter-process-assessment-tool-for-beef-and-sheep

Since 2005, there have been changes in the way the industry operates and hence a revision of this beef tool was appropriate. This revision was developed during a workshop in Adelaide in January 2015, incorporated findings from the investigations undertaken as part of this project and information obtained and observations made during plant visits in November 2014. The revision included the questions/practices in the areas of

- Incoming problem;
- Process' ability to cope; and
- People.

These three "dimensions" of hygienic slaughter and the questions that form part of each, are detailed below.

**Problem posed by incoming livestock**

Questions in this section of the tool try to capture the problem posed by incoming livestock.

- *Do you process veal?*
  Veal, i.e. young cattle, are likely to carry and shed STEC (Cobbold & Desmarchelier 2000, 2002), can have dirty hides and have less fat that can be trimmed. Investigations after the introduction of verification testing for STEC (Meat Notice 2012/3) indicated that manufacturing beef from veal resulted in higher confirmations of STEC than steers and heifers and hence many processors have decided to not process veal. A score of 3 was assigned when veal are being processed (irrespective of proportion) compared with a score of 0 when they are not.
- *What proportion of dairy do you process?*
  Dairy cattle are often harder to process because of their size (cull cows) and the large udders that have to be removed. Investigations after the introduction of verification testing for STEC (Meat Notice 2012/3) indicated that manufacturing beef from dairy cows resulted in higher confirmations of STEC than steers and heifers. A score proportional to the percentage of dairy cattle processed was assigned with the maximum score of 2 when 100% dairy cows are processed.
- *What percentage of cattle are short (less than 8 hours), medium (8-18 hours) and long haul (longer than 18 hours)?*
  Shorter haulage was considered to be better due to shorter time off feed, less opportunity for hide soiling and cross-contamination of cattle with faeces and STEC (Callaway et al. 2013). For the three haulage durations a score proportional to the corresponding percentage of cattle transported was calculated – the maximum scores for these three durations were 1, 2, and 3, respectively.
- *What proportion of cattle comes from feedlots?*
  Cattle from feedlots were found to be more likely to be contaminated with *E. coli* O157 (Dewell et al 2008). These animals are also often considerably dirtier, even after pre-slaughter washing, than cattle not from feedlots. A score proportional to the percentage of feedlot cattle processed was

calculated, with a maximum score of 3 when 100% of cattle processed come from feedlots.

The scores for these questions were summed to give a **problem score** – the higher the score the greater the incoming problem of faecal contamination of hides and likelihood of pathogenic *E. coli* on the hides or in the gut of the animals.

**Effectiveness of the process**

Questions in this section of the tool try to capture the design of the processing operation and the potential for processing to be undertaken hygienically.

- *Are hide-on operations separated effectively from hide-off?*
  Physical separation of hide-on and hide-off areas assist in controlling the air flow and limits the potential for airborne contamination, which is more common around the hide pulling area (Schmidt *et al*. 2012). A process with physically separation was assigned a score of 0, while a process with no such physical separation was assigned a score of 1.
- *Do you process multispecies? If yes, are the slaughter floors/chains completely separated?*
  These two questions, in combination, have a similar intent to the previous question on physical separation of hide-on and hide-off areas. That is, physical separation from other species, *e.g.* sheep, is good practice and reduces the potential for aerosol contamination of carcases from an adjacent slaughter floor/chain. A score of 1 is assigned if other species are processed without physical separation, and 0 otherwise.
- *Average Chain Speed per hour* and *Average staff numbers on slaughter floor*
  The chain speed and staffing numbers largely relate to the size of an abattoir and they can vary considerably between plants. However, in combination they can be used to assess the relative chain speed (animal per hour divided by number of operators) which can be compared across abattoirs. The faster the relative chain speed the less time operators have to perform their operations hygienically. The relative chain speed was multiplied by a penalty factor of 3 to give the score.
- *Do you use a hide decontamination treatment?*
  Hide decontamination treatments have been shown to reduce microbial counts on cattle hides (*e.g.* MLA 2014), and this can relate to microbial counts on the carcase (Elder *et al*. 2000; Fegan et al. 2004; Yang et al. 2015). Consequently, using a hide decontamination step scores 0, while not using one scores 1.
- *Do you rod the oesophagus vertically or horizontally?*
  Rodding vertically, while the animals is still on the landing bed, is preferable as there is less opportunity for spillage. Consequently, rodding vertically scores 0 while horizontal rodding scores 1.
- *Do you bag (& tie) the bung?*
  Bagging and tying the bung substantially reduces the potential for faecal leakage and is therefore scored as 0, while not bagging and tying the bung scores 1.

- *Is the tail removed fully prior to hide removal?*
  Tail flick has been noticed at many Australian abattoirs. Even those that try to control tail flick at the hide puller find it difficult unless the majority of the tail is removed first (not just the bush). Therefore, removal of the tail scores 0 while partial removal or no removal score 1.
- *Do you use a carcase decontamination process?*
  Carcase decontamination processes, such as hot water washing, have been shown to be effective at reducing microbial carcase contamination, especially *E. coli* (see MLA Processor guide 2, in preparation). Because such a decontamination step affects all prior processing steps a multiplicative factor of 0.5 is used when a decontamination step is in place (i.e. reduces the process score by half), while a multiplier of 1 is used when there is not (i.e. has no effect on the process score).
- *Do you process feedlot cattle? If yes, are they always processed at the end of the day?*
  Feedlot cattle are generally much dirtier and hence processing them at the end of the day reduces the potential for cross contamination of workers and equipment, and hence carcases. For this reason, processing feedlot cattle last scores a 0, and 1 otherwise.
- *Do you deep trim along all cutting lines?*
  Cutting lines are potentially more likely to be contaminated at hide opening than other areas. Deep trimming removes these areas and any microbial contamination transferred onto these areas from the hide via knives. Consequently, a score of 0 is allocated for deep trimming, and a score of 1 otherwise.
- *Do you clean and sanitise your boning room at every major work break?*
  While this question relates to the boning room rather than the slaughter operations, it is one practice that can reduce the opportunity for cross-contamination in the boning room throughout the day. A score of 0 is given when this practice is used, and 1 when no or only dry cleaning between breaks is used.

With the exception of carcase decontamination and boning room sanitising, the scores are added up and then multiplied by the carcase decontamination score (0.5 or 1). Finally the score for boning room sanitation is added to give the **process score**.

**People**

Questions in this section of the tool try to capture the contribution of staff to being able to control the problem posed by incoming livestock through good implementation of the process. This dimension has been added to the tool in recognition of the need for skilled staff to achieve good implementation of the process, *i.e.* even a well-designed process can result in high levels of carcase contamination if operators are not sufficiently skilled in carrying out their process steps.

- *What is your annual percentage turnover of operators?*
  High turnover of slaughter personnel was considered “detrimental” to good

processing as there is less knowledge retention and fewer highly skilled staff (Jenson *et al*. 2014). A score was calculated by multiplying the maximum score of 3 by the percentage of staff turnover.

- *What proportion of operators have Certificate II or higher?*
  Hygienic slaughter and dressing has been recognised as requiring a skilled workforce (Jenson *et al*. 2014) and hence a greater proportion of staff with a Certificate II or higher in meat processing was considered good practice. Hence a score was calculated from the percentage of untrained staff (i.e. the complement of trained staff) on a proportional basis, with a maximum score of 2 being achieved when 0% of slaughter staff were trained to Cert. II or higher.

The scores for these questions were summed to give a **people score** – the higher the score the lower the skills basis and retention of staff, and hence the greater the potential for poor implementation of the process.

The problem, process and people scores were added to give a **total score** – the greater the total score the greater the potential for microbial contamination of the carcase.

The answers and scores for the eight plants visited and interviewed are provided in Table 1, along with microbiological summaries (January 2013 and August 2014).

**Table 1: Answers and scores for proposed beef tool questions for plants visited**

| | Plant 1 | | Plant 2 | | Plant 3 | | Plant 4 | | Plant 5 | | Plant 6 | | Plant 7 | | Plant 8 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Ans. | Score | Ans. | Score | Ans. | Score | Ans. | Score | Ans. | Score | Ans. | Score | Ans. | Score | Ans. | Score |
| **Problem posed by incoming livestock** | | | | | | | | | | | | | | | | |
| Do you process veal? | No | 0 | No | 0 | No | 0 | No | 0 | Yes | 3 | No | 0 | Yes | 3 | No | 0 |
| What proportion of dairy do you process? | 20% | 0.4 | 10% | 0.2 | 75% | 1.5 | 80% | 1.6 | 80% | 1.6 | 10% | 0.2 | 10% | 0.2 | 10% | 0.2 |
| What percentage of cattle are short haul (less than 8 hours)? | 60% | 0.6 | 80% | 0.8 | 80% | 0.8 | 95% | 0.95 | 80% | 0.8 | 50% | 0.5 | 80% | 0.8 | 80% | 0.8 |
| What percentage of cattle are medium haul (8-18 hours)? | 28% | 0.56 | 20% | 0.4 | 20% | 0.4 | 5% | 0.1 | 20% | 0.4 | 50% | 1 | 20% | 0.4 | 20% | 0.4 |
| What percentage of cattle are long haul (longer than 18 hours)? | 12% | 0.36 | 0% | 0 | 0% | 0 | 0% | 0 | 0% | 0 | 0% | 0 | 0% | 0 | 0% | 0 |
| What proportion of cattle come from feedlots? | 30% | 0.9 | 0% | 0 | 10% | 0.3 | 1% | 0.03 | 20% | 0.6 | 10% | 0.3 | 10% | 0.3 | 5% | 0.15 |
| **Problem Score** | | **2.8** | | **1.4** | | **3.0** | | **2.7** | | **6.4** | | **2.0** | | **4.7** | | **1.6** |
| **Effectiveness of process** | | | | | | | | | | | | | | | | |
| Are hide-on operations separated effectively from hide-off? | No | 1 | No | 1 | No | 1 | No | 1 | No | 1 | No | 1 | No | 1 | No | 1 |
| Do you process multispecies? | No | | No | | No | | No | | Yes | | No | | No | | No | |
| If yes, are the slaughter floors completely separated? | Yes | 0 | Yes | 0 | Yes | 0 | Yes | 0 | No | 1 | Yes | 0 | Yes | 0 | Yes | 0 |
| Average Chain Speed per hour | 70 | | 75 | | 20 | | 22 | | 70 | | 190 | | 90 | | 92 | |
| Average staff numbers on SF | 46 | 4.6 | 70 | 3.2 | 28 | 2.1 | 15 | 4.4 | 49 | 4.3 | 100 | 5.7 | 90 | 3.0 | 60 | 4.6 |
| Do you use a hide decontamination treatment? | No | 1 | No | 1 | No | 1 | No | 1 | Yes | 0 | No | 1 | No | 1 | No | 1 |
| Do you rod the oesophagus vertically or horizontally? | Hor. | 0 | Hor. | 0 | Hor. | 0 | Hor. | 0 | Hor. | 0 | Vert | 1 | Vert. | 1 | Vert. | 1 |
| Do you bag (& tie) the bung? | Yes | 0 | Yes | 0 | Yes | 0 | Yes | 0 | Yes | 0 | Yes | 0 | Yes | 0 | Yes | 0 |
| Is the tail removed fully prior to hide removal? | No | 1 | No | 1 | No | 1 | No | 1 | No | 1 | No | 1 | No | 1 | No | 1 |
| Do you use a carcase decontamination process? | No | 1 | No | 1 | No | 1 | No | 1 | Yes | 0.5 | No | 1 | No | 1 | No | 1 |
| Do you process feedlot cattle? | Yes | | No | | Yes | | Yes | | Yes | | Yes | | Yes | | Yes | |
| If yes, are they always processed at the end of the day? | Yes | 0 | | | Yes | 0 | Yes | 0 | Yes | 0 | Yes | 0 | No | 1 | Yes | 0 |

| | Plant 1 | | Plant 2 | | Plant 3 | | Plant 4 | | Plant 5 | | Plant 6 | | Plant 7 | | Plant 8 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Ans. | Score | Ans. | Score | Ans. | Score | Ans. | Score | Ans. | Score | Ans. | Score | Ans. | Score | Ans. | Score |
| Do you deep trim along all cutting lines? | No | 1 | No | 1 | No | 1 | No | 1 | No | 1 | No | 1 | No | 1 | No | 1 |
| Do you clean and sanitise your boning room at every major work break? | No | 1 | No | 1 | No | 1 | No | 1 | No | 1 | No | 1 | No | 1 | No | 1 |
| **Process Score** | | **9.6** | | **8.2** | | **7.1** | | **9.4** | | **5.1** | | **11.7** | | **10.0** | | **10.6** |
| **People** | | | | | | | | | | | | | | | | |
| What is your annual turnover % of operators? | 10% | 0.3 | 40% | 1.2 | 20% | 0.6 | 10% | 0.3 | 100% | 3 | 10% | 0.3 | 40% | 1.2 | 10% | 0.3 |
| What proportion of operators have Certificate II or higher? | 10% | 1.8 | 10% | 1.8 | 10% | 1.8 | 10% | 1.8 | 10% | 1.8 | 10% | 1.8 | 10% | 1.8 | 10% | 1.8 |
| **People Score** | | **2.1** | | **3** | | **2.4** | | **2.1** | | **4.8** | | **2.1** | | **3** | | **2.1** |
| **Total Score** | | **14.5** | | **12.6** | | **12.5** | | **14.2** | | **16.3** | | **15.8** | | **17.7** | | **14.3** |
| **Microbial Indicators** | | | | | | | | | | | | | | | | |
| Mean log TVC | | 0.9 | | 0.1 | | 0.7 | | 1.6 | | 1.2 | | -0.3 | | 1.3 | | 0.9 |
| Adjusted *E. coli* Prevalence (%) | | 2 | | 2.7 | | 2.1 | | 4.2 | | 3.2 | | 3.6 | | 7.3 | | 5.4 |
| Adjusted Coliform Prevalence (%) | | 25 | | 6 | | 13 | | 9 | | 6 | | 7 | | 16 | | 11 |
| STEC Confirmations (%) | | 1.3 | | 0.3 | | 0.3 | | 0.4 | | 0.6 | | 0 | | 2.2 | | 2.5 |

The relationship between the revised beef tool total score and the microbiological performance was investigated with the scatter plots shown in Figure 1 – for hot swabbing/boning plants the coliform and *E. coli* prevalence were divided by 3 –yielding adjusted prevalence estimates – to account for the greater likelihood of detecting coliforms and *E. coli* from hot carcases.[2]

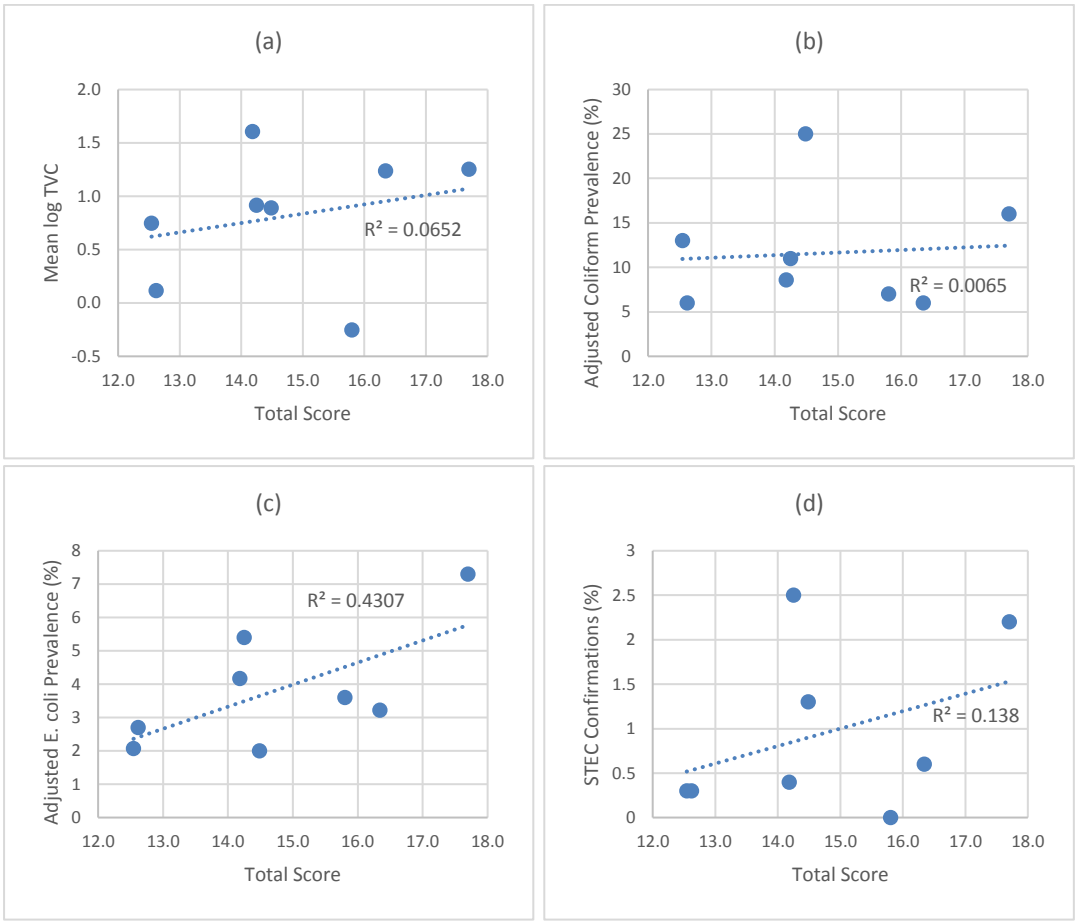


**Figure 1: Scatter plots of microbiological performance versus total score – (a) mean log TVC, (b) adjusted coliform prevalence, (c) adjusted *E. coli* prevalence, (d) STEC prevalence.**

From these plots it can be seen that the strongest relationship exists between adjusted *E. coli* and Total Score, indicating that plants with higher total scores have a tendency to also have higher adjusted *E. coli* prevalence. However, there remains considerable "residual" variability that has not, and possibly cannot, be explained by this tool. Reasons for not finding better relationships may include:

- Too much variation in the system (problem, processing, people), especially on a day-to-day basis, which cannot be captured through "long term" answers and long-term hygiene performance;
- Important variables are missing or unimportant variables have been included and add noise;

[2] This factor was calculated from plants for which investigations on hot and cold carcases had been undertaken, e.g. pasteuriser investigation detailed in the second edition of the MLA Processor guide.

- The weights for the different variables are too small or too large;
- The way the scores have been combined (by summing) for the variables and dimensions (Problem, Process, People) is not correct – interrelationships may require different mathematical approaches.

Given the small number of plants and large number of variables, and potential variables that have not been included, it is impossible to determine which of the four reasons, or combination of reasons, is the case. Feedback on the revised beef tool has been, and is continuing to be, sought from QA Managers at national MINTRAC MI&QA Manager Networking meetings.

Despite the foregoing, the revised tool can still be useful to the industry. In particular, QA Managers can use it to critically evaluate their process and how it is coping with the problems faced on a daily basis.

Work is continuing outside of this project to finalise the revision of the tool.

## 3.2 Objective 2: Propose tests and testing systems

> *Propose tests and testing systems (frequency, microbiological criteria, and statistical methods) that can alert an establishment or national bodies to a significantly increased risk of the detection of pathogenic* E. coli*.*

From the current and previous work it is evident that the majority of Australian processors are achieving low levels of microbial contamination and process performance indicators as measured by existing systems (ESAM, MHA, etc). This is also evidenced by the low frequency with which pathogenic *E. coli* are detected in Australian lots of manufacturing beef destined for grinding in the US (Table 2). While jumps in detection rates were observed in 2008 and 2012, due to changes in testing to N-60 and modification of the enrichment period, the overall rates are very low. This indicates that for the most part Australian processors are managing *E. coli* O157 contamination of carcases well.

**Table 2: Summary of detections of *E. coli* O157 in lots of Australian manufacturing beef destined for grinding in the United States.**

| *Year* | *Number of tests* | *Number (%) of lots with* E. coli *O157 confirmed* |
|---|---|---|
| 1998-2002 | 184,843 | 32 (0.017%) |
| 2005 | 24,029 | 4 (0.016%) |
| 2006 & 2007 | 45,000 | 8 (0.017%) |
| 2008 | 30,647 | 36 (0.117%) |
| 2009 | 34,433 | 35 (0.106%) |
| 2010 | 31,615 | 21 (0.066%) |
| 2011 | Not known | Not known |
| 2012 | 21,791 | 48 (0.22%) |
| 2013 | 18,234 | 39 (0.21%) |

In addition, from the results of a recent risk assessment, it was concluded that the risk of *E. coli* O157 illness from consumption of Australian beef burgers in the US is low (Kiermeier et al. 2015). Furthermore, increases in the stringency of sampling manufacturing beef, over and above current N-60 sampling and testing, have been shown to "provide marginal additional public health benefit" (Kiermeier et al. accepted).

Nevertheless, it would be useful to processors to be able to predict increased risk of detecting *E. coli* O157 or STEC contamination in lots of manufacturing beef, as this would allow them to manage the issue proactively. However, from the work undertaken in this project, and from previous MLA funded work, it has become evident that current test systems do not work for assessing increases in risk of detecting pathogenic *E. coli*. This is due to the following reasons.

1. Results from carcase testing generally result in low detection of *E. coli*. This is due to several factors:
    a. Carcase hygiene is generally "good" (as assessed at present) and even when *E. coli* are detected, the counts are usually low.
    b. Carcases are usually sampled after chilling, which results in lower levels of *E. coli*.[3] Recent research by the University of Tasmania indicates that this effect may not be related to *E. coli* reductions, but due to our inability to culture them effectively after overnight chilling.
    c. The areas that are sampled as part of ESAM are "least likely to be contaminated", *i.e.* not the best sentinel sites.
    d. Each of the three ESAM areas sampled is relatively small (100 $cm^2$).
    e. There is considerable variability between QA staff in terms of how well they swab and are able to remove microbial contamination from the carcase (Saeger *et al*. 2010).
    f. Few carcases are sampled per day, yet there often is large variability between incoming animals (origin, transport distance & duration, species, dirtiness, etc).
    g. Presence of data entry / recording errors.
2. STECs are detected very infrequently in cartons because:
    a. Cartons are usually frozen, which makes detection more difficult (reduction in counts).
    b. Twelve cartons, possibly from multiple production days / periods, are composited for STEC testing. In the case of a detection, it is impossible to tell which carton, or cartons, a STEC originated from.
3. Carcase and carton testing is not aligned.
    a. The only relationship between cartons and carcases is that the cartons are generally produced from the carcases that were slaughtered the day before. In the boning room, belts and equipment quickly equilibrate (see Appendix 2: Relationship between carcase and carton data) and hence carton TVC results bear little resemblance to carcase levels.
    b. With respect to STEC testing results, it is even harder to align the sampled cartons with carcases.

[3] For this reason an adjustment factor of 3 has been used in the revised beef tool to relate hot swabbed with cold swabbed carcase.

Consequently, to increase the chances of relating some hygiene indicator with the risk of pathogenic *E. coli* detection, the following need to be considered.

1. A suitable hygiene indicator that is detected more frequently is required. This may involve:
    a. Sampling more contaminated carcase sites, e.g. bung, belly strip, neck/forelegs (to catch evisceration problems & Halal cut) and along the back (to catch tail flick problems).
    b. Sampling hot rather than cold carcases, which would better identify slaughter problems and remove the confounding effect of chilling.
    c. Sampling larger areas will increase the chances of capturing the organism, especially if used in conjunction with an enrichment test (e.g. for *E. coli*) to allow detection of very low levels of contamination.
    d. Excision sampling would remove variability between sample collectors, and may, to some degree, reduce the need for sampling larger areas.
    e. Testing for a different indicator may be preferable to testing for generic *E. coli*, e.g. coliforms or Enterobacteriaceae. However, it should be noted that even coliforms are possibly not detected frequently enough under current systems (see Table 1) at most plants to be useful for process control monitoring.
2. Better linkage between carcase and cartons is needed to allow for relationships to be detected (provided they exist). Otherwise, the natural variability in the system will overshadow any signal that might be present (Bollerslav *et al*. 2013).
3. Better identification of extent of STEC contamination is needed to enable better tracing back to animal source. This could be addressed by “pre-enriching” samples from each carton separately and then combining aliquots for the molecular screening test. While this approach probably slows down the testing process, it provides considerable more information. In particular, given the way lots are produced (by combining cartons from multiple production periods based on customer requirements), this information would give establishments more information about when there might have been some loss of process control.

In addition, it will be necessary to continue to monitor ESAM / PHI data and to support QA staff to critically appraise their data, *e.g.* to realise that no detectable TVC for any length of time is unrealistic, and that such an outcome is likely the result of an unsuitable microbiological test (*i.e.* wrong dilutions).

# 4 Conclusions and Recommendations

During the course of this project it became apparent that the data that are currently collected often are not sufficient to detect loss of process control and an increased risk of pathogenic *E. coli* detections. There appears to be no relation between the indicator organisms that are regularly collected for regulatory purposes and *E. coli* O157 / STEC detections. As such, it is recommended to investigate alternative methods or indicators, such as coliforms or Enterobacteriaceae, which could alert establishments to a loss of process control and an increased risk of STEC detections, including sampling larger carcase areas, sampling hot carcases rather than chilled carcases, and sampling sites that are more likely to be contaminated than current ESAM sites.

Many plants have developed in-house systems for monitoring their microbiological results and process control and use these data to inform their slaughter floor staff and supervisors. However, there is still scope for establishments to undertake more process investigations, to

gain better understanding of where contamination is added to and removed from the carcase. From plant visits and communications with QA staff it is apparent that many new staff have entered a QA role over the last two years. Consequently, the need for delivering another series of investigation workshops should be appraised.

Through this project, QA managers now have access to the "Processor's Guide to Improving Microbiological Quality" (MLA, 2014), with a second edition in preparation, which they can use to investigate their process for areas of possible improvement. In addition, the industry has benefitted from the statistical and data analysis support received through this project and it is recommended that the provision of such a service is continued in the future, especially to supplement the investigation workshops.

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# 6 Appendix 1: Process control interviews

## 6.1 Background

Australian manufacturing beef destined for grinding in the US is required to be tested for *E. coli* O157 using robust N-60 testing since 2007. More recently, the US Food Safety and Inspection Service (FSIS) has focused on an additional six shiga toxin producing serotypes of *E. coli* (O26, O45, O103, O111, O121, O145), commonly referred to as STEC. These have now also been declared adulterants and the Australian industry has been undertaking testing of manufacturing beef for all seven *E. coli* serotypes since mid 2012. There has been a higher detection rate than anticipated.

Meat and Livestock Australia (MLA) has funded the South Australian Research and Development Institute (SARDI) to investigate potential risk factors and interrogate national microbiological databases (ESAM and Product Hygiene Index) for indications of loss of process control, that is, the ability of plants to control microbial hazards. From this work, it has become apparent that the large amounts of data that are collected in the ESAM and PHI databases are of limited value for identifying increases in overall probability of detection of *E. coli* O157 (and STEC). This is likely due to a range of reasons, including:

1. Generic *E. coli* and other hygiene indicators (TVC and coliforms) on carcases and in cartons are generally low (prevalence and concentration);
2. *E. coli* O157 and STEC are infrequently detected in cartons of beef trim;
3. Process and Product hygiene monitoring results, e.g. Meat Hygiene Assessment (MHA), are generally good;
4. There is no 1-1 relationship between process / carcase monitoring and *E. coli* O157 / STEC testing;
5. Data errors and reliability issues in ESAM and PHI data.

In addition, processing of cattle is usually performed well in Australia and plants generally meet performance objectives, including:

- Ante mortem inspection of cattle;
- Processing systems approved and audited by the Department of Agriculture and third party auditors;
- Pre-operation inspections and hygiene testing performed daily prior to production;
- Meat Hygiene Assessments (MHA) performed;
- Chilling of carcases that meets the Refrigeration Index (RI), where chilling is used;
- Microbiological testing undertaken to verify process control, including ESAM, carton testing involving TVC and *E. coli*, and US export certification testing involving *E. coli* O157 / STEC;
- Undertaking investigations when problems arise or are identified and perform corrective actions.

Furthermore, in the US, pathogenic *E. coli* in beef products continue to result in recalls and foodborne illness outbreaks, despite current control measures, e.g. multiple interventions, and testing requirements. Consequently, internationally and in Australia, focus is being placed on improving process control and better risk management.

Work to date by the South Australian Research and Development Institute (SARDI) has focussed on linking slaughter hygiene and microbiological performance with hazard

detections, but no clear relationships have been established. This indicates that microbiological monitoring of the process alone has limited utility. As such, there remains a need to identify factors that may be contributing to the increased risk of contamination by microbial hazards. Hence, a broader approach is required, taking into consideration other factors that may play a role, including livestock, processing, personnel, training, 'food safety culture', housekeeping, etc.

The purpose of this project was to undertake a qualitative assessment of plants that have, and have not, had high levels of O157 / STEC detections, and identify factors, or combinations of factors, that are hypothesised to affect O157 / STEC contamination. These can later be assessed quantitatively as causal relationships cannot be established from simple observational studies, i.e. they require experimental investigations.

It should be noted that the results presented in this report have been kept general to reduce the risk of plants being identified.

## 6.2 Methods

### 6.2.1 Selection of plants

Plants were selected by the project team so that they would be broadly representative of different geographic regions, size of operation, hot / cold boning and the frequency of *E. coli* O157 and STEC detections. The characteristics of the plants selected are as follows:

- Geographic location: QLD (×2), NSW (×3), VIC & TAS (×3)
- Size of Operation: <200 to 1,500+ animals per day
- Hot / cold boning: 3 hot boning, 5 cold boning (1 sometimes bones warm)
- Percentage of tests where *E. coli* O157 and/or STEC[4] were detected between January 2013 and August 2014 (see also
- Table 4):
    - Low percentage confirmed: Five plants had less than or equal to 1% of tests where STEC were confirmed;
    - High percentage confirmed: Three plants had more than 1% of tests where STEC were confirmed.

### 6.2.2 Plant visits and interviews

The eight plants were subsequently visited during November 2014 and QA managers were asked questions about their operations. In addition, the slaughter floor, boning room and yard areas were visited and operations observed. Some questions were answered by the project team through observation, which were confirmed with the QA Manager where necessary. The questions were broadly categorised as:

- The livestock – questions about the cattle being processed
- Process – questions about the slaughter operations
- People – questions about the staff and training

[4] The *E. coli* serotype O103 was excluded from these calculations as there is currently uncertainty about the validity of test results where *E. coli* O103 was detected.

- Plant & operation – questions about the operation
- Microbiology – questions about microbiological testing and investigations

The questions had been developed prior to the visits by members of the team, based on their experience and work in the industry.

### 6.2.3 Evaluation of results

After each visit, project members discussed the visit, their observations of the plant, and the answers obtained. After all visits had been completed the answers to the questions were collated. We acknowledge that a brief plant visit cannot provide the in-depth knowledge of the operation that a QA Manager has and the responses have to be taken at face value, and these are supplemented with our own observations and knowledge.

Because of the small number of plants and large number of questions it is not possible to undertake a statistical assessment of the responses. Instead a qualitative assessment was undertaken with the aim to identify areas which may contribute to increased risk of carcase contamination.

## 6.3 Results

### 6.3.1 ESAM and *E.* coli O157 / STEC testing results

Summaries of ESAM and *E. coli* O157 / STEC test results, by plant and species, are provided in Table 3 and

Table 4. Of note are the relatively high percentage of *E. coli* detections for plants C, D and E, for Cow / Bull which is likely related to the fact that these plants primarily hot swab/bone. However, the fact that these plant hot swab/bone does not appear to translate into a higher percentage of STEC confirmations as can be seen from

Table 4.

**Table 3: Summary of ESAM results by plant and species (January 2013 and August 2014).**

| Plant | Species | Percentage of kill (%) | Mean $log_{10}$ TVC/$cm^2$ | *E. coli* (%) |
|---|---|---|---|---|
| **A** | Cow/bull | 18 | -0.12 | 6.3 |
| **A** | Steer/heifer | 82 | -0.28 | 2.4 |
| **B** | Cow/bull | 49 | 0.24 | 2.5 |
| **B** | Steer/heifer | 51 | 0.05 | 2.9 |
| **C** | Cow/bull | 100 | 0.76 | 14.1 |
| **D** | Cow/bull | 100 | 1.61 | 16.7 |
| **E** | Cow/bull | 85 | 1.29 | 16.6 |
| **E** | Steer/heifer | 15 | 1.03 | 7.1 |
| **F** | Cow/bull | 18 | 0.72 | 4.9 |
| **F** | Steer/heifer | 82 | 0.96 | 1.3 |
| **G** | Cow/bull | 13 | 1.16 | 3.7 |
| **G** | Steer/heifer | 87 | 1.32 | 8.4 |
| **H** | Cow/bull | 36 | 0.90 | 6.0 |
| **H** | Steer/heifer | 64 | 0.92 | 5.6 |

**Table 4: Summary of *E. coli* O157 and STEC testing results (January 2013 and August 2014).**

| Plant | Number of test | Number of Potentials | Percentage Potentials | Number of Confirmed* | Percentage Confirmed | Percentage of Potentials confirmed |
|---|---|---|---|---|---|---|
| **A** | 3711 | 0 | N/A | 0 | N/A | N/A |
| **B** | 996 | 5 | 0.5% | 3 | 0.3% | 60.0% |
| **C** | 967 | 8 | 0.8% | 3 | 0.3% | 37.5% |
| **D** | 224 | 3 | 1.3% | 1 | 0.4% | 33.3% |
| **E** | 4951 | 108 | 2.2% | 30 | 0.6% | 27.8% |
| **F** | 558 | 34 | 6.1% | 7 | 1.3% | 20.6% |
| **G** | 495 | 41 | 8.3% | 11 | 2.2% | 26.8% |
| **H** | 276 | 22 | 8.0% | 7 | 2.5% | 31.8% |

* Excluding *E. coli* O103, which has been excluded due to uncertainty in the validity of the test results.

### 6.3.2 Livestock

Plants generally processed a range of animals from prime cattle to cull animals – some primarily slaughtering cull cattle (cows and bulls) while others mainly processed steers and heifers. Only Plant G indicated that they processed any veal, but it is not known how many of the STEC confirmations were related to veal.

All plants obtained the majority of cattle slaughtered via direct consignment, although the percentage of cattle from saleyards varied between about 20 to 40% according to the QA Managers. However, there was a tendency for this percentage to vary more for plants with a higher proportion of STEC confirmations.

Six plants processed some portion of feedlot cattle and there was a tendency for plants with lower STEC confirmation percentages to process fewer feedlot cattle and to be located in dryer areas. Nevertheless, at Plant A feedlot cattle were processed more commonly during 3 months of the year to ensure continued processing even though for the remaining time

grass-fed cattle were the norm. In general, QA Managers reported that feedlot cattle per se were not processed differently, but that they were usually dirtier and hence may receive extra soaking and/or washing (including hosing on the landing bed after stunning), or additional trimmers were placed on the line.

None of the QA Managers indicated that they had specific knowledge of feed withholding practices that had been put in place prior to transport, and neither travel distance / duration nor 'stress' was taken into account during slaughter. Travel duration varied considerably between plants and also throughout the year based on where cattle were sourced. One plant encouraged transporting cattle empty, while another had noted problems with burst paunches as a result of overfeeding in their own yards. Two plants with low STEC confirmation percentages routinely rest animals before slaughter. One of these plants instituted this practice earlier in 2014 in response to a high incidence of dark cutting. This plant considered the practice to be beneficial and reported that the incidence of dark cutting had reduced and that they have not failed an *E. coli* window since.

QA Managers reported that the usual practice was to present clean cattle for slaughter and that all cattle were thus routinely washed. The extent of washing however varied between plants as did cattle dirtiness (as noted above, feedlot cattle tended to be dirtier). Most plants used automatic overhead and/or under belly wash systems, with manual hosing for dirtier cattle or dirtier areas. Only one plant washed cattle with recycled water, followed by a potable water rinse – all other plants used potable water and several added additional chlorine. Several plants reported switching to potable water for all washing after experiencing more frequent microbiological problems, i.e. *E. coli* detections as part of ESAM, when they had previously used recycled water (microbiological testing was generally not done on the recycled water supply). One QA Manager also noted that cattle were sequenced based on cleanliness, with dirtier cattle scheduled for the end of the day, to allow more time for washing and to reduce the potential for (visible) cross contamination.

### 6.3.3 Process

None of the plants visited had physical separated hide-on and hide-off areas and slaughter floors were predominantly 'serpentine' in layout. None of the plants visited processed other species, e.g. sheep, on the same slaughter floor.

All plants had been designed, especially in terms of rail height, for the majority of stock that they process. However, most also reported that a small proportion of stock, i.e. large animals, touched the ground. At two plants – one plant with low and one with high STEC confirmation percentage – it was evident that large animals posed a potential problem at the viscera table, with the forelegs and neck either sitting directly on the viscera table or hanging over the side.

Several questions related to the *effective* chain speed, including number of animals slaughtered per hour and number of operators at key positions, such as hide opening cuts, bunging, hide removal and evisceration. Irrespective of the *absolute* chain speed (i.e. animals processed per hour), which largely depends on the size of the plant, operators at all plants were able to undertake their operations without noticeably falling behind while being observed by the project team. Falling behind can happen when operators make a greater effort to follow work instructions while being watched and subsequently struggle because the

‘correct’ operation takes longer than how they normally work. In addition, about half the plants indicated that the chain speed remained constant, while the other half indicated that the speed was varied according to visual defects on the carcase / MHA feedback or proactively for dirtier or larger cattle.

While all plants indicated that air flow was from the cleaner (hide-off) to the dirtier (hide-on) end, only one plant had undertaken any form of checking, a once-off check using paper towel/tissue. During the plant visits, it was apparent that even if the air flow was primarily from clean to dirty, the installation of fans or large air conditioning units could result in different airflow patterns. None of the air intakes were filtered to avoid external airborne contamination and one plant used fly screens on open windows to provide some airflow on the slaughter floor. Air conditioning systems appeared more common in southern plants compared with the hotter and more humid northern plants. In addition, all plants pulled hides downward and no additional airflow controls were present at this station.

The landing area was cleaned between animals at four plants, automatically at two and manually at the other two. At four plants the landing area was also used for an additional cleaning step – at one plant, the anus, rump and hind leg area received additional hosing with potable water while at the other plants, either “Clean Oxide” or “Twin Oxide” were applied to opening cuts or due to be implemented (one plant). Plants using chemical interventions had indicated that they had undertaken microbiological trials to validate the efficacy of the intervention prior to implementation. It is likely that these measures are a response to the STEC detections at these plants.

All but three plants processed Halal and this did not vary throughout the year. Those plants who did process Halal also trimmed the area of the Halal cut after hide removal and at least one plant exported these neck trimmings. There was no apparent relationship between Halal processing and STEC confirmation percentage.

All plants rodded and clipped the oesophagus, although the direction – vertically or horizontally – differed according to the plant set up. Spear cuts were used for hide opening and the bung was bagged and tied at all plants, though only one plant used an automatic bung spear.

Tail flick at the hide removal station had been noticed at most of the plants visited. Consequently, most remove at least the brush. At one plant, most of the tail is removed routinely while at another this is done only in the case of dags on the tail. At other plants operators generally attempted to control the tail manually, but often they had limited success to prevent the flick of the hide when the tail was cleared.

Trimming was fairly consistent across plants with all performing a standard hygiene trim. Some added extra trimmers to trim extra fat and at least two added additional trimmers in response to dirty cattle or increased occurrence of visible defects.

With respect to carcase interventions, one plant was planning on installing a hot water cabinet, while another had installed a full carcase hot water invention. They reported having

validated its efficacy and that *E. coli* detections (ESAM) had become less frequent although TVC levels had increased. The QA Manager indicated that efficacy of the intervention was checked periodically. An analysis of this plant's ESAM data[5] showed that *E. coli* detections after implementation of the hot water intervention were almost half of those prior to the implementation – this reduction was statistically significant (P-value < 0.001) and practically important. Over the same period of time there was a marginal increase in TVC of 0.19 $\log_{10}$ cfu/cm$^2$ (P-value < 0.001).

Lastly, only two plants (A and H) indicated that they had an automatic full body carcase rinse (not hot water decontamination).

### 6.3.4 People

Most plants paid their slaughter floor and boning room staff on an hourly basis, although one plant also provided a bonus based on the number of animals processed. The only plant that paid workers by the animal also had a fixed line speed and thus achieved a comparable system to plants that paid an hourly wage.

The employment numbers varied considerably between plants, from less than 100 to more than 1000 across the whole plant, and consequently the staffing levels on the slaughter floor and boning room also varied between plants. However, plants also dealt with varying staffing levels throughout the day in different ways. Some noted adding extra trimmers or slowing down the chain in response to dirtier cattle or when increased hygiene problems were detected. These approaches tended to be more established at plants with lower STEC confirmation percentage.

With respect to slaughter floor staff turnover, only two plants had a low turnover (<7% per year), although most QA managers noted the largely stable workforce. These plants also reported either not employing seasonal staff or backpackers (417 VISA holders) or, when they did employ such workers, then only on non-critical jobs, such as packing or processing hides. Only one plant (with low STEC confirmation percentage) reported having a primarily transient workforce (approx. 80%) consisting of seasonal staff from overseas, and subsequent slaughter floor turnover in excess of 100%.

All QA Managers reported that all new staff are formally inducted and that the induction covered company policies / HR, Workplace Health and Safety, and Hygiene and Sanitation. The duration of this induction varied from ½ day to 5 days. The longer induction processes included some structured on-line training with a buddy according to the position on the slaughter line for the new operator.

A buddy system is used at all plants to train new workers on the slaughter line. Once the worker is deemed competent their competency is assessed by the supervisor and possibly an on-site trainer / tutor. At most plants, training was performed on the shift that the new staff member continued to work on although at one plant (with high STEC confirmation percentage), training was done on night shift when the chain speed is slower.

[5] The data covered 12 months prior to and 8 months after the implementation of the hot water intervention.

Almost all the plants maintained a skills matrix with competency assessed against work instructions with virtually no assessment of underpinning knowledge. In some cases, this would have been handicapped by language barriers. However, a few had accredited training at Certificate Levels II and III for all their permanent slaughter floor and boning room operators. At one plant (with low STEC confirmation percentage), competency was reassessed every 12 months, including their underpinning knowledge for each task, e.g. potential sources of contamination and cross contamination.

All QA Managers indicated that feedback is given to staff on a regular and ongoing basis, by QA staff or floor supervisors. This was especially so in the case of problems being detected down the line. Feedback usually took the form of a conversation with the supervisor or QA staff and may also involve review of work instructions, retraining or disciplinary action. Some mentioned running daily or weekly review meetings with supervisors about slaughter floor and boning room hygiene (i.e. Zero Tolerance (ZT) defects and MHA) performance. Naturally, feedback usually focussed on “problems” or “negative feedback”, although some plants with lower STEC confirmation percentages indicated that they also tried to provide positive feedback.

### 6.3.5 Plant & operation

Plants reported that 2014 had been very busy, compared with previous years, and some have added a second slaughter and boning shift. Two plants processed ‘only’ five days per week (one with low and one with high percentage of STEC detection), one plant processed six days using a four day roster, while all other plants processed two out of three Saturdays. Three of these plants had no issues finding volunteers for the Saturday shift, although one QA Manager noted that there was also an expectation – staff who had not volunteered were eventually required to work.

Five plants slaughtered during one shift only and of these plants, only one had a second boning shift. The other three plants had two slaughter and boning shifts, and one of these had a high percentage of STEC detections. The QA Manager from this plant indicated that night shift was considered ‘worse’, but did not indicate in what sense night shift was worse. Shift length varied from 7.35 hours to 9.6 hours although the number and duration of breaks were similar (2×30 minutes) – the exception were two plants (both with low STEC confirmation percentages) with shifts in excess of 9 hours, which allowed multiple 5 minutes breaks in addition to the longer breaks.

As indicated earlier, three plants hot boned, and all had low STEC confirmation percentages. All other plants spray chilled overnight and had been doing so for more than 3 years - only one of these plants had changed to spray chilling 12 months ago.

The boning rooms generally consisted of fore- and hindquarter chains with multiple parallel belts for primal, trim and condemned material/bones. Only two plants had a more complex system of belts with pieces of meat being handled by multiple operators. None of the boning rooms were set up to automatically record operators logging on/off, boning performance, etc.

All QA Managers reported that the slaughter floor was fully cleaned and sanitised at the end of each day and some also reported dry cleaning and sanitising contact surfaces during breaks. Boning rooms were also only fully cleaned at the end of each day, with some also dry cleaning during breaks.

### 6.3.6 Microbiology

All plants reported that they tried to minimise the number of different production days that are part of a lot (or port mark), but none restricted lots to a single day or shift. All plants test frozen trim and primals that are destined for grinding in the US for *E. coli* O157 / STEC. Other product may be tested if required by the customer, e.g. fresh trim for McDonalds.

Microbiological screening for *E. coli* O157 / STEC was undertaken by a qualified microbiologist at three plants while the others sent samples to an external laboratory. Two other plants employed microbiologists for non-molecular tests, such as TVC, pre-operation swabs, project work, etc.

All QA Managers reported a lack of confidence in ESAM and PHI due to time lag, irregular reporting and general data quality. Consequently, all used either iLeader or an Excel based system to monitor their microbiological performance on a more real-time basis.

Different plants undertake different additional microbiological testing, such as for shelf-life or to meet specific customer requirements, though these tend to be less frequent than ESAM. Only two plants (B and G) undertook additional daily sampling as part of their process control monitoring.

All but one of the QA Managers reported having undertaken a microbiological investigation, either before implementing a specific intervention or while trying to understand the source of a STEC detection. Four of the eight QA Managers had also undertaken an investigation to identify carcase sites that were more likely to be contaminated, although none continued to monitor these sites on an ongoing basis – three of the plants had low STEC detection percentages and one plant had high STEC detection percentages.

Those plants that have experienced higher STEC confirmation percentages indicated that they have sought help external to their own plant environment. All QA Managers indicated that they attend Mintrac QA meetings/conferences and used other resources such as the MLA website, Meating Place, and food safety mailing list to stay up to date with food safety information.

## 6.4 Discussion

It is important to note that the various aspects and factors of cattle slaughter operations – pre-transport, transport, slaughter, boning – cannot be considered on an individual basis and hence no one factor would be expected to align with low / high STEC confirmation percentage. Instead the problem is likely to be multifactorial and hence there will likely be confounding factors that obfuscate the conclusions. Hence we can only collate the observations made in this qualitative investigation and hypothesise about which factors might contribute to the risk of STEC contamination. These hypotheses may be tested later in an experimental setting and thus help establish causal relationships.

The following questions were developed by the project team after the plant visits, taking into account the observations made, the responses obtained from plant staff and subject matter knowledge of the project team.

### 6.4.1 Livestock

- What is the relationship between the consignment type, i.e. saleyard versus direct consignment, and STEC shedding or contamination of the hides? If there is a relationship, is it because of stress or are there other factors? Is there a relationship with the rate of cattle that are super-shedders, i.e. cattle that excrete high numbers of STEC per g of faeces?
- What is the relationship between feedlots / non-feedlot, i.e. grass versus grain fed, cattle and STEC contamination?
  Fegan et al. (2004) did not find significant differences between prevalence and concentration of *E. coli* O157 in faeces from grain and grass fed cattle. However, the latter test resulted in a P-value of 0.06, which indicates that there may be some difference but that there was not quite enough evidence (i.e. data on *E. coli* O157 detections) to conclude so. In addition, the relationship between feedlots and STEC in faeces in Australia is unknown. Alternatively, it might be because feedlot cattle tend to be dirtier than grass-fed cattle, especially in the colder and wetter southern / south-eastern areas of Australia (Fegan et al. 2009).
- Does the occurrence of super-shedders differ geographically?
- What is the effect of resting cattle prior to slaughter on STEC concentration in faeces. Does resting affect the chances of super-shedding or the number of animals that super-shed?
- What effect does washing cattle have on the STEC contamination of hides and carcases? Does recycled water pose a greater risk for carcase contamination than potable water?

### 6.4.2 Process

- Does contact between viscera and the carcase, e.g. forelegs and neck as was the case at two plants for very large animals, increase the risk of contaminating these carcase parts with STEC?
- What is the effect of changing the chain speed based on cattle cleanliness and visual feedback on carcase contamination?
- How much do aerosolised STEC contribute to carcase contamination?
- How effective are current practices of controlling / preventing tail flick for preventing carcase contamination?
- How effective are slaughter robots, e.g. bung spear, at preventing carcase contamination?

### 6.4.3 People

It appears that slaughter and boning staff are considered a commodity or ‘disposable resource’ by some plants and as ‘skilled labour’ by others. While members of the project team believe that plants with a stable, well trained workforce and where appropriate feedback (both positive and negative) is provide frequently should perform better, the observations from this investigation cannot provide a clear answer (given the multidimensional nature of the operations).

- What effect does staff turn-over and employment of skilled versus temporary labour have on microbiological contamination of carcases?
- What effect does training (basic buddy system and tutoring versus Certificate II or higher) have on microbial contamination of carcases?

- What effect does positive / negative feedback to staff have on microbiological contamination of carcases?

### 6.4.4 Plant / Operation

- Is night shift 'worse' in terms of carcase contamination than day shift? Is this effect consistent for all plants or is it plant specific? If the latter, why is this so and what are the contributing factors?
- How much do boning room operations and contact materials, including conveyor belts, hands, and gloves, contribute to the cross-contamination of primals and trim with STEC?
- How effective is more frequent cleaning and sanitising at reducing or limiting cross-contamination? Do these effects differ between hot / cold boning?

### 6.4.5 Microbiology

It was encouraging that all plants have developed systems that allow them to monitor their microbiological results in a more 'real-time' manner and that some used additional sampling and testing for monitoring process control. However, the details of these systems are uncertain, especially how trends are assessed. For example, we are aware of at least one instance where the actual test results, rather than the log-transformed values, are plotted and assessed for trends. However, because of the skewed nature of microbiological data – most values will be small while very large values can occur occasionally – trends will be dominated by the occasional high counts.

It was also reassuring that microbiological trials had been undertaken at several plants to evaluate the efficacy of interventions before they were implemented. Again, it is unknown how rigorous these trials were and how thoroughly they had been undertaken.

## 6.5 Conclusions

As part of this project, eight plants were visited and the QA Managers were interviewed. All of these plants have been operating throughout 2014 and many have seen a considerable increase in demand for their products. While all plants basically undertake the same operations, they do so in different ways. In addition, their ESAM results and frequency of STEC confirmations differ.

The differences in livestock, staff and operations between plants may or may not contribute, either directly or indirectly, to the frequency with which carcases and end product are contaminated. In addition, the combination of factors may differ between plants and the difficulty is evaluating their individual and combined effects. If it can be established that a combination of factors reduces carcase contamination, then the questions of consistency and cost efficiency naturally arise. For example, would it be cost effective for a plant to train staff to Certificate III when their workforce is largely transient and of non-English speaking background? Or would multiple whole carcase interventions, e.g. lactic acid and hot water wash, have a greater and more consistent / predictable effect on controlling carcase contamination, and one that it easily verified on a regular basis?

By contrast, multiple interventions are already common place on North American slaughter floors and in the boning rooms, likely because of the faster line speeds and differences in production systems and environments. Despite this, recalls of meat due to STEC

contamination and foodborne illness outbreaks continue to occur, which indicates that these multiple interventions can still be overwhelmed by high levels of STEC in and on the incoming cattle. Therefore, all elements of cattle slaughter and processing will likely have a role to play, e.g. as part of a hurdle system.

## 6.6 Acknowledgement

We would like to thank the staff at the establishments visited for their cooperation and for openly sharing their experiences and approaches to managing the ever increasing food safety and requirements.

# 7 Appendix 2: Relationship between carcase and carton data

It has been proposed that micro results from ESAM carcase swabs bear little relation to the carton samples from the same plant. This analysis considers both beef and sheep results, including veal and lamb.

## 7.1 Is there a relationship between Carcase (ESAM) micro results and Carton micro results?

### 7.1.1 Weekly Medians

Weekly medians of carcase and carton results were calculated and plotted against each other, to determine if there is a relationship on a national level. Weekly medians were used as it was expected that in any given week, most plants should have enough results above the limit of detection to result in a non-infinite (-infinity) median TVC for both carcase and carton results.

When all plants are combined (as in Figure 1), there appears to be a slight positive trend. I.e. if carcase micro is higher, carton micro is generally higher. However, a lot of variability remains around the line of best fit, which simplifies the relationship (and doesn't necessarily fit well).

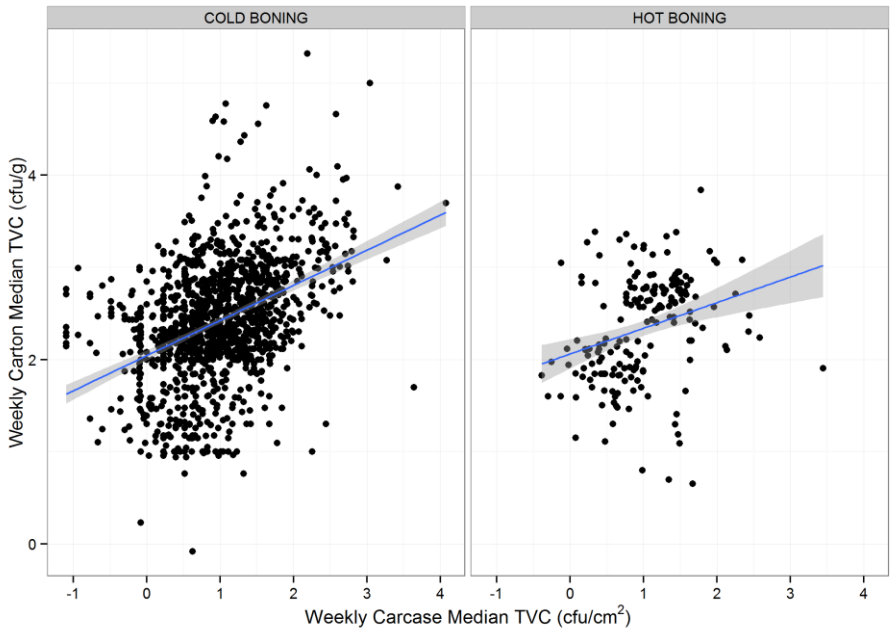


**Figure 1: Weekly median carcase TVC vs. weekly median carton TVC**

On further inspection of individual plants, it appears that most plants do not follow this trend. In fact, most plants do not have a significant relationship between median weekly carcase and carton results. This can be seen in Figure 2, below. Many establishments have a very low range of values for carton testing results (i.e. the vertical spread of carton results is very low. See for example establishment 746). Many plants appear to have very consistent weekly median TVC counts, for both carton and carcase micro.

### 7.1.2 Slope

Figure 3 shows the plants with a significant slope (i.e. there is a statistically significant relationship between carcase and carton micro – coloured blue) along with the magnitude of the slope. In all, there are 6 plants (9761, 8432, 853, 9983, 7627 and 1506) out of 42 with a significant slope, ranging from 0.25 to 0.6 $\log_{10}$ cfu/$cm^2$ /$\log_{10}$ cfu/g (Figure 3 – blue bars). Of these statistically significant results however, none were deemed to be practically important, as most of the slopes were due to one or two extreme results for the given plant.

There is perhaps one other noteworthy plant that can be seen from Figure 2. Plant 1295 appears to have a cluster of points which show very low levels of Carcase TVC, but has carton micro of around 3 log.

### 7.1.3 Daily Medians

Daily medians were then considered, because it was thought that perhaps weekly medians were too insensitive, and were "evening out" the effect of plants that had a high proportion of non-detects in either carcase or carton results. From Figure 4 it can be seen that some plants do have a significant proportion of results below the limit of detection (shown as half circle points on the left or bottom of each cell).

### 7.1.4 Proportion of Non-detects

A proportion of non-detect daily medians for both carcase and carton TVC was calculated to determine which establishments had a significant number of medians below the limit of detection (implying that 50% or more of samples on the given day being below the limit of detection). These establishments were identified by considering the ratio of the proportions of medians above the limit of detection. There were 5 plants which had 50% or more non-detect carton medians than carcase medians (Plants 917, 6828, 8015, 8858 and 10246), shown in Figure 5. These plants were then examined individually, to investigate why they were losing data.

### 7.1.5 Limit of detection

It appears that the labs performing ESAM testing for the 5 plants above were performing too many serial dilutions for the carton samples (and in some cases the carcase samples too – see plant 917 for example). This had the effect of raising the limit of detection, and resulting in a large number of tests being below this limit of detection. This can be seen in Figure 6. Some plants have a high limit of detection for both carcase and carton samples, while for others only carton results appear to be affected.

### 7.1.6 Conclusions

Carton results tend to have little relation to carcase results. Boning rooms appear to have the effect of evenly distributing the contamination, and resulting in reasonably consistent carton results regardless of the carcase results on a given day or week.

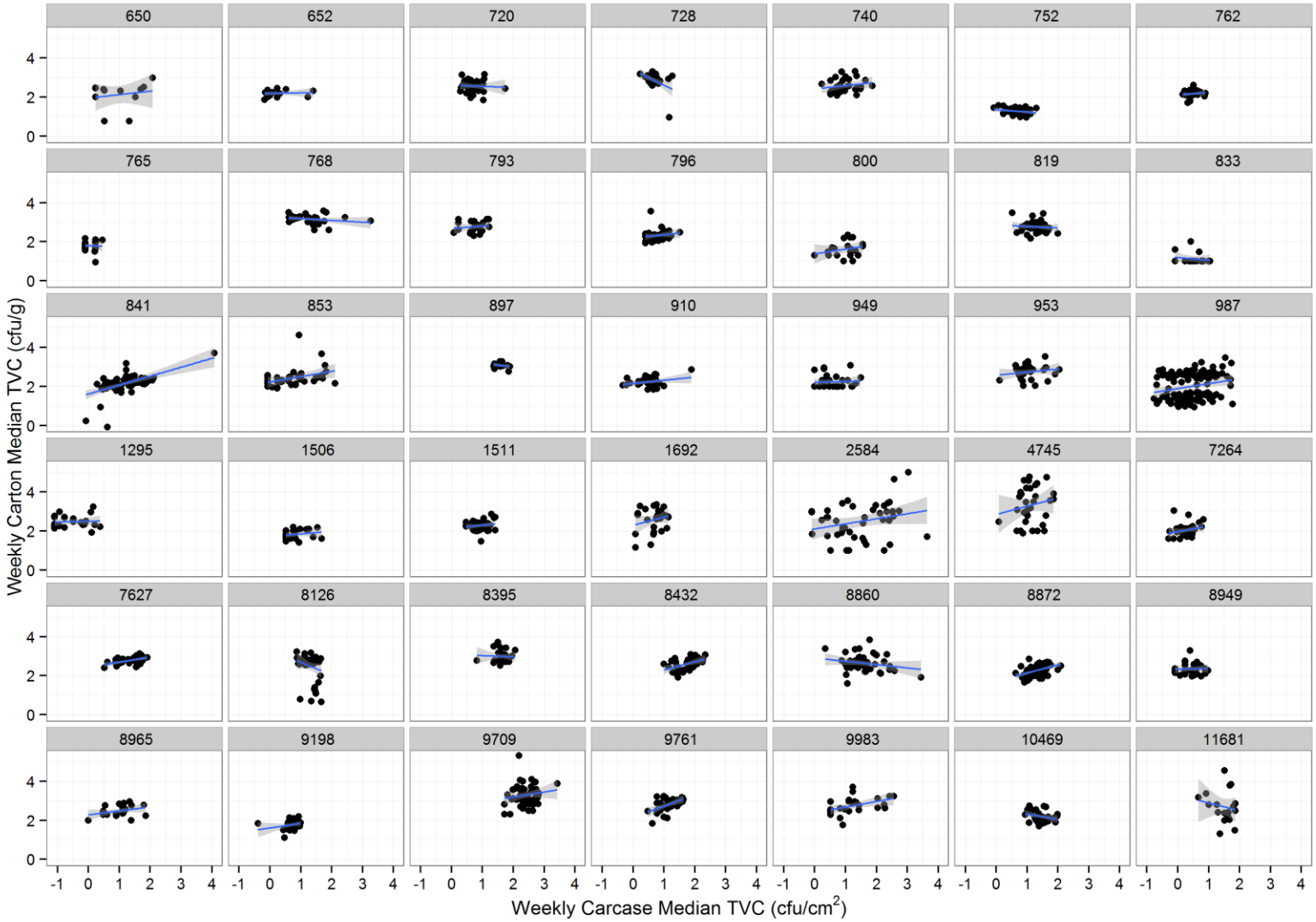


**Figure 2: Carcase vs Carton weekly median TVC for individual plants**

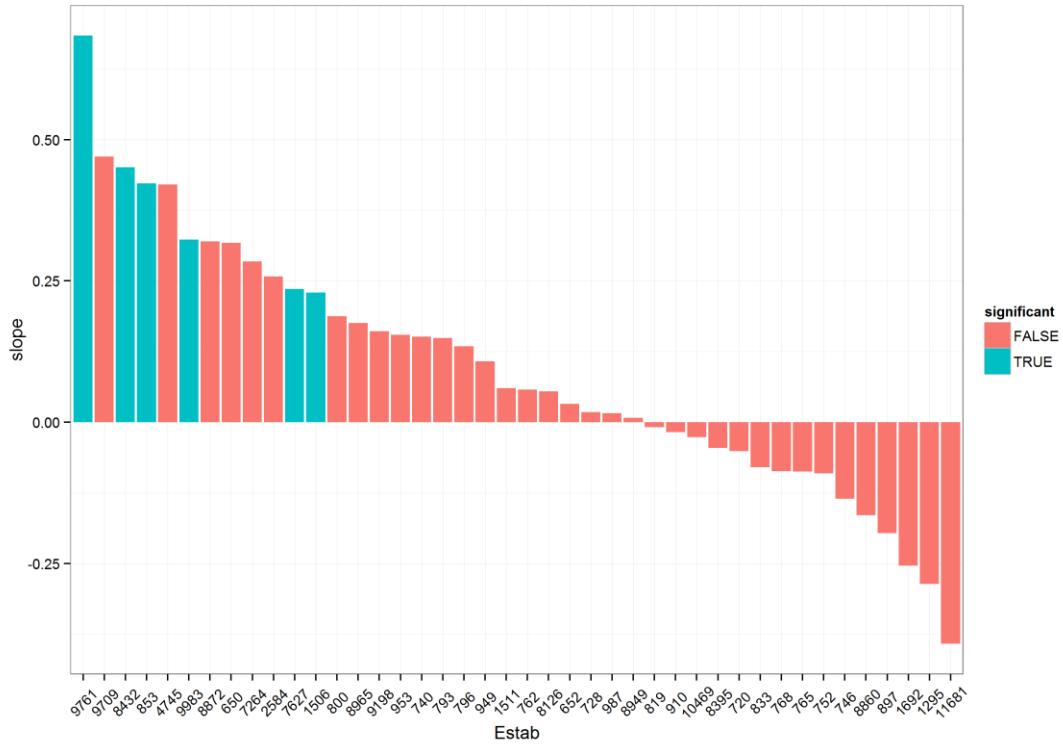


**Figure 3: Magnitude and significance of regression slopes for individual plan**

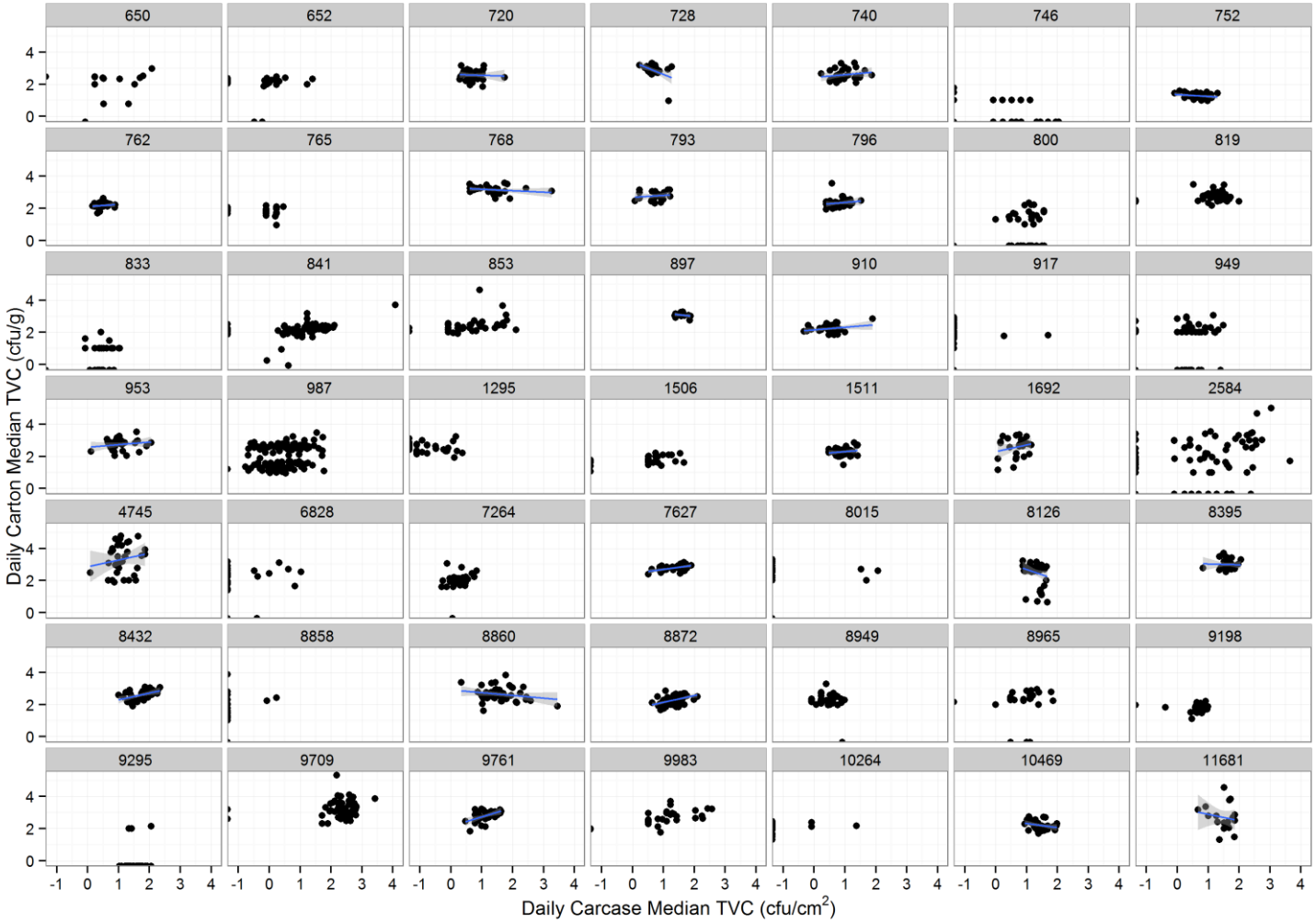


**Figure 4: Carcase vs Carton daily median TVC for individual plants**

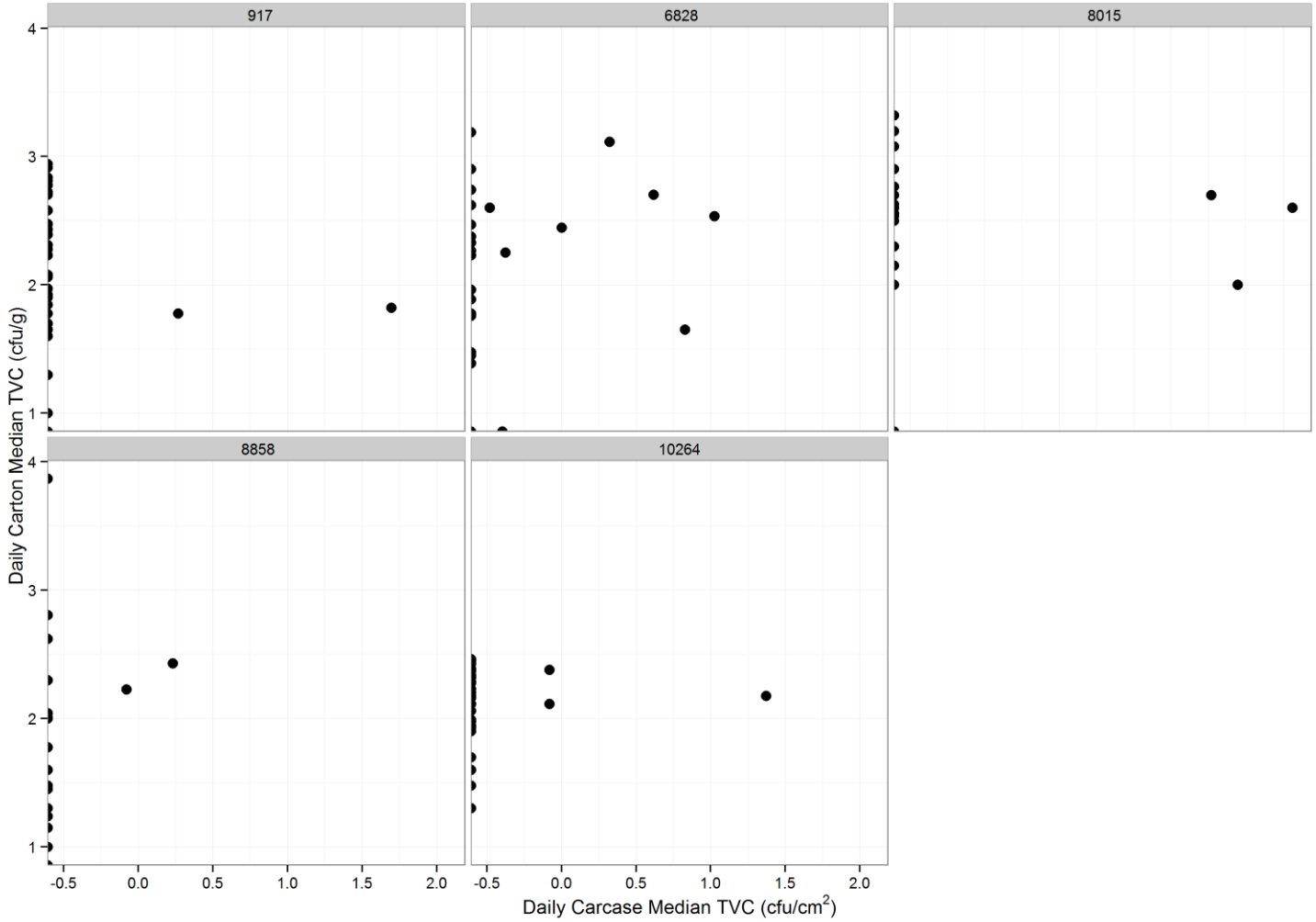


**Figure 5: Daily Carcase and Carton median TVC for plants losing a large proportion of carton results**

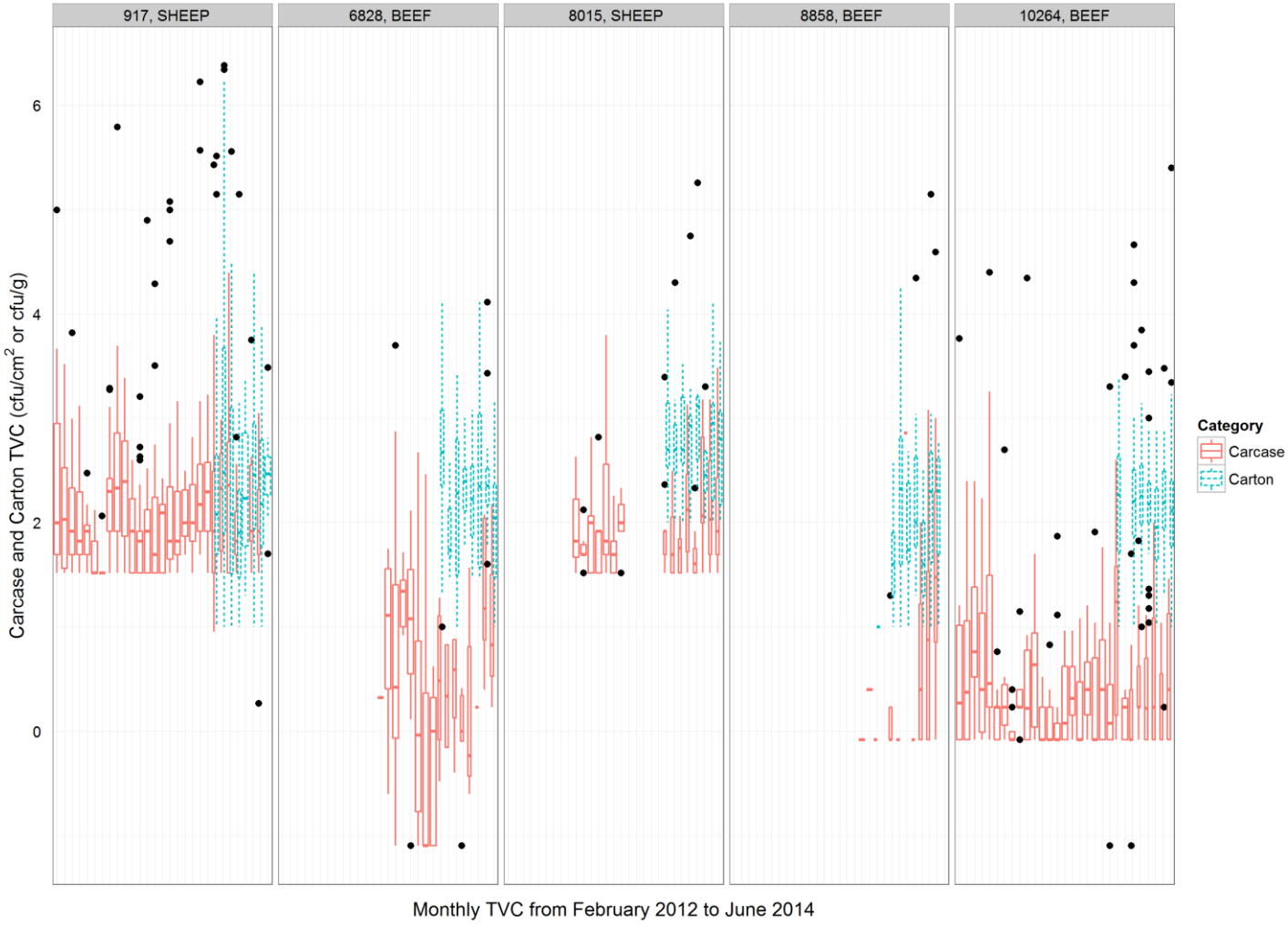


**Figure 6: Monthly carcase and carton TVC boxplots for plants with a large proportion of non-detect carton results**