

# Final report

---

## V.MSF.0448 Microbiological food safety of effluent from animal industries

Project code: V.MSF.0448  
Prepared by: Peter Horchner  
Symbio Laboratories Pty Ltd  
Date published: 30 April 2022

PUBLISHED BY  
Meat and Livestock Australia Limited  
PO Box 1961  
NORTH SYDNEY NSW 2059

Meat & Livestock Australia acknowledges the matching funds provided by the Australian Government to support the research and development detailed in this publication.

This publication is published by Meat & Livestock Australia Limited ABN 39 081 678 364 (MLA). Care is taken to ensure the accuracy of the information contained in this publication. However MLA cannot accept responsibility for the accuracy or completeness of the information or opinions contained in the publication. You should make your own enquiries before making decisions concerning your interests. Reproduction in whole or in part of this publication is prohibited without prior written consent of MLA.

## Abstract

Livestock are a known source of pathogens that could be transferred to the food chain and present a food safety risk without other 'hurdle interventions'. In the event of a foodborne illness outbreak, source attribution studies and investigations increase accountability and potential consequences to the supply chain to learn from.

The project examined microbiological quality of waste and effluent in the red meat supply chain, to identify potential risks which may not have been previously evaluated.

A survey of practices was conducted by desktop survey and site assessments at selected premises including farms; feedlots; saleyards; processing establishments; composters; and transport operators. Samples were collected from feedlots, saleyards, and processing establishments and a pathogen and indicator microbiological tests conducted.

The project has provided baseline data and identified potential risks to other sectors. *E. coli* and coliforms were detected; *Listeria*, *Salmonella* and coagulase-positive staphylococci were not detected in either wastewater or solid waste.

The combination of state environmental control measures over use of effluent, the processes adopted by commercial composters, and the actions of the horticultural sector appear to be acting as a reasonable barrier for transmission of pathogens directly to horticultural crops.

## Executive summary

### Background

Livestock are a known source of pathogens that could unintentionally be directly or indirectly transferred to product within the food supply chain and present a food safety risk. In the event of a foodborne illness outbreak, source attribution studies and epidemiological investigations, which are extending to farm level to identify the primary source of hazards, increase accountability and potential consequences to producers.

The project was initiated to examine the microbiological quality of processed waste and effluent from within the red meat sector, ranging from farms to processing establishments, to provide clarity and identify potential risks which may not have been previously evaluated. It also well accepted that 'Hurdle technology' is a method of ensuring the safety of foods by eliminating or controlling the growth of pathogens, making the food safe for consumption and extending its shelf life through the application of a combination of technologies and approaches. To that end, this project did not consider post treatment effects but acknowledges there exists hurdles along the value chain for example in pre-cut washed produce that for example was grown in fertilised land containing materials from rendering from an abattoir.

### Objectives

The main objective for this aspect of the project was to survey the microbiological quality of processed waste and effluent, ranging from farms to processing establishments, to provide clarity and identify potential risks which may not been previously considered.

The project has been successful in providing baseline data for the first time on a range of pathogens and identifying potential risks with transmission of, most notably, *Escherichia coli* and Shiga toxin-producing *E. coli* (STEC) to other sectors.

### Methodology

The project commenced with a literature review of the tests that need to be conducted, methods for the isolation of bacteria from animal waste and development of a suitable checklist and core constructs for the surveys.

An effluent mapping exercise was completed to assist the identification of sites from which to draw samples for testing.

A survey of current practices was conducted by desktop survey and site assessments of actual practices at selected premises. The sectors included with the numbers of participants surveyed in brackets (desktop, site) were as follows - farms (8, 6); feedlots (21, 7); saleyards (12, 4); processing establishments (24, 6); composters (3, 1); transport operators (1,0).

Samples were collected by the project team and by submission from participants, from 11 feedlots (64 samples); 12 saleyards (73 samples); 24 processing establishments (88 samples) making a total of 225 samples.

Microbial tests conducted on the samples included the following tests:

- Thermotolerant coliforms
- *E. coli*
- *Salmonella* (with serotyping when detections occurred)

- *Listeria monocytogenes*
- *Listeria* spp.
- *Campylobacter* spp.
- Coagulase positive *Staphylococcus*
- *E. coli* O157 & Big 6 STEC<sup>1</sup> Screening
- *E. coli* O157 & Big 6 STEC partial confirmation (part way confirmation to IMS and agglutination for potential positive detections – refer to Section 3.2.2 Testing and Appendix B for further details)

### **Results/key findings**

Based on current practices and test results for the participants in this project, the overall risks of transferring microbiological hazards from processed waste and effluent through the red meat supply chain appear relatively low.

The environmental protection agencies in each jurisdiction appear to control and manage major microbiological food safety related risks by using licensing requirements for the various categories of waste produced in each sector.

### **Benefits to industry**

Based on the results of this project, the red meat supply chain has increased awareness of potential risks from pathogenic bacteria not being fully deactivated by current practices.

Downstream users of processed effluent and wastewater and composted manure, such as the horticulture sector, will benefit from an increased understanding of the hazards present in these products and the risks they present.

### **Future research and recommendations**

It is recommended that MLA consult with several key sectors as well as inform the respective state Environmental Protection Agencies and State Health Departments about control measures in place. It is also recommended that MLA consult further on pathogen prevalence with those composting organic waste and key industry stakeholders especially in horticulture (including nuts).

---

<sup>1</sup> Big 6 STEC refers to the group of non-O157 serogroups of Shiga-toxin producing *Escherichia coli* including *E. coli* O26, O45, O103, O111, O121, and O145 as testing is a requirement by the USDA for beef imported into the US market.

## Table of contents

<b>Abstract .....</b>	<b>2</b>
<b>Executive summary .....</b>	<b>3</b>
<b>1. Background .....</b>	<b>8</b>
<b>2. Objectives.....</b>	<b>8</b>
<b>3. Methodology .....</b>	<b>9</b>
<b>3.1 Literature review .....</b>	<b>9</b>
<b>3.2 Effluent mapping from farm, sale yard, feedlot, processing plant ...</b>	<b>9</b>
<b>3.3 Survey of current practices.....</b>	<b>10</b>
3.3.1 Desktop survey of industry participants.....	10
3.3.2 On-site survey of industry participants .....	11
<b>3.4 Survey of microbiological quality.....</b>	<b>11</b>
3.2.1 Sample Collection .....	11
3.2.2 Testing.....	12
<b>4. Results.....</b>	<b>15</b>
<b>4.1 Literature review .....</b>	<b>15</b>
<b>4.2 Current Practices - Liquid Waste.....</b>	<b>16</b>
4.2.1 Raw discharge to a river/water course .....	16
4.2.2 Raw discharge to fields &/or farms .....	17
4.2.3 Raw discharge – other .....	17
4.2.4 Treated discharge .....	17
4.2.5 Re-use on site .....	18
4.2.6 End Users/Receivers of liquid waste/wastewater.....	15
<b>4.3 Current Practices - Solid Waste.....</b>	<b>19</b>
4.3.1 Manure & lairage/yard wastes.....	20
4.3.2 Paunch .....	21
4.3.3 Downers/dead animals.....	21
4.3.4 Pond crusts from wastewater treatment.....	21
4.3.5 Sludge & Slurry .....	21

4.3.6	Other solid waste.....	22
4.3.7	End Users/Receivers of solid waste.....	17
4.3.8	Other general results.....	26
	Goats.....	18
	Dairy cattle.....	18
<b>4.4</b>	<b>Composting Processes.....</b>	<b>22</b>
4.4.1	Treatment process for the compost.....	22
4.4.2	Accreditation or standard that complies with e.g. AS4454-2012 .....	23
4.4.3	Process Controls .....	23
<b>4.5</b>	<b>Links to Horticulture.....</b>	<b>24</b>
4.5.1	Profile of Horticultural sector.....	24
4.5.2	Pathways into Horticultural sector from Livestock industries .....	25
	Liquid waste/Wastewater.....	25
	Solid Waste/Manure/Compost.....	25
	Macadamia industry .....	25
<b>4.6</b>	<b>Micro testing results .....</b>	<b>26</b>
4.6.1	Liquid Waste/Wastewater.....	27
4.6.2	Solid waste/manure/compost .....	30
<b>5</b>	<b>Key findings.....</b>	<b>20</b>
	<b>5.5 Heading .....</b>	<b>20</b>
	5.5.2 Sub heading .....	20
<b>6</b>	<b>Conclusion and recommendations .....</b>	<b>34</b>
	<b>6.5 Heading .....</b>	<b>21</b>
	6.5.2 Sub heading .....	21
	<b>6.6 Conclusions.....</b>	<b>34</b>
	<b>6.7 Recommendations .....</b>	<b>34</b>
<b>7</b>	<b>References.....</b>	<b>35</b>
<b>8</b>	<b>Appendix .....</b>	<b>39</b>
	<b>8.1 Appendix A – Literature Review .....</b>	<b>39</b>
	8.1.1 Hazard identification .....	39

8.1.2	Quantitative methods for the isolation of bacteria from animal waste .....	40
	Generic E. coli .....	40
	STEC .....	40
	Salmonella.....	40
	Listeria.....	41
	Campylobacter.....	42
	Staphylococcus aureus .....	42
<b>8.2</b>	<b>Appendix B – Approach to confirmation of E.coli O157 and Big 6</b>	
	<b>STEC.....</b>	<b>43</b>
<b>8.3</b>	<b>Appendix C – Questionnaires used for survey of practices .....</b>	<b>46</b>
	<b>Survey of Practices – Microbiological Risks from Waste Material .....</b>	<b>46</b>
<b>8.4</b>	<b>Appendix D – Victoria EPA “Guidelines For Environmental</b>	
	<b>Management Use Of Reclaimed Water”, Table 1 .....</b>	<b>49</b>

## 1. Background

Livestock are a known source of pathogens that could unintentionally be directly (waste, manures) or indirectly (air, dust, water runoff, water source, pests and wildlife) transferred to product within the food supply chain and present a food safety risk. Environmental, demographic, and climate changes have the potential to increase the risk of hazard transmission between agricultural sectors.

In the event of a foodborne illness outbreak, source attribution studies and epidemiological investigations, which are extending to farm level to identify the primary source of hazards, increase accountability and potential consequences to producers. Research funded by the fresh produce industry is investigating some risk factors in food safety e.g. adequately composted manure which may be used for growing fresh produce.

The feedlot industry, with MLA, has published guidelines to help manage and utilise waste and effluent appropriately and safely with minimal negative impact on the environment and surrounding sectors.

The project was initiated to examine the microbiological quality of processed waste and effluent, ranging from farms to processing establishments, to provide clarity and identify potential risks which may not have been previously evaluated. Mapping of the waste and effluent streams in the red meat industry from farm to processors has also been undertaken, with value adding opportunities identified.

## 2. Objectives

The objectives for the project were:

1. A survey of microbiological quality on processed waste and effluent, ranging from farms to processing establishments, to provide clarity and identify potential risks which may not been previously considered.
2. Mapping of the waste and effluent stream in the red meat industry from farm to processor with value added opportunity. (Appendix E)

This report addresses the first objective of the project. The separate MLA reports covering the second objectives is in the appendix 6.5. Overall, with respect to the first objective, the project was successful in identifying some potential risks from waste streams. This was despite having sample numbers reduced from the intended volume as a consequence of the COVID-19 pandemic and frequent lockdowns impacting business continuity and eagerness to participate. There was also reluctance by some participants to join the study, stating concerns about the potential for the project to identify and link hazards directly to their enterprise.



## 3. Methodology

### 3.1 Literature review

As this area is relatively new and there are many unknowns, the project commenced with a literature review. The review covered potential hazards/risks, the tests that need to be conducted, and details of the preferred test methods for the sample matrices involved in this area. The literature review included reviewing previous work referenced in industry standards as they provided insights into the risks that were and weren't seen as likely. Available industry standards for effluent treatment were reviewed to inform and develop a suitable checklist and core constructs for the surveys.

For evaluating test results, the following commonly used standards were considered the most relevant:

- for wastewater, test samples were compared to classes of water and usage permitted as per Victorian EPA guidelines; and
- for compost, tests samples must contain E. coli <100 CFU/g and Salmonella Not Detected/25g which is the requirement to be considered 'compliant' if referencing the Horticultural Industry's Compliant Compost Guidelines<sup>2</sup>.

The literature review also identified preferred quantitative methods for the isolation of bacteria from animal waste. These were used to negotiate methods with the National Association of Testing Authorities (NATA) accredited laboratory.

### 3.2 Effluent mapping from farm, sale yard, feedlot, processing plant

Current effluent processes were mapped by All Energy Pty Ltd by conducting site visits (or similar e.g. video conference with technical images / visual information / process flow sheeting) and associated communications. This was undertaken with a nominated MLA contact/stakeholders for red meat processing plants, saleyards, feedlots and farms.

The emphasis of this part of the project was to define the various waste streams, management options and utilisation options in support of the project.

A separate report was prepared by All Energy Pty Ltd summarising the above and included:

- Mapping of all of the waste streams from farms through to processing facilities and presented in the form of a flow diagram
- Potential value adding/opportunities for waste; and
- Identification of some upcoming waste streams in the next 2 – 3 years with associated micro-survey recommendations (where available).

The All Energy report and mapping exercise was intended to assist the identification of sites from which to draw samples for testing as outlined in section 3.4 below.

---

<sup>2</sup> The Compliant compost guideline is a voluntary industry initiative to provide a verification mechanism for the production of safe compost for the Australian fresh produce industry. The Compliant Compost standard is managed by MRA Consulting Group [www.mraconsulting.com.au](http://www.mraconsulting.com.au)

### 3.3 Survey of current practices

A survey of current practices was conducted in two parts - an initial desktop survey of industry participants to get a broader cross section of information on current practices; and site surveys and assessments of current practices at selected premises.

#### 3.3.1 Desktop survey of industry participants

The initial desktop survey of industry participants was conducted in order to get a broad cross section of information on current practices across each of the main sectors within the scope of this study – including farms, feedlots, saleyards and processing establishments. The survey was done as a series of telephone interviews and other desktop assessments (e.g. satellite images/google maps), and where possible assisted by a structured question and answer style document.

A cross section of establishments from different states, sizes of enterprise, and types of enterprise was selected for the desktop survey. Available industry databases were used to draw suitable candidates for these surveys. Additional contacts were identified from public sources and referrals from survey participants, where possible. Candidates were selected across all states (Queensland, NSW, Victoria, Tasmania, SA and WA) in order to provide a suitable cross section of respondents.

The final numbers of participants for the desktop stage of the project for each sector was as follows:

Sector	Surveyed	Comment
Farms	8	<ul style="list-style-type: none"> <li>• Broadacre farms were seen as lower risk since they mostly well away from horticultural sector, and numbers were lower since they were not likely to gain additional information</li> <li>• Dairies weren't included but may be a risk due to more intensive production system. proximity and concentration of waste streams</li> <li>• Risk areas are seen as farms that are close to horticultural areas and should be sought out</li> </ul>
Feedlots	21	<ul style="list-style-type: none"> <li>• Feedlots range from small to large and were seen as essential to survey due to high volume of effluent especially solids. Most undertake composting to some degree.</li> </ul>
Saleyards	12	<ul style="list-style-type: none"> <li>• There are less saleyards vs feedlots, and volumes are relatively smaller. They commonly work with local councils to control waste.</li> </ul>
Processing establishments	24	<ul style="list-style-type: none"> <li>• Processors produce a significant amount of waste, especially liquids. State EPA arrangements typically in place.</li> </ul>
Composters	3	<ul style="list-style-type: none"> <li>• With widespread use of off-site professional composters, surveys of these companies should be included.</li> </ul>
Transport Operators	1	<ul style="list-style-type: none"> <li>• External transport used extensively. EPA licensing is required for waste.</li> </ul>

Phone interviews with email follow up were used for Farms, Feedlots, Saleyards and Processing establishments.

**Appendix C** contains the questions that were used for the surveys of processors, saleyards, and feedlots. Farm surveys were based on the same themes but less structured.

### 3.3.2 On-site survey of industry participants

In concert with the desktop surveys, site visits were undertaken to assess current practices and adherence to industry standards and protocols. The approach to the site surveys was similar to the desktop surveys and extended to delve deeper into any specific issues or practices of interest that arose from other work in the project. The site surveys covered Feedlots, Saleyards, and Processing establishments selected from desktop survey pool. Farm visits were not seen as necessary initially based on information gleaned from the initial phone surveys and other desktop research conducted.

The COVID-19 pandemic made it difficult to access as many sites as originally planned for the project. Lockdowns and state government declared “Hot Spots” and border closures in various states meant travel restrictions frequently interrupted planned trips to sites. There were also many sites that did not want any non-essential visitors attending their site.

The final numbers of on-site assessments for the project for each sector is as follows:

Sector	Surveyed	Comment
Farms	6	<ul style="list-style-type: none"> <li>• Risk areas are seen as farms that are close to horticultural areas and could be sought out in future in partnership with horticulture stakeholders</li> </ul>
Feedlots	7	<ul style="list-style-type: none"> <li>• Further feedlots could be visited if any innovative practices are identified e.g. pelletising systems for compost or innovative water treatment systems (none visited in this project)</li> </ul>
Saleyards	4	<ul style="list-style-type: none"> <li>• Interactions with trucks and truck washes could be considered a potential risk and could be examined in future (further noting that plant biosecurity which was not in the scope of this project but is a risk issue).</li> </ul>
Processing establishments	6	<ul style="list-style-type: none"> <li>• Processors close to horticultural areas are worth further consideration and investigation if they can be identified (none identified in this project)</li> </ul>
Composters	1	<ul style="list-style-type: none"> <li>• Given the widespread use of off-site professional composters further engagement with this stakeholder group may be warranted</li> </ul>

## 3.4 Survey of microbiological quality

### 3.4.1 Sample Collection

Samples were obtained in two ways – submission by participants from the desktop surveys; and samples collected by Symbio team during site visits.

Sample submission was slightly less than originally planned due to COVID-19 restrictions and enterprises preferring to focus on core business and immediate challenges. The project team managed to gather sufficient samples to make the results meaningful for the major sectors. The following table outlines the number samples collected and submitted by sector.

Sample collection - sector	Total samples
Farms (contributing sites =0)	-
Feedlots (contributing sites =11)	64
Saleyards (contributing sites =12)	73
Processing establishments (contributing sites =24)	88
<b>Total number of samples</b>	<b>225</b>

### 3.4.2 Testing

Microbial tests conducted on the above samples included the following tests:

- Thermotolerant coliforms
- *E. coli*
- *Salmonella* (with serotyping when detections occurred)
- *Listeria monocytogenes*
- *Listeria* spp
- *Campylobacter* spp
- Coagulase positive *Staphylococcus*
- *E. coli* O157 & Big 6 STEC<sup>3</sup> Screening
- *E. coli* O157 & Big 6 STEC partial confirmation (part way confirmation to IMS and agglutination for potential positive detections). The approach used for “confirmation” of *E. coli* O157 and Big 6 STEC was an abbreviated method stopping at latex agglutination as outlined in **Appendix B**. At that stage we know serotype.)

*E. coli* O157 & Big 6 STEC tests were added following the initial literature review and with agreement from MLA. As outlined in the literature review (see **Appendix A**), the environment can become contaminated with STEC allowing transmission of the bacteria within and between animal groups. STEC have been shown to be present in soil and water samples from Australian cattle farms and feedlots and can survive in manure and on pasture for several months. Sheep have also been shown to carry STEC in their faeces. STEC survival in faeces is affected by temperature, with STEC detected for longer periods in faeces stored at lower temperatures e.g., 10°C and 25°C, than at higher temperature e.g., 37°C. Only a small number of STEC are of interest in human disease, therefore testing has been selective along the lines of meat product testing for the USA beef market.

Serotyping of *Salmonella* spp. was to be undertaken as the serotypes may give clues to the original of the salmonellae and also to identify serotypes that may be interest to sensitive markets such as the USA.

<sup>3</sup> Big 6 STEC refers to the group of non-O157 serogroups of Shiga-toxin producing *Escherichia coli* including *E. coli* O26, O45, O103, O111, O121, and O145 as testing is a requirement by the USDA for beef imported into the US market.

### 3.4.3 Test Methods

An overview of the respective test methods is as follows:

#### ***Thermotolerant Coliforms***

Quantitate enumeration of Thermotolerant coliforms was performed using Petrifilm technique. Petrifilm plates are a ready-made culture medium system containing Violet Red Bile nutrients, a cold-water-soluble gelling agent and a tetrazolium indicator that facilitates colony enumeration. A measured amount of sample was plated onto Petrifilm and incubated at 44°C for 24 hours. Thermotolerant coliform colonies were counted at the end of the incubation period. Typical thermotolerant coliform colonies growing on Petrifilm plates produce acid, which causes the pH indicator to deepen the gel colour and gas from lactose fermentation trapped around red colonies.

#### ***E. coli***

Quantitate enumeration of *E. coli* was performed using Petrifilm technique. Petrifilm plates are ready-made culture medium system containing Violet Red Bile nutrients, a cold-water-soluble gelling agent and tetrazolium indicator that facilitates colony enumeration. A measured amount of diluted sample was plated onto Petrifilm and incubated at 35°C for ~48 hours. At the end of incubation, typical *E. coli* colonies were counted.

#### ***Salmonella spp.***

Qualitative testing was performed using real-time Polymerase Chain Reaction (PCR) technology to analyse *Salmonella* species from 25g test samples. The test samples were enriched in buffered peptone water (BPW) and incubated at 37°C for 22 hours. The enriched samples were lysed to release their DNA. The lysate was then used to prepare the PCR reaction which consists of fluorescent dye-labelled probes targeting unique DNA sequences specific to *Salmonella* species, and an internal positive control (IPC). Amplified target DNA, where present, is detected by real-time PCR and analysed using analysis software for result interpretation.

#### ***Listeria monocytogenes***

Qualitative testing was performed using real-time Polymerase Chain Reaction (PCR) technology to analyse *Listeria monocytogenes* from 25 g test samples. The test samples were enriched in *Listeria* enrichment broth and incubated at 37°C for 22 hours. The enriched samples were lysed to release their DNA. The lysate was then used to prepare the PCR reaction which consists of fluorescent dye-labelled probes targeting unique DNA sequences specific to *Listeria monocytogenes*, and an internal positive control (IPC). Amplified target DNA, where present, is detected by real-time PCR and analysed using analysis software for result interpretation.

#### ***Listeria species***

Qualitative testing was performed using real-time Polymerase Chain Reaction (PCR) technology to analyse *Listeria* species from 25 g test samples. The test samples were enriched in *Listeria* enrichment broth and incubated at 37°C for 22 hours. The enriched samples were lysed to release their DNA. The lysate was then used to prepare the PCR reaction which consists of fluorescent dye-labelled probes targeting unique DNA sequences specific to *Listeria* species, and an internal positive control (IPC). Amplified target DNA, where present, is detected by real-time PCR and analysed using analysis software for result interpretation.

***Campylobacter spp.***

Qualitative detection of *Campylobacter* was based on AS 5013.6 and performed by selective enrichment of sample aliquots in media broth and subsequent isolation on specific agar media. A Specific amount of test sample, 25g, was measured/weighed out and enriched in Preston Broth for 48 hours. The enriched sample was analysed by surface spread method on Preston and Skirrow agar media plates. Typical growth on these plates was confirmed by Matrix-assisted Laser Desorption Ionization Time-of-flight (MALDI-TOF) mass spectroscopy.

***Coagulase positive Staphylococcus***

Coagulase positive *Staphylococci* was isolated quantitatively using surface spread method, based on AS 5013.12.1. Specific amount of test sample (25g) was measured weighed out and diluted to perform initial suspension. The specific medium, Baird Parker agar, was inoculated on the surface with specific quantity of test sample suspension and incubated aerobically at 37°C for 48 hours. The identification of isolates was confirmed by Matrix-assisted Laser Desorption Ionization Time-of-flight (MALDI-TOF) mass spectroscopy and coagulase production confirmed biochemically.

***E. coli O157:H7 & Big 6 STEC Screening***

Screening of *E. coli* O157:H7 & non O157 STEC was performed by PCR using Genetic Detection Systems (GDS) biocontrol system, analysing multiple genetic targets for Shiga-toxin producing *E. coli* O157:H7 and other six other non- *E. coli* O157:H7 STEC serotypes (O26, O45, O103, O111, O121, O145) in test samples. *E. coli* O157:H7, where detected, was characterized by H7 plus either of the Shiga toxin genes *stx1* or *stx2* genes while the non-O157 STEC, where detected, were characterized by the *eae* gene plus at least one of the Shiga toxin genes (*stx1* or *stx2*).

A 25g portion of test sample was enriched in proprietary modified enrichment medium, followed by concentration of test analytes by using a proprietary IMS-based sample preparation. PCR amplification of highly conserved DNA sequences for the target organisms with specific primers was performed for genetic analysis of the associated pathogenicity genes.

***E. coli O157:H7 IMS (Partial Confirmation)***

Test samples with screen positive results were considered potential positives and analysed further for confirmation. This method is based on enrichment in a selective broth medium, followed by isolation of *E. coli* O157 using immuno-magnetic separation (IMS). Post concentration process, immunomagnetic particles with adhering bacteria were subcultured onto Modified Rainbow Agar (mRBA) and CT-SMAC. Typical isolates from specific agar medium were tested for agglutination with *E. coli* O157 antiserum.

***Big 6 STEC IMS (Partial Confirmation)***

Samples that were potential positive from screening for Shiga-toxin (*stx*) and Intimin (*eae*) genes were analysed further using real-time PCR for specific pathogenic STEC serogroups. Screen-positive test samples were culturally isolated by immuno-magnetic separation (IMS) process using beads coated with antibodies (major six serogroups) followed by plating on modified Rainbow Agar (mRBA). Typical isolates were purified on Sheep Blood Agar (SBA) and tested for specific O antigens using latex agglutination.

## 4. Results

### 4.1 Literature review

**Appendix A** contains the full literature review. The key points for the purposes of this study are provided here.

Thermotolerant coliforms and *E. coli* are typically used as indicators of faecal contamination although they are not always reliable indicators of the presence of bacterial pathogens, and not all thermotolerant coliforms are of faecal origin. *E. coli* is the most appropriate group of coliforms for the identification of faecal pollution. As the concentration of *E. coli* in animal effluent is expected to be (initially) high it is the preferred indicator organism for inclusion in surveys of the microbiological contamination of animal effluent.

The “Guidelines for fresh produce food safety 2019<sup>4</sup>” was used as a reference document to determine the pathogens of interest for the fresh product industry and their general microbiological requirements. The following bacterial hazards were selected to be included in the survey of effluent from red-meat animal industries:

- *E. coli* (generic)
- *Salmonella* spp. (to be serotyped when detected)
- *Listeria monocytogenes*
- *Campylobacter* spp.
- Coagulase positive *Staphylococcus* and STEC have been included for completeness, however, quantification of these hazards is not essential for this project.

A partial confirmation (to serotype stage on colonies) was included for potential positive detections of *E. coli* O157 and Big 6 STEC. **Appendix B** provides an overview of the partial confirmation method used and a copy of the process flow for the full method.

The majority of fresh produce which does not need to be washed, peeled or cooked, such as bagged salad, fresh fruits and nuts are classified as Ready To Eat (RTE) and need to meet the FSANZ of microbial limits for RTE foods. Table 0 below gives an outline of the microbial requirements.

---

<sup>4</sup> <https://fpac-anz.com/food-safety-guidelines-2019/>

**Table 0. Critical limit on read to eat foods (FSANZ)**

Indicator		Satisfactory	Marginal	Unsatisfactory
<b>E. coli</b>		<3 cfu/g	3 – 100 cfu/g	>100 cfu/g
<b>Total coliforms</b>		<100 cfu/g	100 – 10,000 cfu/g	>10,000 cfu/g
<b>Salmonella spp.</b>		Not detected in 25g	-	Detected in 25g
<b>Listeria spp</b>		Not detected in 25g	<100 cfu/g	>100 cfu/g
<b>Listeria monocytogenes</b>	RTE foods that support growth of <i>L. monocytogenes</i>	Not detected in 25g	-	Detected in 25g
	RTE foods that do not support growth of <i>L. monocytogenes</i>	Not detected in 25g	Detected, <100 cfu/g	>100 cfu/g

## 4.2 Current Practices - Liquid Waste

Based on the desk top review and the waste streams work conducted by All Energy Pty Ltd in the project, the practices that the survey sought to explore for each sector included the following:

- Raw discharge to a river/water course
- Raw discharge to fields &/or farms
- Raw discharge – other
- Treated discharge
- Re-use on site

Results for each of these are summarised below.

### 4.2.1 Raw discharge to a river/water course

- There was only one respondent (processor), that reported discharging of raw wastewater to a river/water course. No feedlot or saleyard enterprises report this practice for the bulk of their wastewater, however, two feedlots and two saleyard respondents reported that surface water runoff did find its way to nearby rivers, especially during a heavy rain event. Field assessment identified two more scenarios where there were nearby rivers that in theory raw wastewater run off would reach streams feeding into rivers. Given there is some research being done on source attribution, it would be worth considering taking some samples from these entry points and testing for *E. coli* serotypes.
- Farm respondents to date have been broad acre producers and the liquid wastes from their livestock fall naturally to ground. Whilst they agreed that it is possible some waste finds its way to a nearby stream or ground water, and this could result in some pathogens theoretically finding their way to horticultural farms, they did not see this as within their control. This survey did not identify any nearby horticultural farms that would be drawing



from the farms close by, however, it may be worth conducting some further sampling and testing in such areas where they can be identified.

- The control measures for this potential source of microbial hazard include the natural separation by distance and time. From a broad perspective, ground water would have a microbial load from 'natural' microflora as well as other added microbes from a range of sources, not just livestock sector. The red meat industry should take note that academics and health departments do conduct attribution studies on the origin of pathogens and with rapid advances in genome testing and there may be more definite linkages identified in future.
- Further to this, Controlling Authorities approve licenses on an enterprise-by-enterprise basis and this process would appear to offer an avenue for identifying and addressing any risks at a local level.

#### **4.2.2 Raw discharge to fields and/or farms**

- There have not been any respondents from processing or feedlot enterprises that report discharging of completely raw wastewater to neighbouring fields or farms, i.e., in all cases there was some form of treatment, at a minimum settle ponds or aerobic ponds.
- One saleyard operator reported wastewater used to irrigate pasture, with excess to sewage treatment plant in times of peak flow. No livestock on paddocks while irrigating.

#### **4.2.3 Raw discharge – other**

- There were two processor and three saleyard respondents that reported discharging of raw wastewater directly to council sewerage treatment plants (STPs) after a solid separation process. This process is regulated with specifications for the wastewater specified and verification testing of physiochemical indicators conducted.
- Council/municipal STPs are themselves regulated and closely monitored and these scenarios are seen as low risk.

#### **4.2.4 Treated discharge**

- Most respondents reported some form of treatment of liquid waste prior to dispatch
- Liquid waste typically drains to an individual or series of ponds (aerobic and anaerobic) for settling and further treatment
- Further treatment varies across enterprises and includes the following measures:
  - wastewater pond where the aerobic digestion takes place, sludge is pumped out and treated wastewater then pumped onto the fields as irrigation water to grow crops (e.g. sorghum or maize) or simply left to evaporate. This practice occurred in all the regional and more remote areas surveyed, especially discharges from processors and feedlots.
  - one respondent reported conducting pre-screening, wastewater filtration with UV treatment of filtered wastewater
  - one respondent reported pre-screening, treatment dissolved and chemical air flotation

- one respondent reported pre-screening, anaerobic and aerated/facultative ponds, final tertiary Dissolved Air Filtration (DAF) and Chlorination
- As above in 4.2.3, where transferring to STPs, respondent typically had some form of solid separation or DAF and then directed to sewer system
- two respondents reported wastewater from a saveall going to Covered Anaerobic Lagoon/s (CAL) which was also used for biogas production and subsequent energy generation
- One saleyard operator reported they implemented a 'seeding' process for the surface of their dam to stimulate growth of the desired organisms
- Processors and saleyards are typically less remote than feedlots and farms which meant that the liquid waste streams for the former were, in many cases, returned to either municipal STPs or wastewater treatment plants (WWTPs) if already treated. WWTPs have well established processes in place, with managed control measures and verification processes in place, include routine testing for biochemical, phys-chem and microbial indicators and therefore risks to other food sectors would be seen as low where this is the case.
- In such cases wastewater is discharged in accordance with trade waste agreements with council and/or water authorities
- For the other enterprises, once treatment is completed, water sprayed onto paddocks or used for irrigation of crops via table drains/flood irrigation.
- Verification testing occurs in operators where the matrix is specified in an EPA agreement; including monthly through to six monthly discharge "grab" samples for parameters such as pH & suspended solids; and more extensive quarterly testing across a range of parameters. EPA control orders for individual sites are publicly available in most states.
- Self-imposed verification testing is also in place for major operators and includes basic micro testing of effluent water where irrigated on site in integrated operations such as feedlot and cropping or processors with their own cropping operations.
- Several respondents queried the closed loop nature of spraying and other use of wastewater for irrigation of crops that are subsequently fed to animals. The query was whether there could be enhanced risk of build-up of pathogenic microbes, especially *E. coli*, that may then be higher at the meat processing stage. It would be worth MLA considering this question further and conducting a targeted trial, such as an attribution study if seen as a potential issue.
- Almost all saleyard respondents, several processors and feedlots raised the issue of the truck wash areas. The truck wash water, by volume, was seen as being a substantial part of run-off water at saleyard depots. Transport operators are required to have EPA approvals. However, the concern raised was that any given wash down was not likely to be 100% effective and could give rise to pathogens from other journeys being mixed in with waste from the standing stock.

#### 4.2.5 Re-use on site

- Re-use of water on site was report for processors, feedlots and saleyards including the following typical uses:
  - belt press service water

- transfer to a Waste Water Treatment Plant
- Cleaning yards & lairages
- Wastewater used to irrigate pasture on site (excess to sewage treatment plant in times of peak flow) but with no livestock on paddocks while irrigating
- First wash for certain offal items that are naturally contaminated anyway (e.g. omasum)
- Mixed with (shandy of) potable water used for stock wash down and/or truck washdown
- Irrigation of Class B water in line with irrigation plan (as per Victorian EPA guidelines – refer **Appendix D** for Classes of water and usage permitted). Verification is in place for this use, covering ground water testing, surface water testing and river water testing quarterly in line with the Environmental Management Plan
- Watering roads at feedlots for dust control.

### 4.3 Current Practices - Solid Waste

Based on a desk top review and the waste streams work conducted by All Energy Pty Ltd in the project, the practices that the surveys have sought to explore for each sector include the following:

- Manure & lairage/yard wastes
- Paunch
- Downers/dead animals
- Pond crusts from wastewater treatment
- Sludge & slurry
- Other non-organic solids that find their way into solid waste were identified but not assessed specifically in this project.

General points about typical practices for solid waste are:

- As for liquid wastes, respondents referred to oversight by Controlling Authorities, namely the respective Environmental Protection Agency in each jurisdiction and felt that practices adopted were considered 'safe' from the public health point of view due to compliance with the respective legislation/regulations
- There was seen to be increased pressure in recent years from the Controlling Authorities. Three respondents reported major EPA challenges, which were related to both waste treatment and also plant biosecurity requirements e.g., Tropical Soda Apple in NSW, which dictated specific requirements and/or amendments to their solid waste management processes.
- Across all sectors the typical practices for treatment of solid waste could be grouped into three groups – transferred to composters no/minimal treatment; on site static or *ad hoc* windrow composting; and controlled composting (time temp parameters). There were no respondents that reporting having sought certification to AS4454: Composts, Soil Conditioners and Mulches

Results to date for each of these are summarised below.

### 4.3.1 Manure & lairage/yard wastes

- Approximately one third of the processors surveyed were sending manure and lairage/yard waste off site to external commercial composters or nurseries
- In all cases, respondents sending manure and lairage/yard waste off site reported that only licensed service operators were used for transporting waste (including their own transporters in some cases) and that control measures were mainly the contract agreements with specified compliance with regulations
- The majority of processors surveyed were composting manure and lairage/yard waste on site to varying degrees, some were paying to have the material removed, others were being paid by commercial composters for the material as an organic resource, where it is blended with other organics, and others were selling it directly to nurseries or farmers
- Numerous respondents in all sectors in Qld referred to the macadamia industry as taking their compost, either directly or indirectly (via a commercial composter or nursery). Given the harvest process for macadamia nuts, and sale of both raw and roasted nuts, this supply chain was considered further in the project (see section below).
- Almost all feedlots and saleyards surveyed were composting on site to varying degrees before sending material away, with the level of sophistication and verification systems varying widely
- Composting practices varied in the level of sophistication. This is related to the volume of waste produced, i.e., the higher the volume of waste, the more sophisticated the system. Large feedlots have a substantially larger volume of manure, and their composting practices were more sophisticated as a result.
- Pens were cleared of solids periodically and stockpiled prior to screening and spreading. The residence time for stockpiled manure compost ranged from being aged for 6 months prior to spreading before being worked into soil directly after spreading to manure being removed from site by an external contractor within one month.
- Many feedlot and saleyards respondents reported that local home gardeners would come and collect composted manure. The use by these gardeners is presumed to be as a soil amendment, however, it is unknown whether the use is entirely for gardens/nursery plants or whether they also grow any crops for their own food.
- Those processors, feedlots and saleyards composting on site typically referred to use of the NSW composting exemption guidelines<sup>5</sup> as their control measures and verification programs for risks from composting – which means actual or intended application of compost to land as a soil amendment at the premises where it is ‘consumed’ (used). Those enterprises either used manure on site and/or sold it off site after composting.
- Quarterly to Annual soil samples were taken to monitor mainly physicochemical parameters. However, NSW and VIC operators reported sending composted soil samples for verification testing for microbiological testing and, in some cases, weed seeds.

---

<sup>5</sup> Refer to <https://www.epa.nsw.gov.au/-/media/epa/corporate-site/resources/wastegrants/rre16-compost.pdf>

### 4.3.2 Paunch

- For most processors, paunch is dewatered and either sent off-site for composting or composted on site with manure
- Some operators (with higher volumes) separately identify their composting rows containing paunch material and diverted this product to different end user markets, based on their view of organic quality rather than microbial risks
- Approximately 15% of processors reporting using processed paunch materials as part of energy generation on site
- Those sending off-site for composting reported sending material only to an EPA-licenced facilities.

### 4.3.3 Downers/dead animals

- For feedlots and processors (that did not take dead stock for rendering), there were reported to be very specific procedures for composting and SOPs were in place accordingly.
- The compost rows for dead stock were separately managed and monitored with controls for the frequency of turning of compost rows with physical observation contributing to suitability of breakdown (see section on composting below).
- Saleyards surveyed did not generally comment on treatment of dead stock, other than one regional operator saying they used incineration.
- Farms interviewed did not conduct any specific composting of dead stock and they were simply left in the field. These respondents stated this was the typical practice as well as agreeing that some farms may choose to incinerate the dead stock.

### 4.3.4 Pond crusts from wastewater treatment

- Approximately 15% of respondents reported they heat treated and sent pond crust for composting off site (maximum of 95°C and maintained above 60°C for more than an hour). In these cases, temperature measurement was used for verification.
- The majority of respondents reported that settlement ponds are dewatered, dug out and sent for composting or composted and used on site.
- Sediment from basin solids and other pond crust were mostly removed and stockpiled as per other manures as above.

### 4.3.5 Sludge & Slurry

- Two operators reporting using pond sludge in their digester plant for energy recovery otherwise it was used on used or sent for composting off site.
- Sludge was not as high a volume as other material therefore depending on the size of the operation, so many sites did not have the volume warrant special treatment simply buried or used for landfill on site or included with other composted material on or off site with no separate or different process involved.

### 4.3.6 Other solid waste

- Processors – Fly Ash from coal fired boilers is also removed and typically sold and taken away to professional composters and used as an ingredient. It has also been reported go to land fill. Given the high temperature of the ashing process, it is highly unlikely there would be any microbes survive the process.
- Inorganic waste was not considered in the project. There were small volumes of material such as baling twine and plastics observed during site visits. These materials were usually removed at screening stage (20mm sieve) of compost and removed.

### 4.3.7 Composting Processes

The practices, controls and verification programs in place for composting range from “static rows, based on guidelines or common practice” to the more sophisticated “operating according to AS4454”. In general, there was a correlation between scale/volume of waste material and the level of sophistication.

Consequently, the term “composting” is used very broadly and has an equally broad meaning or interpretation across the industry players surveyed. From the above section on practices in place, we can see that it cannot be assumed that all players have adequate control over the compost they produce, transfer or sell to other parties. The microbiological testing results below suggest there are some enteric pathogens potentially still present in the current ‘composting’ practices, albeit not *Salmonella* or *Listeria*.

Further to this, with approximately one third of the processors, feedlots and saleyards sending their manure, paunch and lairage waste off site to commercial composters, the practices of the commercial composting and nursery operators might be examined by businesses disposing of their waste in this way.

Given the widespread use of composting of manures, lairage waste, dead stock and paunch material, across processors, feedlots and saleyards, the project sought further questions of participants about the management of these practices (using the questions shown in in **Appendix C**).

Based on data provided during site visits, it appears the temperatures achieved due to the natural composting or fermentation processes are sufficient to achieve pasteurisation and occur regardless of season. Rainfall is a factor all year round and managing compost in the face of rain events is undertaken all year around. The project did not attempt to separate time periods by season and look at composting as an ongoing activity and analysed data as a single data set.

#### *Treatment process for the compost*

Detailed time-temperature and operational procedures were provided in the desktop surveys, which supported the case for well managed composting processes. Site assessments should the procedures were in place as per procedures, albeit that opportunities for ‘less than perfect’ pasteurisation were observed – e.g. windrow turning is performed with endloaders and it is not practical to know whether all material at the surface of the pile finds its way inside where heat is greatest. Procedures that could be outlined covered:

- treatment process for the compost; the overall process e.g. In vessels, windrows
- the total time period for the compost to settle/stand i.e., for a given batch

- how frequently is it turned over/aerated and for how many occasions in its life i.e. in that batch on site

#### *Accreditation or standard that complies with AS4454-2012*

To date there are no meat industry respondents claimed to have certification to AS4454-2012, however, many respondents claimed to operate to this standard, without seeking certification.

#### *Process Controls*

As mentioned above, across all sectors the typical practices for treatment of solid waste could be group into three groups – transferred to composters no/minimal treatment (minimal to no process controls in place); on site static or ad hoc windrow composting (basic controls of dewatering, ad hoc time and temperature approach with ad hoc turning of rows by observations); and controlled composting (detailed time temp parameters as per next point).

The third group reported, and were observed to have the following typical process controls in place:

- Pasteurisation in windrows with a composting management plan, often in accordance with an EPA control order
- The initial composting (3 to 4 weeks) followed by windrow turning and consolidation for the maturation or curing process (further 8 to 12 weeks). The total composting process can take up to 16 weeks (commonly 12 and 14 weeks), depending on the material mix, ambient temperature and other factors
- Once the windrow is constructed, initial composting commences within the windrow. Turning the windrow is required to maintain aeration, moisture content and prevent the development of anaerobic conditions (which cause odours)
- Turning is typically weekly during the initial phase and fortnightly during maturation stage, and subject to weather conditions
- To meet the requirements of Australian Standard 4454-2012, pasteurization of the compost is achieved by maintaining the internal temperature of the windrow above 55°C for three consecutive days
- Windrow temperature checks are meant to be weekly but can vary in practice - initial composting 3- 4week above 55°C then maturation stage 65-70°C for 8-12 weeks.
- Where there are multiple windrows, as is mostly the case, one or more windrows are compiled each week, being added to each day until the end of the week. Some sites were observed to have 15-20+ windrows on site at any one time, at different stages of maturation.
- Calibration of temperature probes is reported to be done monthly on site, with an annual NATA “third party” calibration of temperature probes within +/-1 °C.
- For approved programs, some report conducting an annual NATA laboratory “third party” process validation – with initial & matured samples tested for thermotolerant coliforms. and enterococci. Others have report this was only required initially for approval.

An important point to note is that despite the above practices, some pathogens were still detected (see micro results below), however, there was no quantification of those pathogens and levels of indicators were typical lower than requirements.

### 4.3.8 Links to Horticulture

Given the importance in this project of the linkages to horticulture sector and reporting by respondents of such linkages, it is worth considering this further.

#### *Profile of Horticultural sector*

The major horticulture growing areas in Australia include:

- Goulburn Valley of Victoria
- Murrumbidgee Irrigation Area of New South Wales
- Sunraysia district of Victoria and New South Wales
- Riverland region of South Australia
- Northern Tasmania
- Southwest Western Australia and
- The coastal strip of both northern New South Wales and Queensland.

#### **Fruit**

- There were 2.57 million tonnes of fruit grown in Australia in 2019/20
- Over 1.47 million tonnes of fresh fruit valued over \$4.7 billion was grown and sold in Australia
- The balance, 515,435 tonnes of fresh fruit valued over \$1.4 billion was exported

#### **Vegetables**

- There were 3.69 million tonnes of fresh vegetables grown in Australia in 2019/20
- Over 1.85 million tonnes of fresh vegetables valued over \$4.2 billion was grown and sold in Australia.
- The balance, 210,824 tonnes of fresh vegetables valued over \$275 million exported internationally.

#### **Nuts**

- Tree nut crops grown throughout Australia include almonds, cashews, chestnuts, hazelnuts, macadamias, peanuts, pecans, pistachios and walnuts
- The Riverina and Northern Rivers regions of New South Wales are major producers of almonds, chestnuts, hazelnuts, macadamias, pecans and walnuts
- In Victoria, the Sunraysia, Swan Hill, central west and north-eastern regions of Victoria produce almonds, chestnuts, hazelnuts and pistachios. Mount Hotham in Victoria produces a small amount of pine nuts.
- The Riverland and Adelaide Hills regions of South Australia produce chestnuts, walnuts and, in the Pinnaroo regions, a small amount of pistachios are grown.
- Queensland produces macadamias, pecan and cashews.
- The Swan Valley region of Western Australia produces almonds, chestnuts, and hazelnuts.
- Tasmania produces a small amount of hazelnuts and walnuts.
- 99,835 tonnes of Australian nuts were exported overseas with a value of over \$942 million.

**Source: Hort Innovation Australian Horticulture Statistics Handbooks 2019/20**

It is recommended that the results of this work are communicated to the horticulture sector to assist them to assess risks.



## *Pathways into Horticultural sector from Livestock industries*

### *Liquid waste/Wastewater*

No respondents in this study reported either providing irrigation water directly or spraying wastewater directly onto neighbouring farms with horticultural crops. As this was a survey rather than a census, this is not to say that there may not be cases of this happening. However, it appears that licensing requirement by the state based environmental authorities has addressed this risk by limiting the practice. There is verification testing in place in most states which monitors coliform levels in wastewater. It is recommended that MLA consult with several key sectors as well as inform the respective state Environmental Protection Agencies and State Health Departments about control measures in place.

One potential avenue for transmission of pathogens via liquid waste is casual runoff of surface water into rivers, which are then pumped downstream by horticultural operators and used for (spray or other) irrigation. With the current licensing requirements in place and the trend towards genomics for attribution studies, they may be more information obtained in the future to determine if this theoretical possibility of pathogen transmission occurs in practice. It is recommended that a watching brief on genomics and attribution studies be maintained by MLA.

### *Solid Waste/Manure/Compost*

A much more widespread practice is the transferring of solid waste in the form of dried manure or composted manure directly to the horticulture sector. Although direct transfer was reported to be infrequent, low volumes or ad hoc, it was still reported. However, the indirect transfer of solid waste via composters was reported frequently. It is recommended that MLA consult further on pathogen prevalence with those composting organic waste and key industry stakeholders in horticulture (including nuts).

Respondents reported compost being directly or indirectly transferred to the following horticultural sectors:

- Carrots
- Oranges
- Other citrus farms
- Table grapes
- Nuts, especially Macadamias (see section below)
- Sugar cane farms
- Professional nurseries and turf farms
- Home gardeners (usually local)

### *Macadamia industry*

Given the macadamia industry was mentioned specifically as receiving a significant amount of compost from the red meat sector by participants in the survey (mainly Queensland and Northern NSW), the project reached out to that sector to glean further information. This is only one sector that has direct links, so it used here as an example of what other horticultural sectors may be doing.

Australia is among the world's largest producers of macadamia nuts with ~40,000 tons of nuts harvested from 6 million trees annually. The trees are planted in rows, and mulch; fertilizer and compost; are applied. Nuts are de-husked at harvesting, on farm or at processing establishments, and dried before cracking, roasting, and/or further processing.

*Salmonella enterica* is a common contaminant of macadamia nut kernels and *Salmonella* spp monitoring and verification is widespread in the macadamia industry as the main pathogen of interest. The source of the salmonellae is of interest to the macadamia industry and they have been working with Health Authorities undertaking attribution modelling and associated studies.

In one such study, findings by Munck et al (2019) were that that macadamia nuts were contaminated by direct transmission from animals with access to the plantations (e.g., wildlife and companion animals) or from indirect transmission from animal reservoirs through biosolids-soil-compost. One of their two attribution models attributed 47% of all *Salmonella* detections on macadamia nuts to biosolids-soil-compost. Wildlife and companion animals were found to be the second and third most important contamination sources, respectively. A second model had lesser percentages and varied with season but still established the connection to biosolids as a source.

It is recommended that MLA/AMPC engage with the macadamia industry and health authorities on attribution of *Salmonella*.

#### 4.3.9 Other general results

General points about the enterprises surveyed and potential impacts on waste streams were:

- Most respondents referred to oversight by Controlling Authorities, namely the respective Environmental Protection Agency in each jurisdiction and felt that the practices they adopted were considered 'safe' from a public health perspective as a result of compliance with the respective legislation/regulations (conditions of licence).
- Further to this Controlling Authorities approve licenses on an enterprise-by-enterprise basis and this process would appear to offer an avenue for identify and addressing any risks. Numerous examples of publicly available control orders and approvals for each sector of the red meat industry plus composters and other major waste processors were obtained and reviewed to confirm this was the case.
- Approximately 60% of processors and most saleyards were in close proximity to major regional towns, the remainder of these and most feedlots were in more regional areas.
- Other than respondents near major towns, all had farms within 5km of their operations and most had some type of farm adjoining their enterprise.
- The type of neighbouring farms was, in general, broad acre grazing and/or cropping and therefore low risk of pathogens entering the food chain directly.
- Four respondents reported that they have nearby horticultural farms. Two respondents reported their waste material (solids only) being used directly by horticultural farms (carrots and table grapes).
- Two respondents reported having nearby dairy farms. The mention of dairy farms highlighted a potential risk for the cattle industry. The project didn't explore dairy farms specifically.

#### 4.4 Micro testing results

Microbiological testing data in this project was skewed towards the processing and feedlot sectors as these sectors were the most accessible during the COVID-19 pandemic period. Although there

were not sufficient samples taken across all sectors to allow meaningful statistical analysis for between sector comparisons of pathogen and indicator prevalence, this report provides descriptive statistics for combined (all sector) results as well as the individual sector results.

#### 4.4.1 Liquid Waste/Wastewater

Samples were sought from (mostly) the “treated” or “finished” state as per the dispatch point from the respective enterprise. For example, for many respondents “treated” wastewater refers to the fact it had been through their aerobic pondage system, and therefore the process was as completed, ready to be discharged to waste streams or sprayed onto paddocks.

A selection of raw or in/process wastewater samples was also included for testing where there was no further treatment and/or to determine for the project what the microbiological risks might be prior to further treatment. The collection of samples essentially “before” and “after” some form of treatment was considered worthwhile for seeing risk through the process, albeit not meant to be a proper trial of efficacy as such. The test data summarised below is presented with the delineation between raw/process vs all other forms of “treated” wastewater. “Exceedance” levels for *E.coli* were arbitrarily set at the Class B irrigation water requirement of 100cfu/mL as per Victorian EPA Guidelines (Refer Appendix D – Victorian EPA Guidelines), which set exceedance levels for *E. coli* for Class B and C irrigation water at 100cfu/mL and 1000cfu/mL respectively.

Table 1 provides the results for all sectors combined and Tables 2, 3 and 4 provides the results for the processing, feedlot and saleyard sectors respectively.

**Table 1: Microbiological test results wastewater – all sectors**

All Sectors	LOR – liquid waste	Wastewater – raw/in process (N = 32)	Wastewater – treated/at dispatch from ponds (N = 67)
Thermotolerant Coliforms	10 cfu/mL	Detection rate – 72% Mean <sup>a</sup> – 258 cfu/mL Exceedance rate <sup>b</sup> – 50%	Detection rate – 51% Mean <sup>a</sup> – 113 cfu/mL Exceedance rate <sup>b</sup> – 22%
<i>E. coli</i>	10 cfu/mL	Detection rate – 22% Mean <sup>a</sup> – 101 cfu/mL Exceedance rate <sup>b</sup> – 13%	Detection rate – 24% Mean <sup>a</sup> – 70 cfu/mL Exceedance rate <sup>b</sup> – 3%
<i>Salmonella</i> spp.	ND/25mL	Detection rate – 0%	Detection rate – 0%
<i>Listeria monocytogenes</i>	ND/25mL	Detection rate – 0%	Detection rate – 0%
<i>Listeria</i> spp.	ND/25mL	Detection rate – 0%	Detection rate – 0%
<i>Campylobacter</i> spp.	ND/25mL	Detection rate – 0%	Detection rate – 15%
CP staphylococci	100 cfu/mL	Detection rate – 0% Mean – 0 cfu/mL	Detection rate – 0% Mean – 0 cfu/mL
<i>E. coli</i> O157:H7 IMS (Partial Confirmation)	ND/25mL	Detection rate IMS – 0%	Detection rate IMS – 0%
Big 6 STEC IMS (Partial Confirmation)	ND/25mL	Potential positive rate IMS – 38%	Potential positive rate IMS – 48%
<b>a = Mean is simple arithmetic mean of detected results</b>			
<b>b = Exceedance rate defined as percentage &gt;100cfu/mL</b>			

**Table 2: Microbiological test results wastewater – processing sector**

Processing Sector	LOR – liquid waste	Wastewater – raw/in process (N = 12)	Wastewater – treated/at dispatch from ponds (N = 32)
Thermotolerant Coliforms	10 cfu/mL	Detection rate – 75% Mean <sup>a</sup> – 95 cfu/mL Exceedance rate <sup>b</sup> – 25%	Detection rate – 41% Mean <sup>a</sup> – 45 cfu/mL Exceedance rate <sup>b</sup> – 6%
<i>E. coli</i>	10 cfu/mL	Detection rate – 8% Mean <sup>a</sup> – 1 cfu/mL Exceedance rate <sup>b</sup> – 0%	Detection rate – 25% Mean <sup>a</sup> – 47 cfu/mL Exceedance rate <sup>b</sup> – 3%
<i>Salmonella</i> spp.	ND/25mL	Detection rate – 0%	Detection rate – 0%
<i>Listeria monocytogenes</i>	ND/25mL	Detection rate – 0%	Detection rate – 0%
<i>Listeria</i> spp.	ND/25mL	Detection rate – 0%	Detection rate – 0%
<i>Campylobacter</i> spp.	ND/25mL	Detection rate – 0%	Detection rate – 6%
CP staphylococci	100 cfu/mL	Detection rate – 0% Mean – 0 cfu/mL	Detection rate – 0% Mean – 0 cfu/mL
<i>E. coli</i> O157:H7 IMS (Partial Confirmation)	ND/25mL	Detection rate IMS – 0%	Detection rate IMS – 0%
Big 6 STEC IMS (Partial Confirmation)	ND/25mL	Potential positive rate IMS – 25%	Potential positive IMS – 32%
<b>a = Mean is simple arithmetic mean of detected results</b>			
<b>b = Exceedance rate defined as percentage &gt;100cfu/mL</b>			

**Table 3: Microiological test results wastewater – feedlot sector**

Feedlot Sector	LOR – liquid waste	Wastewater – raw/in process (N = 7)	Wastewater – treated/at dispatch from ponds (N = 15)
Thermotolerant Coliforms	10 cfu/mL	Detection rate – 14% Mean <sup>a</sup> – 110 cfu/mL Exceedance rate <sup>b</sup> – 14%	Detection rate – 60% Mean <sup>a</sup> – 49 cfu/mL Exceedance rate <sup>b</sup> – 7%
<i>E. coli</i>	10 cfu/mL	Detection rate – 0% Mean <sup>a</sup> – 0 cfu/mL Exceedance rate <sup>b</sup> – 0%	Detection rate – 0% Mean <sup>a</sup> – 0 cfu/mL Exceedance rate <sup>b</sup> – 0%
<i>Salmonella</i> spp.	ND/25mL	Detection rate – 0%	Detection rate – 0%
<i>Listeria monocytogenes</i>	ND/25mL	Detection rate – 0%	Detection rate – 0%
<i>Listeria</i> spp.	ND/25mL	Detection rate – 0%	Detection rate – 0%
<i>Campylobacter</i> spp.	ND/25mL	Detection rate – 0%	Detection rate – 0%
CP staphylococci	100 cfu/mL	Detection rate – 0% Mean – 0 cfu/mL	Detection rate – 0% Mean – 0 cfu/mL
<i>E. coli</i> O157:H7 IMS (Partial Confirmation)	ND/25mL	Detection rate IMS – 0%	Detection rate IMS – 0%
Big 6 STEC IMS (Partial Confirmation)	ND/25mL	Potential positive rate IMS – 25%	Potential positive IMS – 40%
<b>a = Mean is simple arithmetic mean of detected results</b>			
<b>b = Exceedance rate defined as percentage &gt;100cfu/mL</b>			

**Table 4: Microbiological test results wastewater – saleyard sector**

Saleyard Sector	LOR – liquid waste	Wastewater – raw/in process (N = 13)	Wastewater – treated/at dispatch from ponds (N = 20)
Thermotolerant Coliforms	10 cfu/mL	Detection rate – 100% Mean <sup>a</sup> – 381 cfu/mL Exceedance rate <sup>b</sup> – 92%	Detection rate – 60% Mean <sup>a</sup> – 236 cfu/mL Exceedance rate <sup>b</sup> – 60%
<i>E. coli</i>	10 cfu/mL	Detection rate – 46% Mean <sup>a</sup> – 118 cfu/mL Exceedance rate <sup>b</sup> – 31%	Detection rate – 40% Mean <sup>a</sup> – 94 cfu/mL Exceedance rate <sup>b</sup> – 5%
<i>Salmonella</i> spp.	ND/25mL	Detection rate – 0%	Detection rate – 0%
<i>Listeria monocytogenes</i>	ND/25mL	Detection rate – 0%	Detection rate – 0%
<i>Listeria</i> spp.	ND/25mL	Detection rate – 0%	Detection rate – 0%
<i>Campylobacter</i> spp.	ND/25mL	Detection rate – 0%	Detection rate – 15%
CP staphylococci	100 cfu/mL	Detection rate – 0% Mean – 0 cfu/mL	Detection rate – 0% Mean – 0 cfu/mL
<i>E. coli</i> O157:H7 Screening	ND/25mL	Potential positive rate – 46%	Potential positive PP – 55%
<i>E. coli</i> O157:H7 IMS (Partial Confirmation)	ND/25mL	Detection rate IMS – 0%	Detection rate IMS – 0%
Big 6 STEC Screening	ND/25mL	Potential positive rate - 100%	Potential positive rate - 90%
Big 6 STEC IMS (Partial Confirmation)	ND/25mL	Potential positive rate IMS – 62%	Potential positive IMS – 80%
<b>a = Mean is simple arithmetic mean of detected results</b>			
<b>b = Exceedance rate defined as percentage &gt;100cfu/mL</b>			

Key points to note from the microbiological results for liquid waste/wastewater are:

- Thermotolerant coliforms and *E. coli* have been detected in both raw and finished product wastewater ready for dispatch. That implies there could be some further interventions required if the wastewater was to be used in conjunction with food production such as horticulture, such as the withholding periods that have already been included in industry guidelines. Detection rates were noted to increase in the ponds vs the raw state.
- There are exceedance levels set for *E. coli* for Class B and C irrigation water (Refer Appendix D – Victorian EPA Guidelines) at 100cfu/mL and 1000cfu/mL respectively there were two detections found above 100cfu/mL (i.e. Class B) and none exceeding 1000cfu/mL. There were no respondents in this survey reporting an approval for Class A.
- There were no detections for *Salmonella*, *Listeria monocytogenes*, *Listeria* spp, Coagulase positive *Staphylococcus* or *E. coli* O157
- There was one site where *Campylobacter* spp., was detected, in a settle pond at end point before dispatch.
- Big 6 STEC Screening had Potential Positive detections at a similar rate to Thermotolerant coliforms and its detection supported what had been identified in the literature review as a potential pathogen of concern

- Big 6 STEC IMS (part way confirmation to IMS and agglutination for Potential Positive detections to serotype stage) found there was likely to be some pathogens confirmed in wastewater at raw and exit points. See further points on STEC in section 4.4.3 below.

#### 4.4.2 Solid waste/manure/compost – by sector

Samples for solid waste were sought from (mostly) the “treated” or “finished” state as per the dispatch point from the respective enterprise. As highlighted in the previous section, there is a range of practices in place across feedlots, saleyards and processors. Therefore sample types were categorised according to the type of process and material in that process as listed above and the test data summarised below presented with the delineation shown according to the different types of material. As for liquids there are more “finished” product samples than “raw/unprocessed”.

Table 5 provides the overall microbiological test results all solid waste, all stages and all sectors combined.

Tables 6, 7, and 8 provide the micro test results for processors, feedlots and saleyards respectively.

**Table 5: Microbiological test results all solid waste, all stages & all sectors**

All Sectors	LOR – liquid waste	Solid Waste – All types, Raw (N = 32)	Solid Waste – All types, partial or fully processed (N = 94)
Thermotolerant Coliforms	10 cfu/g	Detection rate – 50% Mean <sup>a</sup> – 2042 cfu/g	Detection rate – 36% Mean <sup>a</sup> – 1094 cfu/g
<i>E. coli</i>	10 cfu/g	Detection rate – 22% Mean <sup>a</sup> – 1549 cfu/g Exceedance rate <sup>b</sup> – 19%	Detection rate – 17% Mean <sup>a</sup> – 840 cfu/g Exceedance rate <sup>b</sup> – 7%
<i>Salmonella</i> spp.	ND/25g	Detection rate – 0%	Detection rate – 0%
<i>Listeria monocytogenes</i>	ND/25g	Detection rate – 0%	Detection rate – 0%
<i>Listeria</i> spp.	ND/25g	Detection rate – 0%	Detection rate – 0%
<i>Campylobacter</i> spp.	ND/25g	Detection rate – 3%	Detection rate – 2%
CP staphylococci	100 cfu/g	Detection rate – 0% Mean – 0 cfu/g	Detection rate – 0% Mean – 0 cfu/g
<i>E. coli</i> O157:H7 IMS (Partial Confirmation)	ND/25g	Detection rate IMS – 0%	Detection rate IMS – 0%
Big 6 STEC IMS (Partial Confirmation)	ND/25g	Detection rate IMS – 28%	Detection rate IMS – 15%
<b>a = Mean is simple arithmetic mean of detected results</b>			
<b>b = Exceedance rate defined as percentage &gt;100cfu/g</b>			

**Table 6: Microbiological test results all solid waste, all stages – processing sector**

Processing Sector	LOR – liquid waste	Solid Waste – All types, Raw (N = 11)	Solid Waste – All types, partial or fully processed (N = 33)
Thermotolerant Coliforms	10 cfu/g	Detection rate – 9% Mean <sup>a</sup> – 50 cfu/g	Detection rate – 24% Mean <sup>a</sup> – 381 cfu/g
<i>E. coli</i>	10 cfu/g	Detection rate – 9%	Detection rate – 15%

		Mean <sup>a</sup> – 50 cfu/g Exceedance rate <sup>b</sup> – 0%	Mean <sup>a</sup> – 412 cfu/g Exceedance rate <sup>b</sup> – 9%
<i>Salmonella</i> spp.	ND/25g	Detection rate – 0%	Detection rate – 0%
<i>Listeria monocytogenes</i>	ND/25g	Detection rate – 0%	Detection rate – 0%
<i>Listeria</i> spp.	ND/25g	Detection rate – 0%	Detection rate – 0%
<i>Campylobacter</i> spp.	ND/25g	Detection rate – 9%	Detection rate – 3%
CP staphylococci	100 cfu/g	Detection rate – 0% Mean – 0 cfu/g	Detection rate – 0% Mean – 0 cfu/g
<i>E. coli</i> O157:H7 IMS (Partial Confirmation)	ND/25g	Detection rate IMS – 0%	Detection rate IMS – 0%
Big 6 STEC IMS (Partial Confirmation)	ND/25g	Detection rate IMS – 18%	Detection rate IMS – 21%
<b>a = Mean is simple arithmetic mean of detected results</b>			
<b>b = Exceedance rate defined as percentage &gt;100cfu/g</b>			

Table 7: Microbiological test results all solid waste, all stages – feedlot sector

Feedlot Sector	LOR – liquid waste	Solid Waste – All types, Raw (N = 5)	Solid Waste – All types, partial or fully processed (N = 37)
Thermotolerant Coliforms	10 cfu/g	Detection rate – 0% Mean <sup>a</sup> – 0 cfu/g	Detection rate – 11% Mean <sup>a</sup> – 44 cfu/g
<i>E. coli</i>	10 cfu/g	Detection rate – 0% Mean <sup>a</sup> – 0 cfu/g Exceedance rate <sup>b</sup> – 0%	Detection rate – 8% Mean <sup>a</sup> – 19 cfu/g Exceedance rate <sup>b</sup> – 0%
<i>Salmonella</i> spp.	ND/25g	Detection rate – 0%	Detection rate – 0%
<i>Listeria monocytogenes</i>	ND/25g	Detection rate – 0%	Detection rate – 0%
<i>Listeria</i> spp.	ND/25g	Detection rate – 0%	Detection rate – 0%
<i>Campylobacter</i> spp.	ND/25g	Detection rate – 0%	Detection rate – 3%
CP staphylococci	100 cfu/g	Detection rate – 0% Mean – 0 cfu/g	Detection rate – 0% Mean – 0 cfu/g
<i>E. coli</i> O157:H7 IMS (Partial Confirmation)	ND/25g	Detection rate IMS – 0%	Detection rate IMS – 0%
Big 6 STEC IMS (Partial Confirmation)	ND/25g	Detection rate IMS – 0%	Detection rate IMS – 3%
<b>a = Mean is simple arithmetic mean of detected results</b>			
<b>b = Exceedance rate defined as percentage &gt;100cfu/g</b>			

Table 8: Microbiological test results all solid waste, all stages – saleyard sector

Saleyard Sector	LOR – liquid waste	Solid Waste – All types, Raw (N = 16)	Solid Waste – All types, partial or fully processed (N = 24)
Thermotolerant Coliforms	10 cfu/g	Detection rate – 94% Mean <sup>a</sup> – 2175 cfu/g	Detection rate – 83% Mean <sup>a</sup> – 1098 cfu/g
<i>E. coli</i>	10 cfu/g	Detection rate – 38%	Detection rate – 33%

		Mean <sup>a</sup> – 1798 cfu/g Exceedance rate <sup>b</sup> – 38%	Mean <sup>a</sup> – 526 cfu/g Exceedance rate <sup>b</sup> – 17%
<i>Salmonella</i> spp.	ND/25g	Detection rate – 0%	Detection rate – 0%
<i>Listeria monocytogenes</i>	ND/25g	Detection rate – 0%	Detection rate – 0%
<i>Listeria</i> spp.	ND/25g	Detection rate – 0%	Detection rate – 0%
<i>Campylobacter</i> spp.	ND/25g	Detection rate – 0%	Detection rate – 3%
CP staphylococci	100 cfu/g	Detection rate – 0% Mean – 0 cfu/g	Detection rate – 0% Mean – 0 cfu/g
<i>E. coli</i> O157:H7 IMS (Partial Confirmation)	ND/Broth	Detection rate IMS – 0%	Detection rate IMS – 0%
Big 6 STEC IMS (Partial Confirmation)	ND/Broth	Detection rate IMS – 44%	Detection rate IMS – 25%
<b>a = Mean is simple arithmetic mean of detected results</b>			
<b>b = Exceedance rate defined as percentage &gt;100cfu/g</b>			

#### 4.4.3 STEC Detections – by type of effluent

The project collected samples from wastewater which was mostly in ponds in raw state or following the ponding process. A range of solid waste samples were collected, according to practices observed in the field – these included pond crust, sludge, dedicated downer stock compost rows, and dedicated and mixed paunch compost rows. Some raw/unprocessed samples were taken from a selection of sites where the solids were taken away from the site before any significant time was incurred for drying. Given that *E. coli* O157 and STEC are pathogens of interest to red meat and *E. coli* O157 in some fresh food sectors, additional breakdown of the *E. coli* O157 and STEC results to the partial/serotyping stage of confirmation are provided below. *E. coli* O157 was not detected after the potential positive stage in any samples.

There were 67 detections of Big 6 STEC to partial/serotyping stage of confirmation, mostly from the Saleyard Sector where composting is not widely practised to the extent of processors and feedlots. The O-types isolated were as follows:

- 55 x *E. coli* O26
- 6 x *E. coli* O45 (rarely, if ever, confirmed in meat samples)
- 6 x *E. coli* O103 (rarely, if ever, confirmed in meat samples)

Key points to note from the microbiological test results for solid waste:

- Thermotolerant coliforms and *E. coli* were detected in finished composted manure, paunch, combined manure/paunch ready for dispatch. The compost samples must contain *E. coli* <100 CFU/g and *Salmonella* Not Detected/25g to be considered treated in the Compliant Compost guidelines. The fact there have been *E. coli* detections at processor and saleyard sectors means there would need to be further interventions required if the compost was to be sold as “Compliant”. As mentioned above, it was reported for example that some of this material is going to macadamia growers in QLD and NSW, albeit mostly via intermediaries that are commercial specialist composters where further interventions may or may not be in place and effective.



- Thermotolerant coliforms and *E. coli* have been detected in raw material, which is not surprising. However, the prevalence and levels are higher over time in ponds.
- There was a detection for *Campylobacter* spp. in one sample of composted paunch/manure, however, the concentration was below the limit of reporting (<100cfu/g). There was one other detection of *Campylobacter* spp. at one site in raw material.
- There were no detections for *Salmonella*, *Listeria monocytogenes*, *Listeria* spp., Coagulase positive *Staphylococcus* or *E. coli* O157
- There were Big 6 STEC Screening Potential Positive detections for raw paunch, composted paunch/manure, composted manure, with further details in Table 9 above
- Big 6 STEC IMS has identify the presence of *E. coli* O26 in both raw and composted paunch/manure material.
- There may be greater risk associated with paunch material and in downer stock compost than manure only. If that is widespread then there are implications for the processing and feedlot sectors.

Given the findings about the extent of effluent material being sent to the commercial composters, it is recommended MLA/AMPC work with that sector to understand and evaluate the potential risks from that sector as part of the supply chain (of effluent treatment).

The project team also recommends engaging with state based environmental regulators as they are closely controlling all sectors and may change practices in each sector.

## 4 Conclusion and recommendations

### 4.1 Conclusions

The overall conclusions from this project with respect to microbiological quality on processed waste and effluent from the red meat supply chain were as follows:

- Based on current practices and test results for the participants in this project, the overall risks of transferring microbiological hazards from processed waste and effluent through the red meat supply chain appear relatively low
- Liquid waste - based on current practices and test results for the participants in this project, there is the potential for wastewater to carry microbial hazards further down the supply chain, albeit indirect transfers via other waterways are more likely than direct transfer to farms. The classification of raw vs irrigation water and approved uses by regulators is in place, thereby limiting direct use onto horticultural farms. There may still be circumstances where spaying or direct transfer of risk material occurs and therefore further investigation is warranted.
- Solid waste/compost - based on current practices and test results for the participants in this project, there is the potential for solid waste/compost to carry microbial hazards further down the supply chain. The definition and use of the term “composting” varies across industry players with feedlots undertaking systems that were the most in line with composting standards. There may still be circumstances where solid waste and non-compliant compost results in direct transfer of risk material to other industries and therefore further investigation is warranted.
- Regulations - The environmental protection agencies in each jurisdiction appear to control and manage major microbiological food safety related risks by using licensing requirements for the various categories of waste produced in each sector.

### 4.2 Recommendations

The following recommendations are made for future work based on the outcomes of this initial project:

1. Given that approximately one third of the processors, feedlots and saleyards send their manure, paunch and lairage waste off site to commercial composters, it is recommended that the practices of the commercial composting and nursery operators might be examined by businesses disposing of their waste in this way.
2. It is recommended that MLA considers conducting further research on pathogen prevalence in partnership with those composting organic waste – including the industry body/s representing composters and individual firms.
3. It is recommended that the results of this work are communicated to the horticulture sector to assist them to assess risks. It is recommended that MLA consult with several key sectors as well as inform the respective state Environmental Protection Agencies and State Health Departments about control measures in place. It is recommended that MLA maintain a watching brief on genomics and attribution studies being conducted by other parties e.g. State Health authorities, and industry groups such as Macadamia growers.

## 5 References

- Ashbolt, N. J., W. O. K. Grabow and M. Snozzi (2001). Indicators of microbial water quality. In World Health Organization, *Water Quality: Guidelines, Standards and Health*. IWA Publishing, London.
- Australian Standard AS 5013.10 (2009). Food microbiology. Method 10: Microbiology of food and animal feeding stuffs—Horizontal method for the detection of *Salmonella spp.* Standards Australia, Sydney, Australia.
- Australian Standard AS 5013.12.3 (2004). Food microbiology Method 12.3: Microbiology of food and animal feeding stuffs-Horizontal method for the enumeration of coagulase-positive staphylococci (*Staphylococcus aureus* and other species)-Detection and MPN technique for low numbers. Standards Australia, Sydney, Australia.
- Australian Standard AS 5013.15 (2006). Food Microbiology, Method 15: Microbiology of food and animal feeding stuffs—Horizontal method for the detection and enumeration of presumptive *Escherichia coli*—Most probable number technique. Standards Australia, Sydney, Australia.
- Australian Standard AS 5013.24.2 (2009). Food Microbiology. Method 24.2. Microbiology of food and animal feeding stuffs - Horizontal method for the detection and enumeration of *Listeria monocytogenes* - Enumeration method. Standards Australia, Sydney, Australia.
- Australian Standard AS/NZS 4276.20 (2004). Water microbiology. Method 20: Examination for coagulase positive staphylococci, including *Staphylococcus aureus*, by membrane filtration. Standards Australia, Sydney, Australia.
- Australian Standard AS/NZS 4276.7 (2007). Water Microbiology. Method 7: *Escherichia coli* and thermotolerant coliforms – Membrane filtration method. Standards Australia, Sydney, Australia.
- Blaiotta, G., A. Di Cerbo, N. Murru, R. Coppola, and M. Aponte (2016). Persistence of bacterial indicators and zoonotic pathogens in contaminated cattle wastes. *BMC Microbiology* (2016) 16:87.
- Bolton FJ, Coates D, Hutchinson DN, Godfree AF. (1987) A study of thermophilic campylobacters in a river system. *Journal of Applied Bacteriology*, 62: 167-76.
- Brichta-Harhay, D. M., T. M. Arthur, J. M. Bosilevac, M. N. Guerini, N. Kalchayanand and Carter AM, Pacha RE, Clark GW, Williams EA. (1987) Seasonal occurrence of *Campylobacter spp.* and their correlation with standard indicator bacteria. *Applied and Environmental Microbiology*, 53: 523-6.
- Cobbold R and Desmarchelier P. (2000). A longitudinal study of Shiga-toxigenic *Escherichia coli* (STEC) prevalence in three Australian dairy herds. *Veterinary Microbiology*. 71:125-137.
- Cox, P., M. Griffith, M. Angles, D. Deere, and C. Ferguson (2005). Concentrations of Pathogens and Indicators in Animal Feces in the Sydney Watershed. *Applied and Environmental Microbiology*. 71(10):5929-5924.
- Davis, J. A., S. R. Farrah and A. C. Wilkie (2006). Selective growth of *Staphylococcus aureus* from flushed dairy manure wastewater using acriflavine-supplemented mannitol salt agar. *Letters in Applied Microbiology* 42:606-611.

Dhamaa, K., K. Karthik, R. Tiwari, M. Shabbir, S. Barbuddhe, S. Malik and R. Singh (2015). Listeriosis in animals, its public health significance (food-borne zoonosis) and advances in diagnosis and control: a comprehensive review. *Veterinary Quarterly*, 34(4):211-235.

Djordjevic SP, Hornitzky MA, Bailey G, Gill P, Vanselow B, Walker K and Bettelheim KA. (2001). Virulence properties and serotypes of Shiga toxin-producing *Escherichia coli* from healthy Australian slaughter-age sheep. *J Clin Microbiol.* 39:2017-2021

Duffy, L. L., A. Small and N. Fegan (2010). Concentration and prevalence of *Escherichia coli* O157 and *Salmonella* serotypes in sheep during slaughter at two Australian abattoirs. *Australian Veterinary Journal*, 88(10): 399-404.

Fegan, N. and P. Desmarchelier (1999). Shiga toxin-producing *Escherichia coli* in sheep and preslaughter lambs in eastern Australia. *Letters in Applied Microbiology*, 28:335–339.

Fegan, N. G. Higgs, P. Vanderlinde, and P. Desmarchelier (2003). Enumeration of *Escherichia coli* O157 in cattle faeces using most probable number technique and automated immunomagnetic separation. *Letters in Applied Microbiology*, 38: 56-59.

Fegan, N., P. Vanderlinde, G. Higgs, and P. Desmarchelier (2004). Quantification and prevalence of *Salmonella* in beef cattle presenting at slaughter. *Journal of Applied Microbiology* 97(5): 892-898.

Fluit, A. C. (2012). Livestock-associated *Staphylococcus aureus*. *Clinical Microbiology and Infection*, 18:735:744.

Food Science Australia. (2000). STEC Ecology. A final report prepared for Meat & Livestock Australia, November 2000, Project TR.046: STR021.

Fukushima H, Hoshina K and Gomyoda M. (1999). Long-term survival of Shiga toxin-producing *Escherichia coli* O26, O111, and O157 in bovine faeces. *Applied and Environmental Microbiology*. 65:5177-5181.

Grau, F.H. (1988). *Campylobacter jejuni* and *Campylobacter hyointestinalis* in the intestinal tract and on the carcasses of calves and cattle. *Journal of Food Protection*, 51 (11): 857-861.

Henry, R., C. Schang, G. I. Chandrasena, A. Deletic, M. Edmunds, D. Jovanovic, P. Kolotelo, J. Schmid, R. Williamson, and David McCarthy (2015). Environmental monitoring of waterborne *Campylobacter*: evaluation of the Australian standard and a hybrid extraction-free MPN-PCR method. *Frontiers in Microbiology*, 6,74.

Hutchison, M. L., L. D. Walters, S. M. Avery, B. A. Synge, and A. Moore (2004). Levels of zoonotic agents in British livestock manures. *Letters in Applied Microbiology*, 39:207-214.

Hutchison, M. L., L. D. Walters, S. M. Avery, F. Munro, and A. Moore (2005). Analyses of Livestock Production, Waste Storage, and Pathogen Levels and Prevalences in Farm Manures. *Applied and Environmental Microbiology*, 71(3):1231–1236.

International Organisation for Standardisation (2005). ISO 17995:2005, Detection and enumeration of thermotolerant *Campylobacter* spp. Geneva, Switzerland: ISO.

International Organisation for Standardisation (2006). ISO 10272-1:2006, Microbiology of Food and Animal Feeding Stuffs – Horizontal Method for detection and Enumeration of *Campylobacter* spp . Geneva, Switzerland: ISO.

International Organisation for Standardisation. ISO/DIS 6888-1 (1999). Microbiology of the food chain — Horizontal method for the enumeration of coagulase-positive staphylococci (*Staphylococcus aureus* and other species) - Part 1: Technique using Baird-Parker agar medium. Geneva, Switzerland: ISO.

International Organization for Standardization (2002) ISO 6579:2002. Microbiology of food and animal feed-Horizontal method for the detection, enumeration and serotyping of *Salmonella*-Part 2: Enumeration by a miniaturized most probable number technique. Geneva, Switzerland: ISO.

Jain, P., D. Cunningham, J. Chesson and S. Fabiansson (2003). A quantitative risk assessment of microbial emissions from abattoirs. Final Report PRMS.036, Meat and Livestock Australia, Sydney, Australia.

Jiang, X., M. Islam, J. Morgan, and M. P. Doyle (2004). Fate of *Listeria monocytogenes* in Bovine Manure-Amended Soil. *Journal of Food Protection*, 67(8):1676-1681.

Kirby, M. E., M. W. Mirza, T. Leigh, L. Oldershaw, M. Reilly, and S. Jeffery (2019). Destruction of *Staphylococcus aureus* and the impact of chlortetracycline on biomethane production during anaerobic digestion of chicken manure. *Heliyon*, 5:e02749.

Koohmaraie, M. (2007). Enumeration of *Salmonella* and *Escherichia coli* O157:H7 in ground beef, cattle carcass, hide and faecal samples using direct plating methods. *Journal of Applied Microbiology* 103: 1657-1668.

Korajkic, A., B. R. McMinn, and V. J. Harwood (2018). Relationships between Microbial Indicators and Pathogens in Recreational Water Settings. *International Journal of Environmental Research and Public Health*, 15: 2842.

Luedtke, B. E., and J. M. Bosilevac (2015). Comparison of methods for the enumeration of enterohemorrhagic *Escherichia coli* from veal hides and carcasses. *Frontiers in Microbiology*, 6:1062.

McMahon, W. A., V. A. Aleo, A. M. Schultz, B. L. Horter, and K. G. Lindberg (2003). 3M Petrifilm Staph Express Count Plate Method for the Enumeration of *Staphylococcus aureus* in Selected Types of Meat, Seafood, and Poultry: Collaborative Study. *Journal of AOAC International*, 86(5):947-953.

Mellor, G., N. Fegan, L. Duffy, K. McMillan, D. Jordan, and R. Barlow (2015). Enumeration of pathogenic Shiga toxin-producing *Escherichia coli* in Australian beef cattle feces at slaughter. In: VTEC 2015 Boston, 13-16 September 2015; Boston, USA.

Midgley J and Desmarchelier P. (2001). Pre-slaughter handling of cattle and Shiga toxin-producing *Escherichia coli* (STEC). *Lett Appl Microbiol*. 32: 307-311.

Munck N et al, (2019). Source Attribution of *Salmonella* in Macadamia Nuts to Animal and Environmental Reservoirs in Queensland, Australia. *FOODBORNE PATHOGENS AND DISEASE*.

Odonkor, S. T. and J. K. Ampofo (2013). *Escherichia coli* as an indicator of bacteriological quality of water: an overview. *Microbiology Research*, 4:5-11.

Ogden ID, Hepburn NF, MacRae M, Strachan NJC, Fenlon DR, Rusbridge SM and Pennington TH. (2002). Long-term survival of *Escherichia coli* O157 on pasture following an outbreak associated with sheep at a scout camp. *Letters in Applied Microbiology*. 34: 100-104.

- Patriarchi, A., B. Maunsell, E. O'Mahony, A. Fox, S. Fanning, J. Buckley and D. J. Bolton (2009). Prevalence of *Campylobacter* spp. in a subset of intensive poultry flocks in Ireland. *Letters in Applied Microbiology*, 49:305-310.
- Pavic, A., P. J. Groves, G. Bailey, and J. M. Cox (2010). A validated miniaturized MPN method, based on ISO6579:2002, for the enumeration of *Salmonella* from poultry matrices. *Journal of Applied Microbiology*, 109:25-34.
- Premier, R. (2010). Reducing *Listeria* contamination from salad vegetable farms. Horticulture Australia Ltd, Sydney, Australia.
- Public Health England (2018). Detection and enumeration of *Listeria monocytogenes* and other *Listeria* species. National Infection Service. Food, Water and Environmental Microbiology Methods Working Group. PHE Publications, London, England.
- Roberts, M. C., G. Garland-Lewis, S. Trufan, S. J. Meschke, H. Fowler, R. C. Shean, A. L. Greninger, and P. M. Rabinowitz (2018). Distribution of *Staphylococcus* species in dairy cows, workers and shared farm environments. *FEMS Microbiology Letters*, 365(15).
- Samarajeewa, A. D., S.M. Glasauer and K.E. Dunfield (2010). Evaluation of Petrifilm EC method for enumeration of *E. coli* from soil. *Letters in Applied Microbiology*, 50: 457-461.
- Scallan, E., R. M. Hoekstra, F. J. Angulo, R. V. Tauxe, M. Widdowson, S. L. Roy, J. L. Jones, and P. M. Griffin (2011). Foodborne Illness Acquired in the United States—Major Pathogens. *Emerging Infectious Diseases*, 17(1):7-15.
- Shepherd, M. A., V. M. Fleming, T. R. Connor, J. Corander, E. J. Feil, C. Fraser, and W. P. Hanage (2013). Historical Zoonoses and Other Changes in Host Tropism of *Staphylococcus aureus*, Identified by Phylogenetic Analysis of a Population Dataset.
- Sinton, L. W., R. R. Braithwaite, C. H. Hall, and M. L. Mackenzie (2007). Survival of Indicator and Pathogenic Bacteria in Bovine Feces on Pasture. *Applied and Environmental Microbiology*, 73(24):7917-7925.
- Smith, T. C. (2015). Livestock-associated *Staphylococcus aureus*: The United States Experience. *PLoS Pathog* 11(2):e1004564.
- Sobsey, M. D., L. A. Khatib, V. R. Hill, E. Alocilja, and S. Pillai (2006). Pathogens in Animal Wastes and the Impacts of Waste Management Practices on their Survival, Transport and Fate. In *Animal Agriculture and the Environment: National Centre for Manure and Animal Waste Management White Papers*, pp 609-665. St. Joseph, Michigan.
- Sutherland, P. and Porritt, R. (1997). *Listeria monocytogenes*. In: *Foodborne microorganisms of public health significance*. AIFST, Sydney, 1997.

## 6 Appendix

### 6.1 Appendix A – Literature Review

#### 6.1.1 Hazard identification

Many zoonotic agents can be transmitted from animals to humans, including viruses, parasites, and bacteria (Sobsey et. al., 2006). While most viruses are considered host specific, some are or maybe capable of infecting humans

Most parasites associated with animal waste are considered waterborne zoonoses (e.g. Cryptosporidium and Giardia; Sobsey et. al. 2006) and only pose a risk where humans come in contact with contaminated water or where contaminated water is used in the processing of ready-to-eat foods such as horticulture products. An exception to this is *Toxoplasma gondii*. This parasite is commonly found in pigs, although other domestic animals such as sheep and goats are also known to be infected. In 2011, *T. gondii* was the second highest cause of foodborne deaths in the US (Scallan et. al., 2011). Human exposure is generally from consumption of infected undercooked meat, with transmission via animal wastes unlikely (Sobsey et. al. 2006). This review will focus on bacterial pathogens found in animal waste that are associated with foodborne disease.

A risk assessment of the impact of zoonotic diseases on human health (Jain et. al., 2003) identified five bacterial pathogens as being important to the Australian red-meat industry: *Campylobacter jejuni*, *Coxiella burnetii*, *Escherichia coli* (specific serotypes), *Listeria monocytogenes*, and *Salmonella* spp. It should be noted that this work did not consider foodborne routes of transmission. *Coxiella burnetii* is not considered to be a foodborne pathogen with airborne transmission the most likely route of infection.

A study of British livestock manures (Hutchison et. al., 2003) quantified levels of four bacterial pathogens most associated with foodborne illness: *Salmonella* spp., *Campylobacter*, *E. coli* O157, and *Listeria*. Interestingly, both studies did not consider *Staphylococcus aureus* as an important pathogen in animal waste streams, perhaps based on the long-held belief that animal strains are not normally associated with human disease (Fluit, 2012; Shephard et. al., 2013; Smith, 2015). There are suggestions (Shephard et. al., 2013; Smith, 2015) that animal strains may contribute to human disease, particularly in relation MRSA, either through direct exposure or gene transfer in the host. Contrastingly, Roberts (2018) found no genetically related isolates among worker, animal, and environmental samples on dairy farms in the US. Without further study the role of animal strains in human disease remains unclear. Inclusion of *Staphylococcus aureus* in animal waste studies should be considered with caution as true animal strains may complicate any inference drawn between animal waste and human disease. A more risk-based approach would be to examine antimicrobial resistant *Staphylococcus aureus* in waste streams.

Thermotolerant coliforms and *E. coli* are often used as indicators of faecal contamination although they are not always reliable indicators of the presence of bacterial pathogens (Korajkic et. al., 2018). Further, not all thermotolerant coliforms are of faecal origin (Obonkor et. al. 2013). *E. coli* is the most appropriate group of coliforms for the identification of faecal pollution (Ashbolt et. al., 2001). As the concentration of *E. coli* in animal effluent is expected to be high (Blaiotta et. al., 2016; Sinton et. al., 2007; Cox et. al., 2005) it is the preferred indicator organism for inclusion in surveys of the microbiological contamination of animal effluent.

It is proposed that the following bacterial hazards be included in the survey of effluent from red-meat animal industries:

- *E. coli* (generic)
- *Salmonella* spp.
- *Listeria monocytogenes*
- *Campylobacter* spp.
- Coagulase positive *Staphylococcus* and STEC have been included for completeness, however, quantification of these hazards is not recommended as being essential.

### 6.1.2 Quantitative methods for the isolation of bacteria from animal waste

#### *Generic E. coli*

Enumeration of *E. coli* will be dependent on the physiological state of the cells in individual waste streams. Plating techniques, such as Petrifilm, have been used for the enumeration of *E. coli* in animal faeces in previous studies (Fegan et. al., 2003 and MLA, 2007), although Samarajeewa et. al. (2010) found slightly lower counts on Petrifilm compared to a traditional membrane filtration technique when enumerating *E. coli* in manure treated soil samples. However, the difference was not enough to discount the use of Petrifilm for enumeration of all solid or semi-solid effluent streams for generic *E. coli*.

For liquid streams, a traditional membrane filtration technique is recommended (Samarajeewa, 2010; Hutchison, 2004; AS/NZS 4276.7, 2007). If there is a need to quantify thermotolerant coliforms, then an MPN enrichment procedure (AS 5013.15, 2006) or membrane filtration technique (AS/NZS 4276.7, 2007) should be considered. *E. coli* should be quantified in conjunction with thermotolerant coliforms if the latter is included in the study.

#### *STEC*

The environment can become contaminated with STEC allowing transmission of the bacteria within and between animal groups. STEC have been shown to be present in soil and water samples from Australian cattle farms and feedlots (Cobbold and Desmarchelier, 2000; Midgley and Desmarchelier, 2001) and can survive in manure and on pasture for several months (Fukushima et al., 1999; Ogden et al., 2002). Sheep have also been shown to carry STEC in their faeces (Djordjevic et al., 2001). STEC survival in faeces is affected by temperature, with STEC detected for longer periods in faeces stored at lower temperatures e.g. 10°C and 25°C, than at higher temperature e.g. 37°C (Food Science Australia, 2000: STR021).

Only a small number of STEC are of interest in human disease, therefore quantification would need to be selective, if conducted at all. Enumeration of specific serotypes in faecal and environmental samples is complicated and expensive requiring a combination of immunocapture, PCR and MPN techniques (Luedtke, 2015; Mellor, 2015). As it is already known that STEC are likely to occur in waste streams and given that generic *E. coli* levels are an indicator of survival of pathogenic strains, quantification of STEC is not recommended at this time.

#### *Salmonella*

*Salmonella* spp. are commonly found in cattle and sheep at slaughter (Duffy, 2010; Fegan, 2003) and have been isolated from fresh and stored manures (Hutchison, 2004). Hutchison et. al. (2003) used a membrane filtration technique to enumerate pathogens, including *Salmonella*, in faecal samples (samples requiring several clean-up steps prior to filtration). A disadvantage of direct plating



techniques is that the limit of quantification can be high, around 100 CFU/g (Brichta-Harhay, 2007). Others (Duffy, 2010; Fegan, 2005) have used immunocapture in combination with PCR and an MPN technique to enumerate *Salmonella* in faeces.

While direct plating is potentially more accurate than MPN techniques, clean-up of samples and confirmation from plates can be problematic in heavily contaminated samples. However, for relatively clean samples such as treated water, membrane filtration techniques may be more appropriate (see Hutchison et. al., 2004), such techniques should be coupled with presence/absence tests to define contamination of water samples more accurately. Traditionally, standard enrichment protocols in conjunction with MPN (see ISO/TS 6579-2, 2012) have been used for the enumeration of pathogens from heavily contaminated samples.

A similar method was used in an Australian study to enumerate *Salmonella* in poultry samples (Pavic et. al., 2010). The best approach for heavily contaminated samples such as manures, appears to be enrichment (either traditional or using IMS) followed by an MPN/PCR technique. Such an approach could also be used for water samples if this makes laboratory analysis more streamlined.

Confirmation of *Salmonella* spp. should be by the Australian standard AS 5013.10.

### *Listeria*

*Listeria* has been isolated from a wide range of mammals, birds, and insects (Sutherland et al 1997) and are truly ubiquitous in nature (Dhama, 2015). They are associated with animal illness including mastitis, septicaemia and abortion. Feed including silage can be a source of *Listeria* on farm. *Listeria* spp. are commonly found in animal faeces and the farm environment including manure, sludge and water (Dahama, 2015). It is not clear what role animal waste plays in the ecology of listeriosis as most disease is associated with consumption of ready-to-eat foods that have been contaminated in the post processing environment. Given its ubiquitous nature detection in waste streams is likely unavoidable and it is not clear how its quantification will help inform industry practices.

*Listeria* spp. have been quantified in animal manures using membrane filtration techniques (Hutchison, 2004) and have been shown to survive in faeces (Hutchison, 2005). Few studies could be found where enumeration of *Listeria* spp. in faecal samples was undertaken. Jiang et. al. (2004) used a direct plating technique to enumerate *Listeria* in artificially contaminated soil/manure samples. Most standard enumeration techniques rely on direct plating of diluted samples onto *Listeria* selective media (AS 5013.24.2, 2009). This technique is appropriate for food and animal feed samples but may not be suitable for heavily contaminated samples such as faeces.

An Australia study on horticulture products appeared to use standard methods for the detection and enumeration of *Listeria* from manure and water samples without any issues (Premier, 2010). A similar UK standard (Public Health England, 2018) has an expanded scope to include enumeration of environmental samples. Hutchison (2004) describes a membrane filtration/direct plating technique for the enumeration of *Listeria* in environmental samples.

It is recommended that water samples are analysed by membrane filtration followed by plating onto ALOA as per the Australian standard. A resuscitation step may be appropriate (Hutchison et. al., 2004). Traditional enrichment (with or without a PCR step), coupled with MPN, can be use and maybe the easiest method for heavily contaminated samples such as manures.

### *Campylobacter*

*Campylobacter* can be found in natural water sources, presumably through faecal contamination, with survival being greatest during colder months (Carter et al, 1987; Bolton et al, 1987).

*Campylobacter* has also been isolated from the intestinal tract of cattle and calves at slaughter, with younger animals more at risk of being contaminated (Grau, 1988).

Enumeration is complicated by the fragile state *Campylobacter* assumes in response to stress and the lengths that need to be taken to protect the organism during enumeration and recovery. Grau (1988) used direct plating onto selective media for enumeration of *Campylobacter* in cattle and calf faeces and rumen samples. Patriarchi et. al. (2009) used direct plating (ISO 10272, 2006) and a semi-quantitative membrane filtration technique (ISO 17995, 2005) for detection of *Campylobacter* in poultry faecal and water samples, respectively.

An Australian study of ground water systems in Melbourne (Henry et. al, 2015) used a membrane filtration technique coupled with MPN and the Australian Standard to estimate numbers of *Campylobacter*. Hutchison (2004), describes a membrane/direct plating technique for the enumeration of *Campylobacter* from environmental samples. In many studies direct plating techniques are preferred as they are more rapid and less laborious than MPN techniques.

### *Staphylococcus aureus*

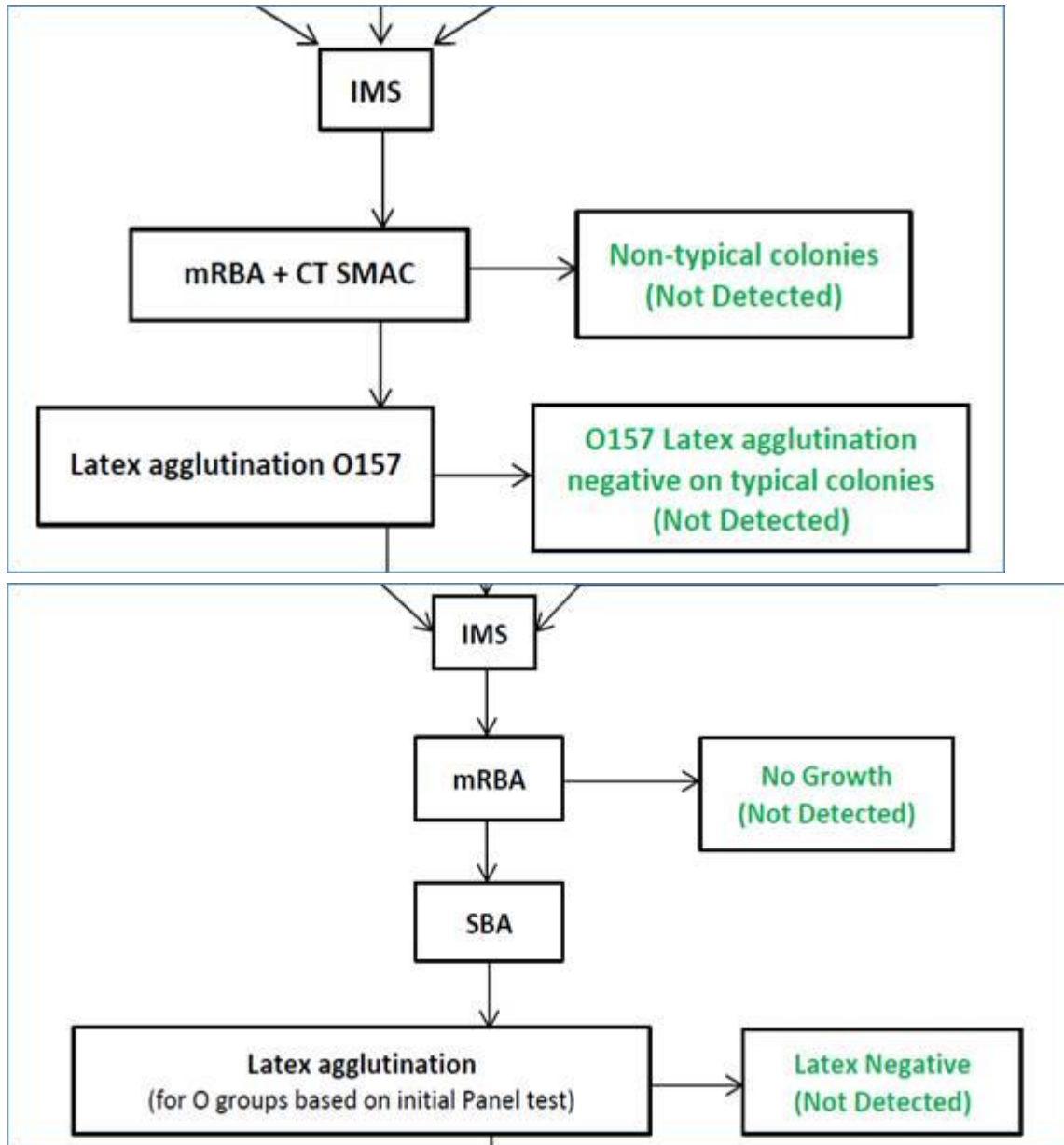
It is likely that most *S. aureus* found in red-meat industry waste streams are of animal origin making their significance in relation to human disease questionable (Smith, 2015). Most studies shown that cross species transmission is rare (Shepherd, 2013), however emergence of antibiotic resistant strains does appear to pose a risk to human health.

*Staphylococcus aureus* are generally enumerated using Baird Parker media, although there appears to be a lack of studies enumerating *Staphylococcus* in animal waste. Davis et. al. (2006) proposed a modified mannitol salt agar (MSA) for the enumeration of *Staphylococcus aureus* in manure wastewater. Mannitol salt agar alone has been used for the enumeration of *S. aureus* in manure (Kirby et. al, 2019). While dehydrated films such as Petrifilm Staph Express Count have been validated for the enumeration of *S. aureus* in foods (McMahon et. al., 2003), it is not clear how they would perform with heavily contaminated samples.

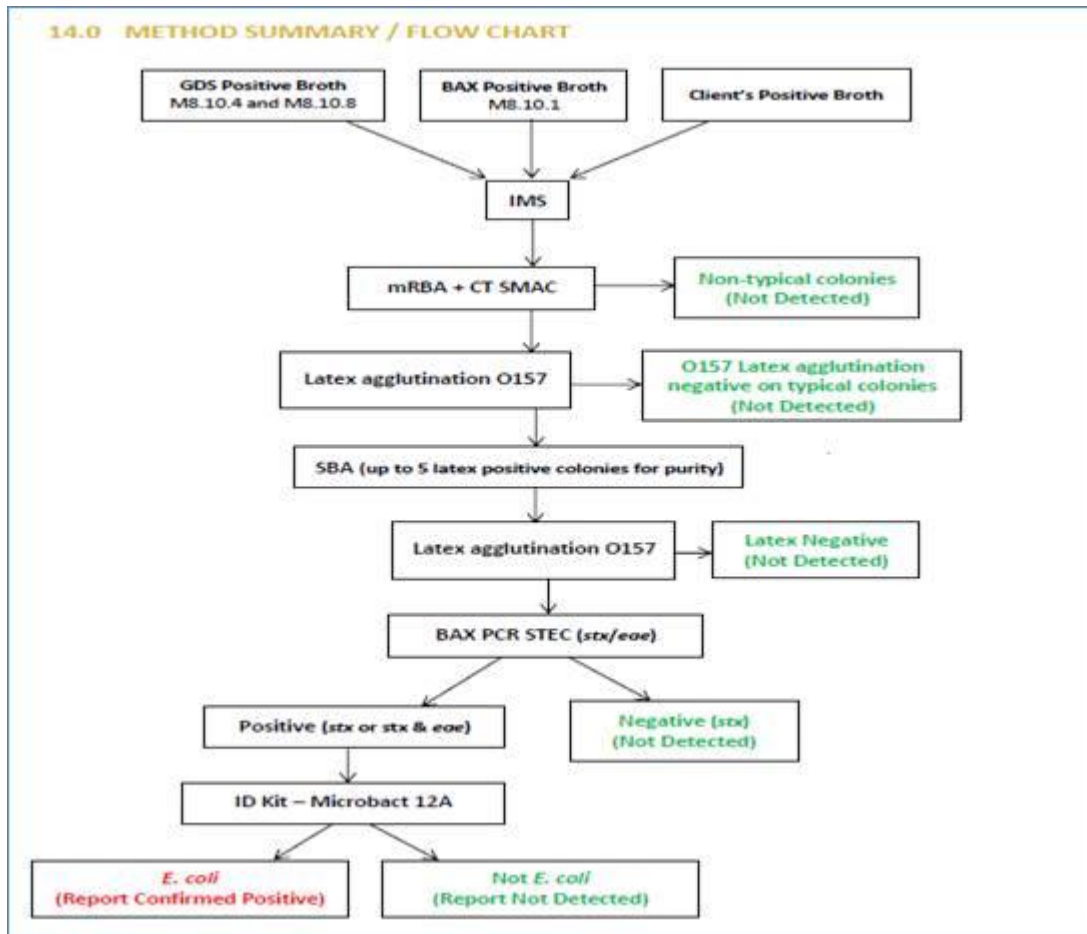
Direct plating onto Baird-Parker medium is still the preferred method of enumeration from food and environmental samples (ISO/DIS 6888-1, 1999). AS 5013.12.3 (2004) describes an MPN procedure for the enumeration of *S. aureus* in environmental samples that may be appropriate when low numbers of *S. aureus* are present in samples. Water samples can be analysed using membrane filtration followed by direct plating onto Baird-Parker media (AS/NZS 4276.20, 2003).

## 6.2 Appendix B – Approach to confirmation of E.coli O157 and Big 6 STEC

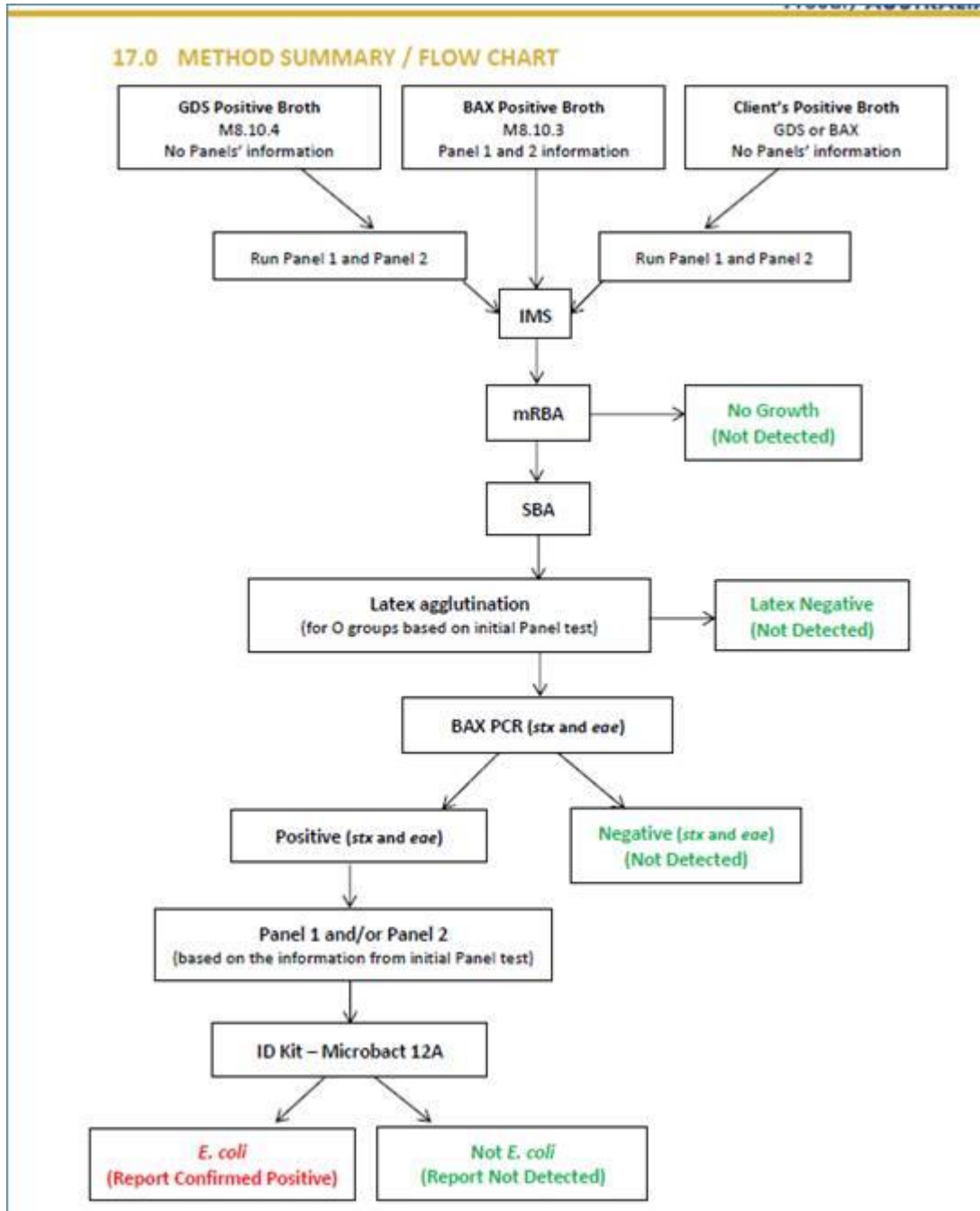
The approach used for “confirmation” of E.coli O157 and Big 6 STEC was an abbreviated method stopping at latex agglutination. At that stage of serology we know serotype. Flow charts for the full confirmation of E.coli O157 and Big 6 STEC are included below.



Full Confirmation method e.g. E.coli O157



Full Confirmation method for STEC (Big 6)



## 6.3 Appendix C – Questionnaires used for survey of practices

### Survey of Practices – Microbiological Risks from Waste Material

#### Background

Livestock are a known source of foodborne pathogens that could unintentionally be directly (waste, manures) or indirectly (air, dust, water runoff, water source, pests and wildlife) transferred to product within the supply chain and present a food safety risk.

MLA/AMPC is conducting a project to examine the microbiological quality of processed waste and effluent, ranging from farms to processing establishments, to provide clarity and identify potential risks which may not been previously considered. Part of the project involves mapping the waste and effluent streams from farm to processors. Of particular interest are control measures in place for microbiological hazards and whether they are seen as effective.

Symbio Laboratories is conducting this part of the project.

Your assistance with this part of the project is greatly appreciated.

#### Approach to questions

Below is a summary table of key topics being examined. It is intended to be an open rather than closed survey, so feel free to make comments seen as relevant. The topics are just a guide and we might follow up to clarify or expand on points as needed.

#### General Information

Company Name:	
Person Responding:	
Contact phone no.:	
Location for this enterprise (town & post code):	
How close is the enterprise to the nearest town?	
Are there any farms within 5km (regardless of discharging to them or not? YES/NO. If yes, what type of farms are they?	

#### Testing

Would you be willing to send samples for bacterial testing (note: project will pay for sampling vessel, shipment and testing? YES/NO

#### Return of response

Please send your response to Mr Peter Horchner at [phorchner@symbiolabs.com.au](mailto:phorchner@symbiolabs.com.au) . Contact phone number is 0407 877 094.

## Site/Enterprise Specific Topics

Type of waste product	Which of the following does your site have? YES/NO	If YES, please outline what happens with the waste?	Control Measures – if YES, what steps are in place (natural or introduced controls measures) to either eliminate or reduce the risk of harmful microbes entering the food chain?	Are there any checking (verification) on the effectiveness of control measures? Including any testing or measurements? If YES, please describe
1. Wastewater	<u>Raw</u> discharge to a river/water course YES/NO			
	<u>Raw</u> discharge to fields &/or farms YES/NO	e.g sprayed or flood irrigated on crops such as grains; fruit or vegetables or other ready-to-eat crops? Or non-agricultural land?		
	<u>Raw</u> discharge – other YES/NO			
	<u>Treated</u> discharge YES/NO	e.g. type of treatment dissolved and chemical air flotation, aeration, clarifiers, chemically dosed saveall, and anaerobic digestion		
	Re-use on site	e.g. stock washing, cleaning yards & lairage or other?		
2. Solids	Manure & lairage/ yard wastes	e.g. Untreated, treated, sent for composting ( <b><i>see below for composting</i></b> ), composted on site, sent for landfill?		
	Paunch	e.g. Dewatered or Un-dewatered? sent for composting, composted on site, sent for landfill?		
	Pond crusts from wastewater treatment	e.g. Untreated, treated, sent for composting, composted on site, sent for landfill?		

Type of waste product	Which of the following does your site have? YES/NO	If YES, please outline what happens with the waste?	Control Measures – if YES, what steps are in place (natural or introduced controls measures) to either eliminate or reduce the risk of harmful microbes entering the food chain?	Are there any checking (verification) on the effectiveness of control measures? Including any testing or measurements? If YES, please describe
3. Sludge & Slurry (if different from the above)				
4. Transporting off site material	Use external providers? YES/NO			

### Composting

Where Composting is undertaken on site...

1. Is there a treatment process for the compost? If YES, what is the overall process? E.g. In vessel? Windrows?	
2. What is the total time period for the compost to settle/stand i.e. for a given batch?	
3. How frequently is it turned over/aerated? And for How many occasions in its life i.e. in that batch on site?	
4. Is there an accreditation or standard that it complies with e.g. AS4454-2012?	
5. Is there any time/temperature monitoring of the compost? What is the target temperature & time to be achieved?	
6. Are there batch controls for separate batches? How does this work? Are new/raw batches separated from treated?	
7. Are there treatment records kept?	
8. Is there calibration of time temperature measuring equipment?	
9. Are there any lab tests done on the compost? If YES how are the samples derived/batched? And What tests are undertaken?	
10 Where is the finished product sent afterwards? E.g. to farms, commercial composters, landfill etc.	



## 6.4 Appendix D – Victoria EPA “Guidelines For Environmental Management Use Of Reclaimed Water”, Table 1

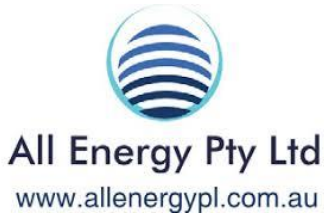
**Table 1. Classes of reclaimed water and corresponding standards for biological treatment and pathogen reduction. (Acceptable uses are detailed in Table 3)**

<b>Class</b>	<b>Water quality objectives - medians unless specified<sup>1,2</sup></b>	<b>Treatment processes<sup>a</sup></b>	<b>Range of uses- uses include all lower class uses</b>
<b>A</b>	Indicative objectives <ul style="list-style-type: none"> <li>• &lt; 10 <i>E.coli</i> org/100 mL</li> <li>• Turbidity &lt; 2 NTU<sup>4</sup></li> <li>• &lt; 10 / 5 mg/L BOD / SS</li> <li>• pH 6 – 9<sup>5</sup></li> <li>• 1 mg/L Cl<sub>2</sub> residual (or equivalent disinfection)<sup>6</sup></li> </ul>	Tertiary and pathogen reduction <sup>7</sup> with sufficient log reductions to achieve: <ul style="list-style-type: none"> <li>&lt;10 <i>E.coli</i> per 100 mL;</li> <li>&lt;1 helminth per litre;</li> <li>&lt; 1 protozoa per 50 litres; &amp;</li> <li>&lt; 1 virus per 50 litres.</li> </ul>	<u>Urban (non-potable):</u> with uncontrolled public access <u>Agricultural:</u> eg human food crops consumed raw <u>Industrial:</u> open systems with worker exposure potential
<b>B</b>	<ul style="list-style-type: none"> <li>• &lt;100 <i>E.coli</i> org/100 mL</li> <li>• pH 6 – 9<sup>5</sup></li> <li>• &lt; 20 / 30 mg/L BOD / SS<sup>8</sup></li> </ul>	Secondary and pathogen (including helminth reduction for cattle grazing) reduction <sup>7</sup>	<u>Agricultural:</u> eg dairy cattle grazing <u>Industrial:</u> eg washdown water
<b>C</b>	<ul style="list-style-type: none"> <li>• &lt;1000 <i>E.coli</i> org/100 mL</li> <li>• pH 6 – 9<sup>5</sup></li> <li>• &lt; 20 / 30 mg/L BOD / SS<sup>8</sup></li> </ul>	Secondary and pathogen reduction <sup>7</sup> (including helminth reduction for cattle grazing use schemes)	<u>Urban (non-potable)</u> with controlled public access <u>Agricultural:</u> eg human food crops cooked/processed, grazing/fodder for livestock <u>Industrial:</u> systems with no potential worker exposure
<b>D</b>	<ul style="list-style-type: none"> <li>• &lt;10000 <i>E.coli</i> org/100 mL</li> <li>• pH 6 – 9<sup>5</sup></li> <li>• &lt; 20 / 30 mg/L BOD / SS<sup>8</sup></li> </ul>	Secondary	<u>Agricultural:</u> non-food crops including instant turf, woodlots, flowers

**Notes to Table 1**

1. Medians to be determined over a 12-month period. Refer table 6 for Notification / reclassification limits.
2. Refer also to Chapter 6 and7, and Waste Water Irrigation Guideline (EPA Victoria, 1991 Publication 168) for additional guidance on water quality criteria and controls for salts, nutrients and toxicants.

## 6.5 Appendix E - Effluent mapping from animal industries



# milestone report

Project code: V.MSF.0448 MS 1  
Prepared by: Max Barnes, Dr Gareth Forde  
All Energy Pty Ltd  
Date published: April 2022

#### PUBLISHED BY

Meat and Livestock Australia Limited  
Locked Bag 1961  
NORTH SYDNEY NSW 2059

## **Effluent mapping from animal industries: farm, sale yard, feedlot, processing plant - Microbiological food safety of effluent from animal industries.**

Meat & Livestock Australia acknowledges the matching funds provided by the Australian Government to support the research and development detailed in this publication.

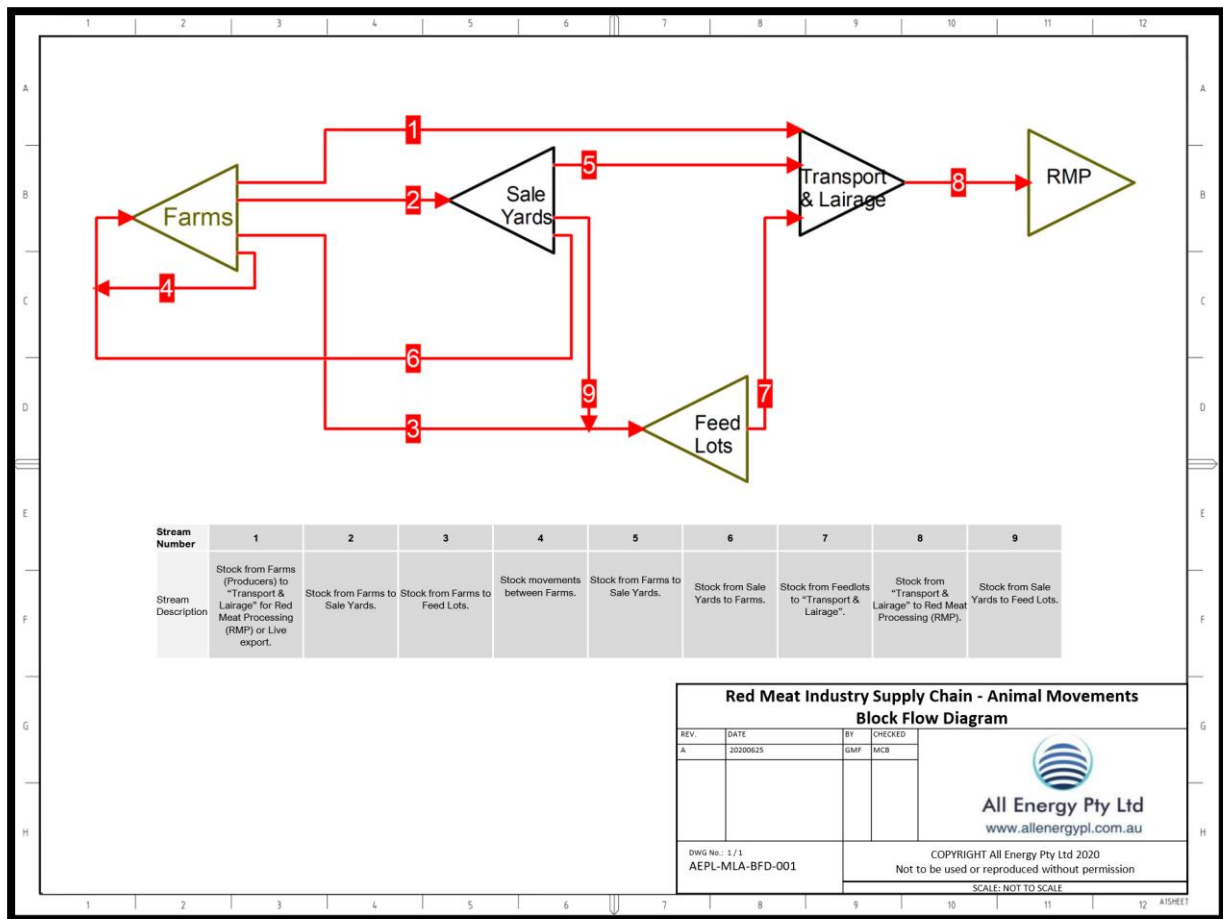
This publication is published by Meat & Livestock Australia Limited ABN 39 081 678 364 (MLA). Care is taken to ensure the accuracy of the information contained in this publication. However MLA cannot accept responsibility for the accuracy or completeness of the information or opinions contained in the publication. You should make your own enquiries before making decisions concerning your interests. Reproduction in whole or in part of this publication is prohibited without prior written consent of MLA.

Abstract

The red meat supply chain is complicated as numerous parties with a wide array of business models operating in all types of physical and geographic environments are involved.

This milestone maps the waste and effluent streams in the red meat industry from farm to processor with identification of value adding opportunities. This is to contribute to a survey of microbiological quality on processed waste and effluent, ranging from farms to processing establishments, to provide clarity and identify potential risks which may not have been previously considered.

The following flow diagram shows the movement of animals (lines) to the main nodes of producers (Farm) to feedlots (FL), red meat processors (RMP), sale yards (SL) with Transport and Lairage including live exports.



**Figure 1:** Block flow diagram of principal livestock movements throughout the red meat industry supply chain. RMP stands for Red Meat Processor; Transport and Lairage includes live export activities.

The key considerations were the different waste streams created at each node, composition, total tonnages, and potential for increased risk / avoidance of food safety risks (i.e. uses; changes) so that the right sampling points can be selected. Compartmentalising each stream to consider best practice through to worst practice.

A literature review suggests that this is first attempt to undertake a mapping of key organic waste streams across the entire red meat industry supply chain including beef and sheep. Hence, a wide array of reference types have been utilised and an associated colour coding used to define the type of reference and to highlight where data gaps currently exist, with an approximate hierarchy from higher to lower quality data inputs:

Primary reference – directly reported
Primary reference – calculated with second primary reference
Primary reference – calculated with assumption
Assumption – certain / logical progression
Assumption – uncertain / TBC
Requires clarification during survey
White – Calculated result based upon other source data

### BEEF – Estimated annual tonnages of key organic streams:

Stream	kg per head per day	Indicative Facility Standing Head	Operational Days pa	Indicative Facility Tonnes pa	Standing Head - Australia	Tonnes pa - Australia	Solids %
Farm-Manure	1.76 – comprised of 0.62 kg solid faeces and 1.14 kg urine	5,000	365	3,212	24,800,000	15,942,857	35%
SY-Slurry	390 - 430	5,458	50	111,889	4,559,807 consignments in 2018-2019	1,869,521	0.5%
SY-Manure	1.76 – as above	5,458	50	480	4,559,807	8,025	35%
FL-Slurry	Highly weather, seasonal & operationally dependant.  Estimate 11.74 kg/head/day	20,000	365	85,722	1,239,563	5,330,121	0.5%
FL-Manure	3.79	20,000	365	75,800	1,239,563	4,697,944	35% (26% as pen scrapings)
RMP – Dissolved Air Float (DAF)	16.5	1000 hpd	250	4,125	6,900,000 head slaughtered per annum	113,850	7%
RMP – Paunch	26	1000 hpd	250	6500	6,900,000	179,400	34%
RMP – Waste Activated Sludge (WAS)	26.3	1000 hpd	250	6575	6,900,000	181,470	12.5%
RMP – green screenings	2.33	1000 hpd	250	583	6,900,000	16,091	20%
RMP – red screenings	3.2	1000 hpd	250	800	6,900,000	22,080	20%
RMP – Plastic	1.63	1000 hpd	250	408	6,900,000	11,261	98%
RMP – Cardboard	1.2	1000 hpd	250	300	6,900,000	8,280	90%
RMP – Anaerobic Digestion (AD) Digestate	0.37	1000 hpd	250	23,441	Unknown number of AD facilities in Australian processing plants encompassing open ponds, crusted ponds, and covered anaerobic lagoons with a wide range of solids handling procedures*.		5%

\* Solids handling procedures include no digestate management (i.e. ponds are allowed to silt up), periodic solids removal (e.g. every 4 – 5 years) to maintain lagoon operational volumes to continuous recovery and dewatering with the destination of the slurry / solids including stockpiles, application to adjacent land and removal off-site.

**SHEEP – Estimated annual tonnages of key organic streams:**

There are significant gaps in the literature regarding sheep in saleyards, feedlots, and processing and associated waste production. Before receiving data during the survey these will be approximated to cattle data by a linear extrapolation assuming a sheep liveweight of 51 kg at slaughter, i.e. 1 sheep = 0.085 SCU.

Stream	kg per head per day	Indicative Facility Standing Head	Operational Days pa	Indicative Facility Tonnes pa	Standing Head - Australia	Tonnes pa - Australia	Solids %
Farm-Manure	2.47	5,000	365	4,504	29,200,000	26,303,804	74%
SY-Slurry	35	6,500	50	11,375	16,600,344	581,012	0.5%
SY-Manure	2.47	6,500	50	803	16,600,344	41,016	74%
FL-Slurry	1	10,000	365	3650	40,685	40,685	0.5%
FL-Manure	2.47	10,000	365	9016	40,685	100,492	74%
RMP - DAF	1.4	1,000 hpd	250	350	29,200,00	40,880	7%
RMP - Paunch	2.2	1,000 hpd	250	550	29,200,00	64,240	34%
RMP - WAS	2.2	1,000 hpd	250	550	29,200,00	64,240	12.5%
RMP – green screenings	0.19	1,000 hpd	250	47.5	29,200,00	5,548	20%
RMP – red screenings	0.27	1,000 hpd	250	67.5	29,200,00	7,884	20%
RMP – Plastic	0.14	1,000 hpd	250	35	29,200,00	4,088	98%
RMP - Cardboard	0.1	1,000 hpd	250	25	29,200,00	2,920	90%
RMP – AD Digestate	0.37	1000 hpd	250	23,441	Unknown number of AD facilities in Australian processing plants encompassing open ponds, crusted ponds, and covered anaerobic lagoons with a wide range of solids handling procedures.		5%

TOTALS: Beef at 25.4 million tpa representing 48.3% of the total, Sheep at 27.2 million tpa representing 51.7% of the total.

Stream	Tonnes pa - Australia	Percentage (%)
Farm-Manure	42,246,661	80.37%
SY-Slurry	2,450,533	4.66%
SY-Manure	49,041	0.09%
FL – Slurry	5,344,971	10.17%
FL - Manure	1,751,429	3.33%
RMP - DAF	154,730	0.29%
RMP - Paunch	243,640	0.46%
RMP - WAS	245,710	0.47%
RMP – green screenings	21,639	0.04%
RMP – red screenings	29,964	0.06%
RMP – Plastic	15,349	0.03%
RMP - Cardboard	11,200	0.02%
RMP – AD Digestate	TBA	0.00%
TOTAL	52,564,867	

## Table of contents

<b>1</b>	<b>Milestone description .....</b>	<b>58</b>
<b>2</b>	<b>Project objectives .....</b>	<b>59</b>
<b>3</b>	<b>Success in meeting the milestone .....</b>	<b>62</b>
	<b>3.1RMI Supply Chain .....</b>	<b>62</b>
	<b>3.2Producers .....</b>	<b>64</b>
	3.2.1 On-Farm Beef Cattle - Solids.....	64
	3.2.2 On-Farm Beef Cattle - Liquids.....	65
	3.2.3 On-Farm Gases.....	65
	3.2.4 On-Farm Sheep .....	65
	3.2.5 On-Farm Opportunities .....	66
	<b>3.3Sale Yards .....</b>	<b>68</b>
	3.3.1 Beef Cattle Saleyards and Solids.....	68
	3.3.2 Beef cattle Saleyards - Liquids .....	68
	3.3.3 Saleyard Gases .....	69
	3.3.4 Sheep Saleyards .....	69
	3.3.5 Saleyard Opportunities .....	69
	<b>3.4Feedlots.....</b>	<b>71</b>
	3.4.1 Feedlot Solids.....	71
	3.4.2 Feedlot Liquids.....	71
	3.4.3 Feedlot Gases.....	72
	3.4.4 Feedlot Opportunities .....	72
	3.4.5 Sheep in Feedlots.....	74
	<b>3.5Processors .....</b>	<b>75</b>
	3.5.1 RMP Solids .....	75
	3.5.2 RMP Water / Liquid Sources and Recycling Options .....	76
	3.5.3 RMP Gaseous Emissions .....	78
	3.5.4 RMP Opportunities .....	79
	3.5.4.1 Forced Aeration Composting with Heat Recovery.....	79
	3.5.4.2 Waste to Energy – Thermal Systems.....	82
	3.5.4.3 Energy from Waste – Anaerobic Digestion .....	84
	3.5.4.4 Lower Grade Oils .....	85
	3.5.4.5 Innovative Solutions – High Value Products from Waste .....	86
	<b>3.6Upcoming Waste Streams in the Next 2-3 Years.....</b>	<b>87</b>
<b>4</b>	<b>Overall progress of the project .....</b>	<b>88</b>
<b>5</b>	<b>Conclusions/recommendations .....</b>	<b>88</b>
<b>6</b>	<b>Bibliography .....</b>	<b>89</b>



## GLOSSARY

AD – Anaerobic Digestion

BSFL – Black Soldier Fly Larvae

BW – Body Weight

CN2030 – Carbon Neutral by 2030 goal of Australian RMI

CSTR – Continuously Stirred Tank Reactor

Cwt – Carcass Weight

DAF – Dissolved Air Flotation

FL – Feedlot

FOG – Fats, Oils, and Grease

GJ – Gigajoule, equivalent to 1,000,000,000 joules

Hpa – Head per Annum

Hpd – Head per Day

Hpw – Head per Week

kWh – Kilowatt-Hour unit of energy commonly used for electrical energy. 1 kWh = 1 kW for 1 hour

LHV – Lower Heating Value a.k.a. net calorific value, measure of energy content after evaporation of water content, contrasted to Higher Heating Value HHV a.k.a. gross calorific value

ML – Megalitre

OpEx – Operational Expenses

pa – Per Annum

RAS – Recirculating Aquaculture System

RMI – Red Meat Industry

RMP – Red Meat Processor

RO – Reverse Osmosis, water purification process

SCU – Standard Cattle Units, 1 SCU = 600 kg liveweight

Scope 1, 2, 3

Scope 1: Direct emissions from owned or controlled sources e.g. emissions from boiler fuel

Scope 2: Indirect emissions from generation of purchased energy e.g. emissions from grid power

Scope 3: Indirect emissions not included in Scope 2 that occur in the value chain upstream, and downstream e.g. transport of product

STEC – Shiga toxin-producing E. coli

SY – Saleyard



TL – Transport and Lairage

Tpa – Tonnes per Annum

Tpw – Tonnes per Week

TS – Total Solids

VS – Volatile solids

W2E – Waste to Energy

WAS – Waste Activated Sludge a.k.a. Aerobic Solids

WWTP – Wastewater Treatment Plant

## **1 Milestone description**

Mapping of the waste and effluent streams in the red meat industry from farm to processor with identification of value adding opportunities. This will contribute to a survey of microbiological quality on processed waste and effluent, ranging from farms to processing establishments, to provide clarity and identify potential risks which may not have been previously considered.

## 2 Project objectives

The primary emphasis of this milestone is to define the various waste streams, management options and utilisation options in support of the project, including:

- Mapping out of all of the waste streams from farms through to processing facilities.
- Potential value adding/opportunities for waste (note: without duplication of Rural R&D for Profit project “Wastes to Profits”).
- Advice on any upcoming waste stream in the next 2 – 3 years with associated micro-survey recommendations (where available).

Livestock are a known source of pathogens that could unintentionally be directly (waste, manures) or indirectly (air, dust, water runoff, water source, pests and wildlife) transferred to product within the supply chain and present a food safety risk. Environmental, demographic, and climate changes have the potential to increase the risk of hazard transmission between agricultural sectors.

In the event of a foodborne illness outbreak, source attribution studies and epidemiological investigations, which are extending to farm level to identify the primary source of hazards, increase accountability and potential consequences to producers.

Research funded by the fresh produce industry is investigating some risk factors in food safety e.g. adequately composted manure used for growing fresh produce.

The feedlot industry, with MLA, has published guidelines to help manage and utilise waste and effluent appropriately and safely with minimal negative impact on the environment and surrounding sectors.

The wider tasks of the project are:

- Literature review of hazards and methods of quantification: As this area is relatively new and there are many unknowns, the project will commence with a literature review. The review will cover potential hazards/risks, the tests that need to be conducted, and details of the preferred test methods for the sample matrices involved in this area. The literature review will also include reviewing previous work referenced in industry standards as they should hopefully provide insights into the risks that were and weren't seen as likely. The focus will therefore be: Hazard identification (including whether others e.g. STEC should be considered), Methods for recovery of organisms from complex matrices, and Review literature referenced in industry standards/docs.
- Survey of practices, protocols and adherence to procedures: The survey of current practices will be conducted in two parts - an initial desktop survey of industry participants to get a broader cross section of information on current practices; and site surveys and assessments of actual practices at selected premises.
- Desktop survey: The initial desktop survey of industry participants will be conducted to get a broader cross section of information on current practices at each of the main sectors within the scope of this study – including Farms, Feedlots, Saleyards and Processing establishments. The survey will be done as a series of telephone interviews. The budget has made provision for the following numbers of survey participants to be interviewed in the respective sectors and has allowed for 30, 60, 60 and 90 minute interviews for Farms, Feedlots, Saleyards and Processing establishments, respectively.

	QLD	NSW	VIC	TAS	SA	WA
<b>Survey of practices, protocols and adherence to procedures</b>						
<b>A. Desktop survey</b>						
Farms	6	6	4	2	2	2
Feedlots	6	6	4	2	2	2
Saleyards	4	4	3	2	2	2
Processing establishments	6	6	3	2	2	2
Total	22	22	14	8	8	8

- On Site survey, including sample collection: Following the desktop surveys, a series of site visits will be undertaken to assess current practices and adherence to industry standards and protocols. The approach to the site surveys will be the same as the desktop surveys, albeit modified to delve deeper into any specific issues of interest that arise from the previous work in the project. It will include Farms, Feedlots, Saleyards, and Processing establishments selected from desktop survey pool. The budget has provision for the following numbers of survey participants to be interviewed in the respective sectors and has allowed for 1-1.5 days each for interviews at Farms, Feedlots, Saleyards and Processing establishments as shown.

<b>Survey of practices, protocols and adherence to procedures</b>						
<b>B. On Site survey, incl sample collection</b>	QLD	NSW	VIC	TAS	SA	WA
Farms	1	1	1	0	0	0
Feedlots	2	2	2	0	0	0
Saleyards	2	2	2	0	0	0
Processing establishments	2	2	2	0	0	0
Total	7	7	7	0	0	0

- Sample Collection: It is proposed to collect samples for testing in two ways – submission by participants from the desktop surveys; and submissions collected by Symbio team during site visits. The aim is to ensure we gather a sufficient number of samples to make the results meaningful, without impacting the project budget (i.e. field collection costs are high if done by personnel travelling to a specific site). The following table outlines the number samples that will be collected and submitted by sector and by state.

	QLD	NSW	VIC	TAS	SA	WA
<b>Sample collection from each site surveyed above - desk top mailed to lab, sites collected and sent to lab</b>						
Farms	24	24	18	6	6	6
Feedlots	30	30	24	6	6	6
Saleyards	24	24	21	6	6	6
Processing establishments	30	30	21	6	6	6
<b>Total number of samples</b>	<b>108</b>	<b>108</b>	<b>84</b>	<b>24</b>	<b>24</b>	<b>24</b>

- Testing: Microbial tests will be conducted as per the scope of the project, on the above samples submitted. This includes the following tests:
  - Thermotolerant coliforms
  - E.coli
  - Salmonella
  - Listeria mono
  - Listeria spp
  - Campylobacter spp
  - Coagulase positive Staphylococcus
- Data Analysis & Reporting - Correlation of practices and microbial quality: Once all the samples have been collected and surveys completed, it should be possible to analyse the results by performing correlations. It should be noted that with pathogens, there may not necessarily be well established correlations with practices undertaken due to the typically infrequent nature of their prevalence.
- Evaluate treatment methods & parameters against guidelines: The project team will compile and evaluate treatment methods and parameters against the industry guidelines using data collected from both the desk top and site surveys plus the test analysis results.
- Propose changes in waste and effluent management to improve quality: Based on the findings from the above work, the project team should be able to identify and propose changes in waste and effluent management to improve microbiological quality. Recommendations for further research/investigation will be made. Note: this aspect of the reporting will be general in nature, i.e. the project has not made provision for re-writing any guidelines as that is a whole process in itself.

### 3 Success in meeting the milestone

#### RMI Supply Chain

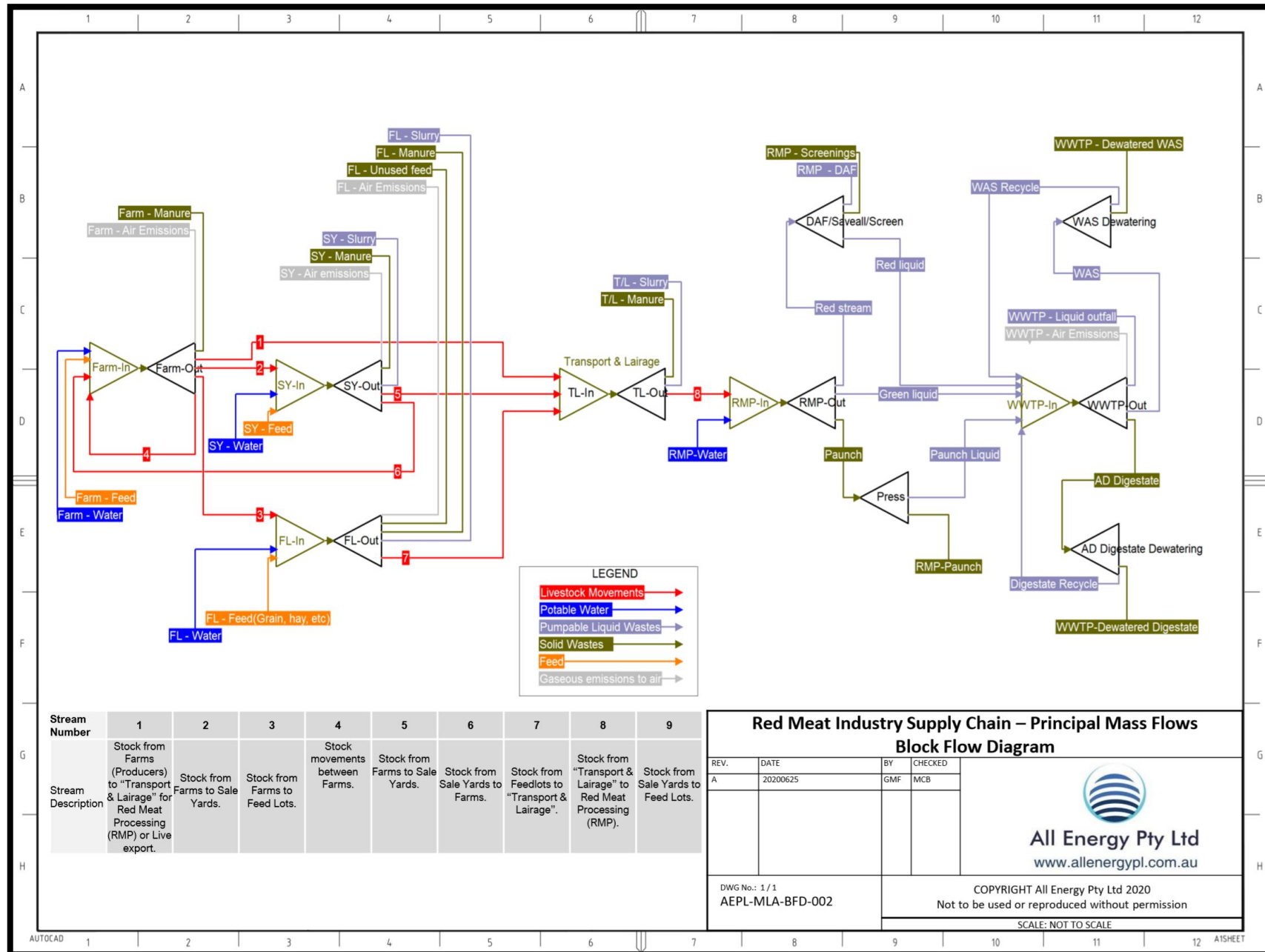


Figure 1: RMI Supply Chain Flow Diagram

## 4 Producers

### 5 On-Farm Beef Cattle - Solids

On-farm solid wastes include mortalities, manure, waste feed. A literature review of MLA works on herd mortality rates appear to be limited to breeder mortality, with many difficulties in sourcing accurate data identified, instead relying on indirect and alternative measures. Mortality rates per annum have been estimated<sup>6</sup> at 2 – 12% with higher mortality rates expected in cows aged 10 years or older (15 – 20%) or in severe drought conditions (>20%). As an indicative estimate, for a national herd of 24.8 million in April 2020<sup>7</sup> at an average 7% per annum mortality and 300 kg liveweight, annual mortality tonnage is estimated at 520,800 tonnes. During the survey, we will attempt to refine this estimate.

Manure production on-farm is very difficult to quantify due to herds distributed over very large surface areas, however, may be approximated as the proportion of feed intake to intensive feeding manure production, of which higher quality research has been commissioned by MLA. As an indicative estimate, for an intensive cattle production estimate of 900 kg manure per head per year at 35% moisture<sup>8</sup> and a ratio of body weight percentage feed consumption of 2% on farm to 2.8% in feedlot<sup>9</sup>, on-farm manure production is estimated at 643 kg per head per year. At 35% solids, the solid (faeces) fraction of manure production is 225 kg per head per year.

Waste feed will be required to be estimated with the assistance of producers as part of the survey stage, as no solid information on producer waste feed was found during the literature review.

A 45.4 kg market lamb produces an estimated 1.81 kg of manure per day<sup>10</sup> (grain finished lambs weigh 35 to 50 kg)<sup>11</sup>. At first view, the mass of manure per sheep appears high. However, when it is considered that 78% of sheep are in self-replacing systems and hence the sheep are older and larger, the 2.47 kg per head of manure for on-farm sheep is reasonable.

There is some evidence from an Australian Bureau of Statistics 2011-12 survey that 35.8% of agricultural operations with animal wastes turn the dry manure into compost, with 63.8% collecting dry manure into piles then spreading with 0.4% sending animal wastes to a digester<sup>12</sup>, however a

---

<sup>6</sup> Henderson, Perkins, and Banney, 2013. Determining property-level rates of breeder mortality in northern Australia: literature review. MLA

<sup>7</sup> <https://www.mla.com.au/globalassets/mla-corporate/prices--markets/documents/trends--analysis/cattle-projections/april-2020-aust-cattle-industry-projections.pdf>

<sup>8</sup> Davis, Watts, and McGahan. Quantification of Feedlot Manure Output for Beef-Bal Model Upgrade. MLA

<sup>9</sup> <http://agriculture.vic.gov.au/agriculture/livestock/beef/feeding-and-nutrition/opportunity-lot-feeding#:~:text=Some%20figures%20as%20a%20guide,depending%20on%20their%20initial%20condition.>

<sup>10</sup> [www.sheep101.info](http://www.sheep101.info), accessed 3 July 2020.

<sup>11</sup> <http://agriculture.vic.gov.au/agriculture/livestock/sheep/feeding-and-nutrition/feedlotting-lambs>, accessed 3 July 2020.

<sup>12</sup> <https://www.abs.gov.au/AUSSTATS/abs@.nsf/DetailsPage/4630.02011-12?OpenDocument>, accessed 13 July 2020



detailed review of the data suggests that the surveys may not accurately reflect current practices and are now dated (e.g. data suggests 133.2 beef cattle feedlots spread dry manure with no sites turning manure into compost).

## 6 On-Farm Beef Cattle - Liquids

Wastewater is not expected to exist on farm in any significant quantity, with on-farm liquids limited to urine, of which humans breathing in dust contaminated by urine and faeces is the primary transmission of Q-Fever. The difficulty with estimating on-farm liquids is that reported manure values account for urine and faeces i.e. 'manure' refers to urine plus faeces<sup>13</sup>; it is also reported that it is difficult to obtain the urine component directly from unconfined animals in the field. As an indicative estimate, the liquid fraction of manure (65%) may be assigned to urine, or 418 kg per head per year. Care should be taken to ensure values reported in the literature are not referring to dairy cattle, of which the manure production is significantly higher than beef cattle, due to the liquid fraction.

## 7 On-Farm Gases

On-farm waste gases include methane emissions from cattle and fugitive emissions from manure, and emissions from distributed generation, petrol and diesel generators etc. Emissions to air are generally considered outside of the scope of microbial food safety.

## 8 On-Farm Sheep

Sheep production statistics reported by MLA<sup>14</sup> used to estimate waste stream production are as follows

Total national flock 67,500,000

### Lamb

- 2019 slaughter 21,200,000
- 475,000 tonnes cwt production
- 22.4 kg cwt per head

### Sheep

- 2019 slaughter 8,000,000
- 188,000 tonnes cwt production

<sup>13</sup> Watts, McGahan, Bonner, and Wiedemann, 2011. Feedlot Mass Balance and Greenhouse Gas Emissions – A Literature Review. MLA

<sup>14</sup> [https://www.mla.com.au/globalassets/mla-corporate/prices--markets/documents/trends--analysis/sheep-projections/mla\\_australian-sheep-industry-projections-2019.pdf](https://www.mla.com.au/globalassets/mla-corporate/prices--markets/documents/trends--analysis/sheep-projections/mla_australian-sheep-industry-projections-2019.pdf)

- 23.5 kg cwt per head
- Small difference in carcass yield between mutton and lamb

The key figure used to estimate live weight at slaughter is the dressing percentage, reported in the range of 37% to 54%<sup>15</sup>, with a mean value of 46% used in this study.

Manure production has been reported in the literature at 13 kg / tonnes body weight / day of urine and 37 kg / tBW / day faeces. Estimates of manure production for the Australian sheep flock are 6,838,989 tonnes per annum of urine and 19,464,815 tonnes per annum of faeces, for a total manure production of 26,303,804 tonnes. At a cwt of 23.5 kg and dressing percentage of 46% (51 kg liveweight), a combined 50 kg / tBW / day of urine and faeces estimates a daily production of 2.47 kg per head. Comparing this similar production to a cow with a liveweight an order of magnitude greater than a sheep, it is assumed that this value is over-reported in the literature.

## 9 On-Farm Opportunities

It is not expected that red meat producers will have significant agency to change their waste management practices, due to highly distributed waste generation over large surface areas, low capital availability, lower scales making most technology choices unviable, and unavailable resources to be allocated towards managing waste.

Micro-scale nutrient recycling may be viable on-farm, if the difficulties in collecting fresh volatile organic matter can be overcome. An example of a micro-scale system is shown below that can digest 45 litres of manure slurry (or 15 litres of manure and 30 litres of water) to create 2 hours of biogas for use in a single burner or for lighting a gas lamp. It is highly recommended that the gas be used in devices outside and that hazard / safety reviews be completed. Within Queensland, such devices are covered under the Petroleum and Gas Act 2004 and hence require appropriate approvals. Payback for this system is approximately 3 years when compared to energy from LPG; hence systems could be considered at points where there is a daily requirement for cooking and/or lighting.

---

<sup>15</sup> [https://www.mla.com.au/globalassets/mla-corporate/prices--markets/documents/minlrs-information-brochures-etc/mla\\_sheep-assessment-manual\\_jan-2017.pdf](https://www.mla.com.au/globalassets/mla-corporate/prices--markets/documents/minlrs-information-brochures-etc/mla_sheep-assessment-manual_jan-2017.pdf)

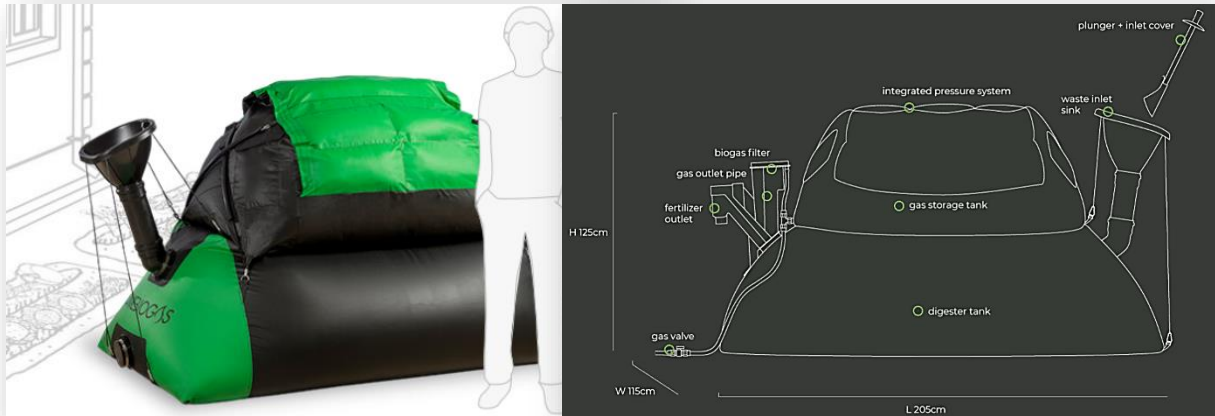


Figure 2: Micro-scale digester for nutrient recycling. Source: Homebiogas<sup>16</sup>.

Emissions Reduction Fund projects that could utilise manure products for on-farm sequestration projects include:

- Soil carbon
- Managed plantations
- Regeneration

<sup>16</sup> <https://www.homebiogas.com/Products/HomeBiogas2>, accessed 13 July 2020.

## 10 Sale Yards

### 11 Beef Cattle Saleyards and Solids

Saleyard throughputs for 2018-2019 are reported<sup>17</sup> at

- NSW – 1,660,000 over 41 saleyards
- QLD – 1,300,000 over 27 saleyards
- VIC – 1,040,000 over 19 saleyards
- SA – 238,242 over 4 saleyards
- WA – 261,225 over 4 saleyards
- TAS – 60,340 over 2 saleyards
- Total – 4,559,807

A daily estimate of manure production is 1.76 kg per head per day at 35% solids, 0.617 kg dry fraction (faeces).

Saleyard mortalities are not assumed to be significantly impacted by the time that livestock are deprived of feed and water before and during the transport journey. Due to less time spent in the saleyard, it is assumed to be more appropriate to estimate mortality rates on a daily basis, which for an average 7% per year equates to 0.019% mortality rate per day. As an indicative estimate, the largest saleyard in the country, the Roma saleyard, lists 5458 cattle booked in for Tuesday 9<sup>th</sup> June 2020<sup>18</sup>, which at an 0.019% daily mortality rate estimates 1.04 mortalities, or 0.419 tonnes at 400 kg liveweight. Other large saleyards in the country include<sup>19</sup>

- Roma, QLD – 319,053 head per annum
- Dalby, QLD – 258,293 head per annum
- Dubbo, NSW – 193,788 head per annum
- NVLX Wodonga, VIC – 175,993 head per annum
- Wagga Wagga, NSW – 172,734 head per annum
- Leongatha, VIC – 138,846 head per annum

### 12 Beef cattle Saleyards - Liquids

The 65% liquid fraction of 1.76 kg per head per day manure production is estimated at 1.15 kg per head per day. This tonnage will be accounted for in the values of saleyard pen washdown and water runoff, which may be estimated as a proportion of processor holding yard washdown tonnage estimated by All Energy in previous RMP projects<sup>20</sup> for AMPC. For a 1000 hpd processor, yard runoff was calculated at 2,590 tonnes per week, thus as an indicative estimate prior to refinement during the survey, a 5458 head per day saleyard runoff is conservatively estimated at 14,136 tonnes per week.

<sup>17</sup> <https://www.beefcentral.com/markets/cattle-saleyard-throughput-state-by-state-2018-19/>

<sup>18</sup> <http://www.mymaranoa.org.au/business/saleyards/sale-numbers-special-lines>

<sup>19</sup> <https://www.beefcentral.com/markets/cattle-saleyard-throughput-state-by-state-2018-19/>

<sup>20</sup> Barnes and Forde, 2020. Aggregated Waste to Energy (W2E). AMPC 2020-1006

This number is likely greatly overestimated as the processor values include belly wash and more intensive cleaning for food safety reasons, but serves for now as a starting estimate.

### **13 Saleyard Gases**

Saleyard waste gases are limited to methane emissions from cattle and fugitive emissions from manure, and emissions from distributed generation, petrol and diesel generators etc. The Scope 3 transportation emissions (i.e. the greenhouse gas emissions associated with the transportation of cattle to and from the saleyard) are outside of the scope of this project.

### **14 Sheep Saleyards**

Sheep saleyard throughput is reported in the MLA 2019 Saleyard Survey<sup>21</sup> as follows

- NSW – 8,556,753 over 26 saleyards
- QLD – 113,110 over 1 saleyard
- SA – 1,176,433 over 3 saleyards
- VIC – 5,107,369 over 17 saleyards
- WA – 1,388,879 over 2 saleyards
- TAS – 257,800 over 2 saleyards

Assuming 50 sale days per annum, indicative standing heads are estimated at

- NSW – 6,582
- QLD – 2,262
- SA – 7,843
- VIC – 6,009
- WA – 13,889
- TAS – 2,578

### **15 Saleyard Opportunities**

General saleyard practices include manure collection and often “free issued” to surrounding farms or use on adjacent lands. Opportunities for managing saleyard wastes include on-site digestion or “hub and spoke” models where smaller distributed saleyard wastes are aggregated at larger centralised, more intensive saleyards making use of economy of scale for the anaerobic digester and associated

---

<sup>21</sup> <https://www.mla.com.au/globalassets/mla-corporate/prices--markets/documents/saleyard-surveys/saleyard-survey-2019.pdf>

engines. However the transportation costs associated with transporting wastes needs to be considered and may be a key reason that a “hub and spoke” model is not economically viable.

## 16 Feedlots

Because feedlots concentrate the livestock in one location, there is significantly greater ability to concentrate and manage wastes to recover value.

### 17 Feedlot Solids

Intensive lot-fed cattle are reported to produce manure at 900 kg per head per year at 35% solids, or 315 kg per head per year dry weight, or 79 kg per head per 3 month feeding period. For the October – December 2019 period where 1,239,563 head were on feed, the pen scrapings (assuming the manure stays upon the pen surface for sufficient time as for the majority of the moisture to evaporate – i.e. pens scraped infrequently) produced during this period are estimated at 97,616 tonnes dry matter. This estimate is verified by MLA where a value of 410 kg TS / SCU / yr equivalent to 102.5 kg TS / SCU / 3 month is reported<sup>22</sup>. Normalising SCU to head with an assumed 500 kg liveweight produces 85.4 kg TS / head / 3 months, or 8.4% error. Another reference states 5.5 kg per head per day<sup>23</sup>.

Waste feed is derived from pen feeding trough clean outs, contamination of grain / straw by mould, fungus, or vermin due to high moisture content and improper storage. This is routinely mixed with scrapings and composted onsite.

Mortality rates are reported in-feedlot at 2% during the time spent on-feed<sup>24</sup>, significantly lower than on-farm due primarily to the higher availability of feed and water. Cattle on feed finished on a record high of 1,239,563 in October – December 2019. Mortalities are estimated at 24,791 for the period of October – December 2019 as an indicative estimate, or 12,396 tonnes at an average liveweight of 500 kg.

### 18 Feedlot Liquids

The liquid fraction of manure is estimated at 146 kg per head per 3 month feeding period, or 181,286 tonnes for the period October – December 2019 as an indicative estimate. A large portion of this tonnage may evaporate if manure sits in an uncovered or partially covered pen for an extended period, and soak into the ground in porous earth pens. This moisture loss may be mitigated by covered pens and cement pad pens. The remaining runoff is influenced by cleaning and dependent on rain events, so will need to be further defined during the survey. The volume of liquid runoff from feedlot pens is dominated by rain events.

---

<sup>22</sup> Tucker et al, FSA Consulting and Rural Directions, 2015. Beef cattle feedlots: waste management and utilisation, Meat and Livestock Australia

<sup>23</sup> <https://mdpi.com>, accessed 3 July 2020.

<sup>24</sup> Watts, McGahan, Bonner, and Wiedemann, 2011. Feedlot Mass Balance and Greenhouse Gas Emissions – A Literature Review. MLA

MLA reports a range of calculated feedlot slurry runoffs for six different 5,000, 10,000, and 25,000 SCU feedlots. Interpolating for an indicative scale of 20,000 SCU estimates 85.7 ML pa runoff, or 11.74 kg per head per day.

## 19 Feedlot Gases

Feedlot gas emissions include methane from cattle, manure, emissions from LPG boilers, and stationary diesel generation. All Energy Pty Ltd's extensive feedlot energy strategy works have estimated that a feedlot located in the Darling Downs region of QLD will consume 0.507 GJ per year per SCU, or 30.5 kg CO<sub>2</sub> equivalent per SCU per year<sup>25</sup>. At a calculated 25.4 kWh per SCU per year, the emissions from a diesel generator are calculated at 21.3 kg CO<sub>2</sub> equivalent per SCU per year, or 8.2 kg for grid power.

## 20 Feedlot Opportunities

Before opportunities for improving effluent management in feedlots can be explored, the problem of low LHV / low methane potential stale manure must be solved. Automated / instant manure collection with autonomous robots has been implemented in the dairy industry before, with manufacturers having commercially ready products available. Implementing robots such as these in feedlots can improve animal health and comfort, and reduce greenhouse gas emissions, collecting the manure whilst fresh for further value adding in anaerobic digestion.



Figure 3: JOZ-Tech JT200 Evo<sup>26</sup>

<sup>25</sup> Australian Government Department of the Environment and Energy, 2017. National Greenhouse Accounts Factors. <https://www.environment.gov.au/system/files/resources/5a169bfb-f417-4b00-9b70-6ba328ea8671/files/national-greenhouse-accounts-factors-july-2017.pdf>

<sup>26</sup> <https://joz.nl/en/oplossingen/manure-robots/>





Figure 4: Lely Discovery 120<sup>27</sup>

All Energy Pty Ltd has analysed anaerobic digestion in the red meat industry in great detail, with a number of scale systems available from high rate modular systems expandable in modules of 50 m<sup>3</sup>, to continuously stirred tank reactor (CSTR) style systems available in standard modules of 2500 m<sup>3</sup>. As an indicative estimate of feasibility, All Energy Pty Ltd has estimated the capital cost of a CSTR in a 20,000 SCU feedlot burning LPG and running on grid power at approximately \$3,100,000, with simple payback of 7 years.

Anaerobic digestion is not known to be utilised in any Australian feed lotting environment. When offsetting LPG for boilers and diesel gensets, paybacks of towards 5 years can be achieved. Continuous stirred tank reactors (CSTRs) provide higher high efficiency and can handle the higher solids percentage of manure, hence are common at USA, Canadian and European feedlots (refer Figure 5). Biogas for cogen and/or boilers make strong economic sense when grain tempering / steam flaking is collocated onsite.

---

<sup>27</sup> <https://www.lely.com/press/2017/06/10/Discovery-Collector/>



Figure 5: twin digesters at a Canadian feedlot produce 630 kilowatts of green power annually while reducing carbon dioxide emissions by more than 10,000 tonnes annually. The facility cost \$CAD 7.1 million when installed in 2014 and can process 25,000 tpa of organics<sup>28</sup>.

## 21 Sheep in Feedlots

The expansion of the lamb and mutton feedlot industry has been suggested by state agriculture and primary industry departments<sup>29,30</sup> to take advantage of the growing export demand for lamb. There has been limited development in this sector to date, however guidelines and checklists are being published by MLA<sup>31,32,33</sup> for establishing intensive sheep and lamb finishing systems.

Due to the infancy of this sector, there has been little to no quantification of waste volumes. This has been identified during this project as a key area for future investigation by MLA.

<sup>28</sup> <https://www.canadiancattlemen.ca/features/feedlot-manure-helps-fuel-7-1-million-bioenergy-plant/>, accessed 3 July 2020.

<sup>29</sup> <http://agriculture.vic.gov.au/agriculture/livestock/sheep/feeding-and-nutrition/feedlotting-lambs#:~:text=The%20following%20are%20some%20guidelines,approximately%20five%20square%20metres%20Flamb>

<sup>30</sup> [https://www.dpi.nsw.gov.au/\\_data/assets/pdf\\_file/0020/193313/Feedlotting-lambs.pdf](https://www.dpi.nsw.gov.au/_data/assets/pdf_file/0020/193313/Feedlotting-lambs.pdf)

<sup>31</sup> <https://www.mla.com.au/globalassets/mla-corporate/extensions-training-and-tools/documents/nationalproceduresandguidelineslambfinishing.pdf>

<sup>32</sup> <https://www.mla.com.au/globalassets/mla-corporate/extensions-training-and-tools/documents/nationalproceduresandguidelineslambfinishing-checklists.pdf>

<sup>33</sup> <https://publications.mla.com.au/login/redirectFrame>

## 22 Processors

Annual slaughter for 2020 was projected by MLA at 6,900,000<sup>34</sup> at a 19% decrease compared to 2019 levels, due primarily to easing of drought pressures.

Major inlet/outlet nodes shown in Figure 1 are:

- Red stream solids separation devices: DAF, save-alls, screens.
- Green stream / paunch press
- Wastewater treatment plants (WWTPs)
- Boilers (non-microbial)

## 23 RMP Solids

Solids, in this report, are materials that are generally not pumpable (or able to be pumped with an impeller style pump). These materials will often have free water associated with them, however, predominantly require a materials handling solution that is not a pump. Solid wastes generated by a processor include non-renderable red stream screenings, paunch usually sent to composting and land application, tannery hair, cattle yard scrapings (may be collected as solid or liquid depending on individual site practices), and waste aerobic sludge. Other solid by-products that are not considered wastes as they are currently value added into a saleable product include hides, bones, fat, meat trimmings, and offal.

---

<sup>34</sup> <https://www.mla.com.au/prices-markets/Trends-analysis/cattle-projections/>

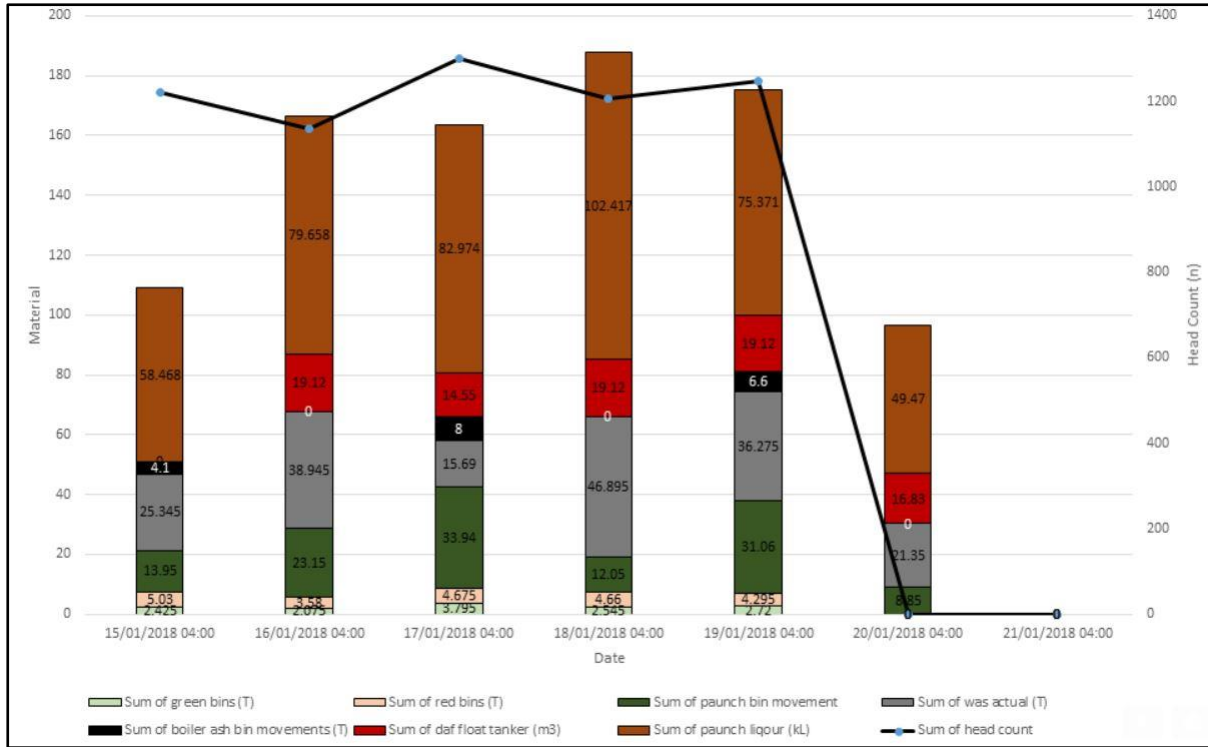


Figure 5: Example Comparative tonnages of different RMP wastes over one typical week.

All Energy has previously surveyed six medium to large processors on their non-recyclable solid waste production in partnership with AMPC<sup>35</sup> and reported that for an indicatively sized facility of 6000 hpw, waste is produced in the following volumes

- DAF solids at 99 tpw
- Paunch at 156 tpw
- Waste activated sludge from aerobic ponds at 158 tpw
- Green stream screenings at 14 tpw
- Red stream screenings at 19 tpw
- Contaminated plastic at 9.8 tpw
- Contaminated cardboard at 7.1 tpw

## 24 RMP Water / Liquid Sources and Recycling Options

There exists the opportunity to reduce potable water costs at RMPs via judicious selection of sources of and uses for recycled water. The following sections considered the “cleanest” source of wastewater and matched it with non-production uses. The following provides a list of the different qualities of water in existence at a typical RMP, in approximate order from highest to lowest quality.

RO make up water for Boiler

Potable + RO for cooling tower make up

Sterilizer water

Used within plant and utility processes

<sup>35</sup> AMPC 2020-1006 Aggregated Waste to Energy

Warm water

Potable water / cold wash water

Biofilter water

Re-use water (e.g. belt press wash)

Cattle wash water

Used sterilizer water

Viscera table wash

RO reject and blow down water

Treated wastewater

Rendering plant liquid wastes / stick water

Paunch press water



Currently sourced from used water. Could be sent through new recycling plant.



Water with minimal contamination currently sent to WWTP that could be sent through recycling plant.



Water with higher level of contaminants with potential for other purposes (Horticulture; Energy from waste).

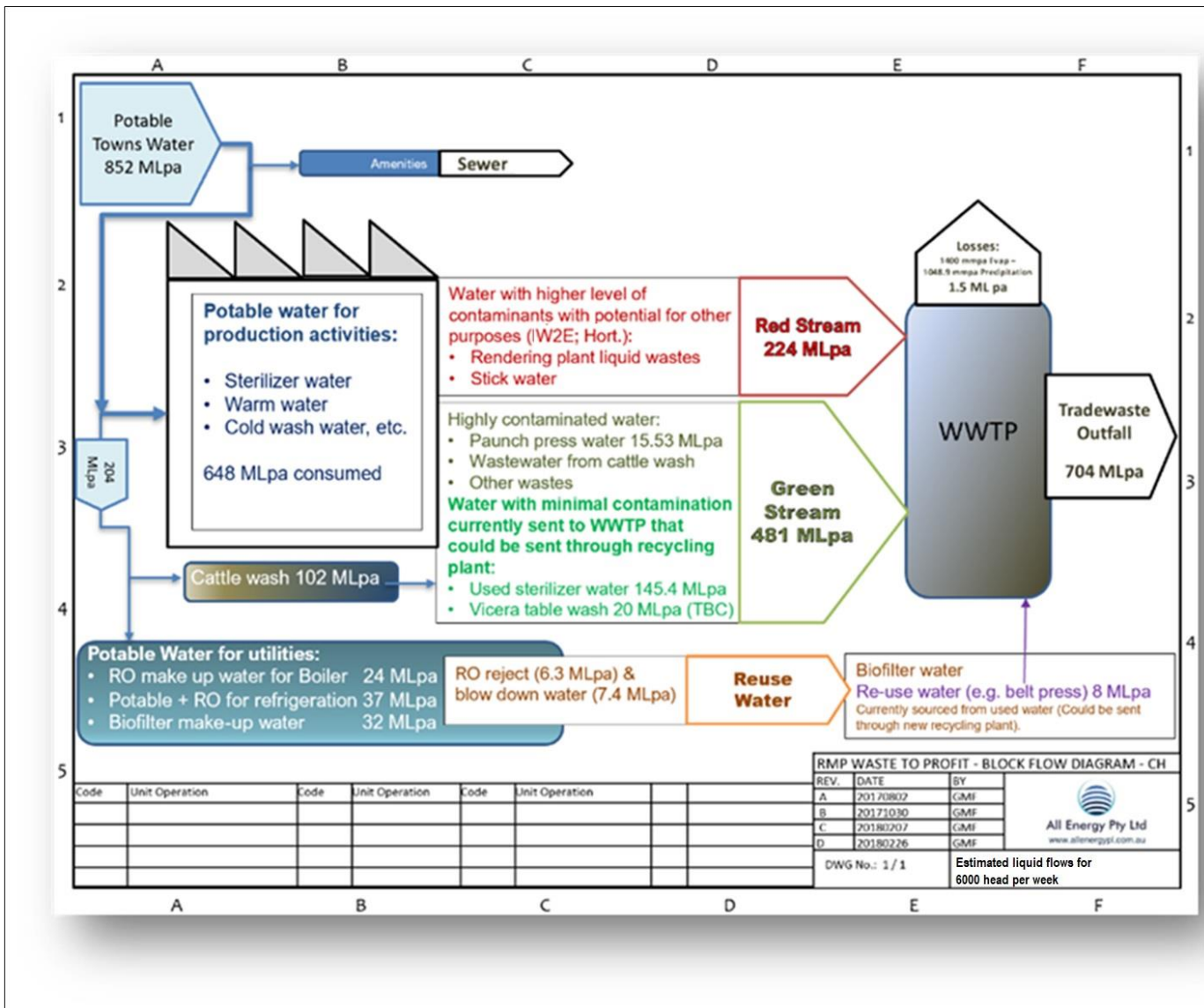


Figure 6: Example 1000 hpd processor water block flow diagram and opportunities

## 25 RMP Gaseous Emissions

Gaseous emissions from processing include boiler flue stacks (Scope 1), emissions from cattle in holding pens prior to slaughter (Scope 1), fugitive emissions from aerobic and anaerobic wastewater treatment plant (Scope 1), and emissions from trucking cattle to site (Scope 3). Of these, the boiler flue stacks are most easy to calculate at 90 kg CO<sub>2</sub> equivalent per GJ of bituminous and sub-bituminous coal, zero reportable carbon emissions for woodchip boilers, 51.4 kg CO<sub>2</sub> equivalent per GJ of natural gas, and 60.2 kg CO<sub>2</sub> equivalent per GJ of LPG, and 73.6 kg CO<sub>2</sub> equivalent per GJ of fuel oil burned.

An indicative value of flue gas emissions for a 1000 hpd processor burning coal was calculated by All Energy Pty Ltd at 17,076 tonnes CO<sub>2</sub> equivalent per annum<sup>36</sup>.

For an indicative 1000 hpd processor, annual power consumption has been calculated by All Energy Pty Ltd at 24,567 MWh or 81.07 kWh / head processed. On grid power burning bituminous coal, annual Scope 2 emissions are calculated at 7,960 tonnes CO<sub>2</sub> equivalent per annum<sup>23</sup>. It can thus be observed that the dominant contributor to site wide emissions are Scope 1 emissions from combustion of thermal fuel, followed by Scope 2 emissions from purchase of electricity at approximately half of Scope 1. Scope 3 trucking emissions are dependent on trucking distances and which operator has control over the fleet, and specific to each individual site.

As an indicative estimate, for a mean liveweight of 600 kg = 1 SCU, the number of head per 12.2m deck is reported at 20. For a double decker B double Higher Mass Limit (HML)<sup>37</sup>, i.e. 4 decks, it is estimated that 80 cattle could be packed with a maximum density of 1.47 square metres per head, with a total payload of 48 tonnes. An online vehicle emissions calculator<sup>38</sup> was used to estimate an emissions intensity of 1.735 kg/km or 0.022 kg/km/SCU. This number depends primarily on the packing density, with a greater emissions value for trucks packed less than 20 head per 12.2m deck.

At an assumed packing density of 30%, it is estimated that a refrigerated B double General Mass Limit (GML) could carry a payload of 25.2 tonnes of processed meat. 1.655 kg/km emissions or 0.066 kg/km/tonne was calculated for finished product. The lower packing density of boxed meat highlights the importance of payload when determining the emissions per tonne.

## 26 RMP Opportunities

### 27 *Forced Aeration Composting with Heat Recovery*

By “sucking” air through a compost heap, the aerobic microbial activity is maintained at a homogenous level hence higher average temperatures and a higher rate of composting is achieved. Further, via a mechanically aerated system the heat is drawn through a single pipe which enables recovery of the heat (i.e. heat exchanged with a closed water loop; refer Figures 8 and 9 below for an example) and could be used for general hot water, boiler water pre-heating or greenhouses.

ADVANTAGES of forced Aeration Composting With Heat Recovery:

- Collection of leachate into a single point
- Collection of gases into a single point – Odourless after biofilter
- Smaller footprint
- Heat recovery
- Water recovery (from condensate)

<sup>36</sup> Barnes and Forde, 2017. Development of a clean, viable, and sustainable energy strategy for red meat processing. MLA/AMPC P.PIP.0739

<sup>37</sup> Higher Mass Limit, greater concessions for payload than General Mass Limit (GML) and Concession Mass Limit (CML)

<sup>38</sup> <http://www.sustainablefreight.com.au/tools-and-programs/emission-calculators/truck-fuel-emissions-and-cost-calculator-and-comparison-tool>

- Higher rate of composting from forced air flow through compost pile / windrow leading to smaller scale, lower capex vs piles being aerated by passive atmosphere with periodic turning
- Tighter control and more homogenous aeration and temperatures throughout pile leading to enhanced pathogen destruction / vector control.
- Avoids need for active turning of piles via mechanical aeration system
- Better economics than in-vessel or enzyme systems.

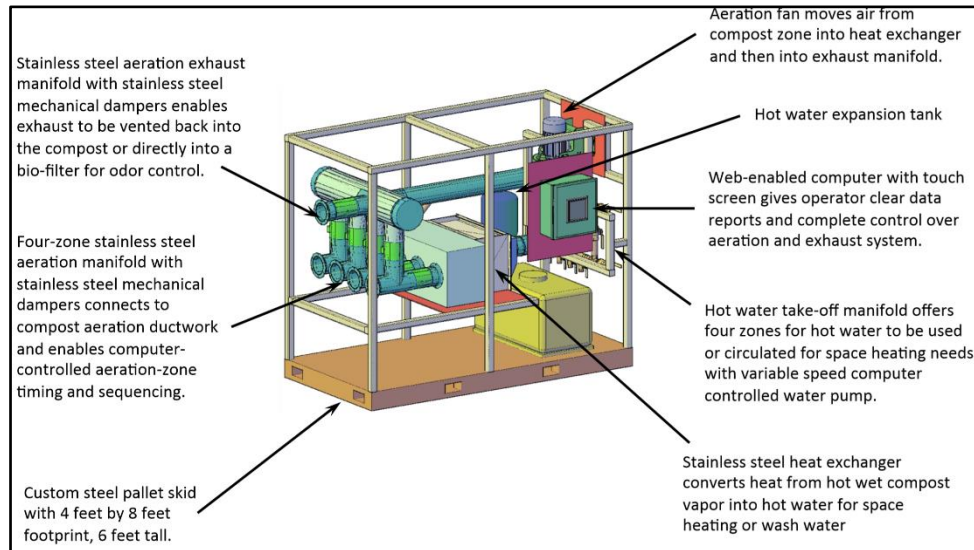


Figure 7: Hot Skid 250R-8. Vendor: Agrilabtech, USA

The system shown can process approximately 6000 – 8400 tpa of compost feedstock. Each Hotbox 250R-8 [8 rows] is expected to deliver 2,292 GJ pa at steady state (half rated capacity). ~72 kWt; with a maximum operation of 5,064 GJ pa at maximum air flow for water pre-heating ( ~164 kWt). Displacing LPG at \$30/GJ, this heating could be valued towards \$151,920 pa. The heat generated over the life of plant (20 years) is estimated at ~\$6 / GJ. The vendor has a range of operating modes, with the maximum thermal recovery for pre-heating of ambient incoming water. Capex fully installed estimated at \$646k (with equipment financing estimated at \$27k per month).



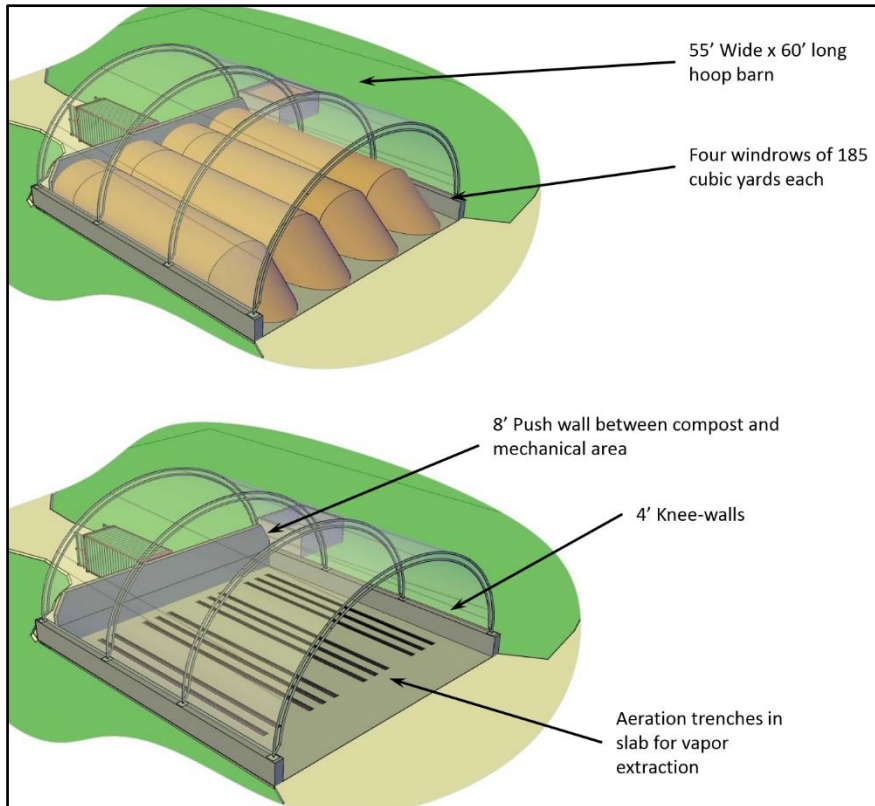




Figure 8: Schematic and example Agrilabtech installations

## 28 Waste to Energy – Thermal Systems

Figure 10 compares various waste to energy technologies by their operating temperature, primary products, opportunities for further processing and value adding, and respective advantages. Figures 11 – 13 expand upon thermal processing options with schematics and process flow diagrams showing key unit operations, inputs, energy and product outputs, and byproducts.

Waste to Energy Tech	Temp. °C	O <sub>2</sub> Level %	Gas Comp				Primary Products	Further processing	Advantages
			CH <sub>4</sub>	H <sub>2</sub>	CO <sub>2</sub>	CO			
Torrefaction	200-320	0					Syngas, bio-char, condensables	Energy pellets	Low temp & pressure
Pyrolysis	400-650	0	9	36	7	17	Syngas, bio-char &/or bio-crude	Power; Fertilizer	Low pressure; No ash
Gasification	650-850	20	3	18	6	24	Heat, syngas, ash	Methanol, hydrogen, syn-diesel	“Clean” syngas
Combustion	850-1000	125			11		Heat and Ash	Power	Heavy metals inert in slag / ash
Anaerobic digestion	35	0	52		48		Digested sludge, treated effluent	Fertiliser	Process wet biomass
Organic Rankine Cycle	~>80		Any heat source. Thermal oil is ideal.				Power		No fuel costs

Figure 9: Comparison of various waste to energy technologies by processing conditions, products, and advantages

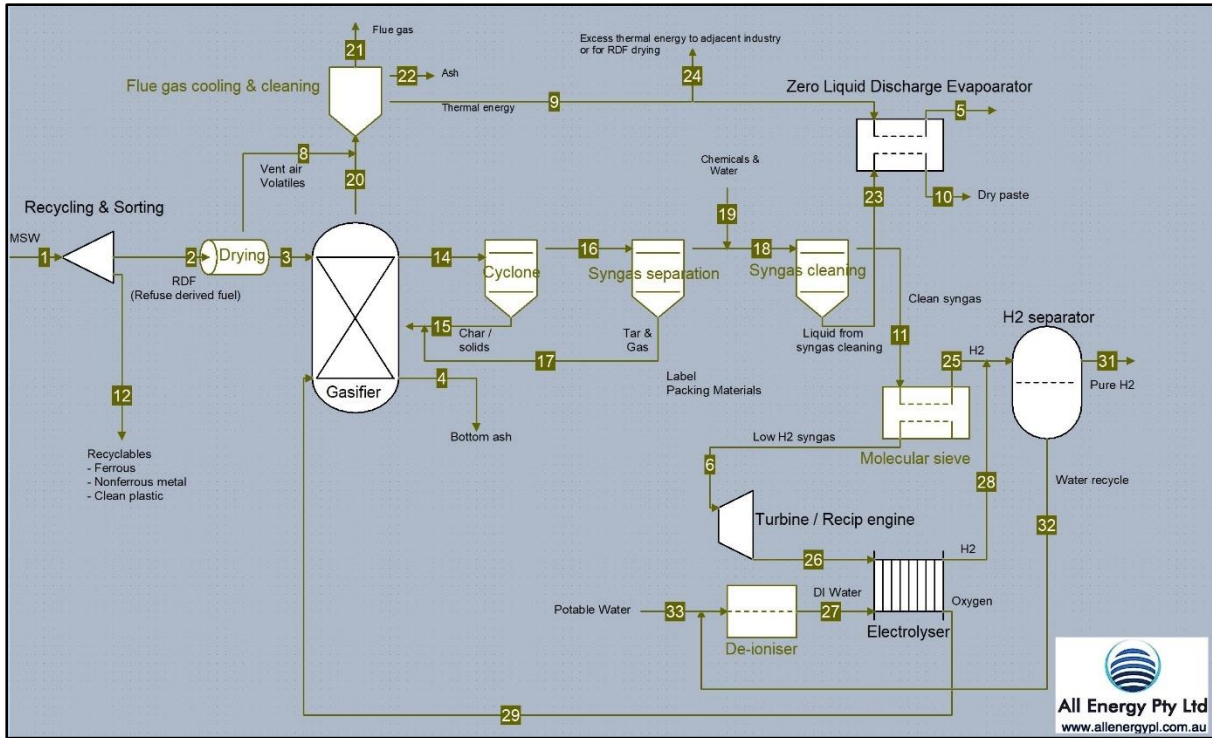


Figure 10: Gasification process flow diagram

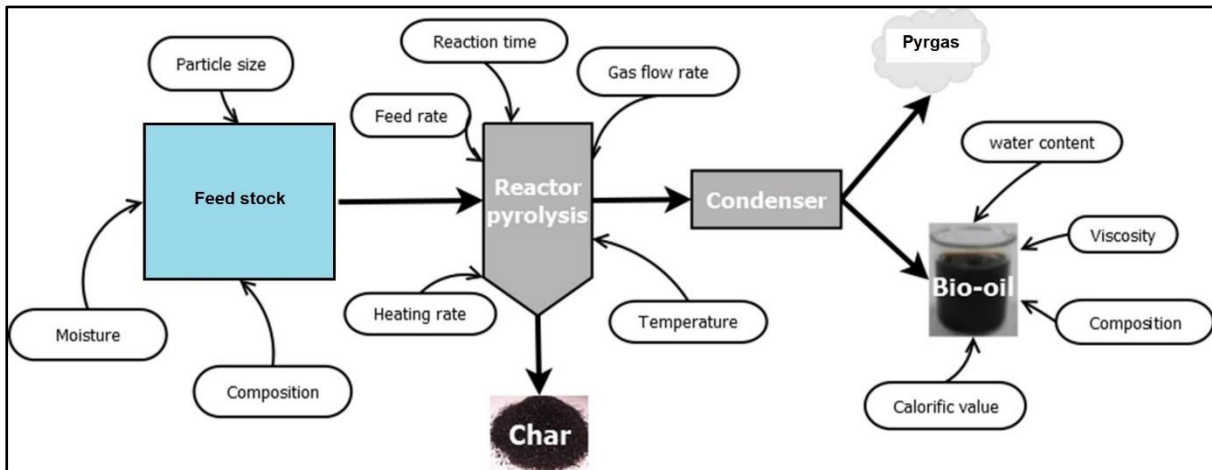


Figure 11: Pyrolysis process flow diagram

Advancements in boiler technology is enabling materials with higher moisture content to be fired in boilers. For the red meat industry, this means that paunch (~60 to 80% moisture), dried manure (~50% moisture) and waste fast streams (e.g. DAF floats) can be blended with air dried hardwood chip (~15%) to create a boiler fuel. An example of a suitable technology is shown in Figure 13 below with the following as a specific example:

**Fuel:**

- paunch at 20% solids and LHV of 1.7 GJ/tonne, 50 tonnes per week.
- airdried hardwood chip at 17.5 GJ/tonne, 97.8 tonnes per week.

**Boiler:** 5.3 MWt understoked, pile burner specifically designed for burning moist biomass.  
Total Capital Investment (TCI): \$3 mil

**Savings:**

- Paunch haulage costs of \$40/tonne, 50 tonnes/week = \$102,000 p.a.
- Natural gas usage at 5.3 MWt, 20 hrs/day, 245 days per year = \$1,449,126 mil p.a.

**OpEx:** Woodchip at \$55/tonne delivered to site, 97.76t/week: \$274,211 p.a.

**Simple payback:** 2.4 years. Payback can be reduced to 1.9 years where recycled construction wood is “free issued” in the place of procuring clean woodchip.

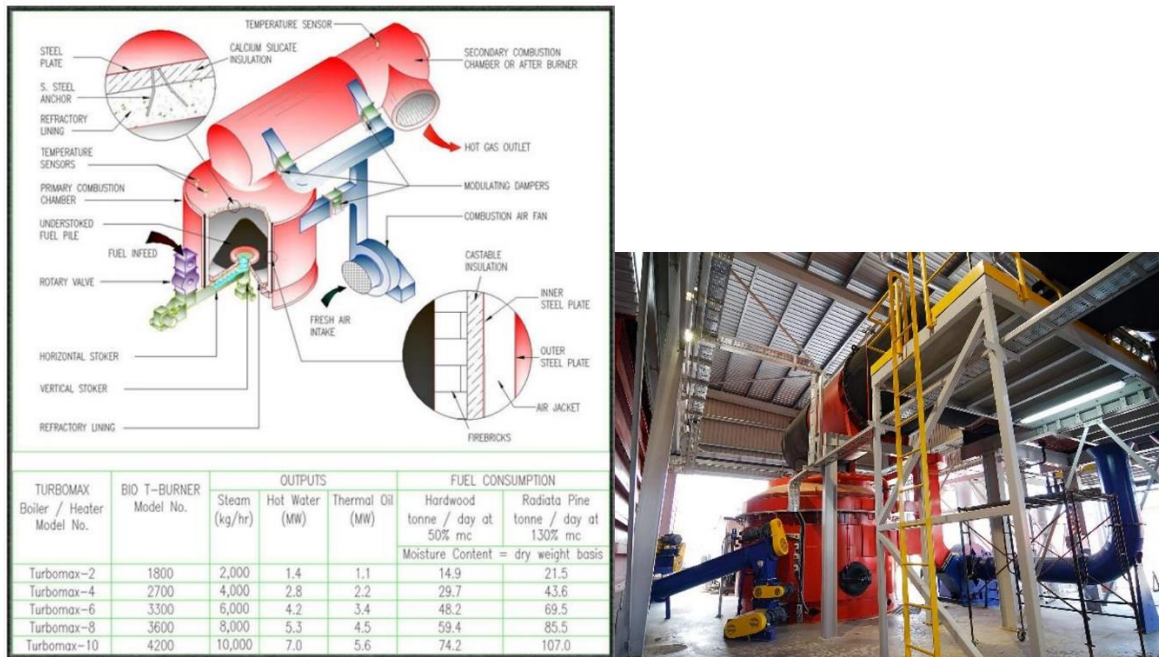


Figure 13: Multi-fuel Biomass Boiler suited for 50 to 60% moisture fuel. Source: Vismamax.

29 Energy from Waste – Anaerobic Digestion

The following Figure 14 shows the results of an analysis of an engineered anaerobic digester (e.g. continuous stirred tank) where the biogas is combusted in a boiler replacing a lagoon which has fugitive biogas emissions.

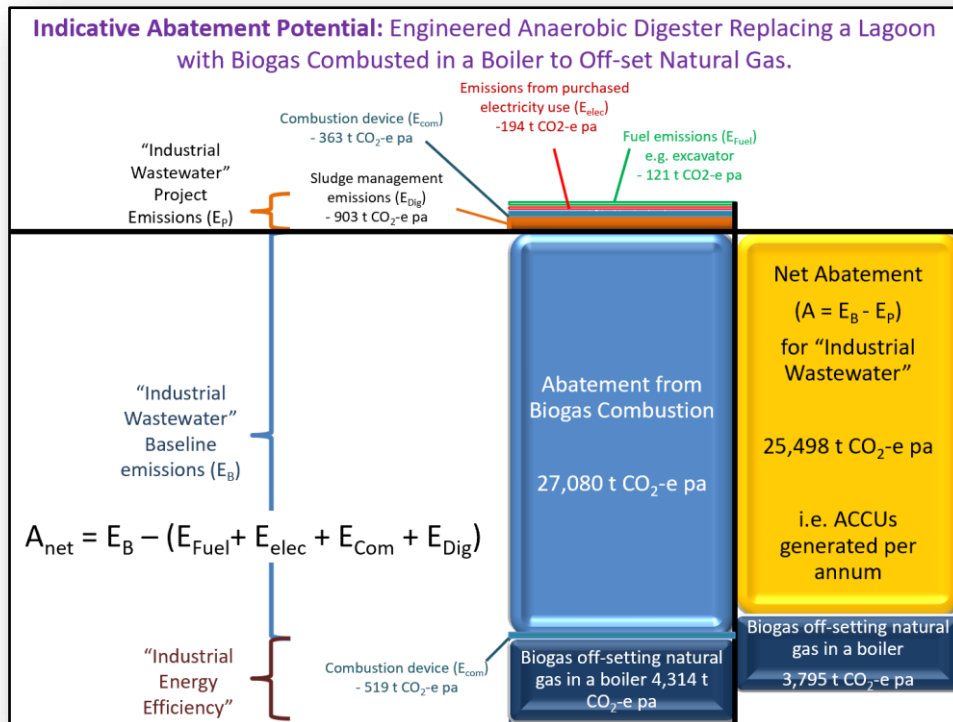


Figure 14: Indicative abatement potential - processor anaerobic digestion

### 30 Lower Grade Oils

Fats, oils and greases (FOGs) suitable for bio-diesel could be recovered from:

- Hydrocyclones on red waste / render waste streams,
- Rendering of mortres,
- Rendering of saveall / red screenings.

The FOGs need to be recovered before dissolved air floatation (DAF) processing as the high levels of oxygen results in oxygenation of the fats and volatile fatty acids which is undesirable for making bio-diesel.

### 31 Innovative Solutions – High Value Products from Waste

Some options that have been considered include:

- Use of solids (paunch and sludge) for growing fungus / moulds that produce high value enzymes;
- Use of organics for growing edible insects e.g. Black Soldier Fly Larvae (BSFL);  
High temperature liquefaction to create fermentable sugars;
- Use of wastewater for aquaculture; and
- Sterilization of solids for mushroom operations.

The table below summarizes a range of options considered for the creation of profit from wastes generated at red meat processing plants (RMPs).

Table 1: Waste to profit options analysis

Scenario	Cap ex \$AUS	Op Ex \$ pa	Revenue \$ pa	IRR	Pay- back	Lag to revenue	Production tpa	Profit pa
1) Aquaculture (RAS)	5.5 mil	3.53 mil	5.325 mil	17.7%	7 yrs	2 yrs	300 live fish	1.798 mil
2) Mushrooms	12.8 mil	9.29 mil	12.350 mil	33.7%	5 yrs	3 months	1,333	3.192 mil
3) BSFL whole live – Manual @ 104 tpa substrate	0.48 mil	0.33 mil	0.989 mil (Assumes 50% of \$4.99 / 25 g )	123% @ 10 yrs	1.8 yrs	12 months	9880 kg pa live larvae	0.584 mil
5) BSFL rendered – Mechanised @ 20 ktpa RMP substrate	3.4 mil	1.30 mil	1.617 mil	11.3% @ 25 years	11 yrs	2 months	433 meal 538 oil	0.318 mil
5) BSFL rendered – Mechanised @ 160 ktpa feedlot + RMP substrate	12.3 mil	5.78 mil	11.6 mil	47% @ 25 years	3 yrs	2 months	3,497 meal 5016 oil	5.79 mil
6) Water recycling	0.43	0.08 mil	0.604 mil	122%	0.8 yrs	0 months	140,888 Class A+ water	0.524

Due to the high levels of contaminants in RMP wastewater streams and the high pellet feeding costs, aquaculture was found to not provide an internal rate of return (IRR) as high as other options available. Whilst waste water streams available at RMPs show nutrient levels considered “good” (cattle wash and viscera table) and “permissible” (outfall to trade waste) that could be suited to horticultural operations, the high microbial levels in the water as well as the high capital and high labour costs means that the technical and financial viability of mushrooms could be lower than systems less susceptible to microbial levels and more automated / low labour horticultural operations.

Black soldier fly larvae (BSFL) operations show the strongest viability for small “niche” operations generating whole and live larvae and also at a large scale (160 ktpa or more of solid wastes) that warrants an automated / mechanised plant with rendering to create a meal (fish meal replacement) and oil. It was found that an automated / mechanised plant for an RMP (i.e. 20 ktpa of wastes) showed lower economic viability compared to the niche and large scale operations. Hence, there is numerical data to support the operation of a small whole larvae facility that could provide brood stock / strain optimization for a much larger BSFL facility producing meal and oil. The most important parameter for the viability of a BSFL project is the sale price of the products. A critical element that is not understood is how the commercial production of whole live BSFL would saturate the market hence decreasing the market value. Due to the infancy of the market, it is not possible to define the market value of Australian manufactured larvae meal and oil, however data from international operations was utilized.

Water recycling of selected “cleaner” wastewater streams (i.e. sterilization and viscera table water) for use in utilities (biofilter, wash downs, cooling towers, boiler make-up) exhibits an excellent economic proposition. Further refinement of the mass balance is required to understand the exact current potable water uses that can be switched to Class A+ water.

## 32 Upcoming Waste Streams in the Next 2-3 Years

Australian RMI Mega-trends:

- Industry wants to double the value of Australian red meat sales by 2030 as the trusted source of the highest quality protein;
- World leading environmental management e.g. CN2030;
- Reduced water usage creating more concentrated waste streams; leading to more intensive waste treatment processes onsite e.g. mechanical aeration;
- Onsite reuse / recycling;
- Stricter waste legislation reducing movement of wastes and increasing landfill levies; and
- Circular economy / industrial ecology: usage of wastes for co-located businesses e.g. composted manure for greenhouses; treated wastewater for intensive greenhouse / horticultural operations.

Key points on potential upcoming waste streams:

- Increased volumes of AD Digestate, with each stream having a unique microbial population. Covered anaerobic lagoons (CALs) are generally dug out every 4 to 5 years whilst continuous stirred tank systems (CSTRs) generate digestate continuously;
- Fats, oils and greases (FOGs) recovered from red streams for sale as lower grade tallow products i.e. biodiesel. The collection of removal of FOGs before DAF systems reduces the volatile fatty acid content thereby increasing the value of the FOGs;
- Feed lotting capacity expansions: more manure and slurry, hence more and larger slurry holding ponds; urban expansion leading to odour and vector issues with feedlots;
- Wastewater recycling leading to more concentrated waste streams e.g. reverse osmosis retentate with higher salinity;

- By-product devaluing: hair and hides with no / low viable market leading to use of this material in rendering with associated increases in render plant waste water;
- Waste / landfill regulatory changes: higher costs / limits on regulated wastes to landfills leading to higher demand for on-site processing
  - Use of paunch and other waste organics in multi-fuel boilers;
- Stricter trade waste limits on tannery and RMP waste waters
  - Concentration of contaminants into more concentrated streams e.g. electro-coagulation;
- Co-location of hydrogen electrolyzers providing “free issue” oxygen and thermal energy to intensify wastewater treatment; low cost ozone for waste treatment;
- Climate change threats:
  - Reduced rainfall but with more extreme weather events; could lead to pond flooding (i.e. release of runoff to surrounding surface water from feedlot retention basins and RMP WWT ponds);
  - Increased droughts leading to intensification of herds into feedlots for drought buffering. Increased feedlot manure and run-off;
  - Increased pathogen / virus monitoring on waste streams; potential for increased bio-security events;
  
- For changing from coal to biomass boilers, less ash tonnage but a cleaner ash more suited to use as a soil conditioner (alkaline pH) and/or blending with compost;
- For changing from natural gas / LPG to biomass boilers, more ash tonnage but a clean ash suited to use as a soil conditioner (alkaline pH) and/or blending with compost; and
- Pyrolysis: generation of char / ash.

### 33 Overall progress of the project

Project is tracking ahead of schedule and to budget, with no managerial interventions required of MLA at this stage.

### 34 Conclusions/recommendations

This milestone has identified the various solid, liquid, and gaseous effluents from the red meat supply chain including on-farm, saleyard, feedlot, and processing with preliminary indicative estimates on volumes of production for typical Australian wide beef and sheep operations. Waste value adding opportunities for each have been suggested, considering the different scales in the supply chain and how this will affect viability. Feedlots and processors are expected to have greater capacity to value add wastes, with saleyards and producers either not having sufficient scale, motivation (lower net thermal and electrical energy costs), or resources to allocate towards waste management.



These estimates can be used to inform and be refined by a survey and testing of industry wastes, on microbiological quality of processed waste and effluent.

## **35 Bibliography**

References are contained in footnotes throughout the report.