final report

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Alternative Onchocerca gibsoni detection and management in cattle

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Executive summary

Due to a perceived risk to public health, that dates back to the early 1900s when quarter beef was regularly exported to the United Kingdom, it was a Department of Agriculture and Water Resources (AQIS) requirement that all Onchocerca nodules be removed from infected beef. Nodules have no public health risk as infection is not transmissible to humans through consumption of affected meat. However, these "extensive inspection" arrangements remain largely in place in Australia, most likely resulting in a cost disadvantage against export competitors (e.g. US, Brazil, Argentina) into the same markets. Based on anecdotal information from both inspection and company staff, the prevalence of beef nodules has been decreasing over the last 20 to 30 years. Carton Meat Assessment records (Boneless Meat Inspection pre-2002) and overseas rejections also point to this downward trend in prevalence in finished product. Despite this information on loss to industry, regional risk and downward prevalence due to modern animal health management there has not been a proportional reduction in post-mortem interventions.

This project aimed to reassess prevalence of Onchocerca nodules (*Onchocerca gibsoni*) across Australia using abattoir surveillance, to re-classify risk areas previously established in 1967 (Seddon). The project also assessed the use of effective but not registered treatments for *Onchocerca sp.* and alternative methods of detection for Onchocerca nodules.

Surveillance was conducted in four abattoirs and boning rooms across Australia to cover the sourcing of cattle from the Northern Territories, South Australia, Queensland, New South Wales and Victoria.

Although this project does not support slaughter floor palpation of briskets as an alternative to boning room palpation and trimming due to a higher level of sensitivity (lesions being found) than specificity (accuracy), for staffing and/or overhead cost reasons this method of detection may still be more beneficial to some companies.

Anthelmintic treatments are not specifically used for *Onchocerca gibsoni* as there are no registered treatments for *Onchocerca* in cattle in Australia. Therefore, collection of this data by itself does not aid a prediction that animals may or may not be present with *Onchocerca gibsoni* nodules. Furthermore, it is not possible to ascertain the region the feedlot cattle were breed – without this information it is not possible to determine whether or not an induction at feedlot is effective to an existing infection noting that the majority of cattle are grain fed for only 100 days.

The research allows for re-classification of the prevalence of *Onchocerca gibsoni* which can be used by establishments to seek alternative arrangement against exist inspection requirements.

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1 Background

Due to a perceived risk to public health, that dates back to the early 1900s when quarter beef was regularly exported to the United Kingdom, it was a DAFF (AQIS) requirement that all *Onchocerca* nodules be removed from infected beef. Nodules have no public health risk as infection is not transmissible to humans through consumption of affected meat. However, these "extensive inspection" arrangements remain largely in place in Australia, most likely resulting in a cost disadvantage against export competitors (e.g. US, Brazil, Argentina) into the same markets. Based on anecdotal information from both inspection and company staff, the prevalence of beef nodules have been decreasing over the last 20 to 30 years. Carton Meat Assessment records (Boneless Meat Inspection pre-2002) and overseas rejections also point to this downward trend in prevalence in finished product.

Despite this information on loss to industry, regional risk and downward prevalence due to modern animal health management there has not been a proportional reduction in post-mortem interventions.

2 Project objectives

The project objectives are:

- Classifying regions on a risk-basis by demonstrating regional prevalence for *Onchocerca gibsoni* thereby informing inspection need
- Define post-mortem inspection in line with pre-slaughter health management that minimises lot-fed animal exposure in medium and high-risk areas i.e. lot-fed cattle as per the definition in the Export Meat and Meat Product orders
- Identify lots to which alternate procedures and arrangements may apply

3 Methodology

3.1 Validation trial design and collaboration

A summary on previous research and background information on onchocerciasis in cattle is provided in Appendix 1. The key points that effected the design of the project methodology were:

- Onchocerciasis in cattle does not pose a human health concern; it is not a food safety concern but rather a food suitability issue.
- *Onchocerca gibsoni* causes no clinical signs or effect to the cattle and therefore is of little significance to producers.
- Infection with *Onchocerca gibsoni* occurs 12 months prior to visible lesions.
- Research shows no significant difference in the attributes of sex, age, altitude or vegetation (with no mention of breed). However, this should be considered in light of the research being over 30 years old and in some cases small sample sizes of the research.
- Previous research showed a significant difference in the initial surveillance where there was an increase in prevalence believed to be due to an increase in expertise in detecting the nodules.

3.1.1 NAMP collaboration and prospective surveillance established

The National Arbovirus Monitoring Program (NAMP) was considered as it potentially offered supporting information to this project due to distribution of arbovirus vector species of insects.

This Program coordinated by Animal Health Australia aims to monitor the distribution of the three economically important arboviruses i.e. insect-borne viruses (bluetongue, Akabane and bovine ephemeral fever virus) and their vectors.

Animal Health Australia (2016), explain that across Australia, the program collects data from:

- serological monitoring of cattle in sentinel herd network
- strategic serological surveys of commercial cattle herds.

Participating cattle producers hang insect traps to identify whether *Culicoides* vectors occur in the area during the testing periods. The monitoring is available in Figure 1.

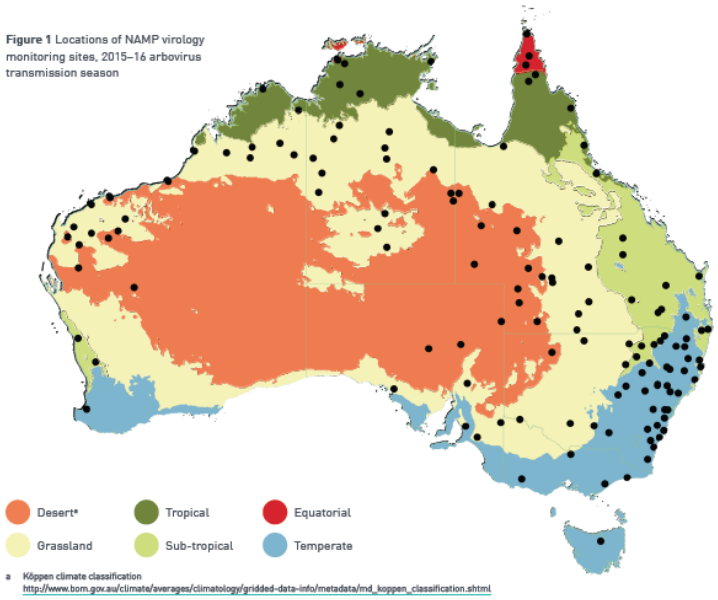

Figure 1: Locations of NAMP virology monitoring sites, 2015-16 arbovirus transmission season (sourced Animal Health Australia 2016).

This information is used to develop the bluetongue virus zone and the associated Australian Bluetongue Zone Map show in Figure 2 as well as the distribution maps of Akabane and Bovine Ephemeral Fever virus, published in the annual NAMP report (2016).

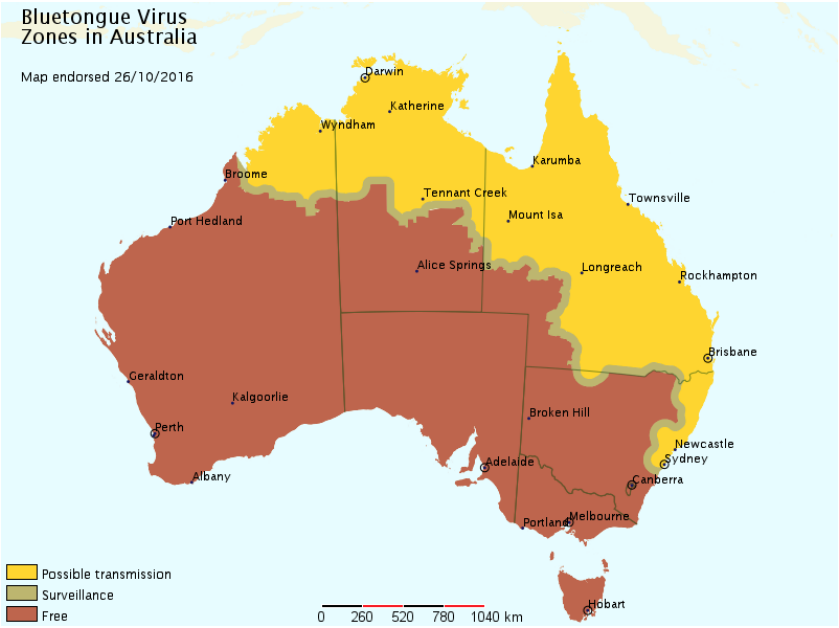

Figure 2: Australian Bluetongue Zone Map (sourced 11 March 2017 and subject to change).

This interactive map is significant to Australian agriculture as it facilitates the export of live cattle, sheep, goats and their genetics. The area or zone defined as bluetongue free is where no detections have occurred for two years. (Animal Health Australia, 2017) The maps (Fig. 3) are developed based on the World Organisation for Animal Health (OIE) guidelines. The bluetongue zonings are assessed and amended from the surveillance data captured through the National Arbovirus Monitoring Program. (2016)

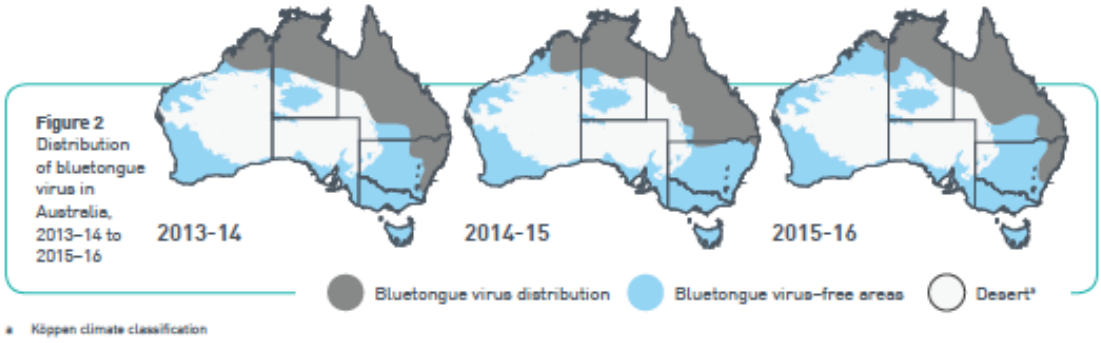

Figure 3: Distribution of Bluetongue virus in Australia, 2013-14 to 2015-16

NAMP collaboration on the Project

The program team met with the NAMP co-ordinator, to discuss collaboration with NAMP and establishment of surveillance for the vectors which may transmit *Onchocerca gibsoni*. During this discussion, how the bluetongue line is established became of primary importance if to be used as a measure of risk to meet the object of this project. Dr Langstaff explained that the Bluetongue zoning is predominantly based of the sentinel herd testing. Cullicoides (fly) traps are used to identify the specific types of flys (*Culicoides brevitarsis*, *Culicoides actoni*, *Culicoides dumdum*, *Culicoides fulvus*, *Culicoides wadai* and *Culex annulirostris*) which spread the arboviruses of significance. The fly trap information is used more for the likelihood of positive testing in sentinel herds and the future selection and positioning of sentinel herds rather than for the bluetongue zoning. Records do not support or substantiate regional demarcation based on fly population. The opportunity to use the

NAMP by broadening it's scope to collect information on possible vector(s) for *Onchocerca gibsoni* was also impractical given that there is no known specific vector other than a member of the *Cullicoides* species i.e. a biting fly.

On this basis the maps developed by Seddons (1967) (Fig.2 in Appendix 1) with regional prevalence holds more value, if revalidated to provide more up to date information on the prevalence of *Onchocerca gibsoni* throughout Australia.

3.1.2 Approved anthelmintic program developed

Under current Australian husbandry practices 80% of cattle are treated with an ivermectin based wormer at weaning (Carr et. al, 2011). On average cattle are weaned between 6-12 months of age depending on the producer's management practices and location of the property. This means that an initial ivermectin treatment at weaning could result in cattle not showing clinical signs (i.e. brisket nodules) until 18-24 months of age, without any additional treatment.

It is also known that the feedlot industry has well established induction programs, which include the available ivermectin treatments options that would be effective for *Onchocerca gibsoni* (Appendix 2).

It is understood commercially that interventions i.e. handling, husbandry practices (such as drafting) or treatment protocols (including preventative treatments) result in a reduction in feed conversion i.e. yield and the potential stress resulting in a reduction in meat quality. For this reason, without compensation producers and feedlotters are very reluctant to add additional treatment protocols unless there is a clear benefit to them. In the case of onchocerciasis, no visible clinic signs or effect on the cattle is evident. It is concluded that without compensation to offset the production losses there is no incentive for producers or feedlotters to include an additional treatment protocol to address *Onchocerca gibsoni*.

However, through discussion with a number of the feedlot vets it is evident that their recommendation and therefore, the existing induction protocols include ivermectin treatment options that would be effective for *Onchocerca gibsoni* (Appendix 2). These can however vary between feedlots as commercial outlets offer substitution of other derivatives at a lower cost.

Therefore, this study aimed to target surveillance of cattle sourced from feedlots with established ivermectin treatment programs that meet the available ivermectin treatments options that would be effective for *Onchocerca gibsoni* (Appendix 2). These cattle were also to be sourced from the higher prevalence areas as much as possible given the location of Australian feedlots.

3.1.3 Feedlot and processor collaboration confirmed

Collaboration was confirmed by processing establishments that source from across the country and collectively cover the majority of Australia (Fig 4). Processing establishments without alternative arrangement were preferenced. However, it is known that processing establishments that source cattle out of central South Australia and the southern areas of the Northern Territory already hold alternative arrangements. It is not commercially viable to ask these establishments to change their practice back to traditional/historic inspection, however original validation of the alternative arrangement will be sourced. In addition to this, three feedlots confirmed their participation.

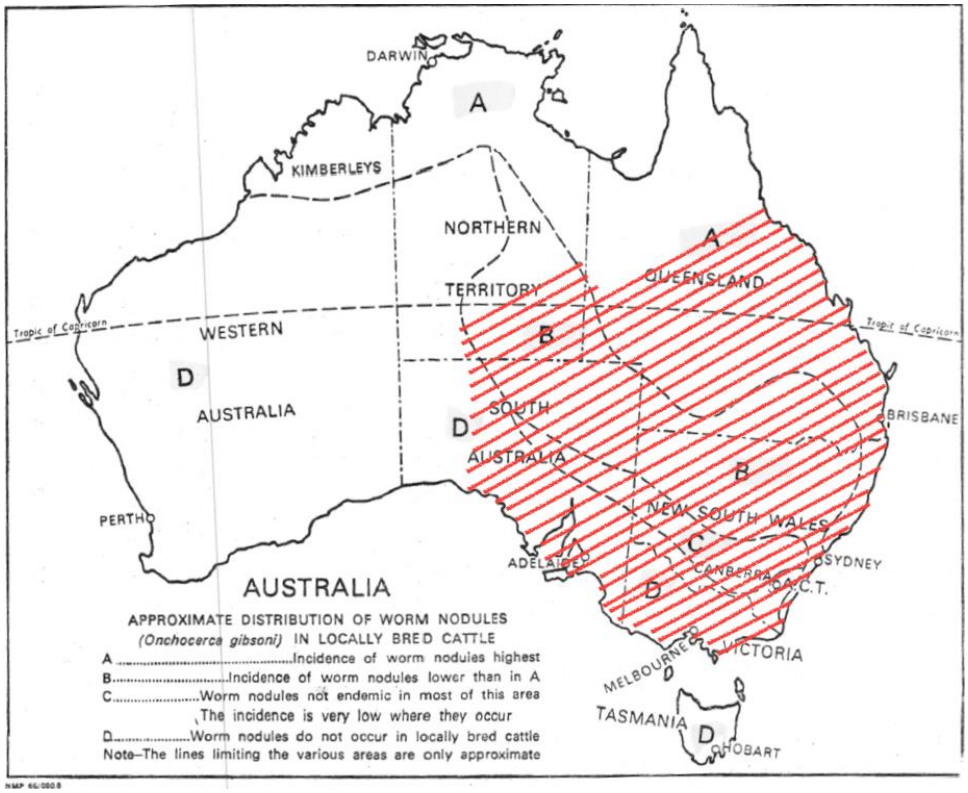

Figure 4. A Map of Australia Zoning *Onchocerca gibsoni* prevalence annotated with the sourcing area (shaded) of the collaborating processing establishments.

3.1.4 Alternative inspection procedure agreed

Providing commercial consideration to the fact that a number of processing establishments hold Department of Agriculture and Water Resources approval for alternative inspection arrangements, a similar alternative inspection procedure was adopted to that in the existing alternative arrangements that were validated through this project.

It is expected that following this project companies will conduct an assessment of contributing factors prior to hazard identification and mitigation steps as detailed in section 8.1.5 (Appendix 1). As imaging technology improves it is expected that processing establishments may want to internally validate these methods of identify the risk.

Sample Alternative Procedure for Hazard Identification that was to be conducted:

On the slaughter floor: Briskets will be palpated and partially boned by freeing from the sternum. If any nodules or worms are found on the cut surface or within the brisket the carcase is identified and assessed as positive to be 'fleeced' out in the boning room.

In the boning room: All carcases identified on the slaughter floor will have all obvious nodules removed through 'fleecing' the briskets.

Boned out brisket products (to be produced with the red bark left in situ) destined to be exported intact will be assessed by palpation of the lateral and medial surfaces only for presence or absence of nodules. In the unlikely event a nodule is detected, procedures would then be such that the red

bark would be fleeced to allow further inspection of the affected piece of product. This product would then be diverted into other meat packs.

3.1.5 Trial protocols established and agreed

Given the research findings the revised aims of the trial protocol were to:

- Re-validate the prevalence of *Onchocerca gibsoni* through the presence or absence of brisket nodules based on Seddon's map (1967),
- Validate that palpation of the brisket is as effective at identifying the risk of onchocerciasis as 'fleecing' of briskets, and
- Validate that any of the available anthelmintic treatment options (Appendix 2) administered within the last 12 months are effective at preventing the presentation of nodules in the brisket.

The following protocol was established and agreed for the trial

Field Surveillance General Method: An entire production day would be sampled from each of the processing establishments. This approach was expected to provide approximately 5000 records, from approximately 100 properties. Given the finding of Holdsworth and Moore (1985) the collection team would start at a processing establishment in the north of the country with a high prevalence to ensure expertise in detecting the nodules. The sampling team would be maintained as a constant throughout the surveillance.

No sampling bias was to be placed on the collection of data based on age, sex, vegetation, altitude of supplying property given that previous research has shown no significant statistical difference, however analysis would be conducted on the surveillance data to validate age and sex given that this research is over 30 years old. Also no sampling bias was to be placed on rainfall of the supplying property or seasonality given the findings of Ottley and Moorhouse (1980). Given the current commercial market for cattle the sampling model was not be accounting for breed.

Re-validation of Prevalence: As much as possible collection days were to occur on production days where the processing establishment sourced more vendor breed cattle. The reason for this was to avoid cattle that had been moved between areas (Figure 4) as it is impossible to know where the animal was infected (if it was). Because nodules take a long time to manifest "no nodules" doesn't necessarily mean "no infection" for all PICs the animal has been moved between. Ideally, a grid-based sampling program would be implemented to cover the target area fully. However, this was logistically not possible and hence opportunistic abattoir-based surveillance was selected. Therefore, abattoir visits were aimed at maximising direct vendor-bred consignments (if / as far as possible), though this could not be guaranteed.

The collected data was to be collated with additional data obtained directly from establishment records on:

- PIC and RFID
- breed
- sex
- dentition data
- body number on the slaughter floor correlated to carcase number in the boning room.

Further sourcing records on the movement of these cattle were to be sourced with MLA's assistance through NLIS.

Validation of an alternative inspection method: At each processing establishment the first day was to be spent on the slaughter floor following the alternative inspection procedure for briskets (Section 3.4) identifying the presence or absence of nodules. One person would palpate the briskets on the slaughter chain whilst a second records any findings from that and the incision. On the second day these same carcases were to be assessed in the boning room during the palpation and 'fleecing' process again to identify presence and absence. One person would palpate the boned brisket whilst the second recorded this information and recorded any findings from the fleecing of the briskets. These later findings would include any other lesions or abnormalities. This means that the same carcases are being assessed through the surveillance.

At processing establishments with alternative arrangements in place the initial validation information for equivalence will also be sourced. It should be noted that the original accepted Seddon data was gathered from both palpation and boning data surveillance.

Validation of available anthelmintic treatments: To ensure 95% confidence that post anthelmintic treatment prevalence was reduced to less than 0.5% a sample size of 598 cattle from treated animals would be collected. The majority of feedlot cattle are produced through zones B, C and D of Seddon's map (Fig 6), as such this could bias the validation of treatment. In an aim to prevent this, it was aimed to collect the majority of feedlot data to validate treatment options from zone B where the existing prevalence is higher (noting this may result in additional collection days). Treatment records would be sourced for feedlot cattle.

3.2 Preparation for the validation trials

Confirmation was provided by processing establishments to allow access and assistance to collect data which covered the sourcing area as shown in Figure 1. The request was made for labour where possible to partially bone the brisket from the sternum prior to chilling of the carcases. Dates were also requested that allowed for the greatest amount of direct sourced and vendor breed cattle. The reason for this is to avoid cattle that have been moved between areas (refer to Figure 4) as it is impossible to know where the animal was infected (if it was). Because nodules take a long time to manifest "no nodules" doesn't necessarily mean "no infection" for all PICs the animal has been moved between. Establishments with alternative arrangements in place were asked for a copy of their initial validation information for equivalence.

Given the finding of Holdsworth and Moore (1985) the collection team started at a processing establishment in the north of the country which predominately sourced cattle from the high prevalence areas of northern and central Queensland and the Northern Territories to ensure expertise in detecting the nodules. Appendix 4 provides photos of *Onchocerca gibsoni* nodules (whole and incised) from briskets. The nodules varied in sizes from 0.5mm to 3cm in diameter, with a firm pea like consistency, predominantly in the smaller nodules, to a capsuled gelatinous consistency similar to a lymph node, however when cut open tightly knit adult worms could be seen.

3.3 Validation trials - Data collection

Data was collected from four processing establishments who's cattle sourcing practices normally covers the majority of the country as shown in Figure 4. The data collected was sourced from 104 properties. The sampling team was maintained as a constant throughout the surveillance.

Two days were spent at each processing establishments, the first on the slaughter floor and the second day in the boning room. In line with the agreed alternative inspection procedure in Milestone 2, all carcase sides had the full brisket palpated on the slaughter floor. Where company labour allowed, the point end of the brisket was partially boned by freeing it from the sternum, this was only available at one of the four establishments given the down turn period. When any nodules were found on the cut surface or identified through palpation within the brisket of the carcase, this was recorded on the Slaughter floor data recording sheets as 'Y' (Appendix 5). An observation on the approximate size and location of the nodule was also recorded. A number of worms were also observed on the briskets of carcases at two of the plants. These were recorded as an observation however only reported as 'Y' where a corresponding nodule was also palpable. This was because the worm could have been due to either infection with *Onchocerca gibsoni* or due to transfer between carcases during processing. To ensure no operator bias in the results of palpation, the team member palpating the carcase had no prior knowledge of the kill agenda and therefore region the cattle had come from. However, it should be noted that palpation may have been a more sensitive than specific method of detecting for *Onchocerca gibsoni* nodules as any enlarged or reactive thoracic lymph nodes would have palpated as a larger, gelatinous nodule.

In the boning room on the second day, all carcases were assessed for presence or absence of nodules using the usual process of palpation and incision/'fleecing' and palpation verification. At all four establishments there a minimum of two points of data collection required either due to two boning chains or the point end of the brisket being boned at a difference area of the chain to the bible end of the brisket. Team members discussed the surveillance project with the boners prior to the start of the shift, asking them to notify the team if any nodules were detected through the shift. At one establishment there were 3 boning chains and on this occasion a company staff member that had previously been a boner on this task, was briefed on the project and assisted in the recording of nodules on the Boning Room data Recording sheets (Appendix 6). Each carcase entering the boning room had an entry marked against it, as by the stage in the boning where identification and recording of data was occurring there was no body number.

At each plant the kill agenda and boning room 'scan in' were requested for the collection days as the order of carcases entering the boning room is not in numerical sequence but in product type and grade. At Plant A this resulted in a reduction of the data set as with two shifts not all the carcases from the first shift of the slaughter floor were boned out in 12hours of recording (a shift and a half).

The collected data was collated with additional data obtained from the establishment records on:

- PIC and RFID
- sex
- dentition data

As breed information was not available for all animals, hump height data was used as an indication of tropical breed content (TBC) and therefore whether the breed of cattle was:

- Unknown (where animal was not MSA eligible and a hump height was not available)
- British breed (where hump height is 45mm or less)
- Crossbreed (where hump height is greater than 45mm but less than 120mm)
  Note: this therefor also includes breeds with tropical breed content (e.g. Droughtmaster, Charbray, Brangus) and crossbreeds (e.g. common crossbreeds such as Santa X Braford, Angus X Santa etc.)
- Tropical breed (where hump height is 120mm or greater)

The PICs provided were used to determine the region within the State where the property was located. For the centre of each region (either the administrative centre i.e. shire council or geographical centre) was used to calculate the latitude and longitude.

Note: The raw data has not been provided as it includes confidential and locational information (e.g. PICs, latitude, longitude) that may allow the identification of the properties and/or producers of the cattle that were assessed as a part of this trial.

As indicated in Milestone 2, anthelmintic treatments are not specifically used for *Onchocerca gibsoni* as there are no registered treatments for *Onchocerca* in cattle in Australia. Therefore, treatments specifically effective on the microfilaricidal stage of the life cycle is rare. Therefore, collection of this data by itself does not aid a prediction that animals may or may be present with *Onchocerca gibsoni* nodules. Furthermore, it is not possible to ascertain the region the feedlot cattle were breed – without this information it is not possible to determine where or not the induction is effective to an existing infection noting that the majority of cattle are grain fed for one 100 days. In addition, one of the establishments/feedlot permissions advised during collection in the boning room that they held an approve variation to the requirements.

Following initial analysis further sourcing records on the movement of cattle were obtained from cattle coming from regions that had a higher than expected prevalence. This information was provided through individual NLIS searches, with the PICs provided once again used to determine the region within the State where the property was located. For the centre of each region (either the administrative centre i.e. shire council or geographical centre) was used to calculate the latitude and longitude. This was done to verify the vendor-bred status of the cattle.

3.4 Data analysis

The data were provided in a Microsoft Excel spreadsheet, and were imported into the statistical software R, v3.5.0 (R Core Team, 2018) for all data manipulations, graphics and analyses. An overview of the information collected is provided in Appendix 7 and the data analysis report is provided in Appendix 8.

The data were loaded into the R software, combining the separate plants; one data set was prepared for the slaughter floor data and one for the boning room data (including plant identifiers). Separately for each data, the detection of *O. gibsoni* was calculated for each carcase as follows:

- For slaughter floor data a carcase detection was recorded if lesions were noted for either of the two sides

- For boning room data, a carcase detection was recorded if nodules were trimmed or detected via palpation (after trimming) for either side.

There are a range of graphical and estimation methods available for spatial data in the R software, through the provision of add-on packages. In particular, for the estimation of prevalence maps the PrevMap package by Giorgi and Diggle (2017) was used, which allows spatial estimation of prevalence maps, including incorporation of covariates.

Diggle and Giorgi (2016) note that a typical feature of most geostatistical problems is a focus on prediction rather than on parameter estimation. As a result, these authors use the standard (bi-nomial) generalised linear models (GLMs) to determine which covariates to include in the geospatial model; a similar approach is used here.

4 Results

4.1 Simple data summaries

A summary of the number of carcases inspected on the slaughter floor and in the boning room, by plant is presented in Table 1. From this table it can be seen that not all carcases on the slaughter floor were captured in the boning room, which is the primary source of information for the prevalence map update. In addition, there were 162 carcases with multiple PICs, which were removed from the data set as they were not vendor-bred, leaving a total of 3,271 carcases for analysis.

Table 1: Summary table of the number of carcases recorded at each plant on the slaughter floor and in the boning room.

Plant	Slaughter Floor	Boning Room
A	825	617
B	1115	1092
C	665	665
D	1187	1059

A summary of the number of carcases inspected, number and percentage of *O. gibsoni* detections in the boning room and on the slaughter floor by sex, breed and dentition, are shown in Table 2. While there appear to be some differences in boning room prevalence between male and female cattle, breed and dentition, these may be correlated with geographic location and hence cannot be interpreted on their own. Interestingly, slaughter floor prevalence is higher than boning room prevalence and the differences between sexes, breeds and dentition are much smaller.

Table 2: Summary table of the number of carcases recorded, number and percentage of *Onchocerca gibsoni* detection in boning room and on the slaughter floor.

	Number of Cattle	Slaughter Floor		Boning Room	
		Det.	%	Det.	%
All					
Total	3271	856	26.2	619	18.9
Sex					
F	1007	250	24.8	317	31.5
M	2264	606	26.8	302	13.3
Breed					
British	12	3	25.0	3	25.0
Cross	2247	580	25.8	320	14.2
Tropical	506	151	29.8	133	26.3
Unknown	506	122	24.1	163	32.2
Dentition					
0	562	140	24.9	51	9.1
1-2	1321	369	27.9	130	9.8
3-4	621	135	21.7	92	14.8
5-6	218	38	17.4	40	18.3
7-8	549	174	31.7	306	55.7

Across the four abattoirs, the animals were sourced from 64 unique PICs. However, due to confidentiality reasons, the geographical location of these PICs could not be used. Nevertheless, it was possible to obtain the PIC region, an administrative grouping of geographically close properties, that each PIC belongs to and an associated geographic location (latitude and longitude), which can be used for mapping. There were a total of 37 PIC regions and hence geographic locations. Given this limited number of locations, which are primarily along the east coast of Australia (see Figure 6) it will be difficult to estimate an updated prevalence map of *O. gibsoni.*

On a PIC basis, the median number of animals inspected was 25, the average was 52.7, while the minimum and maximum number of animals inspected were 1 and 354, respectively. On a PIC region basis, the minimum and maximum number of animals inspected were 1 and 485, respectively, while the median and mean were 62 and 91.2, respectively. A probability histogram of the number of cattle inspected per PIC region is shown in Figure 5.

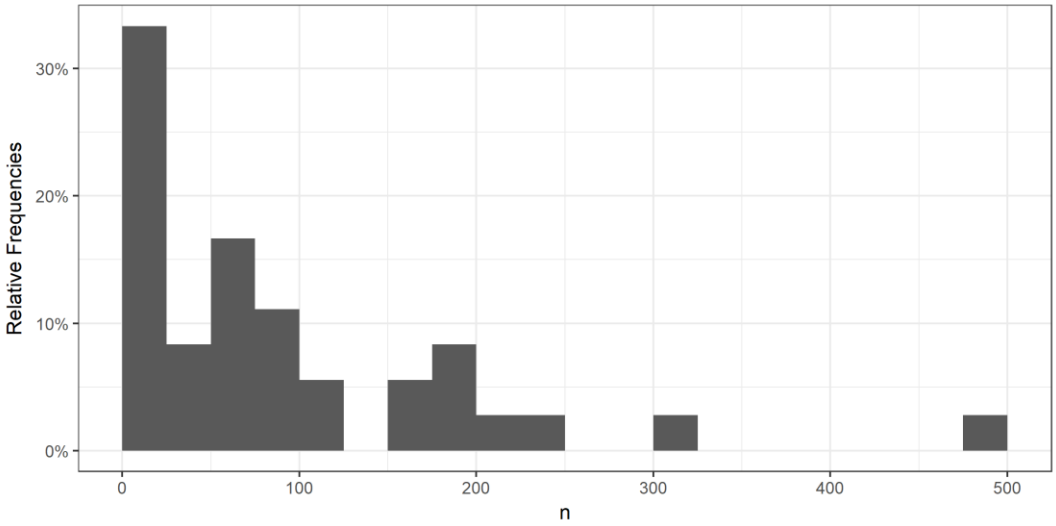

Figure 5: Histogram of the number of cattle consigned per PIC region.

4.2 Prevalence map of *O.gibsoni* in Australia

A Google map was downloaded covering the various PIC regions that were contained in the survey. The map, indicating the location of each of the sampled PIC region centres, is shown in Figure 6. From this map it can be seen that the majority of data is close to the east coast of Australia. Estimation of the prevalence of *O. gibsoni* in regions where data points (i.e. sampled PIC areas) are close together is likely to be better than in regions where there are no data points or where they are far apart.

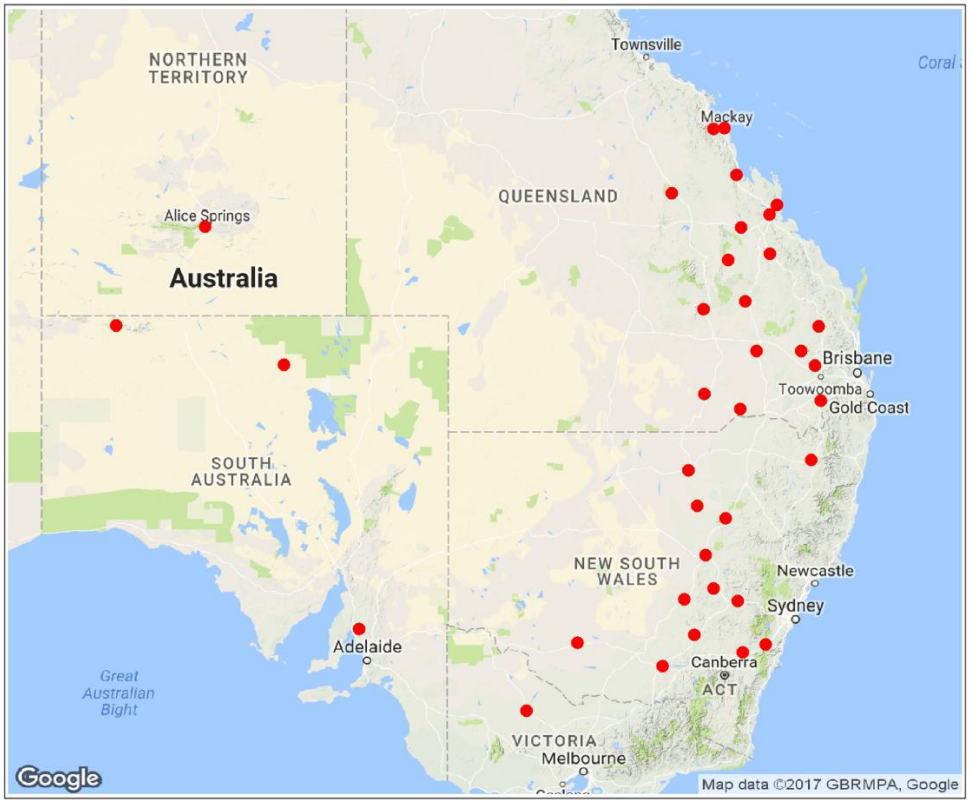

Figure 6: Map of Australia shown the location of each PIC region included in the survey.

Similar maps, showing the number of carcases inspected and the estimated prevalence, for slaughter floor and boning room data, respectively, are provided in Figure 7 and 8 – in these plots the size of the dot relative to the size of the number of carcase sampled and the colour corresponds to prevalence bands. While the boning room data seems to show a reduction in prevalence from north to south, the same pattern is not apparent from the slaughter floor data.

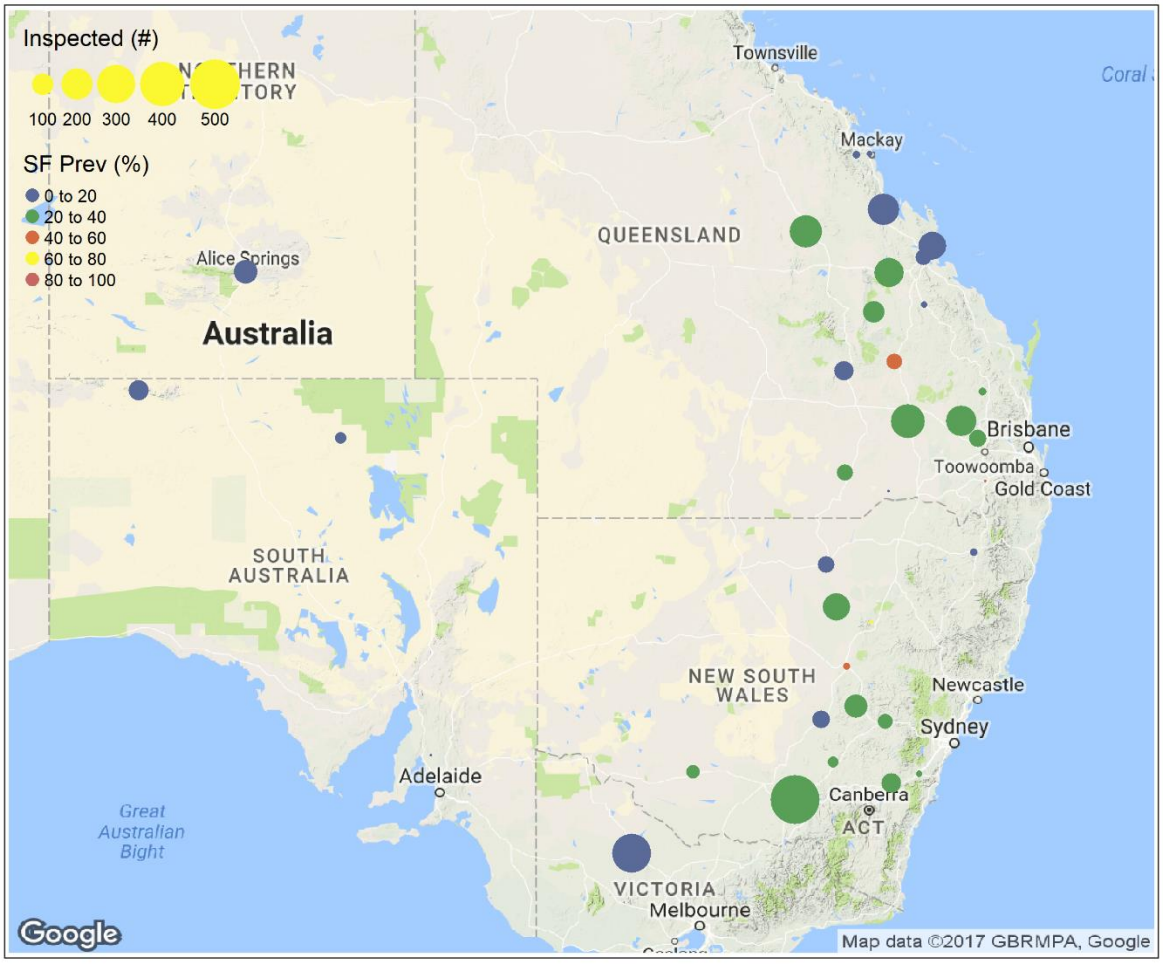

Figure 7: Map of Australia showing the location of each PIC region and estimated prevalence from slaughter floor data

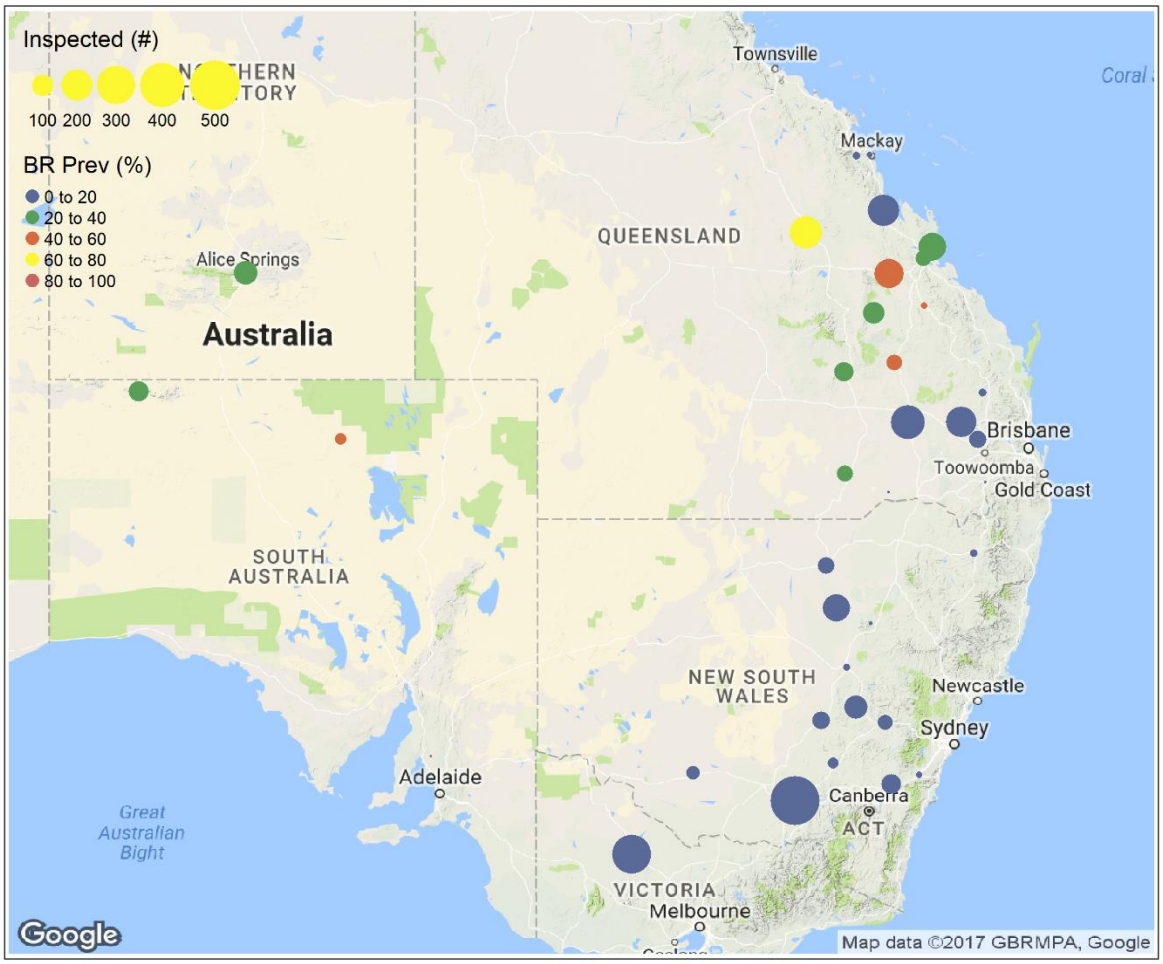

Figure 8: Map of Australia showing the location of each PIC region and estimated prevalence from boning room data

5 Discussion

5.1 Key findings

During this trial palpation was conducted on both the slaughter floor and boning room. Palpation was found to be a more sensitive than specific method of detecting for *Onchocerca gibsoni* nodules on the slaughter floor as any enlarged or reactive thoracic lymph nodes could have palpated as a larger, gelatinous nodule. This could have contributed to the difference in prevalence rates that were demonstrated on the slaughter floor compared to the boning room due to the false positives detected by palpation on the slaughter floor. On this basis it is not recommended that slaughter floor palpation be used as an alternative inspection method to boning room trimming and palpation.

In order to consider the prevalence levels of *Onchocerca gibsoni* detected in this project to the prevalence levels reported by Seddon (1967), Figure 2 of Appendix 1 has been overlaid onto Figure 8 (Figure 9). Despite the different map projections this shows that the results of this project do not support the prevalence previously reported in Seddon (1967). While the Seddon map divides the lower part of Australia into multiple zones with prevalence rates ranging from 0% - 77%, the results of this study demonstrated prevalence of 0 – 20% for Southern Australia through to the NSW – Queensland border with the line circling the south-east Queensland region. This allows for a re-

classification of regional prevalence as shown in Figure 10. It is noted that pure epidemiological principals could be used to dispute this re-classification due to the sample set being relatively small in comparison to the 25 million head cattle herd (ABS 2016, cited by MLA 2017), or the lack of a targeted grid-based sampling framework to ensure full coverage of the country, or the limited knowledge and therefore consideration of *the Onchocerca gibsoni* lifecycle, or the limitation of access to traceback information on all cattle. However, all of these limitations were also present in the initial prevalence mapping by Seddon (1967) with the addition of live-cattle palpation also being used to develop the prevalence levels which is shown here to be highly sensitive, however, not specific.

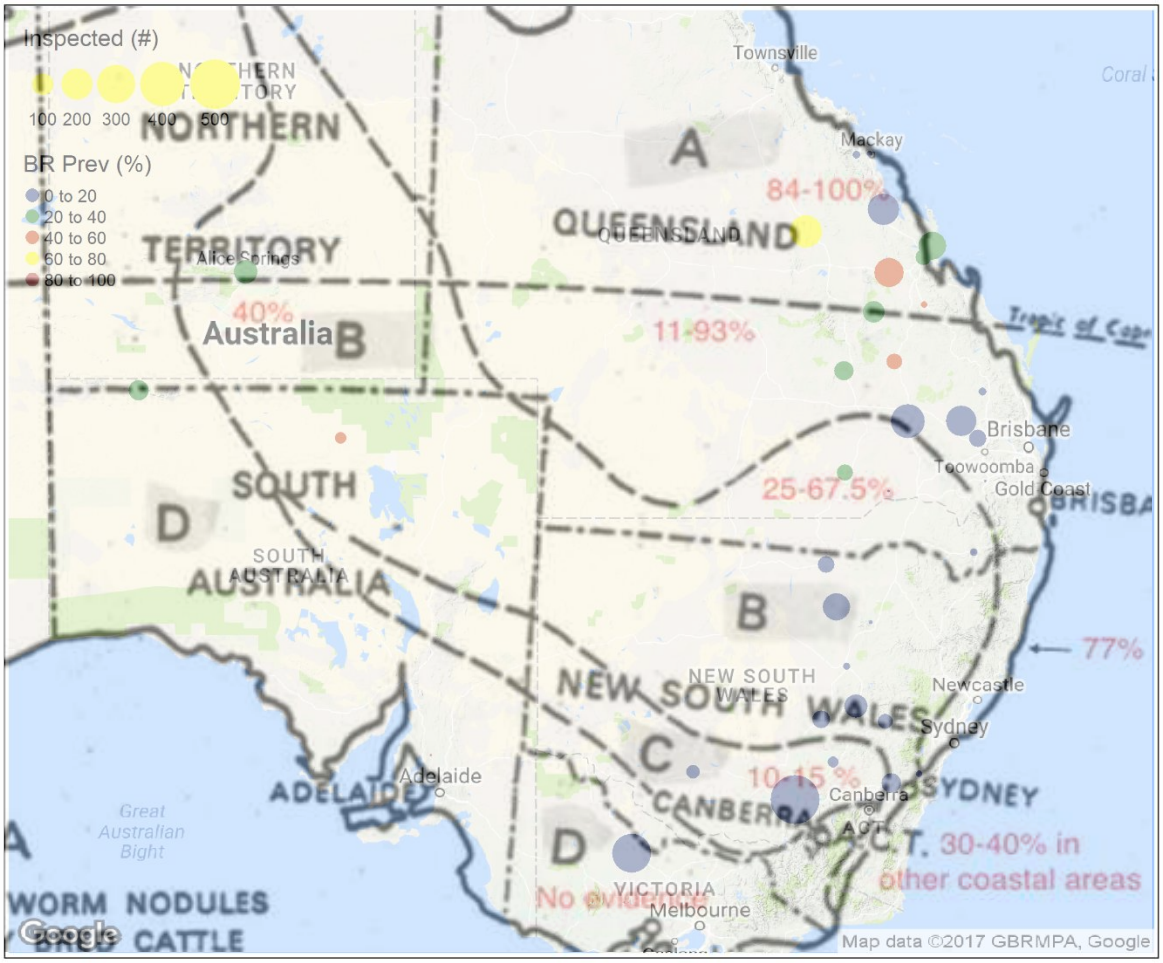

Figure 9: Comparison of project prevalence to Seddon prevalence from 1967.

6 Conclusions/recommendations

Although this project does not support slaughter floor palpation of briskets as an alternative to boning room palpation and trimming due to a higher level of sensitivity (results being found) than specificity (accuracy), for staffing and/or overhead cost reasons this method of detection may still be more beneficial to some companies.

Anthelmintic treatments are not specifically used for *Onchocerca gibsoni* as there are no registered treatments for Onchocerca in cattle in Australia. Therefore, collection of this data by itself does not aid a prediction that animals may or may be present with *Onchocerca gibsoni* nodules. Furthermore, it is no possible to ascertain the region the feedlot cattle were breed – without this information it is

not possible to determine where or not an induction at feedlot is effective to an existing infection noting that the majority of cattle are grain fed for only 100 days.

The research allows for re-classification of the prevalence of *Onchocerca gibsoni* (Figure 10) which can be used by establishments to seek alternative arrangement against exist inspection requirements.

Figure 10: Annotated Google map to indicate the high and low prevalence areas for *Onchocerca gibsoni.*

7 Acknowledgements

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9 Appendix

9.1 Appendix 1: Background of *Onchocerca gibsoni*

9.1.1 Onchocerciasis in Cattle

Onchocerciasis is a condition of a number of species including cattle. It is caused by a nematode (round worm parasite) of the *Onchocerca* species. There are a number of *Onchocerca* species which affect cattle including *Onchocerca gibsoni*, *Onchocerca lienalis* (also known as *Onchocerca gutturosa*), *Onchocerca ochengi* (also known as *Onchocerca dermata*), *Onchocerca armillata*, *Onchococerca dukei* and *Onchocerca stilesi* (Taylor et al 2015). The most economically significant of these to Australian beef production is *Onchocerca gibsoni* as it is the cause of nodules in the brisket of cattle.(Seddon 1967 and Andriessen 2012)

Since 1911 it has been understood that beef nodules were caused by a threadworm (equivalent to a nematode), i.e. an *Onchocerca* sp. and are found in the brisket, both superficial subcutaneous tissue and intermuscularly and on the external surface of the hind limb (Anon 1911). Through whole carcase dissection, Copeman (1978) found that more than 90% of onchocerca lesions are in the brisket. Seddon (1967) clearly describes the location of the nodules found in the brisket as

'*mainly in the triangle formed by the junction of the ribs with the costal cartilages, especially between the fourth and sixth ribs, but often extending forward to the second rib, or posteriorly to the tenth rib*'.

These nodules are formed as a 'worm nest' consisting of tightly coiled and knotted worms. Within these nodules there are a minimum of two worms and at least one of each sex. The nodule are described as presenting between 0.5 – 5 cm in diameter, firm, fibrous, singularly or in clusters (Figure 1). The nodules can contain, live or dead worms, or present as calcified lesions in older lesions. (FAO 1994 and Andriessen 2012).

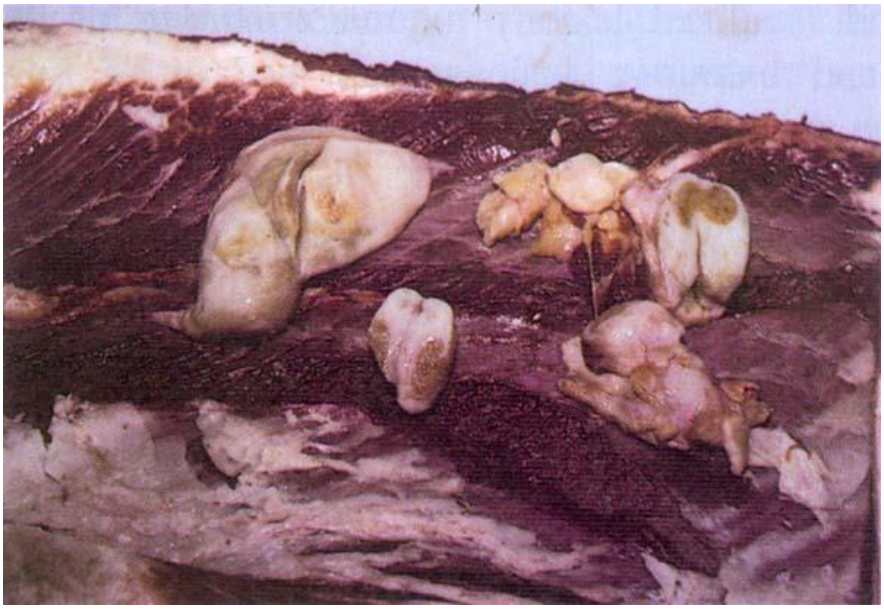


Figure 1: Fig. 94 Firm fibrous nodules of *Onchocerca gibsoni* in the brisket of an ox. (FAO, 1994)

The worms are between 3-5cm if male and 14-20cm if female, with reports of worms more than 50cm in length. Taylor et al (2015) describe the male worms as having a tail with a ventral curve, bearing lateral alae with 6-9 papillae at either side and spicules of unequal size.

These worm 'nest' nodules can be identified through palpation of the affected region, i.e. brisket during ante mortem or on freshly killed carcases (FAO 1994 and Seddon 1967).

9.1.2 Epidemiology of Onchocerciasis

There is limited information known about the life cycle of *Onchocerca gibsoni*. The nodules contain adult worms. The fertilised females shed microfilariae into the cattle's surrounding tissue. These microfilariae migrate through the connective tissue to the upper dermis. A biting insect vector (the intermediate hosts) takes up the microfilariae, which develop to a larvae infective stage. The larvae are then transferred back to cattle by the biting insect feeding on the next animal (FAO 1994, Taylor et al 2015 and Mehlhorn 2001).

The intermediate host is understood to commonly be of the genus *Cullicoides* i.e. a biting midge or fly. However, the specific species of biting insect acting as the intermediate host and vector for *Onchocerca gibsoni* is unknown. Although research attempts were conducted through the 1960s-1980s to identify the vector, Lee et al (1963) strongly suspected *Culicoides brevitarisis* as the vector. However, Ottley and Moorhouse (1980) proved this incorrect whilst demonstrating through laboratory transmission that *Forcipomyia (Lasiohelea) townsvillensis* (also known as *Culicoides townsvillensis*) may be a vector while Holdsworth and Moorhouse (1985) suggest that more knowledge is needed on the originating geographical areas for infected cattle before assessment of vectors be made. This area of research seems to have stopped mostly due to the development of anthelmintic treatment options being developed in the 1990s. Although a significant finding during this period of research was that infection occurs 12 months prior to visible lesions (Ottley and Moorhouse 1980).

In 1911, it was reported in a scientific explanation that beef nodules, i.e. onchocerciasis was found with equal numbers in 2-4 year old bullocks as in older cattle, however this article is not clear as to whether this was refering to the prevalence of the condition or the actual number of lesions in the cattle. (Anon 1911)

In the 1970s bovine onchocercal infections in Queensland was believed to be related to breeds, sex and age of cattle (Beveridge et al, 1979, Ladds et al 1979a,b.), with the view that infection increased significantly with age (Ladds et al 1979b). Holdsworth and Moorhouse (1985) sampled 13,665 cattle from the south east Queensland area from June to November 1982, assessing age (2 to 8 years), sex, property latitude, longitude, altitude and vegetation, noting that rainfall would be insignificant as transmission would have occurred 12 months prior to visible lesions. In this study there was no significant difference in the attibutes of sex, age, altitude or vegetation. No mention is made in this study to breed. There was however a significant difference in the initial 6 weeks of surveillance where there was an increase in prevalence which was believed to be due to an increase in expertise in detecting the nodules.

9.1.3 Prevalence

Onchocercosis is known to occur widespread through tropical and subtropical regions of Asia-Pacific countries, Norhtern and Southern Africa, Europe and USA (FAO 1994 and Merial 2017).

Since 1911 it has been known that the prevalence of onchocerciasis (known at the time as beef nodules) decreases down Australia with little to none found in Victorian bred cattle. (Anon, 1911).

Seddon (1967) provides the detailed information for Australian prevalence on *Onchocerca gibsoni* in Part 1 of Domestic Animals in Australia – Helminth Infestations. This information is summarised in Fig. 2. Seddon's summary of prevalence is based on surveys conducted between 1915 and 1963, describing that some of these surveys included identification of infection by palpation whilst other surveys were through identification of *Onchocerca gibsoni* at slaughter and boning.

It is expected that this distribution of the disease and the variations in prevalence are due to the availability of the vector(s) and stock movements through the country at the time of the research.

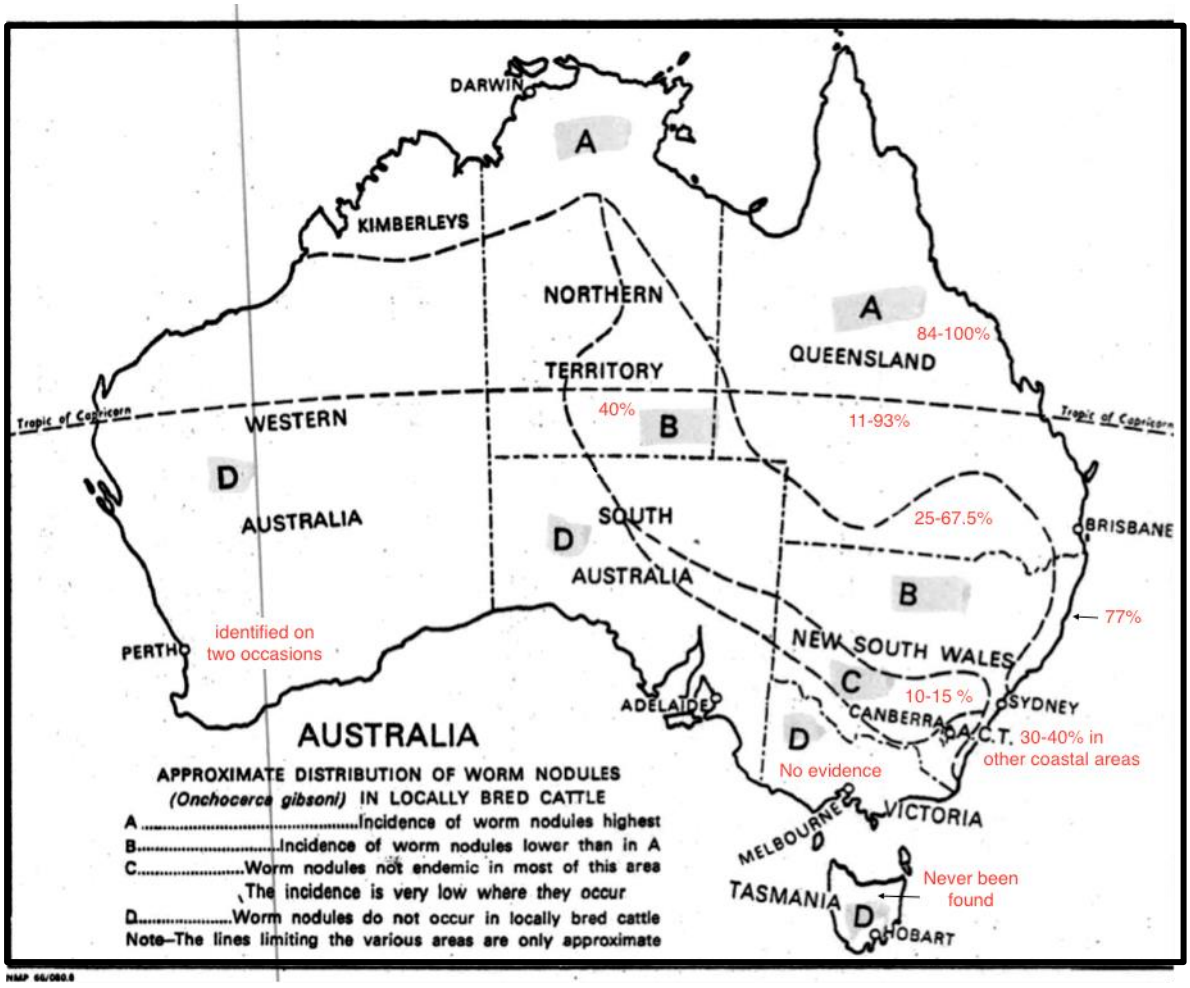


Figure.2: A Map of Australia Zoning *Onchocerca gibsoni* Prevalence annotated based on the associated text. (Seddon, 1967)

In 1979, Ladds et al, found highly significant differences in the prevalence of infection between farms. Although in subsequent research, Holdsworth and Moorhouse (1985) found no statistical difference in prevalence for 13,665 cattle sampled during slaughter from 35 properties around the Lockyer Valley. A lower rate of infestion of 24.5% was also found by Holdsworth and Moorhouse in 1985 through the Lockyer Valley compared to Seddon's earlier summary, which at the time was believed to reflect a difference in vector species.

Anecdotally over the last 20-30 years the presence of nodules and therefore prevalence of *Onchocerca gibsoni* has been decreasing, based on information from both government inspection and processor company staff. This decrease is also evident by the decrease in identification during carton meat assessment records (as Boneless Meat Inspection prior to 2002) and overseas rejections. It is hypothesised that this decrease in prevalence may be due to the introduction and use of anthelminthic treatment options.

9.1.4 Available Anthelmintic Treatment Options

Mehlhorn (2001) provides the following tabulated information on the treatment of *Onchocerca gibsoni* in cattle:

Nonproprietary Name	Dose and Delivery Mode	Mechanism of Action
Avermectins - Ivermectin	0.5mg/kg pour-on or 0.2mg/kg subcutaneous (i.e. injection) or orally (i.e. drench)	Microfilaricidal effects i.e. kills microfilariae
Milbemycins -Moxidectin	0.2 mg/kg; subcutaneous	Microfilaricidal effects
Piperazine Derivatives – Diethylcarbamazine (DEC)	50mg/kg intramuscular injection or 22mg/kg intramuscular injections for 3days or 40mg/kg intramuscular injections or or orally for 3 days	Microfilaricidal effects

Table 1: Summary of Anthelmintic Treatment Options

The microfilaricidal mechanism of action of the treatment means that the spread of *Onchocerca gibsoni* through to the vector is prevented and therefore the spread of the condition is decreased and in turn decreases the prevalence of the disease. This is expected to have contributed to the anecdotal decrease in prevalence of *Onchocerca gibsoni*.

However, the Australian Pesticide and Veterinary Medicine Authority have no treatments registered for *Onchocerca gibsoni* treatment. This is most likely due to *Onchocerca gibsoni* causing no clinical signs or effect to the cattle and therefore is of little significance to producers.

Although Carr et al (2011) state that 80% of Australian producers use an ivermectin based wormer at weaning. We are also aware that the feed lot industry has well established induction programs, which include ivermectin treatments. Given this we have identified the currently label treatment protocols that would be effective for *Onchocerca gibsoni* in Appendix 2.

Preventative treatment control measures are also described by Merial (2017) to reduce *Onchocerca* sp. are to measure and reduce the midge (Culicoides sp.) population and protect animals from the midge.

9.1.5 Regulator Approach and Commercial Concerns

A newspaper article from 1911 explains that, beef nodules had been evident in Queensland cattle since the 1870's however became a commercial and regulator consideration for the beef industry in the early 1900's. In 1911, during the inspection of Australian beef on entry into England, beef nodules were identified. The English inspection of subsequent beef products was increased and this finding was part of the reason for the Australian Federal Government's decision to appoint veterinary inspectors under the export regulations of the time. Although even in 1911 it was fully understood that the worm did not pose a human health concern, the federal requirement was to remove the nodules in infected beef carcases due to food suitability. The economic implications of a decrease in carcase value was noted as the reason for further research to be conducted to identify the vector (Anon 1911 and Seddon 1967).

Under the current Australian Standard for the Hygienic Production and Transportation of Meat and Meat Product for Human Consumption (AS4696:2007) onchocerciasis is listed within Schedule 3 – Ante mortem and Post mortem Dispositions under 2.2 Parasitic Conditions with a disposition of 'Lesions and affected tissue trimmed from the carcase and condemned' (FRSC, 2007). Given that onchocerca nodules are a food suitability concern this inclusion is not questioned.

Within the Meat Hygiene Assessment (DAWR 2002) used by both government inspectors and processing staff to assess meat hygiene, nodules are included as a critical defect, reasonably likely to seriously affect food safety or wholesomeness, if identified on the outside of forequarters. However as previously stated it has been understood since the early 1900's that *Onchocerca gibsoni* is not a public health risk to humans through the consumption of affected meat. The Meat Hygiene Assessment also lists brisket nodules as 'pathology' stating that,

'*Obvious nodules detected on the slaughter floor should be removed intact. When assessing briskets nodules, the nodule search in the boning room is accepted as a further safeguard against this defect. Where product is not boned at an establishment (for example quarter beef) nodule removal is critically assessed after trimming prior to wrapping or load out.'*

In line with these requirements, the Department of Agriculture and Water Resources' own Disposition Notes and Post Mortem Work Instruction for inspection staff, states for *Onchocerca gibsoni* that visible lesions are to be removed and condemned on the slaughter floor (normally removed by company staff) with inspection occurring during the boning process for nodules not evident at the time of post-mortem inspection (DAWR 2010 and DAWR 2013). Briskets are 'fleeced' of the connective tissue between the muscles removing any nodules from infected cattle, prior to the product being packed for market.

However, in practice despite the change in Export Control (Meat and Meat Product) Orders in 2005 which allowed for less prescriptive requirements and more industry ownership of the 'how to comply', a large proportion of processing establishments still comply with volume 3

of the previous export meat manual, a historic approach of ‘fleecing’ all briskets of carcases over 90kg, sourced from all States and Territories except Tasmaina. (Appendix 3). For non-infected cattle, this reduces the weight and therefore price as well as preventing the additional price premium available at sale for intact briskets.

Seddon (1967) stated that *Onchocerca gibsoni* can be identified in a freshly killed carcase through palpation of the brisket or observation of the cut edge of the tissue as the worms emerge, despite over lying skin never being ulcerated. Based on this research a number of processing establishments hold commercially confidential approved alternative arangements with the Department of Agriculture and Water Resources to use an alternative inspection procedure similar to that provided in the trial protocol of this report (section 3.5).

During a meeting of the Inspection Review Expert Panel held in Adelaide, the Department of Agriculture and Water Resources representative explained that the existing Seddon map is still accepted as part of the validation of an alternative arrangement despite its age.

The industry is aware that competing export countries with *Onchocerca gibsoni* (including the US and Brazil) are exporting intact (non-fleeced) briskets to a range of markets that Australia supplies and as such the current practice is costing the Australian beef supply chain approximately $7.7million.

Given that *Onchocerca gibsoni* does not hold a risk of food safety to the consumer but a business risk of suitability between commercial parties, the original inception of this project was to ensure that beef processors were provided with the tools to address *Onchocerca gibsoni* with a risk based approach considering the whole supply chain. This would then allow processors to

- assess contributing factors of incoming cattle based on:
    - prevalence zoning
    - producer type: feedlot verses grassfed, given that a significant proportion of the feeder cattle are sourced from southern regions rather than north Queensland
    - age of stock
    - stock movements: given that the research shows that nodules take 12 months to form
    - anthelmintic treatments information available
- develop an appropriate hazard identification method for their site’s needs considering available resources – this could be the project validated palpation method suggested here, or through other means such as imaging diagnostic tools currently being developed (and consider by the broader Rural R&D for Profit animal health project)
- develop risk mitigation through trimming practices or market assessment. The risk assessment of the receiving market could include commercial specification that might require some fleecing and therefore further checking of briskets but mean that an inclusion of 12mm of fat will increase the cut price.

Therefore the businesses risk assessment can take into account the entire supply chain management including market for product and uses existing practices that do not result in additional costs for producers but decrease overheads and increase potential revenue for the processing sector.

9.2 Appendix 2: Available Anthelmintic Treatment Options

Active constituent	Product Name	Route of administration	Concentration	Dose: Cattle
Ivermectin	Ivomec Antiparasitic Injection for cattle	Injection	10mg/ml	1ml for each 50kg liveweight. For use by subcutaneous injection only
	Genesis Injection	Injection	10mg/ml	1ml/50kg body weight by subcutaneous injection only
	Cattlemax	Injection	10mg/ml	1ml for each 50kg liveweight. For use by subcutaneous injection only.
	Noromectin Antiparasitic Injection for cattle and pigs	Injection	10mg/ml	1ml per 50kg bodyweight by subcutaneous injection only
	Bomectin	Injection	10mg/ml	1ml per 50kg liveweight by subcutaneous injection
	Pastoral Ag Ivermectin Injection for Cattle	Injection	10mg/ml	1ml per 50kg liveweight by subcutaneous injection
	Virbac Virbamec LA Injection Endectocide for Cattle and Pigs	Injection	10mg/ml	1ml per 50kg bodyweight by subcutaneous injection
	Topshot Antiparasitic Injection for Cattle	Injection	10mg/ml	1ml per 50kg bodyweight by subcutaneous injection
	Bovimectin Antiparasitic Injection for Cattle and Pigs	Injection	10mg/ml	1ml per 50kg bodyweight by subcutaneous injection only
	Ivomec Pour-on for cattle	Pour-on	5mg/ml	1ml per 10kg liveweight

Ivermectin	Coopers Paramax Pour on for beef and dairy cattle	Pour-on	5g/L	The formulation should be applied along the topline in a narrow strip extending from the withers to the tailhead. The dose is 1mL for each 10 kg of liveweight
	Genesis Pour-on	Pour-on	10mg/ml	1ml/20kg bodyweight. Apply along the backline of the animal
	Baymec Pour-on for cattle	Pour-on	5g/L	1ml per 10kg bodyweight. Apply along the backline of the animal in a narrow continuous strip extending from the withers to the tailhead.
	Noromectin Pour-on for cattle	Pour-on	5mg/ml	The dose rate is 1ml for each 10kg of liveweight. Apply along the topline in a narrow strip extending from the withers to the tailhead.
	Pastoral Ag Ivermectin Pour-on	Pour-on	5mg/ml	1ml for each 10kg bodyweight
	Virbac Ivermectin Pour-on for Beef and Dairy Cattle	Pour-on	5g/L	1ml/10kg body weight
	Virbac Virbamax Pour-on for Beef and Dairy Cattle	Pour on	10mg/ml	1ml/20kg body weight
	Virbac Virbamec LV Pour-on endectocide for cattle	Pour-on	10mg/ml	1ml/20kg body weight
	IMAX CD Pour-on for cattle	Pour-on	10mg/ml	1ml/20kg body weight. Apply along the topline of the animal
	Ausmectin Cattle Pour-on	Pour-on	5mg/ml	1ml for each 10kg body weight
	Vets Choice Ivermectin Pour-on for Cattle	Pour-on	10mg/ml	1ml/20kg body weight. Apply along the topline of the animal

Ivermectin	Vetmec Pour-on for Cattle	Pour-on	10mg/ml	1ml/20kg body weight. Apply along the topline of the animal
	Bomectin Pour-on for Cattle	Pour-on	10mg/ml	1ml/20kg body weight. Apply along the topline of the animal
	Topshot Pour-on for Beef and Dairy Cattle	Pour-on	5g/L	1ml for each 10kg of liveweight
	Baymec Pour-on LV	Pour-on	10mg/ml	1ml/20kg body weight. Apply along the topline of the animal
	Stockrite Ivermectin Pour-on for Beef and Dairy Cattle	Pour-on	5g/L	1ml for each 10kg of liveweight
	Top End Mectin Pour-on for Cattle	Pour-on	10mg/ml	1ml/20kg body weight. Apply along the topline of the animal
	Toromax Pour-on for Beef and Dairy Cattle	Pour-on	5g/L	1ml for each 10kg of liveweight
	Starmec Pour-on for Beef and Dairy Cattle	Pour-on	5g/L	1ml for each 10kg of liveweight
	Cattlepro Pour-on for Beef and dairy Cattle	Pour-on	5g/L	1ml for each 10kg of liveweight
Ivermectin + Clorsulon	Ivomec Plus	Injection	Ivermectin: 10mg/ml Clorsulon: 100mg/ml	1ml for each 50kg liveweight. For use by subcutaneous injection only
	Virbac Virbamax Plus Antiparasitic Injection for Beef and Dairy Cattle	Injection	Ivermectin: 10g/L Clorsulon: 100g/L	1ml per 50kg liveweight by subcutaneous injection
	Genesis Ultra Injection Broad Spectrum Antiparasitic for Cattle	Injection	Ivermectin: 10mg/ml Clorsulon: 100mg/ml	1ml/50kg body weight by subcutaneous injection only
	Virbac Virbamec Plus Injection Endectocide & Flukicide for cattle	Injection	Ivermectin: 10g/L Clorsulon: 100g/L	1ml per 50kg liveweight by subcutaneous injection

Ivermectin + Clorsulon Ivermectin + Clorsulon	Vetmec F Broad Spectrum Antiparasitic Cattle Injection	Injection	Ivermectin: 10mg/ml Clorsulon: 100mg/ml	1ml/50kg body weight by subcutaneous injection only
	Noromectin Plus	Injection	Ivermectin: 10mg/ml Clorsulon: 100mg/ml	1ml per 50kg liveweight by subcutaneous injection
	Bomectin F Broad-Spectrum Antiparasitic Injection for Cattle	Injection	Ivermectin: 10mg/ml Clorsulon: 100mg/ml	1ml per 50kg liveweight
	Ivaclor Broad Spectrum Antiparasitic Injection for Cattle	Injection	Ivermectin: 10mg/ml Clorsulon: 100mg/ml	1ml for each 50kg liveweight. For use by subcutaneous injection only.
	Baymec Gold Injection	Injection	Ivermectin: 10mg/ml Clorsulon: 100mg/ml	1ml per 50kg liveweight
Triclabendazole+ Ivermectin	Young's Triclamec Cattle	Oral	Triclabendazole:120g/L Ivermectin: 2g/L	5ml per 50kg bodyweight
	Cooper's Sovereign Pour-on Flukicide and Anthelmintic	Pour-on	Triclabendazole:240g/L Ivermectin: 15g/L	1ml per 10kg of liveweight
Bitroxynil + Ivermectin + Clorsulon	Virbac Nitromec Injection Endectocide & Flukicide for Cattle	Injection	Nitroxynil: 340g/L Ivermectin: 6.7 g/L Clorsulon: 67g/L	1.5ml per 50kg liveweight by subcutaneous injection
Fluazuron + Ivermectin	Acatak Duostar Tick Development Inhibitor and Broad Spectrum Pour-on	Pour-on	Fluazuron: 15g/L Ivermectin: 5g/L	5ml/50kg

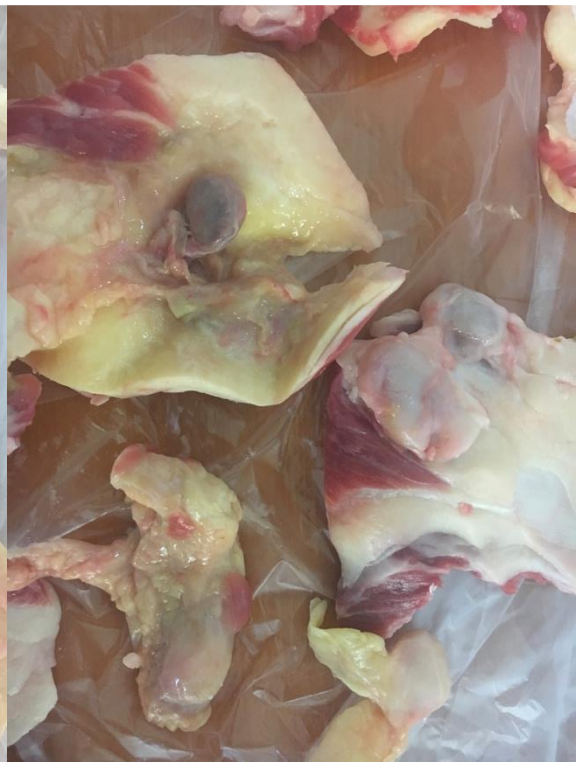
9.3 Appendix 3: Historic Approach to Brisket Preparation and Inspection

(Circa 1996)

Beef brisket preparation applicable to all export boning establishments in all States/Territories except Tasmania, where *beef nodules* are not known to occur:

- applicable to briskets derived from beef carcases of a dressed weight greater than 90 kg and intended for export in boneless form,
- briskets are to be separated from the carcase along a straight line from the point where the first rib joins the first sternal segment to the reflection of the diaphragm onto the 11th rib,
- resources for brisket preparation are available:
    - sufficient tables of sufficient size to enable thorough palpation and incision,
    - sufficient personnel to perform 'beef nodule' inspection, efficiently at establishment production rates,
    - sufficient receptacles (condemned) for the disposal of nodules
- search and detection procedures for beef nodules are performed by establishment operatives as follows:
    - after briskets are boned, slicers are to remove all apparent nodules, discarding them as condemned material in receptacles provided,
    - cuts are to be defatted to an extent that all remaining fat can be thoroughly palpated,
    - the fat on the inside of the point end brisket is to be cut to make 2 or 3 parallel incisions, 1 of which should extend about half the length of the edge of the point end brisket,
    - the fat underlying the sternal end of the 'rib-fingers' is to be lifted and searched,
    - the posterior end of the point end brisket is to be pocketed and the incision continued along the lateral edge of the brisket, the flap turned back and the underlying tissues palpated,
    - leaving one edge attached, the outside muscle of the 'navel end' brisket is to be lifted free and the underlying tissues thoroughly palpated; and
    - the remainder of the navel end brisket is to be sufficiently defatted to allow for an adequate palpation and inspection.

9.4 Appendix 4: Photos of Onchocerca gibsoni nodule



9.5 Appendix 5: Slaughter floor data recording sheet

Plant: Date: Slaughter Floor

Body	Palpation L	Palpation F	Surface (S) Deep (D)	Navel (N) Point- end (P)	Inside (I) Outside (O) Cut (Cut)	Size (10mm, 20mm, bigger)
1						
2						
3						
4						
5						
6						
7						
8						
9						
10						
11						
12						
13						
14						
15						
16						
17						
18						
19						
20						
21						
22						
23						
24						
25						
26						
27						
28						
29						

9.6 Appendix 6: Boning Room data recording sheet

Plant: Date: Boning Room

Carcase sequence No.	Trimming	Palpation	Comment
1			
2			
3			
4			
5			
6			
7			
8			
9			
10			
11			
12			
13			
14			
15			
16			
17			
18			
19			
20			
21			
22			
23			
24			
25			
26			
27			
28			
29			

9.7 Appendix 7: Data details

The excel spreadsheet Onchocerca Data_Plant A - D.xlsx contained a separate sheet for each data collection point (SF = slaughter floor; BR = boning room) at each plant (A D) were data were collected.

The following is a description of the variables in each of the four SF sheets (as provided).

Plant - Anonymous plant identifier (A D)
PIC The Property Identification Code (PIC) from which the animal was consigned; multiple PICs per carcase indicate movement between properties (including saleyards and feedlots).
Latitude - The latitude of the PIC region to which the PIC belongs; the PIC region was used instead of the PIC due to confidentiality reasons.
Longitude - The longitude of the PIC region to which the PIC belongs; the PIC region was used instead of the PIC due to confidentiality reasons.
RFID - The animal's radio frequency identifier, a unique ID associated with the National Livestock Identification Scheme (NLIS)
Mob No - A lot identifier used to link animals consigned together.
Body Number - A plant specific number to identify the carcase
Palpation (leading) - Detection of *O. gibsoni* (Y=yes; N=no) in the leading carcase side
Palpation (following) - Detection of *O. gibsoni* (Y=yes; N=no) in the following carcase side
Comment - A descriptive comment
Stock - Type of animal, synonymous with sex
Sex - Sex of animal
Dentition - Dentition of the animal, a surrogate of age
Breed - Breed of animal, based on hump height; values include British breed, cross breed or tropical breed, or unknown for when the information was not available.

The following is a description of the variables in each of the four BONING ROOM sheets (as provided).

Plant - Anonymous plant identifier (A D)
Body Number - A plant specific number to identify the carcase
Trimming - Identifies whether the carcase had *O. gibsoni* nodules trimmed (Y=yes; N=no), i.e. *O. gibsoni* was detected in the boning room.
Palpation - Identifies whether the carcase had *O. gibsoni* nodules detected via palpation (after the trimming step; Y=yes or N=no), i.e. *O. gibsoni* was detected in the boning room.
Comment A descriptive comment

9.8 Appendix 8: V.RBP.0023 Data Analysis

Attached to final report as a pdf.