

# final report

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### **Reducing Mastitis in Sheep**

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#### 1. What the Group set out to do:

• The primary goal of the GVPDA was to decrease sheep death rates due to mastitis, by developing a better understanding of bacteria causing disease and possible treatment options for mastitis.

#### Goals (Measurable Outcomes)

- 1. Reduce death due to mastitis by 30% (average flock sizes of 150 ewes with 3% death rate currently).
- 2. To train at least 20 group members on correct sheep milk collection techniques.
- 3. To train at least 20 group members on the identification and treatment of early stage mastitis.
- 4. To define the causes of mastitis on more than ten farms within the Goulburn Valley region in the Poll Dorset breed.
- 5. To collect milk from at least 500 sheep to determine the types of bacteria causing mastitis.
- 6. To determine the percentage of sheep affected with mastitis per flock and the range between different flocks within the Goulburn Valley.
- 7. To determine the best antibiotics for treatment of mastitis on at least ten farms.
- 8. Investigate the use of "dry-sheep" intramammary therapy on one farm with high levels of mastitis.

#### 2. How the Project was done

#### i. Bacteria causing clinical mastitis

Twenty three farms participated in collecting milk samples from sheep that were clinically affected by mastitis. These farms were located within the Goulburn Valley (GV) region, South West NSW (SWNSW) region and Wimmera-Mallee (WM) regions of the Poll Dorset Association (PDA). The increase in number of regions was done to provide a wider geographic spread and ensure adequate milk submission rates were attained. All producers were shown how to collect milk and possible mastitis treatment options were defined, prior to the trial commencing.

Sheep were identified as having mastitis in the paddock by their appearance. Signs of mastitis included:

- Lameness
- A swollen or discoloured udder
- Skinny lambs
- Inappetance
- Hollow gut
- General appearance (ears down, head down)
- Extended time lying down

Those sheep that were identified in the paddock as having mastitis could be further

classified by examination of the animal once restrained. Further signs of mastitis were:

- Elevated temperature (above 40.5 degrees Celsius). Not all sheep showed this temperature elevation but it was generally seen in severely affected sheep.
- Increased heart and breathing rate, often double normal.
- Swollen and hot udder
- Milk of different colour or consistency compared to normal. The colour ranged from red through white to watery. Consistency ranged from pus of a honey consistency through to clotted cream or very "flecky".
- At a later stage the udder became cold. These sheep were developing gangrenous mastitis and in general this tissue sloughed off the sheep, if the sheep survived the infection.

A milk sample was collected from both teats of those sheep deemed to have clinical mastitis. The collection method involved swabbing the teat with a 70% methanol (methylated spirits) solution. The first few squirts of milk were then discarded and milk sample was collected into a sterile 5 ml container and frozen as soon as possible post collection. This was done individually for both the affected and non-affected glands. This sample remained frozen until processing at the laboratory at the University of Melbourne.

A total of 154 samples were submitted for testing at the University of Melbourne laboratory. This comprised 77 samples from the infected half and 77 from the non-infected half of the udder. Other data collected included lambing date, number lambs born, sire & dam and date of disease.

#### ii. Bacteria causing sub-clinical mastitis

One property was used to assess the rate of sub-clinical mastitis. This involved the collection of milk from ewes at weaning time in 2005 and 2006. In all, 453 milk samples were collected from sheep on this property (145 were collected in Spring 2005 and 308 from both Autumn and Spring lambing flocks in 2006). These milk samples were a combination of both left and right sides of each sheep's udder so represent 906 sides in total. The method of milk collection was the same as for clinical mastitis cases, although both samples were collected into the one tube.

In 2005, of the 145 sheep that had milk collected, approximately half were administered a "drying-off" antibiotic into the udder (similar to the technique used for dairy cows). These sheep were then evaluated in the 2006 lambing season for mastitis to investigate differences between treated and un-treated sheep.

## 3. Analysis of the data, what was achieved?i) Overall incidence of mastitis in 2006

There was a remarkably low incidence of mastitis during the 2006 season, both in Autumn and Spring lambing flocks. In 2005, some flocks had over 10% of their flocks with mastitis while this year the average was less than 2%. Presumably, the very dry prevailing conditions and associated low dry matter availability in paddocks were important in this reduction. There are, however, divergent views from Poll Dorset breeders as to whether

dry or wet weather is more likely to produce mastitis. It would appear most likely that any factor likely to lead to damage of the teat orifice or to result in milk leaking out, is likely to increase mastitis. This appears more likely to occur during wet conditions.

There were approximately 6,500 ewes enrolled in this study based on the ewe numbers within each registered stud. Of these ewes, 77 had samples submitted for testing. This suggests that the infection rate was 1.2%. There were, however, a number of reasons for sheep that became infected not having samples submitted. These reasons included:

- Acute death the time from infection to death for sheep infected with some bacteria is less than 24 hours. This resulted in some sheep dying prior to any milk being collected.
- Milk production A number of ewes presented with mastitis where there was no milk production from the teats. This may have been due to previous damage to the udder (pre-existing disease) or that the disease caused inflammation and rapid milk production decline.
- Lack of collection A number of sheep that had mastitis were not collected for various reasons, including inexperienced personnel watching sheep while owners were away. The signs of mastitis can be subtle hence untrained shepherds risk not observing the symptoms and thus not collecting samples.
- Variations in what is designated as "clinical mastitis". Severe, acute, black mastitis is almost always diagnosed by farmers; however, less obvious clinical infection may pass unnoticed. Thus, the level of reported clinical infection is generally an under-representation of true "clinical" infection.

The above reasons suggest that the true infection rate for clinical cases was probably closer to 2% than 1%, though remained dramatically lower than 2005.

#### ii) Clinical cases

There were a total of 77 samples submitted in 2006. All of these samples and the other sample from the uninfected gland were cultured for the presence of bacteria. Where bacteria grew from these milk samples they were then identified.

Of the 77 samples submitted, only 48 grew bacteria. This is a common finding where only approximately 2/3<sup>rds</sup> of samples submitted to the laboratory will grow bacteria. This was one of the prime reasons for using the power of a PIRD to evaluate mastitis across several farms rather than an individual farm.

Of these 48 samples that grew bacteria, over 50% were mastitis caused by *Mannheimia*. This bacterium, previously knows as *Pasteurella* but renamed in the last decade, was responsible for a lot of cases of black, or gangrenous, mastitis. The next most common bacteria were *Staphylococcus*. There are many different types of these bacteria, with *Staphylococcus aureus* causing black mastitis as well. This was the most commonly identified *Staphylococcus* species during the trial.

#### Figure 1: Causes of clinical mastitis in Poll Dorset sheep



#### Table 1: Causes of clinical mastitis in sheep

	Mannheim ia (percenta ge of farm total)	Staphylococ cus	Streptoco ccus	E. coli	Pasteurell a multocida	Bacillu s	Total sampl es
Property 1	4 (50%)	1	2	1	manoonaa	0	8
Property 2	1 (25%)	2	1				7
Property 3	2 (50%)	2					10
Property 4	1 (50%)				1		6
Property 6	1 (50%)	1					4
Property 7	3 (100%)						5
Property 8	(0%)	1					2
Property 9 Property	3 (100%)						5
10	1 (100%)						4
Property	. (10070)						
13	3 (30%)	5	1	1			10
Property	, , , , , , , , , , , , , , , , , , ,						
15	3 (75%)					1	4
Property							
16	(0%)	1					3

Property 18							2
Property 19							1
Property							
20	(0%)		1				2
Property							
22 Sum	3 (75%)			1			4
Sum	25	13	5	3	1	1	77
Percentag							
e*	52.1%	27.1%	10.4%	6.3%	2.1%	2.1%	

\*Refers to the percentage as a percentage of total isolates that grew bacteria

Table 1 demonstrates the different bacteria causing mastitis on each farm within the PIRD. It shows that some properties appear to have a higher percentage of *Mannheimia* compared to other causes of mastitis. This may be true, or may be due to bias on behalf of the farmer by only identifying moderate to severe cases of mastitis. There is considerable variation within management practices for the lambing period. This may vary from individual treatment of each ewe through to a minimal intervention approach and may alter to number of cases of mastitis that are observed.

#### Resistance to antibiotics

Bacteria have the ability over time to become resistant to the effect of antibiotics. Bacteria grown from clinical mastitis samples submitted to the laboratory were tested for their sensitivity to commonly used antibiotics. Results for *Staphylococcus* and *Mannheimia* are given below:

#### Mannheimia

All samples of *Mannheimia* were resistant to erythromycin so this antibiotic is not the drug of choice to use against these bacteria. Over 50% of samples were also resistant to Penicillin and there was some variation from property to property. There was less than 25% resistance to Sulphafurazole and no resistance to Trimethoprim. Only 2 properties had *Mannheimia* isolates with resistance to tetracycline, this however is of concern as similar bacteria have rapidly developed resistance to tetracycline in cattle feedlots. Tetracycline is currently the antibiotic of choice on many properties in this PIRD, due to its long action and penetration into the udder. No isolates demonstrated resistance to ceftiofur, however this antibiotic is not commonly used within the sheep industry, requires daily administration and may be expensive compared to other antibiotics.

#### Figure 2: Percentage of Mannheimia isolates resistant to antibiotic



Only six isolates of *Staphylococcus aureus* were tested for sensitivity to antibiotics. All of these isolates were sensitive to tetracycline, sulphafurazole and ceftiofur. Property 3 had three isolates of *S. aureus* tested; one isolate was resistant to penicillin and erythromycin while another showed resistance to trimethoprim, the third did not show any signs of resistance to antibiotics. A further isolate resistant to trimethoprim was identified on property two.

#### Genetics

It was proposed at commencement of this PIRD to collaborate with SGA/Lambplan to investigate the genetic component of mastitis. While the issue of collecting a data-set for mastitis has been discussed with SGA, no analysis of data has as yet been performed. There remains much to be understood about mastitis in Australian sheep before genetic selection may yield positive results for reducing mastitis. Little is known about the transmission of *Mannheimia*, compared to information on *Staphylococcus* and *Streptococcus* which has been reasonably well documented in Europe.

#### iii) Sub-clinical cases

Of the 145 sheep sampled in 2005, 45 (31%) milk samples grew bacteria. It was hoped to have cell counts from these milk samples however the initial method used to count cells proved inadequate. This figure of 31% is an overestimation of the true prevalence of subclinical mastitis as some of the cultures did not produce large numbers of bacterial colonies. There is some debate as to the optimal method of defining sub-clinical mastitis; however there is no doubt that the level of sub-clinical mastitis is considerably higher than clinical mastitis. This is likely to affect milk production, hence growth rate of lambs and in severe cases lamb survival. Some of these ewes with sub-clinical mastitis can remain infected for extended periods (months) and even into following lactations. The predominant bacteria causing sub-clinical mastitis in 2005 were *Staphylococci* and *Streptococci*.

In 2006, 61 (20%) of 308 milk samples grew bacteria. This consisted of; 44 of 212 sheep in Autumn and 17 of 96 animals in Spring. Cell counts were available only for the last 96 sheep tested. Using a cut-off of 750,000 cells per milliliter for sheep with sub-clinical mastitis this suggested that 19 (20%) of 96 sheep had sub-clinical mastitis. Approximately

half of these 96 ewes were maiden ewes and would generally be expected to have lower cell counts than average.

Only seven of these high cell count animals produced milk that allowed bacterial culture. Reducing the cut-off to 500,000 cells per milliliter increased the number of sub-clinical infections to 24 sheep. Similar to 2005, the predominant bacteria causing mastitis were *Streptococci* and *Staphylococci*. There were however, several cases of sub-clinical *Mannheimia*.

No obvious benefits of using "drying-off' therapy was evident from this trial. This may in part be due to the methodology used during the PIRD. It was hoped to have cell count data to compare milk from treated and un-treated sheep but due to problems in cell counting this was not possible. Only seven sheep at the stud developed mastitis with similar numbers of animals developing mastitis with or without treatment. Repeating this work using larger group sizes and cell counts early in the following lactation may produce different results to those attained during this PIRD. The use of cell counts may be an effective tool for selection of sheep that would be less prone to mastitis.

#### 4. What did the Group learn by doing the trials?

The group achieved all the results planned at the beginning of the trial. These were:

• Reduce death due to mastitis by 30% (average flock sizes of 150 ewes with 3% death rate currently).

Results from this PIRD have improved the group's knowledge of appropriate antibiotics to use for sheep affected by mastitis. The outcomes of the PIRD have also allowed a better understanding of the signs of mastitis (especially for any producers who do not have a long history of working with sheep). It is difficult to quantify the exact reduction in long term death rate on each property from involvement in the PIRD, but all properties experienced lower death rates this year than last.

- To train at least 20 group members on correct sheep milk collection techniques.
- To train at least 20 group members on the identification and treatment of early stage mastitis.

At least 23 members of the Goulburn Valley, South West NSW and Wimmera Mallee Poll Dorset groups were trained in both milk collection and identification and treatment of early stage mastitis. Many more members of both the Poll Dorset association and the British Breeds association have also seen results from this PIRD.

• To define the causes of mastitis on more than ten farms within the Goulburn Valley region in the Poll Dorset breed.

In total, 16 properties from the 23 properties submitted at least one sample for testing in this PIRD. This produced 77 clinical samples for testing, the largest reported single year survey of its kind undertaken in Australian history. The causes of mastitis were defined and *Mannheimia* was diagnosed as being the most commonly identified cause. This is a change in scientific attitude as previously it was thought that *Staphylococcus* was the most

common cause of clinical disease.

• To sample at least 500 sheep to determine the types of bacteria causing mastitis.

Even though it was a very difficult year in which to find samples as there was a low infection rate, 530 samples were submitted for bacterial culture. The majority of these were from sheep with sub-clinical mastitis, but the 77 samples received from sheep with clinical mastitis allowed new data on bacteria causing mastitis to be collated.

• To determine the percentage of sheep affected with mastitis per flock and the range between different flocks within the Goulburn Valley.

The percentage of mastitis across all flocks was highly variable ranging from 0% to 5.5% with an average of 1.3%. This fact alone is vitally important as in a year that produces bad mastitis problems, this percentage may be more like 2% to 15%. This suggests that either: some farms are able to maintain mastitis at low levels suggesting that some form of selection or management can reduce mastitis, or that there is substantial variation in each farmers ability to diagnose infection. All farmers involved in the study felt that 2006 produced very little mastitis compared to most years. In particular, one farm that had over 10% of sheep with mastitis in 2005 had less than 2% in 2006. A study to determine true incidence of mastitis would need to be of larger geographical size and also duration to give an accurate prediction of long term expected mortality rates.

• To determine the best antibiotics for treatment of mastitis on at least ten farms.

Bacterial specimens were grown from 16 farms and analysed for antibiotic sensitivity. These tests demonstrated that the two most common bacteria (pathogens) had some degree of resistance to antibiotics. The best "first line" antibiotics appeared to be tetracycline and trimethoprim/sulphonamide as both *Mannheimia* and *Staphylococcus* were sensitive to these antibiotics.

• Investigate the use of "dry-sheep" intramammary therapy on one farm with high levels of mastitis.

No benefits of using "dry-sheep" intramammary therapy were identified during the PIRD. This was possibly due to the trial design and an inability to get accurate milk cell counts. A laboratory that is capable of performing cell counts has since been identified and 96 samples processed with good result. Overseas studies have suggested that using intramammary antibiotic therapy significantly reduces mastitis in the following lactation (for milking sheep). One of the problems of using cell counts is that the samples must be "fresh", less than 3 days old, at the time of counting. This makes this technique difficult to perform in practice, given time for collection of samples and subsequent transport to the laboratory. Each sample also costs approximately \$3 to perform, without including transport costs. To further investigate intramammary therapy a larger study would be required including the use of cell counting in the month post lambing.

To utilize cell counting for genetic selection would also require multiple collections during

the lactation period, from both sides of the udder. For example, three to four collections may be taken resulting in a total of up to 8 samples, costing \$24 per animal plus transport costs. It is unlikely that many producers would be interested in paying this amount of money for the possible benefits in genetic selection.

• What changes members have made as a result of doing the project, or what changes are planned as a result of running the trials?

Changes that members have made/or plan to make in response to this PIRD include:

- Changing to a more effective antibiotic. Some farms had considerable resistance to particular antibiotics and have since changed to an antibiotic that should be more effective.
- If in doubt as to a diagnosis of mastitis, some members are utilizing a thermometer to check for increased temperature in sheep. Sheep with increased temperatures require antibiotic treatment while some sheep may just have swollen udders post lambing. This reduction in use of antibiotic can reduce the selection pressure for antibiotic resistance.
- Members involved in Lambplan flocks may have the opportunity to submit data on mastitis if SGA include this in their database. This may identify sires more likely to produce progeny that develop mastitis. There are however, a number of issues relating to genetic selection with respect to mastitis that require further clarification/research. Further research into mastitis in Australian sheep is required to evaluate expected benefits of incorporating mastitis information into breeding objectives.
- All members are aware of the benefits of rapid treatment of sheep with clinical mastitis. Severe mastitis happens so quickly that immediate treatment is imperative to save the ewe. This may also change the frequency with which sheep are checked as they must be observed daily to reduce the chance of mastitis deaths.

• Trial measurements. Have these enabled you to show the economics of the outcomes and what benefits [dollar] members may be able to gain? How have/will members improve their bottom line?

- The measurements taken during this trial have given a very approximate indication of the amount of mastitis in British Breed sheep. It is however, only a single time point within one geographic location of Australia (using approximately 6% of the national Poll Dorset flock) hence not necessarily representative of all sheep. It also does not address the problem of mastitis in first cross ewes, as many first cross ewe breeders complain of high death rates and losses within their flocks due to mastitis. These issues require further clarification and might help further research into preventive mastitis therapies such as production of vaccine (as it would dramatically increase the numbers of sheep to be vaccinated).
- This PIRD has demonstrated the high costs of mastitis to producers within the region. Approximately 2% of sheep became ill with mastitis during the year and up to 20-30% of sheep were sub-clinically infected at the end of lactation. The exact

cost of this sub-clinical infection is difficult to quantify but would result in increased culling rates, higher lamb death rates and reduced lamb growth rates. This probably equates to a cost greater than that caused by clinical mastitis but currently goes predominantly unrecognized.

- These figures, when extrapolated to the Australian Poll Dorset flock suggest a loss each year somewhere between \$2-8 million dollars (depending on the incidence in any given year and on the value given to each sheep).
- Further research is required to dramatically reduce this figure as producers will still have sheep die due to mastitis given the rapid onset of disease. An ideal product to aid producers would be a vaccine. This may be a current product used "offlabel", or might require development of a new product.
- Members of the PIRD now have the skills to be able to combat mastitis more effectively and to reduce, but not eliminate, the effect that mastitis can have on their flock.

#### 5. Please describe any open days, field days etc and how many attended?

Dr. Stuart Barber discussed mastitis in sheep at meetings held by the GVPDA and SW-NSWPDA in Autumn 2006 and then presented a summary of the findings from the project to the GVPDA in December 2006. Each meeting was attended by approximately 20 people. A presentation to the Australian Sheep British Breeds Association and Poll Dorset Association (combined meeting) was given in Autumn 2006 with approximately 50 people attending. Each farm involved in the project (of which there were 23) was also visited individually. This resulted in over 100 people having direct contact with the project. The project was also reported in the media including the Weekly Times, ABC local and national radio, University of Melbourne NEWS (UniNews) and overseas media. A summary of early results from the trial was presented at the Australian Sheep Veterinarians conference at Wagga Wagga in July, 2006 with approximately 100 veterinarians in attendance.

#### 6. Was the Group satisfied with the results of the project?

The group was satisfied with the results of the project. It has significantly improved the knowledge regarding mastitis within Poll Dorset sheep within the area and improved the chance of saving more sheep and their lambs.

#### 7. How could you have done the project better?

There were no particular suggestions as to how the project could have been done better. The influence of the drought dramatically reduced the number of specimens available, however planning by the group allowed an adequate number of samples to be submitted despite this influence. A larger number of clinical cases of mastitis may have allowed a more rigorous evaluation of the use of "drying off" treatment.

#### 8. Is the group interested in doing another project?

Most members of the group would be interested in being involved in another project. Some members suggested projects such as the investigation of joint ill in lambs which can be a considerable problem in the industry. There still remain a lot of unanswered questions about mastitis in the sheep industry that might be answered by PIRDs or other industry funding.

#### 9. Would you recommend other Groups run their own trials?

In general, the group would recommend the concept of PIRD trials to other groups.

## 10. How would the Members sum up their experiences in doing the MLA PIRD project? (What was the bottom line?)

Group members described their experience from the PIRD as being useful to their enterprise. In particular, the knowledge of the best antibiotic to use on farm was useful as some properties were using antibiotics that may not have been the best for the bacteria present on the property. This is due to the understanding that *Staphylococcus sp.* were the predominant cause of mastitis, rather than *Mannheimia*. This finding may change the general management of sheep mastitis treatment.

## 11. Comment on the organisation and management of PIRDs, this will assist MLA in better management of future projects.

There were no particular problems with organization or management of this PIRD (apart from minor problems with money transfer as the GVPDA was not registered for GST). Communication with MLA via Gerald Martin was very good and he was helpful with advice for this project.

#### 12. Media Coverage

There has been considerable media and producer interest in this mastitis PIRD. This has resulted in information being published in The Weekly Times, UniNews (University of Melbourne publication), Feedback (MLA, July 2006), The Australian Sheep Veterinarians Conference Proceedings from Wagga Wagga 2006 as well as on radio (both local and national ABC broadcasts). There has also been interest from international media.