On behalf of the Countdown team and the University of Melbourne, welcome to the Countdown Symposium 2011 – Tackling Strep uberis; Mastering plant hygiene.

Many at this event will have attended the first ‘International All Stars’ Mastitis Control Symposium in March 2010. As stated at the time, the 2010 Symposium was the largest stand-alone mastitis meeting of its type held in Australia since the early ’90s. Through the enormous adviser support of that day, and subsequent feedback to the Countdown team, convening a follow-up event was seen as a natural next step. Bringing together advisers, wearing many different professional hats from all dairying regions of Australia, to participate in the Symposium is an integral component of Countdown’s aim of re-energising and connecting the regional adviser networks.

Given the success of 2010 we have followed a similar path in planning this year’s event. Three dairying countries are represented by the speakers this year and, in many cases, a lifetime of experience in mastitis control work will be drawn upon for the presentations. Our goal is to have all advisers leave the room today equipped with practical, new, milk quality control ideas they can implement tomorrow with their dairy farmer clients. This day is very focussed on putting relevant, cutting-edge science into practice.

The dual 2011 themes, ‘Tackling Strep uberis; mastering plant hygiene’, have emerged from the extensive dairy industry consultation process embarked upon by Countdown in mid-2010. Farmers, advisers and processors indicated that priorities for the immediate future for the Australian industry were managing clinical mastitis and in particular mastitis caused by environmental bacteria as well as control of bacteria in milking plants from ineffective cleaning practices. The speakers invited this year, and the information they present, are a direct response to the priorities articulated by our industry.

We are also pleased to formally launch at this year’s event a new Countdown Technote FAQ: What are the keys to controlling Strep uberis mastitis in dairy herds? This Technote FAQ is published in the proceedings and will also be available on-line at the Countdown web resource for advisers at www.countdown.org.au.

An event of this scale cannot happen without energetic individuals providing support and resourcing. We gratefully acknowledge our principal sponsor, Boehringer Ingelheim. Jonathan Leslie and his associates have fully supported the organisation of the symposium since its initial planning continuing their contribution from 2010. Without their generosity we would not have this impressive line-up of guest speakers. We also acknowledge the ongoing partnership of Dairy Australia through their direct support of the day and their investment in Countdown. The team at Harris Park Group (who manage Countdown for Dairy Australia) have provided the communication and logistics backbone for today.

Mastitis control remains an important and ever-present part of day-to-day dairy farm management. Milk quality cannot be altered past the farmgate and protection of our international trading markets depends, in part, on our continued ability to demonstrate sound mastitis and cell count control at a farm, regional and national level. Milking machine technicians, processor field staff, veterinarians and other dairy advisers are all critical to this success. The adviser network is the primary conduit from best practice science to best practice on farm.

We hope you enjoy the symposium and the interaction with your colleagues and our speakers. We trust that these proceedings, and the practices and conversations you take back to your farmer clients, will continue to make a positive contribution to milk quality control in Australia.

John Penry
Graeme Mein
Peter Mansell
Acknowledgements

The Countdown project team committee would like to thank the following organisations and individuals for their assistance in the preparation of the Countdown Symposium 2011. In particular we wish to express our gratitude to Jonathan Leslie and the team at Boehringer Ingelheim for being the principal sponsor. We also acknowledge the sponsorship support provided by Dairy Australia and University of Melbourne.

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*The speakers have supplied the papers printed in this document to support their presentations.*
Speakers at the symposium

Scott McDougall
Scott has been in the dairy industry for over 20 years and is currently the managing director of Cognosco. He has worked as a clinical veterinarian, taught at Sydney University and worked as a scientist for Dairy Research Corporation (now DairyNZ). He is actively involved in adapting the Australian InCalf Fertility programme to the New Zealand context and in introducing a new mastitis management and control system. Scott is on the editorial board of the Journal of Dairy Science, and undertakes training courses for vets and herd owners in New Zealand and internationally.

Jakob Malmo
Jakob was a partner who helped develop an 11 veterinarian practice in the Macalister Irrigation Area of Victoria. He continues to work full-time as a cattle specialist in the practice. He provides lectures in cattle medicine and production to veterinary students at the University of Melbourne during the last 2 years of their course and has been involved in providing practical instruction to undergraduates at the Rural Veterinary Unit at Maffra for over 30 years.

He is a Fellow of the Australian College of Veterinary Scientists in the area of Dairy Cattle Management and Disease and is a Veterinary Board Registered Specialist in Veterinary Medicine - Cattle medicine.

His areas of special interest include individual animal cattle medicine, dairy cattle reproduction, mastitis control programs, lameness and dairy cattle production. He is a co-author of the recently released textbook Diseases of Cattle in Australasia.

Josh Wheeler
Josh has wide ranging experience in on farm milk quality and has played an integral role in the development of systems, procedures and resources at a farm and technical level in New Zealand and internationally. In 2010 Josh played the lead role in the development of a new innovative On Farm Quality Management Program for New Zealand’s leading dairy company and this is now implemented on 10,000 dairy farms. Josh is regarded as one of New Zealand’s top dairy consultants and trainers in the fields of on farm milk quality, on farm quality management, and on farm quality auditing. He is now using his expertise internationally and is consulting in Australia, Brazil, and Chile.

John Penry
John graduated from the University of Melbourne in 1990. In 1995 he gained a Masters Degree in Dairy Medicine and Production (Melbourne), followed by a Membership of the Australian College of Veterinary Scientists in Ruminant Nutrition in 1999. John has worked at the Camperdown Veterinary Centre in South-West Victoria since 1991 and has extensive project development and implementation experience across four of the Dairy Australia national programs: Countdown Downunder, InCalf, Grains2Milk and The People in Dairy. Through the Harris Park Group, he has been involved with dairy project design for Animal Health Ireland and Dairy NZ.

He is an associate of the Rural Innovation and Research Group at The University of Melbourne and the current Countdown project leader.

John House
John House is Associate Professor at the Livestock Veterinary Teaching and Research Unit at the University of Sydney, and Director of Bovine Clinical Services. His research focuses on disease control, and he has been project leader for the development of a new molecular test for Mycoplasma and Strept agalactiae mastitis, in addition to working directly with herds dealing with Mycoplasma mastitis infections. His current projects also include epidemiological studies of environmental mastitis pathogens on NSW dairy farms.

Doug Reinemann
Doug is Professor of Biological Systems Engineering at the University of Wisconsin-Madison. Doug conducts an active research program for international post-graduate students and visiting scientists at the UW Milking Research and Instruction Laboratory. He currently chairs the IDF Milking Machine Action Team.
Control of environmental mastitis – the Kiwi perspective

Scott McDougall
Cognosco, Animal Health Centre, New Zealand

Introduction
Environmental mastitis in the New Zealand context means predominantly Streptococcus uberis (McDougall et al. 2007b). About half the clinical mastitis cases in the average NZ dairy farm are due to Strep uberis, while the incidence of Escherichia coli and other Streptococci are generally <5%.

Control plans
The current New Zealand mastitis management plan (Seasonal Approach to Managing Mastitis plan or SAMM plan; Woolford et al. 1995) evolved from the ‘5-point plan’ and simply states the same principles but in a seasonally focused way to match the predominantly spring calving pattern still used in New Zealand. While never formally evaluated, the program was associated with a decline in Bulk Tank Somatic Cell Counts (BTSCC) after its introduction. Clinically, there are herds that apply the SAMM plan principles but still experience a high incidence of clinical mastitis (and BTSCC) particularly early in lactation and in heifers. This is probably not surprising given the emphasis in the original 5-point plan on control of ‘contagious mastitis’, the lack of specific heifer mastitis control techniques and also the emphasis on use of partial rather than whole herd dry cow therapy.

Current New Zealand management systems are generally seasonal, pasture-based systems with relatively low nutritional and infrastructure inputs with limited use of supplementary feeds and no housing of cattle. The result is lower yields and potentially increased exposure to some environmental mastitis pathogens (e.g. the Streptococci) as cattle move from pasture to the milking parlour. Pre-milking udder preparation is generally not done, with reduced sensitivity of detection of clinical mastitis, no stimulation of milk let down before application of the cluster and increased risk of new environmental infections.

Preventing new infections
The focus for preventing new infection has been on milking clean dry teats, including use of teat antisepsis and environmental management; effective maintenance of the milking machine, and use of dry cow therapy (or teat sealants) to prevent new infections and cure existing infections over the dry period.

Poor udder hygiene post-calving has been associated with an increased risk of subclinical mastitis (Compton et al. 2007) and density of animals on pasture, a proxy for increased risk of poor udder hygiene, associated with an increased risk of clinical mastitis in NZ (Parker et al. 2007). Anecdotally, use of small areas to feed supplements, or farm tracks on which mud or faeces pool, may increase the incidence of clinical mastitis. High numbers of Strep uberis are found on areas of the farm where there is a high density of cow traffic, for example the laneways leading to and from the dairy parlour (Lopez-Benavides et al. 2007). While not formally examined, it appears logical that improved cow hygiene via improved management of mud and faeces on the farm may reduce the incidence of mastitis in pasture-based systems.

Application of teat spray three times weekly pre-calving to cows grazed on pasture reduced the abundance of Strep uberis on the teat-end just before calving, and significantly reduced the prevalence of intra mammary infection (IMI) associated with Strep uberis after calving, although the overall incidence of clinical mastitis was not altered (Lopez-Benavides et al. 2009).

Dry cow therapy or use of teat sealants reduced the incidence of new infections over the dry period, especially Strep uberis (Williamson et al. 1995). Increasing the proportion of cows within a herd treated with dry cow therapy reduced the incidence of clinical mastitis over the dry period and reduced BTSCC in the subsequent lactation (McDougall 2003). An internal teat sealant was found to be as effective as dry cow therapy alone in preventing new infections in one NZ study (Woolford et al. 1998) as shown elsewhere. However, a more recent study found dry cow therapy combined with an internal teat sealant reduced the risk of clinical mastitis in the next lactation by 0.63 relative to antibiotics alone (Laven and Lawrence 2008). Additionally, infusion of a teat sealant into primiparous cows around 39 days before calving resulted in a 66% reduction in the incidence of new IMI, a 75% reduction in prevalence of IMI post calving, and a 74% reduction in the incidence of bacteria-positive clinical mastitis compared with no infusion (Parker et al. 2008).
Reducing the interval between calving and first milking of primiparous cows by 10 hours reduced the risk of clinical mastitis post-calving by 45% and there was a linear increase in the incidence of clinical mastitis with increasing interval from calving to first milking in a NZ study (Figure 1; Compton and McDougall 2008).

Loss of more than 0.5 unit of body condition score (BCS) (10-point scale) is associated with an increased risk of udder oedema which is in turn associated with an increased risk of clinical mastitis in primiparous cows (Compton et al. 2007). Negative energy balance pericalving is associated with higher concentrations of non-esterified fatty acids and depressed neutrophil function (Hammon et al. 2006). Logically, management to minimise negative energy balance after calving should reduce pericalving disease, including mastitis. Feeding approximately 25% of the pre-calving diet as pasture hay, with the intention of modulating BCS and preventing excessive lactogenesis pre-calving, failed to reduce the prevalence and incidence of subclinical and clinical mastitis in primiparous dairy cows (Compton and McDougall 2008). In another study, feeding the ionophore monensin before calving to primiparous cows increased BCS at calving, but did not affect the prevalence of subclinical infection or the incidence of clinical mastitis (McDougall et al. 2008). However, use of the ionophore lasalocid was associated with less clinical mastitis (McDougall et al. 2004).

Enhancement of immunological responses by vaccination is an attractive proposition for mastitis control (Leigh 1999). In one NZ study using a killed trivalent Strep uberis vaccine, vaccination tended to reduce the prevalence of Strep uberis compared to unvaccinated cows (0.12 vs. 0.16, p=0.06), but there was no difference in the prevalence of Strep uberis at gland level (0.043 vs. 0.055) or the incidence of clinical mastitis associated with Strep uberis at either cow or gland level (McDougall unpubl. data).

### Reducing the duration of existing infections

Strategies commonly used to reduce the duration of existing infections include antimicrobial treatment of subclinical and clinical mastitis either during lactation, or at the end of lactation, and removal (culling) of infected cows. Parenteral treatment or intramammary infusion of antibiotics during lactation for treatment of clinical mastitis cases in NZ result in bacteriological cure rates of approximately 80% (McDougall 1998; McDougall et al. 2007a,b). Cure rates for Strep aureus remain lower than for other pathogens (McDougall et al. 2007a,b). Penicillin resistance does not appear to be a major issue (i.e. <3%; Petrovksi et al. 2011), at least amongst the streptococci, which allows continued use of penicillin as the first line therapy, particularly in early lactation.

Therapy of subclinical mastitis remains little studied in NZ. In one preliminary study, the cure rate of Strep aureus increased linearly with increasing duration (0, 3 or 6 tubes) of intramammary treatment with 250 mg cefurozime (Shelgren et al. 2007). Similarly the cure proportion of subclinical IMI caused by various pathogens increased with increasing number of daily intramammary treatments of 5g penethamate hydriodide (0.16±0.04, 0.32±0.06 and 0.56±0.02 following 0, 3 or 6 treatments, respectively; p<0.001; Nicole Steele, pers. comm.). However, the economics of treatment remain unclear.

Bacteriological cures rates of between 65% and 100% for major Gram positive pathogens following dry cow therapy are reported from Australasia (Browning et al. 1994; Williamson et al. 1995), similar to most international reports. Dry cow therapy also reduces the new infection rate over the non lactation period by about two-thirds, with the majority of this reduction associated with Strep uberis (McDougall 2010).

### Research priorities

*Streptococcus uberis* is the most common cause of clinical mastitis in pasture-based systems and its control remains a challenge (Leigh 1999). Treatment of clinical and subclinical Strep uberis infection is reasonably successful, with more than a 75% cure rate with both lactating and dry cow therapy. However, as the current control programmes have limited effects on prevalence and incidence of infection with Strep uberis, novel management strategies are required. Molecular techniques are providing new information on the epidemiology of Strep uberis (Zadoks et al. 2005; Lopez-Benavides et al. 2007). Further studies are required to use these findings to develop and validate practical control measures. Vaccination remains an attractive proposition, but there are still technical difficulties, suggesting that a vaccine is still some time away.
Conclusions

Despite considerable research into mastitis epidemiology and treatment under pasture-based systems, further work is required. Current gaps include fully understanding the epidemiology of environmental pathogens, particularly *Strep uberis*, so that better control methods can be implemented.

Despite these gaps, implementation of the basic control measures as outlined in the 5-point plan and its derivative varieties and the addition of whole herd dry cow therapy, use of teat sealant in place of, or in combination with, dry cow therapy in cows and precalving in heifers, and effective monitoring of cows as they move from the colostrum to main mobs enables many herds to manage environmental pathogens effectively.

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Troubleshooting high bacteria counts in bulk milk: what needs cleaning, the machine or the cows?

Douglas J. Reinemann
University of Wisconsin-Madison

Introduction and Overview

I have had a professional fascination with bulk tank cultures for the past 20 years. This odd obsession developed from my interest in the mechanics of milking machine cleaning and sanitation. Over the course of the past two decades I have been involved in numerous situations in which bulk tank bacteria counts were higher than desired, most often approaching or exceeding the legal limit. A remarkable number of these crisis situations share a common thread: an argument between the milking machine service technician (or cleaning chemical supplier) and the farmer over whether the cause of the high bacteria counts was a failure in the cleaning system or the excessive bacteria load from milking wet/dirty cows. A very crude estimate based on my memory of interesting cases is that there are not overwhelming odds for either of these 2 primary sources, but that when the legal limit is being exceeded, it is common that both sources contribute to the crisis situation. In this paper, I will provide the tools that I have found most useful and effective in diagnosing the source of bacteria in bulk tanks, which, in turn, will allow the investigator to focus intervention in the right place to solve the problem.

Bulk tank cultures: know your enemy

A simple, yet powerful, method for diagnosing high bacteria counts in bulk tank milk using the relative relationships between bulk tank standard plate count (SPC), somatic cell count (SCC), laboratory pasteurised count (LPC), and coliform count was presented by Guterbok and Blackmer (1984) and is used as the basis for the NMC Guide, Troubleshooting Cleaning Problems in Milking Systems (Figure 1).

This method uses coliform bacteria as an indicator of the level of environmental contamination (organisms drawn into the milk from the environment – mainly from the skin of teats and udders) in bulk tank milk. There are, however, many other types of environmental bacteria and elevated coliform counts can occur for other reasons. The method also uses the LPC (or thermoduric count) as the primary indicator of a cleaning failure in milking and milk storage equipment. Again, there are many different types of thermoduric bacteria and elevated LPC can occur for reasons other than cleaning failures.

This method relies on the RELATIVE COMPARISON between numbers to formulate a diagnosis. The most common misapplication of the method is the formulation of a diagnosis based on only one of these numbers without considering the relative values of the others. The simple chart in Figure 1 is intended to highlight the diagnostic measure that is the most elevated compared to the others. Note that the different types of counts have different ‘target’ values based on the performance of a typical dairy herd.

Figure 1: Diagnostic chart for bulk tank bacteria counts.
LPCs are correlated with coliform counts because there are thermudric bacteria that are present in the environment. Coliform (and other environmental organisms) make up a larger percentage of the population than thermudric organisms so that the increase in coliform count is larger than the increase in LPC. Following are some examples of the 3-part decision tree to properly implement this diagnostic technique:

Milking wet and/or dirty cows:
- Coliform count is between 100 and 1000 cfu/mL
- LPC is less than Coliform count
- SPC is moderately elevated (5000 – 20,000) cfu/mL

Persistent milking machine cleaning problem:
- LPC is between 100 and 1000 cfu/mL
- Coli less than LPC (probably because of the use of an effective sanitise cycle)
- SPC is moderately elevated (5000 – 20,000 cfu/mL)

Incubation in the milk handling system:
- Coliform count is greater than 1000 (or to numerous to count TNTC)
- LPC is greater than 100 but less than Coliform count (Or TNTC)
- SPC is extremely elevated (greater than 50,000 to 100,000 or TNTC)

Multiple sanitation problems are likely contributing to these elevated counts and further investigation is recommended (strategic sampling from various points in the milk handling system both early and late in the milking process).

Acquiring more information about the specific bacterial species represented in bulk tank milk will improve the power of a diagnosis. Quantitative bulk tank cultures (QBTC) enumerate a range of specific organisms. QBTC is typically focused on types of bacteria related to the level of mastitis in the herd (environmental and contagious). Some of these organisms are also useful in diagnosing sources of environmental contamination of milk, cleaning failures and incubation in milk handling equipment. Following is a summary of sources and growth characteristics of specific bacteria types commonly found in bulk tanks from the excellent review by Murphy and Boor (2008) (additional comments by the author in italics). This continually updated document on the E-extension website is required reading for anyone interested in the diagnosis of bulk tank bacteria counts. Applying this research based information for a broader range of bacteria types will greatly improve your diagnostic abilities.

1. Mastitis organisms:
   a) Mastitis organisms that most often influence bulk milk count are Streptococcus spp., most notably Strep agalactiae and Strep uberis.
   b) Staphylococcus aureus is not a frequent contributor to total bulk tank bacteria count.
   c) Detection of (environmental) mastitis pathogens does not necessarily indicate that they originated from cows with mastitis as environmental mastitis pathogens occur in milk as a result of factors other than mastitis infection.
   d) Correlation of somatic cell responses and bulk tank environmental mastitis organisms is poor.

2. Environmental Contamination
   a) Organisms associated with bedding materials that contaminate the surface of teats and udders include streptococci, staphylococci, spore-formers (or thermudric) coliform, and other Gram-negative bacteria.
   b) Both thermudric (bacteria that survive pasteurisation) and psychrotrophic (bacteria that grow under refrigeration) strains of bacteria are commonly found on teat surfaces. Contamination from the exterior of the udder can influence Lab Pasteurisation Counts (LPC) and Preliminary Incubation Counts (PIC).
   c) Milking heavily soiled cows could potentially result in bulk milk bacteria counts exceeding 10^6 (or 10,000) cfu/mL, although higher coliform (or other environmental bacteria) counts are more likely to occur due to incubation in milk handling equipment. Elevated bulk tank coliform counts can also result from coliform mastitis in the herd.

3. Cleaning and Sanitation
   a) Significant buildup of (thermoduric) organisms in milk residue to a point where they influence the total bulk tank count may take several days to weeks (and are therefore an indication of a persistent cleaning failure). Old cracked rubber parts are also associated with higher levels of thermudric bacteria.
   b) Some types of cleaning failures can also select for faster growing, less resistant organisms, principally Gram-negative rods (coliforms and Pseudomonads) and lactic streptococci and can result in high PIC.
   c) Effective use of chlorine or iodine sanitisers has been associated with reduced levels of psychrotrophic bacteria that cause high PIC.

4. Refrigeration
   a) Elevated psychrotrophic bacteria counts are often associated with poorly cleaned refrigerated bulk tanks.
   b) In milk produced with low initial psychrotrophic populations, psychrotrophic bacteria can quickly become dominant after incubation at 4.4°C (40°F) resulting high PIC.

Statistics: transforming data into information

Bacteria of all types grow at an exponential rate and therefore produce highly skewed distributions. The same is true for the increase in somatic cell counts in cows infected with mastitis. The linear mastitis score was developed to adjust these highly skewed SCC indicators into a linear effect on milk production. A log transformation will convert bacteria count data into a more normally distributed population and give a better estimate of the resulting milk quality effects of increased bacteria counts. Log transformations therefore offer a better yardstick for true deviations in bacteria count data and provide a more accurate assessment of deviations over time.
Investigating the influence of udder hygiene and pre-milking sanitation

A wide variety of bacteria species can be harvested from the skin of teats and udders during milking. The ‘wipe test’ method described in Reinemann et al. (2008) has been used to assess the bacteria population on teat skin both before and after pre-milking sanitation. The results of this method used on three farms using both SPC and DBC methods pre- and post-teat sanitation are shown in Figure 3. These two tests allow a comparison across farms or within a farm over time of the general bacterial condition of teat skin as influenced by farm management and weather conditions. Comparison of the pre- and post-teat sanitation practice can be used to assess the effectiveness of that practice.

DBC technology typically enumerates many more bacteria than plate count methods. Plate count methods rely on the recovery of viable bacteria which form colonies on growth media, whereas DBC technology can enumerate both viable and killed bacteria. In this study the reduction in DBC were used as an estimate of the effectiveness of removing solids from the teat skin in a similar way to the previous studies which used various types of tracer materials. The comparison of viable to viable plus unviable bacteria reductions also allowed for an estimate of the killing action of pre-milking teat disinfectants.

Further insight is gained by measuring the prevalence of specific bacteria types, as illustrated in Figure 4. It is interesting to note large differences in the LPC counts across farms as well as differences in the relative populations of LPC vs. Coli and other environmental bacteria. Experience with this method on a number of farms has indicated that contamination of bulk milk by thermoduric bacteria can occur because of the harvest of these bacteria from teat skin. It is also common for the environmental bacteria to also be present in large numbers in these situations. It is also interesting to note that pre-milking disinfectants are less effective at reducing thermoduric bacteria counts than common environmental organisms such as coliform, streptococcal and staphylococcal bacteria.

Cleaning the milk handling equipment

A procedure for troubleshooting cleaning problems was developed by the machine milking committee of the NMC and last updated in 2004. This method begins with an analysis of bulk tank bacteria cultures. The methods presented above provide further clarification and new developments in this method. The purpose of this exercise is to determine if cleaning problems are the most likely cause of elevated bacteria counts.
There have been innumerable attempts to fix a high bacteria count by focusing attention on the cleaning system when the likely cause of an elevated count (especially a ‘spike’ or short term or single event of a rise in SPC or LPC) was actually caused by environmental contamination. A common response in such a situation is to apply a ‘shock’ treatment to the milking machine in which chemical concentrations and wash water temperatures are greatly increased. This practice usually results in a substantial degradation of the rubber components in the milk handling system resulting in a milking machine that is much more difficult, or impossible, to clean effectively.

If bulk tank cultures indicate that a cleaning failure is the likely cause of elevated bacteria counts, there are a few basic concepts to further determine the causal mechanism.

1. Are the cleaning procedures being implemented properly and with the appropriate frequency?
   a) Proper implementation of the cleaning procedures includes using the appropriate concentration of cleaning chemicals and water temperature.
   i) Check the product labels to ensure that the cleaning chemicals are being used correctly. Increasing water hardness requires increasing detergent concentration for the same cleaning efficacy. Recommendations for the proper detergent concentration should include a test of water hardness in addition to a test of the alkalinity of the cleaning solution as mixed on the farm.
   ii) Water temperatures can be checked with a pocket thermometer.
b) It is often difficult to determine if the cleaning cycles are being executed with the proper frequency. A temperature recorder in the bulk tank can be used to determine if the expected temperature rise occurs at the frequency that cleaning should be performed. Many modern milking machine wash controllers also record this information for the cleaning cycles in the milking machine. These are very useful sources of information and should be used whenever available.

2. Are the cleaning solutions being circulated throughout the entire milking machine?
   a) There are two fundamentally different flow patterns in a milking machine cleaning system.
      i) The milking units and milk meters if present: Cleaning of these components requires that cleaning solutions have long contact times as the agitation and turbulence developed in them is very low. This can be achieved by ensuring that these components are ‘flooded’ or full of cleaning solution, during a majority of the cleaning cycle. This can be assessed by visual assessment, or more accurately by measuring the water flow through each unit during the cleaning cycle with a simple flow meter.
      ii) Flow dynamics in the milk line: In smaller milking machines the milk line may also be cleaned by flooding the milk line. As the diameter and length of the milk line increase this becomes impractical due to the large volume of water required. Air injection provides increased mechanical cleaning action in the milking and reduced the amount of water and cleaning chemical required to achieve effective cleaning. A vacuum recorder can be used to determine if air-injected slug flow results in complete coverage and agitation in the entire milk line. A vacuum drop of at least 15 kPa at each milk inlet on the milk line during each air injection cycle is a good indication that milk line flow dynamics are effective.

Further details of test methods and diagnostics are available in references.

Summary

- Know your enemy: Acquiring information about the specific bacterial species represented in bulk tank milk will improve the power of a diagnosis. Use bulk tank cultures to determine that a cleaning failure is the most likely cause of elevated bacteria counts before trying to ‘fix’ the cleaning system.
- Bacteria count data should be converted into log values so that statistical analysis and process control algorithms are valid and more sensitive.
- The moving average is a useful tool to assess long term trends in bacteria populations in bulk milk.
- Correlation is a useful tool to aid in the diagnosis of the types of bacteria contributing to short-term changes in bulk tank bacteria counts.
- Incubation causes dramatic increases in some bacteria types but not in others. Knowledge of the growth rates of different bacteria types under different conditions will improve diagnostic methods.
- Premilking sanitisation will reduce bacteria population on teat skin but not eliminate bacteria. Cows entering the milking with a higher degree of teat contamination will still have a higher degree of teat contamination for the same method of pre-milking teat sanitisation than cows with lower initial teat contamination.
- Post-milking sanitisers are more effective at reducing the population of some bacteria types (environmental) than others (thermoduric).
- If bulk tank cultures indicate that a milking machine cleaning failure is likely, begin your troubleshooting by checking the simple things first.
  - Is something broken? (Water heater, chemical mixer, valves, switches, etc.)
  - Are chemicals being used properly (according to label directions for concentration adjusted for water hardness, temperature and circulation time)?
- Move to an analysis of flow dynamics after these have been confirmed.

References

The causes of, and solutions to, bacterial downgrades in milk on Australian dairy farms with reference to the approach used in New Zealand

Josh Wheeler
QCONZ

Common problems/faults in Australia over recent years

- Very little use of chlorinated alkalis in the wash routine. Predominately non-chlorinated liquid alkaline.
- Lack of understanding of how chlorine is used. Some utilisation as a sanitiser but not rinsed and not routinely used as part of the hot alkaline wash to remove protein.
- Very little use of Acid Sanitiser detergents in the wash routine to neutralise the alkaline wash and leave the plant sanitised.
- Farmers do not routinely inspect the cleanliness of the milking machine.
- Lack of understanding about what the bacterial grades are, what to look for and how to rectify.
- No industry standard for carrying out a full plant inspection where milk is down-graded.
- Some poorly installed plants and industry does not have an approval system in place for new installations.
- Hot water volumes, especially for the vat wash system, are not adequate.
- Plant wash tub sizes do not match the plant size.
- Lack of knowledge around wash programs.
- Plant wash system is not part of the annual milking machine test so is not reviewed or checked.

Causes of bacterial grades in milk

Milk is basically sterile when secreted into the alveoli. From this point there are three potential contamination points.

1. Within the udder (Mastitis)

   When a cow has mastitis it has a bacterial infection of the udder and can shed large amounts of bacteria into the milk. The amount that mastitis bacteria influences the bulk milk will depend on the bacteria type. Bacterial grades in the bulk milk due to mastitis bacteria are not common when milk is tested using the Bactoscan test as the Bactoscan tester tends to undercount mastitis bacteria.

2. Exterior of the udder

   Bacteria on the exterior of the teat from manure, soil and feed residues left on the teats has the potential to increase the bacteria level. The influence of the exterior of the udder on the bulk milk bacteria levels will depend on the cleanliness of the cow’s environment and how the cows are prepared for milking. Only clean dry teats should be milked.

3. Surface of milking machine, bulk tank and also water source

   The predominant source of bacterial contamination of bulk milk is from unclean milking machine surfaces and/or milk vat surfaces.

   All the above sources can influence the bacterial level of the bulk milk either individually or in combination with the two other sources.

Current bacteria tests

Bactoscan Test

- Bactoscan tests for total bacteria in milk. The level at which milk is down-graded can vary from 50,000 – 100,000 total bacteria per mL. In New Zealand all dairy companies grade milk on the Bactoscan results at >50,000 per mL.
- Bacteria are electronically counted. Testing is reasonably quick (6 minutes). Results are potentially available to the farmer within 24 hours of the milk being collected.
- Milk is randomly tested normally three times per month. If the test result is above the grade level then milk will be tested every consignment until there is a specified number of clear tests. Once this is achieved that supply goes back on random testing.
- All major dairy companies around the world are now using the Bactoscan machine for testing for total bacteria in milk.
- The Bactoscan machine is regularly calibrated against the standard plate count method.
- High total bacteria counts are normally associated with a fresh deposit when found in a milking machine.
• Total bacteria multiply rapidly in warm milk.
• Bacterial contamination through poor quality water supply has caused very few grading problems in New Zealand as long as the last wash is an acid sanitiser and the plant is left to drain and not flushed with raw water.

**Common Causes**
- The milking machine/bulk tank (most common cause >95% of grades)
  - Unclean milking plant/vat
  - Inadequate plant/vat cleaning system
  - Mechanical failure of milking machine components
  - Worn/perished rubberware
  - Inadequate refrigeration
- Poor cow preparation (unclean teats/udder)
- Mastitis

**Thermoduric Test**
- The thermoduric test identifies specifically heat resistant bacteria in milk by firstly pasteurizing the milk sample so that only heat resistant bacteria are present. Then the milk is plated and incubated for 3 days.
- Not all dairy companies test for Thermoduric bacteria. Dairy companies who do test normally set their grading level between 1,500-5,000 per mL. All New Zealand dairy companies test for thermoduric bacteria and all grading levels are set at 1,500 per mL.
- The test at the laboratory takes 3 days to complete. Therefore a thermoduric result should be available to the farmer 4 days after the collection.
- Milk will be randomly tested initially. If the test result is above the determined grade level then it will be tested every consignment until a specified number of consecutive clear tests has been achieved. Once this is achieved the supply goes back on random testing.
• Thermuduric bacteria are slow growing and like an ambient air temperature above 20°C.
• Can survive through the hot wash cycles.
• High thermuduric counts are normally associated with an aged deposit and perished rubberware when found in a milking machine.
• Will multiple in warm milk but at a slower rate than other bacteria.
• Normally found in areas where hot water is getting to and killing off their competition (other bacteria) but not adequately cleaning the surface.
• As far as I'm aware, thermuduric bacteria contamination through a water supply has never been identified as a problem in New Zealand.

Common Causes
• The milking machine/bulk tank (most common cause >95%)
  – unclean milking plant/vat
  – inadequate plant/vat cleaning system
  – mechanical failure of milking machine components
  – worn/perished rubberware
  – inadequate refrigeration.
• Environment (poorly made silage).
• Mastitis (mastitis has not been linked to a thermuduric grade).

Solving bacterial grades

Milking machine inspection
More than 95% of the bacterial downgrades of milk in New Zealand are caused by an unclean milking machine due to one or more of the following:
• mechanical failure
• operator error
• poor machine maintenance and rubberware replacement
• inadequate wash system
• inadequate wash program
To identify what part of the milking machine is unclean a full plant inspection must be carried out. The plant inspection needs to follow the path of the milk from the clusters to the milk vat with every component being inspected. If the plant is the cause then a full plant inspection will find the problem when done thoroughly. As part of the plant inspection the milk refrigeration needs to be checked to ensure it is chilling the milk quickly. Also the plant/vat wash systems and wash programs need to be checked to ensure they are correct.

When a fault is found then you need to figure out why this particular part of the milking machine is not cleaning well and decide what can be done to improve the cleaning.

Other tools for hard-to-solve grading problems
In some situations, a full plant inspection will not find the source of the downgrade and other tools will have to be used to narrow down what is causing the grading situation.

Data Loggers
The installation of data loggers on the milk delivery line and milk vat will allow the evaluation of:
• plant and vat hot wash temperatures
• duration of plant and vat hot washes (contact time)
• effectiveness of milking cooling system.

Point sampling
The sampling of milk at specific points consistently throughout milking can be very effective at narrowing down where the bacterial contamination is occurring. We have used this a lot in the past in New Zealand to help solve difficult grading problems. Point sampling was used 10 years ago to link poorly made silage as the cause of some of our ongoing thermuduric grading situations.

More recently, the amount of situations in New Zealand where we require point sampling to solve the grading problem has reduced. This dairy season it was only used on 4% of the grading situations we were involved in. The reduction in the need for point sampling is due to increased emphasis and training on how to complete a full plant check.

Solution and Prevention
To effectively clean plants you must concentrate on four key areas: Water, Detergents and Sanitisers, Contact Time and Turbulence.

Farm dairy water
Water quality is important for ensuring the water does not contaminate the milking equipment with bacteria, soil or minerals. Poor quality water also affects the ability of detergents and sanitisers to clean the milking equipment. If water quality is poor and thus reducing the effectiveness of the wash then options should be considered to improve the water quality.

Adequate quantities of hot and cold water also needs to be available for effective washing of the plant and vat. The New Zealand requirements ask that there is enough hot water available to wash the plant and vat at the same time with the plant needing 10 litres of hot water per cluster and the vat 2% of the volume of the vat with a minimum of 120 litres.

An example of the NZ requirement for hot water volume:

<table>
<thead>
<tr>
<th>Type of Plant</th>
<th>Hot Water Volume Required</th>
</tr>
</thead>
<tbody>
<tr>
<td>50 Bail Rotary (10 litres per cluster)</td>
<td>500 litres</td>
</tr>
<tr>
<td>30,000 litre silo (2% of the volume)</td>
<td>600 litres</td>
</tr>
<tr>
<td>Total hot water required</td>
<td>1100 litres</td>
</tr>
</tbody>
</table>

Every dairy farm in NZ is audited annually and one of the requirements is that they must meet the above hot water standards. All existing farms have to be within 70% of the
standard and all new dairies within 90% of the standard. All
dairy companies enforce this rigorously. The enforcement of this
standard at a farm level has been one of the main drivers in the
improvement of milk quality at a farm level over recent years.

The quantities of hot water on Australian farms is one of the
things we have noticed to be inconsistent with a lot of farms
having very low quantities available.

**Temperature**

All milk fat melts at temperatures above 70°C therefore we
need to ensure the wash temperatures for the plant and vat
are above 70°C. Hot water also kills bacteria and improves the
reactivity of the detergents and therefore detergents generally
work better in hot water than cold water.

**Table 1: NZ temperature guidelines for hot washing milking
machines.**

<table>
<thead>
<tr>
<th>Wash Temperature Guide</th>
<th>Initial Temp in wash tub</th>
<th>Plant</th>
<th>80-85°C</th>
<th>80°C</th>
<th>Dump temperature of wash</th>
<th>Plant</th>
<th>55°C</th>
<th>Vat</th>
<th>55°C</th>
</tr>
</thead>
</table>

**Detergents and sanitisers**

Detergents and sanitisers are used to remove soil from the
milking plant and kill bacteria. The role the detergents play is
illustrated in Figure 3.

**Acid and acid sanitiser detergents**

Acid detergents remove mineral soils. Acid detergents can
incorporate a sanitiser (Acid Sanitiser Detergent). The sanitiser
is there to kill bacteria. Acid Sanitiser detergents are designed
to be used in the last wash cycle and left to freely drain from
the plant after washing. This provides extended sanitising
protection between milkings.

**Alkaline and chlorinated alkaline detergents**

Alkaline detergents are designed to remove fat. Chlorinated
Alkaline detergents should be used in a wash program. They
should always be used with hot water. The caustic ingredient
removes fat and the chlorine removes protein deposits. The
surfactants help dissolve and retain soils in suspension until
removed from the plant/vat when the wash water is dumped.

Alkaline detergents leave a powdery residue on stainless/
rubberware and must be rinsed from the plant. Chlorine
attacks black rubber components if left in contact for long
periods (between milkings).

**Sanitisers**

A sanitiser is a substance that kills bacteria. The most common
sanitisers you will be familiar with include Quaternary
Ammonium Compounds (QACs) which are found in most acid
detergents, iodine which is found in Iodophors and chlorine. A
sanitiser can also be a condition that kills bacteria rather than a
substance. The best example of this is temperature which will
kill bacteria during pasteurising.

**Typical NZ plant wash program for effective cleaning**

**Morning Plant Wash Routine**

<table>
<thead>
<tr>
<th></th>
<th>Mon</th>
<th>Tues</th>
<th>Wed</th>
<th>Thur</th>
<th>Fri</th>
<th>Sat</th>
<th>Sun</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cold rinse</td>
<td>C</td>
<td>C</td>
<td>C</td>
<td>C</td>
<td>C</td>
<td>C</td>
<td>C</td>
</tr>
<tr>
<td>Chlorinated alkali</td>
<td>H</td>
<td>H</td>
<td></td>
<td>H</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Acid sanitiser</td>
<td>C</td>
<td>H</td>
<td>H</td>
<td>H</td>
<td>C</td>
<td>H</td>
<td>H</td>
</tr>
</tbody>
</table>

Note: The chlorinated alkali wash should be recycled for 7-10 minutes

**Afternoon Plant Wash Routine**

<table>
<thead>
<tr>
<th></th>
<th>Mon</th>
<th>Tues</th>
<th>Wed</th>
<th>Thur</th>
<th>Fri</th>
<th>Sat</th>
<th>Sun</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rinse</td>
<td>C</td>
<td>C</td>
<td>C</td>
<td>C</td>
<td>C</td>
<td>C</td>
<td>C</td>
</tr>
<tr>
<td>Chlorinated alkali</td>
<td>H</td>
<td>H</td>
<td>H</td>
<td>H</td>
<td>H</td>
<td>H</td>
<td>H</td>
</tr>
<tr>
<td>Acid sanitiser</td>
<td>H</td>
<td>H</td>
<td></td>
<td>H</td>
<td></td>
<td>H</td>
<td>H</td>
</tr>
</tbody>
</table>

Note: The afternoon acid sanitiser wash can be carried out in cold water,
but must be done in hot water when in colostrum cows and/or treated
cows are milked through the plant last and/or when bacteria grading.

**Vat wash routine**

<table>
<thead>
<tr>
<th></th>
<th>Mon</th>
<th>Tues</th>
<th>Wed</th>
<th>Thur</th>
<th>Fri</th>
<th>Sat</th>
<th>Sun</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cold Rinse</td>
<td>C</td>
<td>C</td>
<td>C</td>
<td>C</td>
<td>C</td>
<td>C</td>
<td>C</td>
</tr>
<tr>
<td>Chlorinated Alkali</td>
<td>H</td>
<td></td>
<td>H</td>
<td>H</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Acid Sanitiser</td>
<td>C</td>
<td>H</td>
<td>H</td>
<td>H</td>
<td>C</td>
<td>H</td>
<td>H</td>
</tr>
</tbody>
</table>

Note: The chlorinated alkali wash should be recycled for 5-7 minutes.
When milk is collected every second day Alkaline wash every second pickup.

**Turbulence/mechanical**

Milking machines are designed primarily to harvest milk from
the cow and deliver it to the storage tank without causing
adverse damage to the milk being harvested. Operating a
milking machine requires both air and liquid to flow from

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**Figure 3: Detergents and sanitisers and the role they play in
removal of soil and killing bacteria.**
Tackling *Strep uberis*; mastering plant hygiene

The causes of, and solutions to, bacterial down-grades in milk

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the cluster to the receiver. The milk flow through the milkline is laminar flow. However, for an effective clean we require turbulence and therefore a turbulent flow through the milkline into the receiver. Under laminar flow parts of the milkline and receiver will not clean. So the requirements of the wash are completely the opposite to the requirements of milk harvesting.

It is impossible to create effective turbulent flow in a milking machine without the use of a washline injector or flushing pulsator. A flushing pulsator, set correctly, will create the necessary slugging action to clean the milkline and receiver can. Figure 4 illustrates no slugging and slugging action.

Correctly installed flushing pulsators will store water and fire every 35-45 seconds and generate enough of a slug of wash water to fill the milkline. The flushing pulsator needs to fire 10-12 times during the chlorinated alkaline wash to effectively clean the milkline.

**Contact time**

Contact time is essential as it gives the detergent the time to remove soil deposits. The longer the contact time, within limits, the more effective the wash will be. The limiting factor is temperature. As the temperature drops the chance of soil re-depositing increases. To avoid this, recycling must cease before the solution falls below 55°C. For the temperature to be maintained you need to have sufficient hot water quantity and the right initial hot wash temperature.

**Key benefits of contact time**

- Increased contact time between wash solution and surfaces – meaning the detergents have more time to effectively clean
- More chance for the hot water to melt fat and kill bacteria
- Essential to give the flushing pulsator time to effectively clean the milk line
- The main wash that requires recycling is the chlorinated alkaline wash.
- The milk plant/vat should be recycled for between 5-10 mins and for closer to 10 minutes when incurring downgrades

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**Other NZ initiatives that have driven improved milk quality**

**Training**

NZ has basically used a similar format to Countdown but focused on Bacterial grading with a bit of basic mastitis understanding. The industry has developed a course for farmers called Licensed to Milk Stage 1. This course has been delivered to about 8500 farm owners and staff. This course has lifted farmer understanding of:

- what they are being tested for and why
- penalty system and proactive prevention measures
- milking machine function
- plant cleaning
- completing a plant inspection and why it is so important
- basic mastitis detection, prevention and recording

By default the course has also set the guidelines for industry in terms of wash programs and plant inspections.

**Grade ownership**

Industry has ensured ownership of grading problems are the responsibility of the farmer. Dairy companies will assist by phone information or organising a service provider to visit and complete a plant inspection but they will not visit the farm to assist. In the past, dairy companies have visited farms to solve grading problems but this was difficult. If they couldn’t solve the problem then how could they penalise the farmer?

**Penalty system**

Over recent years, the penalty system has been tightened up by all dairy companies. The result is that farmers that grade more lose more money. Financial penalties, in a lot of cases, are incurred from the first grade day. This system has put pressure on farmers to be proactive in plant inspection and plant maintenance.
Dairy company initiated service provider visits

More recently, dairy companies have been proactive in getting service providers on farm quickly to assist the farmers in rectifying the grading problem. The NZ industry uses a grade relief system: if a farmer seeks professional help, the cost will be deducted off the grade penalties.

Conclusion

Solving on-farm grading situations relies heavily on a full plant inspection to identify unclean surfaces. Once the problem is located, it must be fixed to ensure the problem does not occur again.

The plant wash system needs to incorporate a chlorinated alkaline wash in the wash routine to ensure the effective removal of protein and fat. The use of an Acid/Sanitiser combination detergent is very effective at neutralising the alkaline wash and leaving the plant sanitised between milkings.

Effective plant washing relies on correct volumes of hot water for the plant and vat and this must be used at the correct temperatures. The importance of hot water cannot be underestimated in the wash routine.

Education at a farm level has a key role to play in improving on-farm milk quality. Farmers and farm staff need to understand the common causes of bacterial grades, key components of an effective wash routine and how to complete a plant inspection. With this knowledge they are better able to be proactive in plant cleaning and, if they do have a problem, are more likely to rectify it by themselves. This should result in a much quicker turn-around time.
Countdown activities in 2010-11 – recent paths, future journeys

John Penry,* Pauline Brightling, Anne Hope  
Harris Park Group  
* Camperdown Veterinary Centre

Milk quality is important to farm productivity and profitability, and fundamental to the success of the supply chain and the resilience of the dairy industry (Figure 1).

Maintaining milk quality is a complex, multi-factorial, pre-farmgate activity. There is no post farmgate fix for increased cell counts in milk as a result of mastitis infection.

Australia’s milk quality is a combination of outputs from 7,400 dairy farm businesses. Dairy companies let their supply base know the standard of milk quality they want through individual contracts and milk quality payment schemes. This provides a strong commercial signal – but not necessarily the capacity to respond.

To reduce the risk of mastitis, farms need to have consistent milking routines, optimal milking machine performance, and good hygiene at milking, drying-off and calving every day. Knowing what to do and being able to respond appropriately to changes in circumstances is a lot to ask of dairy teams. An ‘enabling’ environment that supports farmers to achieve this (such as having access to dairy service providers, competent staff and good data) requires a collective approach to be achievable and affordable.

Countdown in consultation with industry

The Countdown project team was re-established after the “All Stars” Mastitis Control Symposium in March 2010. Facilitated by Dairy Australia seed funding, a series of industry consultations was embarked upon during April to July of that year. This consultation process was a mixture of meeting based and online surveys, and face to face interactions with processors. Table 1 summarises the common topics of concern identified during this consultation and planning phase.

Following the assimilation of consultation results the Countdown objectives for 2010-11 emerged. Countdown’s prime objectives over that period were to make the core resources for mastitis control widely accessible and to reinvigorate the regional networks of advisers. There would be emphasis on clinical case management and Strep uberis control in response to technical issues identified as high priority by industry sectors.

Activities in detail

**Countdown Core Resources:** The Countdown Farm Guidelines for mastitis control were first published in 1999 and over 14,000 have been distributed. They remain the foundation stone of clear mastitis control messages for farmers. One of the first Countdown activities was editing the Guidelines for inclusion on the Dairy Australia website (www.dairyaustralia.com.au). A second edition of the Guidelines is now in pre-production ready for publication (print and web) later in 2011. Alongside this installation on the Dairy Australia site was a change to the Countdown website (www.countdown.org.au). This site has now become the primary portal for advisers. The site includes
Awards being distributed in June this year to recognise the farms in the lowest 5% of BMCC for the 2011 on-farm mastitis and cell count control. Data collated from Awards are now firmly established as an indicator of excellence.

The Countdown Milk Quality Bulk Milk Cell Count Control: Queensland was part of the post flood response for the Murray region, NSW and Queensland. Mastitis after wet and humid conditions were developed as this, resource sheets dealing with the management of clinical mastitis and, in particular, mastitis caused by Strep uberis. One indicated the need for new resources around managing clinical mastitis, and often specifically Strep uberis (how to reduce exposure or deal with endemically infected herds). Whether the milk pricing signals are helping or hindering action.

Managing clinical mastitis: Industry consultation clearly indicated the need for new resources around managing clinical mastitis and, in particular, mastitis caused by Strep uberis. One significant milestone in this area of activity is the Strep uberis Technote FAQ being launched at the Symposium. In addition to this, resource sheets dealing with the management of clinical mastitis after wet and humid conditions were developed as part of the post flood response for the Murray region, NSW and Queensland.

Bulk Milk Cell Count Control: The Countdown Milk Quality Awards are now firmly established as an indicator of excellence in on-farm mastitis and cell count control. Data collated from all processors in the calendar year 2010 has been employed to recognise the farms in the lowest 5% of BMCC for the 2011 Awards being distributed in June this year.

Table 1: Key issues raised by processors, advisers and farmers in the Countdown industry consultation process

<table>
<thead>
<tr>
<th>Dairy processing companies (8 companies, &gt; 90% of Australian supply)</th>
<th>Advisers (96)</th>
<th>Farmers (260)</th>
</tr>
</thead>
<tbody>
<tr>
<td>• Antibiotic residues in milk associated with mastitis treatments</td>
<td>• Lack of professional milking machine technicians who can install and service milking equipment correctly, find and fix problems competently, and report clearly</td>
<td>• Dealing with clinical mastitis – especially questioning treatment effectiveness, concern about the costs ($ and time) of early detection and treatment, and clinical mastitis at calving</td>
</tr>
<tr>
<td>• Unacceptably high levels of thermoduric bacteria (mainly due to poor cleaning methods, old rubber-ware, poor milking system installation)</td>
<td>• Early signs of breakdown in Countdown's clear, consistent messages</td>
<td>• Environmental mastitis and Strep uberis in freshly calved cows</td>
</tr>
<tr>
<td>• Inadequate numbers or skills of local milking machine technicians</td>
<td>• Need for on-going training (refresher courses and skills development) for advisers and farmers</td>
<td>• Many concerns about the ability and commitment of staff (especially new workers, casual and relief staff)</td>
</tr>
<tr>
<td>• Too many herds with chronically high cell counts</td>
<td>• Ways of improving staff training and commitment to best practice on farm</td>
<td>• Ability to have milking machines operating optimally and access to milking machine technicians willing to do tests</td>
</tr>
<tr>
<td>• Need help with training for their field staff</td>
<td>• Issues about environmental mastitis, and often specifically Strep uberis (how to reduce exposure or deal with endemically infected herds)</td>
<td>• The expense of BMCC control</td>
</tr>
<tr>
<td></td>
<td>• Whether the milk pricing signals are helping or hindering action</td>
<td>• Lack of experts (in some districts)</td>
</tr>
<tr>
<td></td>
<td>• Unwillingness of many farmers to tackle high cell count issues</td>
<td>• Loss of income due to milk quality penalties and low milk price</td>
</tr>
<tr>
<td></td>
<td>• Frustration with having to deal with chronically high cell count herds</td>
<td>• High Bactoscan/TPC results (and questioning the reliability of factory testing).</td>
</tr>
<tr>
<td></td>
<td>• Time lag and lack of useful results from milk cultures from herds</td>
<td></td>
</tr>
</tbody>
</table>

the Mastitis Investigation Pack, Technote updates, newsletter grabs among other items.

Countdown Mastitis Focus has continued to be used by advisers since its inception in 2009, either through the report website (www.mastitisfocus.com.au) or direct from Mistro centre software through a herd improvement organisation (HIO). In response to the floods and wet conditions in Northern Victoria two HIOs commenced production of Mastitis Focus reports, as a monitoring and investigation tool, with every herd test day summary report. For herds enrolled in herd testing, Mastitis Focus has become the first line tool for monitoring and herd investigation work.

Managing clinical mastitis: Industry consultation clearly indicated the need for new resources around managing clinical mastitis and, in particular, mastitis caused by Strep uberis. One significant milestone in this area of activity is the Strep uberis Technote FAQ being launched at the Symposium. In addition to this, resource sheets dealing with the management of clinical mastitis after wet and humid conditions were developed as part of the post flood response for the Murray region, NSW and Queensland.

Bulk Milk Cell Count Control: The Countdown Milk Quality Awards are now firmly established as an indicator of excellence in on-farm mastitis and cell count control. Data collated from all processors in the calendar year 2010 has been employed to recognise the farms in the lowest 5% of BMCC for the 2011 Awards being distributed in June this year.

Work remains to be completed by the Countdown team in updating the mastitis dynamics model used successfully to promote ‘Cell Count Solutions’ through the processors. Many advisers have employed the Benefits of a lowered cell count "step graph" to illustrate the value proposition in sustainably lowering BMCC through adoption of best practice. Updating this resource to reflect current milk pricing and common Quality Payment schemes remains a priority.

Strengthening regional networks: Since early 2009 there have been signs of fracturing in the clear consistent mastitis control messages among advisers through, in part, industry turnover and a continuation of the reduction in adviser numbers in many dairying regions. The extended and severe wet summer conditions in Northern Victoria, NSW and Queensland presented Countdown with a clear need to assist the regional network of advisers to respond to increasing mastitis levels. This response will be outlined later in this paper.

The 2011 Symposium also forms a significant part of re-energising the regional network. As with 2010, all dairy areas and all professional groups are represented at this meeting. The Countdown symposium has emerged as the flagship mastitis control information forum, drawing on international speakers, for Australian advisers.

Cups on cups off: The Countdown Cups on cups off course has been delivered to industry since 2008 and has become one of the most successful, and well subscribed, courses offered by the National Centre for Dairy Education Australia (NCDEA). The course was designed specifically around mastitis control.
tasks during the milking process with the farm team in mind. Since the start of 2010, about 30 courses have been completed utilising a group of Countdown trained advisers.

Countdown’s response to the floods and wet conditions during early 2011

From late December 2010 onwards, parts of northern Victoria, the Riverina, Northern NSW and Queensland experienced extreme weather conditions resulting in widespread flooding, prolonged wet and humid conditions and, in the case of Far North Queensland, the destructive effects of tropical cyclone Yasi.

In the Murray region 136 dairy farms were flood affected. In a small number of cases around the Kerang area the water inundation persisted for more than three months. Around 900 farms in the Murray/Riverina regions experienced persistent wet conditions resulting in farm management problems (increased mastitis, lameness, reduced feedbase quality).

In Queensland and Northern NSW virtually all dairy farms were in disaster declared shires (152 farms in Northern NSW and 582 farms in Queensland including the Atherton Tablelands). Damage ranged from extensive flooding to persistent wet conditions with resultant damage to pastures, tracks, loafing and feeding areas.

Increased mastitis levels, as indicated through elevated BMCC and clinical case rates, were reported widely in the affected regions. All dairy processors reported persistent rises in BMCC and usually in the order of a 50,000 cells/mL increase in the individual factory average for January, February and March (Murray Goulburn, Fonterra, Tatura Milk in the Murray region and Parmalat and Dairy Farmers/National Foods in Queensland). Most factories had an average BMCC just over 300,000 for February (see the BMCC data example in Graph 1). Four large vet practices in Northern Victoria reported increases in lactating cow treatment sales of between 20% and 50%.

The Countdown input to the industry floods response was built around:

- Close liaison with the two flood and wet condition recovery groups facilitated by Murray Dairy, Subtropical Dairy/Queensland Dairyfarmers Organisation and Dairy Australia.
- Use of the existing adviser network to provide the quickest path to farmers with relevant control messages.
- Bolstering and up-skilling of the existing adviser networks, where necessary, given that Countdown had not had a regional presence in either Northern Victoria or Queensland since at least 2009.
- The creation of a resource kit for advisers and farmers using tailored, technically sound mastitis control information extracted from the existing Countdown materials. The creation of this ‘bespoke’ farm guideline set for wet conditions relied on the input of a small group of experienced advisers who co-developed the pack.
- Where possible, Countdown assisted with farmer meetings facilitated through local vet clinics and the Regional Development Programs in both Northern Victoria, NSW and Queensland. Countdown presentation materials were prepared for distribution to advisers running mastitis control meetings for farmers.
- Strong alignment with the processors who were responsible for frontline advice on farm and distribution of the elements of the mastitis control resource kit.

Developments into the near future

Countdown’s role in assisting with the flood and wet conditions recovery response in multiple regions highlighted the continuing leadership role in milk quality the project maintains. Having established momentum again, at a national level, the challenge now is to build on the activities of the last 12 months.

As already indicated, the second edition of the Countdown Farm Guidelines will be published later in 2011. While the architecture of the guidelines will remain the same, the text
will reflect changes seen in the updated Technotes as well as an increased alignment with other resources such as The People in Dairy project. Since 2004 there has been a 38% increase in the number of farms employing people who are not family members – it is likely that many of these people have never been able to become familiar with the contents of the Farm Guidelines.

Continuation of the revitalisation of the adviser networks in the regions remains important in the coming 12 months. The recent experience in multi profession meetings around the flood recovery indicates that engaging with advisers on more than one occasion is necessary to effectively re-build capacity. This is particularly the case where advisers might only have limited number of dairy clients and servicing the dairy sector is not a dominant part of their business. Sitting alongside this is a continuing demand for structured adviser training such as that experienced with the Countdown Adviser Short Course. Whilst this course was modified to incorporate the Mastitis Focus resource in 2009 a substantial re-design of the course is warranted. A course in late 2011/early 2012 is planned to address the needs of the 40 or so advisers who have indicated they would enrol in the next courses.

Changes to the Cups-On Cups-Off farmer course, delivered through the NCDEA, is also likely. Trainers in Northern Victoria have flagged a demand for a drying off section to be incorporated into the course and this will be piloted in July/August this year before a wider roll out.

The scientific review of best practice _Strep uberis_ control and the experiences of the farms in the wet conditions has raised the need for the industry to better understand pre-milking preparation of cows – when do we need to wash and dry teats? Countdown will explore ways to help answer this question.

As Dairy NZ builds its new mastitis control program (Smart SAMM) we are liaising with them as they adapt the Countdown Technotes for their use.

Since May 2011 a multi pathogen bulk tank molecular test kit has been commercially available in Australia (based on a Scandinavian PCR assay). Countdown has a role in crafting the clear messages around the application of this new technology so that it is employed to best effect, both as a screening test and an adjunct test as part of a planned mastitis investigation (Technote 13, 2003).

The recent _Dairy Moving Forward_ review of RD&E priorities has indicated the pressing need for workforce development in the milking machine technician area. Through diverse factors such as the resources boom drain on skilled labour and the reduction in dairy farm numbers, particularly outside of Victoria, milking technician training and industry alignment has become fragmented. Countdown has the potential to play a role in assisting the re-establishment of coordinated industry training for plant testing and fitting in addition to ensuring that technicians view their sector as having a defined and attractive career path.

So, as we approach the end of 2010/11, it is fair to ask the question: can Countdown continue to play a valuable role in the Australian dairy industry? We are one of the few countries in the world with a national milk quality program – only Australia, the Netherlands and NZ have successfully implemented national programs with Ireland and the UK still in active development. The challenges of maintaining best practice around mastitis are ever present as are the need for our national program to respond with clear, concise messages on mastitis control to an ever changing farming environment. Countdown has a strong legacy and a continuing role to play.
Mycoplasma: a re-emerging pathogen

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Mycoplasmas are the smallest known self-replicating prokaryote that lacks a cell wall. The lack of cell wall renders them resistant to antimicrobials that affect the cell wall or its synthesis. They are nutritionally fastidious organisms that grow slowly, generally requiring 3–7 days of growth before colonies become apparent. Growth is best at 37°C in an atmosphere of increased CO₂. Under a dissecting microscope, the colonies have a ‘fried egg’ appearance. Outside their hosts they can survive for long periods of time in cool moist environments surviving 8 months in recycled bedding. Mycoplasmas are susceptible to heat, alcohol, and commonly used teat dips (iodine, chlorhexidine and chlorine).

Disease manifestations
The most notorious species of mycoplasma in cattle is Mycoplasma mycoides subspecies mycoides small colony type, the causative organism for contagious bovine pleuropneumonia – a devastating disease that was introduced into Australia in 1858 and subsequently eradicated in 1967. There are more than 100 reported species of Mycoplasma. Twelve species have been associated with disease in cattle. Mycoplasma bovis is recognised as the most important cause of mycoplasma mastitis. Other Mycoplasma species reported to cause disease in cattle include: M. alkalescens, M. arginini, M. bovigenitalium, M. bovirhinis, M. californicum, M. canadense, M. dispar, M. serogroup 7, Mycoplasma F-38, Acholeplasma laidlawii, and Acholeplasma axanthum. The disease produced by each Mycoplasma species is similar but may vary in severity. M. californicum, M. alkalescens, M. bovigenitalium and M. canadense are reported to be the most common associated with mastitis.

Mycoplasma infections in cattle can range from subclinical to severely debilitating with some leading to death. Subclinically infected cattle carry organisms on mucosal surfaces including nasal, conjunctival, oral, intestinal and genital mucosa and may maintain the organism in the herd. With respect to dairy cattle mastitis, mycoplasmas are highly contagious and can be an economically important cause of milk loss and increased culling of infected cows.

M. bovis spreads to multiple organs and is capable of invading various kinds of host cell. The most common disease manifestations of mycoplasma infections include mastitis, pneumonia, and arthritis. Mycoplasma infections have also been associated with keratoconjunctivitis, otitis, meningitis, endometritis, salpingitis, oophoritis, seminovesiculitis, subcutaneous abscesses, infertility and abortion.

Arthritis associated with M. bovis infection may occur in calves and adults. In calves it is often associated with the respiratory form of the disease and in adults may be associated with mastitis. This condition often arises within 2–3 weeks of housing when cattle and are introduced into freestall facilities and may also follow transportation of calves over long distances.

Global distribution
M. bovis was first isolated in 1961 in the United States from a cow with severe mastitis. It has subsequently been isolated from cattle in many countries: Israel (1964), Spain (1967), France (1974), Britain (1975), Czechoslovakia (1975), Germany (1977), Denmark (1981), Switzerland (1983), Morocco (1988), South Korea (1989), Brazil (1989), Northern Ireland (1993), Republic of Ireland (1994) and Chile (2000). Early investigations of mycoplasma in Australia identified Mycoplasma spp. from seven different serogroups. Since then Mycoplasma alkalescens and Mycoplasma serogroup 7 have been associated with outbreaks of mastitis, arthritis, and abortion in dairy cattle. More recently, Mycoplasma bovis and Mycoplasma californicum have been isolated from disease outbreaks that have manifest, with mastitis and arthritis in adult cows and pneumonia, arthritis, and otitis media interna in young calves.

Transmission
The two primary risk factors for Mycoplasma mastitis in a herd are introduction of diseased animals to the lactating herd and problems with milking time procedures, particularly inadequacies in hygiene. Large expanding herds have been reported to have a greater risk for mycoplasma mastitis. The recent observation of mycoplasma emerging in predominantly larger herds in Australia fits with the experience reported overseas. This may in part reflect herd expansion and the introduction of cattle from multiple sources.

Transmission frequently occurs from animal to animal through direct contact and aerosolisation of respiratory secretions. Mechanical transmission is important in contagious...
mycoplasma mastitis. Contaminated milk can be a source of infection for calves. Risk factors for clinical disease include age, crowding, trauma, concurrent infections, and environmental and transportation stresses. Cow to cow spread of mycoplasma during milking is believed to be an important mode of disease transmission. Mycoplasma species are commonly found on nasal mucous membranes, respiratory and urogenital surfaces, and teat skin.26 A number of investigators have proposed that mycoplasma mastitis may come from mechanical transfer of respiratory and urogenital tract infections to the mammary gland.27,28 There is also evidence that internal transfer of mycoplasma from extra-mammary organ system sites to the mammary gland can occur. Following experimental intra-mammary inoculation of Mycoplasma sp. the organism was subsequently isolated from the blood and other organ systems.29,30 Multiple quarters have been observed to develop mastitis following the inoculation of a single quarter suggesting either haematogenous and or lymphatic transmission from one mammary quarter to the next. Studies investigating the relatedness of mycoplasma isolates from different body sites have reported mixed results. In one study, more than 90% homology in genetic fingerprints was observed in ante-mortem mycoplasma isolates from the nares, ear, eye and urogenital system as those isolated at post mortem from the mammary gland. Similarly, the mycoplasma species associated with an outbreak of pneumonia and arthritis identified the same subtype in mammary and extra-mammary sources. According to these observations it has been hypothesised that transmission between animals may occur via ingestion or aspiration of extra-mammary secretions and haematogenous transfer of mycoplasma from the respiratory system to the mammary gland or joints. However, one study where the genetic profile of mycoplasma mastitis isolates and those collected from nasal and vaginal secretions differed contradicts this theory.31

Ingestion is recognised as a mode of transmission in calves with infections observed following the feeding of contaminated milk to calves. Disease manifestations include pneumonia, otitis, conjunctivitis, and tenosynovitis. Vertical transmission has also been proposed according to the results of a study where infected calves were found to be descendents from infected cows.13 Aerosol spread of infection is another mode of transmission between calves with pneumonia and non-infected calves. The rapid spread of mycoplasma around the United States may have been facilitated by the use of contract calf-rearing facilities where calves from different farms are raised and then returned to their farm of origin. The mucosal surfaces of eyes, nasal cavities, ears and vestibular fossa are colonisation sites for Mycoplasma spp.1 Longitudinal studies suggest that calves may be asymptptomatically infected with mycoplasma and serve as a nidus of infection post parturition, it has subsequently been recommended that surveillance of heifers is incorporated into mycoplasma mastitis control programs.32

Intra-mammary infections acquired from environmental contamination associated with organisms shed from the urogenital tract has been proposed as a potential mode of transmission that could explain outbreaks of mycoplasma mastitis in periparturient heifers33 and dry cows.25,54

Experimental contamination studies suggest that the survival of Mycoplasma bovis in the environment is influenced by the temperature. At 4°C M. bovis survived on sponges for 57 days, in milk for 54, on straw for 20 and on wood and in water for 17 days. At 20°C, the survival period on these materials dropped to one to two weeks, and at 37°C to one week.13

Artificial insemination with infected semen is another potential route of infection.16 The male genital tract can become infected with M. bovis through contact with other animals or, possibly, via a heavily contaminated environment. Infection of the prepuce or urethra by M. bovis leads to an ascending infection of the testes causing orchitis, vesiculitis, decrease of semen quality and ultimately shedding in the semen.33 Mycoplasma contaminated frozen sperm can remain infectious for years and probably represents an overlooked infection source.2,36

The existence of multiple modes of transmission is consistent with the observation that good biosecurity and milking protocols to prevent mycoplasma transmission at milking has not proven to be as effective to control mycoplasma mastitis as it has for other contagious mastitis pathogens.37,38

Disease control and prevention

Implementation of milking time hygiene procedures coupled with identification and then segregation or culling of infected animals has been demonstrated to be an effective control strategy on some farms.26,35 It has been less effective on others supporting the contention that other means of transmission may be important.1 Attention to detail is important when working to control the spread of mycoplasma. Milking procedural strategies recommended to reduce the risk of disease transmission at milking include: wearing of disposable rubber gloves, milker hand cleansing between cows, use of post-milking teat asepsis, and milking unit backflush between cow milkings.26,23,40

A number of herds in Australia have been compromised by diagnostic failure which has resulted in delays to recognition of the disease. Cows with clinical mastitis caused by mycoplasma species often fail to respond to therapy. When the cause of the problem is not recognised producers tend to retreat the cows retaining them in a ‘hospital’ group providing opportunities for disease spread. With increasing numbers of infected cows the challenge to, and the rate of spread to, non-infected cows increases, sometimes exponentially. The hospital pen or group can become a site of disease transmission with cows entering the ‘hospital’ with environmental mastitis and subsequently becoming infected with mycoplasma. The importance of disease transmission in hospital pens was recently reported in a study of mycoplasma-infected dairies in North America.41

The reported duration of mycoplasma disease outbreaks range from less than 2 months to several years.24,32,42 The duration of the herd’s infection at the time of diagnosis will impact the proportion of animals infected and the prevalence of subclinical infections in the herd. Persistence will be favoured by a higher disease prevalence and higher incidence of subclinical infections. The persistence of the disease will also be influenced by numerous management decisions relating to segregation,
culling, milking procedures, nutrition, cow flow and calf management. On larger farms the effectiveness of management decisions are complicated by labour compliance or lack thereof. The need for attention to detail cannot be over emphasised.

Prevention and control of mycoplasma mastitis is facilitated by properly functioning milking equipment and consistent milking routines as outlined in the Countdown Downunder guidelines. Due to the propensity for the hospital pen to become an area of disease transmission it is important to instigate strict hygiene procedures with ‘hospital’ cows. Stripping cows in the hospital pen has the potential to contaminate milkers’ hands and equipment and should be performed judiciously and only when necessary to make treatment decisions. Disease transmission during the procedure of intra-mammary infusion should be considered a potential risk. There are reports of disease outbreaks in dry cows associated with contamination introduced during administration of dry cow therapy. During the initial stages of a disease outbreak when the identity of mycoplasma-infected cows is unknown and the number of infected cows may be high-disease transmission can be reduced by avoiding intra-mammary infusions, alternatively treating new cases of mastitis with systemic antimicrobials such as penethamate, erythromycin, tylosin or oxytetracycline. Following the identification and segregation/culling of mycoplasma-infected cows and establishment of good stable milking routines, normal treatment protocols with intra-mammary formulations can be re-implemented.

During the initial outbreak of mastitis the high risk area for mycoplasma tends to be the ‘hospital’ group. Affected cows tend to have mastitis in multiple quarters, little milk and respond poorly to treatment. According to the nature of the disease these cows are often eventually culled. The risk of disease transmission is significantly reduced if the infected cows can be identified and removed. If the cost of culling is not justifiable, then at least they should be segregated and milked separately. Mycoplasma infections tend to be chronic similar to Staphylococcus aureus. Some cows may appear to recover; however, these cows may remain sub-clinically infected and shed mycoplasma intermittently. It is subsequently difficult to establish if a cow has cleared the infection or if she is shedding intermittently. Generally, it is said, “Once a mycoplasma cow always a mycoplasma cow”.

Calf management practices have the potential to influence the persistence of mycoplasma in the herd. Feeding milk from mycoplasma-infected cows to calves provides an effective method of disease transmission. Disease manifestations in calves include pneumonia, tenosynovitis, otitis and death. Some calves may recover and remain asymptomatic carriers. The herd consequence of this is the persistence of the infection in the herd with the potential re-emergence of the disease when the heifers enter the milking herd. In endemically infected herds mycoplasma may be shed in colostrum so colostral management is important to minimise the risk of disease transmission. Pooling colostrum increases the risk of transmission as it increases the number of calves that could be infected by an infected cow. Methods to reduce the risk of mycoplasma transmission through milk to calves include pasteurisation of milk or the feeding of milk replacer. Colostrum can also be pasteurised to minimise mycoplasma transmission. Pasteurising mastitis milk at 65°C kills M. bovis and M. californicum after 2 min of exposure and M. canadense after 10 min. Exposure to 70°C inactivated M. bovis and M. californicum after 1 min and M. canadense after 3 min. Pasteurising colostrum is more difficult than milk as it has a tendency to coagulate if it is overheated. Installation of pasteurising equipment has other potential benefits such as mitigating risk of salmonellosis, Johnes’s disease, and pestivirus in calves. However, it should be appreciated that the cleaning and maintenance of the equipment requires attention to detail to maintain proper operation. Commercial pasteurisers for milk and colostrum are available in North America.

In endemically infected herds where heifers entering the milking herd may have been infected as young stock, disease may manifest shortly after calving. Clinical manifestations of disease in cows and heifers sub-clinically infected with mycoplasma are likely to manifest when the animals are under stress such as shortly after calving. The incidence of disease in these animals and the risk of disease transmission are exacerbated by problems with the transition period which may be precipitated by nutritional stress, overcrowding and/or poor cow comfort. Fresh cow disease may present as a mastitis and/or lameness problem. Sometimes the lameness problem is not recognised as an infectious disease problem. Affected animals develop diffuse swelling of joints in one or more limb. The swelling is not confined to the joint as the tendon sheaths are also affected. The condition is typically very painful and the animals are frequently culled.

There are currently no effective vaccines available to prevent the diseases caused by mycoplasma in cattle.

Diagnosis

There are a number of diagnostic modalities available to investigate mycoplasma infection status including; culture, serology and molecular detection techniques. There are also several sampling strategies described for identifying infected herds and infected animals within those herds. Sampling strategies include bulk tank surveillance, string or group sampling and individual cow sampling. The most appropriate sampling strategy will depend on the herd’s infection status and the herd’s clinical and subclinical mastitis history. When mycoplasma is first introduced into a herd the dairy manager tends to know that something is wrong even though they may not know what is causing the problem. Observations may include:

• Clinical mastitis that affects multiple quarters on the same cow.
• An increased incidence of mastitis treatment failure.
• Cows with mastitis may look like they have a lot of milk but give very little.
• Although non-specific, the milk from affected glands is often watery with flaky sediment.
• In some instances, the mycoplasma outbreak may be associated with cases of arthritis in adult cows.
Mycoplasma californicum and Mycoplasma bovis have been associated with outbreaks of disease in Australia. A recent study found that most of the outbreaks of mycoplasma mastitis in Australia have been associated with Mycoplasma bovis. Some herds have experienced dual infections involving Mycoplasma bovis and Mycoplasma alkalescens, which is likely to require ongoing monitoring. The following is a generic guide to diagnosing mycoplasma mastitis.

### Clinical Infections

- **Mycoplasma bovis**
- **Mycoplasma alkalescens**

#### Clinical Infections

Clinical infections are usually associated with prolific shedding of the organism (>10^6 mycoplasma per mL). Subclinical infections are associated with more variable shedding with counts: >10^6 approximately 60% of the time, lower numbers approximately 10% and non-detectable shedding approximately 30% of the time. The logistics and costs associated with collecting and submitting fresh milk samples for culture each day are significant. Collecting and freezing milk samples from cows with clinical mastitis for batch submission provides a practical compromise however it should be appreciated that the process of freezing and thawing samples may not have a great impact on clinical samples due to the large numbers shed during clinical disease however it will reduce the sensitivity of culture techniques for detecting subclinical infections where the number of organisms shed may be low. Molecular based assays such as PCR do not require the organism to be viable for detection hence their sensitivity will not be reduced by sample freezing. Regardless of the method of detection utilised approximately 30% of the composite milk samples collected from cows with subclinical infections may not contain mycoplasma yielding a false negative result on the basis of a single sample.

### Herd Monitoring

Herd monitoring of mycoplasma mastitis is often performed through bulk tank cultures or PCR. Isolation of mycoplasma species in herd bulk tank milk is likely to indicate that there is at least one cow in the herd affected with mycoplasma mastitis as environmental contamination of the bulk tank is unlikely. There are limitations to bulk tank sampling that should be considered when interpreting diagnostic results. If a single cow in a hypothetical herd was shedding mycoplasma in its milk, then the number of organism in a bulk tank milk sample (i.e. number of mycoplasma per mL of bulk tank milk) could be calculated by multiplying the number of organisms shed by the infected cow by the milk production for that cow and dividing by the number of days as mycoplasmas grow very slowly. The logic of this latter sampling strategy is that cows with subclinical mycoplasma infections are likely to have an elevated somatic cell count.

Cows infected with mycoplasma should be culled or segregated from the milking herd and milked last. The logistics of maintaining a segregated herd is difficult and carries with it disease transmission risks.

- Following culling/segregation of infected cows the “clean” herd can be monitored using bulk tank sampling and PCR if it is a Mycoplasma bovis infected herd or culture for other species of mycoplasma.
- Milk should be collected for culture from all new cases of clinical mastitis to monitor for re-emergence of disease.
- Bulk tank monitoring is continued on a monthly basis. There are a number of characteristics of mycoplasma infections that are useful to understand when designing sampling and interpreting diagnostic investigations.

#### Herd Monitoring of Mycoplasma Mastitis

- The initial objective of the diagnostic investigation is to define the problem and to identify which cows are infected. If the problem has recently developed it is likely that most of the infected cows will be in the ‘hospital’ group. Longer standing herd infections may result in a mix of clinical and sub-clinical infections with infected cows distributed to the milking herd.
- The costs associated with diagnosing mycoplasma infected herds and cows within these herds can be significant and is likely to require ongoing monitoring. The following is a generic recommendation that may be adjusted to fit with particular herd circumstances.
- Culture all clinical cases of mastitis.
- If the problem is associated with Mycoplasma bovis, collect bulk tank milk samples daily for 3 days and test for Mycoplasma using the PCR assay. If the herd is milking more than 350 cows it would be best to collect more than one sample of bulk tank milk each day i.e. one sample per 350 cows each day and if possible have the milk diverted to different vats so there is less dilution of the latter sample/s. The objective of the tank sampling is to determine if there is evidence of cows with subclinical infections in the herd. If the initial sampling yields a positive result it is not necessary to continue with the 3 days of sampling.
- If the bulk tank samples are positive for mycoplasma there are two options. One is to culture all cows – this option is expensive and not usually necessary. Alternatively, cultures can be performed on milk samples collected from all cows with a somatic cell count greater than 200,000 cells/mL.
for about 9% of the time. Consequently, about 39% of the time, mycoplasma infection would not be detected by testing bulk tank milk samples from a dairy herd with a single infected cow.\(^4\)

The take-home message is that clinical infections will be easier to detect than subclinical infections. Sub-clinically infected cows do not shed the organism all the time so repeat monitoring samples of bulk tank milk are indicated to verify the non-infected status of the milking herd following segregation/culling of infected cows.

In endemically infected herds surveillance of recently calved heifers should also be considered if the heifers are likely to have been exposed to mycoplasma as calves. The re-emergence of mycoplasma in a herd may coincide with exposed heifers calving. We have recently observed this situation on a farm where clinical disease in the lactating herd had ‘resolved’. Stress appears to play a role in these calving-associated disease outbreaks as they appear more common when other aspects of the transition period are compromised.

Serology has been used for the diagnosis of Mycoplasma bovis infection. The risk of a seropositive test has been observed to increase with advancing age and increasing farm size.\(^4\) Care must be taken when interpreting mycoplasma serology given that detection of antibodies in an animal implies exposure, not disease. Mycoplasma bovis exposure may occur by different routes including respiratory, genital, and mammary.\(^1\)

### Treatment

In regard to antibiotics, Mycoplasma species are known to be susceptible to a number of antibiotics \textit{in vitro} such as tylosin, tulathromycin, tilimcosin, spectinomycin, lincomycin, oxytetracycline, and enrofloxacin. Unfortunately, the efficacy of antibiotic drugs for mycoplasma mastitis has generally been poor and antimicrobial therapy is not considered effective.\(^13\)

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Unravelling the mysteries of liner compression

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What is liner compression?
Liner compression (LC) is the mean compressive pressure (expressed in kPa above atmospheric pressure) applied to the inner tissues of the teat apex by the liner during the d-phase of pulsation. One component of LC has been defined as Over-Pressure (OP) by Mein et al. (2003) as the mean compressive pressure, above that required to just start or stop milk flow from the teat, which is applied to the inner tissues of the teat apex by the liner during the d-phase (Figure 1). Note that a new method (Gomez et al. 2010) has been used to measure the OP values reported in this paper. Our new method (OP2) is a ‘dynamic’ method in which pulsation continues throughout the test while the pulsation chamber vacuum is increased in steps of 2 kPa until milk flow is observed. The previous method (OP1) reported by Mein et al. (2003) stops pulsation by removing the short pulse tube from one teatcup. Vacuum is then slowly increased until milk flow is observed. Essentially, OP1 provides a very long d phase of pulsation followed by a slow removal of LC as PCV is increased. OP2 provides continuous pulsation with stepped decreases in LC. The OP2 method produces values about 66% of the OP1 method. This must be taken into account when referring to previous recommendations for OP made with the OP1 method.

Another method to estimate the relative LC of different liners, or individual liners used at different milking vacuum levels, is the Residual Vacuum available for Massage (RVM). This value is obtained by subtracting the vacuum required to collapse the liner (i.e. the liner ‘Touch Point’) from the average claw vacuum (Figure 1). For a given claw vacuum, therefore, RVM is assumed to decline in direct proportion to any increase in the vacuum required to collapse the liner. Touch Point, measured without a teat in the liner, is usually defined as the pressure difference required to collapse the liner to the point where the opposing walls of the liner barrel first touch each other. This measurement was developed for round liners, which exhibit ‘buckling’ behaviour when they collapse. The measurement is easy to make and quite repeatable for round liners. It cannot be applied to triangular and square liners, as the opposing walls of these liner shapes never touch. The ‘touch point’ of the adjacent walls of triangular or square liners is a much more subjective, and therefore less repeatable, measurement and cannot be used as a comparable measurement to the touch point in round liners. We have found that while OP and RVM are correlated, there are substantial differences between these two estimates of LC, especially for certain liner types.

Figure 1: Relationships between Liner Vacuum (LV), Touch Point (TP), Residual Vacuum available for Massage (RVM), Pulsation Chamber Vacuum (PCV), PCV when milk starts flowing from the teat (SMF), Liner Compression (LC) and Over-Pressure (OP). From Mein and Reinemann (2009).
There have been numerous attempts to develop sensors to measure LC and/or some of its components. The measurements resulting from artificial teat sensors are highly dependent on the measurement technology used. These artificial teat sensors have provided valuable information on the relative components of LC as influenced by:
- material properties of the liner
- liner tension
- pressure difference across the liner during the d phase of pulsation
- liner shape

A summary of these terms and the influence of liner properties on LC effects are presented by Mein and Reinemann (2009).

Sensors have also been developed to measure local pressure at the teat-liner interface. LC is NOT the same as point pressures measured in this way. These sensors typically respond to forces that can be transmitted to the inner teat tissue as well as the shear force developed at the teat/liner interface and these two components are usually not differentiated. Point measurements made with a flexible sensor (usually using capacitance devices designed to measure normal forces) can be more than 10 times higher than sensors designed to measure LC and estimates made using OP. This is likely because these flexible sensors are designed and calibrated to measure normal force while signal generated by shear force is greater than the signal generated by normal force.

Teat length, diameter and shape affect LC. For example, short teats may experience a lower LC because the liner cannot apply much compression when it does not have to bend so far to collapse beneath the teat. Very short teats may not even penetrate the liner into its zone of collapse and therefore receive no LC during milking. Wide flat-bottomed teats may experience a relatively high LC because the liner has to bend further and also because the free surface area beneath the teat is increased.

Why is it interesting and/or important?

One purpose of pulsation and LC is to relieve congestion in teat tissues during milking. Vacuum is applied to the teat end during the entire pulsation cycle and also applied to the teat barrel depending on how well the teat barrel seals in the liner barrel. If the teat barrel does not fill the liner cross section, more vacuum will be developed in the liner mouthpiece and expose more of the teat to vacuum levels approaching the claw vacuum. Vacuum applied to teat tissues is the driving force behind teat tissue congestion. As the vacuum level increases and as more of the teat surface is exposed to vacuum, the faster and more severe will be the resulting tissue congestion. The teat-end experiences no LC during the b-phase and the longer the liner is left open, the more teat-end congestion will occur.

LC is applied only to the teat end; the lower 10 to 25 mm, depending on the liner type and teat shape. The teat walls surrounding the sinus receive little or no LC. LC is effective, therefore, at relieving teat end congestion, but not congestion in the teat wall or at the base of the teat. Effective reduction of teat-end congestion results in dramatic increases in the peak milk flow from a teat during the b phase of pulsation and increasing LC generally results in higher peak and average milk flow rates.

Another result of LC is that the skin at the end of the teat is compressed and stretched during the d phase of pulsation. LC is the most important machine effect on the development of teat-end hyperkeratosis or roughened teat-ends.

Choosing a liner and the appropriate vacuum level and pulsation settings is an exercise in balancing the fundamental goals of milking quickly and gently. Too little liner compression results in teat-end congestion during milking and slower milking. Too much liner compression results in excessive teat-end hyperkeratosis and provides no additional benefit in relieving teat-end congestion. Just-right liner compression relieves the congestion at the teat-end and results in minimal teat-end hyperkeratosis, reasonable milking speed and improved cow comfort during milking.

Practical applications of this knowledge

The 2007 revision of International Standard ISO: 5707, Milking Machine Installations – Construction and Performance, states “The user’s manual shall include … sufficient data to be able to choose the liner for a herd,” and “the desired average liner vacuum and/or the desired average liner vacuum during phase b and phase d of the pulsation chamber vacuum records.”

Recent research at the University of Wisconsin Milking Research and Instruction Lab has been conducted to investigate the interaction between liner properties (liner compression, dimensions, materials and shapes) and milking machine settings (vacuum level and pulsation phase durations). The methods developed to predict liner performance can be used by milking machine manufacturers and field advisors to provide guidance to users to choose liners and milking machine settings to balance the goals of milking quickly, gently and completely.

Liner dimensions

The most important aspects of fitting the liner to the cow are the physical dimensions of the liner compared to the physical dimensions of the teat. The most important liner dimensions for liner fit specified in ISO 3918 are:
- Mouthpiece and mid barrel liner diameter
  - mouthpiece and mid barrel diameters range from just under 20 mm to more than 30 mm for commercial liners.
- Mouthpiece Chamber (MPC) Depth, or the distance from the top of the liner to the highest point that the liner is able to fully collapse
  - MPC depth for commercial liners ranges from about 20 mm to more than 45 mm.

The teat is remarkably adaptable in its ability to conform to a liner. Most liners have little or no ability to adapt to different teat sizes. Since we know that there will always be a range of teat sizes and shapes in any herd, the best liner is the liner that will perform well over the widest range of teat sizes.
In order for the liner to apply compression to the end of the teat, the teat-end must be positioned in the part of the liner that is able to collapse and provide this compression. Teats stretch about 40% from their resting length to their length when situated in a narrow bore liner during milking. If we want to apply compression to the lower 25 mm of the teat, a liner with a mouthpiece depth (MPD) of 30 mm will apply full compression to teats that are longer than about 39 mm (minimum teat length = (MPD (mm) + 25 mm)/1.4).

There has been a general trend toward breeding for short teats and first lactation cows have the shortest teats in any herd. American herds typically have about 25% of teats shorter than 39 mm. A liner with mouthpiece depth of 30 mm will thus result in excessive teat end congestion in about 25% of cows in the typical American herd. In addition to the problems generated by insufficient LC, these short teats will also be milked with high mouthpiece vacuum, as the teat is not long enough to create a seal in the liner barrel. The result of this will be ‘rings’ around the base of the teat and congestion and edema of the teat skin.

The relative diameter of the teat compared with the liner barrel also plays a role in the MPC vacuum during milking. Teats can stretch in both directions: they can get ‘fatter’ and ‘longer’. The total volume of the teat in the liner is relatively constant so if teats get ‘fatter’ they will not elongate as much. Wide bore liners (liner bore diameter greater than teat diameters) will cause a teat to get fatter and reduce the ability of the teat to elongate into the zone of effective compression. This increases the minimum teat length that can be effectively massaged during milking.

**Mouthpiece chamber vacuum**

The vacuum developed in the MPC plays a major role in the development of congestion in the teat wall and formation of rings at the base of the teat. Teat wall congestion and teat ringing can also have a significant influence on the ease of unit removal. Our research on liners has indicated that liners with low MPC vacuum reduce the occurrence of ringing and blue teats after milking. In addition these liners also result in conditions that make unit removal much easier. If teat barrels in the MPC region become congested, the “rings” at the base of the teat act to hold the liner on the teat even after vacuum is removed from the claw.

**Liner compression and teat-end hyperkeratosis**

The primary milking machine influence on teat-end hyperkeratosis is LC (environmental conditions and teat size and shape also have a large influence but are not ‘adjustable’ in commercial herds). LC for any individual liner will also increase with the milking vacuum level at which it is used because the pressure difference across the liner is increased during the d phase of pulsation. The most reliable relative indicator of LC for liners milking real cows is the OP as defined above. OP can be measured in the field without specialised sensors and is a more biologically relevant indicator of LC than RVM. OP measurements on a range of commercial liners are presented in Figure 2. The greatest OP values are more than 6 times the lowest OP values over this range of commercial liners. These OP values are highly correlated with teat end hyperkeratosis scores in field studies. In a survey of commercial farms in Wisconsin, herds milked with liners having the highest OP measurements had more than 80% of teats that were roughened and

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**Figure 2: Overpressure in 16 different liners using method OP2.**
cracked while herds milked with liners having the lowest OP measurements had less than 20% of teats that were roughened and cracked. Lower OP results in less hyperkeratosis or teat-end roughness.

**Vacuum level and pulsation settings**

Vacuum level and pulsation settings must be chosen for each liner taking into account the milking technology and management on each individual farm. We have developed methods to predict the effect of milking vacuum level and pulsation settings on milking speed and teat-end congestion for a specific liner. An example of the results of one of these liner performance maps is shown in Figure 3 (a 20 mm bore triangular liner fitted with a vent in the mouthpiece, OP of 5 kPa and mouthpiece depth of 27 mm). This information has been previously unavailable to milking managers and we hope that these methods will be used to take some of the mystery out of milking.

The liner performance map illustrated in Figure 3 is for one specific liner and the specific milk speed and congestion values do not apply to other liners. Liners with different shapes, materials, OP values and differing relationship between OP and claw vacuum will produce different results. There are some general trends, however, that illustrate some basic principles that likely apply to all liners.

The percentage numbers in the body of the chart relate to the relative milking speed, as indicated by average milk flow rates.

- As claw vacuum increases, so does the milking speed.
- As the b phase (milk:rest ratio) increases, so does milking speed until some critical point at each vacuum level at which point milking speed declines with increasing b phase duration due to increasing teat-end congestion.

The effects of these two machine settings are interactive: e.g. there are a number of combinations of claw vacuum and b phase duration to achieve a relative milking speed of 90% of the maximum for this liner. Which is best?

The shades in the body of the chart indicate the degree of teat-end congestion for teats that are longer than about 37 mm, or enough to penetrate into the zone of effective compression for this liner.

- As claw vacuum increases, so does teat-end congestion.
- As the b phase (milk:rest ratio) increases, so does teat-end congestion.

The risk of teat tissue congestion for teats shorter than the minimum length defined by mouthpiece depth is indicated by the colours in the kPa column because these short teats will not receive the full benefit of LC, as described above and congestion is influenced primarily by claw vacuum level.

As an illustration of the use of the specific liner performance map illustrated in figure 3, let us consider a milking parlour with operating vacuum of 48 kPa and claw vacuum of 42 kPa during the peak flow period of milking. Because claw vacuum approaches system vacuum as milk flow rates decline at the end of milking, the expected claw vacuum in the low flow condition would be about 47 kPa. It is useful to measure claw vacuum during both the peak flow and low flow (just before unit removal) periods on a farm in order to assess congestion risk during all phases of milking. In this example we expect a range of 5 kPa in claw vacuum over the range of expected claw vacuum conditions.

The milking speed numbers should be interpreted using the expected claw vacuum during the peak flow period of milking (42 kPa in this example), as the peak flow period makes up the largest portion of the total milking, especially if automatic

<table>
<thead>
<tr>
<th>Claw Vacuum (kPa)</th>
<th>b-phase (ms)</th>
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<tr>
<td>Hg 300 350 400 450 500 550 600</td>
<td></td>
</tr>
<tr>
<td>34 10.0</td>
<td>65% 70% 74% 77% 80% 81% 81%</td>
</tr>
<tr>
<td>36 10.5</td>
<td>68% 73% 77% 80% 81% 82% 82%</td>
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<tr>
<td>37 11.0</td>
<td>71% 75% 79% 82% 84% 84% 84%</td>
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<tr>
<td>39 11.5</td>
<td>74% 78% 82% 84% 86% 86% 86%</td>
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<td>77% 82% 85% 87% 88% 88% 88%</td>
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<td>42 12.5</td>
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<td>85% 89% 91% 93% 94% 93% 92%</td>
</tr>
<tr>
<td>46 13.5</td>
<td>89% 93% 95% 96% 97% 96% 94%</td>
</tr>
<tr>
<td>47 14.0</td>
<td>93% 97% 99% 100% 100% 99% 97%</td>
</tr>
</tbody>
</table>

### Congestion for short teats (<3 cm) indicated by colors in the kPa Column

<table>
<thead>
<tr>
<th>Table colors indicate teat congestion for teats &gt;3 cm in length</th>
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<td>Low</td>
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Figure 3: Milking speed and teat end congestion performance for one specific triangular liner with OP of 5 kPa. NOTE: Does not apply to other liners!
cluster removers are used. The fastest milking condition (91% of the fastest possible condition for this liner) would occur with a b phase setting of 500-550 ms. However, the risk of teat-end congestion would be relatively high during the peak flow period (bordering between yellow and red). During the low flow period of milking (claw vacuum of 47 kPa) this pulsation setting the risk of teat-end congestion borders between high and extreme.

If gentleness were the main priority we might choose a system vacuum level of 42 kPa (range of claw vacuum from 41 kPa during the low flow period to 36 kPa during the peak flow period) and b phase duration of 450 ms. These machine settings for this liner would result in very low teat congestion during the peak flow period and only moderate teat congestion during the low flow period for most cows and would substantially reduce teat-end and teat barrel congestion for cows with short teats. The milking speed for these settings would be 80% of the maximum for this liner, or a reduction of 11% compared with the previous settings. If the average cups-on time were 4 minutes, the change in the average cups-on time would be an increase of about 26 seconds. The lower vacuum setting would also likely result in more complete milking.

We can also estimate the relative HK risk with this liner in these 2 milking conditions. This (triangular) liner has an OP value of 5 kPa and will therefore produce less teat-end HK than a liner with an OP value of 10 kPa. Liners that are designed to optimise peak milk flow rates tend to have OP values of 10 kPa or more. All of the OP measurements presented here were done using a claw vacuum level of 44 kPa. We are still working on predicting how OP changes with the pressure difference across different liner types but can make a rough estimate that OP changes by about 1/4 of the change in claw vacuum. The OP during the low flow period in our fast milking example would be about 6 kPa and about 3 kPa in the gentle milking example. We expect less HK in the gentle milking scenario.

**Summary**

- Choosing a liner and the appropriate vacuum level and pulsation settings is an exercise in balancing the fundamental goals of milking quickly, gently and completely.
- Liner dimensions play a critical role in determining the range of teat sizes that can be effectively massaged. The most important of these dimensions is the depth of the mouthpiece followed by the liner bore.
- Overpressure (OP) is the most practical and biologically relevant measure of the relative compression applied to teats by a liner.
- Increased OP (or LC) will result in increased teat-end hyperkeratosis and will also increase milking speed. The operator must make the final decision on the relative importance of these two goals.
- OP and LC increase with milking vacuum level for each individual liner. The performance maps we are developing for liners indicate the practical effects of milking speed and congestion risk over a range of vacuum and pulsation settings as LC increases with claw vacuum.
- Liners and pulsation settings that maximise milking speed (especially peak flow rates) will also increase the risk of teat congestion and teat-end HK.
- Maximizing the gentleness of milking will generally result in a modest reduction (about 10%) in milking speed (cups-on time). This modest increase in cups-on time will have a smaller effect on the number of cows milked per hour, especially if automatic detachers are used and a maximum milking time per cow is applied.
- We are continuing this work and hope to publish performance maps for a range of liners with differing shape, material and OP in the near future.

**Acknowledgements**

Research on the new methods referred to in this paper was funded by Avon Dairy Solutions.

**References**


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<td>Peter Mansell</td>
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<td>Peter Mansell</td>
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<td>Graeme Mein</td>
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<td>Graeme Mein</td>
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<td>Pauline Brightling</td>
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<td>Pauline Brightling</td>
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<td><strong>Doug Reinemann (US)</strong> Unravelling the mysteries of liner compression</td>
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<td>Q&amp;A session (all speakers in a panel discussion)</td>
<td>Peter Mansell</td>
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<td>Close and thank you (Graeme Mein)</td>
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