Discover the most recent developments on the mastitis research scene in Canada

The CBMRN Gives You the Scoop on its Most Recent Developments in Mastitis Research

Midway through the program of the Canadian Bovine Mastitis Research Network (CBMRN), our research has already begun yielding interesting results. Some projects will pave the way to new technologies in the long term while others generate useful knowledge right away. Such advances coupled with our efforts to transfer new tools and new knowledge will hopefully lead to changes in udder health management practices at the farm over the coming years.

This paper is a sequel to the brochure "Cultivating Knowledge to Maintain Milk Quality" that was published in 2008. Inspired by the success of that first publication, this year, we have opted to move to a document offering as much information in a new reader-friendly newspaper-style format.

While you are quietly enjoying reading your copy of "What's New in the World of Mastitis Research 2008-2009", CBMRN research scientists are hard at work developing strategies and technologies intended to support your work at the farm. We will therefore have a lot more to report in an upcoming issue of this newspaper. Don't miss it!

In the meantime, we hope that you will enjoy this issue!

The CBMRN Team

The CBMRN would like to thank all Provincial Dairy Organisations for the distribution of this newspaper to all Canadian dairy producers.

CBMRN figures:
- 13 financial partners
- 3 donors and sponsors
- 35 members and collaborators
- 7 post-doctoral researchers
- 23 graduate students
- 8 undergraduate students
- 19 technicians
Canada’s Producers and Cows Contribute to Mastitis Research

A genuine gold mine of samples and epidemiological data has recently been made available to experts to paint an accurate picture of the mastitis situation in Canada and find solutions for this major problem.

Through the efforts of a multidisciplinary team covering four laboratories and four collection regional centres in Canada (Figure 1), the CBMRN now boasts a central database that is unique in the world. This database is an invaluable resource for researchers in their quest to find solutions to the mastitis problem on dairy farms.

To build this database, the CBMRN team relied on a large-scale infrastructure anchored by the National Cohort of Dairy Farms. In addition to the staff involved, the success of this undertaking largely depends on the efforts of participating producers. They had to agree to regular visits from technicians, provide milk samples and submit to in-depth interviews designed to gather detailed information about their management practices.

For example, questions were designed to find out about animal housing, bedding material, cleaning frequency and milking procedures. Even the cows had to submit to a close examination of a CBMRN technician to check the condition and cleanliness of their teats, legs and flanks. All this information will be associated with infections diagnosed in each farm and the bacteria isolated in the milk samples provided.

As a result of a cooperation agreement with Valacta and CanWest DHI, production data from each cow enrolled in the Cohort – production, age, days in milk, number of lactations, somatic cell counts, etc. – are also made available to CBMRN researchers and students. All the data regarding the health of the cows, including the medication used, are also recorded for a study on antibiotic resistance.

Figure 1. Distribution of the Core Research Platform activities and CBMRN research activities in Canada

LEGEND

1. Pacific Agri-Food Research Centre - Agassiz
2. University of Calgary
3. University of Saskatchewan
4. Vaccine and Infectious Disease Organization (VIDO)
5. University of Guelph (Ontario Veterinary College)
6. Public Health Agency of Canada
7. University of Guelph (Kemptville Campus)
8. Université de Montréal (Faculté de médecine vétérinaire)
9. McGill University
10. Université de Sherbrooke
11. Université de Montréal (Faculté de médecine vétérinaire)
12. Dairy and Swine Research and Development Centre - Sherbrooke
13. University of Prince Edward Island (Atlantic Veterinary College)

A few figures about the Cohort:

- **91** farms that are representative of the Canadian dairy industry
- **9 000** cows, mostly Holstein
- **134 000** milk samples submitted to bacterial culture
- **17 000** bacterial isolates stored in an ultra-low temperature freezer at the Faculté de médecine vétérinaire of Université de Montréal in Saint-Hyacinthe
- **700** hair samples to create a DNA repository for genetic research

In the spring of 2009, to thank them for their contribution and reward their efforts, six training sessions on knowledge developed by the CBMRN were offered to the producers who participated in the Cohort. Thanks to the samples taken on their farm over a period of two years, every producer was able to get a mastitis profile of his farm through a customized herd report. And since results were presented per farm, province and the entire country, participants were able to compare the situation of their farm with other provinces and Canadian producers.

Training for Cohort producers

In the spring of 2009, to thank them for their contribution and reward their efforts, six training sessions on knowledge developed by the CBMRN were offered to the producers who participated in the Cohort. Thanks to the samples taken on their farm over a period of two years, every producer was able to get a mastitis profile of his farm through a customized herd report. And since results were presented per farm, province and the entire country, participants were able to compare the situation of their farm with other provinces and Canadian producers.
Current Situation of Mastitis in Canada

The analysis of samples from the CBMRN’s National Cohort of Dairy Farms enables us to paint a picture of the present situation of mastitis throughout Canada. It is the most accurate and the most detailed exercise of the kind ever conducted in the country. We no longer need to make extrapolations based on research data from other countries. We now know exactly what is going on in our own backyard.

Clinical mastitis
Typically, at each milking, at least one of every five cows will suffer from clinical mastitis. In other words, it would mean that for a herd of 100 cows, 20 would show visible symptoms of mastitis during the course of a year. Based on data from the CBMRN’s National Cohort of Dairy Farms, the rate of clinical mastitis is precisely 28% in Quebec, 22% in Alberta and 36% in the Maritime and in Ontario.

Our data also show that the rate of clinical mastitis is generally higher in tie stall farms than in free stall farms. Variations in clinical mastitis from one herd to the next are much greater than variations between provinces. For example, Cohort

Bacteria causing clinical mastitis
The same bacteria flourish everywhere in Canada, but not in the same proportions. All those bacteria can be controlled with various management practices.

When milk samples from cows presenting mastitis symptoms are analyzed, the most common pathogen found in Quebec farms is S. aureus with 17.8% of cases. This percentage of S. aureus is the highest in Canada, followed by Ontario (15.4%) and the Maritime (13.4%) (Table 1).

S. aureus is contagious, which means that it can be transmitted from one cow to the next. It is a stubborn enemy which has proven quite troublesome for Quebec producers. In Alberta, producers seem to have found effective ways to keep it at bay. In fact, S. aureus is not the bacterium which causes the most problems to Albertan producers. It is found in only 5.9% of clinical mastitis samples.

It is rather environmental bacteria that cause the most problems to Albertan producers. In the Maritime and Ontario, the main culprits are E. coli and environmental streptococci. In Quebec, the Maritime and Ontario, although they are less prevalent than S. aureus, these same bacteria wreak considerable havoc. Strep. uberis, Strep. dysgalactiae (a bacteria considered to be both environmental and contagious) and Staph. aureus are each found in more or less 8% of clinical mastitis cases.

That being said, it should be pointed out that E. coli is the strain most often found in mastitis cases presenting severe symptoms (31% of the cases of severe clinical mastitis). In addition, it is generally recognized that E. coli is probably involved in a large portion of clinical mastitis cases where no pathogen was identified. “No significant growth” since those bacteria are often rapidly eliminated by the cow’s immune system.

Non clinical infections (without visible symptoms)
Even in the absence of visible signs of mastitis, intramammary infections cause an increase in SCC as well as important production losses, mainly because of damages caused to mammary secretory cells.

Based on CBMRN’s Cohort farms, it is estimated that almost one out of every five (18%) quarters is infected with mastitis pathogens. However, less than one-half of one per cent of these quarters shows visible symptoms: such as abnormal milk (clots, flakes, watery milk), swelling or fever.

Coagulase-negative staphylococci (CNS) are most often involved in subclinical intramammary infections, followed by a close cousin, S. aureus, particularly on Quebec farms. The pattern is more or less the same at the national level, except in Alberta where CNS are followed by Corynebacterium spp. in similar proportions (Figure 2). We have also determined the proportion of new infections occurring during the dry period. To do so, we simply analyzed the milk samples taken immediately after calving in cows that were free of infection at the beginning of the dry period.

Across Canada, the average rate of new intramammary infections is 17% of quarters (Figure 3). To put it plainly, for every five (18%) quarters is infected with mastitis pathogens, one in three (6%) will be free of new infections.

Infections in dry cows
Our analyses have enabled us to assess the proportion of cows that have an intramammary infection at the beginning and at the end of the dry period. Coagulase-negative staphylococci are undoubtedly the most prevalent bacteria during the dry period, followed by S. aureus and Corynebacterium spp. in similar proportions (Figure 2). We have also determined the proportion of new infections occurring during the dry period. To do so, we simply analyzed the milk samples taken immediately after calving in cows that were free of infection at the beginning of the dry period.

Across Canada, the average rate of new intramammary infections is 17% of quarters (Figure 3). To put it plainly, for every five (18%) quarters is infected with mastitis pathogens, one in three (6%) will be free of new infections.

Table 1. Distribution of pathogens by region (%)

<table>
<thead>
<tr>
<th>Pathogen</th>
<th>Quebec</th>
<th>Ontario</th>
<th>Maritime</th>
<th>Alberta</th>
</tr>
</thead>
<tbody>
<tr>
<td>S. aureus</td>
<td>17.8</td>
<td>15.4</td>
<td>13.4</td>
<td>5.9</td>
</tr>
<tr>
<td>E. coli</td>
<td>7.5</td>
<td>11.6</td>
<td>8.9</td>
<td>16.0</td>
</tr>
<tr>
<td>Strep. spp.</td>
<td>8.3</td>
<td>9.6</td>
<td>5.0</td>
<td>8.2</td>
</tr>
<tr>
<td>Strep. uberis</td>
<td>7.5</td>
<td>7.8</td>
<td>9.2</td>
<td>3.8</td>
</tr>
<tr>
<td>Strep. dysgalactiae</td>
<td>7.9</td>
<td>3.2</td>
<td>5.9</td>
<td>3.0</td>
</tr>
<tr>
<td>CNS</td>
<td>2.3</td>
<td>6.5</td>
<td>1.3</td>
<td>3.1</td>
</tr>
<tr>
<td>Klebsiella</td>
<td>1.3</td>
<td>0.9</td>
<td>2.9</td>
<td>8.0</td>
</tr>
<tr>
<td>Others</td>
<td>6.5</td>
<td>6.5</td>
<td>4.3</td>
<td>7.4</td>
</tr>
<tr>
<td>Contaminated</td>
<td>11.6</td>
<td>9.7</td>
<td>8.5</td>
<td>10.7</td>
</tr>
<tr>
<td>No growth</td>
<td>29.4</td>
<td>36.7</td>
<td>40.6</td>
<td>35.9</td>
</tr>
</tbody>
</table>

Data show a spread of 3 to 110 cases per 100 cows per year among farms. The best and the worst 25% of these herds vary from approximately 15 to 39 cases per 100 cows per year.

Of course, part of this variation can be explained by differences among producers in interpreting mastitis cases and in record keeping. For example, one producer may feel that the mere presence of clots in the milk is not a sign of clinical mastitis, while another producer may feel it is.

However, the most credible hypothesis to explain such large variations among herds probably has to do with the implementation of udder health management practices instead.
100 healthy quarters at drying-off (or say 25 cows), we can expect that 17 of them will present a new infection at calving. Rates vary among regions in Canada. Western Canada herds reflect the national average while Ontario herds are slightly above it. In Quebec and the Maritime, dry cows seem to be a little more protected against new infections during the dry period since the rates observed are below the national average. As for cows already infected before drying-off, it is estimated that on average, 74% of infected quarters will heal prior to calving across the country (Figure 4). This recovery rate is attributed to the widespread use of dry treatments.

Infections during lactation

During lactation, CNS and Corynebacteria show up again causing the most new infections (Figure 5). S. aureus behaves a bit differently from these bugs and sticks around for a long time in the udder, causing the chronic infections we all dislike. So even though cows are not getting that many new infections from S. aureus, its long duration makes it look more prevalent across farms. It is also interesting to note that all these most prevalent pathogens are bugs that inhabit the skin of the teat, and are therefore close-by and ready to cause infection. They are also, with the exception of S. aureus, the minor pathogens that don’t usually cause clinical mastitis or severe increases in SCC.

Current Situation of Mastitis in Canada (continued)

Figure 3. New intramammary infection rates per region during the dry period

- West = 17% quarters/period
- Ontario = 23% quarters/period
- Quebec = 15% quarters/period
- Maritime = 12% quarters/period

National average = 17% quarters will be infected during the dry period

Figure 4. Dry period intramammary infection recovery rate per region

- West = 74% quarters/period
- Ontario = 71% quarters/period
- Quebec = 74% quarters/period
- Maritime = 77% quarters/period

National average = 74% of infected quarters heal during the dry period

Figure 5. Prevalence of infections per pathogen during lactation in Canada

- Contaminated 12%
- CNS 5%
- Other 9%
- S. aureus 2%
- S. coagulase-negative 2%
- S. pyogenes 2%
- No significant growth 76%

Highlights of the current mastitis situation in Canada

So, what do we take home with us about the mastitis situation in Canada? Well, the over 134,000 milk samples we collected during the Cohort give us a great picture of what’s going on with mastitis here — and having a grip on what we know is going on is a good place to start. We can now estimate the causes of clinical mastitis regionally and begin to investigate and formulate reasons for the differences we’ve seen. One surprising discovery is that although we expected only a few infections in the clinically normal lactating cows and pre-dry/post-fresh cows, we found quite high incidences of some infections, predominately with minor pathogens such as coagulase-negative Staphylococci and Corynebacterium spp.

We see S. aureus appearing in the top three pathogens in all stages of lactation, and clinical as well as non-clinical mastitis — there is still a lot of room to work on our control of this bug! If anything, farms in the West seem to be controlling it best — knowledge of specific management factors related to controlling mastitis will allow us to do a better job at combating pathogens such as S. aureus. Dry cow therapy seems to be working to cure intramammary infections over the dry period and is still a highly recommended practice. But, as always, we still have room for improvement in trying to decrease the number of new infections occurring on our farms and decrease the long-lasingness of these infections. And we’ll keep working with you to seek out other ways to do just this!

The CBMRN’s Videos

CMT: A Simple and Effective Tool

This video is approximately 5 minutes long and it features the steps involved in performing a CMT for the fast detection of mastitis. Pour, swirl and bam, you can tell if Buttercup has an udder infection! Thanks to a quick test result, you can make the appropriate decision right there and then. Fast, simple and effective, a tool worth discovering… or rediscovering!

Free!

To watch the video, just go to www.mastitisnetwork.org, in the Mastitis Online Resources section and click on the Toolbox. Why wait, watch it today!
Mastitis raises several questions to which research has not found an answer yet, due to a lack of relevant data. Fortunately, at the CBMRN, this particular problem has been solved! With the creation of a comprehensive database on dairy farms that reflects the Canadian dairy industry, there are no limits to what we can hope for!

Kristen Reyher is a veterinarian and a Ph.D. student at the Atlantic Veterinary College of the University of Prince Edward Island, under the supervision of distinguished epidemiologist Ian Dohoo. The CBMRN database, created from the results of the National Cohort of Dairy Farms, represents for her a golden opportunity to test several hypotheses related to the dynamics of mastitis. Using complex mathematical models like artificial neural network, Kristen will study the interactions between healthy and infected quarters of the same cow and between different bacteria that cause mastitis.

Her work will lead, among other things, to the development of a practical protocol for diagnosing mastitis using information readily available at the farm. Here are a few examples of tough questions Kristen will find answers to through her research.

**Is a composite sample as useful as a sample from a single quarter to predict udder infections?**

Every month, an individual milk sample is taken from all the cows of a herd for DHI purposes. Since the milk of all four quarters is mixed together, it provides a composite sample. Could this sample be used to accurately predict the infectious status of a cow? Would they be the same regardless of which bacterium is involved?

Depending on the outcome of this research, veterinary practitioners could have access to new diagnostic strategies that would be inexpensive, readily available and above all, very useful to facilitate the follow-up of a herd’s udder health.

**An Artificial Neural Network**

An artificial neural network is a computational model largely inspired by the functioning of real neurons. Neural networks are generally optimized by statistical type learning methods, so that they classify as statistical applications, but also as artificial intelligence methods. They can be used to study patterns and relationships that exist between several groups of data.

**Do infections caused by minor bacteria actually protect cows from more severe forms of mastitis?**

Major pathogens are well adapted to the mammary gland and they are the main cause of mastitis. Minor pathogens may also cause mastitis, but the damages are usually less significant.

Researchers theorize that the immune system could be stimulated by an initial infection caused by a minor bacterium and might therefore become more effective in combating a subsequent infection caused by a major one. Along those lines, it is conceivable that a cow suffering from an infection caused by a major bacterium would be more susceptible to developing a subsequent infection caused by a minor bacterium. So far, no study has been able to shed light on these issues, but with the help of the data from the Cohort, there is now hope for an answer.

**Christopher Reyher is a veterinarian and a Ph.D. student at the Atlantic Veterinary College of the University of Prince Edward Island,** under the supervision of distinguished epidemiologist Ian Dohoo. The CBMRN database, created from the results of the National Cohort of Dairy Farms, represents for her a golden opportunity to test several hypotheses related to the dynamics of mastitis. Using complex mathematical models like artificial neural network, Kristen will study the interactions between healthy and infected quarters of the same cow and between different bacteria that cause mastitis.

Her work will lead, among other things, to the development of a practical protocol for diagnosing mastitis using information readily available at the farm. Here are a few examples of tough questions Kristen will find answers to through her research.

**Is a composite sample as useful as a sample from a single quarter to predict udder infections?**

Every month, an individual milk sample is taken from all the cows of a herd for DHI purposes. Since the milk of all four quarters is mixed together, it provides a composite sample. Could this sample be used to accurately predict the infectious status of a cow? Would they be the same regardless of which bacterium is involved?

Depending on the outcome of this research, veterinary practitioners could have access to new diagnostic strategies that would be inexpensive, readily available and above all, very useful to facilitate the follow-up of a herd’s udder health.

**An Artificial Neural Network**

An artificial neural network is a computational model largely inspired by the functioning of real neurons. Neural networks are generally optimized by statistical type learning methods, so that they classify as statistical applications, but also as artificial intelligence methods. They can be used to study patterns and relationships that exist between several groups of data.

**Do infections caused by minor bacteria actually protect cows from more severe forms of mastitis?**

Major pathogens are well adapted to the mammary gland and they are the main cause of mastitis. Minor pathogens may also cause mastitis, but the damages are usually less significant.

Researchers theorize that the immune system could be stimulated by an initial infection caused by a minor bacterium and might therefore become more effective in combating a subsequent infection caused by a major one. Along those lines, it is conceivable that a cow suffering from an infection caused by a major bacterium would be more susceptible to developing a subsequent infection caused by a minor bacterium. So far, no study has been able to shed light on these issues, but with the help of the data from the Cohort, there is now hope for an answer.

<table>
<thead>
<tr>
<th>Major pathogens</th>
<th>Minor pathogens</th>
</tr>
</thead>
<tbody>
<tr>
<td>Streptococcus agalactiae / dysgalactiae</td>
<td>Coagulase-negative Staphylococci</td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>Corynebacterium spp.</td>
</tr>
<tr>
<td>Escherichia coli</td>
<td>Klebsiella</td>
</tr>
<tr>
<td>Klebsiella</td>
<td>Streptococcus uberis</td>
</tr>
</tbody>
</table>

**Does the presence of an infection in a quarter influence the risk of developing another infection in another quarter?**

By using samples collected at dry-off, calving or intensive periods of collection, Kristen will assess if there is a link between infections of one or several quarters in the same cow. Is there interaction between quarters or are they four distinct entities acting independently from one another? It has always been believed that the quarters of an udder each did their own thing. However, results from a recent study conducted by another CBMRN research scientist, Herman Barkema, would seem to indicate that an infected quarter would make the others more susceptible to infection. So, what is the real story? Intriguing question...
To Be or not to Be…
an Infection?

Usually, when mastitis is detected and you need to know the bacterium involved, you need to take a milk sample and send it to the lab for bacterial culture. However, the criteria used to determine if there is an infection are not necessarily the same from one laboratory to another.

Signe Andersen, a Ph.D. student from University of Prince Edward Island working under the supervision of Ian Dohoo, has undertaken a project aiming to come up with a definition of an intramammary infection. For a given bacterium, she would like to be able to determine whether or not there is an infection based on a SINGLE milk sample. If she could come up with such a definition that would be both scientific and validated, it would be a great step forward for scientists conducting research on mastitis.

To that end, Signe used different cards indicating the level of somatic cell count, the bacterium that was isolated and the number of colonies that were counted on a culture plate. Based on this information, experts were asked to decide if there was indeed an infection or not. From the analysis of the answers recorded over the course of several sessions and at different times, she finally established two criteria that will serve to narrow down a new definition of what constitutes the infection of a quarter. This definition, which she qualifies as "pseudo-gold standard", is currently tested using milk samples collected in 2007 as part of the National Cohort of Dairy Farms. She will also compare the value of a sample in duplicate and triplicate to that of a single sample.

Although the work is conducted far from the farm, this research could well be very useful to diagnostic laboratories and the scientific community by providing a mastitis standard so that researchers can compare apples with apples!
The Impact of Management Practices on the Incidence of Udder Infections

To date, our knowledge on this topic has essentially been based on prevalence studies. Such studies only provide a static picture of the situation at a given point in time. For instance, we could come to the conclusion that there is a link between wearing gloves during milking and a reduction of the risk of propagating pathogen bacteria. However, we could not determine if wearing gloves helps in preventing new infections. And yet, this information is critical since we can only hope to control mastitis within a herd through the prevention of new infections.

To that end, incidence studies are necessary. However, this type of research requires significant resources, which explains why they are rarely conducted. Having said that, the infrastructure set up as part of the CBMRN National Cohort of Dairy Farms now makes a study of such magnitude possible.

Simon Dubour, veterinarian practitioner and Ph.D. student at the Faculté de médecine vétérinaire de l’Université de Montréal, under the supervision of Daniel Scholl, has collected a mass of data about common management practices, farm type and mastitis control with the help of a detailed questionnaire that was administered in 88 Cohort farms. Concurrently, four series of milk samples were taken over intensive periods of time. For instance, we could come to the conclusion that at any given time over the year, we can expect to see close to 6% of all quarters of the herd with a CNS infection.

Figure 6. A winning condition: cleanliness!

An udder with a score of 3 is three times more likely to get infected
An udder with a score of 4 is seven times more likely to get infected

By linking the data collected through the questionnaire with the results of the bacterial testing performed on the milk samples, it will soon become possible to show which management practices have a significant impact on the occurrence of new udder infections and on what particular bacteria. Preventing the occurrence of new infections will generate both a greater and longer lasting impact on the farm than resorting to short term alternatives such as antibiotic treatments and culling.

What is the difference between prevalence and incidence of mastitis at the farm?

Prevalence
It is the measure of the presence of mastitis within a herd at a specific point in time. Milk samples are collected from a group of cows or from the entire herd and tested using bacterial cultures and somatic cell counts. It is the equivalent of taking a picture of the herd on Day X.

Incidence
It is the measure of the presence of mastitis in a herd over a period of time. Milk samples are collected over a given period of time in order to identify the occurrence of new infections and find out how long they last. The picture of the herd changes on a daily basis.

Many management practices are recognized for their effectiveness in the preventing and controlling mastitis. However, it is presently impossible to establish without question which practices yield the best results, under what conditions and for which bacteria. With the help of the data gathered by the CBMRN, researchers will soon be able to assist producers in identifying which practices are the most appropriate depending on different circumstances.
8- In the World of Mastitis Research - What's New?

Dry-off: No Time to Let Your Guard Down!

Udder infections acquired during the dry period are difficult to diagnose and tend to reduce the subsequent milk production. How frequent are new infections caused by various bacteria during the dry period? What practices are the most effective in preventing such infections? Here is the summary of a study on that particular topic.

Almost 50% of such infections are caused by environmental bacteria and can be prevented through the sole use of an antibiotic treatment at dry-off. In fact, the use of these treatments has virtually eliminated certain major pathogenic bacteria such as Strepococcus agalactiae. Furthermore, a reduction of nearly 80% of infections caused by coagulase-negative staphylococci (CNS) has been observed following the use of an antibiotic treatment at dry-off in herds of the CBMRN’s National Cohort of Dairy Farms.

In order to study the rate of new udder infections during dry-off, Chris Calloway, a Ph.D. student at University of Prince Edward Island under the supervision of Ian Dohoo, obtained milk samples from each farm enrolled in the Cohort, both composite and from individual quarters. They also obtained milk samples from 15 cows at various periods around dry-off: 14 to 30 days, 0 to 14 days before drying off and, 24 to 48 hours, 7 to 14 days after calving. All these samples were sent to the lab for bacterial cultures in order to detect any sign of mastitis.

The bacteria found the most often across Canada in these samples were:
- Coagulase-negative staphylococci;
- Staphylococcus aureus;
- Corynebacterium bovis;
- Environmental streptococci; and
- Other Gram-positive bacteria.

As part of this study, the recovery rate and the incidence of new udder infection during the dry period were calculated per quarter. Chris also performed several analyses per farm taking into account the bacteria involved, management practices, feeding, housing type and the use of dry cow treatment and teat sealants. An economic study was conducted on the costs and benefits of using dry cow treatment.

Chris is currently writing his Ph.D. thesis and he will be submitting scientific articles which will present the findings of his study. Stay tuned!

Until the findings of this study are published, the following fact sheet will jog your memory about the appropriate method for administering an intramammary treatment such as antibiotics at dry-off.

Internal teat sealant would reduce the risk of a new udder infection by close to 50%!

A meta analysis conducted by Chris Calloway on the effects of internal teat sealants, with or without antibiotics, compared to the administration of antibiotics only or the absence of treatment, showed that using internal teat sealants during the dry period reduced the risk of a new udder infection by nearly 50% (ranging from 33 to 75%, depending on the study).

Mastitis prevention: show and tell!

In cooperation with veterinarians, the CBMRN has put together a series of 9 illustrated fact sheets and a computerize calculator intended for dairy producers and the various stakeholders interested in udder health. These fact sheets are designed to be used on the farm to serve as a guide to best udder health management practices. Detailed illustrations are combined with clear and simple explanations that can serve as reference training material!

These fact sheets and the calculator are available online at www.mastitisnetwork.org in the Mastitis Online Resources section, in PDF format easy to read and convenient for printing. They are:
- Cow Cleanliness Assessment, Step-by-step Milking Procedure, California Mastitis Test (CMT), Milk Sample Collection Technique for Bacterial Testing, Administration Technique of Intramammary Treatment in Dairy Cows, Injection Technique for Dairy Cows, Observation of Mammary System Conformation, Teat Abnormalities, Teat Condition Evaluation Table and the Computerized Teat Condition Calculator (Excel based).
Immunizing the Mammary Gland against Mastitis: Quite the Challenge

In 2006, Brian Talbot, a researcher at the Université de Sherbrooke, succeeded in reducing the duration of a mastitis caused by S. aureus by using an experimental vaccine based on DNA of bacteria. Unfortunately, these results could not be duplicated up to now.

The researcher is still focusing on the same objectives, but this time with recombinant proteins as antigens. In 2007, he studied S. aureus to look for proteins that would have the greatest capacity for triggering a strong immune response and he found four of them. After isolating and purifying them, the four antigen proteins did indeed trigger the desired response in mice and then, in cows. Unfortunately, the bovine immune response was detected in the main bloodstream, but with very little effect in the mammary gland. The vaccine has therefore not provided protection against udder infections.

This outcome did not deter Brian Talbot. Even though his work has been completed as part of the CBMRN research program, Brian is pushing forward by focusing on the immunity obtained with the four proteins. Once this aspect will be well controlled, he plans to go back to a double immunization formula – proteins and DNA – in combination with a high-performance adjuvant. He will also look at new ways of administering the vaccine to increase its effectiveness.

There are two other major projects dealing with vaccines against mastitis within the CBMRN. The main issue in this field is not only the quest for the best antigen, but also the appropriate formulation and the optimal mode of administration to maximize the response of the immune system.

Against S. aureus mastitis

Most vaccines against S. aureus are formulated with traditional adjuvants and administered by subcutaneous or intramuscular injection in sites that are away from the mammary gland. Research scientists Andrew Potter and Jose Perez-Casal, from the Vaccine and Infectious Disease Organization, are working on new strategies to increase the effectiveness of these vaccines:

• Formulation of new adjuvants;
• New routes of administration; and
• More specific antigens.

The combination of these components has the capacity of amplifying the immune response in bovines, extending the duration of the protection and reducing the need to administer one or more booster shots.

Since 2006, their research has lead to the identification of the GapC/B protein as an antigen displaying an excellent immune response in dairy cows. Over the last year, this same antigen, administered in different quantities, was used to compare the effectiveness of a vaccine administered by subcutaneous injection with that of a needle-free injection. The results show that a vaccine containing 10 times fewer antigens administered with a needle-free device was just as effective as a conventional dose vaccine injected subcutaneously.

Over the next year, the researchers will focus their efforts on increasing the immune response by developing new vaccine formulations in collaboration with Brian Talbot. These new formulations are intended to extend the immunity provided and reduce the vaccination frequency.

Against coliform mastitis

Since December 2008, Xin Zhao, of McGill University, took over from Grant Tomita, formerly with Université de Montréal, as lead scientist of a research project on a new vaccine against coliform mastitis.

There are already two commercially available vaccines against coliform mastitis in Canada. Research has shown that these two vaccines are useful in reducing the severity and duration of clinical mastitis caused by E. coli.

However, these vaccines require 2 or 3 booster shots to achieve a sufficient production of antibodies. This represents additional workload and costs to the producers. To counter the problem, Grant Tomita had the idea of using microencapsulation of the J5 antigens that form the vaccine. In other words, using a fairly simple laboratory procedure, the antigens are enclosed in polymer microspheres. Once administered to the animal, these beads biodegrade and promote the progressive release of the antigens after a single injection.

The prototype vaccine showed some promise as it stimulated an immune response in the vaccinated animals comparable to that of current commercial vaccines. The next step will require infection testing in cows to verify the effectiveness of the new formulation of the vaccine. Discussions are underway with the biomedical sector to transfer this vaccine to a commercial partner and steps have already been taken to protect the intellectual property of this technology. A story worth following!

Immunology 101

The immune system has several weapons in its arsenal when it is time to defend against an invader. The two main defence modes on which researchers focus their efforts are the production of antibodies and cellular immunity.

Antibody production

When bacteria invade a host, it is analyzed by the immune system which then produces antibodies to eliminate the invader. In the same way, a vaccine which contains portions of the bacterium or the killed bacterium prepares the immune system to quickly recognize the pathogen by giving it the opportunity to manufacture specific antibodies in advance. When the bacterium invades the host, it is rapidly intercepted by the antibodies and destroyed by neutrophils without triggering any symptom. Neutrophils are cells that cause an elevation of the milk cell count during mastitis.

Cellular immunity

The pathogenic bacteria may sometimes hide in a cell, rendering the antibodies ineffective because they are unable to attach themselves to the bacteria. The immune system then activates cellular immunity, the second line of defense, which involves killer cells. These cells are able to recognize the antibody of the bacterium if it is not completely hidden in the bacterium. They then destroy the entire cell and part of its surroundings, causing the inflammation.

That is why it is so important to identify the bacterium antigen correctly so that the immune system can deploy the corresponding weapons.

What is an adjuvant?

In immunology, an adjuvant is a substance that stimulates the immune system and enhances the response to a vaccine, but without any antigenic properties.

Needle-free injection. How does it work?

A specially designed syringe is held against the skin. When activated, it forces a liquid to pass through a very small opening. The ultrathin stream created by this very high pressure penetrates the skin without having to resort to a needle.
Visible mastitis signs
According to CBMRN research, clinical mastitis cases in cows enrolled in the Cohort occurred as follows:

51% of cases: appearance (flakes, clots, watery milk)
38% of cases: swollen udders, reddening, warmth
11% of cases: systemic signs (fever)

WANTED: Bacteria Guilty of Causing Mastitis
Whenever a crime is committed, it is often possible to establish the identity of the culprit through DNA traces left behind on the crime scene. Soon, mastitis causing bacteria will also be treated as common criminals, thanks to new PCR tests developed especially to uncover them. Veterinary practitioners and dairy producers will have the opportunity to play Columbo and potentially have a powerful new diagnostic tool at their disposal to manage mastitis.

A Milk Protein that Protects Against Coliform Clinical Mastitis
Symptoms or visible signs of coliform mastitis are mostly due to the response of the cow’s immune system to the toxins produced by the bacteria. One of these toxins is called lipopolysaccharide (LPS), also known as an endotoxin. When there is an infection, LPS multiply rapidly in the mammary gland and to destroy them, leukocytes secrete a special protein called CD14. This CD14 protein binds to LPS, neutralizing their toxic effect.

The results of a study conducted in the United States suggest that developing a CD14 based product could be effective to prevent the onset of mammary infections caused by E. coli during the dry period. This hypothesis strongly interested Xin Zhao, lead scientist from McGill University, to the point of initiating a project to verify this hypothesis. A positive outcome could open the door to selecting cows that naturally secrete larger quantities of this protein or to administering it as a form of treatment.

Despite several attempts to determine if there is a link between the concentration of CD14, the presence or absence of bacteria in the milk and the somatic cell count, Xin Zhao could not arrive at conclusive results on the beneficial effects of protein CD14. Since the research hypothesis could not be demonstrated clearly enough during the first three years of the project, activities for this project as part of the CBMRN are now terminated.

The technique used to analyze bacteria DNA is polymerase chain reaction better known as PCR. Some laboratories have offered PCR service for mastitis diagnosis for a few years now, but the available tests can only identify a single bacteria at a time and are more costly than traditional bacterial cultures.

However, new PCR tests capable of identifying several pathogens simultaneously are making their way on the market. They are called multiplex PCR. In most cases, these tests can even measure the number of bacteria in the sample. In this case, the PCR is said to be in real time. In a laboratory setting, these tests are very effective and offer a promising avenue for diagnosing mastitis. Still, their actual sensitivity and specificity in the field are still unknown.

Daniel Scholl, Scientific Director of the Canadian Bovine Mastitis Research Network and epidemiology professor at the Faculté de médecine vétérinaire of Université de Montréal, has therefore decided to correct the situation and initiated a large-scale project designed to validate three PCR kits that are or soon will be on the market (see “Taking PCR Tests out for a spin”).

In this project, Daniel Scholl and his team will assess the reliability of PCR tests by evaluating the percentages of false negative (sensitivity) and false positive results (specificity). One and the other both have a significant impact on the farm because a wrong diagnosis can lead to an inappropriate management decision.

To carry out the project, Daniel Scholl and his team can use a phenomenal quantity of samples collected across some 91 farms of the CBMRN’s National Cohort of Dairy Farms. The Cohort provides an excellent representation of the Canadian dairy population. The results of the study will have the advantage of being directly applicable to the context of our farms. Furthermore, the pool of samples includes...
Sensitivity and specificity: what is the difference?

The sensitivity of a test refers to its ability to provide a positive result when there are in fact bacteria present in a milk sample. The specificity of a test refers to its ability to provide a negative result when the sample does not contain the target bacterium.

The higher the sensitivity and specificity values, the more effective the test will be in determining the actual presence or absence of bacteria in the sample.

cows with clinical mastitis (presenting symptoms) and cows in lactation without any apparent symptoms.

There is no perfect diagnostic method to detect mastitis bacteria in milk. The good old bacterial culture, the one we normally use to analyze samples sent to the laboratory, serves as the reference method or gold standard. Therefore, all samples collected as part of the Cohort were first submitted for bacterial cultures, the results of which will then be compared with those from PCR analyses.

Progress report
Currently, 1,765 cases of clinical mastitis are undergoing analysis with the PCR equipments under study. Afterwards, 4,000 milk samples collected from apparently healthy cows in lactation will be analyzed. In the latter, maybe we will find infections that had not been detected by bacterial culture? Will we at last have a way of "seeing" subclinical mastitis? The four bacteria we are looking for in these samples are: Streptococcus aureus, Streptococcus uberis, Escherichia coli and Staphylococcus agalactiae.

All results are expected to be in by the fall of 2010. Concurrently, researchers will try to see if there is a correlation between the specificity and the sensitivity and the lactation phase at the time of the sampling, the somatic cell count, the interval between the collection of the sample and the performance of the analysis, the infectious profile of the herd and the presence of other bacteria.

Impact on the farm
With the pool of milk samples collected through the Cohort, the CBMRN is the only resource in a position to conduct such a large scale and unique project. In early 2011, the results should show the level of accuracy obtained with this new equipment for the diagnosis of mastitis, information that should be of great interest to diagnostic laboratories. As for the farm, diagnosing clinical and subclinical mastitis quickly and accurately would certainly have a major impact on udder health management decisions.

Taking PCR Tests out for a spin

DairyGuard™ from SafeGuard Biosystems Inc., Canada

This system can identify several bacteria in a single milk sample. It is not available on the market yet.

PathoProof™ Mastitis PCR Assay from Finnzymes Oy, Finland

This system can detect 11 different bacteria or groups of bacteria. It has been on the market for three years.

Multiplex Real-Time PCR, University of Tennessee, Knoxville, United States

This PCR test was developed by Stephen Oliver from University of Tennessee. It can identify simultaneously four major mastitis bacteria in real time; that is why the test is called "Multiplex Real-Time PCR".

How does PCR work?
Three components are required to perform a PCR test: a PCR tube (small heat resistant test tube), some reagents and a source of heat. As little as 70 µL of infected milk are required to perform a PCR test. This tiny amount of milk is first subjected to an enzyme reaction which breaks down the cells of the bacteria present in order to release their DNA or genetic code. The DNA strands are then purified and placed in a PCR tube. The following is then added:

• Two reagents which predetermine the gene sequence to be copied (which enables us to target the bacterium we are looking for);
• Nucleotides (A, C, G, T) that make up the DNA code; and
• An enzyme, DNA polymerase, that is capable of reading the genetic code of the bacterium and then creating copies of the specific gene sequence.

This mixture is then subjected to several consecutive cycles – heating and idling – which is how the multiplication of nucleotide sequences occurs. The identification of the copied sequence can be confirmed by specially designed software or by other laboratory procedures.

Is it time to get rid of bacterial culture?

Bacterial culture is a useful and effective tool for the detection of bacteria, but it does have some limitations. A low concentration of bacteria in the sample or selecting the incorrect culture media may lead to a "no growth" result. In some cases, additional cultures or tests must be performed to distinguish between two similar bacteria. Contamination of the milk sample at the farm is always a possibility and can lead to an incorrect diagnosis (4 to 20% of samples). Furthermore, the incubation time creates a waiting period and it may take a while before the results get to the farm. However, bacterial cultures are presently more affordable than PCR tests and are offered by all diagnostic laboratories across the country.

PCR testing promises to overcome the limitations of bacterial cultures and will offer new avenues for research and for managing mastitis among herds. For the time being, bacterial cultures remain a readily available tool that is still relevant to on-farm udder health programs.

References
1. The Genetic Science Learning Center of the University of Utah has developed the learn.Genetics™ Web site, which presents animated demonstrations of many laboratory techniques that are available to students, the faculty and the public. http://learn.genetics.utah.edu/content/labkit/
In the World of Mastitis Research - What's New?

Staphylococcus aureus

Know your Enemy to Prevail

Francois Malouin, research scientist from Université de Sherbrooke, and his team examined 240 strains of S. aureus from the Mastitis Pathogen Culture Collection and discovered interesting associations between the presence or absence of certain genes in S. aureus and clinical mastitis, subclinical mastitis and the formation of biofilm. After experimentally infecting cows with different strains, they analyzed the virulence genes that were expressed. They found that similar genes were expressed in bacteria that cause chronic mastitis.

Impact on the farm
The genes involved in virulence are not all known yet. However, two such genes that have been discovered by François Malouin’s team are currently the subject of two invention disclosures. These genes represent new treatment targets for the development of vaccines or antibiotics to fight against S. aureus mastitis. With such progress, we will get to know the enemy better and we will be able to rely on better weapons to win the battle.

The enemy nests in the udder
The ability of S. aureus to cause intramammary infections represents an awesome example of bacteria adapting to their environment. Here is a short account of how they go about their business. The first stage of a S. aureus infection is the adhesion. The bacteria penetrates into the mammary gland and attaches itself to the gland tissue (A). To do so, at the beginning of its growth, it expresses proteins called adhesins. These proteins recognize the components of the gland, bind to them and allow S. aureus to attach. The bacteria can now multiply without running the risk of getting flushed out during milking.

Once bacteria have reached sufficient numbers, they secrete virulence factors (toxins) that break down the gland tissue to extract nutrients and to migrate to other sites (B). At this stage, the cow’s immune system is already trying to fight the ongoing infection. Symptoms begin appearing and we are now faced with a case of clinical mastitis.

The enemy nests in the udder
The ability of S. aureus to cause intramammary infections represents an awesome example of bacteria adapting to their environment.

Illustration: LE PRODUCEUR DE LAIT QUEBECOIS JULY/AUGUST 2009 page 32

The ability of S. aureus to cause intramammary infections represents an awesome example of bacteria adapting to their environment.
Validation of an On-Farm Mastitis Bacteria Identification System

Providing appropriate treatment for clinical mastitis is an ever-present concern for veterinarians and dairy producers alike. In some cases, administering antibiotics may not be justified because of their poor performance against certain bacteria, such as coliforms, but also because of the high rate of spontaneous recovery from certain mastitis. In addition, the use of antibiotics requires a milk withdrawal period which causes economic losses to the farm.

Using fast diagnostic tools to identify pathogenic bacteria may well be the best option that would contribute to a rationalization in the use of antibiotics without compromising the cure rate. That is the challenge facing the team of Greg Keefe, veterinarian and epidemiologist at the University of Prince Edward Island in a project carried out by Jennifer McCarron, M.Sc. student, and Kimberley MacDonald, Ph.D. student.

Treatment assigned at random
CBMRN technicians enrolled 54 Cohort farms from six provinces to participate in the validation of an on-farm fast diagnostic tool: the PetrifilmTM system. This system had previously been selected during the first stage of this project.

For each case of mastitis of score 1 or score 2, the producer had to make a random selection between a milk culture using the PetrifilmTM test or an antibiotic treatment. A cow suffering from a score 3 mastitis - displaying mastitis visible signs and a rectal temperature above 39.5 °C - was treated immediately according to normal farm procedures. A little over 800 cases of clinical mastitis were selected for this study.

Preliminary results show a reduction of nearly 36 % in the use of antibiotics for the bacterial culture group compared with the immediate antibiotic treatment group. In addition, the milk of these cows could be returned to market 1.5 days sooner than the milk of cows treated with antibiotics. Furthermore, the cost of the on-farm culture would be less than or equal to that of the antibiotic treatment. Several economic parameters have yet to be measured and compared with the help of standard methods.

In the course of the project, the research team also worked on putting together a culture kit that is now available to producers and veterinarians who wish to use it (www.milkquality.ca).

Clinical mastitis

<table>
<thead>
<tr>
<th>Mild (1)</th>
<th>Moderate (2)</th>
<th>Severe (3)</th>
</tr>
</thead>
<tbody>
<tr>
<td>☐ Abnormal milk</td>
<td>☐ Abnormal milk</td>
<td>☐ Abnormal milk</td>
</tr>
<tr>
<td>☑ Normal udder</td>
<td>☐ Abnormal udder</td>
<td>☐ Abnormal udder</td>
</tr>
<tr>
<td>☐ Normal cow</td>
<td>☐ Normal cow</td>
<td>☐ Abnormal cow</td>
</tr>
</tbody>
</table>

The CBMRN just launched a brand new tool in the fight against mastitis in an effort to improve the udder health of Canadian dairy cattle.

Intended for use by veterinarians, this kit offers a customized five-point innovative approach:

1. Udder health and assessment
2. Treatment
3. Infection pressure
4. Milking
5. Cow resistance and the transition period

Developed through a vast consultation initiative with Canadian and international experts, the kit contains ready-to-use educational material that is easy to integrate into daily practice in the form of a binder and a USB flash drive that include:

- Convenient illustrated sheets in full color
- Informational leaflets
- Assessment questionnaires
- Calculation tools
- Videos and much more…

For more information, please visit: www.mastitisnetwork.org.

The Veterinary Kit for Udder Health is currently available in French, but an English version will soon follow.

Dealing with mastitis? Break out your kit!
The immune system is responsible for controlling contagious diseases and is largely governed by genetics. Thus, benefiting from a stronger immune system, some cows are more resistant to diseases and require fewer therapeutic interventions. Their immune system protects them from infectious agents, including mastitis causing bacteria.

Using immunization protocols, Bonnie Mallard, research scientist in immunogenetics at the University of Guelph, characterized the DNA of close to 700 cows from 58 herds of the CBMRN’s National Cohort of Dairy Farms and sorted them into two populations: strong resistance and weak resistance to disease. This invaluable piece of information then gave her the opportunity to discover many genes and proteins that regulate resistance to disease as well as certain production parameters such as the somatic cell count, persistency in milk and yield in milk and proteins. At present, the genotype of 500 Holstein, 100 Jersey and 70 Guernsey bulls is about to be completed to verify their correlation with various health and production parameters.

Benefits from this research:

• The ability to identify resistant cows in commercial herds;
• The development of the estimated breeding value (EBV) for an improved herd health, and the possibility of selecting cows that are more resistant to disease;
• The identification of genes and proteins associated with production parameters;
• Support to the development of effective vaccines;
• The detection of mastitis through certain proteins found in the milk.

A very special strain of S. aureus

Concurrently to the work on resistance genes, Bonnie Mallard and Heba Atalla, Ph.D. student at the University of Guelph, wondered why cows suffering from mastitis caused by Staphylococcus aureus are sometimes so difficult to treat. They hypothesized that these cows might harbour a special strain of S. aureus. To put their theory to the test, Heba cultured milk samples taken from 11 cows suffering from chronic mastitis caused by S. aureus. The results brought to light the strain they were looking for: a small colony variant of S. aureus. This specific strain originates from the ability of the bacteria to transform genetically into a new slow-growth colony that can resist to antibiotics and vaccines. This S. aureus variant exists in human medicine and is unfortunately resistant to numerous antibiotics.

In order to better characterize this strain called SCV Heba3231 along with three other strains of S. aureus, they were inoculated in four groups of five cows that were clinically healthy.

In the group of cows inoculated with SCV Heba3231, researchers observed a mild mastitis with very few visible signs. It seems that this strain also has a great ability to hide inside epithelial cells of the mammary gland, thereby protecting itself from antibiotics.

This study is the first one to show this variant of S. aureus in cows with chronic mastitis. It is believed that this strain could contribute to the extended survival of S. aureus in certain cases of chronic mastitis. Just as for S. aureus, the best way to prevent and control these bacteria is compliance with the NMC Recommended Mastitis Control Program.

Association between the Use of Antibiotics to Treat Mastitis and Antibiotic Resistance

Animals, just as humans, need antibiotics and other antimicrobial agents to prevent or treat diseases they get. The use of these products improves their health and well-being. Yet, the controversy surrounding their use in agriculture comes mostly from the intense non-therapeutic use of antibiotics as growth factors that can lead to the appearance of resistance.

Since mastitis is the first cause for using antibiotics in the dairy industry, it is worthwhile to shed some light on the situation. Do these treatments induce resistance in the bacteria that cause mastitis? This is the question for which Vineet Saini, Ph.D. student at the University of Calgary, under the supervision of Herman Barkema, is attempting to find an answer. The collection of data necessary to conduct this project was carried out in cooperation with the producers of the 91 farms enrolled in the CBMRN’s National Cohort of Dairy Farms. For the purpose of the project, the producers had to collect all empty antibiotic containers used on the farm. They also had to keep a log of all individual treatments, dosage and antibiotics used.

The CBMRN technicians and students then proceeded to make an inventory of the packages and compile all the data in each of the six Canadian provinces participating in the study.

Concurrently, several series of milk samples were taken on all these farms in 2007 and 2008. These samples will be analyzed in order to assess the resistance profile of three common bacteria that cause mastitis: Staphylococcus aureus, Escherichia coli and Klebsiella.

This year, the researchers have completed their inventory of all medication used on the farms. Figure 7 shows the proportion of antibiotic treatments administered by intramammary infusion and by injection. The results of the analysis on antibiotic resistance will be known in 2010.

Where does antibiotic resistance come from?

Experts attribute the rise of antibiotic resistance to three main factors:

1. When an antibiotic is prescribed to fight off a disease that is not caused by bacteria, the treatment is ineffective. The same is true for an antibiotic that is not adequate against the specific bacteria it is supposed to destroy.

2. Inappropriate use. When the correct dosage and duration of treatment are not adhered to, some bacteria survive the treatment and may then develop some resistance to the medication they have been exposed to. The weak will be eliminated, but the strong will survive and multiply. This gives rise to a relapse of the disease in a form that is difficult to treat.

3. Environmental contamination. The modern agri-food industry often integrates antibiotics in the feed of livestock to speed up growth. It is the case with beef, veal, pork, poultry and farmed fish. Antibiotic substances are also sprayed over orchards to prevent or fight against fruit diseases. Residues of antibiotic substances find their way in the food we eat. They also end up in streams and in groundwater, contaminating sources of drinking water. The same is true for medical waste if it is not disposed of adequately.

Source: www.naitreetgrandir.net

Figure 7. Intramammary tubes used to treat mastitis represent 28% of all antibiotics used on dairy farms while injections make up the rest.
**Research Team**

The Canadian Bovine Mastitis Research Network comprises 35 active members and collaborators from Canadian research institutions. The research program also involves international collaborators.

**Members:**

- **Herman Barkema**
  University of Calgary
  barkema@ucalgary.ca

- **Nathalie Bissonnette**
  DSKDC-Agriculture and Agri-Food Canada
  nathalie.bissonnette@agr.gc.ca

- **Émile Bouchard**
  Université de Montréal
  emile.bouchard@umontreal.ca

- **John Campbell**
  University of Saskatchewan
  john.campbell@usask.ca

- **Luc DesCôtes**
  Université de Montréal
  luc.descoteaux@umontreal.ca

- **Trevor DeVries**
  University of Guelph
  tdevries@kemptville.uoguelph.ca

- **Moussa Sory Diarra**
  PARC-Agriculture and Agri-Food Canada
  diarram@agr.gc.ca

- **Ian Dobos**
  University of Prince Edward Island
  idobos@upei.ca

- **Monique Doré**
  Université de Montréal
  monique.dore@umontreal.ca

- **Marcelo Gottschalk**
  Université de Montréal
  marcelo.gottschalk@umontreal.ca

- **Philip Griebel**
  University of Saskatchewan - VIDO
  philip.griebel@usask.ca

- **Neil A. Karrow**
  University of Guelph
  nkarrow@uoguelph.ca

- **Greg Keefe**
  University of Prince Edward Island
  gkeefe@upei.ca

- **François Malouin**
  Université de Sherbrooke
  francois.malouin@usherbrooke.ca

- **Jose Perez-Casal**
  University of Saskatchewan - VIDO
  jose.perez-casal@usask.ca

**Collaborators:**

- **Andrew Potter**
  University of Saskatchewan - VIDO
  andrew.potter@usask.ca

- **Claude Robert**
  Université Laval
  claude.robert@uqal.ca

- **Jean-Philippe Roy**
  Université de Montréal
  jeanphilippe.roy@umontreal.ca

- **Daniel Scholl**
  Université de Montréal
  daniel.scholl@umontreal.ca

- **Henrik Strehn**
  University of Prince Edward Island
  hstrehn@upei.ca

- **Brian Talbot**
  University of Sherbrooke
  brian.talbot@usherbrooke.ca

- **John VanLeeuwen**
  University of Prince Edward Island
  jvanleeuwen@upei.ca

- **Xin Zhao**
  McGill University
  xin.zhao@mcgill.ca

**To Contact Us**

**Réseau canadien de recherche sur la mammite bovine**
Canadian Bovine Mastitis Research Network

**Faculté de médecine vétérinaire**
Université de Montréal
P.O. Box 5000, Saint-Hyacinthe (Québec) Canada J2S 7C6

**Members:**

- **Daniel Scholl**
  Scientific Director
  Phone: 450 773-8521, ext. 8605
  daniel.scholl@umontreal.ca

- **Marie-Ève Paradis**
  Scientific Assistant
  Phone: 450 773-8521, ext. 8621
  marie.eve.paradis1@umontreal.ca

- **Amik L’Esperance**
  Network Manager
  Phone: 450 773-8521, ext. 8619
  amik.lesperance@umontreal.ca

**Collaborators:**

- **James Bellamy**
  University of Prince Edward Island
  bellamy@upei.ca

- **Patrick Boerlin**
  University of Guelph
  pboerlin@uoguelph.ca

- **Vilceu Bordignon**
  McGill University
  vilceu.bordignon@mcgill.ca

**The success of our Network is possible thanks to our funding and research partners, donors and sponsors: the Natural Sciences and Engineering Research Council of Canada and many private and governmental organizations. A special appreciation is dedicated to all the Canadian dairy producers who also participate financially in the research program and are involved in every decision-making level.**