**NE-1748 Multi State Research Project** **Annual Meeting**

**Mastitis Resistance to Enhance Dairy Food Safety**

Annual Business Meeting

Chicago, IL

October 25, 2018

1. The 2018 (FY18) annual business meeting of the NE-1748 Multistate research project was called to order at approximately 5:00 pm.
2. Members present at meeting: 12; stations represented: 11; attendance at conference: 25

Bill Owens Louisiana Louisiana State University

Kasey Moyes Maryland University of Maryland

Ron Erskine Michigan Michigan State University

Sandra Godden Minnesota University of Minnesota

John Middleton Missouri University of Missouri

Pamela Adkins Missouri University of Missouri

John Barlow Vermont University of Vermont

Massimo Bionaz Oregon Oregon State University

Oudessa Kerro Dego Tennessee University of Tennessee

Amanda Stone Mississippi Mississippi State University

Paolo Moroni New York Cornell University

Benjamin Enger Ohio Ohio State University

1. There was no Project Administrator’s report. Current administrative advisor: Kumar Venkitanarayanan, who was not in attendance.
2. Discussed that all members need to fill out an Appendix E and that we need a technical representative at each station. The Appendix E lists all members of the project. Concerned that some members are not participating. We discussed emailing our administrative advisor regarding action to be taken, if any, for those stations that are listed in Appendix E, but no participating in the project. Kasey Moyes agreed to contact the administrative advisor with this concern.
3. Discussed the annual report (needs to be completed within 60d after the meeting). The report will be appended to these minutes and filed in NIMSS.
4. Discussed creating a document listing available microbial isolate collections and other biological specimens that could be made available for use among members. Logistics were discussed. Members were made aware of potential collections (e.g. Southeast Quality Milk Initiative, Canadian Bovine Mastitis and Milk Quality Network, University of Missouri, Washington State University). In the interim it was advised that those seeking materials can contact curators of these collections.
5. The 2018 meeting will be (tentatively) held November 6-8th in Columbia, Missouri.
6. Benjamin Enger was nominated to serve as the NE-1748 secretary, agreed to accept the nomination, and was elected.
7. Meeting was adjourned.

**NE-1748 ANNUAL REPORT WORKSHEET (FY18)**

**OBJECTIVE 1:** Characterize host mechanisms and pathogenic virulence factors associated with mastitis susceptibility and resistance to improve economic outcomes and animal welfare.

Louisiana State University, Ag Center

Member: William E. Owens

Mastitis pathogens continue to be identified from bovine and goat milk samples submitted to this laboratory. Organisms were identified and mastitis reports generated to producers and field agents. Organisms were evaluated for antimicrobial susceptibility to a battery of antibiotics to determine therapeutic efficacy.

Michigan State University

Member: Ronald Erskine, Lorraine Sordillo, Andres Contreras

Collaborators: Phil Durst, Ruben Martinez, Stan Moore

We continued to study the impact of negative energy balance and fat mobilization vascular endothelial and adipose inflammatory responses of in vitro by changing the expression of important inflammatory mediators. In particular, the role of n-3 fatty acid content in altering the profile of vasoactive eicosanoids and the role of poly-unsaturated fatty acids on attenuating endothelial cell inflammatory responses, as well as lymphocyte expression of pro-inflammatory cytokines was a central aim of our work.

Oregon State University

Member: Massimo Bionaz

Collaborators: Gerd Bobe, Charles Estill, Shana Jaff, and Matteo Mezzetti and Erminio Trevisi (Università Cattolica del Sacro Cuore, Italy)

We performed an experiment to assess the effectiveness of feeding a relatively low amount of Se-fortified hay on improving the condition of Se and the effect on anti-oxidative status, immune condition, and transcriptome of liver and mammary macrophages in lactating primiparous. For the purpose we used 18 heifers (10 Jerseys and 8 Holsteins) that were fed ad libitum with TMR and, from approx. 40 days prior parturition to 2 weeks post-partum, were supplemented with 1 kg of Se-fortified Alfalfa hay (3.25 mg of Se/kg DM)/100 kg of body weight or the same amount of a control Alfalfa hay (0.4 mg of Se/kg DM). We collected blood samples for analysis of the immune status, metabolic and inflammatory profiling, antioxidative status, and level of microminerals, including Se. We collected also liver via biopsy and macrophages from milk somatic cells. On the liver we measured level of Se. In liver and macrophages we are measuring whole transcriptome. Our preliminary data indicated that the treatment was effective in increasing the Se in liver and blood, improving the antioxidative status, and improve milk production in Holsteins only. No effects were observed in the immune status or Se in milk.

University of Idaho

Member: Pedram Rezamand

Contributors: Massimo Bionaz (Oregan State University), Chia-Yu-Tsai (University of Idaho)

We have been collaborating with the Oregon State team on PPARg activator (2,4-thiazolidinedione or TZD) and its potential effect on mastitis. We have developed methods to detect various isomers of vitamin A in serum/milk and how they may response to TZD treatment during induced mastitis.

University of Maryland

Member: Kasey M. Moyes

Contributors: Daniel Nelson, John Roche, Theodore Elasser

In collaboration with Dr. Richard Erdman in the Department of Animal and Avian Sciences (ANSC) at the University of Maryland (UMD), we investigated the use of milk instead of mammary tissue biopsies to investigate the response of animals due to treatment in lactating dairy cattle. International collaborations with Dr. John Roche, Dairy NZ in New Zealand, have led to the identification vof neutrophil function during the transition period. In collaboration with Dr. Theodore Elsasser at the Agricultural Research Service within the USDA, we have investigated the effect of dietary vitamin E on the neutrophil response in lactating dairy cattle.

University of Minnesota

Members: S. Godden, E. Royster, L. Caixeta, B. Crooker, R. Fink, T. Schoenfuss

Contributors: L. Fox (WSU), P. Gorden (ISU), D. Nydam (Cornell University), S. Sreevatsan (MSU)

K. Patel, S. Godden, S. Sreevatsan, E. Royster, L. Fox Molecular Epidemiology of Staphylococcus aureus on U.S. Dairy Farms. A prospective cross-sectional multi-state study is being conducted to Describe the prevalence, genotypic diversity, and antimicrobial susceptibility characteristics of Staphylococcus aureus isolates recovered from bulk tank milk from U.S. dairy farms and to identify herd characteristics and management factors associated with risk for presence of multidrug resistant MSSA and of MRSA on U.S. dairy farms. In winter and in summer 2016, bulk tank milk samples were collected from 188 dairy herds from 17 dairy states including. Approximately 150 S. aureus isolates were recovered from bulk tank milk for phenotypic characterization and whole genome sequencing, which was conducted in 2018. Molecular Evolutionary Genetics Analysis (MEGA) software was used to build a phylogenetic tree and to identify resistant isolates (MRSA & MSSA), presence of genes responsible for toxin production such as Staphylococcal enterotoxins (SE a to SE i), Toxic shock syndrome (TSST) in addition to the Panton Valentine Leukocidin (PVL) toxin gene. Data analysis will be completed in fall 2018 with reporting in 2019.

University of Missouri

Members: John Middleton, Pamela Adkins

Contributors: Veronique Bernier Gosselin

Our group has been working to understand the impact of increased heat and humidity on the microbial populations of the milk, feces, rumen, skin, and environment. Understanding the impact of increased heat and humidity on bacterial populations will greatly increase our understanding of the effects of heat stress on the dairy cow as a whole as well as the mammary gland specifically.

University of Tennessee

Members: Gina Pighetti, Oudessa Kerro Dego

Contributors: Reta Duguma Abdi, Desta Beyene Enserum, Susan Headrick, Leszek Wojakiewicz, Caitlin Merill (MS), Jacqueline Vaughn (MS), Tori Couture (MS candidate), Catherine Donnell (BS candidate), Megan O’Brien (visiting PhD scholar)

Regions in the bovine genome have been identified that are associated with phenotypes reflecting the strength of immune response to Streptococcus uberis infection and S. uberis concentrations in milk following intramammary challenge (TN-Pighetti). The first manuscript associated with this research was submitted and published (Siebert et al., 2018). The top candidate is currently being evaluated for its role in mammary epithelial cell defense mechanisms by an undergraduate honor’s student. The second manuscript is nearing completion. Overall, these regions have provided several novel candidate genes that are being examined for their role in mastitis resistance and their potential as novel targets for mastitis control.

Staphylococcus aureus is an important zoonotic mastitis pathogen that has significant effects on animal and human health. The objectives of this study were: 1) evaluate clonal diversity of S. aureus isolates from cases of bovine mastitis 2) determine staphylococcal enterotoxin production patterns 3) evaluate in vitro adhesion and invasion ability of dominant strains on bovine mammary epithelial cell line (MAC-T cells) (TN-Kerro Dego). Of the 111 S. aureus strains isolated, 16 PFGE types (dominant clones) were identified (A – P), with PFGE type M being the most prevalent. Staphylococcal enterotoxin genes seb (11.7%), sec (2.7%), see (0.9%) and/or toxic shock syndrome toxin 1 (tsst-1) (7.2%) were expressed by a subset of isolates, whereas most strains (75.7%) were negative for enterotoxin genes. PFGE types O and M tended to cluster with beta-hemolysin, absence of enterotoxins and susceptibility to antimicrobials. Analysis of in vitro adhesion to and invasion into MAC-T cells showed relatively higher number of O strain adhered to and invaded into MAC-T cells followed by M and I strains, however this differences were not statistically significant.

Utah State University

Member: David Wilson

Contributors: Kerry Rood, Justine Britten, Zhongde Wang, Jacqueline LaRose Kurz, E Jane Kelly

Additional analysis of genome-wide association studies (GWAS) comparing bovine nucleotide differences (SNPs) between cows repeatedly mastitic vs. cows continually free of mastitis was performed. Twenty-seven quantitative trait loci (QTLs) associated with mastitis resistance were identified including 10 novel QTLs; 4 QTLs were for teat length. One QTL includes a guanyl releasing protein gene, a candidate gene for mastitis resistance.

Vermont

Members: John Barlow, David Kerr, Feng-Qi Zhao

The Kerr lab used a primary dermal fibroblast as a model to rank animals based on composite expression of the toll-like receptor 4 gene (TLR4) and lipopolysaccharide (LPS)-induced IL-8 and IL-6 protein production, and then challenged the mammary glands of these animals with the P4 strain of E. coli to determine how difference in rank would affect response to mastitis. They found that high responder animals have an early upregulation in their innate response that is beneficial for bacterial clearance; however, they are equally susceptible to tissue damage caused by an exuberant response to the infection.

**OBJECTIVE 2:** Characterization and manipulation of virulence factors of mastitis pathogens for enhancing host defenses.

University of Maryland

Member: Kasey M. Moyes

Contributors: Daniel Nelson, John Roche, Theodore Elasser

In collaboration with Dr. Robert Peters, ANSC-UMD, and Mr. Dale Johnson, ANSC-UMD, we are currently following 4 New York dairy farms and to gather information regarding production outcomes, financial and lifestyle changes during their transition from conventional to automatic milking systems.

University of Minnesota

Members: S. Godden, E. Royster, L. Caixeta, B. Crooker, R. Fink, T. Schoenfuss

Contributors: L. Fox (WSU), P. Gorden (ISU), D. Nydam (Cornell University), S. Sreevatsan (MSU)

S. Godden, E. Royster, B. Crooker and S. Rowe. Investigation of the relationship between bedding characteristics and intramammary infection in late lactation dairy cows. The major objective of this study is to describe the relationship between bedding characteristics, bedding management and bedding bacteria counts (BBC) in different bedding materials. A secondary objective is to describe the relationship between bedding characteristics and herd-level measures of udder health for late lactation quarters and cows for specific pathogen groups of interest (e.g. Gram-negative organisms; Gram-positive organisms). Eighty herds were enrolled from 10 dairy states using one of four bedding types; new sand, reclaimed sand, manure solids or other organic materials. In summer, 2017 and winter 2018, sampling of each herd included collection of duplicate aseptic quarter milk samples from late lactation cows (> 180 DCC) as well as new and used bedding samples. Milk and bedding samples are being analyzed by bacterial culture. Additionally bedding samples will undergo testing to measure pH, organic matter and dry matter. Data analysis is ongoing with final results and reports expected in early 2019.

Patel, K., S. Godden, E. Royster, J. Timmerman, B. Crooker, and L. Fox. Investigation of the relationship between bacteria counts, bedding characteristics and bedding management practices with udder health and milk quality on dairy farms. The primary objective of this study is to conduct a multi-state, multi-herd cross-sectional observational study to describe the relationship between bedding bacteria counts and udder health and to identify goals (cutpoints) for interpreting BBC test results. A secondary objective is to identify bedding characteristics and bedding management strategies that are associated with lower BBC and improved udder health. One hundred-eighty eight herds were enrolled from 17 dairy states with the assistance of herd veterinarians or mastitis researchers. Herds used either new sand, reclaimed sand, manure solids or other organic bedding materials. New and used bedding samples, collected from the bedding storage area or from the back of stalls, respectively, and bulk tank milk samples were collected twice from each herd during summer and winter of 2016. Bedding samples were cultured and analyzed to measure pH, organic matter and dry matter. Herd level DHIA test day data describing udder health measures were recorded. Data analysis is ongoing with results expected to be reported in late 2018 and early 2019.

Godden, S., T. Schoenfus, R. Fink, J. Timmerman, C. Gebhart, E. Royster, S. Wells. Field validation of Matrix-Assisted Laser Desorption Ionization Time of Flight Mass Spectrometry (MALDI-ToF) for the identification of dairy microorganisms critical for safety and quality. The first objective is to develop and evaluate extraction methods to improve the MALDI-ToF method's ability to detect Mycoplasma bovis as well as individual species of Bacillus and Paenibacillus of importance to udder health, food safety and food quality. The second objective is to complete a field validation study using 2,500 bacterial isolates Report Date 02/17/2016 Page 1 of 3 Accession No. 233101 Project No. MIN-62-021 Multistate No. NE1048 derived from bovine mastitis samples and processed dairy foods. From this we will describe the diagnostic test characteristics of MALDI-ToF, as compared to 16S rDNA sequencing (gold standard) to identify 24 important dairy microorganisms critical to animal health, food safety and food quality. If the MALDI-ToF method proves to be accurate, then its adoption will strengthen the capacity, quality, and possibly the scope of diagnostic services performed by the VDL to support the MN dairy industry. Lab work and data analysis has been completed. A manuscript is in the final stages of preparation to be submitted in fall 2018.

University of Tennessee

Members: Gina Pighetti, Oudessa Kerro Dego

Contributors: Reta Duguma Abdi, Desta Beyene Enserum, Susan Headrick, Leszek Wojakiewicz, Caitlin Merill (MS), Jacqueline Vaughn (MS), Tori Couture (MS)

1) Assess the level of antimicrobial resistance by S. aureus isolates (n=239) collected in TN from 2004-2016. In total, 82 (34.3%) of the S. aureus isolates were found to be resistant at least to 1 of the 10 antimicrobials, whereas 157 (65.7%) of the 239 isolates were pan-susceptible. Overall, the prevalence of AMR among S. aureus varied from as low as 1.3% for ceftiofur to as high as 25% for sulphadimethoxine. The prevalence of AMR was widely distributed throughout the farms at a varying proportion for different antimicrobials tested. Out of all tested isolates, 25.5% were single drug resistant, 4.6% were double drug resistant, and 4.2% were multidrug resistant (resistant to more than three antimicrobials). Of the multidrug-resistant isolates, seven (70%) were also oxacillin-resistant isolates. This research was published earlier this year (Abdi et al., 2018).

2) Assess the level of antimicrobial resistance in isolates (n=197) collected with the Southeast Quality Milk Initiative (KY, MS, TN). The isolates evaluated were: 34% Staphylococcus aureus, 21.3% Streptococcus uberis, 18.3% Streptococcus dysgalactiae, 17.8% Escherichia coli, 6.6% Klebsiella pneumoniae and 2% Klebsiella oxytoca. Ceftiofur showed the highest efficacy on the most isolates followed by cephalothin, but K. pneumoniae isolates were resistant to most of the antimicrobials tested. Isolates from subclinical mastitis had higher MIC (P > 0.05) compared with isolates from clinical mastitis for some antimicrobials. The 197 isolates showed 32 different AMR patterns, which varied with farms and states. Compared with S. aureus and Str. dysgalactiae, E. coli and Klebsiella spp. had widespread resistance to pirlmycin and higher MIC for most of the drugs tested. These results suggested that confirmatory diagnosis and subsequent sensitivity testing would be a prerequisite to treat these mastitis pathogens effectively

3) Assess antimicrobial resistant bacteria and genes associated with dairy cattle production systems. From 4 TN dairies, 80 samples were collected from different farm environments: bulk tank milk, pooled feces from the floor (feces), soil from dairy cattle manure fertilized land, and soil from adjacent land with no access of dairy cattle were collected in 5 replicates. In general, E. coli , Staphylococcus spp., Enterococcus spp., and Salmonella spp. had different patterns in these environments indicating adaptation and micro-ecological niches of each farm. Assessment of bulk tank milk identified different levels of antimicrobial resistant bacterial species on each farm: tetracycline- or cefotaxime-resistant (n=6, farm A), tetracycline- or nalidixic acid-resistant (n=3, farm B), tetracycline- or cefotaxime- or nalidixic acid-resistant (n=2, farm C) and cefotaxime- and nalidixic acid-resistant (n=1, farm D). The presence of resistance to a third generation cephalosporins (cefotaxime) indicates the need for further evaluation of dairy cattle production system to identify the resistome reservoir and determine critical control points to prevent potential spread among humans, animals and environment.

Vermont

Members: John Barlow, David Kerr, Feng-Qi Zhao

The Barlow lab has developed a multilocus sequence typing scheme for S. chromogenes. A manuscript is in preparation describing the characteristics of this strain typing scheme. Briefly, 120 isolates were examined during development of this scheme. We find the majority of the strain types (STs, n=33, 72 %) are connected in a core network consisting only of single and double locus variants; these 33 STs account for 96 of the 120 isolates in the sample population. The network pattern is indicative of a recent clonal expansion with a single strain as a founder, consistent with recent epidemiological evidence that S. chromogenes is a host adapted species of cattle. Members from Washington (Fox) and Belgium (De Vliegher) collaborated in this project.

**OBJECTIVE 3:** Assessment and application of new technologies that advance mastitis control, milk quality, and dairy food safety.

Louisiana State University, Ag Center

Member: William E. Owens

Novel food grade products continue to be evaluated for antimicrobial activity. Novel products were evaluated using the AOAC in vitro test for germicidal activity. The products were also tested against mastitis pathogens in the MIC/MBC test.

Michigan State University

Member: Ronald Erskine, Lorraine Sordillo, Andres Contreras

Collaborators: Phil Durst, Ruben Martinez, Stan Moore

As part of a multistate USDA-NIFA funded project, we developed an on-farm evaluation system for milk quality and reduction of antibiotic use. The evaluation system (Quality Milk Alliance) assesses traditional practices related to mastitis control as well as a novel integration with social and communication barriers on the part of dairy producers and employees. We are developing novel applications for VaDia vacuum analysis as a tool to determine milking efficiency in dairy herds. These standards will be used to evaluate milking protocols and equipment performance and have the potential to provide an on-farm education platform for producers and/or employees regarding milking protocols.

Mississippi State University

Member: Amanda Stone

Collaborators: Mauricio Xavier

This year we worked to evaluate the effects of Imrestor, an immunomodulatory vaccine, on mastitis incidence and milk quality. This year was also the last year for the Southeast Quality Milk Initiative project funded by the USDA to advance milk quality improvements in the southeastern United States.

University of Maryland

Member: Kasey M. Moyes

Contributors: Daniel Nelson, John Roche, Theodore Elasser

In my laboratory at UMD, we are focused on the identification of new treatment strategies to reduce the use of antibiotics for treatment of bovine mastitis on farm to reduce the use of antibiotics commonly used to treat bovine mastitis.

University of Minnesota

Members: S. Godden, E. Royster, L. Caixeta, B. Crooker, R. Fink, T. Schoenfuss

Contributors: L. Fox (WSU), P. Gorden (ISU), D. Nydam (Cornell University), S. Sreevatsan (MSU)

S. Rowe, S. Godden, E. Royster, D. Nydam, A. Lago. Lockout study. Randomized equivalence study evaluating the efficacy of two commercial internal teat sealants in dairy cows. The objective of this study is to complete an equivalence study comparing the efficacy of LOCKOUT™ (Boehringer Ingelheim Vetmedica, Inc.) versus Orbeseal® (Zoetis) when administered in conjunction with a long acting antibiotic (Ab) at dry off in lactating dairy cows. Dry-off and enrollment of approximately 840 cows (Lockout = 420; Orbeseal = 420) from 5 commercial dairy herds in NY, WI, MN and CA was completed during summer 2018. Collection of post-calving samples and follow-up of lactation health and performance for the first 100 DIM will continue into late fall, 2018. Data analysis and reporting will be completed in 2019.

S. Rowe, S. Godden, E. Royster, D. Nydam, P. Gorden, A. Lago, R. Bicalho. Selective Dry Cow Therapy on US Dairy Farms: Impact on Udder Health, Antimicrobial Use and Economics. The major objective is to complete a multi-location noninferiority randomized clinical trial to evaluate the effect of applying 2 different SDCT programs, a culture-guided program and an algorithm-guided program (vs BDCT) on measures of quarter health, cow health and performance, antibiotic use and economics. Dry-off and enrollment of approximately 1,200 cows (400 per treatmentgroup) from 7 commercial dairy herds in NY, MN, IA and CA was completed during summer 2018. Collection of post-calving samples and follow-up of lactation health and performance for the first 120 DIM will continue into late fall, 2018. Data analysis and reporting will be completed in 2019.

Caixeta, L.S., Morley, P.S., Belk, K., Garry, F.B., Scheu, S., Parker, J, Noyes, N., Mijares, S., Hanes, A. Effect of selective dry cow therapy on the milk microbiome of dairy cattle with low somatic cell count. The purpose of this study was to determine the effects of SDCT on udder health and milk microbiome in separated milk cell pellets and cream in post-partum dairy cattle. 16S rRNA amplicon sequencing was performed and bioinformatics analysis of the sequence data is ongoing in order to identify differences between pellet and cream microbiomes for pre- and post-partum samples.

University of Missouri

Members: John Middleton, Pamela Adkins

Contributors: Veronique Bernier Gosselin

In the past year, our group has evaluated non-aureus staphylococcal (NAS) mastitis in dairy heifers and dairy goats. Results in heifers showed an association between skin colonization and intramammary infection (IMI) for some species of NAS and changes in NAS populations on heifer teat skin as animals age. In goats, we investigated the persistence of IMI with NAS during lactation and over the dry period. Results showed that IMI present at ≤ 10 days in milk (DIM) are more likely to persist in lactation than IMI that occur > 10 DIM. Some NAS species appear to be host-adapted in goats and NAS IMI can persist over the dry period. MALDI-TOF was validated for identifying NAS species isolated from goat’s milk. We are also currently working on a project in collaboration with the University of Kentucky to understand the association between housing environment and staphylococcal species isolated from bulk tank milk, teat skin, and bedding.

We are working to understand the effect of intramammary pirlimycin treatment on the fecal microbiome of lactating dairy heifers. This study intends to further the understanding of the broader impacts of intramammary antimicrobial usage and any unintended effects on the microbial ecology on the dairy farm.

University of Tennessee

Members: Gina Pighetti, Oudessa Kerro Dego

Contributors: Reta Duguma Abdi, Desta Beyene Enserum, Susan Headrick, Leszek Wojakiewicz, Caitlin Merill (MS), Jacqueline Vaughn (MS)

1) Assessing the potential of Staphylococcal surface proteins as vaccine candidates to minimize risk and severity of S. aureus-based mastitis. Strain variation and limited knowledge of common immunogenic antigens are among major constraints to developing an effective vaccine. Surface proteins from nine genetically distinct S. aureus isolates from bovine mastitis cases were extracted and evaluated for immune-reactivity. Multiple immuno-reactive surface proteins were common across the nine strains, several of which reacted strongly. across the nine strains tested. The conserved immune-reactive surface proteins may serve as potential candidates for a vaccine to control S. aureus mastitis in dairy cows.

2) Evaluate the immune responses and protection against S. aureus IMI in dairy cows vaccinated with Staphylococcus aureus surface proteins (SASP) and Staphylococcus chromogenes surface proteins (SCSP) during early dry period. Vaccinated cows had increased milk and serum antibody titers and reduced bacterial shedding of S. aureus in milk following challenge. Interestingly, SCSP vaccine cross-protected against S. aureus clinical mastitis thus suggesting its potential as immunogenic antigens to control bovine S. aureus mastitis.

3) Evaluate the protective effects of Staphylococcus chromogenes surface proteins (SCSP) as vaccine antigens to control mastitis during early lactation. Our preliminary results showed that three series of consecutive vaccinations of dairy cows with SCSP at drying off (D0), 21 days after drying off (D+21) and 40 days after drying off (D+40) induced increased immune responses during early lactation in vaccinated cows compared to control cows. The subsequent experimental challenge of vaccinated cows with the heterologous strain of S. aureus resulted in protection from clinical mastitis, reduced number of bacterial shedding in milk, and lower SCC in milk compared to control cows. Overall conclusion from results of early dry period and early lactation studies: The SCSP vaccine cross-protected vaccinated cows from S. aureus clinical mastitis, reduced number of bacterial shedding in milk and somatic cell counts showing its promising immunogenic potential to control mastitis in dairy cows.

Utah State University

Member: David Wilson

Contributors: Kerry Rood, Justine Britten, Zhongde Wang, Jacqueline LaRose Kurz, E Jane Kelly

Cows with a single high SCC quarter were treated IMM with casein hydrolysate (CH), non-hydrolyzed casein (NHC), or cessation of milking only (negative; N). Following treatment and involution of the mastitic quarter for remainder of lactation, decreases in cow-level SCC (-966,000/ml) and milk production (-14%) with 3 remaining lactating quarters were significant. Among treatments, decrease in cow-level SCC (-1,150,000/ml) for the CH group was significant. All treated quarters returned to milk production after calving; proportion of total-cow milk production (24%) was not different from pre-treatment or from 25% of total milk. After calving, treated quarters’ decrease in SCC was significant for CH (-2,763,000/ml) and N (-5,324,000/ml). Quarters with positive milk culture before treatment had 88% (14/16) bacterial cure (no isolation of the same bacteria for 3 weeks following calving).

Data analysis nearly complete for study of casein hydrolysate (CH) intramammary infusion for cessation of lactation in all 4 quarters at time of dry-off at the end of lactation. Udder halves were compared between control (dry cow antibiotic treatment plus teat sealant) with 4 treatment groups, each including CH, one being CH alone. All quarters of all cows showed no signs of clinical mastitis or cow discomfort and all returned to milk production following calving. All udder halves were not significantly different from 50% of total-cow milk production regardless of treatment group. Involution, SCC and bacteriological results between treatment groups are being analyzed.

Vermont

Members: John Barlow, David Kerr, Feng-Qi Zhao

The Barlow lab contributed to a study examining the transmission dynamics of Corynebacterium species causing persistent intramammary infections (IMI) in dairy herds. Using a mathematical model and empirical data from 2 dairy herds, we demonstrated that the rate of new infections was significantly related to preexisting IMI in both farms, underscoring the importance of preexisting Corynebacterium spp. IMI for the transmission of Corynebacterium spp. within groups of lactating dairy cattle. NE1748 members from New York (Schukken and Grohn) collaborated in this research.

**WORK PLANNED FOR THE COMING YEAR, LISTED BY OBJECTIVE:**

**OBJECTIVE 1**

LSU:

Organisms isolated from cases of mastitis from goats and cows will continue to be identified and tested for antimicrobial susceptibility.

University of Idaho:

We are collecting samples form periparturient cows (-28 d through +21 d, relative to expected parturition date), their dry off, close-up and lactation ration, and their calves (1-3d old) along with udder health, milk SCC and similar data from a large dairy unit (10000+ milking cows). One of the goals is to establish relations among mammary health measures, serum metabolites (lipid soluble vitamins, including vitamin A isomers), and calves passive immunity.

University of Maryland:

In my laboratory at UMD, we are currently fast beginning the use of new treatment strategies to reduce the use of antibiotics for treatment of bovine mastitis on farm to reduce the use of antibiotics commonly used to treat bovine mastitis. Furthermore, we are caught industry professionals regarding improving the immune response during a mastitis challenge in lactating dairy cows. We are currently collaborating with others internationally to investigate the role of neutrophils during the transition period. We are currently forwarding with others at the USDA to investigate the mammary gland response during dietary vitamin E E in lactating dairy cattle.

University of Minnesota:

K. Patel, S. Godden, S. Sreevatsan, E. Royster, L. Fox Molecular Epidemiology of Staphylococcus aureus on U.S. Dairy Farms. Data analysis will be completed in fall 2018 with reporting in 2019.

S. Rowe, S. Godden, E. Royster, D. Nydam, A. Lago. Lockout study. Randomized equivalence study evaluating the efficacy of two commercial internal teat sealants in dairy cows. Collection of post-calving samples and follow-up of lactation health and performance for the first 100 DIM will continue into late fall, 2018. Data analysis and reporting will be completed in 2019.

S. Rowe, S. Godden, E. Royster, D. Nydam, P. Gorden, A. Lago, R. Bicalho. Selective Dry Cow Therapy on US Dairy Farms: Impact on Udder Health, Antimicrobial Use and Economics. Collection of post-calving samples and follow-up of lactation health and performance for the first 120 DIM will continue into late fall, 2018. Data analysis and reporting will be completed in 2019.

S. Godden, E. Royster, B. Crooker and S. Rowe. Investigation of the relationship between bedding characteristics and intramammary infection in late lactation dairy cows. Data analysis is ongoing with final results and reports expected in early 2019.

Patel, K., S. Godden, E. Royster, J. Timmerman, B. Crooker, and L. Fox. Investigation of the relationship between bacteria counts, bedding characteristics and bedding management practices with udder health and milk quality on dairy farms. Data analysis is ongoing with results expected to be reported in late 2018 and early 2019.

Godden, S., T. Schoenfus, R. Fink, J. Timmerman, C. Gebhart, E. Royster, S. Wells. Field validation of Matrix-Assisted Laser Desorption Ionization Time of Flight Mass Spectrometry (MALDI-ToF) for the identification of dairy microorganisms critical for safety and quality. A manuscript is in the final stages of preparation to be submitted in fall 2018.

University of Missouri:

Over the next year, we plan to finish two projects related to the effects of heat and humidity on microbial populations of the dairy cow.

University of Tennessee:

Regions in the bovine genome have been identified that are associated with phenotypes reflecting the strength of immune response to Streptococcus uberis infection and S. uberis concentrations in milk following intramammary challenge. We will submit a manuscript evaluating novel phenotypes. Target the top 1-3 candidates and assess their role in mastitis resistance and potential as novel targets for mastitis control.

Utah:

One publication accepted, others submitted for publication regarding the genomic study described above.

Vermont:

The Zhao lab will investigate the effects of bacterial toxins [lipopolysaccharide (LPS) from E. coli and lipoteichoic (LTA) acid from S. aureus] and inflammatory cytokines (TNF-α, IL-1β, and IL-6) on i) apoptosis and viability and ii) milk protein gene expression in primary bovine mammary epithelial cells (bMECs). Primary bMECs have been isolated from five lactating cows. These cells will be treated with i) different doses of LPS or LTA and ii) different does of TNF-α, IL-1β, or IL-6 for various times. Next, we will treat the cells with the combinations of cytokines and bacterial toxins (LPS+TNF-α, LTA+TNF-α, LPS+IL-1β, LTA+IL-1β, LPS+IL-6, and LTA+IL-6) using the lowest effective dose of each cytokine and toxin. The viability of the cells will be analyzed using the Abcam Cell Viability Assay kit. The cell apoptosis rate will be determined by the terminal dUTP nick end-labeling (TUNEL) assay. mRNA expression of α-s1-, α-s2-, β- and κ- caseins, α-LA, and β-LB will be analyzed by qRT-PCR.

**OBJECTIVE 2**

Vermont:

The Barlow lab will initiate a study comparing milk quality and mastitis prevalence on organic dairy herds utilizing different bedding management practices. NE-1748 members from Minnesota (Godden), Missouri (Adkins), and Montreal (Dufour) will collaborate in this research. The Barlow lab will quantify potential associations between S. chromogenes strain types and intramammary infection phenotypes. We will use whole genome sequencing to characterize S. aureus and S. chromogenes strains with different in vitro phenotypes.

**OBJECTIVE 3**

LSU:

Plant based products will continue to be evaluated for antibacterial activity and tested for their potential activity as mastitis therapeutics.

University of Missouri:

In the coming year, work will continue to be focused on the epidemiology of staphylococcal mastitis in dairy cows. Several publications will be completed and submitted relative to our work on NAS mastitis in goats. We also plan to continue working on the effects of intramammary antimicrobials on fecal bacterial populations.

University of Tennessee:

1) Evaluate protective effects of Staphylococcus chromogenes surface proteins (SCSP) Vaccine against Staphylococcal and other mastitis pathogens in dairy cows through natural exposure 2) Identification and management of critical control points for antimicrobial resistant bacteria in dairy farms aimed at reducing urgent and serious level threat antibiotic resistant bacteria, 3) Evaluate the potential of a nutritional supplement to enhance the effectiveness of selective dry cow therapy when used in conjunction with an internal teat sealant.

Utah:

One publication, others in preparation regarding the casein hydrolysate studies described above.

**PUBLICATION LIST**

**Peer-Reviewed Literature**

Michigan State University:

Putman, A. K., J. L. Brown, J. C. Gandy, L. Wisnieski, and L. M. Sordillo. 2018. Changes in biomarkers of nutrient metabolism, inflammation, and oxidative stress in dairy cows during the transition into the early dry period. J Dairy Sci. 2018 Aug 1. pii: S0022-0302(18)30689-1. doi: 10.3168/jds.2018-14591. [Epub ahead of print]

University of Idaho:

Scholte, C.M., P. Rezamand\*, Z. M. Amiri, K.C. Ramsey, C. Tsai, and M.A. McGuire. 2017. The effects of elevated subcutaneous fat stores on fatty acid composition and gene expression of pro-inflammatory markers in periparturient dairy cows. J Dairy Sci 100:2104-2118.

University of Maryland:

Y. Qu, T. H. Elsasser, C.M. Scholte, M. Garcia and K.M. Moyes. 2018. The effects of feeding mixed tocopherol oil on whole-blood respiratory burst and neutrophil immunometabolic-related gene expression in lactating dairy cows. J. Dairy Sci. 101:4332-4342.

K.M. Moyes, L. Ma, D. Johnson, S. Fultz and R.R. Peters. 2017. Estimating and quantifying the production outcomes and lifestyle changes for small–to medium sized dairy farms regarding the transition from conventional to automatic milking systems in the Northeast region: A case study report. Biomedical Journal of Scientific and Technical Research. DOI: 10.26717/BJSTR.2017.01.000610.

K.M. Moyes, L. Ma, D. Johnson, S. Fultz and R.R. Peters. 2018. Estimating and quantifying the production outcomes and lifestyle changes for small–to medium sized dairy farms regarding the transition from conventional to automatic milking systems in the mid-Atlantic region: A case study report. In Natl. Mastitis Counc. Ann. Mtg. Proc., Glendale, AZ. Natl. Mastitis Counc., Inc., Madison, WI.

C.M. Scholte, D.C. Nelson, S. Linden, T.H. Elsasser, M. Garcia, Y. Qu and K.M Moyes. 2018. Short Communication: Recombinant bacteriophage endolysin, PlyC, is non-toxic and does not alter blood neutrophil response in lactating dairy cows. J. Dairy Sci. DOI: 10.3168/jds.2017-13908.

University of Minnesota:

Afifi, M., F. Kabera, H. Stryhn, J. Roy, L. Heider, S. Godden, W. Montelpare, J. Sanchez and S. Dufour. 2018. Antimicrobial-based dry cow therapy approaches for cure and prevention of intramammary infections: a protocol for a systematic review and meta-analysis. Animal Health Research Reviews. 19:74-48. https://doi.org/10.1017/S1466252318000051

Patel, K., S. Godden, E. Royster, J. Timmerman, B. Crooker. N. McDonald. 2017. Pilot Study: Impact of using a Culture-Guided Selective Dry Cow Therapy Program Targeting Quarter-Level Treatment on Udder Health and Antibiotic Use. Bov Pract. 51:48-57

Godden, S., E. Royster, J. Timmerman, P. Rapnicki and H. Green. 2017. Evaluation of an automated milk leukocyte differential test and the California mastitis test for detecting intramammary infection in early and late lactation quarters and cows. J Dairy Sci. 100:6527-6544.

University of Missouri:

Rainard P, Foucras G, Fitzgerald JR, Watts JL, Koop G, Middleton JR. 2018. Knowledge gaps and research priorities in Staphylococcus aureus mastitis control. Transbound Emerg Dis. 65(Suppl 1):149-165.

Cameron M, Perry J, Middleton JR, Chaffer M, Lewis PJ, Keefe G. 2018. Short Communication: Evaluation of MALDI-TOF mass spectrometry and a custom reference spectra expanded database for identification of bovine-associated coagulase-negative staphylococci. J Dairy Sci. 101(1):590-595.

Adkins PRF1, Dufour S, Spain J, Calcutt M, Reilly T, Stewart G, Middleton JR\*. 2018. Cross-sectional study to identify staphylococcal species isolated from teat and inguinal skin of different aged dairy heifers. J Dairy Sci. 101(4):3213-3225.

Adkins PRF1, Dufour S, Spain J, Calcutt M, Reilly T, Stewart G, Middleton JR\*. 2018. Molecular characterization of non-aureus Staphylococcus spp. from heifer intramammary infections and body sites. J Dairy Sci. 101(6):5388-5403.

Bernier Gosselin V1,4, Lovstad J4, Dufour S, Adkins PRF, Middleton JR\*. 2018. Use of MALDI-TOF to characterize staphylococcal intramammary infections in dairy goats. J Dairy Sci. 101(7):6262-6270.

Bernier Gosselin V1, Dufour S, Zhang M, Middleton JR\*. 2018. Sensitivity and specificity of a competitive ELISA using frozen-thawed milk or serum for the diagnosis of small ruminant lentivirus infection in goats using a Bayesian latent class model. Small Ruminant Research. [Epub ahead of print 6 Aug 18] https://doi.org/10.1016/j.smallrumres.2018.08.004

Adkins PRF, Middleton JR. 2018. Methods for diagnosing mastitis. Vet Clin North Am Food Anim Pract. Invited article. In Press.

University of Tennessee:

Abdi R. D., B. E. Gillespie, J. Vaughn, C. Merrill, S. I. Headrick, D. Ensermu, D. H. D’Souza, R. A. Almeida, S. P. Oliver, Getahun E. Agga and O. Kerro Dego. 2018. Antimicrobial Resistance of Staphylococcus aureus Isolates from Dairy Cows and Genetic Diversity of Resistant Isolates. Foodborne Pathogens and Disease, 15(7): https://doi.org/10.1089/fpd.2017.2362.

Almeida RA, O. Kerro Dego, and A. G. Rius. Effect of heat stress on adherence to and internalization of Streptococcus uberis into bovine mammary epithelial cells. 2018. J Dairy Res, 85: 53–56. https://doi.org/10.1017/S0022029917000875.

Couture, V.L., GM Pighetti, LG Schneider, RA Almeida, SP Oliver, PD Krawczel. Behavioral response to the clinical and subclinical effects of a Streptococcus uberis mastitis challenge in Holstein dairy cows. J Dairy Sci (submitted, 8/31/2018)

Kull JA, PD Krawczel, and GM Pighetti. 2018. Short communication: Evaluation of an automated method for assessing white blood cell concentration in Holstein dairy cows. Vet Immunol Immunopathol. 197:21-23. doi: 10.1016/j.vetimm.2018.01.002

Oudessa Kerro Dego, R. A. Almeida, Maria E. Prado, S. I. Headrick, M. J. Lewis, C. Young, B. E. Gillespie, L. J. Siebert, G. M. Pighetti, R. D. Abdi, D. B. Ensermu and S. P. Oliver. 2017. Functionally effective immune responses of dairy cows vaccinated with Streptococcus uberis adhesion molecule during the transition period. Ann Vaccines Immunization 3(1): 1015.

Siebert L, ME Staton, S Headrick, M Lewis, B Gillespie, C Young, RA Almeida, SP Oliver, and GM Pighetti. 2018. Genome wide association study identifies loci associated with milk leukocyte phenotypes following experimental challenge with Streptococcus uberis. Immunogenetics 70(9):553-562. doi: 10.1007/s00251-018-1065-3

Utah:

Britten JE, Rood KA, Wilson DJ: Intramammary infusion of casein hydrolysate for involution of single mastitic mammary quarters elevating cow-level somatic cell count. Adv Dairy Res 6:3:1-6, 2018.

**Abstracts**

Mississippi State University:

Xavier, M.X.S., D.D. McGee, J.A. Brett, A.E. Stone. Evaluation of mammary and uterine health and production parameters of dairy cows treated with pegbovigrastim. 2018. American Dairy Science Association Discover Conference. Chicago, IL.

Stone, A., B. Bowman, M. Denny. On-farm bacteriologic milk culturing: Producer perception and decision impact. 2018. American Dairy Science Association Annual Meeting. Knoxville, TN.

Bowman, B.L., M.D. Denny, A.E. Stone. Adoption of on-farm bacteriologic milk culturing: Evaluating farmer perception and impact on management decisions. 2018. National Mastitis Council Annual Meeting. Tucson, AZ.

Enger, K., C. Petersson-Wolfe, R. A. Almeida, D. T. Nolan, P. D. Krawczel, J. Bewley, A. E. Stone, S. H. Ward, S. P. Oliver, and G. M. Pighetti. 2018. Microorganisms isolated from subclinical intramammary infections present in dairy cattle from the southeast United States. #T201. J Dairy Sci Vol 101 Suppl 2. p285.

University of Idaho:

C. Y. Tsai, F. Rosa, M. Bionaz, and P. Rezamand. 2018. Effects of 2, 4- thiazolidinedione on milk fatty acid profile and vitamins in dairy goats with subclinical mastitis. In: Proceedings of the 51 st Annual Pacific Northwest Animal Nutrition Conference: p. 7.

University of Maryland:

Y. Qu, T.H. Elsasser, S. Kahl, M. Garcia, C.M. Scholte, E.E. Conner, G.F. Schroeder

and K.M. Moyes. 2017. The determination of concentrations of tocopherol isoforms in whole tissues and mitochondria via high-performance liquid chromatography after short-term supplementation in dairy cows. J. Dairy Sci. 100(Suppl 1):1.

M.A. Crookenden, C.G. Walker, A. Heiser, J.J. Loor, K.M. Moyes, J.K. Kay, S. Meier,

A. Murray, V.S.R. Dukkipati, M. Mitchell and J.R. Roche. 2016. miRNAseq from whole blood over the transition period. J. Dairy Sci. 99(Suppl 1):85.

Cynthia M. Scholte, T.H. Elsasser, S. Kahl, D.C. Nelson, D. Biswas and K.M. Moyes.

2018. Effects of citrus oil fractions on Escherichia coli P4 growth and on bovine neutrophils. American Dairy Science Association (ADSA) Annual Meetings, Knoxville, TN.

University of Minnesota:

Scheu S., Huebner, K., Parker, J., Mijares, S., Hanes, A., Garry, F., Belk, K., Morley, P., Caixeta, L.S. “Effect of selective dry cow therapy on udder health and milk microbiome in dairy cattle with low somatic cell count.” 51st Annual Conference of the American Association of Bovine Practitioners, 2018.

K. Patel, S. Godden, E. Royster, J. Timmermen, B. Crooker and L. Fox. 2018. Investigation of the relationship between bacteria counts, bedding characteristics and bedding management practices with udder health on dairy farms using manure solids bedding: preliminary results. 51st American Association of Bovine Practitioners Annual conference, Phoenix, AZ, USA. September 12-15, 2018.

K. Patel, S. Godden, E. Royster, J. Timmermen, B. Crooker and L. Fox. 2017. Investigation of the relationship between bacteria counts, bedding characteristics and bedding management practices with udder health on U.S. dairy farms: preliminary results.” Mastitis Research Workers Conference, Chicago, IL, USA. November 1 - 3, 2017.

K. Patel, S. Godden, E. Royster, J. Timmermen, B. Crooker and L. Fox. 2017. Investigation of the relationship between bacteria counts, bedding characteristics and bedding management practices with udder health and milk quality on dairy farms: preliminary results. Points of Pride Research Day. College of Veterinary Medicine, University of Minnesota, Saint Paul, MN. October 4, 2017.

Rowe, S. Investigation of the relationship between bedding bacteria count and intramammary infection in late lactation dairy cows. American Association of Bovine Practitioners, Phoenix, AZ, September 2018

Rowe, S. Investigation of the relationships between towel laundering practices, towel bacteria count and intramammary infection in late lactation dairy cows. American Association of Bovine Practitioners, Phoenix, AZ, September 2018

Rowe, S. Investigation of the relationship between bedding materials, bedding characteristics, and intramammary infection in late lactation dairy cows. Mastitis Research Workers Conference, Chicago, IL, November 2017

University of Missouri:

McKinnon E, Bernier Gosselin V, Royal A, Rindt H, Middleton JR, Adkins PRF. 2018. Evaluation of milk leukocyte populations in relation to caprine arthritis-encephalitis virus in dairy goats. Proceedings of the 41st Annual CVM Research Day (Phi Zeta). 25 May 2018. Abstract #23.

Placheta L, Middleton JR (Co-Sponsor), Borchers M, Bewley J, Adkins P (Co-Sponsor). 2018. The effect of housing environment on teat skin staphylococcal populations. Proceedings of the 41st Annual CVM Research Day (Phi Zeta). 25 May 2018. Abstract #16.

Bernier Gosselin V, Middleton JR (Sponsor), Dufour S. 2018. Infection dynamics of staphylococcal intramammary infections in dairy goats. Proceedings of the 41st Annual CVM Research Day (Phi Zeta). 25 May 2018. Abstract #36.

Schmidt R, Ierardi R, Placheta L, Adkins PRF, Middleton JR. 2018. Epidemiol0gy of mastitis pathogens on Amish Dairy Farms in Missouri. National Veterinary Scholars Symposium. Texas A&M University, College Station, Texas. August 2-5, 2018.

Wattenburger K, Schmidt R, Placheta L, Middleton JR, Adkins PRF. 2018. Evaluation of aseptic techniques used to collect bovine milk samples. National Veterinary Scholars Symposium. Texas A&M University, College Station, Texas. August 2-5, 2018.

University of Tennessee:

Abdi R. D., D. Ensermu, J. Vaughn, C. Merrill, B. Gillespie, O. Kerro Dego. Staphylococcus aureus cell surface proteins extraction and evaluation of immunogenicity. The 98th Annual Conference of Research Workers in Animal Diseases 2017 in Chicago, Illinois, p81, December 3 – 5. http://crwad.org/wp-content/uploads/2017/11/CRWAD-2017-Author-Index-and-Abstracts.pdf

Abdi R. D., B. E. Gillespie, S. I. Headrick, G. M. Pighetti, R. A. Almeida, S. P. Oliver, and O. Kerro Dego. Antimicrobial resistance patterns of bacterial isolates from cases of mastitis in dairy cows. American Dairy Science Association Annual Meeting, June 24 – 27, 2018, Knoxville, TN.

Kerro Dego O., R. D. Abdi and R. A. Almeida. Experimental S. aureus mastitis teat-dip Infection model for evaluation of efficacy of vaccine against S. aureus IMI. American Dairy Science Association Annual Meeting, June 24 – 27, 2018, Knoxville, TN.

Utah:

Britten JE, Wilson DJ, Rood KA: Intramammary casein hydrolysate alone or combined with other treatments when drying off dairy cows. J Dy SciVol. 101, E-Suppl. 2:177, 2018.

Vermont:

Korkmaz FT, Elsasser TH, Kerr DE. 2018. Variation in fibroblast expression of toll-like receptor 4 and lipopolysaccharide-induced cytokine production between animals predicts control of bacterial growth but not severity of Escherichia coli mastitis. J Dairy Sci. Aug 29. pii: S0022-0302(18)30784-7. doi: 10.3168/jds.2017-14372. [Epub ahead of print] PMID: 30172411.

Dalen G, Rachah A, Nørstebø H, Schukken YH, Gröhn YT, Barlow JW, Reksen O. 2018. Transmission dynamics of intramammary infections caused by Corynebacterium species. J Dairy Sci. 101(1):472-479. doi: 10.3168/jds.2017-13162.

**Conference Proceedings**

Michigan State University:

Erskine, R.J. Dumpster Diving, Somatic Cell Counts and Antimicrobials: What Do They Have in Common? Seminar presented to the Annual Meeting of the Academy of Dairy Veterinary Consultants. Reno, NV, October, 2017.

Erskine, R.J. Selective Dry Cow Therapy: It’s Complicated. DAIReXNET Webinar, October 18, 2017.

Erskine, R. J. Random Thoughts on Parlor Efficiency. Seminar presented to the North Florida/South Georgia Dairy Management Meeting, Quitman, GA, November 16, 2017.

Erskine, R. J. Barn Yard Pharmacology and Milking Efficiency. Discussions and workshops presented to the Dairy Production Medicine Certificate Program for Veterinarians, Ontario Veterinary College, Guelph, ONT, December 5-6, 2017.

Erskine, R. J. BLV: The Silent Thief Seminar presented at the Michigan Veterinary Medical Conference, Lansing, MI, January, 2018

Erskine, R. J., M. Borek-Stine, R. Moore-Foster. Engaged Employees: The Key to Quality Milk. Shortcourse presented at the 58th Annual Meeting of the National Mastitis Council, Tucson, AZ, February 2, 2018.

Erskine, R. J. Bi-modal Milk Ejection and Milk Yield-Is There a Connection? Seminar presented at the Meeting of the National Mastitis Council, Milan, Italy, June 12, 2018.

Erskine, R. J. and D. Thompson. The Cows Speak to Us About Milking- Are You Listening? Shortcourse presented at the Regional Meeting of the National Mastitis Council, Guelph, ONT, June 20, 2018.

Erskine, R. J. Selective Dry Therapy-A Good Tool For The Mastitis Toolbox, If…….. Seminar presented at the Regional Meeting of the National Mastitis Council, Guelph, ONT, June 21, 2018.

University of Maryland:

Invited: C.M Scholte#. 2017. LACTATION BIOLOGY SYMPOSIUM: Alternatives to antibiotic treatment for bovine mastitis. American Animal Science Association Annual Meeting, July 8-12, Baltimore, MD.

University of Minnesota:

S. Godden, K. Patel, E. Royster, J. Timmerman, B. Crooker and L. Fox. 2018. Relationships between bedding bacteria counts, bedding characteristics and udder health: We have a lot more to learn. 2018 International Bovine Mastitis Conference. Milano, Italy. June 11-13, 2018.

S. Godden, K. Patel, E. Royster, J. Timmerman, B. Crooker and L. Fox. 2018. Relationships between bedding bacteria counts, bedding characteristics and udder health. National Mastitis Council Regional Meeting. Guelph Ont. Canada. June 20-22, 2018. Pp. 18-27.

University of Missouri:

Bernier Gosselin V, Adkins PRF, Middleton JR. 2018. Use of strain-typing of staphylococci to determine persistence of intramammary infection in dairy goats. Proceedings of the 57th Annual Meeting of the National Mastitis Council, Tucson, AZ.

Placheta L, Middleton JR, Borchers M, Bewley JM, Adkins PRF. 2018. Association of housing environment and staphylococcal species isolated from teat skin of dairy cattle. Proceedings of the 57th Annual Meeting of the National Mastitis Council, Tucson, AZ.

University of Tennessee:

Abdi, R. D., J. Vaughn, C. Merrill, S. M. Cantwell, B. E. Gillespie, R. A. Almeida, S. I. Headrick, G. M. Pighetti, P. Krawczel, J. Keflot, J. M. Bewley, S. P. Oliver and O. Kerro Dego. 2017. Antimicrobial Resistance Patterns of Staphylococcus aureus Isolates from Cases of Bovine Mastitis. Proceedings of National Mastitis Council, p 60, 56th meeting January 28 – 31, 2017, Trade winds Island Grand Resort, St. Pete Beach, Fl.

Utah:

Wilson DJ: J5 mastitis vaccinates’ milk production change and reproductive performance following clinical mastitis. Proc 10th World Congress of Vaccine: 45, 2017.

Britten JE, Wilson DJ, Rood KA: Casein hydrolysate as a possible adjunct to or replacement treatment for current antibiotic therapies used at dry-off in dairy cows. Proc 57th Ann Mtg Natl Mast Council:238-239, 2018.

Britten JE, Wilson DJ, Rood KA: Effect of casein hydrolysate on bovine mammary involution: changes to milk composition and preliminary histological evaluation. Proc Animal, Dairy and Veterinary Sciences Student Research Symposium:35, 2018.

**Poster Presentations**

University of Idaho:

C. Y. Tsai, F. Rosa, M. Bionaz, and P. Rezamand. 2018. Effects of 2, 4- thiazolidinedione on milk fatty acid profile and vitamins in dairy goats with subclinical mastitis. In: Proceedings of the 51 st Annual Pacific Northwest Animal Nutrition Conference: graduate poster competition (January 2018, Boise, Idaho).

University of Minnesota:

Rowe, S. Selective dry cow therapy on U.S. dairy farms: impact on udder health, antimicrobial use and economics. Minnesota Dairy Health Conference, Saint Paul, MN, April,18-19, 2018.

University of Tennessee:

Abdi R. D., J. R. Dunlap, D. B. Ensermu and O. Kerro Dego. Staphylococcus aureus surface proteins extraction methods with immunoproteomics and electron microscopic study. American Dairy Science Association Annual Meeting, June 24 – 27, 2018, Knoxville, TN.

Merrill C., D. B. Ensermu, R. D. Abdi, B. E. Gillespie, J. Vaughn, S. I. Headrick, K. Hash, T. B. Walker and O. Kerro Dego. Protective effects of staphylococcal surface proteins as vaccine antigens to control mastitis in Dairy cows. American Dairy Science Association Annual Meeting, June 24 – 27, 2018, Knoxville, TN.

Merrill C., Desta B. Ensermu, Reta Duguma Abdi, Barbara E. Gillespie, Jacqueline Vaughn, Raul A. Almeida, S. P. Oliver, Susan I. Headrick, Kody Hash, Tate Bradley Walker and O. Kerro Dego. Evaluation of the immune response and protection in dairy cows vaccinated with immunodominant Staphylococcal surface proteins. The 98th Annual Conference of Research Workers in Animal Diseases, p187, December 1-5, 2017, Chicago Marriott, Downtown Magnificent Mile, Chicago, Illinois. http://crwad.org/wp-content/uploads/2017/11/CRWAD-2017-PRESENTATION-SCHEDULE.pdf

Merrill C., D. B. Ensermu, R. D. Abdi, B. E. Gillespie, J. Vaughn, R. A. Almeida, S. P. Oliver, S. I. Headrick, K. Hash, T. B. Walker and O. Kerro Dego. Protective Effects of a new Staphylococcal vaccine against mastitis in dairy cows. UT Beef and Forage Center annual Meeting, Dec 19, 2017 at the University of Tennessee, Knoxville, TN.

Vaughn J., R. D. Abdi, B. E. Gillespie, C. Merrill, D. B. Ensermu and O. Kerro Dego. Genetic Diversity and Associated Enterotoxin Production Patterns of S. aureus Isolates from Cases of Bovine Mastitis. American Dairy Science Association Annual Meeting, June 24 – 27, 2018, Knoxville, TN.

Vaughn J., R. D. Abdi1, B. E. Gillespie1, C. Merrill1 and O. Kerro Dego. Genetic Diversity of Staphylococcus aureus from Cases of Bovine Mastitis. UT Beef and Forage Center annual Meeting, Dec 19, 2017 at the University of Tennessee, Knoxville, TN.

Vaughn J., R. Duguma Abdi, B. Gillespie, C. Merrill, O. Kerro Dego. 2017. Genetic diversity and enterotoxin production profiles of Staphylococcus aureus strains from cases of bovine mastitis. The 98th Annual Conference of Research Workers in Animal Diseases, P144, December 1-5, 2017, Chicago Marriott, Downtown Magnificent Mile, Chicago, Illinois.http://crwad.org/wp-content/uploads/2017/11/CRWAD-2017-Author-Index-Abstracts.pdf

**Web articles**

Michigan State University:

Launstein E., and R. J. Erskine. Revisited article: You are here on the somatic cell count map

http://qualitymilkalliance.com/2018/07/25/revisited-article-you-are-here-on-the-somatic-cell-count-map/

Erskine, R. J. How is Your Milking Efficiency? Part 1.

http://qualitymilkalliance.com/2018/05/01/how-is-your-milking-efficiency-part-1/

**Mississippi State University:**

Becker, C. and A. Stone. 2018. Dry cow therapy: Aseptic infusion. Mississippi State University Extension Factsheet. P3291.

Becker, C. and A. Stone. 2018. Dry cow therapy: Choosing the best protocol for your dairy. Mississippi State University Extension Factsheet. P3290.

**Public outreach**

Pighetti, GM. High quality milk: Farm to table. Knoxville Academy of Nutrition and Dietetics, 9/26/2018, 24 attendees.