Research Focus

By Heather Huson

Genomic research and selection improve cow health

Genomic selection, using modern techniques to evaluate DNA, has been part of the US dairy industry since 2008. Use of genetic variants for marker assisted selection has improved accuracy and led to a faster rate of gain in production, health, and conformation traits worldwide. Genomic research is an integral part of this system because it identifies new and improved markers for current traits, as well as markers for novel traits. Research projects within our laboratory focus on the genetic regulation of mastitis and lameness, two of the most costly diseases in today's dairy industry. The core of our research relies on associating phenotypic characteristics of lameness or mastitis with genomic markers in an analysis known as a genome-wide association study (GWAS). A GWAS is particularly powerful to identify genetic variants that effect complex traits, including mastitis and lameness, that are influenced by multiple genes and have a greater degree of environmental regulation. In turn, genomic evaluations, using research findings, are very effective at selecting complex traits that can be more difficult to characterize and track efficiently on farm. Figure 1 shows the typical workflow of our research projects and end application to farm management. Our lab has identified some of the preliminary results of the phenotypic or trait characterization recorded for subsequent genetic analysis. Our goal in both projects is to improve our understanding of the biological pathways involved in these traits and to generate a genetic marker panel to use in genomic evaluations for selection of optimally healthy and high producing dairy cattle.

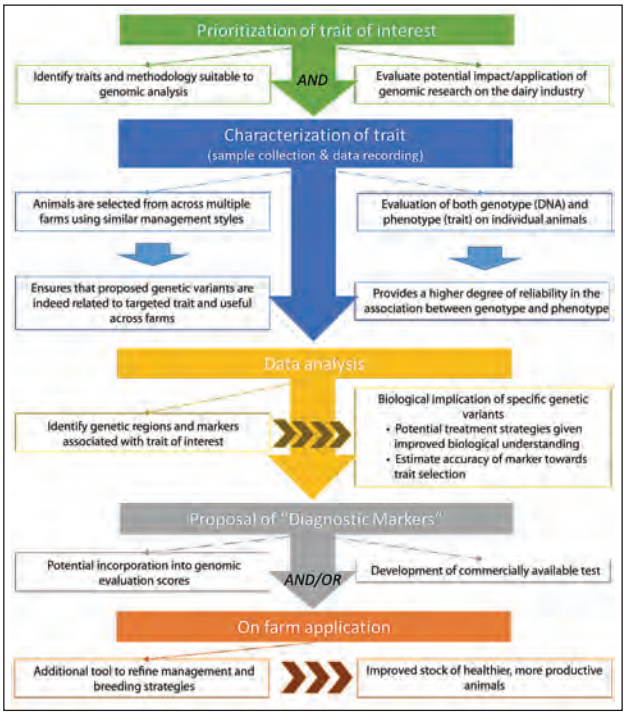
Projects focus on genetic regulation of mastitis and lameness, some of the most costly diseases in the dairy industry.

Selecting for mastitis resistant cows through the integration of teat and udder conformation, somatic cell count, and milk microbiome sequencing data – *Asha Miles.*

Somatic cell score (SCS) was added to the evaluations in 1994 to address the occurrence of mastitis infection within the US dairy cattle population, noting the impact of this disease on health and production. However, mastitis is a complex trait attributed to genetic and environmental effects - thus SCS alone does not inform mastitis susceptibility or resistance. To address this deficiency, we are investigating the genetic mechanisms underlying mastitis. We have enrolled 500 dairy cows across two commercial farms in Upstate New York. In the first phase we phenotyped all cows by characterizing their teats and udders, and taking milk samples for somatic cell count and microbiome analysis at six key physiological time points in lactation (Fig. 2). A blood sample was taken from each cow to extract genomic DNA and genetically characterize, referred to as genotyping, each individual for 777k single-nucleotide polymorphisms (SNP) spanning the entire bovine genome.

Beginning with an epidemiological analysis of this data, we are looking for associations between the physical traits an animal portrays and the occurrence of subclinical mastitis (linear score > 4) and clinical mastitis (farmer-diagnosed). We observed that prevalence of subclinical mastitis varies greatly by the physical trait an animal has, and also by parity and the stage of lactation (Fig. 3). Further, by plotting linear scores at different time points against each other, we can identify populations of cows that have chronic mastitis problems or populations of consistently healthy cows (Fig. 4). This will be particularly useful for the genetics research phase where we compare discrete populations of cows and identify genetic variants that are likely responsible for these differences.

Figure 1.



Once all individuals are genotyped, we will perform a genome wide association study (GWAS), to identify genetic markers associated with clinical mastitis, subclinical mastitis, and consistently healthy cows. Using the method demonstrated in Figure 4, we will designate cows that remain in the lower left quadrant for every sampling period as consistently healthy, and cows that remain in the upper right quadrant for every sampling period as having chronic mastitis problems. Complex traits like mastitis susceptibility or resistance are best

Figure 2. Proposed sampling periods based on key physiological transitions in the dairy cow.

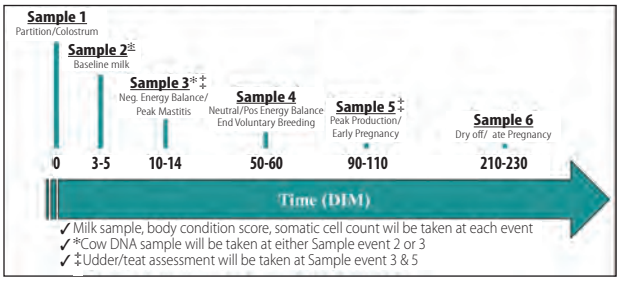
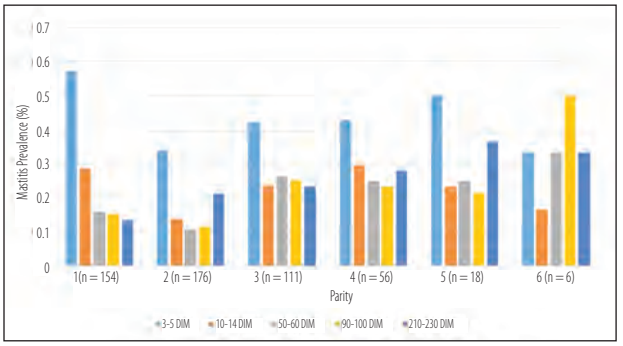


Figure 3: Mastitis prevalence (new and existing cases over the total population at risk) across five sampling time points, stratified by parity.



investigated by a GWAS because they are designed to distinguish common mutations carried by a large number of unrelated individuals. This enables us to identify genetic variants likely responsible for complex traits despite the influence of multi-gene traits, gene interactions, and the environment. In addition, metagenomic (milk bacteria) DNA will be extracted and sequenced from each milk sample to develop a milk microbiome profile for each animal. A similar GWAS can be performed to determine whether a cow is genetically predisposed to populations of healthy commensals colonizing the udder or if they are genetically susceptible to certain pathogens.

The development of genetic profiles associated with mastitis susceptibility or resistance will assist in the selection of optimally producing dairy cows. The diagnostic markers identified from this research will be proposed to the Council on Dairy Cattle Breeding and USDA dairy cattle research scientists for evaluation and introduction into the genetic marker panel used for genomic evaluations of US Holsteins.

Reducing lameness by optimizing digital cushion thickness – *Cassandra Stambuk.*

Lameness is the presentation of abnormal locomotion regardless of cause. It is the manifestation of disease(s), not a disease itself. Lameness can be due to environment, diseases, management practices, biomechanics, and even genetics. It is hard to improve because multiple issues can contribute to its presence. For example, lameness may have a high prevalence on a farm, but specific diseases such as dermatitis or white line disease will have a low prevalence. The best way to reduce lameness is to find a way to decrease the influence of multiple diseases at once. Previous research in Dr. Rodrigo Bicalho's laboratory at Cornell's Veterinary College showed that the thickness of the digital cushion, a fat pad between the bone and the hoof wall, is

Figure 4. Categorization of cows as New Cases (o), Chronic (Δ), Healthy (□), and Cures (*), based on linear score of somatic cell count comparing 3-5 DIM (x-axis) and 10-14 DIM (y-axis).

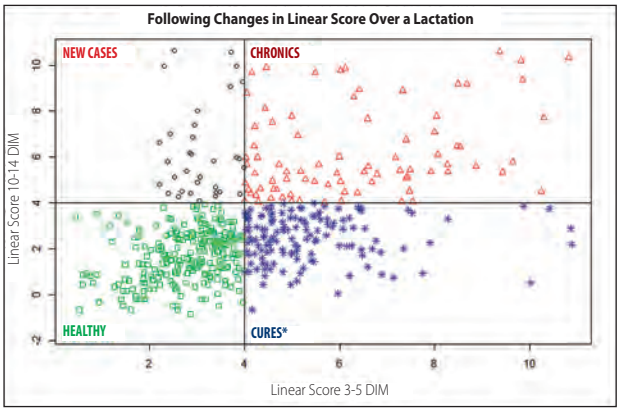
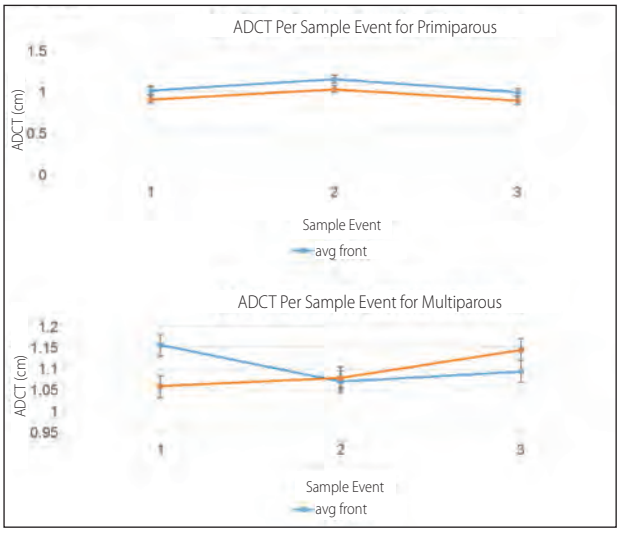


Figure 5. Graphs comparing the average digital cushion thickness (ADCT) of primiparous and multiparous animals separately across the first three sample events. Avg front = ADCT front hoof; Avg hind = ADCT hind hoof.



associated with lameness, particularly sole ulcers and white line disease, in Holstein dairy cattle. Therefore, investigating the genetics that contribute to digital cushion thickness in the bovine hoof is a novel approach to reduce lameness by combating multiple diseases.

Our laboratory split the research into two phases. Phase one is an intensive longitudinal study, which to our knowledge has never been done. We are following about 200 Holstein cows through one lactation, and sampling the digital cushion thickness at four time points. From this data, and the characterization of the digital cushion, we are choosing two time points to use in phase two. In phase two, we will add about 400 more cows, including 300 Jersey cows. In both phases we will record other traits, such as height, body condition score, locomotion, lesion, and production (Table 1). The cows are on commercial farms in Upstate New York.

Some of the trends we identified in the preliminary data agree with previous research. For example, multilevel modeling shows a significant difference between the digital cushion thickness in samples one and three of animals in parity = 1 to those of animals in

Please turn to page 42

Dietary mineral sources and feeding rates in the transition period
continued from page 32

Consistent with higher intake, cows fed MIN-AD had lower blood non-esterified fatty acid (NEFA) concentrations prepartum and tended to have lower NEFA concentrations postpartum.

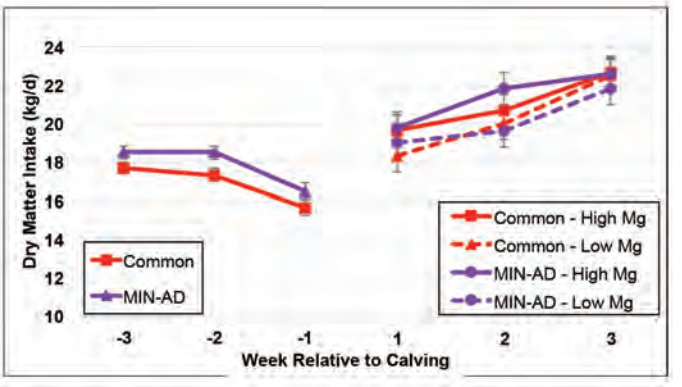
Aside from inclusion in diets as a mineral source, MIN-AD is also incorporated into diets as a buffer. Postpartum, this additional buffer could be aiding in rumen health as cows transition onto higher starch fresh cow diets. While rumen pH fluxuations aren't typically thought of as an issue in the prepartum period in high forage, low starch diets, there may be some unknown benefits of supplementary buffers in the prefresh diet. Additionally, dolomite minerals, the chemical structure of the calcium and magnesium complexed with carbonate in MIN-AD, have previously been shown to increase passage rate, which could influence intake in the transition period.

The take home message. While varying dietary source of calcium and magnesium, and level of magnesium postpartum, did not influence plasma calcium status in this trial, the exceptionally healthy study population may have masked potential effects of the treatment diets. Some important take home points from this trial include:

• Lower blood magnesium in this trial did not impair calcium status, intake or performance suggesting that blood magnesium concentrations in fresh cows should be interpreted cautiously.

• Feeding MIN-AD improved dry matter intake and blood NEFA concentrations, warranting further investigation into strategic use of specific dietary mineral sources in transition cow diets. ❐

Figure 3. Dry matter intake in the prepartum period was higher for cows fed Ca and Mg primarily from MIN-AD as opposed to common sources.



Brittany Leno (bms279@cornell.edu) is a graduate student in the Cornell University Department of Animal Science.

Genomic research and selection improve cow health
continued from page 35

parity > 1. Previously, it was noted that the digital cushion of primiparous cows are different than those of multiparous cows. Additionally, the digital cushion thickness of primiparous cows seem to follow a more distinct change in digital cushion across time. On the other hand, multiparous cows show more variation with their digital cushion thickness (Fig. 5). This could be due to differing fatty acid composition between heifers and cows.

Once both phases are completed, the final step is to compare the trait data, primarily digital cushion thickness, to over 777K genetic variants spanning the genome. By looking at the whole genome we hope to determine possible genetic markers associated with digital cushion thickness. Characterizing the digital cushion in Holsteins and Jerseys allows us to compare genetic variants within and across breeds. Once these are determined, results will be proposed to breed associations, industry, and the Council for Dairy Cattle Breeding, to potentially incorporate into genetic evaluations. Characterizations of legs and hooves are not well represented in genetic evaluations and thus far have shown little impact towards decreasing lameness. Lameness research needs growth and attention to improve animal well-being and to reduce economic loss to farmers. ❐

Table 1. Traits recorded per sample event

	Sample Event 1 245-266 DCC	Sample Event 2 1-30 DIM	Sample Event 3 91-120 DIM	Sample Event 4 >270 DIM
Blood	X			
BCS	X	X	X	X
Lesion	X	X	X	X
Locomotion	X	X	X	X
Ultrasound	X	X	X	X
Height	X			X

Heather Huson (hjh3@cornell.edu), is the Robert & Anne Everett Professor of Dairy Cattle Genetics with the Cornell University Department of Animal Science.

Starch and fiber in fresh cow rations
continued from page 39

From this project we determined that our LF diet likely had adequate amounts of fiber to foster a healthy rumen environment at the level of starch we achieved. Our HF diet, however, limited cows via gut fill and therefore showed negative responses in intake, milk production, and blood metabolites, mainly in weeks three and four postpartum. It is important to note that the HF cows responded quickly to a diet change, meeting the LF cows level of intake, production, and blood metabolites within a week after the diet switch.

In this study we found that it is possible to limit fresh cows via gut fill in the few weeks after calving, however, the limitation is not permanent and can be relieved quickly by feeding a more digestible diet. Although the fiber level in our low fiber diet was not low enough to show any detrimental effects, it is important to note that this is just for the level of starch we achieved. Formulation of fresh rations should occur being mindful of both the starch and fiber levels, specifically $uNDF_{240}$, to maximize both energy density of the diet and rumen health. ❐

Sarah Williams is a Ph.D. student and Thomas Overton (tro2@cornell.edu) is a professor in the Department of Animal Science at Cornell and Director of PRO-DAIRY.