

**PACIFIC NORTHWEST ANIMAL NUTRITION
CONFERENCE**

2022

PROCEEDINGS

Environment, climate change, and ag sustainability

*Frank Mitloehner, PhD
Director, CLEAR Center
Professor and Air Quality CE Specialist
Department of Animal Science
University of California, Davis*

INTRODUCTION

Climate change is a global issue that requires comprehensive and far-reaching solutions across all economic and demographic jurisdictions. The Paris Climate Agreement, adopted in 2015, sets out a global framework to address harmful climate impacts by limiting additional global warming to well below 2 degrees Celsius (°C) (1.5 °C goal). The accord recognizes regional differences and the need for specific actions across all jurisdictions, including developed economies providing leadership and assistance to developing nations in their climate mitigation efforts.

California continues to lead the United States and world in implementing measures to achieve emissions reductions of greenhouse gases (GHGs) that advance climate change. Toward this end, California has established ambitious goals for reducing GHG emissions (Senate Bill 32) by 40 percent by 2030 and 80 percent by 2050. Senate Bill 1383 (2016) also established specific goals for reducing short-lived climate pollutants (SLCPs), such as methane, by 40 percent from 2013 levels. Ultimately, California is working toward a goal of “net-zero” carbon emissions by 2045 (Executive Order B-55-18).

The U.S. dairy industry recently announced efforts to address climate change, boldly aiming for carbon neutral or better (net zero climate impact) by 2050 (Innovation Center for U.S. Dairy, 2020). As part of these important efforts, California’s dairy farms are leading change and making significant progress in reducing the amount of GHG emissions released into the environment. Producing a glass of milk from a California dairy cow generates 45 percent less GHG emissions today than it did 50 years ago. This finding, recently published in the Journal of Dairy Science, comes from a life-cycle assessment of California dairy farms in 1964 and 2014, conducted by researchers at the University of California, Davis (Naranjo et al., 2020). Significant advancements in farming efficiency, feed crop yields, veterinary care, sustainable feed practices, and animal nutrition have helped reduce the environmental footprint of individual cows. Building on these gains, more can be done to lower the climate footprint of milk production in the coming decade.

California’s dairy farmers are working closely with the California Department of Food and Agriculture (CDFA) and the California Air Resources Board (CARB) to further reduce dairy methane emissions. As the efforts continue, it is also important to improve our understanding

of how methane and other GHGs contribute to climate impacts, as we seek to limit warming. Leading climate scientists are now recognizing that moderately reducing methane emissions can quickly stabilize the climate pollutant's powerful impact, and further reductions can actually offset the far more damaging impacts of carbon dioxide (CO₂), which accumulate in the atmosphere for hundreds of years.

California's Greenhouse Gas Emissions

California, the fifth largest economy in the world, is responsible for about 1 percent of all global GHG emissions. More than 80 percent of California's emissions come from the transportation (41 percent), industrial (23 percent) and electrical (16 percent) sectors. Even though California is the United States' largest agricultural producer—producing fruits, vegetables, nuts, livestock, and other commodities for much of the U.S. and world—the sector's GHG contribution is only 8 percent of the state's total. California's largest-in-the-nation dairy sector accounts for about half of the agricultural share, or 4 percent of the state's total GHG emissions. The U.S. dairy sector accounts for 2 percent of the nation's total GHG emissions.

While CO₂ is the primary GHG driving climate warming, methane (CH₄), nitrous oxide (N₂O), and refrigerants are also important GHGs in California. According to CARB, carbon dioxide accounts for about 83 percent of California's GHG inventory. In comparison, methane accounts for 9 percent, and N₂O accounts for about 3 percent. In addition to knowing how much of each gas is being emitted, understanding how each gas causes actual warming is most critical to fully understanding and addressing climate change. Recent work by leading climate scientists at the Oxford Martin School and Environmental Change Institute at Oxford University has shed light on important differences among these GHGs and their impact on climate change (Lynch, 2019).

Methane emissions are generated by a number of processes, both those resulting from human related activity (anthropogenic) and natural (biogenic). Fossil-fuel methane (more commonly known as "natural gas") results from the process of extracting coal or oil, or from leakage during the extraction, storage, or distribution of natural gas for homes and businesses. Fossil methane is largely converted to CO₂ when we burn natural gas in our homes, factories, buildings, and other businesses.

Biogenic methane emissions are created by wetlands, rice cultivation, and ruminant livestock, as well as the waste sector, when microbes digest organic matter in our landfills and sewage treatment plants. Animal agriculture activity (all livestock) in California represents the largest source of biogenic methane emissions, accounting for roughly 55 percent of all human-related methane emissions in the state. California is the largest dairy state, producing roughly 18.5 percent of the nation's milk (USDA, 2019). The dairy livestock sector accounts for about 45 percent of all methane emitted in the state (CARB, 2015), primarily from two sources. Roughly half (55 percent) of dairy methane emissions come from manure management (storage, handling, and utilization), and the remaining 45 percent comes from enteric emissions.

In ruminant animals, methane is produced during manure decomposition as well as during enteric fermentation, where microbes decompose and ferment plant materials in the first compartment of their stomach, known as the rumen. This methane is expelled by the animal through belching.

Fossil Methane vs. Biogenic Methane

Fossil methane impacts the climate differently than biogenic methane. Fossil methane, such as natural gas, is carbon that has been locked up in the ground for millions of years and is extracted and combusted in homes and businesses. The burning of fossil methane directly transfers carbon that was stored in the ground (geologic carbon) into the atmosphere as CO₂. That carbon continues to accumulate and persist in the environment, contributing to climate change for hundreds of years. Bottom line: Fossil methane increases the total amount of carbon in the atmosphere, which drives warming.

Biogenic methane from cows is part of a natural carbon cycle, where after about 12 years it is removed from the atmosphere. As part of photosynthesis, plants capture CO₂ from the atmosphere, absorbing the carbon and releasing oxygen. That carbon is converted into carbohydrates in the plant, which are then consumed by the cows, digested, and released from the cows as methane (CH₄). After about 12 years in the atmosphere, that methane is oxidized and converted into CO₂. These carbon molecules are the same molecules that were consumed by cows in the form of plants. As part of the biogenic carbon cycle, the carbon originally utilized by the plant is returned to the atmosphere, contributing no net gain of CO₂.

Global Warming Potential of California's Primary Greenhouse Gases

Each GHG captures and retains heat at a unique rate, known as its global warming potential or GWP (as shown in Table 1 as GWP 100). For example, CH₄ has 28 times the warming potential of CO₂ over a 100-year period. Understanding how emissions impact global climate; however, requires consideration of not just the potency, but also how long each type of GHG will last in the atmosphere (atmospheric lifetime).

This is particularly important for methane, as it is a SLCP, with emissions breaking down after about 12 years (Farlie 2019; Lynch, 2019). In contrast, a significant proportion of CO₂ emissions are expected to persist in the atmosphere for hundreds of years, or even longer (Farlie, 2019; Lynch, 2019). As a result, the treatment of all GHGs as CO₂ equivalent (CO₂e) using GWP—and failure to consider the atmospheric removal of SLCPs—misrepresents the impact of methane on future warming (Frame et al., 2018; Cain, 2018). Recognizing this shortcoming, leading climate scientists expanded on GWP and developed GWP* (GWP-Star), which quantifies a GHG's actual warming potential, instead of just its CO₂ equivalence, by factoring in how much more or less methane is being emitted from a source over a period of time. GWP* appropriately builds on the conventional GWP approach employed in typical reporting of GHG emissions (Lynch, 2019). GWP* recognizes the rate and degradation of methane emissions, in addition to the total

amount of CO₂ and other long-lived gases emitted (Lynch, 2019; Cain, 2018; Frame et al., 2018).

Climate Impact Potential/GWP* (GWP-Star)

Recognizing the important differences in how methane and carbon dioxide affect climate change is critical to quantifying their actual climate impacts. GWP* was developed to better and more completely account for the warming impacts of short- and long-lived gases and better link emissions to warming (Cain, 2018). GWP* is still based on GWP, but recognizes how different gases such as methane affect warming (Cain, 2018).

Because CO₂ emissions last in the atmosphere for so long, they can continue to impact warming for centuries to come. New emissions are added on top of those that were previously emitted, leading to increases in the total atmospheric stock or concentration of CO₂. As a result, when additional CO₂ is emitted, additional global warming occurs (Frame et al., 2018).

In contrast, methane emissions degrade in the atmosphere relatively quickly, after about 12 years, and do not act cumulatively over long periods of time. For a constant rate of methane emissions, one molecule in effect replaces a previously emitted molecule that has since broken down. This means that for a steady rate of methane release—as emitted by a constant number of dairy cows, for example—the amount of methane in the atmosphere (concentration) stays at the same level and does not increase. As a result, when a steady amount of methane is emitted for more than 12 years, no additional global warming occurs (Frame et al., 2018).

This improved understanding of how short-lived versus long-lived emissions affect climate differently is critical to addressing further global warming. Limiting climate change requires that we bring emissions of CO₂ and other long-lived GHGs down to net-zero (Frame et al., 2018). For methane, however, it is possible to have steady ongoing emissions that do not result in additional warming (Frame et al., 2018).

This does not mean that methane can or should be ignored. Increasing methane emissions would result in significant warming. Because of its short-lived atmospheric lifetime, reducing methane emissions can lead to a drop in atmospheric concentration relatively quickly. So, reducing methane emission rates presents an important mitigation opportunity, which could reverse some of the warming the planet has already experienced (Lynch, 2019). Put simply, a reduction in methane emissions has climate cooling effects (Cain, 2018).

Climate-Neutral Dairy: Achievable in California's Near Future

Understanding how methane impacts global warming is critical to understanding the role of dairy production as a contributor to climate change. California's dairy sector is an excellent case in point. It is no longer growing and expanding production. The number of milk cows raised in the state reached a peak in 2008, around the same time that California passed its first climate policy (2006). Since then, the number of cows has declined by a little more than 7 percent

(CDFA, 2017). Total milk production has also decreased in recent years. As a result, the amount of methane in the atmosphere contributed by California milk production is less today than in 2008, as more methane is being removed from the atmosphere each year through its natural breakdown process (biogenic methane cycle) than is created by fewer dairy cows.

California dairy farms are also taking important, voluntary steps to further reduce methane from farms by installing anaerobic digesters designed to capture methane. Other projects, such as compost pack barns and solid separators, are designed to reduce methane production on farms. More than 213 dairy methane reduction projects have been incentivized with state funds to date (CDFA, 2019). These efforts alone are expected to achieve more than 2.2 million additional metric tons of GHG reduction each year, as the projects continue to be implemented (CDFA, 2019). Hundreds of additional dairy methane reduction projects are expected in future years.

As discussed earlier, enteric emissions (belching) from cows account for a significant share (45 percent) of total dairy methane emissions in California. Identifying solutions to reduce these emissions will also be necessary to meet state goals. While research into enteric emission mitigation is being conducted, and some feed additives show promise, commercially proven and cost-effective solutions are not yet available (Webinar on CARB's Analysis of Progress Toward Achieving Methane Emissions Target from Dairy and Livestock Sector, 2020).

Dairy farms also create other GHGs, such as CO₂ and nitrous oxide (N₂O), from the use of farm equipment for dairy management and the utilization of manure for growing crops. These emissions account for about 20 percent of all GHGs produced by the dairy production sector (Naranjo et al., 2020). Reducing or offsetting these emissions will also be necessary for the state's dairy production sector to achieve climate neutrality, or the point at which operations and resulting emissions are stable and no longer adding to global warming (no net global warming impact). California dairies are also reducing the amount of CO₂ they emit into the atmosphere through the adoption of solar energy and electrification of feed mixing and water pumping operations. Fossil fuel use per unit of milk produced has dropped by 58.5 percent from 1964 to 2014 (Naranjo et al., 2020). As dairy methane emissions are reduced further below current levels, then resulting cooling effects can offset some of the remaining CO₂ and other gases contributed by dairy production.

Conclusions

A continued focus on methane is necessary, as it is a powerful GHG and an important contributor to climate change. Under all scenarios, methane is significant, second only to carbon dioxide in terms of its overall contribution to global, human-driven climate change (Lynch, 2019). Over the last decade, global methane concentrations have increased (Lynch, 2019). Agriculture, including animal agriculture, is partially responsible for the increase, as dairy and meat production and consumption continue to expand globally, particularly in low- and middle-income countries. That notwithstanding, evidence is growing that shale gas production is a larger source of methane emissions than previously assumed (Howarth, 2019). Like every

sector of the global economy, agriculture must do its part if we are to succeed in achieving the overarching goal of limiting global warming. Equally important, California acting alone cannot accomplish significant global dairy methane emission reductions.

Recognizing how methane impacts global climate is also critical to assessing whether the state and world are on track to meet the goals of the Paris Agreement and limit warming to well below 2°C. Comparing GHGs with each other using GWP* preserves the link between emissions and warming or cooling of the atmosphere (Schleussner et al., 2019). It also provides an informative and better suited way to assess the relative merits of different options for reducing GHG emissions, especially in ambitious mitigation scenarios (Cain, 2019). More accurate expression of mitigation efforts in terms of their direct contribution to future warming also better informs burden-sharing and long-term policies and measures in pursuit of ambitious global temperature goals (Allen, 2018; Schleussner et al., 2019).

Reducing methane emissions and achieving climate neutrality is no small undertaking. California is among the most efficient producers of milk and dairy products, and its life-cycle carbon footprint (per gallon of milk produced) is among the lowest of any region in the world. Achieving these or similar levels of production efficiency (more milk with fewer cows) is a critical first step for other dairy regions to begin stabilizing methane emissions and work toward climate neutrality. The impact of such an accomplishment would have profound climate effects. Attaining California's level of production efficiency in all global dairy production regions could reduce total global GHG emissions by as much as 1.73 percent (E. Kebreab, calculations based on Naranjo et al., 2020 and FAO & GDP, 2018).

A full understanding of the potential climate impact of all greenhouse gases is also important in ensuring effective policies are developed to address methane and other flow pollutants in line with their effects. Dairy production primarily produces flow emissions (80 percent is methane) with smaller amounts of stock emissions, such as CO₂ and N₂O (Naranjo et al., 2020). Policy or consumption decisions that trade off and result in greater concentrations of CO₂ and N₂O, while reducing methane, may ultimately leave a warmer planet behind in the long term (Frame et al., 2018).

Adopting sustainable farming practices to vastly improve production efficiency is probably the single-most important step other dairy-producing countries can take to begin to stabilize regional and global methane emissions and begin to achieve climate neutrality. The United Nations Food and Agriculture Organization (FAO) estimates that improved management practices alone could reduce net global methane emissions by 30 percent (FAO, 2019). These efforts will be critical to reduce livestock methane emissions and present important opportunities for reaching global climate mitigation targets. Further reductions in methane emissions will lead to atmospheric concentrations falling relatively quickly, which could reduce some of the warming already experienced (Lynch, 2019).

References

- Allen, M.R., Fuglestedt, J.S., Shine, K.P., Reisinger, A., Pierrehumbert, R.T., & Forster P.M. (2016). New use of global warming potentials to compare cumulative and short-lived climate pollutants. *Nature Climate Change*. 6. 773–6. Retrieved from <https://www.nature.com/articles/nclimate2998?cacheBust=1508877188307>
- Allen, M.R., Shine, K.P., Fuglestedt, J.S., Millar, R.J., Cain, M., Frame, D.J., & Macey, A.H. (2018). A solution to the misrepresentations of CO₂-equivalent emissions of short-lived climate pollutants under ambitious mitigation. *npj Climate and Atmospheric Science*. 1(16). Retrieved from <https://www.nature.com/articles/s41612-018-0026-8>
- California Air Resources Board. (2019, August 12). California 2017 Greenhouse Gas Inventory. Retrieved from https://www.arb.ca.gov/cc/inventory/data/tables/ghg_inventory_bygas.pdf
- California Air Resources Board. (2015). California’s methane inventory based on the 2015 edition the CARB greenhouse gas inventory. Retrieved from <https://www.arb.ca.gov/ghg-slcp-inventory>
- California Department of Food and Agriculture. (2019, September 18). CDFA Awards Nearly \$102 Million for Dairy Methane Reduction Projects [Press release]. Retrieved from https://www.cdfa.ca.gov/egov/Press_Releases/Press_Release.asp?PRnum=19-085
- California Department of Food and Agriculture Dairy Marketing, Milk Pooling, and Milk and Dairy Foods Safety Branches. (2017). [California dairy cows and milk productions]. Unpublished raw historical data.
- Cain, M. (2018). Guest post: A new way to assess ‘global warming potential’ of short-lived pollutants. *Carbon Brief*. Retrieved from <https://www.carbonbrief.org/guest-post-a-new-way-to-assess-globalwarming-potential-of-short-lived-pollutants>
- Cain, M., Lynch, J., Allen, M.R., Fuglestedt, D.J. & Macey, A.H. (2019). Improved calculation of warming- equivalent emissions for short-lived climate pollutants. *npj Climate and Atmospheric Science*. 2(29). Retrieved from <https://www.nature.com/articles/s41612-019-0086-4>
- Dairy Cares. (2019, August 28). Cows vs Cars? [Video file]. Retrieved from <https://www.youtube.com/watch?v=RW8BclS27al&vl=en>
- Dairy Industries International. (2019). Sustainability project aims for net zero climate impact in US Dairy. Retrieved from <https://www.dairyindustries.com/news/32149/sustainability-project-aims-for-net-zero-climate-impact-in-us-dairy/>
- Fairlie, S. (2019). A Convenient Untruth. *Resilience*. Retrieved from <https://www.resilience.org/stories/2019-05-10/a-convenient-untruth/>

FAO. (2019). Five practical actions towards low-carbon livestock. Rome. Retrieved from <http://www.fao.org/documents/card/en/c/ca7089en/>

FAO and GDP. (2018). Climate change and the global dairy cattle sector – The role of the dairy sector in a low-carbon future. Rome. 36 pp. Licence: CC BY-NC-SA- 3.0 IGO. Retrieved from <https://dairysustainabilityframework.org/wp-content/uploads/2019/01/Climate-Change-and-the-Global-Dairy-Cattle-Sector.pdf>

Frame, D., Macey, A.H., & Allen, M. (2018). Why methane should be treated differently compared to long-lived greenhouse gases. The Conversation. Retrieved from <https://theconversation.com/why-methane-should-be-treated-differently-compared-to-long-lived-greenhouse-gases-97845>

Howarth, R. W. (2019). Ideas and perspectives: is shale gas a major driver of recent increase in global atmospheric methane?. Biogeosciences, 16, 3033–3046. Retrieved from <https://doi.org/10.5194/bg-16-3033-2019>

Lynch, J. (2019). Agricultural methane and its role as a greenhouse gas. Food Climate Research Network, University of Oxford. Retrieved from <https://foodsource.org.uk/building-blocks/agricultural-methane-and-its-role-greenhouse-gas>

Naranjo, A., Johnson, A., Rossow, H., & Kebreab, E. (2020). Greenhouse gas, water, and land footprint per unit of production of the California dairy industry over 50 years. Journal of Dairy Science. 103, 3760-3. Retrieved from [https://www.journalofdairyscience.org/article/S0022-0302\(20\)30074-6/fulltext](https://www.journalofdairyscience.org/article/S0022-0302(20)30074-6/fulltext)

Schleussner C., Nauels, A., Schaeffer, M., Hare, W., & Rogelj, J. (2019). Inconsistencies when applying novel metrics for emissions accounting to the Paris agreement. Environmental Research Letters. 14(12). Retrieved from <https://iopscience.iop.org/article/10.1088/1748-9326/ab56e7/meta>

United States Department of Agriculture, National Agricultural Statistics Service. (2019). Milk Production, Disposition, and Income 2018 Summary. Retrieved from <https://usda.library.cornell.edu/concern/publications/4b29b5974>

Ruminal Microbiome: What is new about their contributions to ruminal fermentation and digestion and ruminant productivity?

T. G. Nagaraja
University Distinguished Professor
Department of Diagnostic Medicine/Pathobiology
College of Veterinary Medicine
Kansas State University
Manhattan, Kansas 66506-5800.
Tel: 785-532-1214
Cell: 785-341-6342
Email: tnagaraj@vet.k-state.edu

Introduction

Ruminants, particularly cattle, sheep, and goats, are important production animals for meat and milk to humans worldwide. Their importance comes from their unique digestive tract equipped with a specialized region called foregut or reticulo-rumen that carries out microbial digestion. Because of the microbial contribution to digestion, they are capable of converting fiber-based feeds, with or without grains, into high quality, protein-rich products like milk and meat. The reticulum and rumen, which are practically one compartment, are inhabited by a variety of microbes that work in concert to breakdown feeds to produce energy (volatile fatty acids; VFA), protein (microbial cells) and other nutrients like vitamins (microbial cells) to the host. The production of VFA, mainly from carbohydrates, is central to the ruminal fermentation because the process provides energy (ATP) for microbial growth, which serves as the major source of protein to the host, but also provides the animal with the precursors necessary to generate energy (mainly acetate), glucose (mainly propionate), and lipid (mainly acetate and butyrate). The fermentation of nitrogenous compounds is also an integral process because it provides the molecules (amino acids and ammonia) necessary to build microbial cell protein. In addition to the provision of nutrients, ruminal microbes are linked to host physiology, including the development of ruminal epithelium, most likely involving the modulation of host gene regulation by VFA.

Despite the global importance of ruminants and the tremendous progress that has been made to improve efficiency of milk and meat production, the rumen remains an under investigated, hence, under-characterized, microbial ecosystem. The description that 'rumen is a black box', first made several decades ago, is still applicable. At one time, rumen was the most extensively investigated anaerobic ecosystem. However, in the past 15 years, human gut microbial studies have far outpaced rumen microbiology. The human gut microbiome studies were part of the National Institute of Health-funded Human Microbiome Project, a logical extension of the Human Genome Project, to study the distribution and evolution of the constituent microorganisms in the human body (Llyod-Price et al., 2016). The impetus for the gut microbiome studies is largely because of the recognition that gut microbes have profound impact on human health and diseases (Cani et al., 2018).

Ruminal Microbes

A simple microscopic examination of ruminal fluid reveals a complex and diverse microbial

impact on human health and diseases (Cani et al., 2018).

Ruminal Microbes

A simple microscopic examination of ruminal fluid reveals a complex and diverse microbial population (Figure 1A, B, C). The population includes members of all three domains of life: Bacteria, Archaea (methanogens) and Eukarya (fungi and protozoa). The bacterial activities are absolutely essential for ruminal function and survival of the ruminant host; however, the archaeal and eukaryotic domains are not indispensable. Of the three domains, bacteria are the dominant population and most extensively investigated. Additionally, as in most microbial ecosystems, rumen also possesses acellular organisms called bacterial viruses or bacteriophages as well as fungal and protozoal phages. The structure and contribution of the viral community is the least investigated and hence not much is known about their role.

Molecular ‘Omics’ Methods

Initial molecular techniques were based on amplification of nucleic acids by polymerase chain reaction (PCR), both conventional and real-time, and restriction fragment length polymorphic analyses, such as ribotyping, pulsed-field gel electrophoresis, denatured gradient gel electrophoresis for identification and genetic typing. In recent years, research on rumen microbial ecology has expanded and exploded because of high-throughput and high-resolution nucleic acid sequence (DNA and RNA) and chemical separation and identification methods for protein and metabolites analyses. The advances in nucleic acid sequencing and bioinformatics analyses (whole genome sequencing, Amplicon sequencing and Metagenomics) have enabled researchers to analyze whole genome of an organism, community composition and function of an ecosystem by culture independent methods. DNA sequence information provides insight into physiologic and metabolic potential based on the whole genome, microbial community composition (‘who are there?’), but does not provide a direct measure of the function (‘what are they doing?’), although potential function can be deduced from the genes identified. Therefore, analysis that measure gene expressions or transcription of DNA to messenger RNA, called (meta)transcriptomics, translation of mRNA into protein, called (meta)proteomics, or ultimately production of products or metabolites, called metabolomics, are necessary to delineate functional profiling of the microbial community in the rumen.

The explosive growth in the study of gut microbes is because of the development of high-throughput and high-resolution molecular methods to unravel the community composition and functional role in the ecosystem.

Genomics of Ruminal Microbes

Genomics is the science of sequencing, mapping, and analyzing the entire complement of genetic information of an organism. Essentially, it is a genetic blueprint that provides complete information on the evolution and physiology of the organism. The process provides raw sequences that need to be assembled and annotated (read) to provide biological meaning. The process has become so inexpensive and common, the technique has become routine and often a starting point for characterizing and analyzing the metabolic potential of an organism. The

first rumen bacterial species that was genome sequenced was *Fibrobacter succinogenes*, a dominant fibrolytic bacterium (Jun et al., 2007). A global project on a comprehensive genomic analysis of ruminal microbes was initiated, somewhat similar human gut microbiome project. The Hungate 1000 project (www.Hungate1000.org.nz), a global initiative launched in 2012, was designed to provide a reference set of rumen microbial genome sequences from cultivated ruminal bacteria, archaea, fungi and ciliated protozoa. The database, which are publicly available, enables researchers to analyze the physiology and metabolic potential of the organism with regard to ruminal function. At the beginning, genome sequences were 43 available for 14 bacterial species (belonging to 11 of 88 known genera in the rumen) and one methanogen. As many as 501 organisms (belonging to 73 of 88 genera) have been sequenced, referred to as Hungate genome catalog (Seshadri et al., 2018). Anaerobic fungal genomes have been difficult to sequence because of their high adenine and thymine content, repeat-sequences, complex physiology and unknown ploidy (Edwards et al., 2017). So far, whole genomes of five fungal species have been sequenced and are publicly available; however, there are no genomic sequence data on ciliated protozoa of the rumen.

The genomic sequence of an organism can provide comprehensive information on the metabolic potential. As an example, the genome of *Fibrobacter succinogenes*, a dominant fibrolytic organism, was the first ruminal bacterium to be sequenced and annotated (identification and analysis of the genes). The organism contains 3,252 genes coding for proteins and of those at least 104 genes were identified as coding for enzymes involved in plant cell wall degradation, including 33 genes for cellulose enzymes (Suen et al., 2011). Biochemical studies before genomic sequencing had only identified a dozen or so enzymes in *F. succinogenes* involved in cell wall digestion. The information gleaned from genomics of fibrolytic bacteria not only provides more information on fiber digestion in the rumen, but could potentially lead to identification of novel fibrolytic enzymes for commercial exploitations such as exogenous enzymes as feed additives or their use in biofuel production (Hess et al., 2011).

Amplicon Sequencing and Metagenomics. Sequence-based taxonomic profiling of a microbiome are carried out by amplifying 16S rRNA genes or by whole-metagenome shotgun sequencing. Amplicon sequences of 16S rRNA (reads) are commonly grouped into clusters, called as ‘operational taxonomic units (OTUs)’, which are then assigned to specific taxa based on sequence homology to a reference genomic sequence. In shotgun metagenomics, sequencing methods are applied to millions of random genomic fragments of DNA extracted from ruminal contents. The shotgun sequence reads are used to determine community composition, either by considering the reads individually or by first assembling them into contigs, which are then compared to a reference catalog of microbial genes or genomes. Such community analyses allow researchers to carry out taxonomic profiling of the microbial community to answer the question, ‘who are present?’ in the rumen. Taxonomic profiling of microbial species in the rumen have been performed on the different ruminant species (cattle, sheep, goats, and buffaloes) in relation to animal to animal variation, diet changes, ruminal disorders (acidosis, bloat, liver abscesses, low-milk fat syndrome), feed efficiency, milk production, methane production, maternal influence, feed additives, and seasonal changes, etc. (Denman et al., 2018; McCann et al., 2014). The utility and applicability of the rumen microbial profiling by molecular techniques are best evidenced by a study published by

Henderson et al. (2015). The study to assess the effects of diet, animal species and geographical location on ruminal microbial population involved 742 ruminal content samples from 32 animal species located in 35 countries. The differences in microbial communities were predominantly attributable to diet, and host factors were less influential. The protozoal communities were variable, but dominant bacteria and archaea were similar among all samples, and across animal species, diet, and geographical region a core microbiome was present (Henderson et al. (2015).

Metatranscriptomics. The metatranscriptomics, also called RNA-seq, involves sequencing all of the RNA produced by a microbial community, except ribosomal RNA, which is first depleted before sequencing. The RNA preparation is essentially messenger RNA (mRNA), which is converted to DNA, called complementary DNA (cDNA), for sequencing. A few of the studies on metatranscriptomics have focused on carbohydrate-degrading enzymes associated with microbes adherent to the fiber (Dai et al., 2015; Comtet-Marre et al., 2017). These studies have confirmed culture-base studies that major bacterial activities of fiber degradation were associated with species of the genera *Fibrobacter*, *Prevotella* and *Ruminococcus*, but also indicated large contribution of fungal and protozoal species.

Metaproteomics. Protein is the ultimate product of gene function, therefore, measuring protein abundance provides a more direct indicator of the functional activity of the microbes. The high-throughput method of measuring proteins and their abundance, called metaproteomics, involves mass-spectrometry-based shotgun quantification of peptide mass and abundance. The peptides are then associated with full-length proteins by sequence homology-based searches against reference databases, similar to data bases available for DNA and RNA sequences. Studies on metaproteomics of ruminal fluid are limited (Snelling and Wallace, 2017; Deusch and Seifert, 2015). The study by Deusch and Seifert (2015) identified in excess of 2,000 bacterial, 150 archaeal, and 800 fungal and protozoal proteins in the fiber adherent fraction of the ruminal digesta.

Metabolomics. The metabolomics refers to the detection, identification, and often quantification of metabolites and other small molecules in microbial communities. It is not done by predictions based on genomic information, instead, the analysis relies on techniques, such as high performance liquid chromatography, to separate chemicals, which are then identified and quantified by mass spectroscopy. Ruminal VFA analysis, a widely used technique in ruminal fermentation studies, is an example of a metabolomics. However, metabolomics, as defined now, is a more comprehensive chemical analysis that detects and quantifies all possible chemicals present in a sample. Metabolomic analysis to study the link between microbes and metabolites have been studied in several gut ecosystems. The first study on metabolomics of ruminal fluid was published by Ametaj et al (2010). The study measured ruminal metabolites of dairy cows fed diets with increasing proportions of grain. The results showed unhealthy alterations in the metabolites (increased methylamine, dimethylamine, N-nitrosodimethylamine, endotoxin, ethanol, phenylacetyl glycine, etc.) in ruminal fluid of cows fed higher amounts of grains. What is not known how these alterations are linked to ruminal dysfunction.

Culture vs. Molecular methods

The understanding of the relationship between microbial community and rumen function has generally been based on culture-based analysis, particularly of bacteria and to some extent of fungi. Bacteria are the most predominant organisms in the rumen ranging from 10 to 100 billion per g and account for up to 50% of the microbial cell mass. Rumen bacterial cultivation began almost 8 decades ago with the development of anaerobic techniques, referred to as Hungate's techniques. A simple microscopic examination of ruminal contents has shown morphologically distinct bacteria, such as *Lampromedia*, *Oscillospira*, etc., which have not been cultivated yet. An advantage of microbial community analysis with nucleic acid-based techniques is that ruminal content samples need not be processed immediately to maintain viability and can be archived and processed at convenience. However, with the development and application of a variety of cultivation-independent, molecular techniques, it has become clear that cultivation-based methods have only identified approximately 10 to 20% or less of the total microbial population harbored in the rumen

Ruminal Microbiome

A number of microbiome studies have attempted to relate or link community composition to rumen function and dysfunction. Jami et al (2014) reported that certain physiological parameters, such as total milk yield and milk fat yield correlated with the abundance of certain bacteria in the rumen. Xue et al (2019) reported that rumen bacterial richness and the relative abundance of several bacterial taxa were significantly different between dairy cows with high and low milk protein production. In a study that compared cows with high and low milk protein and fat percentages, concentrations of total VFA, acetate, propionate, and butyrate in high-producers were higher compared to low-producers (Wu et al., 2021). Also, the two groups displayed differences in 38 most abundant species, and genus *Prevotella* accounted for 68.8% of the species with the highest abundance in the high producers. A number of studies have addressed the link or relationship of ruminal microbiome to feed efficiency, a most important trait in the cattle production systems. Bacterial profiles in the rumens of efficient cattle (low residual feed intake) indicated differences in abundances of genera, *Butyrivibrio*, *Lactobacillus*, *Prevotella*, *Ruminococcus*, and *Succinivibrio* compared to inefficient cattle (high RFI; Myer et al., 2015). Li and Guan (2017) have compared cattle with high or low efficiency based on microbiome, metatranscriptomic and carbohydrate enzyme analyses. Three bacterial families (*Lachnospiraceae*, *Lactobacillaceae*, and *Veillonellaceae*) were more abundant in inefficient cattle, and they displayed greater abundance for 30 metabolic pathways and 11 carbohydrate active enzymes, whereas the efficient cattle displayed greater abundance for two metabolic pathways and one carbohydrate active enzyme. The authors suggested that rumen microbiomes of inefficient cattle are more metabolically diverse than those of efficient cattle. A detailed description of the microbiome studies in relation to hydrogen and methane production is given below.

Ruminal Microbiome: Hydrogen and Methane Production and Methane Mitigation Strategies

Hydrogen is a key product in the rumen and is produced by fermentation of both fiber and starch. The hydrogen is used in several hydrogen-sink reactions, of which, methane

production by archaeal population is the major route in the rumen (Figure 2). The utilization of H₂ in an ecosystem that does not have oxygen is critical to prevent increases in the concentration of H₂ and prevent disruption of the normal functioning of microbial enzymes involved in oxidation-reduction reactions. The production of H₂ by one species and utilization by another species, referred to as 'inter species H₂ transfer', is a major microbial interaction in the rumen (Figure 3). The interaction is thermodynamically favored to re-oxidize intracellular reduced cofactors, such as NADH FADH, FDH, etc. because of the ability of methanogens to decrease H₂ concentration. Therefore, in the presence of methanogens or other H₂-consuming reactions, such as succinate- or propionate producers, H₂-producers shift fermentation away from formate, lactate and ethanol (products that do not yield ATP) to acetate (a product that yields ATP). The additional ATP results in higher growth, production of more enzymes, hence higher digestibility.

Although methanogens account for 1 to 4% of the total microbial population in the rumen, methanogenesis represents a major pathway to utilize hydrogen. The methanogens in the rumen are distributed free in ruminal fluid, attached to feed particles, associated with ciliated protozoa, and even attached to ruminal epithelium. Methanogens associated with protozoa and epithelium are novel phylotypes (or species), and the role of methanogens associated with ruminal epithelium has not been identified. Methanogens associated with ciliated protozoa can be intracellular, called endosymbionts, or on the surface, called ectosymbionts. Intracellular methanogens are found inside most of the common protozoal species. In contrast, the extracellular methanogens are less numerous and only 30 to 50% of the protozoan cells carry them. Protozoa produce hydrogen in large amounts in a specialized organelle called hydrogenosomes (similar to mitochondria). This hydrogen is utilized by methanogens that are inside or outside the protozoan cell, and the association represents an important microbial interaction in the rumen.

There are only a limited number of substrates that methanogens are capable of utilizing for methanogenesis. In the rumen the major substrates are CO₂ and hydrogen, and formate, a product of many bacteria, particularly fiber digesters. Formate accounts for approximately up to 18% of ruminal methane. There are three major pathways of ruminal methanogenesis (Figure 4):

- a. Hydrogenotrophic pathway in which H₂ is used as electron donor to reduce CO₂ to methane.
- b. Methylotrophic pathway in which methyl group of methanol or methylamines is reduced to methane
- c. Acetoclastic pathway in which the methyl group of acetate is reduced to methane.

Methane is a waste product, hence, it is expelled into the environment, which results in the loss of energy (2 to 15% of feed energy) to the animal and a anthropogenic source of greenhouse gas to the environment. Methane, as a potent greenhouse gas, is a major contributor, next only to CO₂, of global warming. Methane is more potent than CO₂ and estimated to account for 14% of total global greenhouse gas emissions. About 25% of the anthropogenic methane emissions are due to gut fermentations in livestock, particularly ruminants.

Although there is no relationship between methanogen abundance in the rumen to

production efficiency of the animal, the species composition of methanogenic population is different between efficient and inefficient cattle (Zhou et al. 2009). In a study that used metagenomics analysis, a significantly higher abundance of *Methanobrevibacter* was detected in the rumen of high-methane producing steers compared to low-methane producers (Wallace et al., 2015). Interestingly, a couple of studies in sheep have noted differences in rumen microbiome beyond methanogens in relation to low- or high- methane producers (Kittelmann et al., 2014; Kamke et al., 2016; Wallace et al., 2015). Two bacterial genera, *Sharpea* and *Kandleria* (Kumar et al., 2018) were associated with low methane production. A metagenomic and metatranscriptomic study conducted by Kamke et al. (2016) confirmed the relative abundance of *Sharpea* was greater in low-methane producing sheep compared to high methane producing sheep. Not much is known about these two bacterial genera, except they are anaerobic and produce predominantly D-lactic acid from sugars. Not surprisingly, another organism that is significantly enriched in low methane producers is *Megasphaera elsdenii*, a major lactic acid-fermenting bacterium in the rumen (Kamke et al., 2016; Shabat et al., 2016). Thus, methanogenesis not only is related to methanogens but also other components of the microbiome, particularly lactic acid producers and fermenters. It is possible that lactic acid pathway (production and fermentation) may be central to the production of VFA as an alternative sink to methanogenesis (Mizrahi and Jami, 2018).

Because ruminal methanogenesis results in the loss of energy, therefore, for a number of years, a major focus of researchers has been to develop an effective strategy to inhibit methane production in the rumen. The strategies that have been investigated can be broadly categorized to intervene at the following three stages of methane production (Figure 5):

1. Inhibit or reduce production of major precursors of methane production (H₂ and formic acid).
2. Divert hydrogen to alternate hydrogen-sink reactions in the rumen, which include lactate, propionate and valerate production, acetate production by reduction of CO₂, and reduction of fumarate, nitrate and sulfate.
3. Eliminate or reduce methanogens in the rumen.

Because methane is the major scavenger of hydrogen in the rumen, methane inhibition results in hydrogen accumulation. It is generally assumed that hydrogen accumulation will inhibit re-oxidation of reduced cofactors like NADH and adversely affect the microbial fermentation. Therefore, strategies to mitigate methanogens should consider alternatives to sink hydrogen in the fermentation process (Wright and Klive, 2011). However, no negative effects of methane inhibition have been shown possibly because none of the methods tested inhibit 100% of methane production. Even an effective compound like bromochloromethane (BCM), which reduces methane production by about 80%, had no negative effective effects on feed intake and digestibility in goats (Mitsumori et al., 2012). Although several inhibitors of methane production were effective in in vitro studies, they were reported to be ineffective in in vivo studies.

A promising compound appears to be 3-nitroxy propanol (3-NOP), an analog of the Coenzyme M that inhibits methyl coenzyme M reductase, which is present in all methanogens and is the terminal step in methanogenesis (Ermler et al., 1993). Several studies have shown that

including 3-NOP in diets of dairy cows (Hristov et al., 2015) and beef cattle (Vyas et al., 2016) decreased methane emissions (up to 60%) with no negative effect on ruminal fermentation and animal productivity. Furthermore, inclusion of monensin in the diet had no significant interaction with the effects of 3-NOP (Vyas et al., 2018)

Researchers in New Zealand (Attwood et al., 2011; Leahy et al., 2010) have sequenced and analyzed the genome of *Methanobrevibacter ruminantium*, a major ruminal methanogen, and have identified methanogen-specific genes that code for critical enzymes for methane production, which can potentially be targeted for mitigation. The organism contains a large number of genes that encode for surface adhesion like proteins, which may be involved in mediating close association with hydrogen-producing bacterium or protozoa in the rumen. These proteins can potentially be used as antigens in a vaccine to induce antibodies to inhibit ruminal methanogens.

Conclusions

Rumen is inhabited by a dense population of microbes, which include members of all three domains of life: Bacteria, Archaea (methanogens) and Eukarya (fungi and protozoa), as well as viruses. The fermentative activities of these microbes convert complex organic feedstuffs into energy and protein, which are then used by the host for growth and production. Molecular methods to analyze bacterial community composition have identified a number of novel bacterial genera and species, which have not been cultured, therefore, nothing is known about their role in ruminal fermentation. Anaerobic fungi are the most active and effective fibrolytic organisms because of their combined mechanical (ability to penetrate plant structures) and enzymatic activities. Although ciliated protozoa contribute to digestibility of feeds and VFA production, their overall role in ruminal fermentation and contribution to the host nutrition is still an area of considerable debate and controversy. Rumen viral community analysis has identified a number of viral types and of those a small population have a significant similarity to known viruses. Viruses may be the driving factor in the evolution and stability of microbes in the rumen. Before the advent of molecular techniques, the understanding of the ruminal microbes and their contribution to the host nutrition was based on classical culture methods. In recent years, there is explosive growth on the culture-independent methods, which have provided identity and quantity of microbes and have vastly expanded our understanding of the community composition. These studies are providing answers to who is there, and how many, but provide limited information on what are they doing. Cultivation and functional characterization of species and strains of microbes identified by molecular methods remain a major challenge to rumen microbiologists. An increased functional understanding of the microbiome of the rumen as well as that of the hindgut of ruminants is essential to develop novel approaches to manipulate to improve food animal production.

References

Attwood, G. T., E. Altermann, W. J. Kelly, S. C. Leahy, L. Zhang, and M. Morrison. 2011. Exploring rumen methanogen genomes to identify targets for methane mitigation strategies. *Anim. Feed Sci. Technol.* 166-167:65-75.

Ametaj, B. N., Q. Zebeli, F. Saleem, N. Psychogios, M. J. Lewis, S. M. Dunn, J. Xia and D. S. Wishart. 2010. Metabolomics reveals unhealthy alterations in rumen metabolism with increased proportion of cereal grain in the diet of dairy cows. *Metabolomics* 6:583-594.

Cani, P. D. 2018. Human gut microbiome: Hopes, threats, and promises. *Gut* 67:1716-1725.

Comtet-Marre S, N. Parisot, P. Lepercq, F. Chaucheyras-Durand, P. Mosoni, E. Peyretailade, A. R. Bayat, K. J. Shingfield, P. Peyret and E. Forano. 2017. Metatranscriptomics reveals the active bacterial and eukaryotic fibrolytic communities in the rumen of dairy cow fed a mixed diet. *Front. Microbiol.* 8:67.

Dai X, Y. Tian, J. Li, X. Su, X. Wang, S. Zhao, L. Liu, Y. Luo, D. Liu, H. Zheng, J. Wang, Z. Dong, S. Hu and L. Huang. 2015. Metatranscriptomic analyses of plant cell wall polysaccharide degradation by microorganisms in the cow rumen. *Appl. Environ. Microbiol.* 81:1375-1386.

Denman, S. E., D. P. Morgavi, and C. S. McSweeney. 2018. Review: The application of omics to rumen microbiota function. *Animal.* 12:233-245.

Deusch S. and J. Seifert. 2015. Catching the tip of the iceberg – evaluation of sample preparation protocols for metaproteomic studies of the rumen microbiota. *Proteomics* 15:3590–3595.

Edwards, J. E., R. J. Forster, T. M. Callaghan, V. Dollhofer, S. S. Dagar, Y. Cheng, J. Chang. 2017. S. Kittelmann, K. Fliegerova, A. K. Puniya, J. K. Henske, S. P. Gilmore, M. A. O'Malley, G. W. Griffith and H. Smidt. 2017. PCR and omics based techniques to study the diversity, ecology and biology of anaerobic fungi: Insights, challenges and opportunities. *Front. Microbiol.* 8:1657.

Ermler U, W. Grabarse, S. Shima, M. Goubeaud, and R. K. Thauer RK. 1997 Crystal structure of methyl-coenzyme M reductase: the key enzyme of biological methane formation. *Science* 278(5342):1457–1462

Henderson G, F. Cox, S. Ganesh, A. Jonker, W. Young and P. H. Janssen. 2015. Rumen microbial community composition varies with diet and host, but a core microbiome is found across a wide geographical range. *Scientific Reports* 5,14567.

Hess, M., A. Sczyrba, R. Egan, T-W. Kim, H. Chokhawala, G. Schroth, S. Luo, D. S. Clark, F. Chen, T. Zhang, R. I. Mackie, L. A. Pennacchio, S. G. Tringe, A. Visel, T. Woyke, Z. Wang and E. M. Rubin. 2011. Metagenomic discovery of biomass-degrading genes and genomes from cow rumen. *Science* 331:463-467.

Hristov, A. N., J. Oh, F. Giallongo, T. W. Frederick, M. T. Harper, H. L. Weeks, et al.. 2015. An inhibitor persistently decreased enteric methane emission from dairy cows with no negative effect on milk production. *Proc. Natl. Acad. Sci. USA* 112:10663-10668.

Li F, Guan LL. 2017. Metatranscriptomic profiling reveals linkages between the active rumen microbiome and feed efficiency in beef cattle. *Appl. Environ. Microbiol.* 83(9):e00061-17.

Jami, E., B. A. White, I. Mizrahi. (2014). Potential role of the bovine rumen microbiome in modulating milk composition and feed efficiency. *PLoS One*, 9:e85423.

Jun, H. S., M. Qi, J. K. Ha, and C. W. Forsberg. 2007. Fibrobacter succinogenes, a dominant fibrolytic ruminal bacterium” Transition to the post genomic era. *Asian-Australas J. Anim. Sci.* 20:802-810.

Kamke J., S. Kittelmann, P. Soni, Y. Li, M. Tavendale, S. Ganesh, P. H. Janssen, W. Shi, J. Froula and E. M. Rubin. 2016. Rumen metagenome and metatranscriptome analyses of low methane yield sheep reveals a Sharpea-enriched microbiome characterized by lactic acid formation and utilization. *Microbiome* 4, 56.

Kittelmann, S., C. S. Pinares-Patino, H. Seedorf, M. R. Kirk, S. Ganesh, J. C. McEwan, and P. H. Janssen. 2014. Two different bacterial community types are linked with the low-methane emission trait in sheep. *PLoS One* 9:e103171.

Kumar S., B. P. Treloar, K. H. Teh, C. M. McKenzie, G. Henderson, G. T. Attwood, S. M. Waters, M. L. Patchett, P. H. Janssen. 2018. Sharpea and Kandleria are lactic acid producing rumen bacteria that do not change their fermentation products when co-cultured with a methanogen. *Anaerobe* 54:31-35.

Leahy, S. C., W. J. Kelly, E. H. Altermann, R. S. Ronimus, C. Yeoman, D. M. Pacheco, D. Li, Z. Kong, S. McTavish, C. Sang, S. C. Lambie, P. H. Janssen, D. Dey, G. T. Attwood. 2010. The genome sequence of the rumen methanogen *Methanobrevibacter ruminantium* reveals new possibilities for controlling ruminant methane emissions. *PLoS One* 5:e8926.

Llyod-Price, J., G. Abu-Ali, and C. Huttenhower. 2016. The healthy human microbiome. *Genom. Med.*8:51.

McCann, J. G., T. A. Wickersham, and J. L. Loo. 2014. High-throughput methods redefine the rumen microbiome and its relationship with nutrition and metabolism. *Bioinform Biol. Insights.* 8:109-125.

Mitsumori, M., T. Shinkai, A. Takenaka, O. Enishi, K. Higuchi, Y. Kobayashi, Y., et al. 2012. Response in digestion, rumen fermentation and microbial populations to inhibition of methane formation by a halogenated methane analogue. *Br. J. Nutr.* 108: 482-491.

Myer, P. R. T. P. Smith, J. E Wells, L. A. Kuehn, and H. C. Freely (@015). Rumen microbome from steers differing in feed efficiency . *PLoS One*, 10:e0129174.

Seshadri R., S. C. Leahy, G. T. Attwood, K. H. Teh, S. C. Lambie, A. L. Cookson, E. A. Eloef-Fadrosh, G. A. Pavlopoulos, M. Hadjithomas, N. J. Varghese, D. Paez-Espino. Hungate project collaborators, R. Perry, G. Henderson, C. J. Creevey, N. Terrapon, P. Lapebie, E. Drula, V. Lombard, E. Rubin, N. C. Kyrpides, B. Henrissat, T. Woyke, N. N. Ivanova and W. J. Kelly. 2018. Cultivation and sequencing of rumen microbiome members from the Hungate1000 collection. *Nat. Biotechnol.* 36:359-367.

Shabat S. K. B., G. Sasson, A. Doron-Faigenboim, T. Durman, S. Yaacoby, M. E. B. Miller, B. A. White, N. Shterzer and I. Mizrahi. 2016. Specific microbiome-dependent mechanisms underlie the energy harvest efficiency of ruminants. *ISME J.* 10: 2958-2972..

Snelling, T. J., and R. J. Wallace. 2017. The rumen microbial metaproteome as revealed by SDS-PAGE. *BMC Microbiol.* 17:9.

Suen, G., P. J. Weimer, D. M. Stevenson, F. O. Aylward, J. Boyum, J. Deneke, C. Drinkwater, N. N. Ivanova, N. Mikhailova, O. Chertkov, L. A. Goodwin, C. R. Currie, D. Mead, P. J. Brumm. 2011. The complete genome sequence of *Fibrobacter succinogenes* S85 reveals a cellulolytic and metabolic specialist. *PLoS One* 6:e18814.

Vyas D, A. W. Alemu, S. M. McGinn, S. M. Duval, M. Kindermann, and K. A. Beauchemin. 2018. The combined effects of supplementing monensin and 3-nitrooxypropanol on methane emission, growth rate, and feed conversion efficiency in beef cattle fed high-forage and high-grain diets. *J. Anim. Sci.* 96:2923–22938.

Vyas D, S. M. McGinn, S. Duval, M. Kindermann, and K. A. Beauchemin. 2016. Effects of sustained reduction of enteric methane emissions with dietary supplementation of 3-nitrooxypropanol on growth performance of growing and finishing beef cattle. *J. Anim. Sci.* 94:2024–2034.

Wallace R. J., J. A. Rooke, N. McKain, C-A. Duthie, J. J. Hyslop, D. W. Ross, A. Waterhouse, M. Watson and R. Roehe. 2015. The rumen microbial metagenome associated with high methane production in cattle. *BMC Genomics* 16:839.

Wright, A-D.G. and A. Klive (2010). Does the complexity of the rumen microbial ecology preclude methane mitigation? *Animal Feed Sci. Technol.* 166-167:248-253.

Wu, X., Huang, S., Huang, J., Peng, P., Liu, Y., Han, B., and Sun, D. (2021). Identification of the potential role of the rumen microbiome in milk protein and fat synthesis in dairy cows using metagenomic sequencing. *Animals.* 11:1247.

Xue, M.Y. H. Z. Sun, X. H. Wu, L. L. Guan, J. X. Liu. (2019). Assessment of rumen bacteria in dairy cows with varied milk protein yield. *J. Dairy Sci.*, 102:5031–5041.

Zhou M. I., E. Hernandez-Sanabria, and L. L. Guan. 2009. Assessment of the microbial ecology of ruminal methanogens in cattle with different feed efficiencies. *Appl. Environ. Microbiol.* 75:6524-6533.

Figure 1A. Ruminal microbes by size

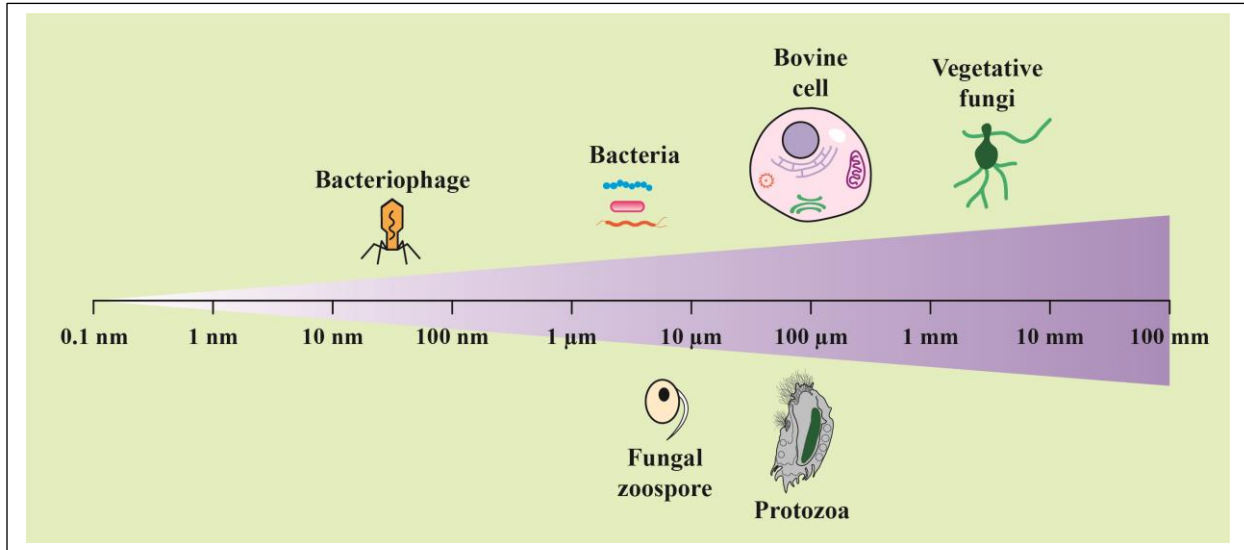


Figure 1B. Ruminal microbes by numbers

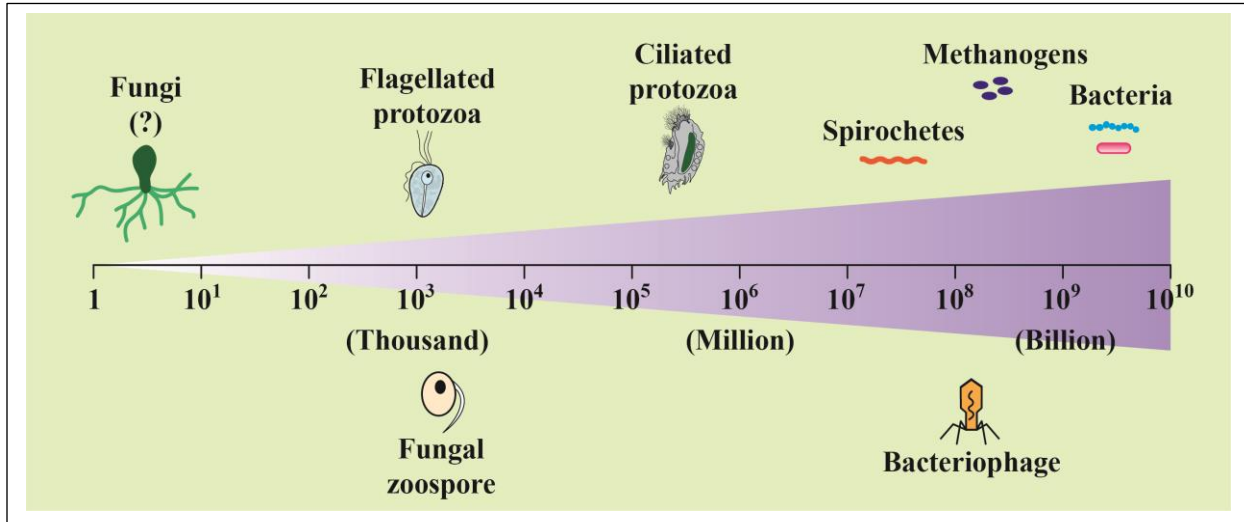


Figure 1C. Ruminal microbes by proportion

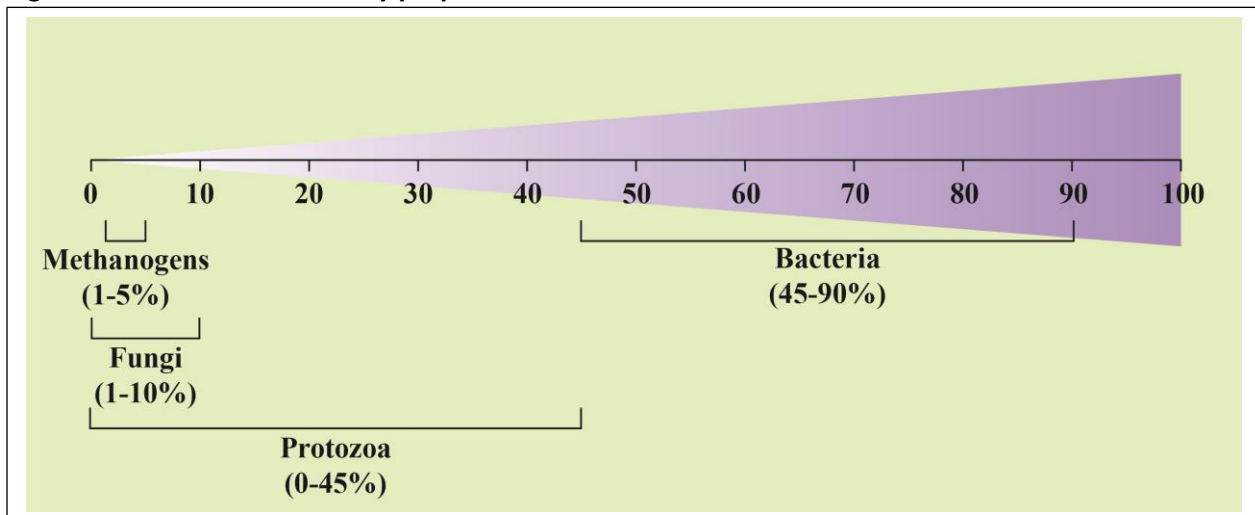


Figure 2. Hydrogen utilizing reactions in the rumen

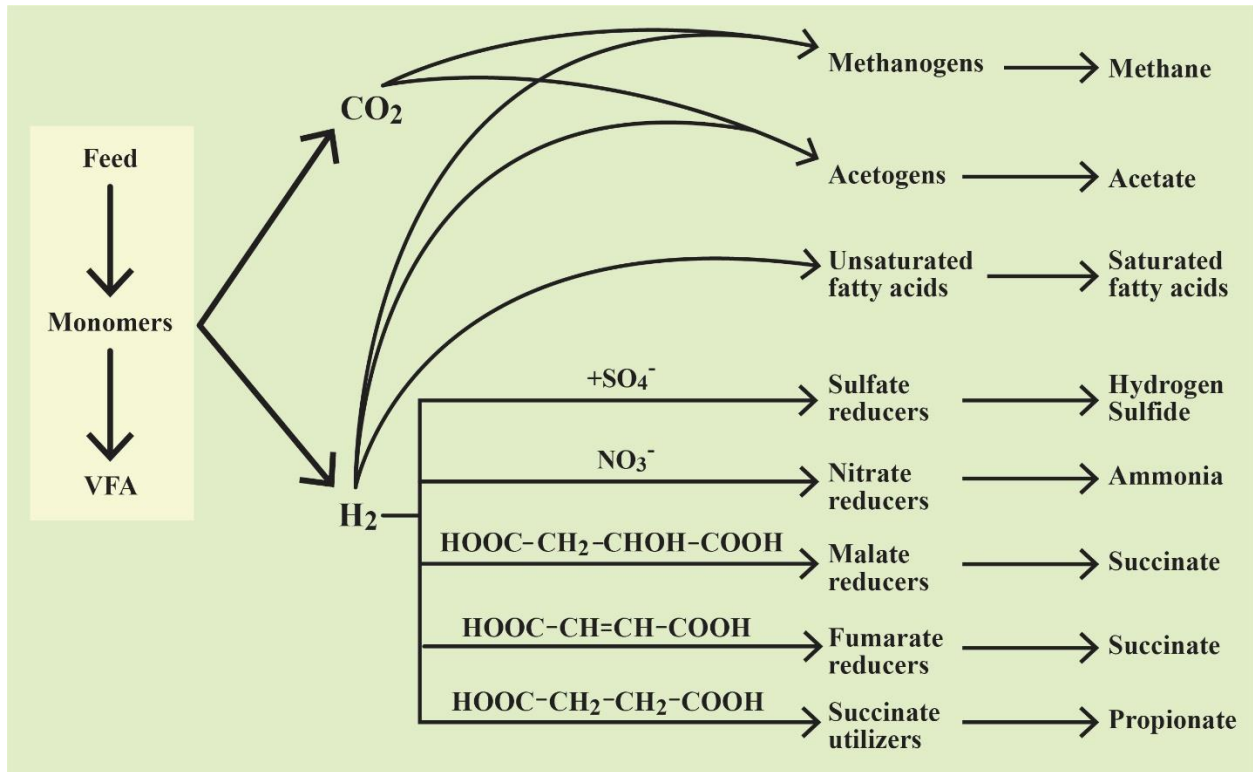


Figure 3. Interspecies hydrogen transfer in the rumen

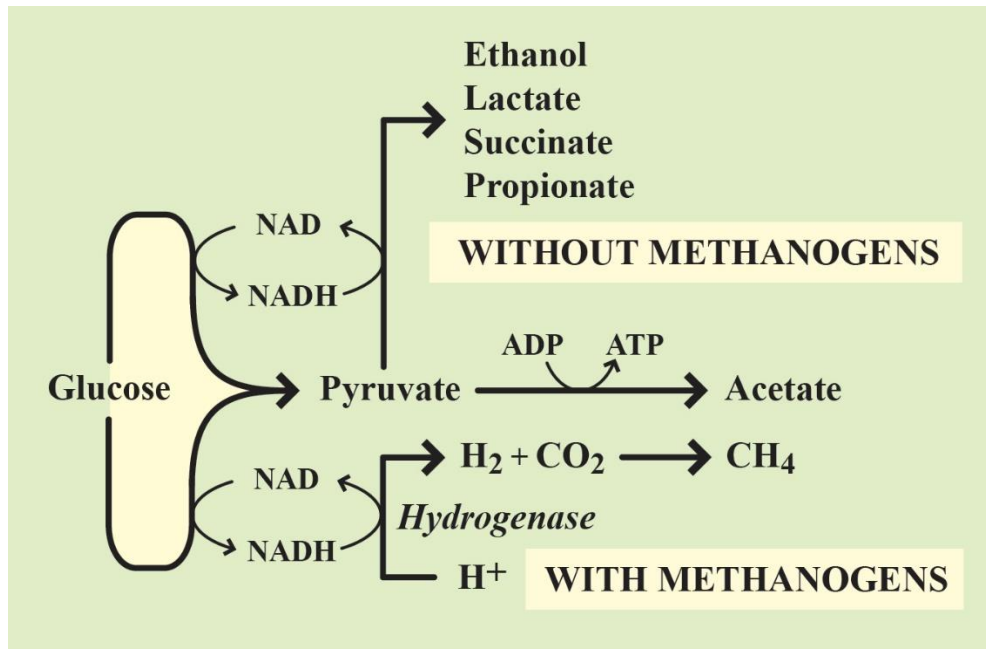


Figure 4. Major and minor pathways for methane production in the rumen

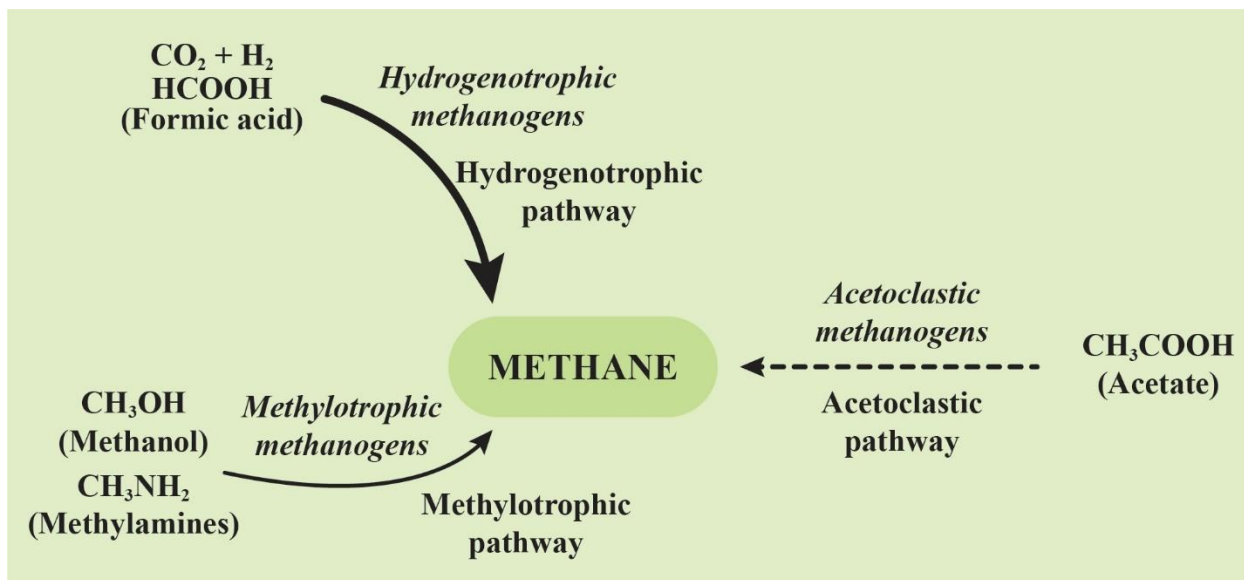
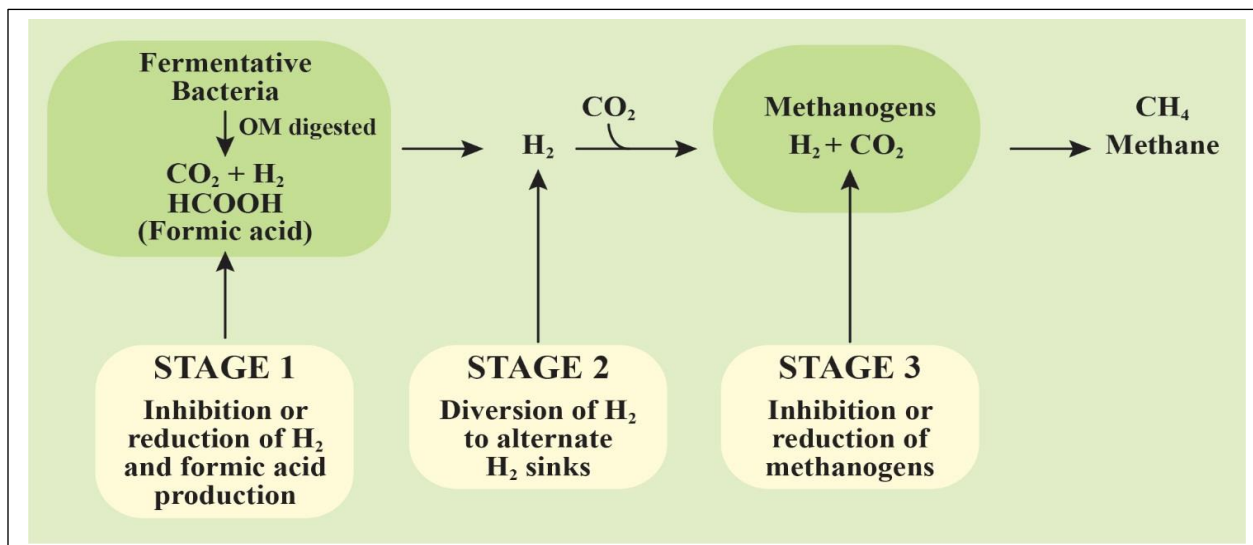


Figure 5. Stages in ruminal methanogenesis for intervention to inhibit methane production.



Economics of improved dairy reproduction considering conventional, sexed and beef semen

Albert De Vries
University of Florida
devries@ufl.edu

Introduction

Dairy cattle reproduction efficiency is important for profitability of dairy farms. Average 21-day cow pregnancy rate as measured by DHIA in 2016 was about 19% (De Vries, 2016) and is increasing. Phenotypic daughter pregnancy rates, as calculated by the CDCB (2022) from days open data, has increased since 2000 by at least 5 percentage points. Cow pregnancy rates greater than 30% are becoming more common. Breeding values for daughter pregnancy rate have increased less since 2000. Most of the improvement in reproduction efficiency in the last two decades is due to management.

We know from older studies that there is a diminishing value to greater reproduction efficiency. Although a 100% pregnancy rate is still the goal, this means that less could spent to continue to increase reproduction efficiency. For example, figure 1 shows how greater pregnancy rates are associated with increased profit per cow per year for six studies conducted a decade ago (Overton and Cabrera, 2017). In figure 1, net return gain is set at \$0 at 10% pregnancy rates for all studies. The increases in profit in figure 1 are a mixture of net gains that include the cost of the technology to achieve that change in pregnancy rate, and gross gains that do not include the cost to obtain the change in pregnancy rate. In all six studies, a greater pregnancy rate leads to a greater profit. Even in the two studies that report pregnancy rates over 30%, profit keeps increasing. The studies in figure 1 were all conducted assuming conventional semen. Calves were sold for a fixed price, and cow cull rates were independent of the number of calves produced. Genetics were not considered in these studies.

In the last decade we have seen a dramatic uptake of reproduction options like genomic testing, sexed semen, and beef-on-dairy (Fourdraine, 2022). The question is how the value of improving reproduction efficiency depends on these expanded options. One hypothesis is that dairy reproduction efficiency is worth more when using combinations of these options, compared to the traditional use of conventional semen only, because genetic merit of the herd can be higher and more valuable calves can be sold. The objective of this paper is therefore to explore how the value of improving reproduction efficiency depends on the combinations of sexed, conventional, and beef semen, with and without the use of genomic testing.

Herd budget calculator

Like the studies in figure 1, I used logic and detailed calculations to obtain results. A herd budget calculator spreadsheet was developed with the goal to evaluate the genetic and economic consequences of changes in prices, reproduction, and strategic mating. The calculator has virtual heifers and cows and many biological inputs such as milk production curves, feed intake, forced culling, and many prices. Herd profit is expressed per milking cow per year because profit should be expressed per most limiting factor, which is milking capacity on many farms.

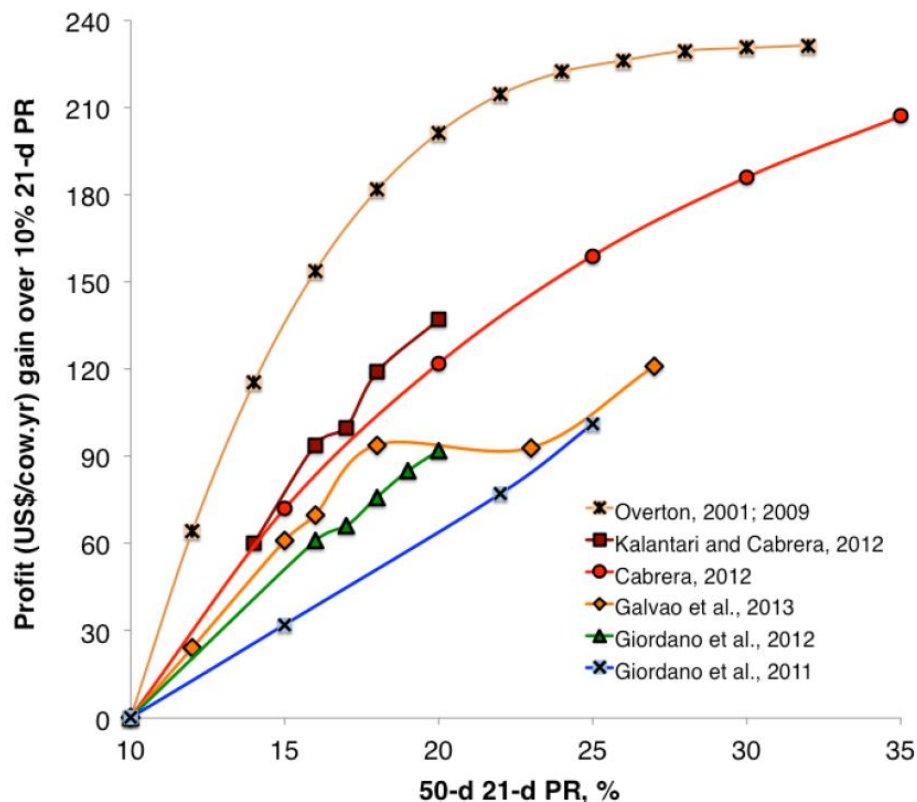


Figure 1. Profit gain of increasing 21-d pregnancy rate (PR) at a 50-day voluntary waiting period reported in six older studies that were conducted around a decade ago. Profit per cow per year was standardized at \$0 at 10% PR. The costs to obtain the increased pregnancy rates were included within some studies but not in others. Source: Overton and Cabrera (2017) (with permission). Published also in De Vries (2016). All studies assumed that only conventional semen was used. Genetics was not considered.

Strategic mating refers to how the different semen types are used within the herd. For example, it is now common to use sexed semen in heifers and beef semen in older cows. Crossbred calves out of a beef-on-dairy mating are generally worth more than surplus purebred dairy calves. Conventional semen may or may not be used. Perhaps genetic merit differences within the same lactation and breeding number are considered and the best animals receive sexed semen while others receive conventional or beef semen. Genomic testing may be used so animals can be better ranked for genetic merit, which may affect culling and mating decisions. Many combinations are possible. A strategic mating plan often depends on the farm's wish to produce a certain number of dairy heifer calves to replace cows in the future. It is usually not clear what strategic mating plan maximizes profitability.

Genetic assumptions

The calculator puts an economic value on the genetic lag of the herd. Genetic lag is the genetic difference between the best available service sires and the average cow in the herd. The idea is that sires contain the best available genetic “package” and cows are on average lagging in genetic merit. Genetic lag is therefore an opportunity cost. It is money not made because the genetics in the herd is older than what is available on the market.

Genetic progress in sires is approximately \$75 predicted transmitting ability (PTA) of Lifetime Net Merit Dollars (NM\$) per year (CDCB, 2022). The result is that on average the genetic merit in heifers is greater than in younger cows, and even greater compared to older cows. But there is genetic variation within the same age, as figure 2 shows. You can find some good cows with higher genetic merit than some heifers. Genomic testing improves the reliability of estimates of genetic merit, which means more certainty about their true genetic merit.

The value of improving reproduction efficiency depends on the genetic merit of the service sires and potential dams in several ways. First, we could create a surplus of dairy heifer calves and keep only the number we need to replace cows, by selling the genetically worst calves. This practice reduces the genetic lag. Secondly, we can use the genetic merit of individual heifers and cows for mating decisions, for example by mating heifers and cows with the highest genetic merit to sexed semen. A simpler strategy is to only look at the age of the animals for mating decisions. Age is a good predictor of genetic merit.

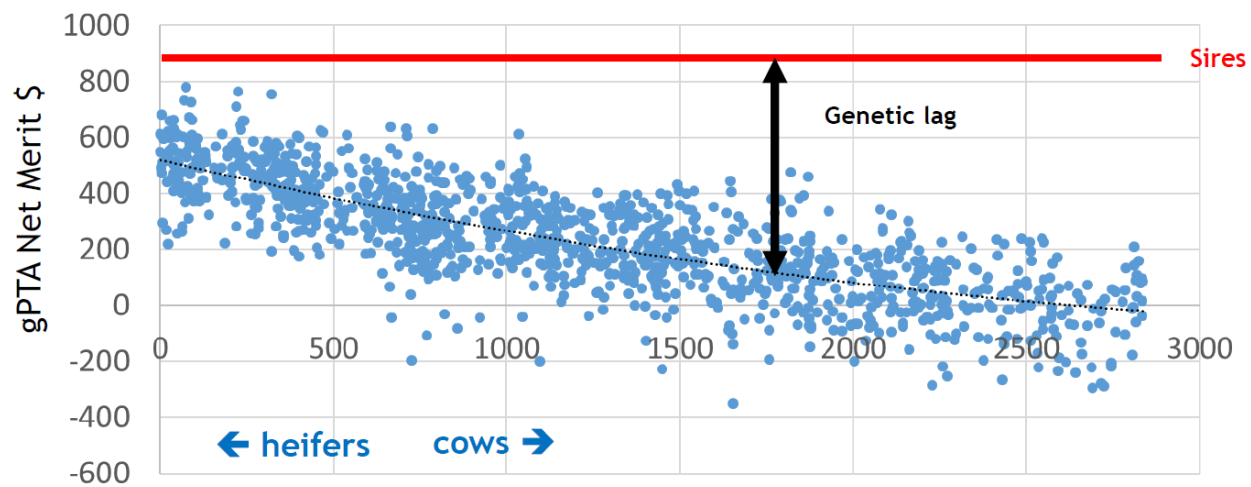
The goal of a genetics program is to reduce the genetic lag, but this goal needs to be balanced with the productive life of the animals in the herd. A high cow cull rate would result in a young herd but a short average productive life. Such a strategy would lead to high cow turn over costs, too few mature cows, and is likely less profitable (De Vries, 2020).

In the calculator, the genetic trend of PTA of NM\$ was set at \$75 per year. Within the same age, the variation in the PTA of NM\$ had a standard deviation of \$197 before selection of surplus dairy heifer calves (if any). The PTA of NM\$ of sires was set at \$1000. The traditional reliability PTA of NM\$ in calves was set at 20%. For heifers it was 21%. For cows in lactations 1 to 4+, the traditional reliability increased from 40% to 48%. While traditional reliabilities come for free, genomic testing of calves at an assumed cost of \$50 per test increased reliabilities from 71% for calves to 74% for cows in lactations 4+. Thus, the genomic test information could be used to first select surplus dairy calves better and later for mating decisions with beef, conventional, or sexed semen.

Crossbred (beef x dairy) calves were sold for \$200. Surplus dairy heifer and bull calves were sold for \$50 per head. The value of the kept dairy heifer calves was the result of the genetic merit of the sire and dam and, if any, surplus dairy calf selection. Insemination expenses were \$35 for sexed dairy semen, and \$15 for beef and conventional dairy semen. In addition, there were costs for feed, and other variable cost for heifer, milking cow, and dry cows. Mature cows produced more milk than first lactation cows.

Heifers were eligible for insemination between 400 and 550 days of age. Cows were eligible for insemination between 70 and 300 days in milk. The number of inseminations of open animals

depended on the 21-day service rate and conception rate (Table 1). Annual cow cull rate was set at 37% for the baseline reproductive efficiency level. The cow cull rate decreased when reproductive efficiency increased because fewer animals would be culled for failure to get pregnant on time.



1,247 animals genomic tested at the UF Dairy Unit

Figure 2. Genomic predicted transmitting ability (PTA) of the economic selection index Net Merit (NM\$) for 1247 animals at the University of Florida Dairy Unit. The graph illustrates the trend and variation in PTA of NM\$ of animals, and the genetic lag of two animals with the service sires, which are assumed to have a PTA of NM\$ of \$900. The goal of a genetics program is to reduce the genetic lag balanced with the longevity of the animals in the herd.

Lactations 0 (heifers) to 4 (older cows) each had four distinct breeding numbers with choices of the fraction of the type of semen (beef, conventional, sexed). Greater breeding numbers had choices equal to the 4th breeding number choices. Thus, there were 20 different breeding opportunities (ages) where the fractions of semen types could be varied. For example, the mating decisions for first inseminations in first lactation cows could be 26% beef semen, 43% conventional semen, and 31% sexed semen (total 100%).

The herd budget calculator was used to evaluate profit per milking cow per year at five levels of reproduction efficiency. These five levels are shown in table 1. In increasing order of reproduction efficiency, the levels are referred to as baseline, improved, good, great, and best.

Table 1. Input assumptions of 21-d service rates and conception rates for five levels of reproduction efficiency.

	baseline	improved	good	great	best
Heifers					
21-day service rate	70%	70%	70%	70%	70%
beef semen conception rate	44%	50%	56%	62%	68%
conventional semen conception rate	44%	50%	56%	62%	68%
sexed semen conception rate	40%	46%	51%	57%	62%
Lactation 1					
21-day service rate	60%	60%	60%	60%	60%
beef semen conception rate	36%	41%	46%	51%	56%
conventional semen conception rate	36%	41%	46%	51%	56%
sexed semen conception rate	32%	36%	41%	45%	50%
Lactation 2+					
21-day service rate	60%	60%	60%	60%	60%
beef semen conception rate	32%	36%	41%	45%	50%
conventional semen conception rate	32%	36%	41%	45%	50%
sexed semen conception rate	28%	32%	36%	40%	43%

Conception rates of second and later inseminations are 90%, 80%, 70% of the conception rate in the table for heifers. For cows, later inseminations have 90% of the conception rate in the table. Sexed semen conception rates of $\geq 4^{\text{th}}$ inseminations are 72% of those in the table.

Mating Strategies

In addition, four mating strategies were evaluated at each of the five levels of reproduction efficiency. Each strategy was evaluated with and without genomic testing.

The **conventional** strategies used only conventional semen on all heifers and cows. Surplus dairy heifer calves were sold. The genetically best dairy heifer calves were kept after ranking based on traditional or genomic reliabilities. Genetic reliabilities were only used to select surplus dairy calves, but obviously not for mating decisions.

The **beef-by-age** strategies used 100% sexed semen on heifers and the youngest cows until enough dairy heifer calves were produced to replace culled cows. The remainder of the cows were inseminated with beef semen. No conventional semen was used. Genetic reliabilities were only used for some mating decisions, but not to select surplus dairy calves because close to no extras were produced.

The **beef-by-PTA** strategies also used only beef and sexed semen. Heifers and cows below a threshold of PTA of NM\$ received beef semen whereas animals above the threshold received sexed semen. The threshold was varied such that just enough dairy heifer calves were produced to replace culled cows. Genetic reliabilities were used extensively for mating decisions because a fraction of the dams was below the threshold in all 20 breeding numbers.

The **optimal** strategies were discovered by varying the fractions of beef, conventional and sexed semen for every one of the 20 breeding numbers, and allowed the generation of surplus

dairy calves. This optimization was done with a non-linear solver (a mathematical optimization technique). Because a great number of combinations are possible, there was no guarantee that the absolute best mating strategy could be found.

All analyses were done for a herd with 1000 milking cows.

Results

Figure 3 shows results for the (near) optimal mating strategies using genomic testing of calves, to illustrate some of the key statistics of the calculator. The 20 breeding numbers are shown vertically. The left column within one of the five reproduction efficiencies shows the fraction beef semen (Be), the middle column the fraction conventional semen (Co), and the right column shows the fraction sexed semen (Se). A bigger fraction is identified by a thicker line. The three fractions add up to 100%.

The breeding numbers show an increasing fraction of beef semen for older cows. Sexed semen is primarily used in heifers and first lactation cows. There is some use of conventional semen, primarily in breedings 4 and greater in heifers. The strategies generate only the number of dairy heifer calves needed to replace culled cows (surplus 0%). The annual cow cull rate decreased from 37% for the baseline (lowest) reproduction efficiency to 30% for the best reproduction efficiency. Therefore, the number dairy heifer calves kept per year decreased from 558 to 397 per 1000 milking cows per year. The cow pregnancy rate increased from 18% for the baseline scenario to the 30% for the best scenario.

The lower annual cow cull rate increased the average age of the cows in the herd from 3.91 years for the baseline reproduction efficiency to 4.13 for the best reproduction efficiency. This implies that the genetic lag increased because more cows got older. However, the age of the dams of the kept dairy calves decreased from 2.78 years to 2.66 years. The PTA of NM\$ of the kept dairy heifer calves increased by \$13.

Profit per milking cow increased from \$533 for the baseline reproduction efficiency to \$811 for the best reproduction efficiency. This gain is mostly due to more mature cows, lower replacement costs, reduced genetic lag, and an increase in the value of sold crossbred calves. Therefore, at 30% cow pregnancy rate (best), the profit per milking cow per year was \$278 greater than at 18% cow pregnancy rate (baseline).

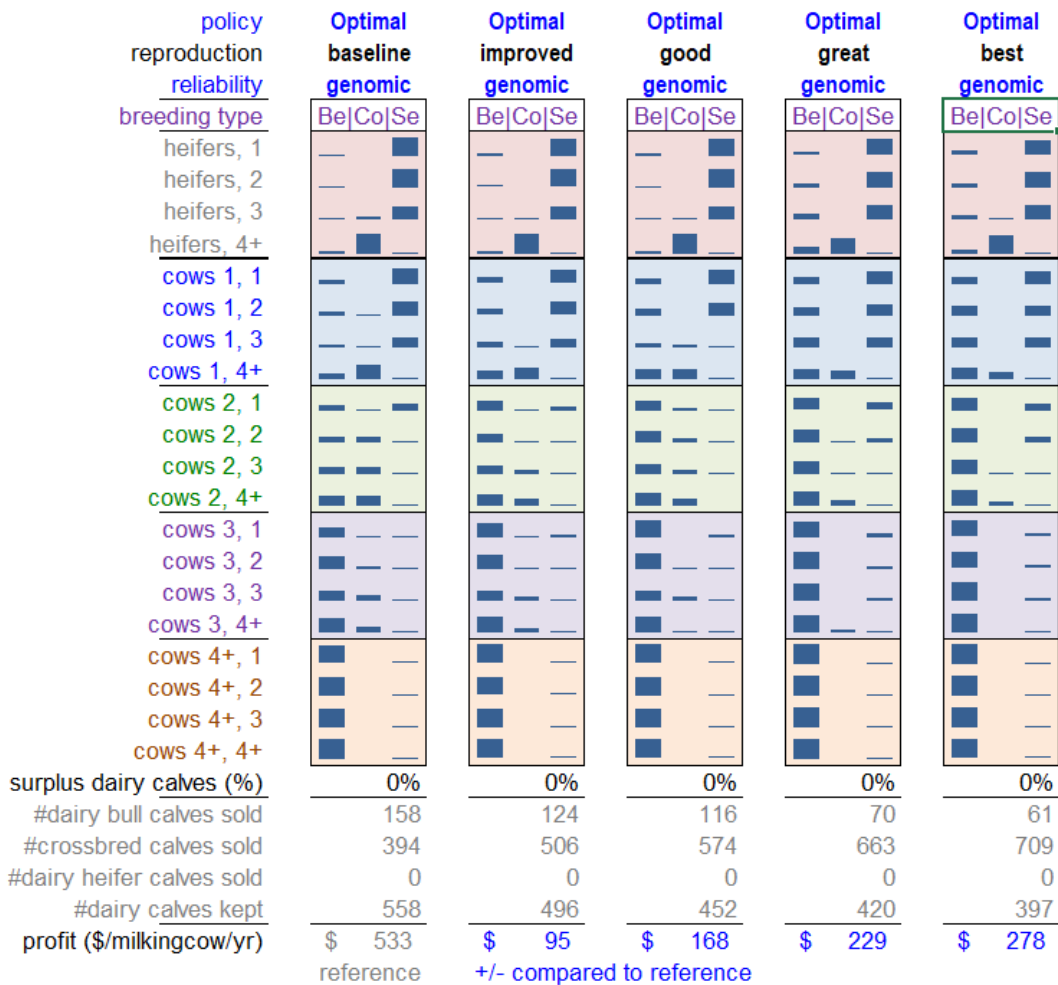


Figure 3. Visual display of the mating strategies for the optimal scenario with genomic testing for the five reproduction efficiencies in a herd of 1000 milking cows. The left column is the fraction beef semen (Be), the middle column is the fraction conventional semen (Co) and the right column is the fraction sexed semen (Se). A thicker line implies a greater fraction. Be + Co + Se = 100%. Profit per milking cow per year increased by \$278 in the best reproduction efficiency compared to the baseline reproduction efficiency.

Table 2 shows profit per milking cow per year for all evaluations. The cow pregnancy rates varied slightly among mating strategies. The beef-by-age, beef-by-pta, and optimal strategies all resulted in substantial greater profit compared the use of conventional semen only. The optimal strategy was not really optimal because the profit was slightly less than for the beef-by-age and beef-by-pta strategies. This is due to the solver not being able to find the best strategy.

Both optimal strategies did not generate a surplus of dairy heifer calves. Genomic testing of dairy calves was only used later in their lives for mating decisions. A beef-by-age mating strategy did not benefit from any genomic testing information. Therefore, its profit was lower for the genomic reliabilities compared to the traditional reliabilities for all five reproduction

efficiencies. Genomic testing was profitable in the other strategies when reproduction efficiency was great to best, but not at the lower reproduction efficiencies.

Table 2 shows that profitability clearly increased with better reproduction efficiency. Second, the increase was greater for all strategies when genomic testing is used. Third, the increase was the lowest for the conventional semen strategies, both with traditional and genomic reliabilities. The value of improved reproduction efficiency was greater in the strategies where more valuable beef calves were produced. Figure 4 shows increases in profit per milking cow per year for the mating strategies with genomic testing. The beef-by-pta and optimal strategies benefit the most from increases in reproduction efficiency. The finding that improvements in reproduction efficiency is greater when beef-on-dairy is practiced was also reported in Sweden (Clasen et al., 2020) and Wisconsin (Cabrera, 2022).

Table 2. Profit per milking cow per year (\$) for the five reproduction efficiencies (baseline to best), four mating strategies, and with or without genomic testing.

	baseline	improved	good	great	best
cow pregnancy rate	18%	21%	24%	28%	31%
mating strategy					
		traditional reliabilities			
conventional	489	541	583	617	646
beef-by-age	546	634	700	753	795
beef-by-pta	549	638	707	763	808
optimal	547	626	707	764	805
mating strategy					
		genomic reliabilities			
conventional	471	533	582	621	654
beef-by-age	523	614	683	737	779
beef-by-pta	539	634	707	765	813
optimal	533	628	701	762	811

Conclusions

There is a decreasing economic return on improving reproduction efficiency when reproduction efficiency is already higher. Reproduction options such as sexed semen, beef semen, and genomic testing allow dairy producers to make strategic mating decisions on heifers and cows. Strategic mating opportunities increase the value of improving reproduction efficiency significantly compared to the use of conventional semen only. Mating strategies beef-by-age and beef-by-pta are likely to be near optimal.

References

- Cabrera, V. 2022. Economics of using beef semen on dairy herds. JDS Communications 3 (article in press) <https://doi.org/10.3168/jdsc.2021-0155>
- CDCB (Council on Dairy Cattle Breeding). 2022. Genetic and phenotypic trend. Daughter Preg Rate. <https://queries.uscdcb.com/eval/summary/trend.cfm> Accessed 1/9/2022

Clasen, J. B., M. Kargo, S. Østergaard, W. F. Fikse, L. Rydhmer, and E. Strandberg. 2020. Genetic consequences of terminal crossbreeding, genomic test, sexed semen, and beef semen in dairy herds. *Journal of Dairy Science* 104:8062–8075. <https://doi.org/10.3168/jds.2020-20028>

De Vries, A. 2016. What is the optimal pregnancy rate? Is being too efficient a benefit or hindrance? *Proceedings Dairy Cattle Reproduction Council*, Columbus, OH. Pages 5-13.

De Vries, A. 2020. Symposium Review: Why revisit dairy cattle productive lifespan? *J. Dairy Sci.* 103: 3838-3845. <https://doi.org/10.3168/jds.2019-17361>

Fourdraine, R. 2021. Maximize returns from your beef x dairy breeding program in 2022. *Progressive Dairy*, 12/31/2021. <https://www.progressivedairy.com/topics/a-i-breeding/maximize-returns-from-your-beef-x-dairy-breeding-program-in-2022>

Overton, M.W. and V.E. Cabrera. 2017. Monitoring and quantifying the value of change in reproductive performance. In: Beebe DK (ed.), *Large dairy herd management* (3rd edn.), USA. pp. 549-564.

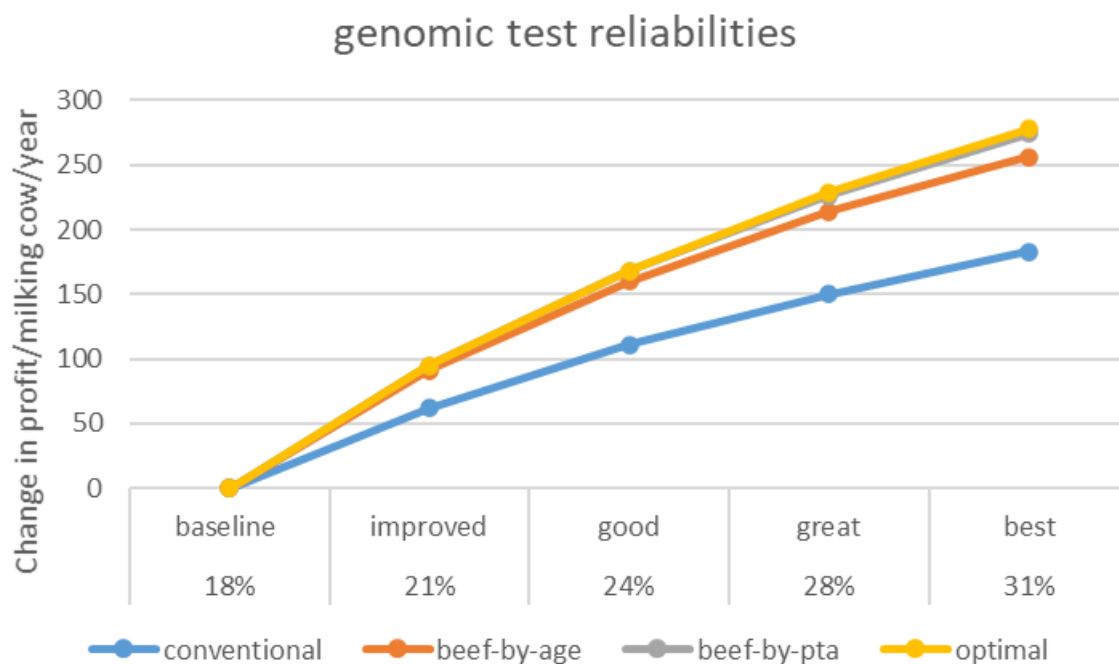


Figure 4. Change in profit per milking cow per year compared to the lowest reproduction efficiency (baseline; 18% cow pregnancy rate) for four mating strategies. The beef-by-age, beef-by-pta, and optimal mating strategies show greater benefit from increased reproduction efficiency than the mating strategy that uses conventional semen only.

Predicting economic implications of weaning programs in young dairy calves

Jim Quigley, PhD, PAS, Dpl. ACAS
Cargill, Inc.
jqigley@provimi-na.com

Introduction

Considerable research has been reported in the past 15 years regarding effects of feeding and management on growth of calves. Traditional methods of feeding limited milk or calf milk replacer (**CMR**) have generally been replaced by increased quantities of liquids prior to weaning as several studies have indicated that early life nutrition may impact future milk production (Soberon et al., 2012, 2013; Gelsinger et al., 2016). Ages at weaning have also been evaluated (Eckert et al., 2015; Khan et al., 2016) even to 17 weeks of age (Schwarzkopf, et al., 2019). Incorporating new data into recommendations for on-farm management is required, particularly to meet farm-specific goals such as maximal rate of gain vs. lowest cost.

In the past 10 years, the calf research team at Provimi North America (a Division of Cargill, Inc.) has evaluated numerous changes to liquid feeding programs and calf starter formulation to better understand their effects on health and growth to four months of age. Many of these studies were summarized in a meta-analysis by Hu et al. (2020). We also reported several studies that evaluated composition of calf starters and indicated that starch supports greater levels of BW gain compared to NDF (Hill et al., 2010; Chapman et al., 2016; Hill et al., 2016a, b; Dennis et al., 2018a, b; Quigley et al., 2018). We found that starch more rapidly stimulates rumen development and nutrient digestibility and differences in NDF digestion may still exist as late as 16 weeks of age. Further, greater amounts of liquid fed preweaning delays rumen development and slows changes in nutrient digestion. The net effect of high fiber starters and high milk allowances is delayed rumen development. Weaning calves without these considerations often results in calves with poor post-weaning growth (“weaning slump”) and predisposition to disease due to stress. Our data suggests that formulation of starter and amount of liquid fed can affect preparation for weaning and should be considered in any calf management recommendations.

Most farmers monitor neither calf BW nor intake on a routine basis. Therefore, it is difficult to evaluate effects of changes in nutrition or management in real time. However, it is possible to use a modeling approach to estimate effects of different programs on preparation for weaning and expected growth in these programs. We have developed a model of growth that allows modification of feeding programs to meet on-farm goals and predict growth, intake, cost, and efficiency to 4 months of age.

Assumptions Used in Model Development

Model framework

The underlying structure for modeling calf growth is prediction of nutrient requirements and estimation of nutrient supply using the 2001 Dairy NRC calf sub-model (NRC, 2001). This model predicts BW gain allowed by metabolizable energy (**ME-gain**) supply and also BW gain allowed by supply of apparently digested protein (**ADP-gain**). The lesser of ME-gain and ADP-gain is the expected BW gain for a calf at a given point in time using feeds and environment provided to the model.

Prediction of BW gain at a given day was expanded to predict daily growth from 3 to 112 days of age. A calf is assumed to weigh 42 kg at 3 days of age, although this may be adjusted by the user. Then, ME-gain and ADP-gain are predicted using the 2001 dairy sub-model. Then, BW on day 4 is calculated as day 3 BW plus the lesser of ME-gain or ADP-gain on day 3. The process is repeated for day 4 to day 112, resulting in a BW growth curve. The model calculates feed intake, so feed costs provided to the model allow calculation of total feed costs and efficiencies of growth. Daily intake of liquid (milk or CMR) is entered by the user. Daily dry feed intake (**DFi**) from concentrates and forages is predicted by the model using inputs for ages at which feeds are offered and any upper limits on voluntary intake.

The initial growth model based on the 2001 Dairy calf sub-model over-predicted growth of calves, particularly in the two months of life. We attempted to improve prediction of nutrient supply by adjusting digestibility of liquids, ME values of calf starters, and improved predictions of DFi.

es u ds

Nutrient supply models generally ignore the maturation of intestinal digestibility with advancing age (e.g., NRC, 2001). However, several studies have documented effects of age on digestibility of nutrients in liquid feed for preweaned dairy calves. For example, Terosky et al. (1997) reported that N digestibility in calves fed milk replacers containing whey or skim milk proteins increased from approximately 70% to 90% from 2 to 8 wk of age. Others reported similar increases with advancing age, generally concluding that digestibility increases to approximately 3 wk (Arieli et al., 1995; Terosky et al., 1997; NRC, 2001) or 5 wk (Guilloteau et al., 2009) of age. Recently, Quigley et al. (2021b) reported results of a meta-analysis that determined effects of age on digestibility on liquid feeds in calves prior to weaning. Change in DM digestibility with advancing age is in Figure 1. Models of nutrient supply from milk or milk replacer were adjusted in the updated model using equations from Quigley et al. (2021b) for DM, N, and fat and actose digestibility. These adjustments to the base model reduce BW gain in the first 30 days of life, particularly when calves are fed large amounts of CMR.

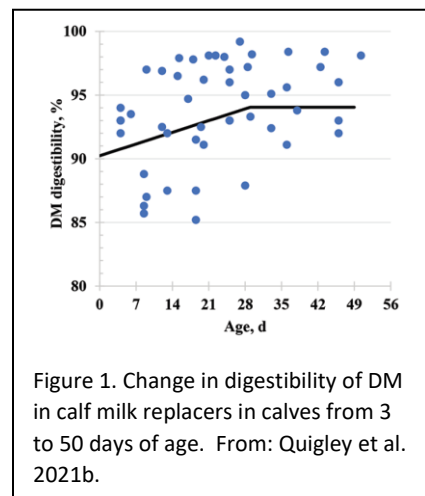


Figure 1. Change in digestibility of DM in calf milk replacers in calves from 3 to 50 days of age. From: Quigley et al. 2021b.

es s e

A critical factor affecting growth of calves during the first 4 months of life is their ability to digest nutrients from dry feed. Prior to initiation of rumen development, digestion of fibrous feeds is extremely limited in young calves as digestibility of NDF in calves prior to about 8 weeks of age is quite low (Quigley et al., 2019a, b). Further, digestion of starch is limited for the first six weeks or so of life (reviewed in Quigley et al., 2019a); therefore, the calf's ability to extract ME from starters is clearly less prior to significant rumen development. The 2001 Dairy NRC calf sub-model ignores effects of rumen development on calculation of ME from dry feed. This results in an over-estimation of energy available to the calf. Energy limits growth in most scenarios, so this over-estimate of ME supply artificially increases growth predictions. Recently, we published a comprehensive review of factors affecting changing digestion in calf starters (Quigley et al. 2019a, b). We reported that intake of non-fiber carbohydrate (NFCi) is the key driver in hanging nutrient total tract nutrient digestion in calves to 4 months of age. Early in life, digestion of carbohydrates is limited; calculation of calf starter ME indicated that for the first few weeks of life, calves extracted only 40-60% of the energy in starters.

Thus, calf performance is over-estimated.

We also reported that cumulative NFCi rather than NFCi at a given point in time (Quigley et al., 2019b). This is logical, as NFCi is associated with rumen development, and cumulative NFCi would be more highly associated with total rumen development rather than NFCi on a given day. We found that when cumulative NFCi reached 15 kg, the ME in calf starters calculated using measured digestibility values were similar to those predicted by the 2001 Dairy NRC. Incorporating this adjustment into predictions of calf growth dramatically improves predictions of growth in calves to 4 months of age.

The ME values of concentrates and forages are adjusted by using adjustments in Quigley et al. (2019b) based on cumulative NFCi. The model determines daily NFCi and adjusts concentrate and forage ME. Result of this adjustment is reduction of ME supply and reduced rate of gain, particularly prior to weaning. High forage diets or starters containing more NDF also reduce ME supply and reduce BW gain.

eed e

Predictions of dry feed intake in calves less than about 4 months are quite limited. Tedeschi and Fox (2009) predicted intake in Holstein calves to 200 d fed a diet of milk and ad libitum forage to simulate conditions of beef calves. Silva et al. (2019) reported two non-linear equations to predict calf starter intake in Brazilian Holstein and Holstein × Gyr calves to 64 d of age. Separate models were developed for calves fed more or less than 5 L of milk per day. Predictions did not follow calves after weaning, nor did equations accurately predict intake when equations were used in data from calves in the U.S. (unpublished data). Also, the 2001 Dairy NRC did not predict DFi in calves, but assumed that calves would consume sufficient ME from dry feed to meet ME requirements after ME intake from milk or milk replacer was considered.

Recently, we developed several linear and non-linear equations to predict dry feed intake in calves from zero to four months of age under various feeding conditions, including high and low levels of milk feeding, ages at weaning, composition of starter, and inclusion of forage (Quigley et al., 2021a). The data set used in model development contained more than 60,000 individual daily observations of intake from 1,235 Holstein calves collected from 30 experiments at our research stations in the United States and Europe. The simplest non-linear equation was $1.4362 \times e^{[-4.6646 + 0.5234 \times \text{MEgap}] \times \text{EXP}(-0.0361 \times \text{Age})} + 0.0025 \times \text{Age} \times \text{MEgap}$ ($R^2 = 0.92$, concordance correlation coefficient = 0.96, and mean square error of prediction = 0.11 kg); where MEgap (Mcal/d) = difference of daily metabolizable energy (ME) requirement and ME intake from milk replacer; Age = age of calf (d) from 3 to 114. The ME requirement was based on 2001 Dairy NRC models.

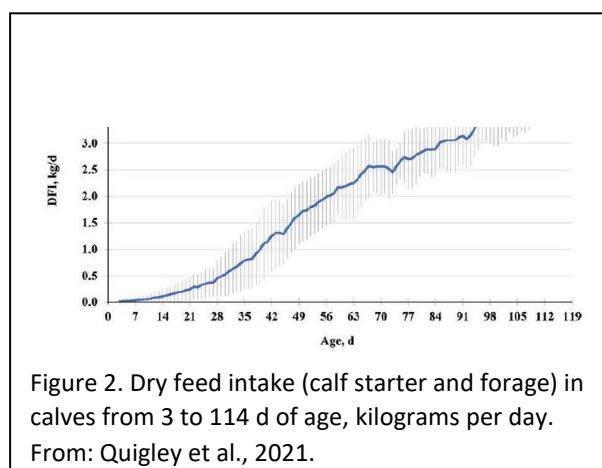


Figure 2. Dry feed intake (calf starter and forage) in calves from 3 to 114 d of age, kilograms per day. From: Quigley et al., 2021.

Use of this updated DFI prediction equation increased the accuracy and precision of growth predictions in the model and made growth predictions more sensitive to intake of liquid (which reduces DFi and slows rumen development).

Application

We adjusted the 2001 Dairy calf sub-model with updated models of DFI and feed digestibility to predict calf performance to 4 months of age. Equations to predict DFi, CMR digestion and ME content of dry feed were built into a growth prediction engine for calves from 0 to 4 months of age, using Microsoft Excel. The application is called “GPS LITE” for “Growth Prediction System LITE”. Inputs include calf birth BW, feed composition, liquid feeding program, ages at which feeds are offered, and limitations (maximum amounts of feeds offered). The program calculates DFi and digestibility of nutrients from CMR and dry feed to calculate ME and ADP supply, then estimates growth for each using requirement equations from the 2001 Dairy NRC calf sub-model. Average daily gain using the minimum of ME-allowable and ADP-allowable gain is added to the BW on a given day to calculate change in BW from zero to four months of age.

An example of the model is in Figure 3. In this example, inputs to the model are in yellow highlighted cells. Calves are offered a CMR containing 24% CP and 17% fat from day 3 of age. Starter is offered from day 3, Grower feed from day 57 and forage (grass hay containing 8% CP and 60% NDF) is offered from day 42. Maximum offered is set to 99 kg, meaning that there is no maximum intake. Forage is offered for ad libitum consumption. The Liquid feeding program includes offering calves 480 g of CMR solids per day to d 7, then 1,200 g/d from d 7 to d 42 and 800 g/d from day 43 to weaning on d 49.

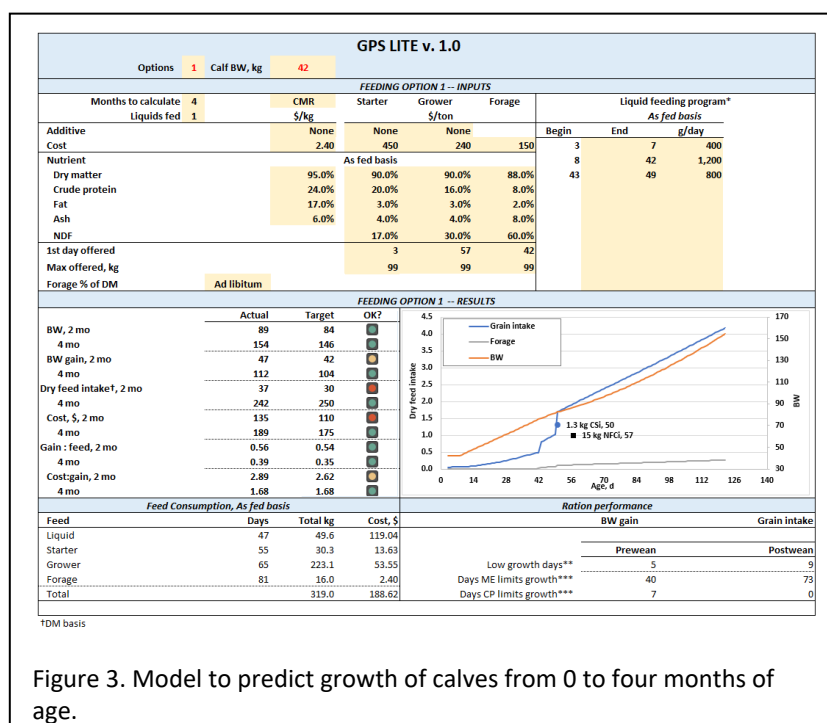


Figure 3. Model to predict growth of calves from 0 to four months of age.

Results are presented in the middle section of the spreadsheet. Predicted growth, intake, cost, and efficiency statistics are reported at two and four months and compared to targets based on doubling birth BW by two months of age and growth of 1 kg/d from two to four months. Targets for costs are based on an optimal growth scenario previously determined. Total intake and intake of each feed are reported in the bottom left quadrant of the results worksheet.

The graph in the middle section of the Worksheet displays changes in BW and intake of concentrates and forage. Also, predicted ages at which calves consume 15 kg of NFCi and 1.3 kg of DFi are reported. Critical value of 15 kg of NFCi is based on data from Quigley et al. (2019a, b) and 1.3 kg of DFi is based on a meta-analysis conducted as part of the 2021 Nutrient Requirements of Dairy Cattle (2021 NASEM) wherein the age at which contribution of microbial N to total abomasal N reached levels similar to those of adult cattle was determined as an

indicator of maturing rumen function. These two statistics provide a reasonable indication of the age at which calves will be ready to be weaned— i.e., the age at which calves will not experience low growth prior to or after weaning.

In this example, calves reach the two targets at 50 and 57 days, respectively. Because calves are weaned at 49 days of age, the number of “low growth” days is higher than optimal. We defined “low growth” days as <400 g of ADG/d preweaning and <700 g/d postweaning. Though data are limited, we believe that calves experience low ADG experience increased stress which may predispose them to disease. Calves experienced a total of 14 low growth days in this scenario, primarily due to a short weaning phase (7 days), and high amounts of milk consumed. This effectively reduced NFCi and rumen development, consequently reducing digestibility of nutrients and ME available from dry feed.

The liquid feeding program was adjusted slightly to reduce the number of low growth days (Figure 4). The amount of CMR offered during the first week was increased from 450 to 700 g/d, which eliminated the preweaning low growth days. To eliminate the low growth days postweaning (which occurred immediately after weaning), the weaning age was increased from 49 to 60 days, with a longer weaning transition and the amount of CMR offered was reduced from 800 to 700 g/d. This change effectively resulted in increased DFi and NFCi so that calves were fully prepared for weaning. Statistics in the graph in Figure 4) show that ages at which calves were prepared for weaning were 55 and 60 days.

Cost of a feeding program is calculated as the sum of daily intakes of each feed and totals calculated at 2 and 4 mo of age. Cost targets are initially set according to a moderate CMR feeding program (700 g of solids/d) with weaning at 60 d of age and predicted starter intake in the program. These inputs may be adjusted by the user. Efficiency of growth (gain to feed ratio) and cost per unit of BW gain are calculated. Efficiency targets are also calculated from gain and intake (cost) targets.

While scenario 2 reduces the months of age, number of low growth days, total cost, and cost per unit of number of low growth days, total cost, and cost per unit of BW gain are increased to the point at which they exceed targets by greater than about 10 to 15% (yellow and red traffic lights). Using an iterative process, it is possible to develop a third scenario that achieves the goal of zero low growth days while maintaining or reducing costs of the overall program.

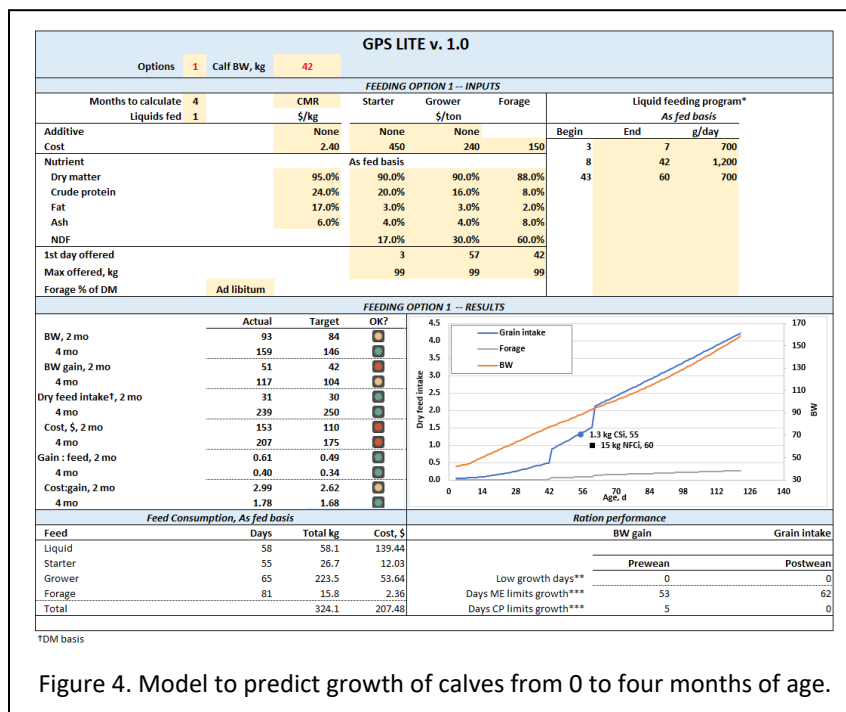


Figure 4. Model to predict growth of calves from 0 to four months of age.

In scenario 3, the amount of CMR was reduced, though age at weaning was maintained at 60 d, based on the statistics in the graph (Figure 5).

Comparison of the three scenarios at two and four months of age is in Table 1. Total intake was generally similar at 2 and 4 mo of age, though the costs varied significantly, both at 2 and 4 mo of age. Calves fed scenario 2 (greatest amount of CMR) gain more BW by 2 mo of age, but there was less of a difference by 4 mo of age due to lower post-weaning growth.

Calves in scenario 3 had greatest post-weaning BW gain greater intake and higher dry feed ME due to earlier DFi. The net result of these comparisons is that the most efficient scenario is #3, though feeding large amounts of CMR (scenario 2) results in greatest ending BW.

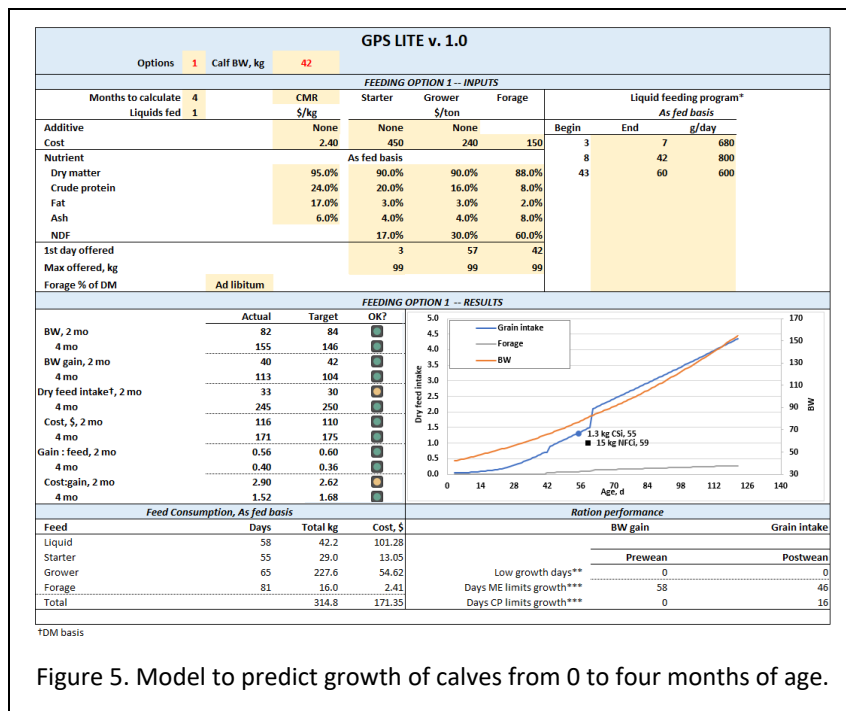


Figure 5. Model to predict growth of calves from 0 to four months of age.

Summary

Providing farmers with useful and actionable feeding and management recommendations based on sound science and research data will assist them to make reasoned decisions when actual on-farm data measurement is lacking. Simulation of growth can provide reasonable estimates of calf performance while simultaneously providing important statistics from which decisions can be made. Our approach provides farmers with reasonable estimates of growth and allows users to evaluate different feeding programs to meet on-farm goals.

Item	Scenario 1	Scenario 2	Scenario 3
Total CMR, kg	50	58	42
Low growth days	14	0	0
Dry feed intake, kg			
2 mo	37	31	33
4 mo	242	239	245
Cost, \$			
2 mo	135	153	116
4 mo	189	207	171
BW gain, kg			
2 mo	47	51	40
4 mo	112	117	113
G : F, kg/kg			
2 mo	0.56	0.61	0.56
4 mo	0.39	0.40	0.40
Cost/gain, \$/kg			
2 mo	2.89	2.99	2.90
4 mo	1.68	1.78	1.52

Table 1. Comparison of two management strategies for calves to 4 months of age.

References

- Arieli, A., J. W. Schrama, W. Van Der Hel, and M. W. A. Verstegen. 1995. Development of metabolic partitioning of energy in young calves. *J. Dairy Sci.* 78:1154–1162. [https://doi.org/10.3168/jds.S0022-0302\(95\)76732-7](https://doi.org/10.3168/jds.S0022-0302(95)76732-7).
- Chapman, C. E., P. S. Erickson, J. D. Quigley, T. M. Hill, H. G. Bateman II, F. X. Suarez-Mena, and R. L. Schlotterbeck. 2016. Effect of milk replacer program on calf performance and digestion of nutrients with age of the dairy calf. *J. Dairy Sci.* 99:2740–2747. <https://doi.org/10.3168/jds.2015-10372>.
- Dennis, T. S., F. X. Suarez-Mena, T. M. Hill, J. D. Quigley, R. L. Schlotterbeck, and L. Hulbert. 2018a. Effect of milk replacer feeding rate, age at weaning, and method of reducing milk replacer to weaning on digestion, performance, rumination, and activity in dairy calves to 4 months of age. *J. Dairy Sci.* 101:268–278. <https://doi.org/10.3168/jds.2017-13692>.
- Dennis, T. S., F. X. Suarez-Mena, T. M. Hill, J. D. Quigley, R. L. Schlotterbeck, R. N. Klopp, G. J. Lascano, and L. Hulbert. 2018b. Effects of gradual and later weaning ages when feeding high milk replacer rates on growth, textured starter digestibility, and behavior in Holstein calves from 0 to 4 months of age. *J. Dairy Sci.* 101:9863–9875. <https://doi.org/10.3168/jds.2018-15319>.
- Eckert, E., H.E. Brown, K.E. Leslie, T.J. DeVries, and M. A. Steele. 2015. Weaning age affects growth, feed intake, gastrointestinal development and behavior in Holstein calves fed an elevated plane of nutrition during the preweaning stage. *J. Dairy Sci.* 98:6315–6326. <https://doi.org/10.3168/jds.2014-9062>.
- Gelsinger, S. L., A. J. Heinrichs, and C. M. Jones. 2016. A meta-analysis of the effects of preweaned calf nutrition and growth on first-lactation performance. *J. Dairy Sci.* 99:6206–6214. <https://doi.org/10.3168/jds.2015-10744>.
- Guilloteau, P., R. Zabielski, and J. W. Blum. 2009. Gastrointestinal tract and digestion in the young ruminant: Ontogenesis, adaptations, consequences, and manipulations. *J. Physiol. Pharmacol.* 60(Suppl. 3):37–46
- Hill, T. M., H. G. Bateman II, J. M. Aldrich, and R. L. Schlotterbeck. 2010. Effect of milk replacer program on digestion of nutrients in dairy calves. *J. Dairy Sci.* 93:1105–1115. <https://doi.org/10.3168/jds.2009-2458>.
- Hill, T. M., J. D. Quigley, H. G. Bateman II, F. X. Suarez-Mena, T. S. Dennis, and R. L. Schlotterbeck. 2016a. Effect of milk replacer program on calf performance and digestion of nutrients in dairy calves to 4 months of age. *J. Dairy Sci.* 99:8103–8110. <https://doi.org/10.3168/jds.2009-2458>.
- Hill, T. M., J. D. Quigley, F. X. Suarez-Mena, H. G. Bateman II, and R. L. Schlotterbeck. 2016b. Effect of milk replacer feeding rate and functional fatty acids on dairy calf performance and digestion of nutrients. *J. Dairy Sci.* 99:6352–6361. <https://doi.org/10.3168/jds.2015-10812>.
- Hu, W., T. M. Hill, T. S. Dennis, F. X. Suarez-Mena, K. M. Aragona, J. D. Quigley, and R. L. Schlotterbeck. 2020. Effects of milk replacer feeding rates on growth performance of Holstein dairy calves to 4 months of age, evaluated via a meta-analytical approach. *J Dairy Sci.* 103(3):2217-2232. <https://doi.org/10.3168/jds.2019-17206>.

- Khan, M. A., A. Bach, D. M. Weary, and M. A. G. von Keyserlingk. 2016. Invited review: Transitioning from milk to solid feed in dairy heifers. *J. Dairy Sci.* 99:885–902. <http://dx.doi.org/10.3168/jds.2015-9975>.
- NASEM. 2001. *Nutrient Requirements of Dairy Cattle*. 8th rev. ed. National Academies Press.
- NRC. 2001. *Nutrient Requirements of Dairy Cattle*. 7th rev. ed. National Academies Press.
- Quigley, J. D., T. S. Dennis, F. X. Suarez-Mena, C. E. Chapman, and T. M. Hill. 2021a. Models to predict dry feed intake in Holstein calves to 4 months of age. *J Dairy Sci.* 104:5539-5556. <https://doi.org/10.3168/jds.2020-19581>.
- Quigley, J. D., T. M. Hill, T. S. Dennis, F. X. Suarez-Mena, and R. L. Schlotterbeck. 2018. Effects of feeding milk replacer at 2 rates with pelleted low-starch or texturized high-starch starters on calf performance and digestion. *J. Dairy Sci.* 101:5937–5948. <https://doi.org/10.3168/jds.2017-13851>.
- Quigley, J. D., W. Hu, J. R. Knapp, T. S. Dennis, F. X. Suarez-Mena, and T. M. Hill. 2019a. Estimates of calf starter energy affected by consumption of nutrients. 1. Evaluation of models to predict changing digestion. *J. Dairy Sci.* 102:2232–2241. <https://doi.org/10.3168/jds.2018-15353>.
- Quigley, J. D., W. Hu, J. R. Knapp, T. S. Dennis, F. X. Suarez-Mena, and T. M. Hill. 2019b. Estimates of calf starter energy affected by consumption of nutrients 2. Effect of changing digestion on energy content in calf starters. *Dairy Sci.* 102:2242–2253. <https://doi.org/10.3168/jds.2018-15354>.
- Quigley, J. D., T. S. Dennis, F. X. Suarez-Mena, T. M. Hill, and K. M. Aragon. 2021b. Meta-analysis of effects of age on intestinal digestibility of liquid feeds in young calves. *J. Dairy Sci. Comm.* <https://doi.org/10.3168/jdsc.2020-0057>.
- Schwarzkopf, S., A. Kinoshita, J. Kluess, S. Kersten, U. Meyer, K. Huber, S. Dänicke, and J. Frahm. 2019. Weaning Holstein calves at 17 weeks of age enables smooth transition from liquid to solid feed. *Animals (Basel)*. 9:1132. <https://doi.org/10.3390/ani9121132>.
- Soberon, F., E. Raffrenato, R. W. Everett, and M. E. Van Amburgh. 2012. Pre-weaning milk replacer intake and effects on long-term productivity of dairy calves. *J. Dairy Sci.* 95:783–793. <https://doi.org/10.3168/jds.2011-4391>.
- Soberon, F., and M. E. Van Amburgh. 2013. Lactation Biology Symposium: The effect of nutrient intake from milk or milk replacer of preweaned dairy calves on lactation milk yield as adults: A meta-analysis of current data. *J. Anim. Sci.* 91:706–712. <https://doi.org/10.2527/jas.2012-5834>.
- Silva, A. L., T. J. De Vries, L. O. Tedeschi, and M. I. Marcondes. 2019. Development of equations, based on milk intake, to predict starter feed intake of preweaned dairy calves. *Animal* 13:83–89. <https://doi.org/10.1017/S1751731118000666>.
- Tedeschi, L. O., and D. G. Fox. 2009. Predicting milk and forage intake of nursing calves. *J. Anim. Sci.* 87:3380–3391. <https://doi.org/10.2527/jas.2009-2014>.
- Terosky, T. L., A. J. Heinrichs, and L. L. Wilson. 1997. A comparison of milk protein sources in diets of calves up to eight weeks of age. *J. Dairy Sci.* 80:2977–2983. [https://doi.org/10.3168/jds.S0022-0302\(97\)76264-7](https://doi.org/10.3168/jds.S0022-0302(97)76264-7).

The value of forage quality when feeding dairy cows

Marcos Inacio Marcondes¹, Ícaro Rainyer Rodrigues de Castro², Marcelo Barros de Abreu², Luiz Ferraretto³

¹Washington State University, Pullman, WA 99164

²Universidade Federal de Viçosa, Viçosa, MG, Brazil, 36570-900

³University of Wisconsin, Madison, WI, 53706

Introduction

Dairy farms are looking for cost-effective feeding strategies to find opportunities to increase profitability. In this sense, feedstuffs correspond to about 40 to 60% of the total milk production cost. Thus, nutritional programs start with adequate forages programs. In the United States, corn silage is the most common ensiled crop used to feed dairy cows (Ferraretto et al., 2018). Corn silage provides a large amount of energy per kilo of dry matter than other traditional forages (Grant and Adesogan, 2018). Starch and fiber are the primary sources of energy for dairy cows fed corn silage-based diets and, therefore, understanding these components and how they change their digestibility is essential to improve milk production or reduce feed costs through enhanced feed efficiency.

It is important to highlight that corn silage has higher starch than other common cereal grains such as barley, oats, sorghum, and wheat compared (Table 1). The total energy available to the cow is usually a function of the dietary starch and its total tract starch digestibility. The starch digestibility changes according to the differences in cereals, as described in Table 1.

Table 1. Starch composition and digestibility of different cereal grains.

Cereal grain	Starch, % of DM	Ruminal Starch Digestibility, % of starch intake	Total Tract Starch Digestibility, % of starch intake
Barley	57.8	70.8 (46.1 - 91.0)	94.3 (76.1 - 99.5)
Corn	70.4	53.2 (9.7 - 80.2)	91.7 (69.5 - 99.4)
Oats	44.6	NA	NA
Sorghum	72.3	48.1 (NA)	83.5
Wheat	67.6	78.9 (59.1 - 95.1)	93.9 (86.3 - 99.1)

Starch

Dairy farmers are seeking silage-specific hybrids to address the high energy requirements of high-producing dairy cows. In this sense, starch is the most significant energy source in the corn silage, ranging from 26.8 to 36.8 % of DM in the last year (Cumberland lab; Figure 1). The high starch on current hybrids is linked to specialized selection programs in the last decades (Ferraretto and Shaver, 2015).

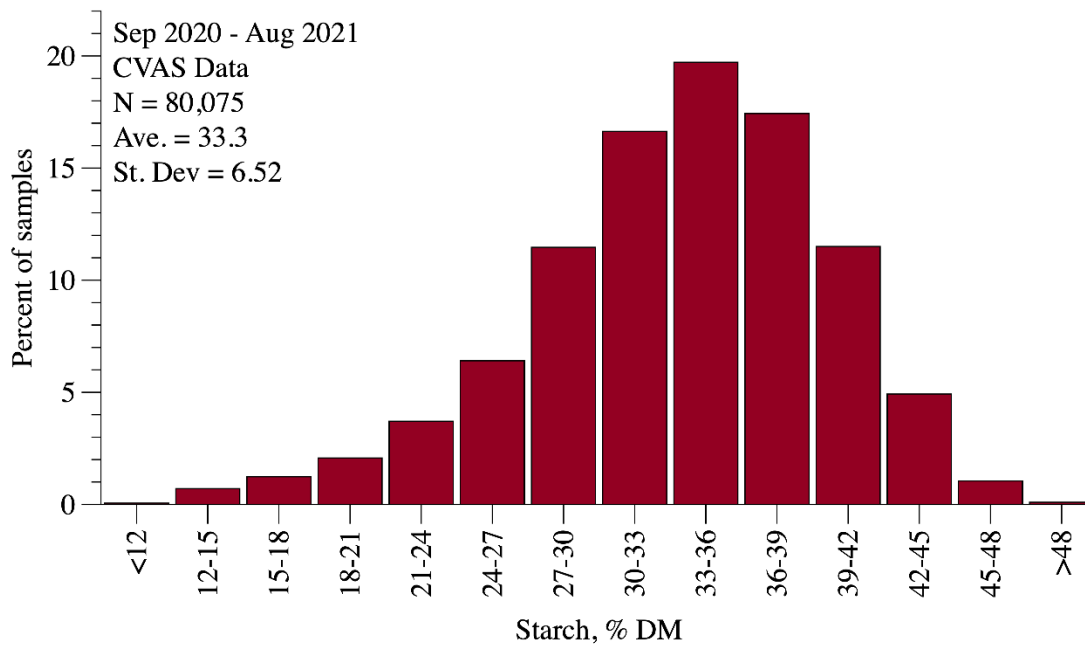


Figure 1. Distribution of starch content (% DM) in corn silage samples analyzed in the Cumberland Valley analytical services. Source: Web page Hoard's Dairyman Sept. 13, 2021.

The proportion of starch is an important consideration for choosing a hybrid that allows higher milk production per ton of DM. Furthermore, the starch digestibility will determine how much of that starch will be available for cow utilization. Thus, improvements in corn silage nutritional quality and components digestibility are reached by changes in kernel and stalk characteristics. For instance, hybrids with a greater proportion of floury endosperm are preferred over those with a greater proportion of vitreous endosperm. This is because floury endosperm has a greater starch digestibility than vitreous (Giuberti et al., 2014). The starch molecule in the vitreous endosperm is most involved with prolamins, which are hydrophobic proteins that confer resistance to digestion (Figure 2).

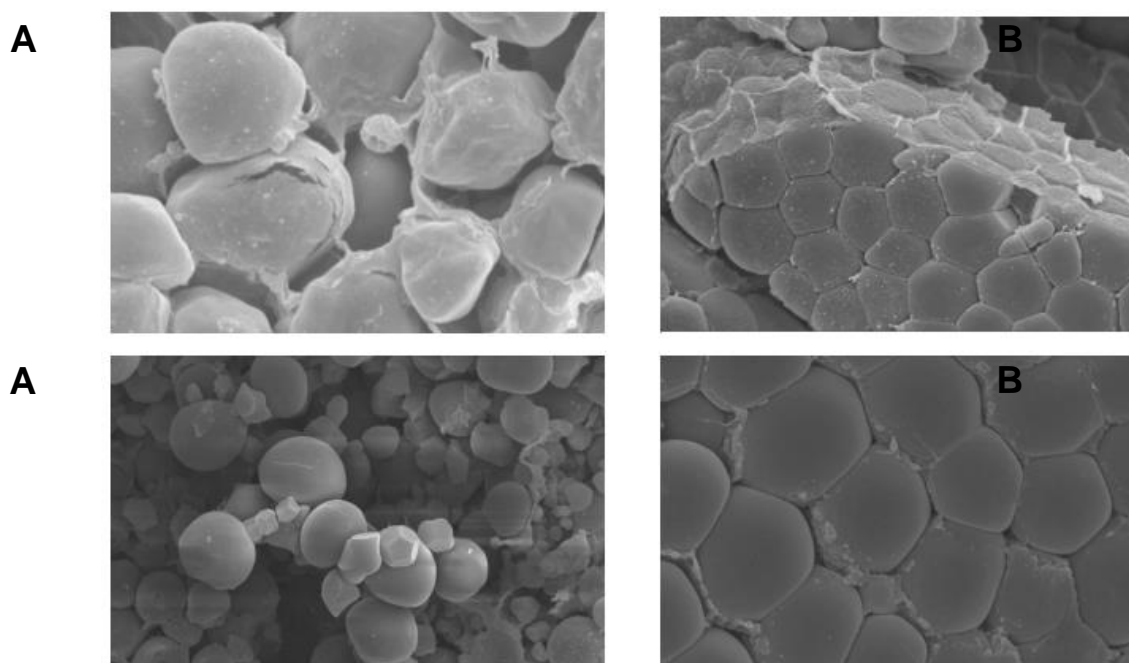


Figure 2. Representation of floury (A) and vitreous (B) endosperm. Adapted from Davide et al., (2009). Thesis, UFLA repository. Federal University of Lavras, Lavras, MG, Brazil.

The vitreousness is a laboratory parameter used to evaluate the percentage of vitreous to floury endosperm; hence, starch digestibility decreases as hybrid vitreousness increases. This relationship can be observed in the previous publication by Correa et al. (2002) (Figure 3).

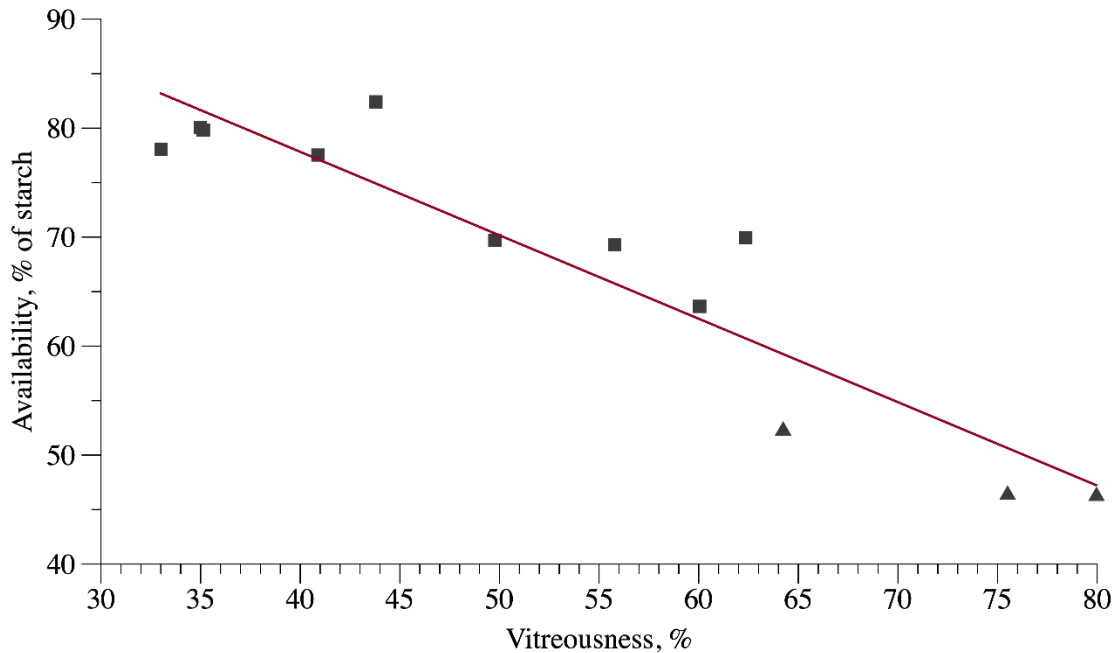


Figure 3. Relationship between corn kernel vitreousness and ruminal in situ starch availability measured in three U.S. dent (■) and Brazilian flint (▲) hybrids harvested at the matured stage of maturity and two U.S. dent (■) hybrids harvested at half milk line, black layer, and maturity stages of maturity. Adapted from Correa et al. (2002)

The combination of starch content and digestibility would affect the diet's energy density, affecting milk yield and/or feed efficiency. The starch digestibility of corn silage is influenced not only by the hybrid starch content and endosperm type but also by parameters defined at harvest. For instance, stage of maturity, particle size, and silage stocking time are recognized parameters used to manipulate starch digestibility (Ferraretto et al., 2013).

Maturity

The literature extensively documented that starch content increases with advances in the maturity stage (Ferraretto et al., 2014). Furthermore, a meta-analysis by Ferraretto et al. (2018) has demonstrated that not only starch increased with maturity but also kernel vitreousness. In that

study, vitreousness increases as the kernel DM concentration increases (Figure 4), suggesting that high maturity at harvest would negatively impact starch digestibility.

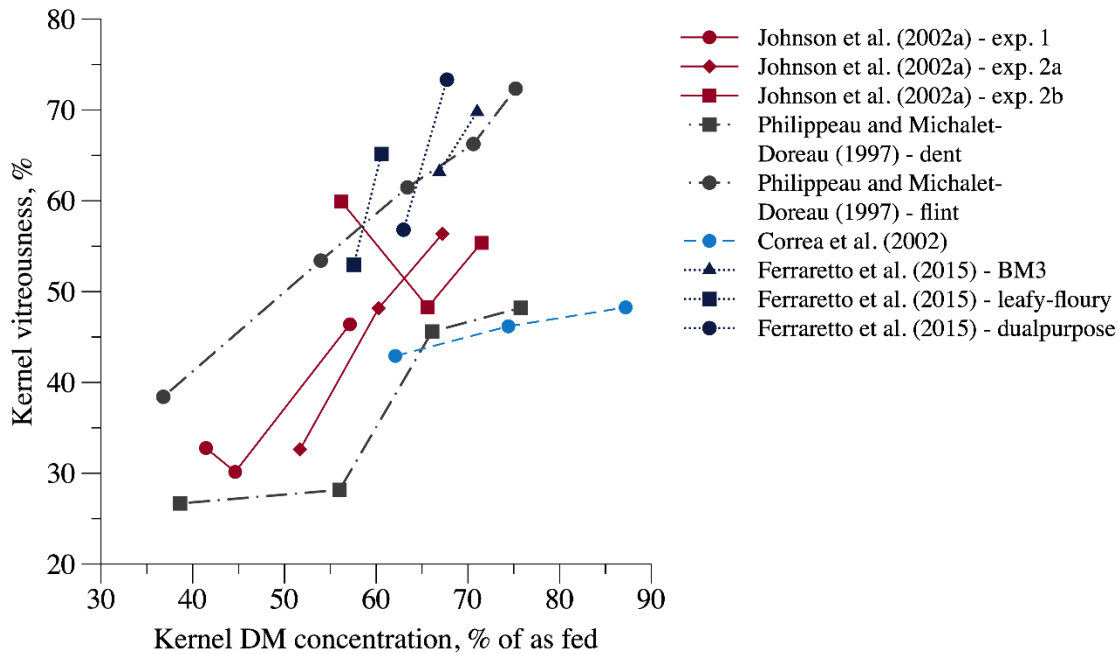


Figure 4. Relationship between DM concentration and vitreousness in corn kernels. (adapted from Ferraretto et al., 2018)

In addition, Ferraretto and Shaver (2012) observed an interaction between maturity and particle size on total-tract starch digestibility (TTSD). In this meta-analysis, the TTSD was increased by the mechanical process of corn silage diets containing 32 to 40% of DM. In the same study, TTSD was 5.9 and 2.8% units greater for silage processed using 1 to 3 mm roll gap settings than processed or unprocessed corn silage with 4 to 8 mm (Figure 5). Additionally, cows fed with processed corn silage produced 1.8 kg more than cows fed with unprocessed corn silage.

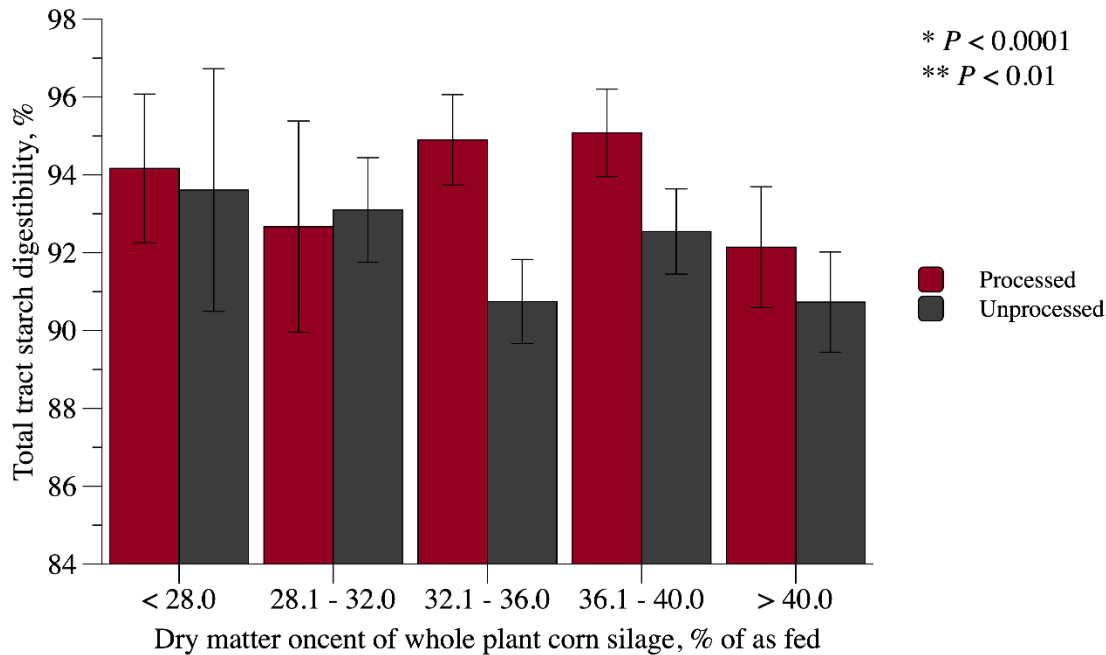


Figure 5. Effect of kernel processing and DM content of whole-plant corn silage on total-tract digestibility of dietary starch. Adapted from Ferraretto and Shaver, (2012)

Particle size

The Kernel processing score (KPS) is used to assess the level of kernel damage after harvest (Ferreira and Mertens, 2005). In this essay, the amount of starch passing through a 4.75 mm screen indicates a score for the kernel corn processing. Samples with more than 70% pass-through 4.75 mm indicate a good kernel process, which is associated with high starch digestibility. Meanwhile, samples retained above the 4.75 mm sieve suggest poor processing, linked to low starch digestibility. Dias Junior et al. (2016) observed an increase in *in situ* starch digestibility when unfermented kernels were split from two to third-six pieces. In that study, 60% of the kernels broken in one-fourth were retained at 4.75 mm sieve, suggesting that broken kernels in four parts were not enough to reach a good KPS score.

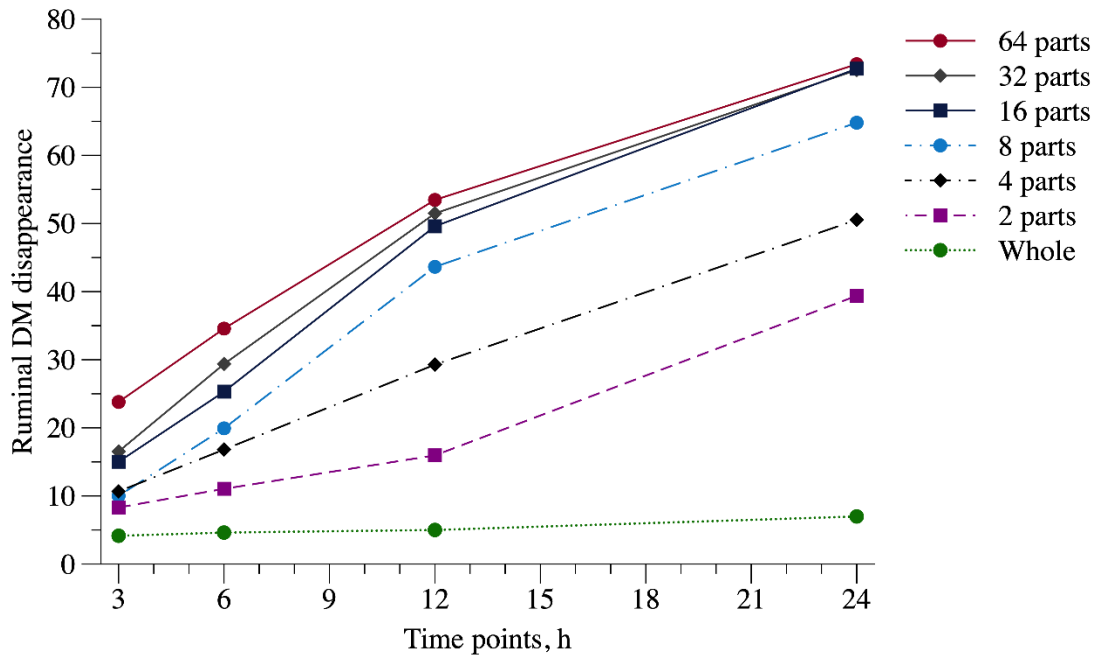


Figure 6. Ruminal in situ DM disappearance (% of DM) of unfermented kernels. Adapted from Dias Junior et al., (2016)

For practical formulation use, the NASEM (2021) has adopted TTSD digestible coefficients based on corn silage DM (Table 2). However, it's important to highlight that starch digestibility can be changed by mean particle size and length of fermentation. The effect of particle size on TTSD can be illustrated for dry ground corn, whereas TTSD increase as the particle size decrease (Table 2). Thus, nutritionists must consider adjustments in the digestibility coefficient to account for these missing values for corn silage. Rémond et al. (2004) and Weiss (2021) demonstrated how to modify starch digestibility for particle size in semi-flint or dent corn. In summary, TTSD in dent and semi-flint corn would decrease by 2.6% and 7.5% units per 1 mm increase in mean particle size, respectively.

Table 2. Total tract starch digestibility of dairy diets containing selected corn grain sources.

Feeds	Total-tract starch digestibility (% starch)
Corn silage, less than 30% DM	91
Corn silage, 32 o 37% DM	89
Corn silage, more than 40% DM	85
Dry ground corn, fine grind (< 1,250 um)	91
Dry ground corn, medium grind (1,500 to 3,250)	89
Dry ground corn, coarse grind (> 3,500 um)	77
High-moisture corn, fine grind (< 2,000 um)	96
High-moisture corn, coarse grind (> 2,000 um)	94
Steam flaked corn	94

Adapted from (Ferraretto, 2021a)

As mentioned, the storage length of corn silage is also associated with a change in starch digestibility. For instance, Kung et al. (2018) summarized the effects of prolonged silage storage on the in vitro TTSD. In that review, starch digestibility has widely increased from 0 to 90 days of fermentation, whereas starch digestibility slightly increases after 120 days of storage (Figure 7). Thus, research supports that new silage would be fed only between 90 to 120 days after ensilage to maximize starch digestibility.

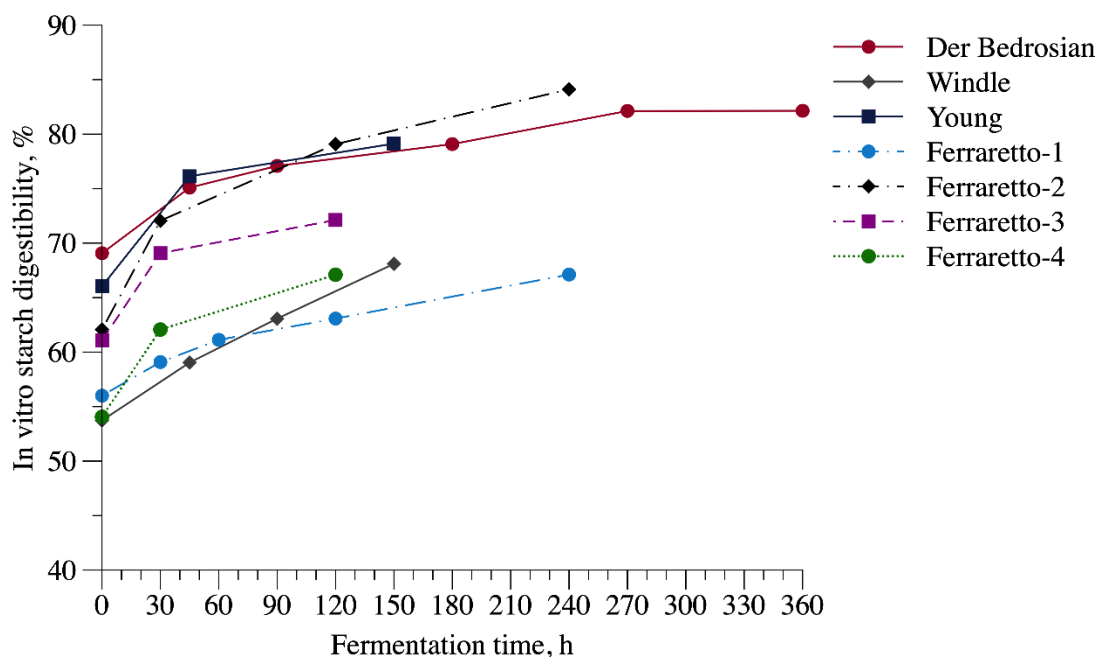


Figure 7. Effect of days of ensiling on ruminal *in vitro* starch digestibility. Adapted from Kung et al. (2018)

Thus, dairy producers have frequently increased the corn silage storage time aimed to increase starch digestibility. Therefore, commercial feed analysis laboratories have adopted assays to report the rate of disappearance data (%/h) calculated using *in vitro* or *in situ* starch digestibility. Predicted values of ruminal starch and whole tract starch digestibility can be calculated using these disappearance rates. Ferraretto (2021) has exemplified the effect of corn silage storage time on the TTSD, TDN, NEL, and milk per ton (Table 3).

Table 3. Effect storage length on the nutritional parameters of corn silage.

Storage length, days	0	30	120	240
ivSD, % of starch	58.9	65.2	71.2	75.6
Starch kd, %/h	14.5	17.5	21.8	23.4
Predicted total tract starch digestibility, % of starch	93.6	94.5	95.3	95.6
TDN, % of DM	72.4	72.7	73.0	73.1
NEL, Mcal/kg	1.62	1.63	1.64	1.64
Milk per ton, kg	1765	1776	1789	1793

In this simulation, the predictions of TTSD have increased as the corn silage storage time increased. In addition to the greater TTSD, the prediction of energy supply (NEL) and milk per ton have also increased with the advanced storage time. Therefore, inventory planning would be set up cautiously to guarantee corn silage availability. One important aspect is that ensiling time does not attenuate differences in starch digestibility caused by hybrids or maturity. Moreover, hybrid choices and harvest time decisions are also important for the total energy available from corn silage.

Stover fraction

Milk production is primarily limited by energy intake on high-production dairy cows. Especially cows in early lactation are normally consuming less than their demand. Limitations in intake are frequently caused by low forages NDF digestibility, which is associated with greater rumen fill and hence, reduced milk production. According to Oba and Allen (1999), each 1% improvement in NDF digestibility corresponds to increases in DMI and 4% fat-correct milk of 0.40 and 0.55 lb/d, respectively. The reduced digestibility of NDF is mainly caused by lignin, an indigestible component of the NDF fraction. Thus, an increase in forage digestibility is often accomplished by reducing lignin NDF concentration (Grant and Ferraretto, 2018).

Most corn stover fraction improvements are relative to fiber digestibility (Sattler et al., 2010). For instance, the brown midrib (BMR) mutant hybrid has a reduced proportion of lignin compared to conventional hybrids (Sattler et al., 2010). Therefore, it is frequently associated with a greater NDF digestibility compared to conventional hybrids. In a meta-analysis, Ferraretto and Shaver (2015) have demonstrated greater ruminal and total-tract NDF digestibility for BMR hybrids (Table 4). Cows fed BMR hybrids produced, on average, 1.5 and 1.0 kg/d more milk and

3.5% fat-corrected milk, respectively, compared to cows fed conventional hybrids. However, it is important to point out that not all BMR hybrids have greater yield than conventional hybrids (Adesogan et al., 2019). Thus, we should use caution when choosing a BMR hybrid and correctly manage the forage inventory.

Table 4. Effect of corn silage hybrids with different stalk characteristics on adjusted least square means for ruminal and total NDF digestibility as well as for lactating performance by lactating cows

Item	CONS ¹	BMR ²	P-value
NDF ruminal digestibility, % of intake	37.0	40.8	0.16
NDF total-tract digestibility, % of intake	42.3	44.8	0.001
Milk yield, kg/d	37.2	38.7	0.001
3.5 FCM	37.6	38.6	0.01

¹CONS = conventional, dual-purpose, isogenic, or low to normal fiber digestibility hybrids;

²BMR = brown midrib hybrid; Adapted from (Ferraretto and Shaver, 2015).

Another factor that is related to improving corn silage fiber digestibility is increasing the harvesting height. This practice is not only associated with increasing the silage energy content but also decreasing the NDF and lignin content on the ensiled material. It is typically adopted by producers that meet or exceed their forage plan. Consequently, producers that harvest corn at a higher height would need less concentrate per unit of milk. Ferraretto (2021b) demonstrated that silage NDF decreased 2.5 and 5.0 % units when harvest height was increased to 10 and 20 inches, respectively, compared to standard harvest height at 6 in (Table 5). In that simulation, the silage starch increased 2.2 and 4.1% units for harvesting whole-plant corn silage at 16 and 26 inches, respectively, compared to harvest at 6 inches. Increased in vitro NDF digestibility was also predicted as harvest height was increased. In contrast, the yield has decreased by 0.4 and 1.0 ton/acre as the harvesting height was increased.

Table 5. Predicted effects of chop height on whole-plant corn silage nutrient composition, digestibility, and yield.

Item	Normal chop height ¹	Simulation ²	Simulation ²
Cutting height, inches	6	16	26
NDF, % of DM	37.7	35.2	32.7
Starch, % of DM	37.5	39.6	41.6
ivNDFD ³ , % of NDF	49.6	52.6	53.6
Yield, ton/acre	8.9	8.4	7.9

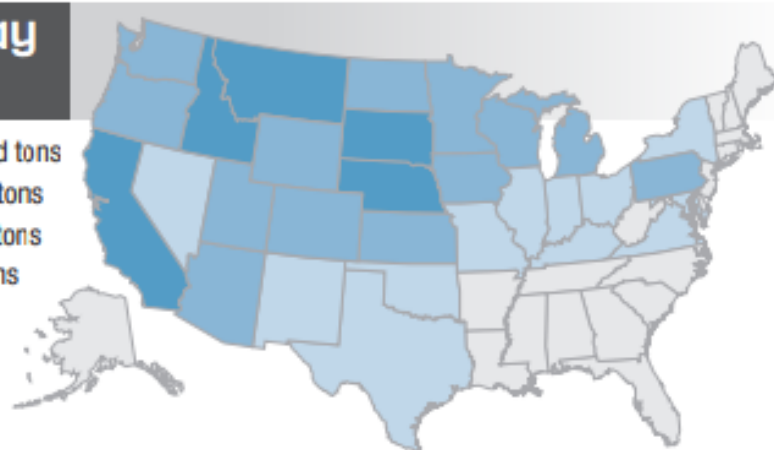
¹Data from Ferraretto et al. (2017); ²Predicted using equations from Paula et al. (2009); ³Ruminal in vitro NDF digestibility.

Alfalfa

Alfalfa is one of the most common forages used to feed cows among US dairies (Ghelich Khan et al., 2016), and most of it is fed as alfalfa hay. In 2020, the harvested area and production of alfalfa hay were estimated at 16 million acres and 53.1 million tons, respectively (USDA, 2020). This production represents about 18% of the total forage harvested in that same year. According to nutritional attributes, alfalfa hay is quality-classified as supreme, premium, good, fair, and low (Table 4; USDA, 2021). The attributes are related to energy availability and crude protein content (Table 3). In November 2021, alfalfa hay prices in the Pacific Northwest averaged \$260, \$238, \$221 to premium, good, and fair alfalfa hay, respectively (USDSA Hay Markets). As a standard forage, alfalfa is considered a compliment forage to corn silage due to its nutritional attributes. As a high-protein forage, it helps to support the requirements of protein of high-production cows. Despite its high soluble protein, alfalfa also has a high amount of protein escaping ruminal degradation, which decreases the necessity of supplementing undegradable nitrogen.

Total alfalfa hay production

- more than 5,000 thousand tons
- 3,000 to 5,000 thousand tons
- 1,000 to 3,000 thousand tons
- 100 to 1,000 thousand tons
- 0 to 100 thousand tons



Total U.S. tons harvested in thousands of tons

● Silage	20.2 tons/acre	137,729
● Other hay	2.1 tons/acre	73,745
● Alfalfa	3.3 tons/acre	53,067
● Greenchop	6.4 tons/acre	29,342
Combined total		293,883

Figure 8. Total alfalfa hay production in 2020. Source: National forage review (2020 U.S. forage statistics)

Table 6. Alfalfa hay quality designation guidelines.

Quality	ADF ¹	NDF ²	RFV ³	TND-100% ⁴	TDN-90% ⁴	CP ⁵
Supreme	< 27	< 34	> 185	> 62	> 55.9	> 22
Premium	27 - 29	34 - 36	170 - 185	60.5 - 62	54.5 - 55.9	20 - 22
Good	29 - 32	36 - 40	150 - 170	58 - 60	52.5 - 54.5	18 - 20
Fair	32 - 35	40 - 44	130 - 150	56 - 58	50.5 - 52.5	16 - 18
Utility	> 35	> 44	< 130	< 56	< 50.5	< 16

¹Acid detergent fiber; ²Neutral detergent fiber; ³Relative feed value (An index for ranking cool-season grass and legume forages based on combining digestibility and intake potential. Calculated from ADF and NDF); ⁴Total digestible nutrients.

The provision of physically effective fiber is another beneficial factor of feeding alfalfa. Fiber stimulates cows' rumination and salivation, which results in rumen buffering. Besides lower NDF content, alfalfa has a higher content of lignin than traditional forages, an indigestible

fiber component. The lignin content increases according to the different alfalfa growth stages, reducing the fiber digestibility and affecting energy supply (Figure 9).

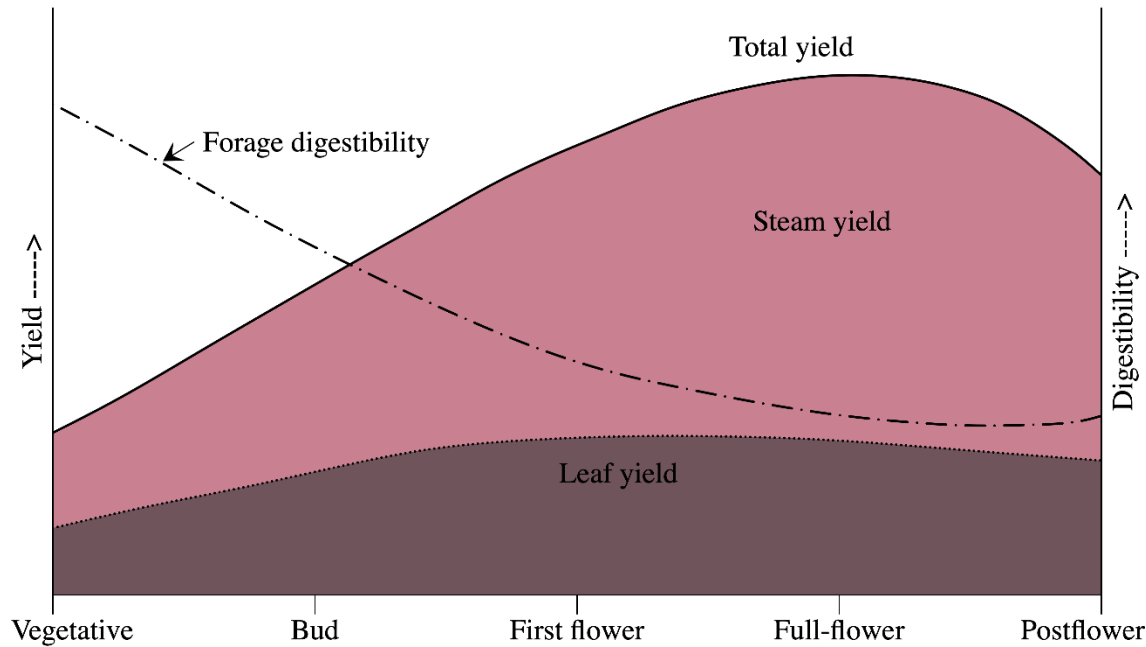


Figure 9. Table 5. Relative forage yield and quality at different alfalfa growth stages. Adapted from Orloff and Putnam (2004).

Thus, new selections technologies such as reduced lignin and condensed tannins are used to improve the nutritional attributes. Reduced alfalfa lignin directly increases fiber digestibility and consequently the energy supply. Reduced lignin had a 10 to 15% decrease in lignin content, which increased 10 to 15% in the relative forage quality (RFV) (Adesogan et al., 2019). As lignin in the plant increase as the advance of maturity, the reduced lignin allows a greater harvest window compared to conventional alfalfa. Weakley et al. (2008) has evaluated the effects of feeding two transgenic alfalfa down reduced lignin (COMT and CCOMT) to dairy cows. In that study, fiber digestibility was greater in the two reduced lignin alfalfa hay than conventional

alfalfa (Table 7). In addition, cows fed with the COMT gene down-regulated produced 2.6 lb/d compared to cows fed conventional alfalfa.

Table 7. Effect of feed alfalfa reduced lignin on the fiber digestibility and milk yield.

Alfalfa hay type ¹	CP (% DM)	NDF (% DM)	NDFD (% NDF)	Milk, lb/d
COMT Inactive	18.1	31.1	53.5**	84.7*
COMT Active (control)	18.4	29.3	42.5	82.1
CCOMT Inactive	18.1	42.5	48.6**	84.5
CCOMT Active (control)	18.3	31.1	44.5	86.7

¹TMR diets - 50% alfalfa hay, 10% corn silage, 40% concentrate; *Significant, P<0.10;

**Significant P< 0.01; Source: Weakley et al. 2008 J. Dairy Sci. Supple. 1

Alfalfa is also commonly fed as silage. Hoffman et al. (1998) reported greater milk production (+1.6 kg/d) for cows fed with alfalfa silage than cows fed perennial ryegrass silage. Broderick (1985) has evaluated the effects of feeding alfalfa silage to corn silage as sole forage in the diet of lactating cows. In the two trials, cows fed about 60% of alfalfa silage as forage had similar milk production and 4% fat corrected milk that cows fed primarily corn silage (Table 8). Furthermore, cows had the same milk performance when fed alfalfa silage or alfalfa hay. Broderick (1985) concluded that high-quality alfalfa silage is essentially equal to corn silage for milk production, reducing the problem from milk fat depression (trial 1).

Table 8. Production and milk components of cows fed with alfalfa silage, corn silage, and alfalfa hay.

Item	Dietary forage (Trial 1) ¹			Dietary forage (Trial 2) ¹		
	60% AS	60% CS	79% CS	63% AS	60% AH	60% CS
Milk, kg/d	26.4 ^a	26.1 ^a	23.9	29.8 ^a	29.4 ^{ab}	30.3 ^a
4% FCM ³	25.1 ^a	24.1 ^a	22.9 ^b	28.3 ^a	28.0 ^{ab}	29.2 ^a
Fat, %	3.72 ^a	3.50 ^b	3.74 ^a	3.68	3.70	3.86
Protein, %	3.16	3.18	3.21	3.11 ^b	3.11 ^b	3.32 ^a

^{ab}Means in row within each trial different superscript differ (P<0.05); ¹Proportion of dietary dry matter from alfalfa silage (AS), alfalfa hay (AH), or corn silage (CS); ²Far corrected milk.

In summary, alfalfa has been the most common forage used to feed dairy cows. The combination of the high energy and protein content accounts for more of this choice. In addition, improvements in fiber digestibility through technologies such as reduced lignin increase the potential to use alfalfa in dairy diets, which might be associated with lower feed costs or greater animal performance.

Alternative forages for the Pacific Northwest

Due to climate changes and predictions for drier conditions (lower rainfall and less water for irrigation), there is a search for alternative feeds in dairy operations that require less water usage, fit the production system that integrates forage production, promoting sustainability and regenerative agriculture (Rockström et al., 2017).

Considering the water availability situation, the current scenario is that 40% of the U.S is in a drought; much of the Western half of the United States is in the grip of a severe drought of historic proportions (Table 10). When it comes to the Pacific Northwest, 100% of this region is experiencing abnormally dry conditions, with more than a fifth of the region enveloped in exceptional drought — the most severe category outlined by the U.S. Drought Monitor (NIDIS, 2021).

U.S. Drought Monitor

November 23, 2021
 (Released Wednesday, Nov. 24, 2021)
 Valid 7 a.m. EST

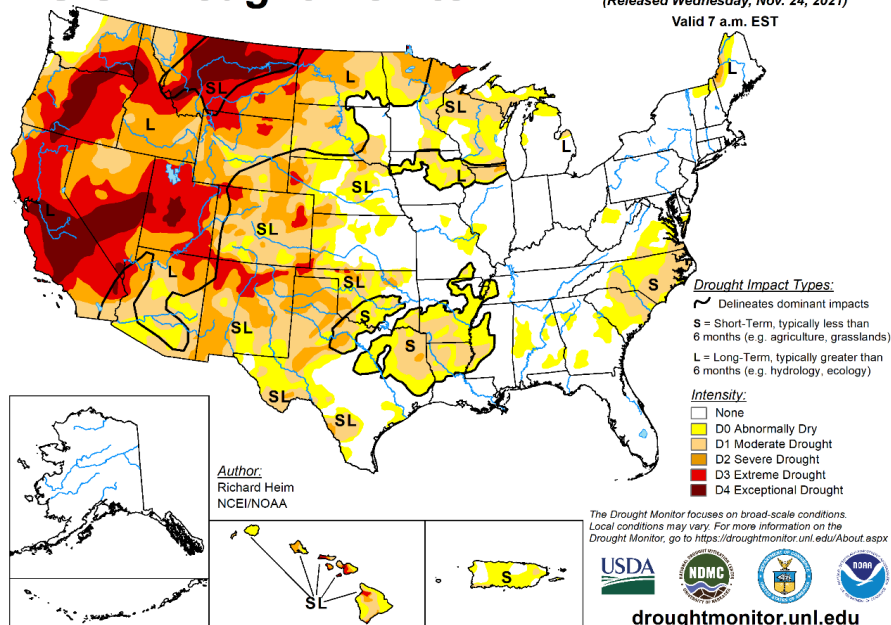


Figure 10 – Current drought situation in the U.S. according to categories. Source: Heim (2021).

The primary direct economic impact of drought in the agricultural sector is crop failure and pasture losses. These costs are often passed on to consumers through increased prices and/or they may be offset through government disaster assistance programs.

The instability of livestock feed prices has forced farmers from integrated agriculture systems to search for alternative feed resources to replace traditional grains without compromising the feed quality or animal performance. This would improve the relationship between crop and livestock production; thus, increasing the stability of feed prices and the ability of producers to cope with climate changes, water shortage, and soil depletion (Condon et al., 2015).

Wheat (*Triticum aestivum*)

Wheat (*Triticum aestivum*) is gaining acceptance as an alternative for more sustainable silage production due to its productivity, nutritional quality (Meinerz et al., 2011), and smaller water requirement (McKenzie and Woods, 2011). This crop has a world annual production of over 735 million tons, being among the largest crop cultivated globally and an essential source of carbohydrates for millions of people (FAO, 2015). Wheat ranks third among U.S. field crops in planted acreage, production, and gross farm receipts, behind corn and soybeans. In 2020/21, U.S.

farmers produced a total of 1.8 billion bushels of winter, durum, and other spring wheat from a harvested area of 36.7 million acres (USDA, 2021b). Washington’s Whitman County produces more wheat than any other county in the United States (WGA, 2021)

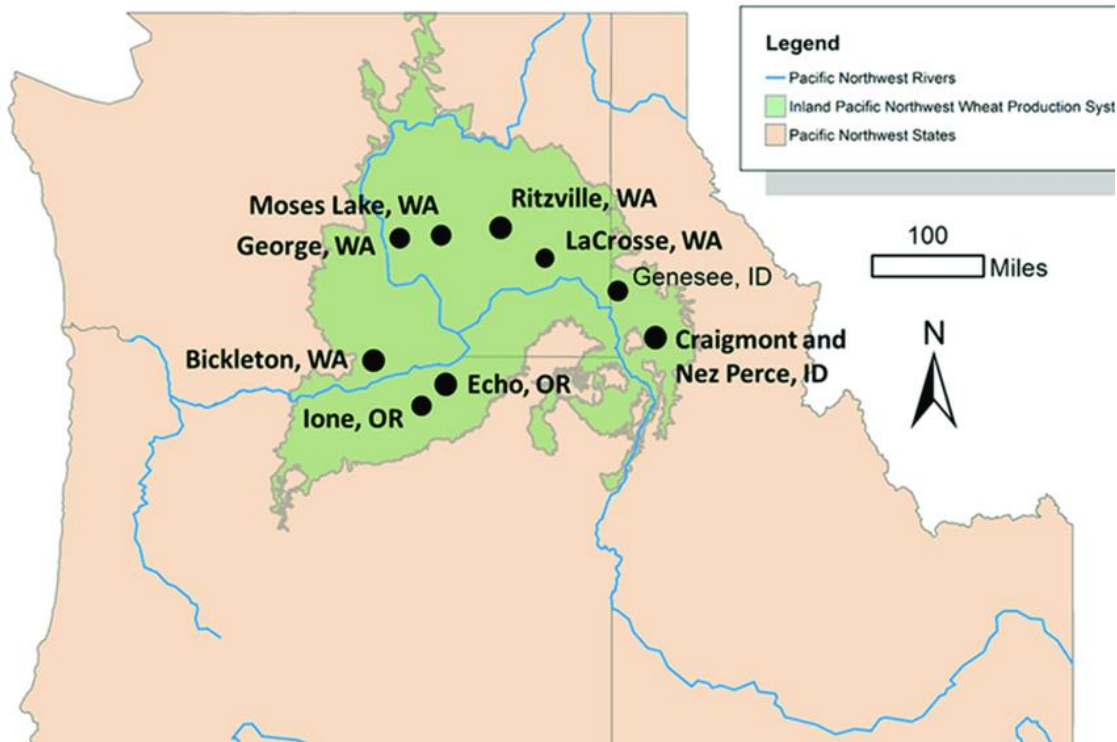


Figure 11. The major wheat-producing area of the inland Pacific Northwest. Source: REACCH (2020).

Despite its potential, wheat has predominantly been a minor forage for livestock in the United States, even with available data supporting its use. When alternative forages, such as wheat silage, fully replace corn silage in a ration, starch content decreases and fiber increases. Thus, normally, energy density of the diet is reduced, potentially impacting MY (Sutton et al., 1998). Therefore, a full substitution was not recommended in the past. However, no study has been done evaluating the replacement of corn silage with WS while maintaining standardized dietary starch levels; thus, standardizing energy levels (Table 9).

Table 9. Current experimental levels of replacement testing wheat as a feed alternative.

Product	Treatment	Main results	Reference
Long wheat hay (HL), short wheat hay (HS) or wheat silage (SI)	30% of TMR DM	Concentrated TMR containing only 30% to 32% wheat forages, HS is better than HL or SI at preventing feed sorting and increasing intake. Replacing HL with SI (containing 20% spikes mass) increased DM digestibility and intake of digestible DM, and resulted in higher yields of milk, 4% FCM and ECM by lactating cows.	(Shaani et al., 2017)
Wheat (<i>Triticum aestivum</i>) silage (WS)	10% of the diet DM	Apparent total-tract digestibility of DM and OM was decreased. The diet resulted in higher urinary urea excretion, higher milk urea N, and lower milk N efficiency than the CS diet. WS decreased CO ₂ emission, but MY may decrease slightly (3%). At MY of around 42 kg/d, WS can partially replace CS DM and not affect DM intake.	(Harper et al., 2017)
Untreated wheat straw (UWS) or WS silage (treated with sodium hydroxide, molasses and wheat grain; TWSS)	1) control (20% alfalfa hay (AH) and 20% corn silage (CS); 2) UWS (13% AH, 13% CS, and 13% UWS) and 3) TWSS (13% AH, 13% CS, and 14.3% TWSS)	The yield of 4 % FCM did not differ between the cows offered the control or TWSS diets (P>0.05). Milk fat contents by cows fed TWSS diet were higher than those fed control diet (P<0.05). Overall, partly substitution of the diet forage by the TWSS (13% of diet DM) had no effects on the digestibility and FCM yield compared with the cows offered control diet, but led to improvement of these traits than the cows offered UWS diet.	(Ghasemi et al., 2016)

Canola (*Brassica spp.*)

Canola is grown in 29 states in the U.S., ranging from just a few hundred acres in some states to 1.7 million acres in North Dakota. According to the latest report from the U.S. Department of Agriculture's Farm Services Agency, there were 2.2 million acres of canola planted in the United States in 2021 (Figure 12). Major production regions in the U.S. include the Northern Plains, Pacific Northwest (PNW), and Southern Great Plains. Montana, Washington, and Idaho are the top producing states after North Dakota (U.S. Canola Association, 2021).

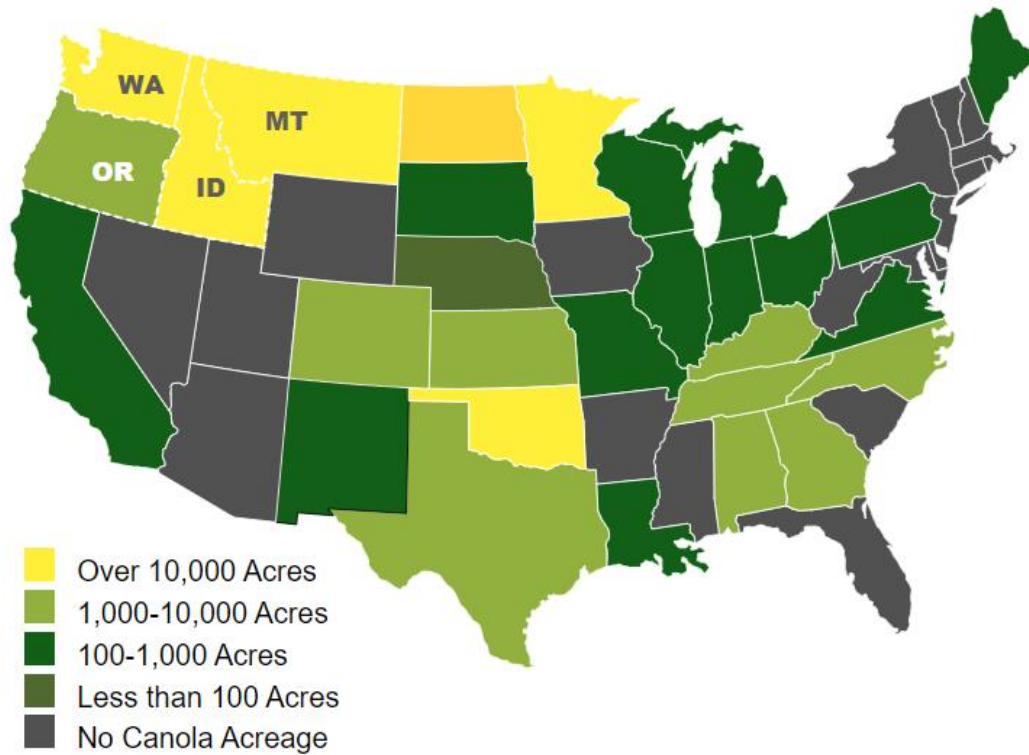


Figure 12. Main Canola cultivation places in Pacific Northwest in 2021. Source: PNW Canola Association (2021).

Brassica plants are frost, heat, and drought-resistant, making them an excellent choice as an alternative crop. Forage rape produces high yields of DM (8–15 t of DM/ha) in a relatively short period (60–120 d), has low establishment costs, uses water and nitrogen efficiently, and has high concentrations of digestible DM and CP. Further, is highly digestible (75–85% DOM), with 10–20% of CP, 18–20% of NDF and high concentrations of readily fermentable carbohydrates such as starch (6–11%), sugars (10–15%), and pectin (9%).(Keim et al., 2020).

Brassicac have long been used as forage for livestock, mainly in temperate grazing systems (Chakwizira et al., 2014). However, significant variations in animal response occurred among experiments testing Brassica forages (Barry, 2013; Table 10).

Table 10. Response data observed in experiments testing Brassica forages in dairy animals.

Product	Treatment	Main results	Reference
Forage rape silage	(1) control, (2) 30% FRS, and (3) 45% FRS	Including FRS to dairy cow diets, up to 45% of diet DM, improved MY due to changes in VFA and predicted microbial N flow, and had no negative impact on dairy cow health or sensory characteristics of milk.	(Keim et al., 2020)
Brassica forages	Control, turnip, or rape silage	Supplementation with turnip or rape modified the profile of FA in blood plasma and milk, increasing the saturated fraction, mainly short- and medium-chain FA, and decreasing the mono- and polyunsaturated FA. Cheeses made with milk from animals fed turnip and rape were differentiated by increased odor, flavor, spiciness, bitterness, and acidity.	(Seguel et al., 2020)

Triticale (*X Triticosecale*)

Triticale is an intergeneric hybrid of wheat (*Triticum* sp.) × rye (*Secale cereale* L.) (Kavanagh et al., 2010). The latest U.S. agricultural census conducted in 2017 reported that triticale grain was harvested from 33,000 ha with 3,700 ha of the total from the state of Washington (USDA-NASS, 2017)

The original goal for producing triticale was to produce a new cereal crop that combined the superior agronomic performance and the end-use qualities of wheat with the stress tolerance (both biotic and abiotic) and adaptability of rye, making it more suitable for the production in marginal areas (acidic, saline, or soils with heavy metal toxicity). Further, possible effects of climate change in terms of reduced rainfall or a change in the pattern of rains (IPCC 2014) call for the need to research alternative forage sources better adapted to those scenarios (Thornton et al., 2009). However, despite having many advantages over wheat, global triticale production is still very low (Colín-Navarro et al., 2021).

The low adoption of triticale is due to factors including production concerns, availability of end-use markets, production economics, policy, and competition from wheat. However, new triticale cultivars often have a significantly higher grain yield than wheat cultivars, with plumper more uniform kernels (Meale and McAllister, 2015) that possess desirable nutritional characteristics for inclusion in lactating cows (Mikulła et al., 2011). Besides, triticale has been used to prevent soil erosion during bare soil periods. Preserving the soil is critically important for continued crop productivity, and therefore has long-term benefits.

Triticale has been evaluated as forage for dairy cattle since the 1970s with good results in terms of yield and nutritive value (Fisher, 1972). Thus, it represents a viable alternative for feeding

livestock, given its high DM production and multi-purpose utilization (Table 11). It can be grazed, made into hay, or ensiled, and has the additional advantage of a slow decrease in nutritive value as the plants progress through their growth stages (Mendoza-Elos et al., 2011; Salcedo et al., 2014).

Table 11. Experimental results found testing triticale in different levels as an alternative for feeding dairy animals.

Product	Treatment	Main results	Reference
Triticale silage (TS)	10% of the diet DM	Digestibilities of NDF and ADF were increased in the TS diet compared to the control diet (CS). The diet resulted in higher urinary urea excretion, higher milk urea N, and lower milk N efficiency than the CS diet. Enteric CH ₄ emission/kg of ECM was highest in the TS diet, but MY may decrease slightly (3.51%). At milk production of around 42 kg/d, TS can partially replace CS DM and not affect DM intake.	(Harper et al., 2017)
Triticale hay (TH)	0% TH, 9.0% AH and 7.4% TH	No effect was observed on ECM production because of a compensatory linear effect of increasing milk fat concentration with the incorporation of TH in the diet. Total-tract NDF digestibility tended to increase linearly by 18.5%, but no differences were detected for urinary urea-N excretion and N utilization estimated as milk N	(Santana et al., 2019)
Triticale silage (TS)	5.0 and 7.5 kg DM/d	Providing TS to grazing dairy cows in small-scale dairy farms during the dry season, when herbage growth is limited, was a viable option to sustain moderate MY of 12 kg/cow/d. There was no benefit in providing 7.5 kg DM/cow/day of TS over 5.0 kg DM/cow/day as there were no differences in MY, milk composition, body condition score or live weight.	(González-Alcántara et al., 2020)

Barley (*Hordeum vulgare*)

Barley is one of the first crops domesticated by humans and remains a popular food source. It is a short-season, early maturing crop and is likely the world's oldest cultivated grain. It is produced in a variety of climates in both irrigated and dry-land production areas. In terms of harvested area, barley is second only to corn, at 47 million hectares worldwide in 2017. Barley competes with corn and sorghum as a feed grain. It has higher protein contents than corn, which reduces the need for protein supplements in feed rations. However, it lacks some of the other nutritional elements present in corn. In general, feed barley prices are approximately 85% of corn prices on a per bushel basis (AgMRC, 2021).

Barley grows well in cool and dry conditions. As a result, U.S. barley production is concentrated in the Northern Plain states and the Pacific Northwest (WGC, 2021; Figure 13). U.S. producers harvested 2.2 million acres of barley in 2020 with an average yield of 77.2 bushels/acre.

Total production in 2020 was 170.8 million bushels. From that, in 2020, Idaho was the leading U.S. state in terms of barley production. That year, some 55 million bushels of barley were produced in Idaho. Montana was another major producer of barley in the United States, at 45.67 million bushels (Shahbandeh, 2021).

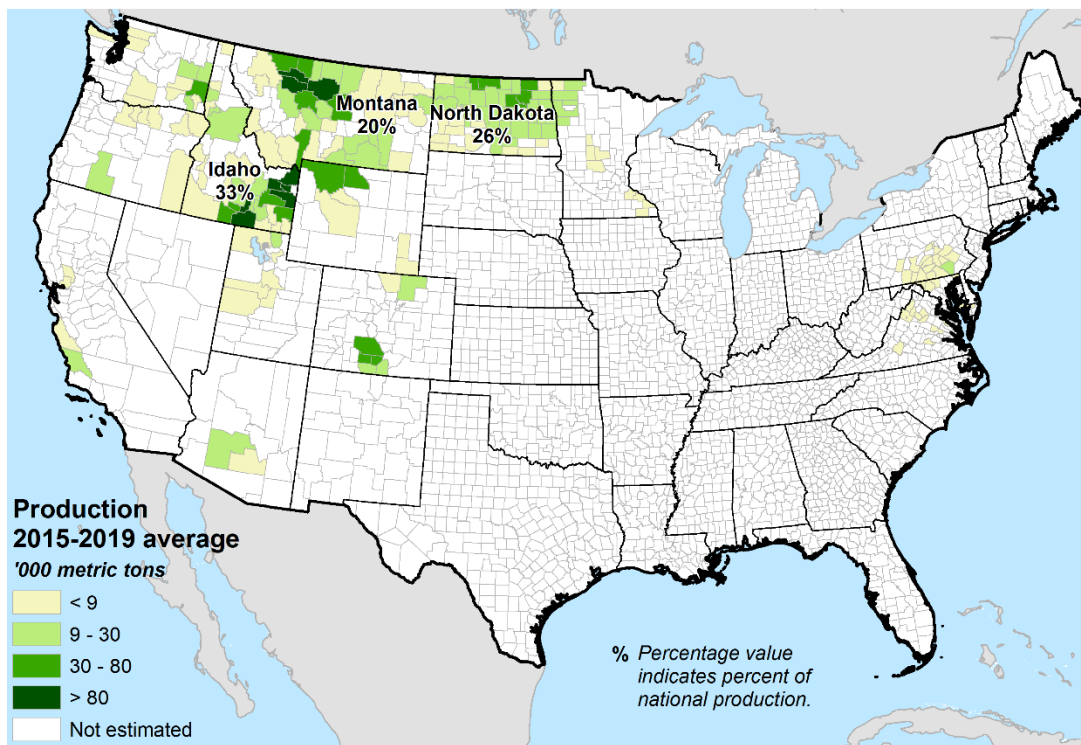


Figure 13. United States: Barley Production Map. Source: USDA (2021c).

However, because of ongoing drought conditions and an unusually long heatwave that has gripped much of the state, Idaho barley yields and total production are expected to decrease considerably this year compared with 2020 (Sean Ellis, 2021). A similar drop in production is expected for other states, such as Washington. According to the USDA, while Washington farmers planted around 70,000 acres in 2021 — down 90,000 in 2020 — to 2021 harvest and yield plummeted to 2.6 million bushels at 38 bushels-per-acre from 6.4 million bushels at 90 bushels-per-acre in 2020 (Featherstone, 2021).

Barley grain is a valuable feedstuff for several different classes of ruminants. When properly processed, mixed, and fed, barley is an excellent feed grain. It can be used as a supplement in forage rations for replacement heifers and as an energy and protein source (Table 12).

Table 12. Experimental data of barley in different levels of substitution in dairy animal diets.

Product	Treatment	Main results	Reference
Hulled or hull-less barley	(1) 45% forage and hulled barley as the sole grain source, (2) 65% forage and hulled barley as the sole grain source, (3) 45% forage and hull-less barley as the sole grain source, and (4) 65% forage and hull-less barley as the sole grain source.	DMI tended to be lower for the diet with 65% forage and hulled barley than for the rest of the diets (24.4 vs. 26.6 kg/d). Neither the type of barley nor the F:C ratio affected MY (41.7 kg/d). Barley type did not affect milk fat or protein concentrations. Feeding LF diets decreased milk fat concentration from 3.91% to 3.50%.	(Yang et al., 2018)
Barley silage (BS)	Barley varieties with different digestible fiber concentrations	Cows fed BS with relatively higher ruminal ivNDFD did not show significant difference from the cows fed other BS varieties with lower ruminal ivNDFD in MY and total chewing activity.	(Refat et al., 2017)
Barley silage (BS)	(1) 0% CS and 54.4% BS in the TMR (0% CS), (2) 27.2% CS and 27.2% BS in the TMR (27% CS), and (3) 54.4% CS and 0% BS in the TMR (54% CS)	CH ₄ production adjusted for DM or gross energy intake increased as the amount of CS decreased in the diet. Decreasing the CS proportion in the diet reduced N utilization.	(Benchaar et al., 2014)

Conclusion

Globally, atmospheric greenhouse gases continue to rise, and it is becoming increasingly apparent that adaptation may be the only viable option to ensure the future food needs of humanity. Furthermore, due to climate changes, the search and adoption of alternative crops that are capable of producing high grain yields on marginal lands under arid conditions with minimal inputs (i.e., fertilizer, pesticide, water) as compared to other cereal grains are becoming more and more necessary as demands for sustainable livestock continue to rise.

Based on the information, these alternatives can be successfully included in the diets of dairy cows, taking into account some specific limitations of each crop (Table 13). Therefore, they represent a way to reduce costs, without considerable changes in milk production and composition, especially in places where the use of corn silage is limited by economic, environmental, or logistical factors. Thus, the main recommendations that can be used are present in Table 13.

Table 13. Recommendation for alternative crops in the Pacific Northwest

Crop	Amount	Effect	Reference
Wheat silage	10 to 13 % of the diet DM	No effect on DMI and digestibility; higher milk fat and no effects on the FCM and ECM yields.	(Ghasemi et al., 2016; Harper et al., 2017)
Canola silage	Up to 45% of diet DM	Improved MY due to changes in VFA and predicted microbial N flow, modification in the profile of FA in blood plasma and milk and had no negative impact on dairy cow health	(Keim et al., 2020; Seguel et al., 2020)
Triticale silage	10% of the diet DM or up to 5.0 kg DM/day	Digestibilities of NDF, ADF, urinary urea excretion and milk urea N were increased, while milk N efficiency was reduced. Enteric CH ₄ emission/kg of ECM was highest in the TS diet, but MY may decrease slightly (3.51%), depending on the animal milk yield and composition, body condition score, and weight.	(Harper et al., 2017; González-Alcántara et al., 2020)e
Barley silage	Up to 65% in diet DM	No effect in MY, milk fat or protein concentrations. Diets with low forage (45%) had milk fat decreased from 3.91% to 3.50%. Diets with high barley showed CH ₄ production adjusted for DM or gross energy intake increased and reduced N utilization	(Benchaar et al., 2014; Yang et al., 2018)

References

- Adesogan, A.T., K.G. Arriola, Y. Jiang, A. Oyebade, E.M. Paula, A.A. Pech-Cervantes, J.J. Romero, L.F. Ferraretto, and D. Vyas. 2019. Symposium review: Technologies for improving fiber utilization. *J. Dairy Sci.* 102:5726–5755. doi:10.3168/jds.2018-15334.
- AgMRC. 2021. Barley Profile. Accessed.
- Barry, T.N. 2013. The feeding value of forage brassica plants for grazing ruminant livestock. *Anim. Feed Sci. Technol.* 181:15–25. doi:10.1016/j.anifeedsci.2013.01.012.
- Benchaar, C., F. Hassanat, R. Gervais, P.Y. Chouinard, H. V. Petit, and D.I. Massé. 2014. Methane production, digestion, ruminal fermentation, nitrogen balance, and milk production of cows fed corn silage- or barley silage-based diets. *J. Dairy Sci.* 97:961–974. doi:10.3168/jds.2013-7122.
- Broderick, G.A. 1985. Alfalfa Silage or Hay Versus Corn Silage as the Sole Forage for Lactating Dairy Cows. *J. Dairy Sci.* 68:3262–3271. doi:10.3168/jds.S0022-0302(85)81235-2.
- Chakwizira, E., P. Johnstone, A.L. Fletcher, E.D. Meenken, J.M. de Ruiters, and H.E. Brown. 2014. Effects of nitrogen rate on nitrate-nitrogen accumulation in forage kale and rape crops. *Grass Forage Sci.* 70:268–282. doi:10.1111/gfs.12109.
- Colín-Navarro, V., F. López-González, E. Morales-Almaráz, F. de J. González-Alcántara, J.G. Estrada-Flores, and C.M. Arriaga-Jordán. 2021. Fatty acid profile in milk of cows fed

triticale silage in small-scale dairy systems in the highlands of central Mexico. *J. Appl. Anim. Res.* 49:75–82. doi:10.1080/09712119.2021.1884082.

Condon, N., H. Klemick, and A. Wolverson. 2015. Impacts of ethanol policy on corn prices: A review and meta-analysis of recent evidence. *Food Policy* 51:63–73.

doi:10.1016/j.foodpol.2014.12.007.

Correa, C.E.S., R.D. Shaver, M.N. Pereira, J.G. Lauer, and K. Kohn. 2002. Relationship between corn vitreousness and ruminal in situ starch degradability. *J. Dairy Sci.* 85:3008–3012.

doi:10.3168/jds.S0022-0302(02)74386-5.

Dias Junior, G.S., L.F. Ferraretto, G.G.S. Salvati, L.C. de Resende, P.C. Hoffman, M.N. Pereira, and R.D. Shaver. 2016. Relationship between processing score and kernel-fraction particle size in whole-plant corn silage. *J. Dairy Sci.* 99:2719–2729. doi:10.3168/jds.2015-10411.

FAO. 2015. Food and Agriculture Organization of the United Nations, Food Outlook Biannual Report on Global Food Markets. Accessed.

Featherstone, C.H. 2021. Wheat, Barley Harvests Hit Hard by Drought. Accessed.

Ferraretto, L.F. 2021a. Dietary fiber and starch for dairy cows: new perspectives from the Nutrient Requirements of Dairy Cattle study report. Pages 49–60 in VIII International simposium of dairy cattle. Simleite.

Ferraretto, L.F. 2021b. Fiber and starch digestibility in corn silage. Pages 88–94 in California Animal Nutrition Conference, Sacramento, CA.

Ferraretto, L.F., P.M. Crump, and R.D. Shaver. 2013. Effect of cereal grain type and corn grain harvesting and processing methods on intake, digestion, and milk production by dairy cows through a meta-analysis. *J. Dairy Sci.* 96:533–550. doi:10.3168/jds.2012-5932.

Ferraretto, L.F., and R.D. Shaver. 2012. Effect of corn silage harvest practices on intake, digestion, and milk production by dairy cows. *Prof. Anim. Sci.* 28:141–149.

doi:10.15232/S1080-7446(15)30334-X.

Ferraretto, L.F., and R.D. Shaver. 2015. Effects of whole-plant corn silage hybrid type on intake, digestion, ruminal fermentation, and lactation performance by dairy cows through a meta-analysis. *J. Dairy Sci.* 98:2662–2675. doi:10.3168/jds.2014-9045.

Ferraretto, L.F., R.D. Shaver, and B.D. Luck. 2018. Silage review: Recent advances and future technologies for whole-plant and fractionated corn silage harvesting. *J. Dairy Sci.*

101:3937–3951. doi:10.3168/jds.2017-13728.

- Ferraretto, L.F., K. Taysom, D.M. Taysom, R.D. Shaver, and P.C. Hoffman. 2014. Relationships between dry matter content, ensiling, ammonia-nitrogen, and ruminal in vitro starch digestibility in high-moisture corn samples. *J. Dairy Sci.* 97:3221–3227. doi:10.3168/jds.2013-7680.
- Ferreira, G., and D.R. Mertens. 2005. Chemical and physical characteristics of corn silages and their effects on in vitro disappearance. *J. Dairy Sci.* 88:4414–4425. doi:10.3168/jds.S0022-0302(05)73128-3.
- Fisher, L.J. 1972. Evaluation of Triticale Silage for Lactating Cows. *Can. J. Anim. Sci.* 52:373–376. doi:10.4141/cjas72-042.
- Ghasemi, E., G.R. Ghorbani, and M. Khorvash. 2016. Effect of feeding untreated wheat straw or ensiled wheat straw treated with NaOH, molasses and wheat grain on performance of lactating dairy cows. *Anim. Sci. J.* 05:33–46.
- Ghelich Khan, M., S.Y. Yang, J.S. Eun, and J.W. MacAdam. 2016. 1596 Nitrogen excretion of lactating dairy cows fed an alfalfa hay– or birdsfoot trefoil hay–based high-forage diet. *J. Anim. Sci.* 94:776–776. doi:10.2527/jam2016-1596.
- Giuberti, G., A. Gallo, F. Masoero, L.F. Ferraretto, P.C. Hoffman, and R.D. Shaver. 2014. Factors affecting starch utilization in large animal food production system: A review. *Starch/Staerke* 66:72–90. doi:10.1002/star.201300177.
- González-Alcántara, F. de J., J.G. Estrada-Flores, E. Morales-Almaraz, F. López-González, A. Gómez-Miranda, J.I. Vega-García, and C.M. Arriaga-Jordán. 2020. Whole-crop triticale silage for dairy cows grazing perennial ryegrass (*Lolium perenne*) or tall fescue (*Lolium arundinaceum*) pastures in small-scale dairy systems during the dry season in the highlands of Mexico. *Trop. Anim. Health Prod.* 52:1903–1910. doi:10.1007/s11250-020-02206-9.
- Grant, R.J., and A.T. Adesogan. 2018. Journal of Dairy Science Silage Special Issue: Introduction. *J. Dairy Sci.* 101:3935–3936. doi:10.3168/jds.2018-14630.
- Grant, R.J., and L.F. Ferraretto. 2018. Silage review: Silage feeding management: Silage characteristics and dairy cow feeding behavior. *J. Dairy Sci.* 101:4111–4121. doi:10.3168/jds.2017-13729.
- Harper, M.T., J. Oh, F. Giallongo, G.W. Roth, and A.N. Hristov. 2017. Inclusion of wheat and triticale silage in the diet of lactating dairy cows. *J. Dairy Sci.* 100:6151–6163. doi:10.3168/jds.2017-12553.

- Heim, R. 2021. U.S. Drought Monitor. Accessed.
- Hoffman, P.C., D.K. Combs, and M.D. Casler. 1998. Performance of Lactating Dairy Cows Fed Alfalfa Silage or Perennial Ryegrass Silage. *J. Dairy Sci.* 81:162–168. doi:10.3168/jds.S0022-0302(98)75563-8.
- Kavanagh, V.B., L.M. Hall, and J.C. Hall. 2010. Potential hybridization of genetically engineered triticale with wild and weedy relatives in Canada. *Crop Sci.* 50:1128–1140. doi:10.2135/cropsci2009.11.0644.
- Keim, J.P., J. Daza, I. Beltrán, O.A. Balocchi, R.G. Pulido, P. Sepúlveda-Varas, D. Pacheco, and R. Berthiaume. 2020. Milk production responses, rumen fermentation, and blood metabolites of dairy cows fed increasing concentrations of forage rape (*Brassica napus* ssp. *Biennis*). *J. Dairy Sci.* 103:9054–9066. doi:10.3168/JDS.2020-18785.
- Kung, L., R.D. Shaver, R.J. Grant, and R.J. Schmidt. 2018. Silage review: Interpretation of chemical, microbial, and organoleptic components of silages. *J. Dairy Sci.* 101:4020–4033. doi:10.3168/jds.2017-13909.
- McKenzie, R.H., and S.A. Woods. 2011. Crop water use and requirements.
- Meale, S.J., and T.A. McAllister. 2015. Triticale. E. F., ed. Springer, Cham.
- Meinerz, G., C. Olivo, J. Viégas, J. Nornberg, C. Agnolin, R. Scheibler, T. Horst, and R. Fontaneli. 2011. Silage of winter cereals submitted to double purpose management. *Rev. Bras. Zootec.* 40:2097–2104.
- Mendoza-Elos, M., E. Cortez-Bacheza, G. Rivera-Reyes, J.A. Rangel-Lucio, E. Adrio-Enríquez, and F. Cervantes-Ortiz. 2011. Época y densidad de siembra en la producción y calidad de semilla de triticale (*X Triticosecale* Wittmack).. *Agron. Mesoam.* 22:309. doi:10.15517/am.v22i2.11804.
- Mikulła, R., W. Nowak, J.M. Jaśkowski, P. Maćkowiak, and E. Pruszyńska Oszmalek. 2011. Effects of different starch sources on metabolic profile, production and fertility parameters in dairy cows. *Pol. J. Vet. Sci.* 14:55–64. doi:10.2478/v10181-011-0008-9.
- NIDIS. 2021. U.S. Crops and Livestock in Drought. Accessed.
- Oba, M., and M.S. Allen. 1999. Evaluation of the importance of the digestibility of neutral detergent fiber from forage: Effects on dry matter intake and milk yield of dairy cows. *J. Dairy Sci.* 82:589–596. doi:10.3168/jds.S0022-0302(99)75271-9.
- Orloff, S., and D. Putnam. 2004. Balacing yield, quality , and persistence. Pages 197–208 in

2004 34th California Alfalfa Symposium, San Diego, CA.

Paula, E.M., B.A. Saylor, J. Goeser, and L.F. Ferraretto. 2009. Influence of cutting height on nutrient composition and yield of whole-plant corn silage through a meta-analysis.

PNW Canola Association. 2021. The Pacific Northwest Is a Prime Canola Growing Region. Accessed.

REACCH. 2020. Regional Approaches to Climate Change for Pacific Northwest Agriculture - Farmer-to-Farmer Case . Accessed.

Refat, B., D.A. Christensen, J.J. Mckinnon, L.L. Prates, J. Nair, A.D. Beattie, W. Yang, T.A. McAllister, and P. Yu. 2017. Evaluation of barley silage with varying ruminal in vitro fiber digestibility on lactation performance and chewing activity of lactating dairy cows in comparison with corn silage. *Can. J. Anim. Sci.* 98:177–186. doi:10.1139/cjas-2016-0191.

Rémond, D., J.I. Cabrera-Estrada, M. Champion, B. Chauveau, R. Coudure, and C. Poncet. 2004. Effect of corn particle size on site and extent of starch digestion in lactating dairy cows. *J. Dairy Sci.* 87:1389–1399. doi:10.3168/jds.S0022-0302(04)73288-9.

Rockström, J., J. Williams, G. Daily, A. Noble, N. Matthews, L. Gordon, H. Wetterstrand, F. DeClerck, M. Shah, P. Steduto, C. de Fraiture, N. Hatibu, O. Unver, J. Bird, L. Sibanda, and J. Smith. 2017. Sustainable intensification of agriculture for human prosperity and global sustainability. *Ambio* 46:4–17. doi:10.1007/s13280-016-0793-6.

Salcedo, G., P. Barcelo, and P. Lazzeri. 2014. Potencial productivo y nutritivo de los triticales de nueva generacion. Pages 1–8 in *Pastos y PAC 2014–2020, 53° Reunión Científica de la Sociedad Española para el Estudio de los Pastos*. Sociedad Española para el Estudios de los Pastos, Potes (Cantabria, Spain).

Santana, O.I., J.J. Olmos-Colmenero, and M.A. Wattiaux. 2019. Replacing alfalfa hay with triticale hay has minimal effects on lactation performance and nitrogen utilization of dairy cows in a semi-arid region of Mexico. *J. Dairy Sci.* 102:8546–8558. doi:10.3168/jds.2018-16223.

Sattler, S.E., D.L. Funnell-Harris, and J.F. Pedersen. 2010. Brown midrib mutations and their importance to the utilization of maize, sorghum, and pearl millet lignocellulosic tissues. *Plant Sci.* 178:229–238. doi:10.1016/j.plantsci.2010.01.001.

Sean Ellis. 2021. Drought, Heat Will Impact Idaho Barley Production. Accessed.

Seguel, G., J.P. Keim, E. Vargas-Bello-Pérez, C. Geldsetzer-Mendoza, R.A. Ibáñez, and C.

Alvarado-Gilis. 2020. Effect of forage brassicas in dairy cow diets on the fatty acid profile and sensory characteristics of Chanco and Ricotta cheeses. *J. Dairy Sci.* 103:228–241. doi:10.3168/jds.2019-17167.

Shaani, Y., M. Nikbachat, E. Yosef, Y. Ben-Meir, I. Mizrahi, and J. Miron. 2017. Effect of feeding long or short wheat hay v. wheat silage in the ration of lactating cows on intake, milk production and digestibility. *Animal* 11:2203–2210. doi:10.1017/S1751731117001100.

Shahbandeh, M. 2021. Leading Barley Producing U.S. States, 2020. Accessed.

Thornton, P.K., J. van de Steeg, A. Notenbaert, and M. Herrero. 2009. The impacts of climate change on livestock and livestock systems in developing countries: A review of what we know and what we need to know. *Agric. Syst.* 101:113–127. doi:10.1016/j.agsy.2009.05.002.

U.S. Canola Association. 2021. What Is Canola? Accessed.

USDA-NASS. 2017. Census of Agriculture. Washington, DC: U.S.

USDA. 2020. 2020 national forage review.

USDA. 2021a. Hay Quality Designation Guidelines. Accessed November 26, 2021. <https://www.ams.usda.gov/market-news/hay-reports>.

USDA. 2021b. USDA ERS - Wheat. Accessed.

USDA. 2021c. United States: Barley Production. Accessed.

Weiss, W.P. 2021. Update on estimating energy supply and energy requirements for dairy cowstle. Pages 55–61 in Proc. of the California Animal Nutrition Conference, Sacramento, CA.

WGA. 2021. The History of Washington State Wheat. Accessed.

WGC. 2021. Facts About Washington State Barley . Accessed.

Yang, Y., G. Ferreira, C.L. Teets, B.A. Corl, W.E. Thomason, and C.A. Griffey. 2018. Effects of feeding hulled and hull-less barley with low- and high-forage diets on lactation performance, nutrient digestibility, and milk fatty acid composition of lactating dairy cows. *J. Dairy Sci.* 101:3036–3043. doi:10.3168/jds.2017-14082.

Protein Supplementation for Beef Cattle

Evan C. Titgemeyer

Department of Animal Sciences and Industry

Kansas State University

Manhattan, KS 66506-1600

etitgeme@ksu.edu

Valuation of protein sources for beef cattle

For supplementing protein (or nitrogen [N]) to beef cattle, the primary concern is providing enough ruminally available N (**RAN**) to meet the needs of the ruminal microbes to ensure that ruminal fermentation maximizes energy availability. In short, we feed protein to optimize fermentation and maximize the energy availability from the diet. When the N/protein needs of the ruminal microbes are met, the flow of microbial protein to the small intestine along with the amount of ruminally undegraded protein (**RUP**) provided by common dietary ingredients will, in most cases, meet the needs of most beef cattle for absorbable amino acids.

Most models that calculate beef cattle performance predict the amount of microbial protein that flows out of the rumen, and this estimate is important for predicting the amount of RAN is required to meet the microbes needs. Ruminal microbes can obtain RAN either directly from ruminally degraded protein (**RDP**) in the diet or from recycled urea-N.

Recycling of urea-N to the gastrointestinal tract, and presumably that to the rumen, is generally similar when RDP or digestible RUP are included in the diet, at least under conditions where RAN is limiting (Wickersham et al., 2008, 2009). However, RDP will additionally provide its N directly to the microbes and, thus, it is better able to meet the microbes' needs than equivalent amounts of RUP. In other words, RDP provides the microbes with N from the degraded protein as well as N from recycled urea, whereas RUP only provides microbes with N from recycled urea. Therefore, in typical situations for most types of beef cattle production, RDP will be of greater value than RUP because RAN is the most critical nutrient provided by the dietary protein.

For most situations in the beef cattle industry, we can roughly equate the value of a protein source with its ability to provide RAN, either directly as RDP or via recycled urea-N. In this context, ruminal degradability and postruminal digestibility of RUP are the factors that will affect the ability of protein sources to provide RAN.

In the work of Wickersham et al. (2008, 2009), digestible RUP led to slightly more of the supplemental N being recycled to the rumen (98%) than did RDP (66%). Because the protein sources were provided on an equal total N basis, the RDP provided significantly more RAN to the cattle than did the RUP. The efficiency of urea recycling decreases as RAN supply increases, which might suggest, therefore, that recycling might be quite similar between RDP and RUP,

when providing equal amounts of RAN. Thus, for calculations presented herein, I simplified the relationship between protein supply and urea recycling by using an intermediate value of 80% to predict the recycling of urea from either RDP or RUP. As such, the RAN supply from a protein source could be calculated as: $RDP + (0.80 \times \text{digested N})$, regardless of where the N is digested.

Using the 80% estimate for urea recycling from either RDP or digestible RUP, the calculations in Table 1 are designed to consider the effects of ruminal degradability of protein as well as of the indigestible N content on the value of a protein source in providing RAN. Protein degradabilities were set to range from 20 to 80% of the feed's N, and indigestible protein ranged from 0 to 40% of the total N. Few feedstuffs would have values outside of these ranges. From the calculations in Table 1, there are clearly disadvantages to increasing the RUP content of a feedstuff, even if there is no detrimental effect on the amount of indigestible N. At the same time, there are additional detrimental effects on RAN if there are increases in indigestible protein, regardless of the ruminal degradability. Additionally, the effect on RAN of a 30% shift in ruminal N degradability (such as decreasing from 80% to 50%) is greater than the effect of a 30% change in indigestible protein. A 30% change in ruminal degradability is a real-world possibility if a feedstuff with highly degradable protein were treated to reduce ruminal degradation (e.g., heating of soybean meal). In contrast, 30% of total protein being indigestible would represent a rather poor quality feed, likely with extensive heat damage. Feedstuffs with large concentrations of indigestible protein are generally more likely to have large concentrations of RUP, so it would be unlikely to find a feedstuff with high RDP along with a large fraction of indigestible protein. In contrast, it is possible to find feeds, such as quality ring-dried blood meal, that would have a large fraction of RUP along with very small amounts of indigestible protein (i.e., the RUP is well digested in the small intestine).

With the viewpoint that the main goal of protein supplementation to most beef cattle diets should be to provide RAN, it is obvious that we primarily want to select protein sources that are extensively degraded in the rumen (high RDP) and that also have extensive small intestinal digestion of any RUP that is present. The question then becomes: How can we effectively measure these two characteristics in feedstuffs in a manner that is accurate, fast, and inexpensive?

Many protein systems consider the *in situ* Dacron bag method as an acceptable way to assess ruminal degradation of feed proteins. Most routine analyses with this approach use a single time point for the incubation to improve throughput and reduce cost. By increasing the number of time points, it is possible to more thoroughly fractionate a feedstuff's protein and determine the rate of degradation for the potentially degraded fraction; this allows RDP to be calculated across a range of passage rates. If a single time point is used, the time of incubation is critical. For example, companies marketing to the dairy industry, where high RUP is valued, are likely to utilize a shorter incubation time to elevate the estimate of their product's RUP concentration. In the beef industry where greater RDP should be valued, longer incubation times might be preferred for marketing purposes, but few feedstuffs are specifically marketed on the basis of a high ruminal protein degradability. This may be because few feeds have greater RDP than the commonly available solvent soybean meal, alfalfa, and urea.

About 50 years ago, Goering et al. (1972) identified acid detergent insoluble N (**ADIN**) as a useful measure of indigestible protein in heat-damaged forages. Based on the success of ADIN as a measure of indigestible protein in heated forages, a number of researchers have assessed ADIN as a measure of indigestible protein in various feedstuffs, and this concept still remains in some models. For non-forage protein sources, there is not a direct relationship between ADIN content and indigestible protein, suggesting that ADIN cannot be used as an accurate assessment of indigestible protein. For example, Nakamura et al. (1994) measured total tract N digestibilities of various sources of distillers grains in lambs, and they found no relationship between ADIN content of the distillers grains and the N digestibility. This agreed with previous work from Nebraska where the ADIN fraction of the feed was not found to be indigestible (Britton et al., 1987). Visual assessment of the color of SBM or distillers grains can provide some qualitative information about heat damage in feeds. Several studies have verified the expected conclusion that DDGS that have experienced more heating have a greater ADIN concentration and a darker color. Cromwell et al. (1993) showed a general relationship between dark color and ADIN concentration of dried distillers grains, although most of the samples in that study were from beverage plants and not from fuel alcohol manufacturers. Cromwell et al. (1993) demonstrated that darker DDGS had lower lysine contents and led to worse performance of pigs fed protein-limiting diets. Lower lysine concentrations reflect irreversible binding of lysine in Maillard reaction products, which would be expected to increase both RUP and indigestible N. In contrast, Nakamura et al. (1994) observed different colors among their distillers grains as well as large differences in ADIN concentrations, yet total tract digestibility of N did not differ appreciably among sources, suggesting that color and ADIN may not be perfect predictors of the ability of distillers grains to provide RAN to cattle.

In my opinion, the best option for assessing RUP concentration and postruminal digestion remains the three-step procedure described by Calsamiglia and Stern (1995). This procedure estimates ruminal digestion using a 16-hour in situ ruminal fermentation followed by sequential treatment with acid-pepsin and pancreatin to determine small intestinal digestion of the RUP. For reasons noted above, some users of this approach select shorter time points for the ruminal fermentation to better reflect rapid ruminal passage from cattle with high feed intakes. The three-step procedure does not directly estimate indigestible protein, but large intestinal disappearance of N from a supplemental protein source is unlikely to be large, so the estimate of indigestible RUP from the three-step procedure should be a reasonable estimate of unavailable N.

Certainly there are some aspects of the three-step procedure that are not ideal. Most importantly, ruminally cannulated cattle are required, which increases complexity of the assay as well as run-to-run variation. One could argue that the data are directly applicable only to feeding conditions that match the diet fed to the cannulated cattle. Moreover, the cost and length of the assay are concerns. Some commercial labs will provide data from the three-step procedure; most commercial analyses are conducted for feeds destined for use in the dairy industry where high RUP concentrations are valued, but it may be worthwhile for feedstuffs destined for beef cattle as well (although for different reasons). Using data collected from the

three-step procedure, one could compare the value of protein sources for the beef industry as $RDP + 0.8 \times \text{total tract digestible protein}$.

Lysine supplementation for growing cattle limit fed corn-based diets

Although most beef cattle will have their metabolizable protein requirements met by supplies of microbial protein and RUP contained in common dietary ingredients, there may be cases where beef cattle require protein/amino acid supplementation to achieve optimal performance. Limit-fed, rapidly growing cattle might be a situation where responses to protein supplementation might be expected. Growing cattle have protein deposition rates that are greater than finishing cattle. Moreover, when growing cattle are limit fed, the goal is typically to achieve near maximal rates of protein deposition, while limiting the amount of fat deposition. To limit fat deposition, energy intake is restricted, either by feeding a diet with a low energy concentration or by restrictedly feeding a more energy-dense diet. In cases where energy intake is restricted, microbial protein synthesis will be limited by the availability of fermentable energy. This in turn will decrease supplies of microbial protein. In addition, corn protein is known to be particularly deficient in lysine. Thus, if protein supply is limiting in calves fed corn-based diets, then lysine might be the most limiting amino acid.

Recently, we conducted a trial to assess the benefit of supplementing ruminally protected lysine to limit-fed steers (255 kg). The steers were predominantly Angus-cross and were implanted with Revalor G. The control diet contained 10% dry-rolled corn, 29.5% steam-flaked corn, 40% Sweet Bran, and 13% hay. Treatments included: control, 0.129% Smartamine-ML (**Lys-3**, providing roughly 3 g/d metabolizable lysine), 0.259% Smartamine-ML (**Lys-6**, providing roughly 6 g/d metabolizable lysine), and 0.89% blood meal (**BM**, providing roughly 3 g/d metabolizable lysine). Calves were limit-fed once daily at 2.4% of body weight (dry matter basis). Relative to control over the 77-day growing phase, supplementing Lys-3 increased body weight gain 8.7 kg, whereas Lys-6 increased body weight gain by 4.7 kg (Table 2). The BM treatment, which should have provided the same amount of lysine as Lys-3, did not increase body weight gain.

Following the growing phase where the treatments were applied, steers were shipped to a commercial feedyard where they were finished on a common diet for an average of 195 days. At slaughter relative to control, Lys-3 steers had 3.4 kg greater carcass weights, Lys-6 steers had 7.1 kg greater carcass weights, whereas BM steers had carcass weights no greater than control. This data provides for some interesting observations. During the growing phase, 3 g/d lysine was more effective than 6 g/d lysine in improving performance. Yet, when the cattle were finished on a common diet, steers fed Lys-3 maintained their advantage over the controls, but the higher level of lysine (Lys-6) during the growing phase led to better finishing performance and the heaviest carcasses. Also interesting was the inability of BM, which was designed to provide the same amount of lysine as Lys-3, to modify either growing phase or finishing phase performance. These results raise the possibility that Lys-6 somehow programmed the cattle for better performance during the finishing phase when the identical diets were fed. We were unfortunately unable to measure feed intake by treatment during the finishing phase, so it is possible that finishing-phase feed intake was different among treatments. However, ribeye

areas were slightly larger and back fat depth was slightly less for cattle that received Smartamine-ML during the growing period, and the slight decreases in back fat might suggest that feed intake was not greatly increased by lysine supplementation during the finishing phase.

Methionine and choline effects on health of receiving cattle

We have recently been studying supplementation to growing cattle of methionine and other compounds containing methyl groups. Some data would suggest that the amino acid methionine or the methyl-containing compound choline could reduce inflammation and fatty liver in periparturient dairy cows (Grummer, 2008; Zhou et al., 2016a,b). In a growth study with receiving beef heifers, Grant (2020) supplemented ruminally protected methionine as Smartamine-M. Methionine supplementation did not affect performance, which was an expected result because the corn-based diet was predicted to provide adequate amounts of methionine. We were most interested in evaluating effects on health performance, but unfortunately, from a research perspective, morbidity rates were extremely low and therefore could not be assessed. However, over time, plasma haptoglobin, a measure of hepatic inflammation, became lower ($P = 0.05$) for heifers that received supplemental methionine than for control heifers.

We are now in the midst of replicated growth studies with receiving heifers to assess effects of supplementation with ruminally protected methionine or ruminally protected choline. Our hypothesis is that either methionine or choline might improve immune response of heifers, leading to less morbidity and/or better responsiveness of sick heifers to treatment. Although pathogens cause respiratory disease, an animal's overactive immune response can sometimes be more detrimental to health than the pathogen itself. Thus, taming of an overstimulated immune system could be of value.

We conducted an experiment to evaluate the effects of choline supplementation to steers maintained under conditions where methyl group supply was designed to be either increased and decreased relative to control. The methyl group status of the steers did not appear to affect our measures of immune function, but choline supplementation tended to reduce plasma haptoglobin as well as in vitro neutrophil phagocytosis after a lipopolysaccharide challenge. These responses suggest a modification of the immune response that might lead to less self-damage in response to an overly activated immune system.

Supplementation of guanidinoacetic acid to growing cattle

Methionine is often a limiting amino acid for lactating dairy cattle, and it has been shown to be the most limiting amino acid in ruminal microbial protein. Across a number of research projects, we have shown that supplemental methionine is used with a lower efficiency than are various other essential amino acids (Titgemeyer, 2012). Over time, this led us to consider the role that methionine plays as a methyl group donor, with the thought that methionine's use as a methyl group donor might lead to a catabolism rate greater than for other amino acids. There are hundreds of reactions for which methionine serves as a methyl group donor, but the two quantitatively most important reactions are synthesis of creatine and choline.

Creatine is a vitamin-like compound that can be synthesized by the body in a two-step process. In the first step, glycine and arginine (two amino acids) are used to synthesize guanidinoacetic acid (**GAA**). The GAA is then methylated to form creatine. The regulatory step in this process is the synthesis of GAA, whereas all of the available GAA is methylated to creatine, independent of the body's needs. Thus, we started studying GAA supplementation as a potential means of modifying methyl group availability because the supplemental GAA would consume methyl groups from methionine. Our initial goal was to create a methyl group deficiency. Although GAA supplementation to cattle led to some minor increases in plasma homocysteine (Ardalan et al., 2020, 2021), which is a hallmark of methyl group deficiency, we never generated an extreme methyl group deficiency with GAA supplementation.

Recently, some research from China has demonstrated huge improvements in performance of finishing Angus bulls in response to GAA supplementation. Bulls started the trials at 400-450 kg, and were fed diets containing on average 36% corn silage and 29% ground corn (13.5% CP, 40% NDF, 36% NFC). Across three studies, 0.6 g GAA/kg dry matter increased average daily gains during 60- to 90-day feeding periods (Li et al., 2020; Liu et al., 2021a,b). Gains increased by an average of 24%, whereas efficiency was improved by an average of 16% when 0.6 g GAA/kg dry matter was added to the diet. Presumably this response relates to the conversion of GAA to creatine, which was a limiting factor for growth of muscle tissues. If translatable to the U.S. beef finishing industry, this response to GAA supplementation would be a game changer.

Although we have not supplemented GAA to finishing cattle, we have observed some small changes in nitrogen retention (a measure of whole body protein deposition) in growing cattle. In one study, GAA was able to slightly increase N retention when steers were provided adequate amounts of methionine, but not when they were methionine deficient (Ardalan et al., 2021). This makes sense, because methionine is required for the methylation of GAA to creatine. In another study, GAA led to small decreases in N retention, independent of methionine status (Speer, 2019). In a third study (Grant et al., 2021), N retention was slightly increased when GAA was supplemented, independent of methionine status. Taken as whole, we have not observed large growth responses to GAA supplementation, but our models have been designed more to effect a methyl group deficiency than to assess growth responses. Future research in this area will be particularly interesting.

Acknowledgements

The concepts related to assessing the value of proteins for meeting RAN requirements of cattle was presented at the Plains Nutrition Council in 2017, and much of the related text is from that publication. Research related to lysine utilization by growing cattle was conducted by Katie Hazlewood, whereas that related to methionine utilization in receiving heifers and choline supplementation to steers was conducted by Maddie Grant. Work with GAA supplementation was conducted by Mehrnaz Ardalan, Hannah Speer, and Maddie Grant. Financial support for these projects was provided by Adisseo, Evonik Industries AG, Balchem, Kansas State University Global Food Systems Innovation Program, USDA National Institute of Food and Agriculture Hatch project 1001435, and USDA-NIFA-AFRI grant 2020-67015-30826.

References

- Ardalan, M., E. D. Batista, and E. C. Titgemeyer. 2020. Effect of post-ruminal guanidinoacetic acid supplementation on creatine synthesis and plasma homocysteine concentrations in cattle. *J. Anim. Sci.* 98(3):1-9, skaa072, <https://doi.org/10.1093/jas/skaa072>
- Ardalan, M., M. D. Miesner, C. D. Reinhardt, D. U. Thomson, C. K. Armendariz, J. S. Smith, and E. C. Titgemeyer. 2021. Effects of guanidinoacetic acid supplementation on nitrogen retention and methionine flux in cattle. *J. Anim. Sci.* 99(6):1-12, skab172. <https://doi.org/10.1093/jas/skab172>
- Britton, R.A., T.J. Klopfenstein, R. Cleale, F. Goedecken, and V. Wilkerson. 1987. ADIN as a measure of N digestibility in protein supplements. *J. Anim. Sci.* 65(Suppl 1):465(Abstr.).
- Calsamiglia, S., and M.D. Stern. 1995. A Three-step in vitro procedure for estimating intestinal digestion of protein in ruminants. *J. Anim. Sci.* 73:1459-1465.
- Cromwell, G.L., K.L. Herkelman, and T.S. Stahly. 1993. Physical, chemical, and nutritional characteristics of distillers dried grains with solubles for chicks and pigs. *J. Anim. Sci.* 71:679-686.
- Goering, H.K., C.H. Gordon, R.W. Hemken, D.R. Waldo, P.J. Van Soest, and L.W. Smith. 1972. Analytical estimates of nitrogen digestibility in heat damaged forages. *J. Dairy Sci.* 55:1275-1280.
- Grant, M.S. 2020. The effects of methyl donors and modulated methyl group status on health and performance in growing cattle. M.S. thesis. Kansas State University. <https://krex.k-state.edu/dspace/handle/2097/40781>
- Grant, M.S., M.D. Miesner, and E.C. Titgemeyer. 2021. Effects of guanidinoacetic acid, creatine, and choline on protein deposition and creatine status in growing cattle. *Kansas Agricultural Experiment Station Research Reports: Vol. 7: Iss. 1.* <https://doi.org/10.4148/2378-5977.8032>
- Grummer, R. R. 2008. Nutritional and management strategies for the prevention of fatty liver in dairy cattle. *Vet. J.* 176:10-20.
- Li, S.Y., C. Wang, Z.Z. Wu, Q. Liu, G. Guo, W. J. Huo, J. Zhang, L. Chen, Y. L. Zhang, C. X. Pei, and S. L. Zhang. 2020. Effects of guanidinoacetic acid supplementation on growth performance, nutrient digestion, rumen fermentation and blood metabolites in Angus bulls. *Animal* 14(12):2535-2542.
- Liu, C., C. Wang, J. Zhang, Q. Liu, G. Guo, W.J. Huo, C.X. Pei, L. Chen, and Y.L. Zhang. 2021a. Guanidinoacetic acid and betaine supplementation have positive effects on growth performance, nutrient digestion and rumen fermentation in Angus bulls. *Anim. Feed Sci. Technol.* 276:114923. <https://doi.org/10.1016/j.anifeedsci.2021.114923>
- Liu, Y.J., J.Z. Chen, D.H. Wang, M.J. Wu, C. Zheng, Z.Z. Wu, C. Wang, Q. Liu, J. Zhang, G. Guo, and W.J. Huo. 2021b. Effects of guanidinoacetic acid and coated folic acid supplementation on growth performance, nutrient digestion and hepatic gene expression in Angus bulls. *Br. J. Nutr.* 126(4):510-517. doi: 10.1017/S0007114520004341
- Nakamura, T., T.J. Klopfenstein, and R.A. Britton. 1994. Evaluation of acid detergent insoluble nitrogen as an indicator of protein quality in nonforage proteins. *J. Anim. Sci.* 72:1043-1048.
- Speer, H.F. 2019. Efficacy of guanidinoacetic acid supplementation to growing cattle and relative bioavailability of guanidinoacetic acid delivered ruminally or abomasally. M.S.

- thesis. Kansas State University. <https://krex.k-state.edu/dspace/handle/2097/39730>
- Titgemeyer, E.C. 2012. Protein and amino acids for growth. *J. Anim. Sci.* 90(E-Suppl. 3):739.
- Wickersham, T.A., E.C. Titgemeyer, R.C. Cochran, E.E. Wickersham, and D.P. Gnad. 2008. Effect of rumen-degradable intake protein supplementation on urea kinetics and microbial use of recycled urea in steers consuming low-quality forage. *J. Anim. Sci.* 86:3079-3088.
- Wickersham, T.A., E.C. Titgemeyer, R.C. Cochran, and E.E. Wickersham. 2009. Effect of undegradable intake protein supplementation on urea kinetics and microbial use of recycled urea in steers consuming low-quality forage. *Br. J. Nutr.* 101:225-232.
- Zhou, Z., O. Bulgari, M. Vailati-Riboni, E. Trevisi, M. A. Ballou, F. C. Cardoso, D. N. Luchini, and J. J. Loor. 2016a. Rumen-protected methionine compared with rumen-protected choline improves immunometabolic status in dairy cows during the peripartal period. *J. Dairy Sci.* 99:8956-8969.
- Zhou, Z., M. Vailati-Riboni, E. Trevisi, J. K. Drackley, D. N. Luchini, and J. J. Loor. 2016b. Better postpartal performance in dairy cows supplemented with rumen-protected methionine compared with choline during the peripartal period. *J. Dairy Sci.* 99:8716-8732.

Table 1. Amount of ruminally available N (RAN) provided by protein sources with different proportions of ruminally degradable protein (RDP) and indigestible protein

Indigestible protein, % of total N	RDP, % of total N		
	20	50	80
	----- RAN, % of feed N -----		
0	100	130	160
10	92	122	152
20	84	114	144
30	76	106	NA
40	68	98	NA

RAN was estimated as: $RAN = RDP + 0.80 \times (\text{digestible protein})$, where digestible protein equals RDP plus intestinally digested RUP. The 0.80 coefficient is based on the assumption that 80% of RDP as well as 80% of digestible RUP will be recycled to the rumen as urea.

NA: more than 20% indigestible protein is not compatible with 80% of total protein as RDP.

Table 2. Response of growing cattle to lysine supplementation during the growing phase

Item	Treatment ¹				SEM	Lysine (<i>P</i> -value)	
	Control	Lys-3	Lys-6	BM		Linear	Quad
Bodyweight, kg							
Day 0	249.1	247.9	248.6	248.7	1.45	0.83	0.60
Day 77	393.7	401.3	397.9	392.8	3.87	0.45	0.26
DM intake, kg/d	7.66	7.73	7.68	7.63	0.061	0.77	0.41
Daily gain, kg/d	1.88	1.99	1.94	1.87	0.042	0.32	0.12
Gain:feed, kg/kg	0.247	0.259	0.254	0.247	0.0040	0.25	0.08

¹Lys-3 = 0.129% of diet as Smartamine ML. Lys-6 = 0.259% of diet as Smartamine ML. BM = 0.89% of diet as blood meal.

Table 3. Response of finishing cattle to lysine supplementation during the growing phase

Item	Treatment ¹				SEM	Lysine (<i>P</i> -value)	
	Control	Lys-3	Lys-6	BM		Linear	Quad
Daily gain, kg/d	1.37	1.37	1.42	1.38	0.06	0.17	0.39
Slaughter wt ² , kg	672.8	678.1	683.8	672.1	5.9	0.20	0.98
Carcass weight, kg	434.2	437.6	441.3	433.7	3.8	0.20	0.99
Ribeye area, cm ²	94.7	97.7	97.5	96.0	1.6	0.05	0.18
Back fat, cm	1.87	1.68	1.80	1.79	0.060	0.36	0.04
Choice + Prime, %	98.3	97.1	99.2	95.5	2.3	0.75	0.53

¹Cattle received treatments only through the 77-day growing phase. See Table 2.

²Calculated from hot carcass weights and average dressing percentages.

General Management Considerations to Improve Success of Artificial Insemination and Natural Service Conception Rates

R.N. Funston¹, G.A. Perry², and M.F. Smith³

¹University of Nebraska, West Central Research and Extension Center, North Platte;

²Department of Animal and Range Sciences, South Dakota State University, Brookings;

³Division of Animal Sciences, University of Missouri, Columbia

Introduction

Although artificial insemination is the most powerful tool available for genetic improvement, cow calf producers have been slow to adopt this technology due to the time and labor associated with estrous detection and a market structure that until recently has not provided an incentive to cow calf producers for genetic improvement. However, adoption of fixed-time artificial insemination protocols (FTAI) has been increasing due to a changing market structure that recognizes and provides an economic incentive for genetic improvement (e.g. premiums) combined with the development of FTAI protocols that result in pregnancy rates similar to AI following detection of estrus. FTAI protocols that result in pregnancy rates similar to AI following detection of estrus result in calves being born early in the calving season resulting in more pounds of calf weaned, which is a tremendous economic gain. A successful FTAI program is dependent upon optimization of the number of healthy cycling females at the beginning of the breeding season, careful attention to sire selection, implementation of an appropriate estrus synchronization protocol, low stress cattle handling, purchase of high quality semen, proper semen handling and insemination technique, and good nutritional management before and after FTAI. Most importantly, implementation of a successful FTAI program requires careful thought and attention to detail. The purpose of this paper is to review the major factors affecting the success of a FTAI program. Emphasis will be given to management considerations that should be implemented before, during, and after FTAI.

Importance of Early Conception

Calving date for first calf heifers may impact cow longevity and productivity. Calving late in year one increases the proportion of cows that either calve later next year or do not conceive (Burriss and Priode, 1958). Research has indicated heifers having their first calf earlier in the calving season remained in the herd longer and had greater calf weaning weights compared with heifers that calved later in the calving season (Cushman et al., 2013). Therefore, heifers calving earlier in the calving season have greater potential for longevity and lifetime productivity. Decreasing the calving period has far reaching implications across the cow-calf enterprise and beyond. Calf age is the single most important factor impacting weaning weight in cow-calf operations so herds with more concentrated calving distributions are expected to have heavier weaning weights compared with herds that do not. Effects of calving early in the calving season potentially extend much further into beef systems, including improved pregnancy percentages and subsequent calving distributions the next calving season, increased cow longevity, lower replacement rate, positive influences on carcass quality and value, reduced labor requirements, increased returns on feed inputs and improved overall sustainability.

The importance of maximizing the proportion of cows that conceive early in the breeding season cannot be overemphasized in a beef herd. Data from the University of Nebraska reported that heifers born during the first 20 days compared to the second or third 20 days of the calving season had greater weaning weights, prebreeding weights, and precalfing weights; more heifers cycling by the start of the breeding season; and higher pregnancy rates. Heifers that conceive early in the breeding season have greater longevity in the herd which increases profitability. Furthermore, steer progeny born during the first 20 days compared to the second or third 20 days of the calving season had greater weaning weights, increased hot carcass weights, higher marbling score, and greater carcass value (Funston et al., 2012a). Consequently, the advantages of calves born early include improved end product quality as well as increased reproductive performance of heifers. Management strategies for increasing the proportion of early calving heifers and cows are discussed below.

Factors Affecting Pregnancy Rate

When it comes to reproductive management the things you do well do not compensate for the mistakes you make. Instead, the mistakes you make cancel out all the things you do well. This is best illustrated by examining the primary factors that affect pregnancy rate. In an AI program, pregnancy rate is the product of estrous detection rate and conception rate (Pregnancy rate = estrous detection rate x conception rate; see definitions below). The following definitions can be applied to an entire breeding season or to the synchronized period (period of time during which estrus is expressed after treatment with an estrus synchronization protocol [normally 5 to 7 days]).

Pregnancy rate – total number pregnant during the breeding season/ number of females exposed to breeding (expressed as a percent).

Estrous detection rate – total number of females detected in estrus/number of females exposed to breeding (expressed as a percent).

Conception rate – percentage of females that become pregnant to a designated insemination.

The effect of a decrease in estrous detection rate and/or conception rate on pregnancy rate can be seen in Table 1. Assume that 100% of the heifers have attained puberty and that you are able to detect 95% of the heifers in estrus during the synchronized period. With a conception rate of 70% the pregnancy rate would be: 95% estrous detection rate x 70% conception rate = 67% pregnancy rate! If we hold conception rate at 70% and decrease estrous detection rate to 75%, due to fewer animals cycling or less time spent detecting estrus, the pregnancy will be reduced to 53%. Alternatively, if estrous detection rate remains at 95% but conception rate is decreased to 50% due to compromised semen quality or poor insemination technique, the pregnancy rate would decrease to 48%. Finally, a decrease in both estrous detection rate and conception rate will decrease pregnancy rate from 67% to 38%. Therefore, understanding the effect of estrous detection rate and conception rate on pregnancy rate and the importance of attention to detail in every aspect of an estrus synchronization program is essential!

Table 1: Effect of estrous detection rate and conception rate on pregnancy rate in cattle.

Estrous detection rate	Conception rate	Pregnancy rate
95%	70%	67%
75%	70%	53%
95%	50%	48%
75%	50%	38%

Things to Do before Estrus Synchronization and Fixed-time Artificial Insemination

Where do I start?

When implementing an estrus synchronization and AI program the first decision should be where to start. Estrus synchronization and AI do not have to be used in combination. Estrus synchronization can be used in combination with natural service or AI. There are clear benefits to reproductive management of a herd from using estrus synchronization in combination with natural service (e.g. increase the proportion of females that conceive early). Two estrus synchronization protocols that are relatively low cost and have been effective in combination with natural service include: 1) Feed MGA (0.5 mg/hd/day) for 14 days to heifers and turn bulls in 10 days after MGA withdrawal, and 2) Turn in bulls (day 1 of breeding season) and inject all heifers and cows with prostaglandin F_{2α} (PGF) on day 4. Advantages of the preceding MGA protocol include no trips through the chute and a portion of the prepuberal heifers will be induced to cycle earlier; however, you have to feed MGA daily for 14 days and each heifer needs to receive the correct dose. The advantage of the PGF protocol is that you only have a single trip through the chute (PGF injection); however, all the heifers and cows need to be cycling in order to respond to PGF. Prepuberal heifers or noncycling cows will not respond to PGF since they do not have a corpus luteum. Once you become comfortable with implementing an estrus synchronization protocol in combination with natural service it is not difficult to make the next step to using AI instead of natural service.

What can I expect in terms of pregnancy rate?

When beginning an AI program it is essential to have realistic expectations regarding the pregnancy rate. As previously discussed, pregnancy rate is the product of estrous detection rate and conception rate. It is important to remember that a pregnancy rate of 67% to a single insemination is good whether you are talking AI or natural service. For natural service, expected pregnancy rates are normally 60 to 70% during 21 days of breeding assuming the bulls are fertile and that 100% of the heifers and cows are cycling. However, a pregnancy rate of 60 to 70% over 21 days is unusually high for natural service since rarely are all the heifers and cows cycling at the start of the breeding season. In a FTAI program, all the cows are injected with GnRH (to synchronize ovulation) and inseminated at a predetermined time. Since there is no estrous detection with FTAI, estrous detection rate becomes the proportion of heifers and cows that ovulate in response to GnRH injection at insemination. It is normal for the pregnancy rate to be higher following FTAI compared to protocols that are dependent upon estrous detection since ovulation is induced and semen is deposited in all the cows in a FTAI protocol. In an estrous detection protocol only the females detected in estrus are inseminated and females that are anestrous or not detected in estrus are not inseminated.

Are my heifers and cows good candidates for an estrus synchronization protocol?

The first question to ask is “Over the past few years what has been the pregnancy rate in my heifers or cows after a 60 to 80 day breeding season?” If the pregnancy rate at the end of the breeding season has been less than 85% there may be management issues that should be addressed before initiating a synchronization and AI program. If the pregnancy rate in your herd over the past few years has been $\geq 85\%$ then you need to evaluate whether your heifers and cows are good candidates for an estrus synchronization and AI program.

Criteria for heifers. Studies in numerous species provide evidence that diet during development can mediate physiological changes necessary for puberty. In cattle, several studies have reported inverse correlations between postweaning growth rate and age at puberty and heifer pregnancy rates. Thus, postweaning growth rate was determined to be an important factor affecting age of puberty, which in turn influences pregnancy rates. This and other research conducted during the late 1960s through the early 1980s indicated puberty occurs at a genetically predetermined size, and only when heifers reach their target BW can increased pregnancy rates be obtained.

Guidelines were established indicating replacement heifers should achieve 60 to 65% of their expected mature BW by breeding. Traditional approaches for postweaning development of replacement heifers used during the last several decades have primarily focused on feeding heifers to achieve or exceed an appropriate target BW and thereby maximize heifer pregnancy rates. Intensive heifer development systems may maximize pregnancy rates, but not necessarily optimize profit or sustainability. Since inception of target BW guidelines, subsequent research demonstrated that the growth pattern heifers experience before achieving a critical target BW could be varied. Altering rate and timing of BW gain can result in compensatory growth periods, providing an opportunity to decrease feed costs. Recent research has demonstrated that feeding replacement heifers to traditional target BW increased development costs without improving reproduction or subsequent calf production relative to development systems in which heifers were developed to lighter target BW ranging from 50 to 57% of mature BW (Funston et al., 2012b). A more comprehensive discussion of heifer development will be presented by Dr. John Hall.

Heifers that will be used for breeding purposes should not have received growth promoting implants. Previous studies report that implanting heifers within 30 days of birth impairs uterine function and decreases subsequent pregnancy rates. Implanting heifers as yearlings is also detrimental to reproduction (Tibbitts et al., 2017).

Criteria for postpartum cows. To increase the number of cows cycling at the beginning of the breeding season, they should have calved unassisted, be in good body condition at calving, disease-free, and allowed an adequate period of recovery from calving to the subsequent breeding season. Postpartum cows that are good candidates for an estrus synchronization program normally meet each of the following criteria: 1) body condition score at calving of ≥ 5 (1= emaciated; 9 = obese), 2) mean postpartum interval of the cows to be synchronized should be ≥ 40 days at the beginning of the protocol. This does not mean that each cow should be ≥ 40 days postpartum but that the mean of the entire group to be synchronized should be ≥ 40 days. If the estrus synchronization protocol you plan to use includes CIDR administration, each cow should be a minimum of 21 days postpartum at the time of CIDR insertion, and 3) low incidence of calving difficulty since dystocia will lengthen the postpartum interval.

How do I choose an AI sire and where do I obtain the semen?

Sire selection is of critical importance and can have a long term effect within a herd, particularly when heifers are retained as replacements. When choosing a sire the following questions need to be addressed: 1) Will I raise my own replacement heifers or purchase them? and 2) How will I market my calves? Answers to the preceding questions will determine the traits that need to be emphasized. If a producer raises his or her own replacement heifers then selection pressure should be placed on maternal traits such as milk, maternal calving ease, stayability, etc.

However, if replacement heifers are purchased off the farm then emphasis on maternal traits in your herd would not be important. When selecting a sire, you need to think about how you will be paid (e.g. pounds of weaning weight, carcass weight, carcass quality) and let this affect your sire selection decisions. Producers that sell their calves at weaning need to place selection pressure on preweaning growth; whereas, producers that retain ownership and market their calves on a grid should emphasize carcass weight, marbling, and ribeye area.

Other genetic traits that have been demonstrated to influence the capacity of a cow to sustain reproduction and be retained include traits that contribute to calving difficulty, level of milk production, and mature size. The genetic changes that have occurred in response to selection for growth and milk production over the last several decades (American Angus Association; American Hereford Association; American International Charolais Association) have undoubtedly resulted in greater nutritional demand to sustain these production traits, leading to greater challenges in sustaining reproduction in nutrient sparse environments. The concept of interaction between genetic potential for production and environment is the basis for recommendation that producers consider selection of breed type or genetic potential of their cattle to match production environment. Converse to this strategy of matching genotype to environment is the recommendation that producers feed their heifers and cows to some target weight or BCS, without consideration of the environmental abundance of associated resources, in an attempt to assure relative high rates of reproductive success. An alternative interpretation of this approach may be that modification of the nutritional environment is needed to sustain a high production potential genotype. The long-term sustainability of this approach needs to be given greater consideration (Roberts et al., 2015).

Expected progeny differences (EPDs) are an effective selection tool and are reported to be 7 to 9 times more effective at generating a response to selection than focusing on measurements of individual performance, which is strongly affected by environment. Use AI sires with high accuracy EPDs and where the semen has been collected from a certified semen services (CSS) facility. Avoid using unproven bulls and do not be hesitant to seek advice from individuals in the AI industry to help make this important management decision.

Another consideration when selecting a sire is whether the bull's semen has worked in FTAI programs. Differences among sires in pregnancy rate to FTAI have been noted; however, the same differences in pregnancy rate may not occur when cows are detected in estrus and inseminated according to the AM/PM rule. Therefore, just because an AI sire has a good conception rate following estrous detection does not ensure he will perform equally well when ovulation is induced and insemination occurs at a predetermined time. It is a good idea to ask an AI representative if there is information available regarding how a bull has worked in a FTAI program.

Which estrus synchronization protocol should I choose?

When choosing an estrus synchronization protocol there are a number of issues to consider including whether you want to detect estrus and inseminate according to the AM/PM rule, inseminate at a predetermined time, or detect estrus for 72 to 84 hr (depending upon the protocol) and inseminate any cows not detected in estrus at a fixed-time. There is an estrus synchronization protocol sheet for heifers and cows that appears in the catalogs of the major AI companies and there are protocols that fit each of the preceding synchronization approaches. Other items to consider include the proportion of females that are cycling as well as the time, labor, and cost of the protocol.

If a significant number of animals are not cycling at the time of implementing an estrus synchronization program, it will be necessary to utilize a progestin-based protocol. Two progestin products that are commercially available for estrous synchronization include Melengestrol Acetate (MGA) and the CIDR (Controlled Internal Drug Release). An advantage of progestin treatment is that a proportion of the prepuberal heifers and anestrus postpartum cows will be induced to begin cycling. In cycling heifers, administration of MGA or CIDRs does not affect the time of corpus luteum regression. However, once corpus luteum regression has occurred, progestin administration can prevent a cow or heifer from showing estrus and ovulating. Consequently, progestin administration in cows that have experienced corpus luteum regression will delay the expression of estrus and ovulation until after progestin withdrawal. At the start of a breeding season, most herds consist of a mixture of cycling and anestrus females. An effective estrous synchronization protocol must be able to induce a fertile estrus or ovulation in both anestrus and cycling heifers and cows. A short luteal phase usually occurs in prepuberal heifers and postpartum beef cows following the first ovulation (Perry et al., 1991; Werth et al., 1996). This short exposure to progesterone is believed to be necessary for reprogramming the reproductive axis to resume normal estrous cycling. Therefore, in herds that have a large proportion of prepuberal heifers or anestrus cows, progestin pretreatment before induction of ovulation can initiate estrous cycling status and eliminate or at least reduce the occurrence of short estrous cycles.

When should I administer the prebreeding vaccines?

Reproductive diseases, including bovine viral diarrhea (BVD), vibriosis, leptospirosis, and infectious bovine rhinotracheitis (IBR), can induce abortion in cattle and decrease profitability (Daly 2007ab). Consequently, a prebreeding vaccination program in combination with careful attention to biosecurity practices and reducing stress/disease transmission within a herd should be included in a herd health program. Since time and labor associated with trips through the chute have been a deterrent to implementing an estrus synchronization program, many producers want to combine prebreeding vaccines with administration of estrus synchronization products. A common question is “Can I administer prebreeding vaccines in combination with estrus synchronization products without decreasing the pregnancy rate to AI?” The answer to this questions depends on how quickly immunity will be established following vaccination and whether or not the vaccine itself will adversely affect reproductive performance and(or) the response to an estrus synchronization protocol (Daly, 2007b). In regards to the first issue, there is a lag time between vaccination and the establishment of immunity that will depend upon factors such as: 1) whether or not the animals were previously vaccinated, and 2) the type of vaccine – modified live (MLV) or killed vaccine. In general, animals that were previously vaccinated will

respond more quickly than animals that are naive to the vaccine and the immune response is normally more rapid to MLV compared to killed vaccine.

Injection of heifers with the IBR virus (wild type and modified live) around the time of breeding resulted in ovarian lesions (particularly within the corpus luteum; Van Der Maaten and Miller 1985; Smith et al., 1990) and decreased conception rates (Miller et al., 1989; Chiang et al., 1990; Miller 1991). Several studies report that vaccinating naive heifers with MLV around time of breeding decreased pregnancy success (Miller et al., 1989; Chiang et al., 1990; Miller 1991). Furthermore, when heifers were vaccinated intravenously with MLV the day after breeding, necrotic lesions were found in the CL and ovaries 9 to 14 days after vaccination and heifers with severe luteal damage had decreased concentrations of progesterone (Van Der Maaten et al., 1985). Heifers vaccinated with a MLV vaccine on the day of the second PGF injection had decreased conception rates compared to control heifers not only for the insemination immediately after vaccination but also for the subsequent insemination. Vaccinated heifers had a first service conception rate of 30% and a second service conception rate of 57%; however, control heifers had a first service conception rate of 78% and a second service conception rate of 100% (Chiang et al., 1990). Furthermore, heifers infected with IBR at or near estrus had disrupted luteal function. In most heifers the next estrous cycle was normal, but in some heifers normal estrous cycles were delayed for up to two months (Miller and Van Der Maaten, 1985). However, when heifers that were previously vaccinated against IBR were administered IBR vaccine either at estrus synchronization or 30 days before insemination there was no detrimental effect of vaccination on pregnancy rate to fixed-time AI or overall pregnancy rate. (Stormshak et al., 1997). Although the latter studies report that administering IBR vaccine at initiation of estrus synchronization to heifers previously vaccinated at weaning did not reduce pregnancy rate, an advantage of administering prebreeding vaccines 30 days or more before insemination is that there is adequate time for the build up of immunity before the heifers are inseminated.

General recommendations for prebreeding vaccinations include the following: 1) Replacement heifers should be vaccinated before and at weaning. The immune response of an individual heifer to a single vaccination is not known; therefore, heifers should receive an initial vaccination followed by a booster when dictated by the vaccination protocol, 2) Both heifers and cows should receive a booster vaccination approximately 30 days before breeding. If it is absolutely necessary to give a modified live vaccine less than 30 days prior to breeding, the vaccine should be administered as soon as possible and only to animals that were vaccinated both before and at weaning. Animals that have not previously been vaccinated (naïve animals) should not be vaccinated near the time of breeding. For additional information on reproductive diseases and the timing of prebreeding vaccines the reader is referred to Daly (2007ab).

Things to Do at Estrus Synchronization and Fixed-time Artificial Insemination

Animal identification and facilities

Individual animal identification and accurate records allow producers to manage animals on an individual basis. When handling animals for synchronization, double check their ear tags for legibility and clip hair from the ears to facilitate reading the tags. Records should include detailed calving, breeding, and pregnancy information. At insemination, document the animal ID, date, time, AI technician, and sire. These records will allow producers to track the reproductive efficiency of individual animals, as well as the skill of the technician.

Stress can suppress the expression of estrus and decrease conception rates. Working facilities should be designed to minimize stressing animals during handling. A well-designed facility will include sorting pens, a crowding tub, and an operable head gate or breeding box for animal restraint. The facility requirement will vary depending on the number and type of animals that will be inseminated as well as the estrus synchronization protocol being used. With a fixed-time AI program, facilities should be sufficient to handle the insemination of all animals within 2 to 3 hrs. Many AI companies or county extension offices have portable breeding chutes available to producers if needed.

Cattle temperament and pregnancy rate

Temperament will vary among animals and is both a safety and production (growth, reproduction, carcass quality) issue. In general, an excitable temperament is a fear-based response that is not breed dependent and can adversely affect reproduction (Cooke, 2010). Three common methods of evaluating temperament in cattle include exit velocity, chute score, and pen score. Exit velocity is a measurement of the speed with which an animal covers a specific distance after release from a squeeze chute and can be measured in feet per second or on a 1 to 5 scale (1 = slow; 5 = very fast). Chute score is a measure of an animal's behavior in a squeeze chute (1 = quiet; 5 = excited) and pen score is a measure of an animal's response to a person when it enters a small pen and interacts with a person inside the pen (1 = quiet; 5 = excited). An excitable temperament in beef cattle is reported to decrease feed intake (Brown et al., 2004; Nkrumah et al., 2007), alter metabolism and nutrient partitioning (Cooke et al., 2009a; Cooke et al., 2009b), and decrease the probability of pregnancy during the breeding season compared to calm herd mates (Cooke, 2010). Attempts to adapt beef females to handling had a beneficial effect on pregnancy in replacement heifers but not older cows (Cooke, 2010). When *Bos indicus*-cross heifers were exposed to four weeks of human interaction and handling, temperament was improved and there was an increase in the proportion of heifers that reached puberty by 12 months and an increase in the proportion of heifers that become pregnant early in the breeding season (Cooke, 2010).

Implementation of an estrus synchronization protocol

Estrus synchronization protocols must be followed precisely. Each product must be administered at the correct dose on the correct day (refer to protocol sheet) and in some cases at the right time of day. For example, the interval from PGF to GnRH and insemination must be in accordance with what is recommended in the protocol sheet (e.g. 66 ± 2 hr for the CO-Synch + CIDR protocol). The recommended time of insemination relative to PGF injection is based on research trials and should be strictly adhered to. In addition, estrus synchronization products must be stored, handled, and administered correctly. For specific tips on handling estrus synchronization products see Figures 1 and 2. Should a mistake occur in product administration or the treatment timeline seek advice immediately from a veterinarian, an extension agent specializing in reproduction, or a representative from an AI company. To minimize the probability of making a mistake, a good practice is to write each of the days of treatment, the product name, dose to be administered, and the day of insemination on a calendar and ask a trusted veterinarian, extension specialist, or AI company representative to review it before beginning the protocol. The Synchronization Planner is an excellent tool to aid in the planning of a synchronization program (beefrepro.info under resources).

Understanding the basic principles of the bovine estrous cycle and how the products synchronize estrus and ovulation can be helpful in reducing the probability of administering the wrong product on a certain day. For more information on how estrus synchronization protocols synchronize estrus and ovulation refer to the article in the appendix entitled “Physiological Principles Underlying Synchronization of Estrus” or see the web based course entitled “Fundamentals of Beef Reproduction and Management:Focus on Estrus Synchronization (http://animalsciences.missouri.edu/extension/beef/estrous_synch/).

Figure 1. Proper handling and administration of GnRH and PG products.

- All injections of GnRH and PG products should be given intramuscularly (IM)
- Wear latex gloves when administering GnRH and PG products
- An 18 gauge 1 ½ inch needle is recommended for these injections
- Change needles frequently
 - Make sure that injection sites are free of manure and dirt, which may cause infection
 - Injecting cattle during wet weather increases the potential for infection
- **Always** follow manufacturer’s recommended storage, dosage and administration procedures

What should I do if a storm is going to hit near during the synchronized period?

A storm or major low pressure system may affect the pattern of expression of estrus in cattle during the synchronized period. Depending upon the temperature change or level of stress there may be a decrease in estrus expression during the synchronized period. If utilizing a FTAI protocol you should inseminate at the scheduled time regardless of estrus expression, provided the heifers or cows meet the criteria for being good candidates for an estrus synchronization program (see previous section). Alternatively, if using a protocol that requires estrous detection you should inseminate according to estrus expression (AM/PM rule) and consider using a cleanup AI (inject GnRH) at 72 to 84 hr after PGF injection.

Proper insemination technique

High pregnancy rates to FTAI are dependent upon a series of events including proper storage and thawing of semen as well as depositing semen in the correct location (uterine body). When synchronizing heifers or cows for FTAI an important question to ask is “How many animals can I (we) inseminate properly in a designated period of time?” The answer to the question will determine how many heifers or cows you synchronize and whether you will require assistance with the insemination process. Representatives of AI companies are available to assist with the entire estrus synchronization and AI process. They can assist you with choosing an appropriate FTAI protocol, administration of the estrus synchronization products, sire selection, purchase of semen, and insemination. If you choose to inseminate the heifers or cows yourself remember that the location of semen placement within the reproductive tract will have a significant impact on pregnancy rates. It is important to deposit the semen in the body of the uterus (target area) and not the cervix. Deposition in the cervix will significantly reduce the pregnancy rate to FTAI; whereas, placing the semen beyond the uterine body into one or both of the uterine horns is not beneficial. During the artificial insemination process it is important to know where the tip of the

AI catheter is at all times. Some helpful tips when performing AI include: pay careful attention to the storage of semen, make sure the thaw unit is at the correct temperature (95°F), and follow the AI company's recommendations for thawing semen.

Figure 2. Proper handling and administration of progestins for estrus synchronization.
Controlled Internal Drug Release (CIDR)
1) Be sure to wear protective (e.g. latex) gloves when handling CIDR inserts.
2) The CIDR applicator should be rinsed in a disinfectant solution (Nolvasan or Chlorohexidine). There should be two buckets each containing a disinfectant solution. The applicator should be washed free of debris in the first bucket and then rinsed clean in the second. By keeping the same washing sequence the disinfectant in the second bucket will remain relatively clean for a sustained period of time. This sequence of events will improve sanitation from animal to animal and reduce the likelihood of infection.
3) Fold the wings of the CIDR and insert it into a clean applicator. The CIDR will protrude approximately one inch from the applicator.
4) Apply lube to the end of the applicator.
5) Wipe the vulva clean before inserting the applicator.
6) When inserting the CIDR make sure that the nylon tail is curved downward. If the tail is pointed upward it will be easier for other animals to pull out the CIDR thus reducing retention rate.
7) Gently insert the applicator containing the CIDR in an upward manner similar to the insertion of an AI catheter.
8) Push the applicator as far forward as possible, deposit the CIDR by pressing the plunger, and slowly remove the applicator.
9) To prevent other animals from removing the CIDR, the nylon tail can be clipped such that only 2 ½ inches protrude from the vulva.
10) At CIDR removal, gently but firmly pull on the nylon tail until it is removed. Be sure to dispose of the CIDR properly.
Melengestrol Acetate (MGA)
1) MGA is an orally active feed additive that should be fed once a day at the recommended dose - 0.5 mg in a 3 to 5 lb carrier. Do not top dress MGA on other feeds. Provide adequate bunk space - 18-24 inches per animal.
2) Allow heifers to adjust to carrier prior to MGA administration.
3) MGA is approved by the FDA for heifers intended for breeding (suppression of estrus) and for heifers fed in confinement for slaughter for increased rate of weight gain, improved feed efficiency, and suppression of estrus.
4) Use of MGA as part of any estrus synchronization protocol in beef cows constitutes and extra label use of medicated feed that is prohibited by the Animal Medicinal Drug Use and Clarification Act and regulation 21 CFR 530.11(b).

Things to Do after Fixed-time Artificial Insemination

Nutrition

Regardless of whether you are developing heifers to attain a target weight or feeding cows to attain adequate body condition at calving ($BCS \geq 5$), nutrition prior to calving and up to the start of the breeding season is of obvious importance. However, nutrition following breeding can also affect embryonic development and survival. A dramatic change in diet or feed intake following FTAI that results in weight loss can negatively impact pregnancy rate.

Heifer development systems will vary depending upon availability of pasture, forage, and supplements. In some cases heifers are developed on pasture or native range and provided a supplement such as dried distillers grains plus solubles (DDGS). Alternatively, heifers may be developed in a feedlot and not have access to pasture or range until near the start of breeding. A study was conducted to evaluate the preceding management strategies for heifer development (Salverson et al., 2009). Heifers were developed on pasture with a DDGS supplement or maintained in a feedlot until estrus synchronization and turnout to grass in the spring. Heifers developed on pasture gained more weight following turnout and had higher pregnancy rates compared to heifers developed in the feedlot. It is not clear whether the increased weight gain in pasture-developed heifers was due to differences in grazing behavior and/or physiological differences between groups. Interestingly, grazing behavior preferences are learned relatively early in a calf's life (Provenza and Balph, 1988) and heifers that grazed from weaning to breeding had better grazing skills during the subsequent grazing season compared to heifers maintained in a feedlot (Olson et al., 2002; Salverson et al., 2009). Therefore, in the preceding study heifers developed on pasture were likely able to graze more efficiently which resulted in a higher average daily gain on pasture and a higher pregnancy rate. In summary, it is essential to ensure that heifers and postpartum cows do not experience significant weight loss following AI. Although the strategy to feed heifers to initiate reproduction and feed the cow herd to sustain reproduction is widely propagated in the beef cattle industry, long-term implications that this approach has on overall production efficiency are not well documented. This management approach removes most, if not all, selection against less efficient animals in a herd. Recent reviews describe benefits of developing heifers to lower target weights than currently recommended by feeding less feed or lower-quality feeds (Funston et al., 2012b) and managing cows with periods of limited or insufficient nutrient availability (Funston et al., 2012c) to enhance production efficiency. The underlying strategy of this approach is that maintaining animals at lighter BW reduces NEm and provides greater opportunity for compensatory responses to small improvements in nutrient environment. It is also expected that implementation of this approach for lifelong management results in adaptation or selection of cows and their offspring that maintain reproductive function under limited nutrient environments, such as occurs during drought or extreme winter stress and in semiarid or arid landscapes, to a greater extent than animals developed or maintained with plentiful or unlimited feed inputs (Roberts et al., 2015).

When can I ship cattle after breeding?

In beef cattle, fertilization rate is frequently 90 to 100% however, pregnancy rate by day 30 to 40 after a single insemination rarely exceeds 70% and calving rate is even lower. Embryonic and fetal mortality may represent the largest economic loss to cow-calf producers (Geary 2006). Pregnancy losses before day 42 post insemination are generally referred to as embryonic loss and

range from 20 to 44% (Humbolt, 2001); whereas, pregnancy losses after day 42 are called fetal loss and are approximately 4% in beef cattle. Factors affecting embryonic/fetal loss are numerous and include genetic abnormalities, fescue toxicosis, plant toxins, excess protein, heat stress, reproductive diseases, an effect of the sire, and handling or shipping stress. In some cases producers ship cattle a long distance to summer or winter pasture following estrus synchronization and AI. Therefore, a common question is “Will shipping stress decrease the pregnancy rate to FTAI?” Shipping cattle on a trailer can induce stress and lead to embryonic/fetal mortality. Pregnancy losses are believed to be due to changes in the uterine environment that adversely affect embryo growth and development. The effect of time of shipping on pregnancy rates following insemination is shown in Table 2. Transporting cattle on a trailer decreased pregnancy rates by about 10% between days 5 and 42 after insemination and by 6% between days 45 and 60. The best time to ship cattle is before synchronization or within 4 days of FTAI.

Table 2. Effect of time of transport after insemination on pregnancy rates.†

	Days after insemination that transportation occurred			
	1 to 4	8 to 12	29 to 33	45 to 60*
Synchronized pregnancy rate	74%	62%	65%	
% pregnancy loss compared to transportation on days 1 to 4		12%	9%	6%*
Breeding season pregnancy rate	95%	94%	94%	

*Loss in heifers compared to percent pregnant prior to transportation (pregnancy determined by transrectal ultrasonography).

†Data adapted from Harrington et al., 1995, and T.W. Geary unpublished data

How do I determine what may have gone wrong during a FTAI program?

Occasionally the pregnancy rate following FTAI is much lower than expected. Trying to identify the root cause of a decreased pregnancy rate can be a daunting task due to the countless factors that can impact pregnancy rate following AI. When trying to trouble shoot what went wrong you should systematically work through the possibilities and not assume anything was done correctly – evaluate all the possibilities! A list of questions that may provide a systematic approach to identifying the problem is provided in Figure 3. Additional points to consider are included below.

What are the most common mistakes when implementing an estrus synchronization and AI program?

One of the most common problems accounting for reduced pregnancy rates following FTAI is that the heifers or cows do not meet the criteria for being good candidates for an estrus synchronization and AI program (see previous section). The second problem is poor choice of an estrus synchronization protocol and (or) protocol compliance. If you have a mixture of cycling and anestrus animals at the beginning of estrus synchronization treatment, you need to use a protocol that includes a progestin (e.g. CIDR or MGA).

Figure 3. Was pregnancy rate to FTAI lower than expected?

1) What was the pregnancy rate in your heifers or cows after 60 to 80 days over the past few years? If less than 85% there may be other issues that should be addressed before initiating an estrus synchronization and AI program.
2) What was the nutrition (protein, energy, phytoestrogens, etc) and mineral program before and after FTAI?
3) Did the animals meet the criteria for being good candidates for an estrus synchronization protocol (see earlier section)?
4) Did you use fixed-time AI or did you breed following detection of estrus? If you inseminated following detection of estrus, how frequently did you detect estrus (when did you begin and when did you end), what criteria did you use for detecting estrus, and when did you inseminate relative to detecting estrus?
5) What bull did you use and is there evidence that semen from this sire has resulted in acceptable pregnancy rates when using fixed-time AI or AI following estrous detection?
6) What protocol did you use and exactly when did you administer each of the products? You will need to confirm that the correct products were administered at the correct dosages and at the correct times. It is helpful to record on a calendar which product was administered on a particular day so you can check back to see if a mistake was made.
7) Was the biological activity of the various products compromised? You will need to verify that the products were not out of date and were stored and administered properly.
8) If using fixed-time AI, when did you inseminate the heifers or cows? Did you record who inseminated each animal? This will be helpful in identifying if there is a technician problem.
9) Where did you obtain the semen, how was it stored, and was the semen thawed correctly?

Progestin treatment will increase the proportion of prepuberal heifers and anestrus cows that will respond to the protocol. Furthermore, it is essential that each heifer or cow receives the correct estrus synchronization product, at the correct dose, and on the appropriate day. A third problem is that the facilities don't allow the cattle to be inseminated properly within a 2 to 3 hr time period and/or cause undue stress on the cattle. With a FTAI protocol you have to carefully consider how many animals you can inseminate properly within the designated time period (e.g. 66 ± 2 hr for CO-Synch + CIDR protocol) with a minimum of stress. As previously mentioned, renting a breeding barn (Figure 1) or contracting with an AI company that has breeding barns available can alleviate the problems associated with marginal facilities.

Biological activity of the estrus synchronization products

It is not uncommon to hear someone blame a particular estrus synchronization product or the protocol for poor results. The commercially available products are effective when properly stored and administered. Furthermore, the protocols have been shown to consistently work in a variety of environments. The estrus synchronization protocols listed in the AI catalogs published

by Select Sires, ABS Global, Genex, and Accelerated Genetics have been thoroughly tested in the field in a number of herds by numerous investigators in many states. Rarely does one find that the biological activity of a particular product has been compromised provided the product has been stored properly, administered at the appropriate dose on the correct day of the protocol, and that the expiration date has not been exceeded. It is not uncommon for PGF or GnRH products to be administered at the wrong dose or to be injected subcutaneously instead of in the muscle. Intramuscular injections should be administered using an eighteen-gauge, 1.5 inch needle to minimize the possibility of back flow.

Potential problems associated with feeding melengestrol acetate (MGA)

Occasionally there can be problems with feeding melengestrol acetate (MGA) if you don't pay attention to a few simple guidelines (Figure 3). The most common problem is that a heifer does not receive the correct dose (0.5 mg/hd/day). If a heifer does not receive enough MGA she may express estrus during the period of MGA feeding. Therefore, it is a good idea to watch the heifers for estrous activity as they come to the bunk. Alternatively, if a heifer receives more than the appropriate dose, expression of estrus may be delayed following the end of MGA feeding. To ensure that each heifer has an opportunity to receive the correct dose, MGA should be fed once daily in 3 to 5 pounds of carrier and each heifer should have 18 to 24 inches of bunk space. To be confident there is adequate bunk space and to train the heifers to come to the bunk it is a good idea to feed the carrier without MGA for a few days before the start of MGA treatment. At the end of 14 days of MGA feeding, heifers will express estrus within 2 to 5 days; however, heifers should not be inseminated at this estrus since pregnancy rates will be reduced. Be sure to inseminate the heifers at the designated time specified in the protocol.

Potential problems associated with CIDRs

Controlled Internal Drug Release (CIDR) is an intravaginal device that contains progesterone and acts like an artificial corpus luteum. Information on the proper handling and administration of CIDRs is provided in Figure 3. Normally there are few problems associated with CIDR treatment. CIDRs should not be inserted in cows that are less than 21 days postpartum because the probability of inducing cyclicity is low. CIDR insertion should be performed as cleanly as possible in order to reduce the risk of spreading disease (see Figure 3). When removing CIDRs it is not uncommon to detect a whitish discharge which is due to vaginal irritation from the wings of the CIDR and does not necessarily mean the animal has a vaginal infection. A difference in conception rate or pregnancy rate has not been detected between CIDR-treated animals that do or do not have a discharge.

Summary

There are significant benefits to genetic improvement and reproductive management that can be gained from the implementation of an estrus synchronization and AI program in heifers and postpartum beef cows. Artificial insemination in beef cattle is more practical than ever due to advances in FTAI, identification of sires with highly accurate EPDs, and a market structure that can identify and reward producers for the quality of their cattle. Above all, a successful estrus synchronization and AI program is dependent upon being proactive, well organized, and attention to detail. The success of these systems hinges on many factors. A check list of tips for a successful estrus synchronization and AI program is provided in Figure 4.

Figure 4. Check list of tips for a successful estrus synchronization and AI program.

Things to do before fixed-time artificial insemination
<ul style="list-style-type: none"> • Keep accurate calving, breeding, and pregnancy records. • Animal identification should be clear and easily readable. • Ensure herd health and disease prevention with a well-designed prebreeding vaccination protocol. Vaccinate females a minimum of 30 days before the breeding season begins. • Decide which estrus synchronization protocol best fits your breeding program, facilities, and personnel (see protocol sheets in AI catalogs). • Ensure all products are purchased and on-hand prior to initiation of the protocol. • Prepare the calendar of actions to ensure protocol compliance.
Sire selection
<ul style="list-style-type: none"> • Determine if you will purchase or raise replacement heifers. • Decide how you will market your calves. • Select proven AI sires with high-accuracy EPDs that match performance goals. • Purchase semen from a Certified Semen Services (CSS) collection facility. • Prepare or update your semen inventory. • Make sure females meet the criteria for being good candidates for estrus synchronization.
Heifer criteria
<ul style="list-style-type: none"> • Heifers may be developed utilizing a variety of resources, research over the past decade has demonstrated acceptable pregnancy rates in heifers developed from 50 – 57% of mature body weight. • Expose additional heifers beyond replacement needs to determine how your genetics responds to a lower input system if implemented.
Cow criteria
<ul style="list-style-type: none"> • Synchronize and inseminate only cows with BCS at calving of ≥ 5 (1 = emaciated; 9.0 = obese). • The average days postpartum of the group of cows to be synchronized should be ≥ 40 by the start of estrus synchronization and experience a minimum of dystocia.
Things to do at the time of estrus synchronization and artificial insemination
<ul style="list-style-type: none"> • Meticulously follow the estrus synchronization protocol! • If detecting estrus, spend as much time observing the animals as possible. • Use a minimum of one person to detect estrus per 100 head of cattle. • Use estrous detection aids to facilitate visual observation of estrus. • Use a properly trained technician for AI.
Things to do after fixed-time artificial insemination
<ul style="list-style-type: none"> • To distinguish between AI and bull bred pregnancies at pregnancy diagnosis, you should wait approximately 10 days to turn in clean up bulls after AI. • Pregnancy check by 75 days after AI via ultrasound or 80 to 90 days after AI via rectal palpation to distinguish AI from bull bred pregnancies. • If cattle need to be shipped do so between days 1 to 4 after AI and avoid shipping cattle between days 5 to 42 after AI. • Maintain breeding females on an adequate nutrition and mineral program.
PAY ATTENTION TO DETAILS!

Literature Cited

- Brown, E. G., G. E. Carstens, J. T. Fox, M. B. White, T. W. Welsh, Jr., R.D. Randel, and J.W. Holloway. 2004. Relationships between temperament and performance traits of growing calves. In: 2004 Beef cattle research in Texas. Available at: http://animalscience.tamu.edu/ansc/beef/bcrt/2004/brown_erin.pdf.
- Burris, M. J., and B. M. Priode. 1958. Effect of calving date on subsequent calving performance. *Journal of Animal Science* 17:527-533.
- Chiang, B. C., P. G. Smith, K. E. Nusbaum, and D. A. Stringfellow. 1990. The effect of infectious bovine rhinotracheitis vaccine on reproductive efficiency in cattle vaccinated during estrus. *Theriogenology* 33:1113-1120.
- Cooke, R. F., J. D. Arthington, D. B. Araujo, and G. C. Lamb. 2009a. Effects of acclimation to human interaction on performance, temperament, physiological responses, and pregnancy rates of Brahman-crossbred cows. *J. Anim. Sci.* 87:4125-4132.
- Cooke, R. F., J. D. Arthington, B. R. Austin, and J. V. Yelich. 2009b. Effects of acclimation to handling on performance, reproductive, and physiological responses of Brahman-crossbred heifers. *J. Anim. Sci.* 87:3403-3412.
- Cooke, R. 2010. Effects of temperament and animal handling on fertility. *Applied Reproductive Strategies in Beef Cattle Proceedings August 5 and 6, Nashville, Tennessee* pages 255-263.
- Cushman, R. A., L. K. Kill, R. N. Funston, E. M. Mousel, and G.A. Perry. 2013. Heifer calving date positively influences calf weaning weights through six parturitions. *J. Anim. Sci.* 91:4486-4491.
- Daly, R. 2007a. Control of infectious reproductive disease: The role of biosecurity. *Applied Reproductive Strategies in Beef Cattle Proceedings September 11 and 12, Billings, Montana*, pages 197 – 208.
- Daly, R. 2007b. Timing of reproductive vaccinations in beef cattle herds. *Applied Reproductive Strategies in Beef Cattle Proceedings September 11 and 12, Billings, Montana*, pages 209 – 214.
- Funston, R.N., J.A. Musgrave, T.L. Meyer, and D.M. Larson. 2012a. Effect of calving distribution on beef cattle progeny performance. *J. Anim. Sci.* 90:5118-5121.
- Funston, R. N., J. L. Martin, D. M. Larson, and A. J. Roberts. 2012b. Physiology and Endocrinology Symposium: Nutritional aspects of developing replacement heifers. *J. Anim. Sci.* 90:1166–1171.
- Funston, R. N., A. F. Summers, and A. J. Roberts. 2012c. Alparma Beef Cattle Nutrition Symposium: Implications of nutritional management for beef cow-calf systems. *J. Anim. Sci.* 90:2301–2307.
- Geary, T. W. 2006 Management strategies to reduce embryonic loss. *Applied Reproductive Strategies in Beef Cattle Proceedings October 3 and 4. Rapid City, South Dakota*, pages 167 – 175.
- Humbolt, P. 2001. Use of pregnancy specific proteins and progesterone assays to monitor pregnancy and determine the timing, frequencies, and sources of embryonic mortality in ruminants. *Theriogenology* 56:1417-1433.
- Miller, J. M. 1991. The effects of IBR virus infection on reproductive function of cattle. *Vet. Med.* January: 95-98. Miller, J. M., M. J. Van Der Maaten, and C. A. Whetstone. 1989. Infertility in heifers inoculated with modified-live bovine herpesvirus-1 vaccinal strains

against infectious bovine rhinotracheitis on postbreeding day 14. *Am. J. Vet. Res.* 50:551-554.

- Nkrumah, J. D., D. H. Crews, Jr, J. A. Basarab, M. A. Price, E. K. Okine, Z. Wang, C. Li, and S. S. Moore. 2007. Genetic and phenotypic relationships of feeding behavior and temperament with performance, feed efficiency, ultrasound, and carcass merit of beef cattle. *J. Anim. Sci.* 85:2382-2390.
- Olson, K. C., J. R. Jaeger, and J. R. Brethour. 1992. Growth and reproductive performance of heifers overwintered in range or drylot environments. *Journal Production Agriculture* 5:72-76.
- Perry, R. C., L. R. Corah, G. H. Kiracofe, J. S. Stevenson, and W. E. Beal. 1991. Endocrine changes and ultrasonography of ovaries in suckled beef cows during resumption of postpartum estrous cycles. *J. Anim. Sci.* 69:2548-2555.
- Provenza, F. D., and D. F. Balph. 1988. Development of dietary choice in livestock on rangelands and its implications for management. *J. Anim. Sci.* 66:2356-2368.
- Roberts, A. J., M. K. Petersen, and R. N. Funston. 2015. BEEF SPECIES SYMPOSIUM: Can we build the cowherd by increasing longevity of females? *J. Anim. Sci.* 93:4235-4243.
- Salverson, R. R., H. H. Patterson, G. A. Perry, D. Young, and M. L. Gibson. 2005. Evaluation of performance and costs of two heifer development systems. *South Dakota State University Beef Report.* p. 13-18.
- Smith, P. C., K. E. Nusbaum, R. P. Kwapien, D. A. Stringfellow, and K. Driggers. 1990. Necrotic oophoritis in heifers vaccinated intravenously with infectious bovine rhinotracheitis virus vaccine during estrus. *Amer. J. Vet. Res.* 51:969-972.
- Stormshak, F., C. M. Tucker, W. E. Beal, and L.R. Corah. 1997. Reproductive responses of beef heifers after concurrent administration of vaccines, anthelmintic and progestogen. *Theriogenology* 47:997-1001.
- Tibbitts, B. T., H. R. Nielson, K. H. Ramsay, and R. N. Funston. 2017. Growth and reproductive performance of yearling beef heifers implanted with Revalor G in the Nebraska Sandhills. *Prof. Anim. Sci.* 33:92-96.
- Van Der Maaten, M. J., and J. M. Miller. 1985. Ovarian lesions in heifers exposed to infectious bovine rhinotracheitis virus by non-genital routes on the day after breeding. *Vet. Micro.* 10:155-163.
- Werth, L. A., J. C. Whittier, S. M. Azzam, G. H. Deutscher, and J. E. Kinder. 1996. Relationship between circulating progesterone and conception at the first postpartum estrus in young primiparous beef cows. *J. Anim. Sci.* 74:616-619.

Rick Funston is a professor and reproductive physiologist at the University of Nebraska. He received his BS from North Dakota State University, MS from Montana State University, PhD from the University of Wyoming, and completed post-doctoral work at Colorado State University. His research on lighter heifer development is receiving national attention/adoption; research on fetal programming effects on postnatal calf performance including carcass characteristics and reproduction has received national and international recognition; and he is a team member of nationally recognized beef systems research. In extension, he provides leadership and subject matter expertise for educational programs in cow-calf production and reproductive management.

The High Fertility Cycle

Paul M. Fricke¹, Milo C. Wiltbank¹, and J. Richard Pursley²

¹Department of Dairy Science, University of Wisconsin – Madison

²Department of Animal Science, Michigan State University

Corresponding author: pmfricke@wisc.edu

SUMMARY

- Over the past two decades, a reproduction revolution has occurred in the dairy industry in which average 21-day pregnancy rates have more than doubled from around 14% to more than 30% in many herds.
- Much of this increase in reproductive performance has been driven by development and adoption of fertility programs.
- Despite the dramatic increase in 21-day pregnancy rates, substantial variation exists among herds using the exact same reproductive management suggesting that factors other than fertility programs can affect fertility.
- Change in body weight or body condition score postpartum or during the periparturient period dramatically affects embryo quality, reproductive outcomes, and transition cow health.
- Although some cows lose body weight or body condition score after calving, some cows maintain, whereas some cows even gain body weight or body condition score during this time.
- Surprisingly, milk production during early lactation is not affected based on body condition score change during the first 3 weeks postpartum; however, peak milk measured near 60 DIM was less in both primiparous and multiparous cows that either gained or maintained compared to cows that lost body condition during the 1st 30 DIM.
- The high fertility cycle coupled with the dramatic increases in reproductive performance due to the development and adoption of fertility programs is a new paradigm that we can now use to explain much of the variation in reproductive performance among herds.
- The high-fertility cycle: How timely pregnancies in one lactation may lead to less BCS loss, fewer health issues, greater fertility, and reduced early pregnancy losses in the next lactation.

INTRODUCTION

Over the past two decades, a reproduction revolution has occurred in the dairy industry. Twenty years ago, the 21-day pregnancy rate in U.S. dairy herds averaged about 14% with conception rates rarely exceeding 40%. In 1998, the annualized 21-day pregnancy rate goal was 20% which few herds could achieve. Today, the average 21-day pregnancy rate in the U.S. exceeds 21% with more than 60% of DRMS Holstein herds achieving 21-day pregnancy rates greater than 20% with average conception rates that exceed 50% in high-producing Holsteins. The development of fertility programs and their adoption by the dairy industry over the past decade has largely driven

this reproduction revolution (Carvalho et al., 2018). Fertility programs, such as Double-Ovsynch or G6G protocols for first timed AI not only increase the AI service rate, but also increase pregnancies per AI (P/AI) beyond that achieved based on AI to a detected estrus (Santos et al., 2017). Despite this increase in reproductive performance, many veterinarians, nutritionists, and consultants observe dramatic variation in reproductive performance among herds that manage reproduction using the exact same reproductive management programs. Although on-farm protocol compliance with complex fertility programs that require multiple treatments across many days remains an issue, it cannot explain all of this variation among herds.

The “Britt Hypothesis”

In 1992, Dr. Jack Britt sorted 76 lactating Holstein cows based on whether they Lost (Lost, n = 30) or Maintained (n = 46) BCS during the first 5 weeks after calving (Britt, 1992). Body condition scores were recorded for the first 10 weeks after calving for these two groups of cows (Figure 1).

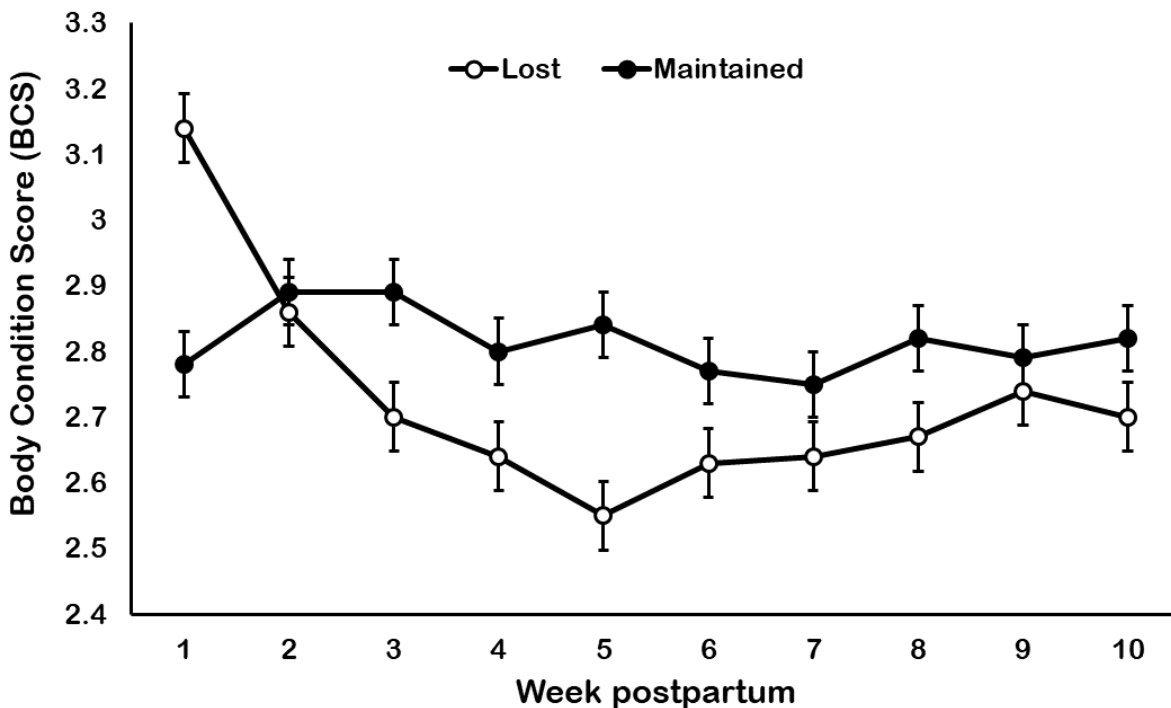


Figure 1. Change in body condition score (BCS) in Holstein cows (n = 76) during the first 10 weeks postpartum. Cows were sorted into two groups based on whether they Lost (Lost, n = 30) or Maintained (n = 46) BCS during the first 5 weeks postpartum. Adapted from Britt (1992).

Cows that maintained BCS post calving had a greater conception rate at first service than cows that lost BCS post-calving (Table 1). Based on these data, Dr. Britt speculated that high producing cows which experience severe weight losses during the first 3 to 5 weeks after calving presumably subject their developing follicles to adverse metabolic conditions associated with the rapid weight loss that compromises fertility later during lactation at first insemination (Britt, 1992). The results from three recent studies; two from the University of Wisconsin - Madison, and one from

Michigan State University, support Dr. Britt's observation from 1992 and challenge the long-held assumption that all cows normally lose BCS after calving.

Table 1. Results of retrospective analysis of data from Holstein cows sorted based on BCS change during the first 5 weeks postpartum. Adapted from Britt, 1992.

Item	Lost	Maintained
n	30	46
BCS ¹ change		
Week 1 to 5	-0.58 ^a	+0.06 ^b
Week 5 to 10	+0.17 ^a	-0.02 ^b
Interval to first ovulation (d)	23.3 ^a	17.2 ^b
Milk yield		
Mean during first 70 d (lbs)	60	58
Mean 305 d lactation (lbs)	18,198	17,941
Interval to first AI (d)	82.9	84.9
Conception rate		
First service (%)	25 ^a	62 ^b
All services (%)	42 ^a	61 ^b

^{a,b}Items with different superscripts differ (P < 0.05)

¹Body condition scores based on a 1 (thin) to 5 (fat) scale.

Effect of body weight change on embryo quality

The first study from the first paper (Carvalho et al., 2014) included an experiment in which lactating Holstein cows (n = 71; 27 primiparous and 44 multiparous) were weighed weekly from calving until 10 weeks postpartum. Cows were divided into quartiles based on percent body weight change from the first week after calving (Figure 2). The quartile analysis divided cows based on those that gained weight (First Quartile), maintained weight (Second Quartile), slightly lost weight (Third Quartile), and dramatically lost weight (Fourth Quartile), and the majority of the body weight change occurred during the first 3 weeks postpartum (Figure 2). Cows in the Fourth Quartile that dramatically lost weight had increased NEFA concentrations during the first 3 weeks after calving, whereas NEFA concentrations did not differ at 10 weeks postpartum when superovulation and embryo flushing was performed (Carvalho et al., 2014).

To assess embryo quality, cows were superovulated using a modified Double-Ovsynch protocol. All cows were inseminated and flushed by two technicians, and cows were inseminated twice at 12 and 24 h after GnRH treatment. Seven days after GnRH treatment, ova/embryos were recovered using a nonsurgical shallow uterine horn flushing technique. Embryo characteristics were affected based on body weight quartile in which cows in the Fourth Quartile that dramatically lost weight during the first 3 weeks postpartum had overall poorer embryo characteristics than cows in the other three quartiles (Table 2).

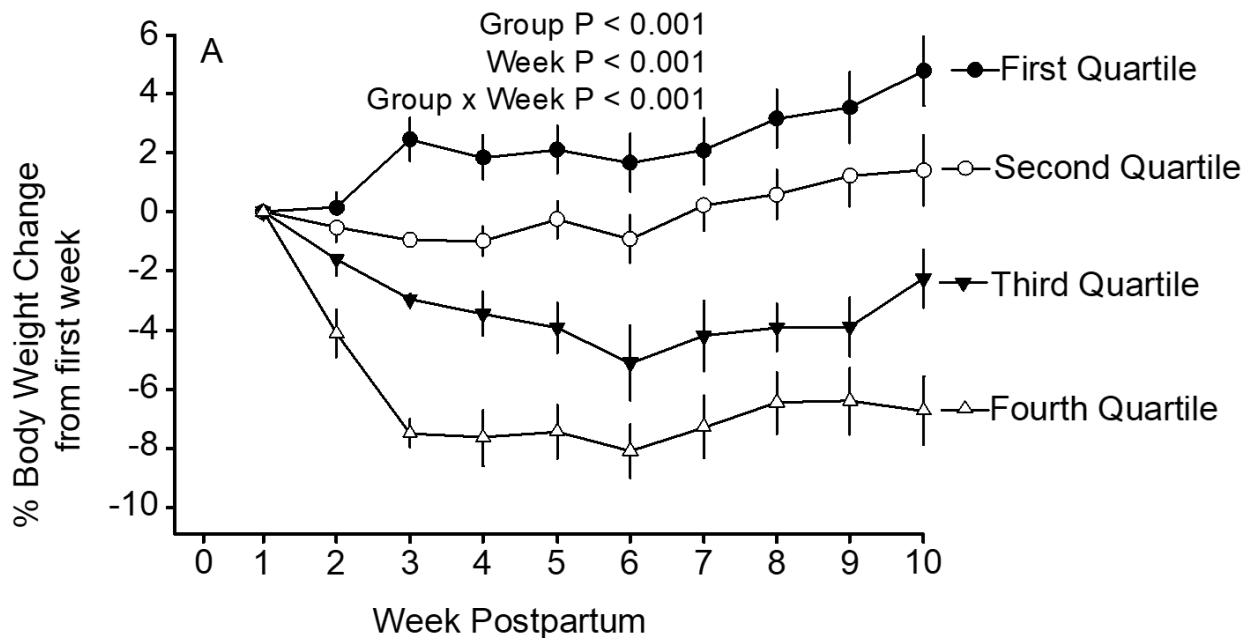


Figure 2. Quartile analysis of percent body weight change from the first week postpartum in Holstein dairy cows. Adapted from Carvalho et al. (2014).

Table 2. Embryo characteristics of lactating Holstein cows based on body weight change¹ from first to third week postpartum. Adapted from Carvalho et al. (2014).

Item	Fourth Quartile	Third Quartile	Second Quartile	First Quartile	P
CL (number)	18.4 ± 2.6	18.4 ± 1.7	19.0 ± 1.7	16.0 ± 2.0	0.67
Fert structures (#)	7.6 ± 2.1	7.3 ± 1.1	4.8 ± 1.1	5.8 ± 1.4	0.43
Deg embryos (#)	2.7 ± 0.7 ^a	1.7 ± 0.7 ^{ab}	0.7 ± 0.2 ^b	0.6 ± 0.2 ^b	0.02
Quality 1 & 2 (#)	4.2 ± 1.4	5.3 ± 0.9	3.9 ± 1.1	4.9 ± 1.4	0.47
Quality 1, 2 & 3 (#)	4.9 ± 1.6	5.6 ± 0.8	4.1 ± 1.1	5.3 ± 1.4	0.49
Fertilized (%)	76.9 ± 7.1	77.0 ± 6.6	77.6 ± 7.6	78.4 ± 7.1	0.99
Degenerate (%)	35.2 ± 8.5 ^a	12.6 ± 4.6 ^b	14.5 ± 6.3 ^b	9.6 ± 3.7 ^b	0.02
Quality 1 & 2 (%)	38.0 ± 8.7 ^{b,B}	61.3 ± 8.2 ^{ab,A}	60.6 ± 9.4 ^{ab,A}	63.4 ± 8.6 ^{a,A}	0.14
Quality 1, 2 & 3 (%)	41.7 ± 8.8 ^{b,B}	64.4 ± 8.2 ^{ab,A}	63.1 ± 9.3 ^{ab,A}	68.9 ± 8.7 ^{a,A}	0.13
Degen of Fert (%)	46.9 ± 9.6 ^{a,A}	17.4 ± 6.4 ^{b,B}	24.8 ± 9.3 ^{ab,A}	16.2 ± 7.0 ^{b,B}	0.04
1 & 2 of Fert (%)	48.4 ± 9.5 ^b	78.3 ± 6.6 ^a	72.6 ± 9.5 ^a	77.7 ± 7.4 ^a	0.05
1, 2 & 3 of Fert (%)	53.2 ± 9.6 ^{b,B}	82.6 ± 6.4 ^{a,A}	75.2 ± 9.3 ^{a,AB}	83.8 ± 7.0 ^{a,A}	0.04
Recovery Rate (%)	45.6 ± 7.4	55.1 ± 6.9	35.4 ± 6.7	45.3 ± 5.8	0.25

^{a,b}Items with different superscripts within the same row differ (P < 0.05).

^{A,B}Items with different superscripts within the same row differ (P < 0.15).

¹First quartile = gaining body weight; Fourth quartile = most body weight loss.

Effect of BCS change after calving on fertility

The second study from the first paper (Carvalho et al., 2014) included a retrospective analysis in which 1,887 Holstein cows from two commercial dairy farms in Wisconsin were submitted to a Double-Ovsynch protocol for first timed AI, and BCS was evaluated at calving and 21 days after calving. Overall, 42% of cows lost BCS, 36% of cows maintained BCS, and 22% of cows gained BCS during the first 3 weeks of lactation (Table 3).

Table 3. Effect of BCS change on pregnancies /AI (P/AI) for cows on Farm 1 and 2 classified as losing, maintaining or gaining BCS from parturition to three weeks postpartum. Adapted from Carvalho et al. (2014).

Item	BCS ² change		
	Lost	Maintained	Gained
All cows			
% of cows, (n)	41.8 (789/1887)	35.8 (675/1887)	22.4 (423/1887)
P/AI at 40 d, % (n/n)	25.1 (198/789) ^c	38.2 (258/675) ^b	83.5 (353/423) ^a
P/AI at 70 d, % (n/n)	22.8 (180/789) ^c	36.0 (243/675) ^b	78.3 (331/423) ^a
Pregnancy Loss, % (n/n)	9.1 (18/198)	5.8 (15/258)	6.2 (22/353)
BCS at parturition	2.93 ± 0.01 ^a	2.89 ± 0.02 ^b	2.85 ± 0.02 ^b
BCS at 21 DIM	2.64 ± 0.01 ^c	2.89 ± 0.02 ^b	3.10 ± 0.02 ^a
ECM (kg/d) ¹	30.9 ± 0.4	31.5 ± 0.4	28.7 ± 0.4

^{a,b,c}Items with different superscripts within the same row differ (P < 0.05).

¹Mean Energy Corrected Milk from calving to 21 DIM.

²Body Condition Score was evaluated at calving and at 21 DIM based on a point 5 scale.

Similar to the experiment by Britt (1992), energy corrected milk (ECM) did not differ among cows based on BCS change (Table 3). Most impressively, P/AI 40 d after timed AI was only 25% for cows that lost BCS, 38% for cows that maintained BCS, and was 84% for cows that gained BCS. It is important to note that there were dramatic farms effects in this study in which one farm had most of the cows that gained BCS (Carvalho et al., 2014). Based on data presented thus far, the key question is: can we increase the proportion of cows that gain BCS after calving? The next study by Barletta et al. (2017) helps us to answer this question.

Effect of BCS change during the periparturient period on reproduction and health

In the second study (Barletta et al., 2017), BCS change was evaluated in 233 Holstein cows from 3 weeks before the expected date of calving until 3 weeks after calving (Table 4). Similar to the experiment by Carvalho et al. (2014), P/AI 30 d after AI for cows submitted to first timed AI was 18% for cows that lost BCS (28% of cows), 27% for cows that maintained BCS (23% of cows), and 53% for cows that gained BCS (49% of cows). Average milk production during the first 3 weeks of lactation did not differ among cows based on BCS change during the periparturient period.

Table 4. Effect of changes in body condition score (BCS) during the transition period on pregnancies per artificial insemination (P/AI) and pregnancy loss. Adapted from Barletta et al. (2017).

Item	Change in BCS ¹			P-value
	Gained	Maintained	Lost	
Cows, % (no./no.)	28 (69/245)	22 (54/245)	50 (122/245)	
P/AI 30 d, % (no./no.)	53.0 (35/66) ^a	26.9 (14/52) ^b	18.3 (21/115) ^b	< 0.01
P/AI 60 d, % (no./no.)	45.5 (30/66) ^a	25.0 (13/52) ^b	15.7 (18/155) ^b	< 0.01
Pregnancy loss, % (no./no.)	14.3 (5/35)	7.1 (1/14)	14.3 (3/21)	0.79

^{a/c}Within a row, items with different superscripts differ (P < 0.05).

¹BCS was evaluated during the transition period (-21 to 21 d) using a 5-point scale.

In addition to increased fertility, cows that gained BCS during the periparturient period were also healthier, with less than 40% of these cows experiencing more than one health event, whereas greater than 60% of cows that lost BCS after calving experienced more than one health event (Table 5).

Table 5. Effect of changes in body condition score (BCS) during the transition period (-21 to 21) on incidence (%) of retained placenta, mastitis, ketosis and pneumonia for cows that lost, maintained, or gained BCS. Adapted from Barletta et al. (2017).

Item	Change in BCS ¹			P-value
	Gained	Maintained	Lost	
n	66	52	116	
Metritis	19.70 (13/66)	21.20 (11/52)	23.30 (27/116)	0.85
Mastitis	16.70 (11/66) ^b	17.30 (9/52) ^{a,b}	29.30 (34/116) ^a	0.09
Ketosis	15.20 (10/66)	19.20 (10/52)	26.70 (31/116)	0.18
Pneumonia	9.10 (6/66)	11.50 (6/52)	14.70 (17/116)	0.55
> 1 Health problem	39.4 (26/66) ^b	46.2 (24/52) ^b	62.9 (73/116) ^a	0.007

In this study by Barletta et al. (2017), the major factor associated with BCS change during the transition period was BCS 3 weeks before expected calving. Only 34% of cows with BCS less than 3.0 lost BCS during the transition period, whereas 51% of cows with BCS = 3.0 lost BCS and 92% of cows with BCS > 3.0 lost BCS. So, how can we ensure that more cows gain BCS after calving? Nearly all of the cows in the study by Barletta et al. (2017) that gained BCS during the transition period had a BCS less than 3.0 3 weeks before calving. Thus, calving cows at a lower BCS was associated with less BCS loss, greater fertility, and fewer health issues. Based on data presented thus far, the next question is: how do I prevent calving cows with a high BCS? The final study provides the answer to this question.

The High Fertility Cycle

The final study evaluated BCS change within 1 week of calving until 30 days after calving in 851 Holstein cows on a commercial dairy farm in Michigan (Middleton et al., 2019). This study linked previous calving intervals of individual cows to BCS changes after calving. Calving interval is

determined by the fixed interval of gestation length and the highly variable interval of calving to conception. Thus, cows with longer calving intervals during the previous lactation took longer to get pregnant than cows with shorter calving intervals. In this study, cows with longer calving intervals in the prior lactation had greater BCS at calving and lost BCS during the first 30 days after calving. In agreement with the first two studies (Carvalho et al., 2014; Barletta et al., 2017), cows that maintained or gained BCS after calving had greater conception rates, less pregnancy loss, and were healthier than cows that lost BCS after calving (Middleton et al., 2019). Amazingly, even when cows with health problems were removed from the data set, differences in conception rates and pregnancy losses in favor of cows that maintained or gained body condition during the 1st 30 DIM were maintained. An excellent overview of the results from this study is captured by the title of the paper: The high-fertility cycle: How timely pregnancies in one lactation may lead to less BCS loss, fewer health issues, greater fertility, and reduced early pregnancy losses in the next lactation (Figure 3).

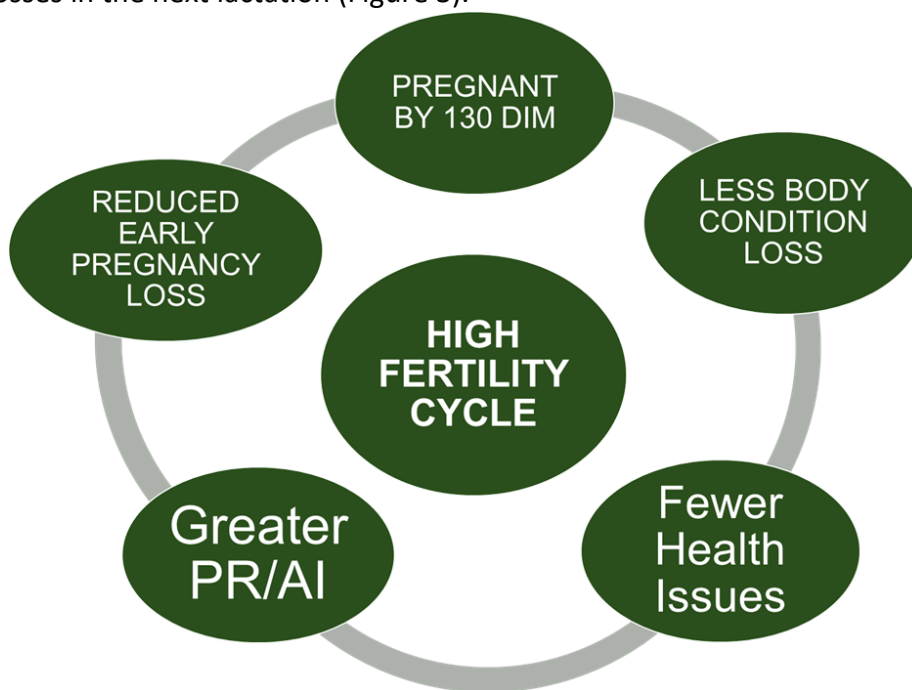


Figure 3. The high-fertility cycle: How timely pregnancies in one lactation may lead to less BCS loss, fewer health issues, greater fertility, and reduced early pregnancy losses in the next lactation. Adapted from Middleton et al. (2019).

CONCLUSION

Based on the collective results from these studies we can now clearly define a relationship in which herds that manage to get their cows pregnant rapidly after the end of the voluntary waiting period calve cows at a lower BCS which in turn leads to more cows maintaining or gaining BCS after calving. Cows that maintain or gain BCS after calving have greater fertility than cows that lose BCS. The High Fertility Cycle coupled with the dramatic increases in reproductive

performance due to the development and adoption of fertility programs is a new paradigm that we can now use to explain much of the variation in reproductive performance among herds. The goal of every farm should be to strive to get their cows into the high-fertility cycle and keep them there. The following are key considerations to achieve this: 1) implement BCS monitoring for transition cows 3 weeks before calving, at calving, 3 weeks after calving, and at AI; 2) use fertility programs to help get cows pregnant quickly after the end of the voluntary waiting period; 3) set a hard cutoff for the number times individual cows will be inseminated; and 4) consider nutritional strategies to prevent late lactation cows from gaining too much body condition.

REFERENCES

- Barletta, R. V., M. Maturana Filho, P. D. Carvalho, T. A. Del Valle, A. S. Netto, F. P. Rennó, R. D. Mingoti, J. R. Gandra, G. B. Mourão, P. M. Fricke, R. Sartori, E. H. Madureira, and M. C. Wiltbank. 2017. Association of changes among body condition score during the transition period with NEFA and BHBA concentrations, milk production, fertility, and health of Holstein cows. *Theriogenology* 104:30-36.
- Britt, J. 1992. Impacts of early postpartum metabolism on follicular development and fertility. Pages 29–43 in *Proc. Annu. Conv. Am. Assoc. Bovine Pract. Am. Assoc. Bovine Pract.*, Auburn, AL.
- Carvalho, P. D., V. G. Santos, J. O. Giordano, M. C. Wiltbank, and P. M. Fricke. 2018. Development of fertility programs to achieve high 21-day pregnancy rates in high-producing dairy cows. *Theriogenology* 114:165-172.
- Carvalho, P. D., A. H. Souza, M. C. Amundson, K. S. Hackbart, M. J. Fuenzalida, M. M. Herlihy, H. Ayres, A. R. Dresch, L. M. Vieira, J. N. Guenther, P. M. Fricke, R. D. Shaver, and M. C. Wiltbank. 2014. Relationships between fertility and postpartum changes in body condition and body weight in lactating dairy cows. *J. Dairy Sci.* 97:3666-3683.
- Middleton, E. L., T. Minela, and J. R. Pursley. 2019. The high-fertility cycle: How timely pregnancies in one lactation may lead to less body condition loss, fewer health issues, greater fertility, and reduced early pregnancy losses in the next lactation. *J. Dairy Sci.* 102:5577-5587.
- Santos, V. G., P. D. Carvalho, C. Maia, B. Carneiro, A. Valenza, and P. M. Fricke. 2017. Fertility of lactating Holstein cows submitted to a Double-Ovsynch protocol and timed artificial insemination versus artificial insemination after synchronization of estrus at a similar day in milk range. *J. Dairy Sci.* 100:8507-8517.

Adipose tissue as an integrator of metabolic and inflammatory signals in periparturient cows

G. Andres Contreras, DVM MS PhD

Department of Large Animal Clinical Sciences, College of Veterinary Medicine,
Michigan State University, East Lansing, MI 48824

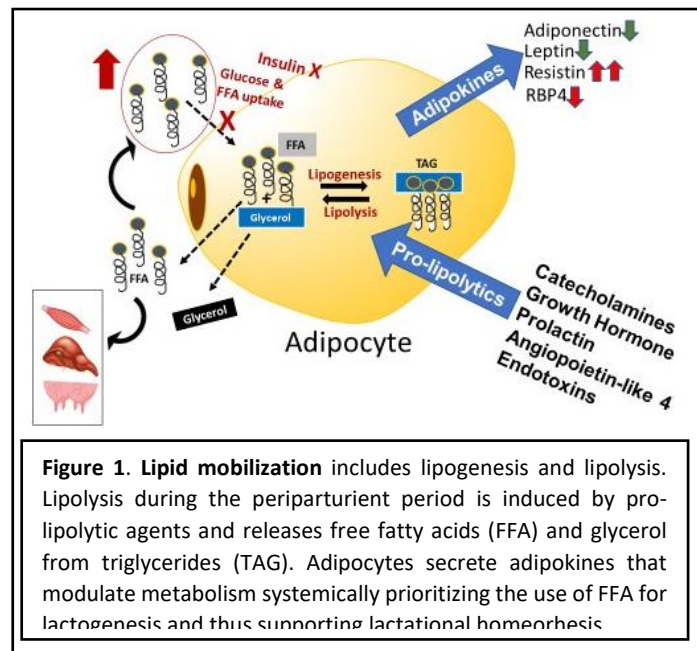
The adipose tissue (AT) is a multisite organ that participates in the endocrine and metabolic adaptations to the onset of lactation in periparturient dairy cows. Three primary AT functions secure a continuous supply of energy to maintain milk secretion and bodily function during periparturient negative energy balance. First, AT releases free fatty acids from triglycerides molecules stored in its adipocytes through lipolysis. Second, AT cellular components secrete peptides, also termed adipokines, that directly and indirectly adapt other organs to use free fatty acids (FFA) as energy substrates (Contreras et al., 2017). Third, the AT undergoes a remodeling process during the periparturient period due to the rapid loss of lipid reserves. This process includes infiltration of anti-inflammatory macrophages that promote the differentiation of new adipocytes capable of buffering the FFA excess accumulated during the first 2-3 weeks after parturition (Contreras et al., 2018). This summary paper highlights metabolic and endocrine functions of AT that are necessary for an effective transition from the dry period to early lactation in dairy cows.

Lipid mobilization

Within the adipocytes, FFA are constantly esterified (i.e., lipogenesis) and hydrolyzed (i.e., lipolysis) to and from triglyceride molecules. This process is known as lipid or fat mobilization. Around parturition due to the negative energy balance, the rate of lipolysis surpasses that of lipogenesis.

Consequently, the AT releases FFA into circulation. Lipolysis can be broadly divided into basal and demand lipolysis (Lafontan and Langin, 2009). The rate of basal lipolysis increases with adipocyte volume. For this reason, over-conditioned cows that have large

adipocytes release more FFA at basal conditions than lean cows (De Koster et al., 2016). Around parturition, demand lipolysis is regulated hormonally. Its primary activators are catecholamines, growth hormone, angiopoietin-like 4, and prolactin (Figure 1). However, lipolytic signals can be exacerbated during infectious and metabolic diseases by the presence of endotoxins in blood that directly activate adipocyte lipases and impair the response of adipocytes to insulin (Chirivi et al., 2021).



The rapid increase in demand lipolysis during the periparturient period coincides with a drastic reduction in lipogenesis. This change is related to low plasma insulin, adipocyte insulin resistance, and the AT's inflammatory responses. All these factors inhibit the transcription of genes that promote de novo lipogenesis and triglyceride assembly. In healthy cows, as lactation progresses, energy balance becomes positive, plasma insulin returns to pre-calving levels, and AT lipolysis and inflammation are reduced. In contrast, cows that do not transition well into lactation exhibit an impaired response to the anti-lipolytic effects of insulin driven by chronic AT inflammation leading to lipolysis dysregulation (Contreras et al., 2015).

Adipokines as regulators of metabolic function

The AT controls systemic energy homeostasis by modulating the availability of energy-dense FFA and by secreting adipokines that have endo-, para-, and autocrine functions. These peptides are produced by the adipocytes and immune and vascular cells that reside in AT. Although there are over 200 adipokines described, only adiponectin, leptin, resistin, and retinol-binding protein 4 (RBP4) have been studied in dairy cattle in detail. It is important to note that the periparturient secretion patterns of these adipokines support lactation energy needs by redirecting glucose to the mammary gland, increasing FFA flow to the liver and epithelial cells in the mammary gland, and modulating energy intake [(Giesy et al., 2012), Figure 1].

Adiponectin enhances systemic insulin sensitivity and reduces lipolysis. Around parturition, its plasma content drops from 35 µg/mL during the dry period to <20 µg/mL immediately postpartum (Singh et al., 2014). Also, the expression of its receptors is reduced during the first three weeks postpartum (Saremi et al., 2014). Reflecting its anti-lipolytic effects, plasma adiponectin is negatively associated with circulating FFA (Kabara et al., 2014). Similar to adiponectin, plasma *leptin* peaks during the dry period (>6 ng/mL), and then its plasma concentration is reduced to <4 ng/mL by the first week after calving (Holtenius et al., 2003). Importantly, over-conditioned cows exhibit higher plasma leptin pre-calving than lean animals (Leon et al., 2004). This difference explains, in part, the more dramatic drop in dry matter intake and higher rates of lipolysis observed in cows with high body condition scores. Since leptin reduces appetite, its low postpartum levels promote the return of DMI to pre-calving levels.

In contrast to adiponectin and leptin, the synthesis of *resistin* increases during the periparturient period. Adipocytes and AT macrophages are the primary sources of this adipokine. Resistin promotes lipolysis by inhibiting insulin signaling and promoting inflammatory responses within the AT (Park et al., 2017). In dairy cows, plasma resistin concentrations rise from 45 ng/mL during the dry period to values above 75 ng/mL postpartum (Reverchon et al., 2014). Body condition score is positively associated with resistin production by AT macrophages (Reverchon et al., 2014). Therefore, over-conditioned cows will have higher circulating resistin compared to lean cows, making them more susceptible to excessive lipolysis and AT inflammation. Finally, *RBP4* is a potent inhibitor of adipocyte glucose uptake that also impairs the differentiation of preadipocytes into adipocytes (Romacho et al., 2014). Plasma

levels of this adipokine fall from >50 mg/mL one week before parturition to <30 mg/mL immediately after calving (Abd Eldaim et al., 2010). By inhibiting AT glucose utilization, RBP4 ensures energy prioritization to the mammary gland; however, impairing adipogenesis reduces the capacity of AT to buffer FFA excess predisposing to lipolysis dysregulation.

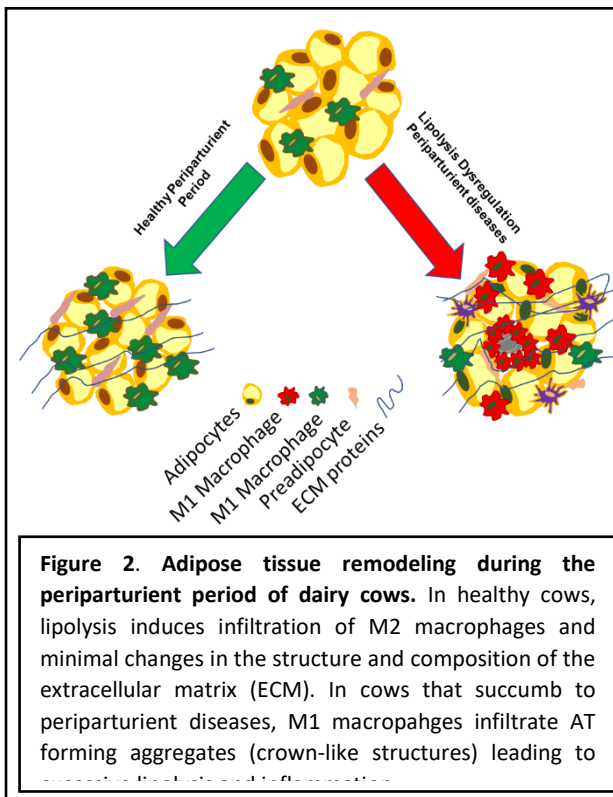
Adipose tissue remodeling

Lipolysis in AT induces a remodeling process within the organ characterized by a moderate inflammatory response with infiltration of immune cells, the proliferation of cells that are precursors of adipocytes, and production of lipid mediators of inflammation (Contreras et al., 2015, Contreras et al., 2017).

Macrophages are the primary immune cell infiltrating AT during lipolysis. The specific inflammatory phenotype of these mononuclear cells has been broadly classified in classical (M1), which have active pro-inflammatory responses, and alternative phenotype (M2), which promote inflammation resolution. The central role of M2 macrophages in AT is to remove products of lipolysis that can be toxic to the cell, such as FFA and triglycerides (Kosteli et al., 2010). For this reason, moderate infiltration of M2 macrophages into AT is beneficial for periparturient cows. During negative energy balance states, healthy dairy cows have a balanced mixture of M1 and M2 phenotype macrophages in AT (Contreras et al., 2016). When periparturient lipolysis is excessive and protracted, macrophages aggregate, forming

crowns-like structures and polarizing towards the M1 phenotype [Figure 2, (Contreras et al., 2015, Newman et al., 2019)]. These macrophages secrete potent cytokines such as TNF α and interleukin 6 that impair insulin signaling leading to a vicious circle where AT inflammation exacerbates lipolysis, aggravating AT inflammation. It is important to note that TNF α and interleukin-6 can activate lipolysis directly in adipocytes (Chirivi et al., 2021).

Regarding the proliferation of adipocyte precursors, this change is directly associated with the drastic changes in the volume of fat depots. During the first 40 days after calving, AT mass is reduced by 25-35% (Akter et al., 2011). Although not demonstrated in dairy cattle, rapid body weight loss induced by extended caloric restriction causes adipocyte death and the release of lipid remnants (Kosteli et al., 2010). As a response, preadipocytes proliferate to generate new adipocytes that replenish fat cell populations in a process termed adipogenesis. An adequate



adaptation to periparturient negative energy balance requires adipogenesis to support the buffering of FFA and other products of lipolysis that are toxic to cells.

Lipolysis induces the production of lipid mediators of inflammation in AT that are released into circulation. The activity of the lipases that break down triglycerides, such as hormone-sensitive lipase, releases linoleic, arachidonic, and other polyunsaturated fatty acids that are the substrate for prostaglandins and oxylipids (Contreras et al., 2020). The synthesis of these mediators of inflammation in the AT is probably one of the significant mechanisms sustaining the low-grade inflammation described by several authors in periparturient cows (Bradford and Swartz, 2020).

Adipose tissue dysregulation during periparturient diseases

The periparturient period is the lactation stage with the highest risk for metabolic and infectious diseases in dairy cows. Periparturient health events pose severe welfare issues and result in significant economic losses associated with decreased milk production, cost of treatment, and culling (USDA, 2015). To make things more complicated, periparturient illnesses often are presented as complexes of metabolic and inflammatory/infectious diseases (Probo et al., 2018). Two significant risk factors for increased disease susceptibility around parturition are lipolysis dysregulation (described above) and the dramatic increase in circulating endotoxins (e.g., Lipopolysaccharide (LPS) and lipoteichoic acids). Remarkably, common periparturient diseases such as mastitis, metritis, pneumonia, and metabolic events such as ruminal acidosis, heat stress, and parturition, often result in high circulating levels of LPS (Dickson et al., 2019). In humans and rodent models of disease, the inflammatory response to endotoxins, especially LPS, impairs the metabolic function of AT (Hersoug et al., 2018). In periparturient cows, experimental LPS exposure was associated with a higher incidence of displaced abomasum and placental retention and changes in metabolic parameters, including low plasma cholesterol and high β -hydroxybutyrate and FFA (Zebeli et al., 2011). The profile of these parameters indicates that endotoxemia possibly induces the development of lipolysis dysregulation in bovine AT.

The possible mechanisms by which the endotoxemia associated with multiple periparturient diseases triggers AT dysfunction are twofold. First, endotoxins activate lipolysis in dairy cows by three mechanisms (Chirivi et al., 2021): 1) binding to TLR4 increases the levels of intracellular cAMP through a calcium-dependent pathway (Song et al., 2007, Moon et al., 2011), leading to the activation of hormone-sensitive lipase. 2) TLR4 binding to LPS stimulates the activation of NF- κ B that triggers the synthesis of pro-inflammatory cytokines, including TNF α (Lu et al., 2008). The latter promotes lipolysis by impairing the expression/function of perilipin, causing the thinning of the protein envelop of the lipid droplet and making it more susceptible to the action of hormone-sensitive lipase (Laurencikiene et al., 2007). 3) The activation of the mitogen-activated protein kinase /extracellular signal-regulated kinase (MAPK and ERK1/2). This pathway activates beta-adrenergic receptors that ultimately trigger the lipolytic activity of hormone-sensitive lipase (Zu et al., 2009, Hong et al., 2018). In contrast to bovines, in rodent adipocytes, LPS activates lipolysis preferentially by ERK1/2, as these species are resistant to NF κ B triggered lipolysis (Zu et al., 2009, Bergan et al., 2013, Chi et al., 2014). In periparturient

dairy cows, lipolytic responses in AT are increased upon LPS exposure indicating that endotoxemia can potentiate AT responses to common stimulants of postpartum lipolysis such as catecholamines.

The second mechanism by which endotoxins may induce AT dysfunction is by altering the inflammatory phenotype of AT macrophages. Endotoxins promote macrophage M1 polarization; therefore, exposure to these bacterial byproducts early in the periparturient period may predispose cows to excessive lipolytic response during NEB postpartum. However, endotoxin-driven M1 polarization in AT may be affected by the degree of adiposity and by the development of endotoxin tolerance (Komegae et al., 2019). Therefore, future studies need to evaluate the effects of endotoxins on the phenotype of AT immune cells and its impact on metabolic function during the periparturient period.

Conclusion

Our knowledge of AT biology in periparturient dairy cows has advanced dramatically since the 1990s. However, there are still gaps in our understanding of the changes that occur during the periparturient period in AT. The role of AT remodeling on the homeorhetic adaptation to lactation, including the responses of AT to infectious and inflammatory diseases is unclear. Also, the impact of the anatomical differences on the immunobiology of AT depots and the endocrine function of fat tissues is unknown. Filling these gaps will support the development of preventive and corrective nutritional or pharmacological interventions to maintain an effective periparturient AT function.

REFERENCES

- Abd Eldaim, M. A., A. Kamikawa, M. M. Soliman, M. M. Ahmed, Y. Okamoto-Ogura, A. Terao, T. Miyamoto, and K. Kimura. 2010. Retinol binding protein 4 in dairy cows: its presence in colostrum and alteration in plasma during fasting, inflammation, and the peripartum period. *The Journal of dairy research* 77(1):27-32.
- Akter, S. H., S. Haussler, S. Danicke, U. Muller, D. von Soosten, J. Rehage, and H. Sauerwein. 2011. Physiological and conjugated linoleic acid-induced changes of adipocyte size in different fat depots of dairy cows during early lactation. *J Dairy Sci* 94(6):2871-2882.
- Bergan, H. E., J. D. Kittilson, and M. A. Sheridan. 2013. PKC and ERK mediate GH-stimulated lipolysis. *Journal of molecular endocrinology* 51(2):213-224.
- Bradford, B. J. and T. H. Swartz. 2020. Review: Following the smoke signals: inflammatory signaling in metabolic homeostasis and homeorhesis in dairy cattle. *Animal* 14(S1):s144-s154.
- Chi, W., D. Dao, T. C. Lau, B. D. Henriksbo, J. F. Cavallari, K. P. Foley, and J. D. Schertzer. 2014. Bacterial peptidoglycan stimulates adipocyte lipolysis via NOD1. *PloS one* 9(5):e97675.
- Chirivi, M., C. J. Rendon, M. N. Myers, C. M. Prom, S. Roy, A. Sen, A. L. Lock, and G. A. Contreras. 2021. Lipopolysaccharide induces lipolysis and insulin resistance in adipose tissue from dairy cows. *J Dairy Sci*.
- Contreras, G. A., J. De Koster, J. de Souza, J. Laguna, V. Mavangira, R. K. Nelli, J. Gandy, A. L. Lock, and L. M. Sordillo. 2020. Lipolysis modulates the biosynthesis of inflammatory lipid mediators derived from linoleic acid in adipose tissue of periparturient dairy cows. *J Dairy Sci* 103(2):1944-1955.

- Contreras, G. A., E. Kabara, J. Brester, L. Neuder, and M. Kiupel. 2015. Macrophage infiltration in the omental and subcutaneous adipose tissues of dairy cows with displaced abomasum. *J Dairy Sci* 98(9):6176-6187.
- Contreras, G. A., C. Strieder-Barboza, and J. De Koster. 2018. Symposium review: Modulating adipose tissue lipolysis and remodeling to improve immune function during the transition period and early lactation of dairy cows. *J Dairy Sci* 101(3):2737-2752.
- Contreras, G. A., C. Strieder-Barboza, and W. Raphael. 2017. Adipose tissue lipolysis and remodeling during the transition period of dairy cows. *Journal of animal science and biotechnology* 8:41.
- Contreras, G. A., K. Thelen, S. Schmidt, C. Strieder-Barboza, C. Preseault, R. Raphael, M. Kiupel, J. Caron, and A. Lock. 2016. Adipose tissue remodeling in late-lactation dairy cows during feed restriction-induced negative energy balance. *J Dairy Sci* 99.
- De Koster, J., W. Van den Broeck, L. Hulpio, E. Claeys, M. Van Eetvelde, K. Hermans, M. Hostens, V. Fievez, and G. Opsomer. 2016. Influence of adipocyte size and adipose depot on the in vitro lipolytic activity and insulin sensitivity of adipose tissue in dairy cows at the end of the dry period. *J Dairy Sci* 99(3):2319-2328.
- Dickson, M. J., S. K. Kvidera, E. A. Horst, C. E. Wiley, E. J. Mayorga, J. Ydstie, G. A. Perry, L. H. Baumgard, and A. F. Keating. 2019. Impacts of chronic and increasing lipopolysaccharide exposure on production and reproductive parameters in lactating Holstein dairy cows. *Journal of Dairy Science*.
- Giesy, S. L., B. Yoon, W. B. Currie, J. W. Kim, and Y. R. Boisclair. 2012. Adiponectin deficit during the precarious glucose economy of early lactation in dairy cows. *Endocrinology* 153(12):5834-5844.
- Hersoug, L.-G., P. Møller, and S. Loft. 2018. Role of microbiota-derived lipopolysaccharide in adipose tissue inflammation, adipocyte size and pyroptosis during obesity. *Nutr Res Rev* 31(2):153-163.
- Holtenius, K., S. Agenas, C. Delavaud, and Y. Chilliard. 2003. Effects of feeding intensity during the dry period. 2. Metabolic and hormonal responses. *J Dairy Sci* 86(3):883-891.
- Hong, S., W. Song, P. H. Zushin, B. Liu, M. P. Jedrychowski, A. I. Mina, Z. Deng, D. Cabarkapa, J. A. Hall, C. J. Palmer, H. Aliakbarian, J. Szpyt, S. P. Gygi, A. Tavakkoli, L. Lynch, N. Perrimon, and A. S. Banks. 2018. Phosphorylation of Beta-3 adrenergic receptor at serine 247 by ERK MAP kinase drives lipolysis in obese adipocytes. *Mol Metab* 12:25-38.
- Kabara, E., L. M. Sordillo, S. Holcombe, and G. A. Contreras. 2014. Adiponectin links adipose tissue function and monocyte inflammatory responses during bovine metabolic stress. *Comp Immunol Microbiol Infect Dis* 37(1):49-58.
- Komegae, E. N., M. T. Fonseca, S. da Silveira Cruz-Machado, W. M. Turato, L. R. Filgueiras, R. P. Markus, and A. A. Steiner. 2019. Site-Specific Reprogramming of Macrophage Responsiveness to Bacterial Lipopolysaccharide in Obesity. *Frontiers in immunology* 10(1496):1496.
- Kosteli, A., E. Soguru, G. Haemmerle, J. F. Martin, J. Lei, R. Zechner, and A. W. Ferrante, Jr. 2010. Weight loss and lipolysis promote a dynamic immune response in murine adipose tissue. *J. Clin. Invest.* 120(10):3466-3479.
- Lafontan, M. and D. Langin. 2009. Lipolysis and lipid mobilization in human adipose tissue. *Progress in lipid research* 48(5):275-297.

- Laurencikiene, J., V. van Harmelen, E. Arvidsson Nordstrom, A. Dicker, L. Blomqvist, E. Naslund, D. Langin, P. Arner, and M. Ryden. 2007. NF-kappaB is important for TNF-alpha-induced lipolysis in human adipocytes. *Journal of lipid research* 48(5):1069-1077.
- Leon, H. V., J. Hernandez-Ceron, D. H. Keislert, and C. G. Gutierrez. 2004. Plasma concentrations of leptin, insulin-like growth factor-I, and insulin in relation to changes in body condition score in heifers. *Journal of animal science* 82(2):445-451.
- Lu, Y.-C., W.-C. Yeh, and P. S. Ohashi. 2008. LPS/TLR4 signal transduction pathway. *Cytokine* 42(2):145-151.
- Moon, E. Y., Y. S. Lee, W. S. Choi, and M. H. Lee. 2011. Toll-like receptor 4-mediated cAMP production up-regulates B-cell activating factor expression in Raw264.7 macrophages. *Experimental cell research* 317(17):2447-2455.
- Newman, A. W., A. Miller, F. A. Leal Yepes, E. Bitsko, D. Nydam, and S. Mann. 2019. The effect of the transition period and postpartum body weight loss on macrophage infiltrates in bovine subcutaneous adipose tissue. *J Dairy Sci* 102(2):1693-1701.
- Park, H. K., M. K. Kwak, H. J. Kim, and R. S. Ahima. 2017. Linking resistin, inflammation, and cardiometabolic diseases. *Korean J Intern Med* 32(2):239-247.
- Probo, M., O. B. Pascottini, S. LeBlanc, G. Opsomer, and M. Hostens. 2018. Association between metabolic diseases and the culling risk of high-yielding dairy cows in a transition management facility using survival and decision tree analysis. *J Dairy Sci* 101(10):9419-9429.
- Reverchon, M., C. Ramé, J. Cognié, E. Briant, S. Elis, D. Guillaume, and J. Dupont. 2014. Resistin in dairy cows: plasma concentrations during early lactation, expression and potential role in adipose tissue. *PloS one* 9(3):e93198-e93198.
- Romacho, T., M. Elsen, D. Rohrborn, and J. Eckel. 2014. Adipose tissue and its role in organ crosstalk. *Acta physiologica (Oxford, England)* 210(4):733-753.
- Saremi, B., S. Winand, P. Friedrichs, A. Kinoshita, J. Rehage, S. Dänicke, S. Häussler, G. Breves, M. Mielenz, and H. Sauerwein. 2014. Longitudinal profiling of the tissue-specific expression of genes related with insulin sensitivity in dairy cows during lactation focusing on different fat depots. *PloS one* 9(1):e86211-e86211.
- Singh, S. P., S. Haussler, J. J. Gross, F. J. Schwarz, R. M. Bruckmaier, and H. Sauerwein. 2014. Short communication: circulating and milk adiponectin change differently during energy deficiency at different stages of lactation in dairy cows. *J Dairy Sci* 97(3):1535-1542.
- Song, J., M. J. Duncan, G. Li, C. Chan, R. Grady, A. Stapleton, and S. N. Abraham. 2007. A novel TLR4-mediated signaling pathway leading to IL-6 responses in human bladder epithelial cells. *PLoS pathogens* 3(4):e60.
- USDA, A. 2015. NAHMS Dairy 2014. U. S. D. o. Agriculture, ed. APHIS, APHIS.
- Zebeli, Q., S. Sivaraman, S. M. Dunn, and B. N. Ametaj. 2011. Intermittent parenteral administration of endotoxin triggers metabolic and immunological alterations typically associated with displaced abomasum and retained placenta in periparturient dairy cows. *Journal of Dairy Science* 94(10):4968-4983.
- Zu, L., J. He, H. Jiang, C. Xu, S. Pu, and G. Xu. 2009. Bacterial endotoxin stimulates adipose lipolysis via toll-like receptor 4 and extracellular signal-regulated kinase pathway. *The Journal of biological chemistry* 284(9):5915-5926.

Managing extensive winter grazing systems in arid/semi-arid environments

H.A. (Bart) Lardner

University of Saskatchewan, Saskatoon, Saskatchewan

Email: bart.lardner@usask.ca

Introduction

The most expensive aspect of a beef cow operation is the wintering of dry pregnant cows. Reducing the feeding of conserved forage while maintaining or increasing cow performance utilizing alternative forage systems could lower overall costs of production (Volesky et al. 2002). Greater reliance on the cow rather than equipment for forage harvesting is one method for reducing feed costs (D'Souza et al. 1990). The cost of wintering beef cows in the prairie region of Canada and United States is the single largest cost of beef production, accounting for 60-65% of the total cost of production in a cow-calf operation (Larson 2008). Providing wintering beef cows enough feed to meet their nutrient requirements while avoiding waste resulting from over-feeding provides a means of controlling and reducing these costs.

Extensive grazing systems to be discussed include stockpile grazing perennials (Hitz and Russell, 1998; Meyer et al., 2009; Kulathunga et al., 2018), swath grazing annuals (Kelln et al., 2011; Kumar et al. 2012), and grazing cereal crop residues (McCartney et al., 2006; Van De Kerckhove et al., 2011; Krause et al., 2013). Bale grazing (Kelln et al., 2011; Lardner et al., 2018), and grazing whole plant corn (Lardner, 2012; Jose et al. 2020) will also be discussed. Using cool or warm season annuals for in-field grazing may allow producers to reduce winter feeding costs, while animal activity and deposition of manure nutrients (Jungnitsch et al. 2011) directly on the land may be beneficial to soils and subsequent crop production (Kelln et al. 2012). There has been considerable research conducted on nutrient management associated with extensive winter grazing systems (Schoenau and Davis, 2006; Jungnitsch et al., 2011; Smith et al., 2011). Finally, extensive grazing systems can decrease costs for harvesting, transportation, labor, yardage and manure removal relative to the conventional drylot system (Nayigihugu et al., 2007).

Annual Forages

Several annual forage crops, including barley (*Hordeum vulgare* L.), oat (*Avena sativa* L.), golden German foxtail millet (*Setaria italica*), kale (*Brassica oleracea*), turnip (*Brassica rapa*) and corn (*Zea mays*) have shown promise in cow-calf grazing systems (Lardner, 2003; McCartney et al. 2008). However, the economic use of these crops should be compared to other annual cereals (May et al. 2007; McCartney et al. 2009). In warm and moister regions, corn is traditionally grown as either a grain crop or silage. The remaining stover or crop residue is usually grazed during late fall or winter with weaned beef calves or dry pregnant beef cows (Klopfenstein et al. 1987; Poland et al. 2003). Recently in western Canada and some northwest US states, there has been interest in grazing standing whole plant corn to avoid the costs of conventional harvesting

and storage. However, corn production has been limited to areas receiving a minimum of 2000-2400 corn heat units (Aasen and Bjorge 2009).

Stockpile Grazing

Stockpiling forage is the practice of accumulating forage biomass during summer and fall and grazing it after the growing season (Hitz and Russel, 1998; Riesterer et al., 2000). Grazing stockpiled perennial forages can be an excellent alternative to more expensive hay or silage feeding in drylot pens. However, stockpiled forages are usually mature and due to leaf senescence, can be moderate to poor in nutritive value. Yet, stockpiled forage can meet dry cow nutrient requirements in early to mid-gestation (Table 1) when requirements are less compared to lactating cows (Poore and Drewnoski, 2010; Kulathunga et al., 2016). Stockpiled forage can be grazed from October to early December, or until weather and snow conditions prevent grazing, or can be used in early spring, before new pasture growth (Kulathunga et al., 2016). Stockpiling perennial forage species for fall and winter grazing has been shown to be a cost effective alternative to traditional drylot feeding (Baron et al., 2005). Costs are reduced through the minimization of harvesting, hauling, feeding and manure removal (Kulathunga et al., 2016). Labor can be reduced by 25 percent in comparison to conventional wintering of beef cows (Riesterer et al. 2000). The efficacy of a stockpile system depends on species selection, accumulation period, soil nutrition management and weather (Baron 2004). Depending on the forage quality, it can be grazed any time after pasture ceases to be productive, usually in September, and graze well into December, possibly longer if weather conditions such as ice and snow do not prevent grazing (Riesterer et al. 2000), with no effect on cow condition (Table 2).

Swath Grazing

Swath grazing is a method of extending the grazing season, where an annual cereal crop is swathed at a defined stage, left in windrows in field for grazing (Aasen et al., 2004; Kelln et al., 2011). For swath grazing of annual cereals, the producer needs to balance yield with the potential weathering (May et al., 2007). In a study by May et al. (2007), later seeding dates resulted in higher quality forage, although the yield was reduced. Suggestion is that ideally oat and barley be seeded from May 20-25, to optimize utilization of soil moisture and cool temperatures (May et al., 2007). If swaths are large enough, cattle can access the feed through up to 45 cm (1.5 ft) of snow. The cows then graze the swaths in fall and winter and sometimes in the following spring (Aasen et al., 2004). These swathed annuals generally meet the nutritional requirements of the cow in mid-gestation (NASEM, 2016) when the temperature is in the thermo-neutral zone (Aasen et al. 2004). Access to the swaths should be controlled with portable electric fence, allocating 3 to 4 day supply of forage (Karn et al. 2005). McCartney et al. (2004) explains that pregnant beef cows can be managed using swath grazing and can result in savings of 50% through decreasing or eliminating the expenses of harvesting, hauling and feeding the forage as well as reduced manure removal costs. Kumar et al. (2012) reported that backgrounding calves on quality swathed barley or millet forage (Table 3) in field paddocks did not adversely affect performance compared with backgrounding calves in a traditional DL pen system (Table 4).

Bale Grazing

Bale grazing is another method to extend the grazing season and optimize nutrient management. Smith et al. (2011) describes bale grazing as a system to optimize the benefits of manure nutrients, by placing round bales on a field site and grazing at a higher stocking density. Bale grazing systems can be managed either as intensive where baled forage is hauled out to the bale grazing site and placed in a grid pattern or more extensively where bales are left where they are ejected from the baler (SMA 2008). With bale grazing there is a need to restrict forage access, using portable electric fence, and 3 to 4 d allocation of forage made available, which reduces wastage and facilitates manure deposition throughout the field (Lardner, 2018). Management of the site is required, as Kelln et al. (2011) explains that bale grazing has the least uniform distribution of manure nutrients of the winter grazing systems.

Cattle have poor N retention and most of the N is excreted in the feces and urine (Kelln et al. 2012). Erickson and Klopfenstein (2001) reported that feedlot yearlings retained only approximately 10 percent N and excreted the remaining 90 percent. Another advantage of cattle directly depositing manure in the field is that manure that is deposited in a drylot feeding pen can be subjected to nutrient losses due to volatilization, making it less valuable (Kelln et al. 2012). Nutrient benefits are accessible through this system as bale grazing at a density of 63 bales/ha (25 bales/acre) can equate to about 34 kg of N available to the plant in the following season (SMA 2008). Jungnitsch et al. (2011) reported significant improvement in soil fertility and greater pasture growth where manure and urine were deposited during winter in-field bale grazing.

Highlights from Jungnitsch et al. (2011) reported soil inorganic N amounts, measured in spring following winter grazing, were 3 times greater on bale graze sites compared to unfertilized sites. Forage DM yields were 3 to 4 fold greater on winter feeding sites compared to unfertilized sites. Recovery of N and P in pasture forage was approximately 30-40% of original feed N and 20-30% of original feed P on beef cattle winter feeding sites. Finally, recovery of N and P in pasture forage was only 1% of original feed N and 3% of original feed P from pen manure applied sites. In addition, Lardner et al. (2018) backgrounded weaned steers on supplemented bale grazing systems, showing an alternative to drylot backgrounding (Table 5). Kelln et al. (2011) reported costs averaged 10% lower for bale grazing compared to drylot feeding over a 3-year study. With a reduction in cost and a reduction of labor associated with overwintering cows, bale grazing is a viable alternative to drylot pen feeding.

Grazing Crop Residues

There has been renewed interest in the use of crop residues in beef-cow diets because of their potential to reduce winter feed costs (Krause et al. 2013). Cereal crop residues such as barley, wheat, oat and triticale grown in the western prairies are potential sources of feed for overwintering beef cows (McCartney et al. 2006). Costs can be reduced by leaving crop residues in the field and having cows graze it (McCartney et al. 2006). Cereal chaff consists of smaller particles than straw and includes glumes, hulls, seed heads, short straw, leaf materials, weed

seeds, and whole or cracked kernels that were separated from harvest grain (McCartney et al. 2006; (Figure 1). These fractions vary in palatability and digestibility depending on the crop variety and the time at harvest, harvest method and weathering of the residue (Van De Kerckhove et al. 2011).

Because of the low protein and high fiber content of cereal crop residues, a study by Krause et al. (2013) compared the effects of grazing either oat or pea residues versus drylot pen-feeding grass-legume hay on cow performance, reproductive efficiency, estimated dry matter intake (DMI), and winter system costs. The CP level of the pea residue was higher than oat residue and the pea residue had similar CP and TDN as the mixed hay (Krause et al. 2013). But despite this, the cows consuming the pea crop residue had lower DM intake and reduced nutrient intake and found that this was likely due to the lower palatability of the crop (Krause et al. 2013). Cows grazing pea residues for 63 days had lower body weight change than cows grazing oat residues or drylot hay fed cows. On average, total costs for the oat and pea residue grazing strategies were \$0.77 and \$0.59 cow/d less than drylot (\$2.13 cow/d), respectively. Grazing crop residue for part of a cow's winter feeding program has cost advantages over pen feeding hay; however, environmental conditions (snowfall, temperature) dictate forage accessibility.

In the northern Great Plains, wintering cows on cornstalk residue is a common practice. With the adaptation of low heat unit corn varieties there is great potential to graze corn residues in beef cow wintering systems. Fernandez-Rivera and Klopfenstein (1989) demonstrated cornstalk residue is of adequate quality for growing cattle immediately following harvest. The nutrient profile of cornstalks is well established, with crude protein (CP) levels reported to be from 3.3 to 5.5%, which does not meet the requirements for a gestating cow or heifer (NASEM, 2016). Protein supplementation may be required to increase intake and digestibility of low-quality forages during winter (DelCurto et al., 1990; Bowman and Sanson 2000). Research in Nebraska reported that although cornstalk residue is typically low in CP, the relatively low CP requirement of early gestation beef cows may be met due to selective grazing of crop residue components, provided the cow has the ability to selectively graze (Warner et al. 2011).

Grazing corn residues also offer an opportunity to lower feed costs and extend the grazing season (Wilson et al. 2004). Although, the main concern when grazing corn residues is that protein content and energy digestibility are low because the plant is harvested at late maturity (Klopfenstein et al. 1987). Cows grazing corn residues may need to be provided a supplement earlier than cows grazing stockpile forages. Digestibility of the diet is high initially, but declines with time due to selection of the more digestible parts early (Wilson et al. 2004). Access to the corn residue should be controlled to minimize wastage and improve utilization (McGeough et al. 2018).

Grazing Whole Plant Corn

Grazing standing whole plant corn is a management system that makes sense to many western US and Canadian cow-calf producers, to extend the grazing season and reduce feed costs per cow per day. However, the equipment, seed, fertilizer cost, and unfamiliarity with growing corn

for grazing often deters producers from trying it themselves. Early grazing corn research in western Canada, evaluated several corn varieties for beef cows (Lardner, 2002) and backgrounding programs for weaned beef calves (Lardner, 2003a). Corn should be seeded early as with an early frost, there is an appropriate amount of leaf and grain on the plants to optimize cow nutrition (May et al. 2007). In central Saskatchewan, corn grazing studies showed that early maturing varieties provided excellent late-season grazing either grazed in a swath or as standing crop during the winter (Lardner 2003a; Jose 2020). Strip grazing is highly recommended when grazing the field with allocation of enough grazing corn for a 3 to 4 day supply. By limiting the grazing area, animals are forced to consume both high- [cobs] and low-quality [stalk, husk, leaves] structures of the corn plant (Lardner et al. 2012). Ensure a balanced mineral program is provided and a good supply of high quality drinking water is also available to the grazing animals.

There are several concerns when managing grazing corn with beef cows. Excessive cob intake may lead to digestive disturbances such as acidosis and founder due to potential grain overload. Adapting cows to grain supplementation for 7 to 10 days before turning into cornfields can minimize this concern. Recent work by Jose et al. (2020) is evaluating ruminal pH of ruminally cannulated heifers fitted with indwelling ruminal pH probes. Cows were field grazing either whole plant corn or swathed whole plant barley or drylot fed barley hay in pens in a 3x3 Latin Square design. Forage was allocated on a 3 d basis and pH values were summarized. Data suggests that in yr 1, beef cows grazing barley swaths faced maximum acidic challenge compared to cows grazing standing corn or fed barley hay. However, in yr 2, SARA conditions were observed for cows grazing whole plant corn (Jose et al. 2020).

Additional strategies to transition animals to grazing corn include supplying extra roughage in the form of supplemented hay/forage bales, or limiting the daily cornfield grazing time and ensuring cows are full prior to accessing the crop. It will take 7 to 10 days for the rumen to adjust to the new diet. Another issue can be nitrate toxicity; however the highest level of nitrate concentration in the plant is the lowest part of the stalk. This plant structure is typically consumed last by the grazing animal; therefore the potential for nitrate issues is unlikely. Finally, animals should be monitored daily to evaluate body condition and remaining crop material and managed for 90-95 percent utilization of available forage.

A recent study was conducted in east central Saskatchewan to evaluate several corn varieties for extended grazing with beef cows (Lardner et al. 2012). Five different corn varieties were seeded with a corn planter June 1, at 65,500 seeds/ha (26,200 seeds/acre) with a row spacing of 750 mm (30 inches) and depth of 37 mm (1.5 inches). The field was sprayed with glyphosate 11 June at 3.8 L/ha (1.5 L/ac). Corn varieties included five varieties, ranging in crop heat units of 2050 to 2250. Total CHU's at the site from 1 April to 31 October 2011 were 2417 CHU. Dry matter yield in September 2011 ranged from 10.8 to 11.8 tonne/ha (4.1 to 5.7 ton/acre) (Table 7).

Forage quality in corn will vary according to cultivar and seeding date with early-maturing cultivars having higher CP (11 to 12 %) than later maturing cultivars. May et al. (2007) noted in

their study that corn was marginal in meeting the CP requirements of third trimester pregnant beef cows. Energy and protein requirements for a 680 kg (1500 lb) pregnant beef cow in second trimester are 7.8% CP and 50% TDN (NASEM 2016). Corn quality was determined at two different times, in September at the end of the growing season and again in November, coinciding with the start of grazing with beef cows (Lardner et al. 2012). September samples included submission of whole plant, leaf and grain+cob from each variety and November samples were only whole plant. Crude protein content of the whole plant for all varieties ranged from 6.4 to 8.1 percent (Table 8). Corn leaf CP levels ranged from 7.4% for P7443R to 13.6% for HLSR06. Grain+cob CP levels ranged from 10.9% for DKC2754 to 12.9% for HLSR06 (Table 4). Total digestible nutrient (TDN) content of whole plant for all varieties ranged from 68.6 to 70.8 percent. Corn leaf TDN levels ranged from 49.7% for P7443R to 60.6% for DKC2754. Grain+cob TDN levels ranged from 89.3% for P7443R to 90.8% for P7213R (Table 4). At start of grazing in November, CP levels ranged from 6.7 to 9.7%, while TDN levels ranges from 57.1 to 66.5 percent (Table 8). Overall, energy levels of most corn varieties would meet nutrient requirements of grazing dry, pregnant beef cows, however CP may be limiting for late gestation cows, suggesting the need for supplementation.

Producers are encouraged to calculate costs according to their own individual situation. The cost per cow per day is calculated by dividing the crop production costs per acre by the grazing days per acre. Crop production costs should be calculated for each variety and compared to alternative grazing systems. Lardner (2012) reported total crop expenses ranged from \$205 to \$223/acre (Table 5). In addition, \$/cow/day ranged from \$0.70 to \$1.42/day and averaged \$0.94/day (Table 5). It is important to note that costs will vary from operation to operation.

Conclusion

With the need for beef producers to find alternative methods for managing cattle in economically challenging times, extensive systems appear to be valuable options in terms of improved economics and nutrient management. Through the reduction in feed costs and returns from manure excretion directly in the field, winter management of beef cattle can be more efficient. However, caution should be observed when choosing the system that best fits an individual's beef cattle operation.

This type of extensive grazing strategy demands a well-managed program, starting with forage crop choice and continuing with close monitoring of animals during the grazing period. For more on extensive grazing systems, several videos are available at:

<https://www.youtube.com/user/WSTRNBEEF/videos>.

References

- Aasen, A., Baron, V. S., Clayton, G. W., Dick, A. C., and McCartney, D. H. 2004. Swath grazing potential of spring cereals, field pea and mixtures with other species. *Can. J. Plant Sci.* 84: 1051-1058.
- Aasen, A. and Bjorge, M. 2009. Alberta forage manual. Alberta Agriculture and Rural Development, Edmonton, AB. pp 348.
- Baron, V. S., Dick, A. C., Bjorge, M., and Lastiwka, G. 2004. Stockpiling potential of perennial forage species adapted to the Canadian western prairie parkland. *Agron. J.* 96: 1545-1552.
- Baron, V. S., Dick, A. C., Bjorge, M., and Lastiwka, G. 2005. Accumulation period for stockpiling perennial forages in the western Canadian prairie parkland. *Agron. J.* 97: 1508-1514.
- Bowman, J. P. and Sanson, D.W. 2000. Energy/protein supplementation considerations for grazing ruminants. Oregon State University Station Bulletin SB 683. Strategic supplementation of beef cattle consuming low-quality roughages in the western United States. Oregon State University Agriculture Experiment Station.
- DelCurto, T., Cochran, R.C., Corah, L.R., Beharka, A.A., Vanzant, E.S. and Johnson, D.E. 1990. Supplementation of dormant tallgrass-prairie forage: II. Performance and forage utilization characteristics in grazing beef cattle receiving supplements of different protein concentrations. *J. Anim. Sci.* 68: 532-544.
- D'Souza, G.E., Marshall, E.W., Bryan, W.B. and Prigge, E.C. 1990. Economics of extended grazing systems. *Amer. J. Alternative Agr.* 5: 120-125.
- Fernandez-Rivera, S. and Klopfenstein, T.J. 1989. Diet composition and daily gain of growing cattle grazing dryland and irrigated cornstalks at different stocking rates. *J. Anim. Sci.* 67:590-596.
- Hitz, A. C. and Russell, J. R. 1998. Potential of stockpiled perennial forages in winter grazing systems for pregnant beef cows. *J. Anim. Sci.* 76: 404-415.
- Jose D, Larson, K, McKinnon JJ, Penner GB, Damiran D & Lardner HA. 2020. Effect of winter-feeding system on beef cow performance, ruminal fermentation, and system costs. *App. Anim. Sci.* Vol 36: 731-744.
- Jungnitsch, P. F., Schoenau, J. J., Lardner, H. A., and Jefferson, P.G. 2011. Winter feeding beef cattle on the western Canadian prairies: Impacts on soil nitrogen and phosphorus cycling and forage growth. *Agric. Ecosyst. Environ.* 141: 143-152.
- Karn, J.F., Tanaka, D.L., Liebigh, M.A., Ries, R.E., Kronberg, S.L. and Hanson, J.D. 2005. An integrated approach to crop/livestock systems: Wintering beef cows on swathed crops. *Renew. Agric. Food Syst.* 20: 232-240.
- Kelln, B.M., Lardner, H.A., McKinnon, J.J., Campbell, J.R., Larson, K. and Damiran, D. 2011. Effect of winter feeding system on beef cow performance, reproductive efficiency and system cost. *Prof. Anim. Sci.* 27: 410-421.
- Kelln B.M., Lardner H.A., Schoenau, J.J. and King, T. 2012. Effects of beef cow winter feeding systems, pen manure and compost on soil nitrogen and phosphorous amounts and distribution, soil density and crop biomass. *Nut. Cycling Agro. Ecosys.* 92: 183-194.
- Klopfenstein, T., Roth, L., Fernandez-Rivera, S. and Lewis, M. 1987. Corn residues in beef production systems. *J. Anim. Sci.* 65: 1139-1148.
- Krause, A.D., Lardner, H.A., Mckinnon, J.J., Hendrick, S., Larson, K., and Damiran, D. 2013.

- Comparison of grazing oat and pea crop residue versus feeding grass-legume hay on beef-cow performance, reproductive efficiency, and system cost. *Prof. Anim. Sci.* 29: 535–545.
- Kulathunga, D.G.R.S., Penner, G.B., Schoenau, J.J., Damiran, D., Larson, K. and Lardner H.A. 2016. Effect of perennial forage system on forage characteristics, soil nutrients, cow performance and system economics. *Prof. Anim. Sci.* 32: 784-797.
- Kumar, R., Lardner, H.A., Christensen, D.A., McKinnon, J.J., Damiran, D. and Larson, K. 2012. Comparison of alternative backgrounding systems on beef calf performance, feedlot finishing performance, carcass traits and system cost of gain. *Prof. Anim. Sci.* 28: 541-551.
- Lardner, H. A. 2002. Comparison of grazing corn varieties. Western Beef Development Centre Fact Sheet #2002-02. Western Beef Development Centre, Lanigan, SK. 4pp.
- Lardner, H. A. 2003a. Backgrounding calves on swathed corn and barley. Western Beef Development Centre Fact Sheet #2003-02. Western Beef Development Centre, Lanigan, SK. 4pp.
- Lardner, H. A. 2003b. Extending the grazing season with turnips. Western Beef Development Centre Fact Sheet #2003-03. Western Beef Development Centre, Lanigan, SK. 4pp.
- Lardner, H.A., Larson, K. and Pearce, L. 2012. Winter grazing beef cows on standing corn. Western Beef Development Centre Fact Sheet. #2012-03. Western Beef Development Centre, Lanigan, SK. 4pp.
- Lardner, H.A., Pearce, L., and Damiran, D. 2017. Evaluation of low heat unit corn hybrids compared to barley for forage yield and quality on the Canadian Prairies. *Sustain. Agric. Res.* 6: 90.
- Lardner HA, Larson K, Christensen DA, McKinnon JJ, Añez-Osuna F, Damiran D, Simili da Silva M and Clark L. 2018. Performance of stocker cattle winter bale grazing when supplemented with either wheat-based DDGS or barley grain in western Canada. *Prof. Anim. Sci.* 34: 460-468.
- Larson, K. 2010. 2008 Saskatchewan cow-calf cost of production analysis. WBDC Fact Sheet #2010-01. Western Beef Development Centre, Lanigan, SK, Canada.
- May, W.E., Klein, L.H., Lafond, G.P., McConnell, J.T. and Phelps, S.M. 2007. The suitability of cool- and warm-season annual cereal species for winter grazing in Saskatchewan. *Can. J. Plant Sci.* 87: 739–752.
- McCartney, D., Okine, E. K., Baron, V. S., and Depalme, A. J. 2004. Alternative fall and winter feeding systems for spring calving beef cows. *Can. J. Anim. Sci.* 84: 511-522.
- McCartney, D. H., Block, H. C., Dubeski, P. L. and Ohama, A. J. 2006. The composition and availability of straw and chaff from small grain cereals for beef cattle in western Canada. *Can. J. Anim. Sci.* 86: 443-455.
- McCartney, D., Fraser, J. and Ohama, A. J. 2008. Annual cool season crops for grazing by beef cattle. A Canadian review. *Can. J. Anim. Sci.* 88: 517-533.
- McCartney, D., Fraser, J. and Ohama, A. J. 2009. Potential of warm-season annual forages and *Brassica* crops for grazing: A Canadian Review. *Can. J. Anim. Sci.* 89: 431-440.
- McGeough, E. J., D. J. Cattani, Z. Koscielny, B. Hewitt, and K. H. Ominski. 2018. Annual and perennial forages for fall/winter grazing in western Canada. *Can. J. Plant Sci.* 98:247-254.
- Meyer, A. M., Kerley M.S., Kallenbach, R.L. and Perkins, T.L. 2009. Comparison of grazing stockpiled tall fescue versus feeding hay with or without supplementation for gestating and lactating beef cows during winter. *Prof. Anim. Sci.* 25: 449-458.

- National Academies of Sciences, Engineering, and Medicine (NASEM). 2016. Nutrient requirement of beef cattle. Eighth revised edition. The National Academics Press, Washington, DC.
- Nayigihugu, V., Schleicher, A.D., Koch, D.W., Held, L.J., Flake, J.W. and Hess, B.W. 2007. Beef cattle production, nutritional quality, and economics of windrowed forage vs. baled hay during winter. *Agron. J.* 99: 944-951.
- Poland, W., Carr, P., Nelson, J., Sedivec, K., Hyren, P., Manske, L. and Miller, W. 2003. Making extended grazing work in North Dakota. In NCRA 225. Improved grazing systems for animal production. North Dakota State University, Dickinson Research Extension Center, Dickenson, ND.
- Poore, M. H., and M. E. Drewnoski. 2010. Review: Utilization of stockpiled tall fescue in winter grazing systems for beef cattle. *Prof. Anim. Sci.* 26:142-149
- Riesterer, J. L., Undersander, D. J., Casler, M. D. and Combs, D. K. 2000. Forage yield of stockpiled perennial grasses in the Upper Midwest USA. *Agron. J.* 92:740-747
- Riesterer, J. L., Undersander, D. J., Casler, M. D. and Combs, D. K. 2000. Forage yield of stockpiled perennial grasses in the Upper Midwest USA. *Agron. J.* 92:740-747.
- Saskatchewan Ministry of Agriculture. 2008. Bale Grazing. [Online] Available: <http://www.agriculture.gov.sk.ca/Default.aspx?DN=7d86096d-566b-4c5d-b9c6-7019b64b9728>.
- Schoenau, J.J., and Davis, J. G. 2006. Optimizing soil and plant responses to land-applied manure nutrients in the Great Plains of North America. *Can. J. Soil. Sci.* 86: 587-59.
- Smith, A., Schoenau, J., Lardner, H.A., and Elliott J. 2011. Nutrient export run-off from an in-field cattle overwintering site in East-central Saskatchewan. *Water Sci. Tech.* 64: 1790-1795.
- Van De Kerckove, A.Y., Lardner, H.A., Walburger, K., McKinnion, J.J., and Yu, P. 2011. Effects of supplementing spring-calving beef cows grazing barley crop residue with a wheat-corn blend dried distillers grains with soluble on animal performance and estimated dry matter intake. *Prof. Anim. Sci.* 27: 219-227.
- Volesky, J.D., Adams, D.C. and Clark, R.T. 2002. Windrow grazing and baled-hay feeding strategies for wintering calves. *J. Range Manage.* 55: 23-32.
- Warner, J.M., Martin, J.L., Hall, Z.C., Kovarik, L.M., Hanford, K.J. and Rasby, R.J. 2011. The effects of supplementing beef cows grazing cornstalk residue with a dried distillers grain based cube on cow and calf performance. *Prof. Anim. Sci.* 27: 540-546.
- Wilson, C.B., Erickson, G.E., and Adams, D.C. 2004. A review of corn stalk grazing on animal performance and crop yield. *Nebraska Beef Rep.:* 13–15.

**Table 1. Chemical composition of stockpiled forage and round bale hay (% DM)
(Kulathunga et al., 2016)**

Item	Forage system ¹		SEM	P-value
	SPF	HAY		
October				
OM	92.1	90.3	2.13	<0.01
CP	10.7	10.0	0.47	0.24
ADF	42.0	41.6	0.53	0.64
NDF	61.8	60.0	0.53	0.02
P	0.21	0.22	0.005	0.06
Ca	0.70	0.70	0.000	0.88
TDN ²	52.5	52.7	0.74	0.76
DE	2.33	2.35	0.026	0.53
December ³				
OM	91.1	90.8	0.34	0.66
CP	9.5	8.7	0.34	0.16
ADF	45.6	44.5	0.99	0.46
NDF	66.8	64.0	0.87	0.04
P	0.13	0.10	0.013	0.17
Ca	0.62	0.66	0.040	0.46
TDN	50.5	51.8	1.12	0.46
DE	2.2	2.25	0.04	0.45

¹SPF = stockpiled perennial grass-legume forage grazed in field paddocks; HAY = round bale grass-legume hay fed in drylot pens.

²Calculated using Penn State equations (Adams, 1995).

³December forage samples in yr 1 were considered unreliable due to laboratory problems; therefore only yr 2 and 3 December samples analyzed.

Table 2. Cow performance grazing either stockpile forage or drylot fed hay bales over 3 yr (Kulathunga et al., 2016)

Item	Forage system ¹		SEM	P-value
	SPF	HAY		
Body weight ² , kg				
Initial	651.3	645.3	2.80	0.10
Final	674.9	677.3	4.85	0.69
Change	23.6	32.0	5.17	0.20
Rib fat, mm				
Initial	3.4	3.3	0.27	0.74
Final	4.9	4.2	0.31	0.18
Change	1.5	0.9	0.20	0.22
Rump fat, mm				
Initial	3.6	3.3	0.42	0.63
Final	4.5	4.1	0.34	0.38
Change	0.9	1.0	0.16	0.96
BCS ³				
Initial	2.6	2.6	0.06	0.47
Final	2.7	2.7	0.06	0.42
Change	0.1	0.1	0.05	0.37

¹SPF = stockpiled perennial grass-legume forage grazed in field paddocks; HAY = round bale grass-legume hay fed in drylot pens.

²Cow BW adjusted for conceptus growth.

³BCS = body condition score (1 = emaciated; 5 = obese; Lowman et al., 1976).

Table 3. Effect of backgrounding system on DMI and consumed nutrients over 3 yr (Kumar et al., 2012)

Item	Backgrounding system ¹			SEM	P-value
	BAR	MILL	DL		
DMI, kg/d	7.76	6.81	7.53	0.447	0.32
CP, kg/d	0.92	0.90	0.75	0.105	0.19
NDF, kg/d	3.25	3.16	3.84	0.286	0.23
TDN, kg/d	4.28	3.51	3.89	0.518	0.27

¹BAR = swathed barley grazing; MILL = swathed millet grazing; DL = drylot pen feeding.

Table 4. Effect of backgrounding system on beef calf performance over 3 yr (Kumar et al., 2012)

Item	Backgrounding system ¹			SEM	P-value
	BAR	MILL	DL		
Performance					
Initial BW, kg	207.1	207.3	207.7	8.46	0.96
Final BW, kg	288.1 ^a	269.4 ^b	290.7 ^a	7.65	0.01
ADG, kg/d	0.8 ^a	0.6 ^b	0.8 ^a	0.03	0.01
BW change, kg	77.9 ^a	59.0 ^b	79.9 ^a	4.39	0.01

^{a,b}Means within a row with different superscripts differ ($P < 0.05$).

¹BAR = swathed barley grazing; MILL = swathed millet grazing; DL = drylot pen feeding.

Table 5. Effect of supplementation on beef steer performance while winter bale grazing over 2 yr (Lardner et al., 2018)

Item	BARL ¹	DDGS	50:50	SEM	P-value
Initial BW ² , kg	228	228	230	7.7	0.79
Final BW ² , kg	322	331	329	7.3	0.10
Gain, kg	94	103	99	2.4	0.07
ADG, kg/d	0.87	0.97	0.92	0.02	0.07

¹BARL = steers supplemented with 100% barley; DDGS = steers supplemented with 100% wheat DDGS; 50:50 = steers supplemented with 50% wheat DDGS + 50% barley.

²Shrunken BW calculated as 96% of liveweight according to NASEM (2016).

Table 6. Effect of wintering system on beef cow performance over 3 yr (Krause et al., 2012)

Item	Treatment ¹			SEM	P-value
	DLPF	OATG	PEAG		
BW, kg					
Initial	650.3	660.9	648.0	6.67	0.39
Final	707.1 ^a	683.3 ^a	651.7 ^b	7.56	0.01
BW change, kg					
Final	65.9 ^a	26.5 ^b	3.7 ^c	3.92	0.01
BCS ²					
Initial	2.6	2.8	2.8	0.07	0.23
Final	2.8	2.7	2.6	0.08	0.16
Change	0.2 ^a	-0.1 ^{ab}	-0.2 ^b	0.05	0.01
Rib fat, mm					
Initial	3.8	4.9	4.7	0.40	0.14
Final	5.5 ^a	5.0 ^{ab}	3.6 ^b	0.45	0.03
Change	1.6 ^a	0.1 ^b	-1.1 ^c	0.25	<0.01
Rump fat, mm					
Initial	3.8	5.4	4.9	0.55	0.14
Final	7.0 ^a	5.8 ^{ab}	4.2 ^b	0.58	0.01
Change	3.2 ^a	0.4 ^b	-0.8 ^b	0.41	<0.01

¹DLPF = drylot pen feeding; OATG = grazing oat residue in field paddocks; PEAG = grazing pea residue in field paddocks.
²BCS = Body condition score (1 = emaciated to 5 = grossy fat; Lowman et al., 1976).
^{a-b}Means (*n* = 9) within a row and with different letters differ (*P* < 0.05).

Table 7. Dry matter yield of corn varieties (Lardner et al., 2012)

Item	P7443R	DKC 27-54	P7535R	HLSR06	P7213R
Crop Heat Unit	2100	2175	2100	2250	2050
Dry matter, %	40.1	50.3	37.0	38.1	49.4
t/acre, wet	11.8	11.4	10.9	10.8	11.4
t/acre, DM	4.75	5.74	4.04	4.13	5.64

Table 8. Nutrient composition of corn varieties (Lardner et al., 2012)

Item ^z	P7443R	DKC 27-54	P7535R	HL SR06	P7213R
<i>September</i>					
CP, %					
Whole plant	7.8	7.7	6.4	8.1	7.0
Leaves	7.4	13.1	12.0	13.6	13.0
Grain+Cob	12.3	10.9	11.4	12.9	11.2
TDN, %					
Whole plant	69.7	70.8	68.6	69.2	68.7
Leaves	49.7	60.6	60.5	59.7	55.1
Grain+Cob	89.3	90.3	90.1	89.8	90.8
<i>November^z</i>					
CP, %					
Whole plant	7.7	8.5	8.7	9.7	6.7
TDN, %					
Whole plant	62.1	63.0	64.7	66.5	57.1

^zwhole plant

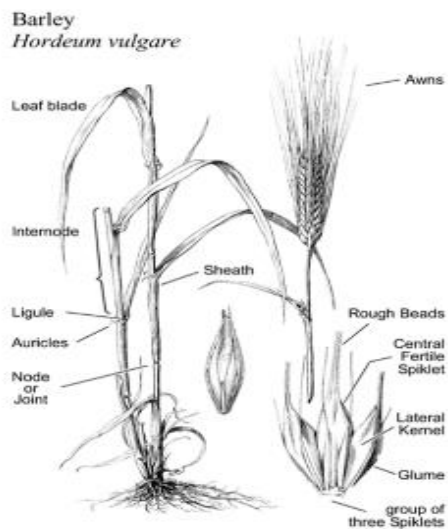


Figure 1. Parts of a cereal plant (McCartney et al. 2006).

Bull Development and Nutrition: Fertility and Beyond

Carl R. Dahlen,

Center for Nutrition and Pregnancy and Department of Animal Sciences

North Dakota State University

701-231-5588; Carl.Dahlen@ndsu.edu

Introduction

To maximize reproductive efficiency in their herds, beef producers pay close attention to nutritional and other managerial inputs. Following suit, many research efforts throughout the world have focused on aspects of managing breeding females. Substantially less science-based information is available, however, regarding the nutritional management of bulls. The United States beef industry is dominated by herds that rely solely on bull breeding. The percentage of operations that relied only on bull breeding was 95.5% and of beef cows maintained, 98.4% were at least exposed to a bull during the breeding season (NAHMS 2020).

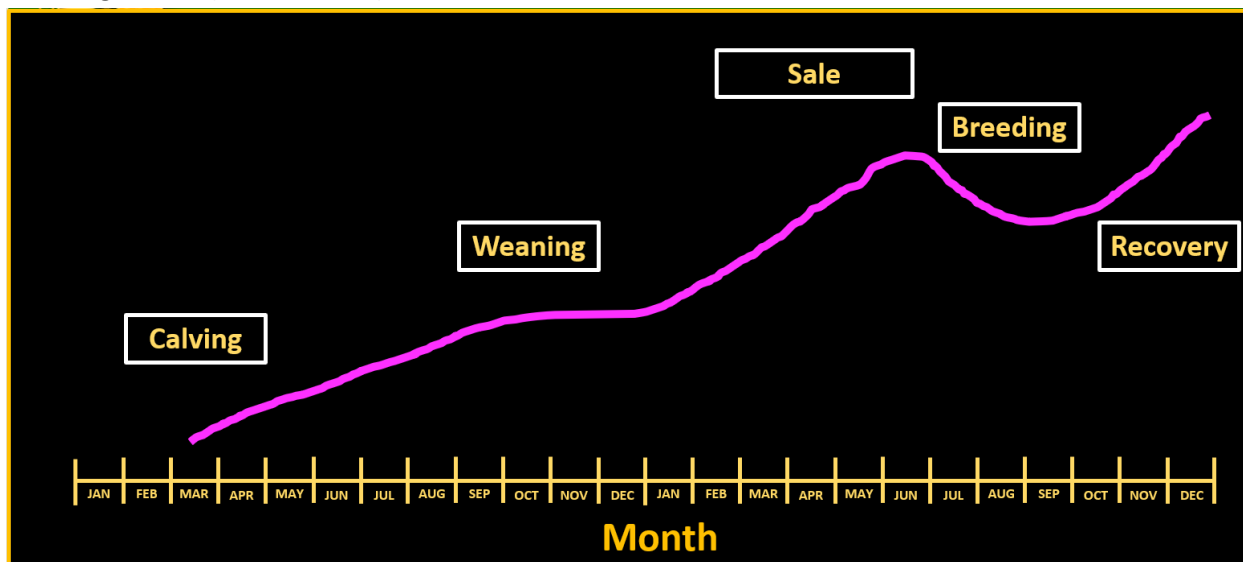


Figure 1. Schematic of bull body weight and key developmental milestones over the first 2 years of life.

Over the course of a bull's life and even within a year in his life, dramatic and dynamic changes in body weight and plane of nutrition are occurring (Figure 1). The pre-weaning, post-weaning, and post-breeding periods each present unique periods of potential management intervention. In addition, effects of relatively short-term feeding decisions (i.e. how should I feed my bulls today) may have long-term impacts that extend to their offspring.

Pre-Weaning Period.

During the pre-weaning period young bulls are managed with their dams; suckling milk and grazing the same pastures or consuming the same stored feeds as their dams. Though everything may look tranquil at this time there are major transitions occurring in the developing testicles that will have lifelong impacts on future sperm production. The period from 1 to 5 months is when seemingly subtle hormonal signals are responsible for proliferation of the Sertoli cell population. Sertoli cells are important because

there is a maximal number of developing sperm cells that each Sertoli cell will be able to protect and nourish later in life. The population of Sertoli cells is set by 5 months of age, but the more Sertoli cells found in the testis the greater the potential daily sperm production as an adult (Berndtson et al., 1987).

Nutrition before 6 months of age is likely the most important influence of future sperm production potential. Research in both beef and dairy bulls has found that enhancing early life nutrition can result in enhanced Sertoli cells populations, testicle size, and sperm production along with a reduced age at puberty (Brito et al., 2007; Brito et al., 2012; English et al., 2018; Kenny and Byrne, 2018). It is also important to note that negative effects of nutritional deficiencies experienced before puberty cannot be overcome by enhancing nutrition after weaning (Thundathil et al., 2016).

From a practical standpoint we need to avoid nutritional deficiencies when possible and consider options for enhancing plane of nutrition when practical. Bulls born from cows had larger scrotal circumference than bulls from heifer dams (Lunstra et al., 1988), likely due to the greater milk production in cows leading to greater bull body weight. Cases of poor pasture quality or availability put further nutritional strain on developing bulls and may need to be mitigated. Providing creep feed is a way to enhance pre-weaning nutrient delivery to bulls that may be practical, but heifer calves receiving excess creep feed could have reduced future milk production (Buskirk et al., 1996). If multiple pasture cells and associated labor are available there may be merit in dividing pastures by calf sex and applying sex-specific targeted management. If cooperators herd are being used as embryo transfer recipients of sale bulls be sure to choose multiparous cows with a good history of mothering ability and milk production.

Post-Weaning Period.

Post-weaning development strategies vary among producers with some raising bulls to gain at aggressive rates whereas others prefer a moderate gain approach. Gains from 2.2 to 3.5 lb/day were reported to be “safe” targets for bull development from 6 to 16 months of age (Brito et al., 2012) but a quick look at bull test reports or sale catalogs will show some bulls growing faster than 5 lb/day. For normal sperm development testicular temperature needs to be maintained 4 to 6 ° cooler than body temperature. High grain diets have been shown to increase scrotal fat and temperature (Bourgon et al., 2018), leading to an increased proportion of sperm with morphological abnormalities, and overall reduced motility.

Genetics and growth potential of specific bulls play a large role in gain that can be achieved and bulls need to be managed to ensure they are putting on weight as muscle and not excess fat. We have completed that first year of a 3-year effort comparing growth rates of 2.5 and 4.0 lb/day in Balancer bulls and have not observed any alterations in sperm properties (Crouse et al., unpublished).

An example of reproductive characteristics during the bull development period is found in Table 1. The 36 bulls used in this example were all embryo transfer calves from a single sire gestated and mothered by multiparous crossbred Angus cows. Average birthweight was 85 lb and bulls gained 2.7 lb per day while suckling. No management interventions (creep feed, etc.) were in place to provide extra nutrients during the pre-weaning period and weaning weight averaged 610 lb. After weaning we targeted a gain of 3.5 lb per day for 112 days. By 9 months of age a portion of the bulls were already pubertal, and all bulls had reached puberty and were classified as satisfactory breeders by the age of 13 months.

Item	Bull age in Months ²					SEM	P-Value
	9	10	11	12	13		
Body weight, lb	705	778	896	1006	1101	9.24	<0.01
Scrotal circumference, cm	30.0	32.0	34.5	35.7	36.8	0.26	< 0.01
Ejaculate volume, mL	2.0	4.3	7.2	7.8	8.0	0.5	< 0.01
Concentration, million/mL	32.0	56.0	73.4	124.5	115.6	14.4	< 0.01
Total sperm, million	68	277	536	1048	937	142	< 0.01
Pubertal, %	22.2	72.2	88.8	94.4	100.0	0.05	<0.01
Motile, %	28.4	51.9	57.5	58.7	72.7	3.5	< 0.01
Progressive, %	9.0	39.9	46.6	47.6	61.5	3.1	< 0.01
Slow, %	3.6	6.2	3.6	7.0	2.6	2.0	0.36
Static, %	55.7	36.6	41.3	40.8	27.3	3.8	< 0.01
Morphology							
Proximal droplet, %	12.8	6.2	4.4	4.2	3.5	1.1	< 0.01
Bent tail, %	8.2	7.5	5.4	3.5	3.0	1.6	0.07

After a development period bulls are often sold. During the time of transition to a different environment and different diets bulls can lose a significant amount of weight. A Canadian study found that bulls lost an average of 2.6 to 4.8 lb per day (depending on breed) for 45 to 50 days after a sale (Barth et al., 1995). Recommendations for transitioning bulls to new environments toward breeding season include acclimating to forage, exercising on pastures, and targeting gain according to desired trajectory (Walker et al, 2009).

Breeding and Post-Breeding Periods.

Before being turned out to breeding pastures every bull should receive a breeding soundness exam (BSE; Koziol and Armstrong 2018). A BSE will evaluate semen and a bulls' physical characteristics (eyes, teeth, feet and legs, accessory sex glands) to determine suitability for breeding. Once a BSE is passed (or an alternative bull is identified), bulls can be turned out to pastured and monitored to ensure they are actively seeking and successfully breeding cows. During the breeding season bulls can experience dramatic weight loss; between 100 to 400 lb (Boyles et al., 2011; Walker et al., 2009; Hersom and Thrift, 2008). Continue close monitoring during the breeding season and be ready to replace injured or overly-fatigued bulls. Bulls losing weight during breeding season must be managed to regain the weight lost during the breeding season (Barth 2013), and sufficient nutrients need to be delivered to account for additional body growth. We monitored a group of bulls and saw continued weight increase as bulls matured from 1500 lb. at 2 years of age to 1750 lb. at 3 years of age to 1870 lb. at 4 years of age (Dahlen et al, unpublished)

Producer decisions determine the point at which bulls begin losing and gaining weight relative to the breeding season (Figure 2). In some scenarios, bulls begin losing weight only after being placed with

females in breeding pastures. These bulls are then managed to gain weight, reaching targeted weight just before the subsequent breeding season. Bulls in this scenario would be in a **positive plane of nutrition** over the time course of sperm development (spermatogenesis). In an alternative scenario bulls may start losing weight before the breeding season. Perhaps these bulls experienced a recent change in environment and diet after purchase, or perhaps they were managed to gain weight over winter and needed to be cut back to get into “breeding shape” ahead of the breeding season. In either instance, these bulls would be on a **negative plane of nutrition** leading up to the breeding season. When we evaluate these two common production scenarios together we see a major divergence in plane of nutrition leading up to the breeding season.

Spermatogenesis is a continual process that takes roughly 61 d for sperm development, followed by up to 14 d residence in the epididymis prior to ejaculation. The net result is that sperm used to inseminate a cow today likely began the process of development up to 75 d before. Thus, divergence in plane of nutrition likely exposes sperm to different hormonal profiles and metabolic substrates during the time of spermatogenesis, residence in the epididymis, and upon combination with seminal plasma at ejaculation. The consequences of sperm development in these differing metabolic states resulting from divergent nutrition remain underexplored.

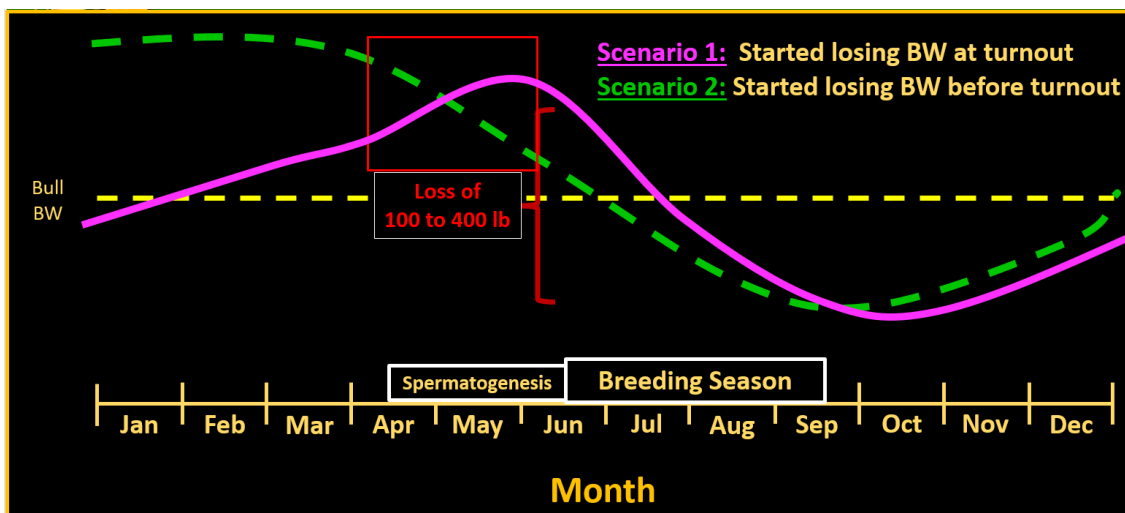


Figure 2. Body weight fluctuations of mature bulls over the course of a year. Figure depicts common production scenarios that result in different planes of nutrition during sperm development.

The Unknown: Does Nutrition During Sperm Development Impact Future Offspring?

The concept of developmental programming is that post-natal physiology is dictated, in part, by pre-natal factors such as maternal metabolism and other environmental factors (Barker and Clark, 1997). While the first evidence of programming effects were observed in humans during periods of major food shortages (Schulz, 2010), research in livestock species has revealed that maternal nutrition and management can impact offspring growth and performance (Caton et al., 2019; Dahlen et al., 2021). However, these studies and those of many other researchers have been confined to evaluations of the impact that dam management has on her offspring.

Research efforts in other species have shown that messages carried in a sires' semen after exposure to alcohol, drugs, or an obese state result in altered offspring development (Baber and Koren, 2015; Fullston et al., 2017). Impacts of sire obesity can be long lasting with differences observed all the way out to post-pubertal semen development in male offspring (Fullston et al., 2015). In addition, low protein diets fed to male mice altered offspring gene expression, resulting in heavier offspring with lower bone density compared with offspring from sires fed adequate protein (Watkins et al., 2017). However, paternal programming effects haven't been extensively studied or characterized in our livestock species.

One of the long-term goals of our research group is to determine the impacts of divergent bull nutrition on indicators of fertility and offspring outcomes. To date we have imposed treatments where bulls were managed on a positive or negative plane of nutrition (targeting a gain or lose 12.5% of their original body weight) for a 112-day period. This project was repeated over 2 years to allow for collection of a portfolio of samples from each individual bull in both a positive and negative plane of nutrition. Semen was collected on the last day of the experiment each year and cryopreserved for further analysis and breeding.

Any specific changes observed in semen and offspring are likely driven through epigenetic changes in sperm as a result of sire nutrition or management (Teperek et al., 2016). When we evaluated sperm for changes in sperm gene expression we saw differential expression of 769 genes (Diniz et al., unpublished) with a portion of differentially expressed genes being related to epigenetic mechanisms. These results indicate that plane of nutrition during spermatogenesis is altering messages in sperm that could influence fertility and also be delivered with the sperm at the time of fertilization. Messages delivered at the time of fertilization could subsequently influence growth and development of the resulting calf crop.

Our future efforts in this area include evaluation of *in vitro* fertilization rates and pregnancy rates in females after artificial insemination. Once calves are born we will evaluate their growth, metabolism, and reproductive responses. In addition, continued evaluation of these F1 calves through their reproductive stages will produce F2 generation offspring that will provide insight into whether any multigenerational effects of divergent sire nutrition are observed.

Summary

Early-life nutrition is extremely important to set the stage for timing of puberty and future sperm production. In cases where nutrient deficiencies are anticipated active steps should be taken to enhance nutrients available to pre-weaning calves. Post-weaning growth should be monitored closely to allow for adequate growth without excess fat accumulation. Monitor bulls closely during the breeding season to identify injuries and ensure bulls are actively breeding. Manage bulls to regain lost condition after the breeding season and allow for additional growth in younger bulls. Implications of specific timing and patterns of body weight gain on sperm characteristics and offspring outcomes are currently unknown but offer an exciting avenue for future discovery.

Literature Cited

Baber, M., and G. Koren. 2015. Investigating the fetal and postnatal effects of paternal alcohol exposure in mouse offspring: a review. *J Popul Ther Clin Pharmacol* 22(1):e57-58.

- Barker, D. J., and P. M. Clark. 1997. Fetal undernutrition and disease in later life. *Rev Reprod* 2(2):105-112.
- Barth, A. D., W. F. Cates, and R. J. Harland. 1995. The effect of amount of body fat and loss of fat on breeding soundness classification of beef bulls. *Can Vet J* 36(12):758-764.
- Berndtson, W. E., G. Igboeli, and B. W. Pickett. 1987. Relationship of Absolute Numbers of Sertoli Cells to Testicular Size and Spermatogenesis in Young Beef Bulls. *Journal of Animal Science* 64(1):241-246.
- Bourgon, S. L., M. Diel de Amorim, T. Chenier, M. Sargolzaei, S. P. Miller, J. E. Martell, and Y. R. Montanholi. 2018. Relationships of nutritional plane and feed efficiency with sexual development and fertility related measures in young beef bulls. *Anim Reprod Sci* 198:99-111. doi: 10.1016/j.anireprosci.2018.09.007
- Barth, A. D. 2013. *Bull Breeding Soundness* 3rd Ed. Western Canadian Association of Bovine Practitioners.
- Brito, L. F., A. D. Barth, R. E. Wilde, and J. P. Kastelic. 2012. Effect of growth rate from 6 to 16 months of age on sexual development and reproductive function in beef bulls. *Theriogenology* 77(7):1398-1405. doi: 10.1016/j.theriogenology.2011.11.003
- Brito, L. F. C., A. D. Barth, N. C. Rawlings, R. E. Wilde, D. H. Crews, P. S. Mir, and J. P. Kastelic. 2007. Effect of improved nutrition during calthood on serum metabolic hormones, gonadotropins, and testosterone concentrations, and on testicular development in bulls. *Domest Anim Endocrin* 33(4):460-469. doi: 10.1016/j.domaniend.2006.09.004
- Buskirk, D. D., D. B. Faulkner, W. L. Hurley, D. J. Kesler, F. A. Ireland, T. G. Nash, J. C. Castree, and J. L. Vicini. 1996. Growth, reproductive performance, mammary development, and milk production of beef heifers as influenced by prepubertal dietary energy and administration of bovine somatotropin. *Journal of Animal Science* 74(11):2649-2662.
- Caton, J. S., M. S. Crouse, L. P. Reynolds, T. L. Neville, C. R. Dahlen, A. K. Ward, and K. C. Swanson. 2019. Maternal nutrition and programming of offspring energy requirements. *Transl Anim Sci* 3(3):976-990. doi: 10.1093/tas/txy127
- Dahlen, C. R., P. P. Borowicz, A. K. Ward, J. S. Caton, M. Czernik, L. Palazzese, P. Loi, and L. P. Reynolds. 2021. Programming of Embryonic Development. *Int J Mol Sci* 22(21)doi: 10.3390/ijms222111668
- English, A. M., D. A. Kenny, C. J. Byrne, H. Sauerwein, C. Uhr, M. A. Crowe, C. Staub, S. M. Waters, and S. Fair. 2018. Role of early life nutrition on regulating the hypothalamic-anterior pituitary-testicular axis of the bull. *Reproduction* 156(4):283-297. doi: 10.1530/REP-17-0671
- Fullston, T., N. O. McPherson, J. A. Owens, W. X. Kang, L. Y. Sandeman, and M. Lane. 2015. Paternal obesity induces metabolic and sperm disturbances in male offspring that are exacerbated by their exposure to an "obesogenic" diet. *Physiol Rep* 3(3)doi: 10.14814/phy2.12336
- Fullston, T., N. O. McPherson, D. Zander-Fox, and M. Lane. 2017. The most common vices of men can damage fertility and the health of the next generation. *J Endocrinol* 234(2):F1-F6. doi: 10.1530/JOE-16-0382
- Hersom, M and T. Thrift. 2008. *Nutritional Management of Bulls*. University of Florida AN211. Available at: [AN21100.pdf \(ufl.edu\)](https://www.ufl.edu/~an211/AN21100.pdf)
- Kenny, D. A., and C. J. Byrne. 2018. Review: The effect of nutrition on timing of pubertal onset and subsequent fertility in the bull. *Animal* 12(s1):s36-s44. doi: 10.1017/S1751731118000514
- Koziol, J.H. and C.L. Armstrong. 2018. *Society for Theriogenology Manual for Breeding Soundness Examinations of Bulls*. Second Ed. Society for Theriogenology, Pike Road, Alabama.
- Lunstra, D. D., K. E. Gregory, and L. V. Cundiff. 1988. Heritability estimates and adjustment factors for the effects of bull age and age of dam on yearling testicular size in breeds of bulls. *Theriogenology* 30(1):127-136. doi: 10.1016/0093-691x(88)90270-1
- NAHMS. National Animal Health Monitoring System. 2020. Part I. Beef Cow-calf Management Practices in the United States, 2017. Fort Collins, CO. Pp. 49- 51.

- Schulz, L. C. 2010. The Dutch Hunger Winter and the developmental origins of health and disease. *Proc Natl Acad Sci U S A* 107(39):16757-16758. doi: 10.1073/pnas.1012911107
- Teperek, M., A. Simeone, V. Gaggioli, K. Miyamoto, G. E. Allen, S. Erkek, T. Kwon, E. M. Marcotte, P. Zegerman, C. R. Bradshaw, A. H. Peters, J. B. Gurdon, and J. Jullien. 2016. Sperm is epigenetically programmed to regulate gene transcription in embryos. *Genome Res* 26(8):1034-1046. doi: 10.1101/gr.201541.115
- Thundathil, J. C., A. L. Dance, and J. P. Kastelic. 2016. Fertility management of bulls to improve beef cattle productivity. *Theriogenology* 86(1):397-405. doi: 10.1016/j.theriogenology.2016.04.054
- Walker, J., G. Perry, R. Daly, and K. Olson. 2009. Bull management and nutrition. *Proceedings, the range beef cow symposium XXI*.
- Watkins, A. J., S. Sirovica, B. Stokes, M. Isaacs, O. Addison, and R. A. Martin. 2017. Paternal low protein diet programs preimplantation embryo gene expression, fetal growth and skeletal development in mice. *Biochim Biophys Acta* 1863(6):1371-1381. doi: 10.1016/j.bbadis.2017.02.009

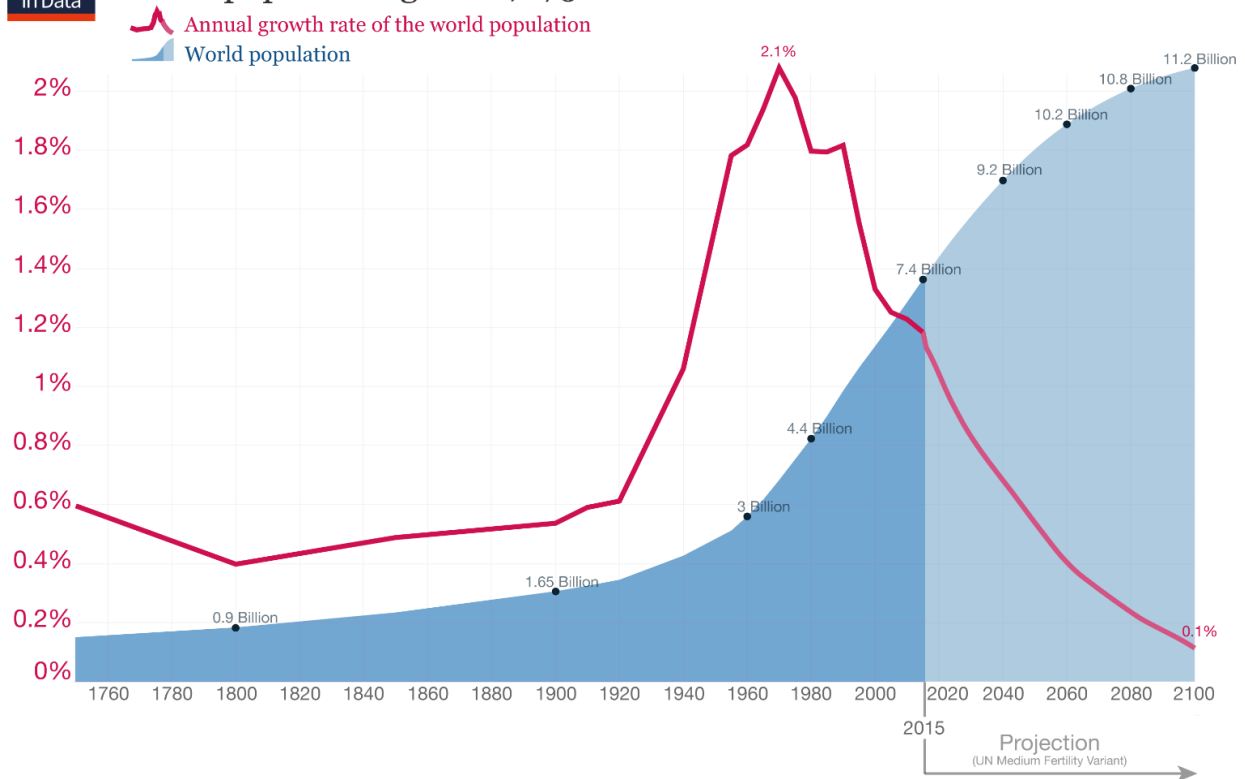
Sustainable Animal Agriculture in 21st century

Robert Collier
Professor & Head
Animal Veterinary and Food Sciences
University of Idaho
875 Perimeter Drive
Moscow, ID 83844-2330
rcollier@uidaho.edu
208-884-9489

Introduction

Although the human population of the earth has been growing since the end of the Bubonic Plague of 1348, it has grown most rapidly since the end of the second world war. Coupled with decreasing mortality rates this has resulted in explosive population growth (Figure 1). However, as also noted in this figure the actual rate of population growth peaked in the late 1960's and has declined rapidly since. Ultimately, this will lead to a leveling of the world population at around 11.2 billion people in 2100. In addition to a rising population many consumers around the world are opting to increase the high-quality protein content of their diet (meat, milk and eggs). Structural changes in diets, will continue to determine shifts from staples to livestock products and fruit and vegetables. Thus, use of livestock products in human diets will increase in Asia, Latin America and Africa. According to estimates compiled by the Food and Agriculture Organization (FAO), by 2050 we will need to produce 60 per cent more food for a world population of 9.3 billion. Increasingly, public concerns around agricultural production practices and the "sustainability of our food production systems" have become part of the demands on our agricultural systems. Thus, in addition to increasing our food supply by 60 per cent our agricultural production systems are challenged to do so with less arable land, during a period of world-wide climate change with improved animal welfare practices and in a more sustainable manner.

World population growth, 1750-2100



Data sources: Up to 2015 OurWorldInData series based on UN and HYDE. Projections for 2015 to 2100: UN Population Division (2015) – Medium Variant. The data visualization is taken from OurWorldInData.org. There you find the raw data and more visualizations on this topic.

Licensed under CC-BY-SA by the author Max Roser.

In the last 50 years, there has been a remarkable emergence of innovations and technological advances that are generating promising changes and opportunities for sustainable agriculture, yet at the same time the agricultural sector worldwide faces numerous daunting challenges. Not only is the agricultural sector expected to produce adequate food, fiber, and feed, and contribute to biofuels to meet the needs of a rising global population, it is expected to do so under increasingly scarce natural resources and climate change. Growing awareness of the unintended impacts associated with some agricultural production practices has led to heightened societal expectations for improved environmental, community, labor, and animal welfare standards in agriculture.

What is Sustainable Agriculture?

The word sustainability must include a component that considers social values (Thompson, 1997, 2007). The current US legal definition (US Code Title 7, Section 3103) states: “an integrated system of plant and animal production practices having a site-specific application that will over the long-term: satisfy human food and fiber needs, enhance environmental quality and the natural resource base upon which the agriculture economy depends, make the most efficient use of nonrenewable resources and on-farm resources and integrate, where appropriate, natural biological cycles and controls, sustain the economic viability of farm

operations, and enhance the quality of life for farmers and society as a whole.” von Keyerslinkg et al. 2013.

The three pillars of sustainability are shown in Figure 2 and include environment, economic and social components. All three components must be addressed to provide sustainability to an agricultural enterprise.

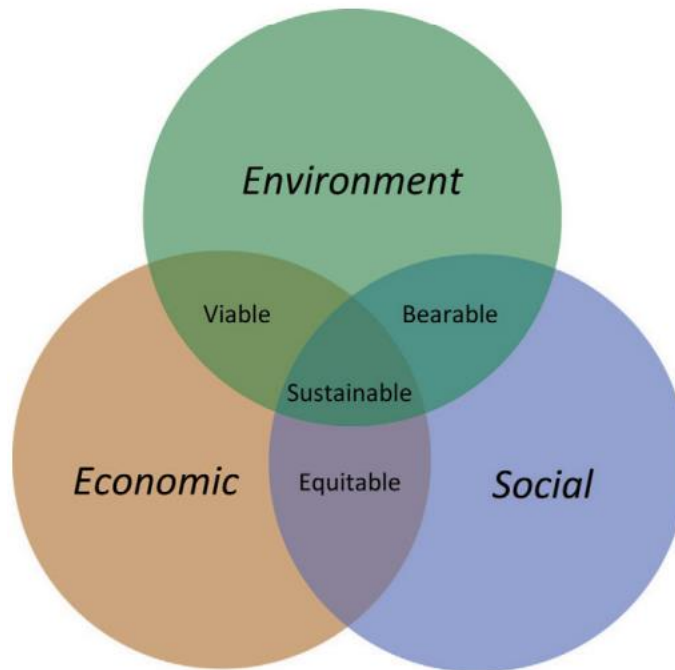
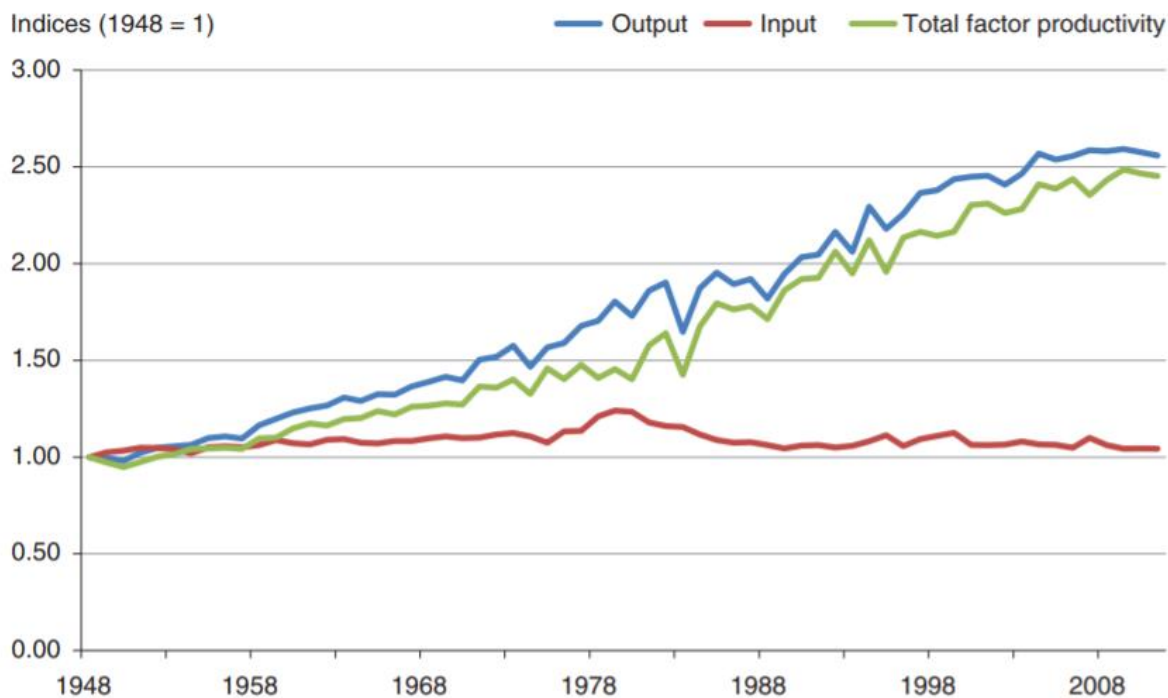


Figure 2. The three pillars of sustainability from von Keyserlingk et al. 2013

Since the 1940's the majority of research in agriculture has focused on increasing agricultural productivity. This has resulted in dramatic improvements in yield and efficiency of agricultural food production as shown in Figure 2.



Source: USDA, Economic Research Service productivity accounts.

Figure 3. Agricultural productivity growth accounted for most output growth between 1948 and 2011

This figure represents all of agriculture and the animal agriculture components are included in the data used for the figure. Thus, improving agricultural productivity has contributed to historically low food prices in the U.S. since the end of the Second World War. It has also contributed greatly to the reduction in carbon footprint of animal agriculture.

However, this measure of agricultural efficiency does not encompass all of the components of agricultural sustainability. As pointed out by von Keyserlingk et al. in their review on sustainability “This approach alone will not address the gaps in knowledge and educational needs ... for many aspects of the sustainability of dairy production and consumer understanding. We also require transformative research that allows for whole system redesign (Reganold et al., 2011).” These additional concerns include animal welfare, use of immigrant workers, carbon footprint, consumer input and economics.

In addition to the challenges of developing a holistic sustainable model for agriculture the animal agricultural sector faces the additional challenge of alternative protein sources to animal products as a disruptive force in the economics of animal agriculture. Alternative dairy, was analyzed by the consulting group of Ernst and Young, 2021 who provided a very bullish estimate of upwards of 60% market share by 2040, and they stated that this is a strong analog for alternative meat. The alternative meat forecast of 9% by 2030 compares favorably with the experience of alternative milk, which has penetrated American households and now accounts for 15% of all dollar sales of retail milk. Ernst and Young 2021. They further state that the additional components of the alternative dairy category, including cheese, is advancing

technologically and continues to attract increasing levels of investment. If social, environmental and regulatory drivers (e.g., water or carbon taxes) are factored into conventional protein production, market shares for alternative meat and dairy could surpass their base forecast considerably. Ernst and Young, 2021. However, the Purdue Center of Commercial Agriculture recently reported that the majority of 400 producers (86%) surveyed expected the alternative protein market share would be only 1-10 per cent in 5 years.

This emerging shift could explain why even though aggregate consumption of meat-based proteins worldwide is increasing, the overall growth rate is expected to decline by half. Plant-based food (the largest source of alternative protein) sales rose 17 percent in 2018 and the use of alternative protein as a food ingredient in consumer products is predicted to continue growing. Currently, the market base for alternative protein is approximately \$2.2 billion compared with a global meat market of approximately \$1.7 trillion, making the growth rate of the alternative proteins marginal to the overall meat market.

The best way for animal agriculture to respond to all of these challenges is to improve overall sustainability of animal agriculture which will counter many of the public perception issues around animal welfare and environmental impacts of animal products and keep animal products price competitive. If the dairy industry meets its target of zero carbon footprint of the dairy industry by 2050 it is difficult to imagine that alternative protein production could match that performance. This research effort to improve sustainability of animal agriculture has to be a multidisciplinary approach to include both agronomic and animal agriculture components to increase sustainability. Examples of these will be provided during the presentation.

References

- Dongoski, R. 2021. When might the term alternative protein be obsolete. [EY Food and Agribusiness | EY - US](#)
- FAO. 2006a. World agriculture: towards 2030-2050. Interim report. Rome. http://www.fao.org/fileadmin/user_upload/esag/docs/Interim_report_AT2050web.pdf
- Reganold, J. P., D. Jackson-Smith, S. S. Batie, R. R. Harwood, J. L. Kornegay, D. Bucks, C. B. Flora, J. C. Hanson, W. A. Jury, D. Meyer, A. Schumacher Jr., H. Sehmsdorf, C. Shennan, L. A. Thrupp, and P. Willis. 2011. Transforming U.S. Agriculture. *Science* 332:670–671.
- Thompson, P. B. 1997. Sustainability as a norm. *Phil. and Tech.* 2:75–93.
- Thompson, P. B. 2007. Agricultural sustainability: What it is and what it is not. *Int. J. Agric. Sustain.* 5:5–16.
- von Keyserlingk, M. A. G, N. P. Martin, E. Kebreab, K.F. Knowlton, R.J. Grant, M. Stevenson, II, C. J. Sniffen , J. P. Harner III , A. D. Wright ,and S. I. Smith.2013. Invited review: Sustainability of the US dairy industry. *J. Dairy Sci.* 96:5405-5425.

55th Annual Pacific Northwest Animal Nutrition Conference

January 17-18, 2022
Boise, Idaho

Graduate Student Abstracts

GRADUATE STUDENT POSTER ABSTRACTS

167 - 174

Effect of weaning pace and age on the health measures and tissue gene expression of inflammatory markers in Holstein dairy calves

B.C. Agostinho, A. Wolfe, C.Y. Tsai, D.E. Konetchy, A.H. Laarman, and P. Rezamand.

Evaluation of Forage Quality of Alfalfa from 200 Varieties Produced in the Pacific Northwest

S. Dreger, D.A. Llewellyn, O.S. Norberg, S. Fransen, G. Wang, D. Combs, G. Shewmaker, E. van Santen, L.X. Yu

Management Strategies to Reduce Negative Health Outcomes in Transported Pre-weaned Calves

K.K. Elmore, P. Rezamand, D. Konetchy, M. Chahin, B.C. Agostinho, A.H. Laarman, and G.E. Chibisa

Evaluation of Growth, Meat Quality, and Sensory Characteristics of Wool, Hair and Composite lambs

M.L. Heimbuch, J.B. Van Buren, B.S. Epperson, O.F. Kayleen, S.M. Jepson, J.A. Nasados, D.A. Vinci, W.J. Price, K.R. Vierck, D.E. Konetchy, P.D. Bass, and M.J. Colle.

The effect of feeding supplemental zeolite (clinoptilolite) of two different particle sizes on measures of nitrogen utilization and nutrient digestibility in finishing beef heifers

C.A. Myers, M.E. de Haro Marti, M. Chahine, and G.E. Chibisa

Impact of maternal nutrition on postnatal growth of crossbred beef steers

K.F. Oliver, J.B. Van Buren, J.B. Hall, M.L. Heimbuch, S. Jepsen, B. Epperson, J.A. Nasados, P.D. Bass, and M.J. Colle

The effects of allyl isothiocyanate inclusion as an additive on whole-plant corn silage

L.D.M. Pereira, P. Rezamand, B.C. Agostinho, G.L.D. Vigne, D. Volpi, N.N. de Mello, Q.G. Tavares, P. Schmid, M. Zopollatto

Impacts of heifer post-weaning intake classification on performance measurements of lactating and non-lactating two-, five- and eight-year-old Angus beef females

K.R. Wellnitz, C.T. Parsons, J.M. Dafoe, S.A. Wyffels, D.L. Boss, T. DelCurto, and M.L. Van Emon

Effect of weaning pace and age on the health measures and tissue gene expression of inflammatory markers in Holstein dairy calves

B. C. Agostinho¹, A. Wolfe², C. Y. Tsai¹, D. E. Konetchy¹, A. H. Laarman², P. Rezamand¹.

¹ Department of Animal, Veterinary & Food Sciences, University of Idaho, Moscow, Idaho, ² Agricultural, Life and Environmental Sciences, University of Alberta, Edmonton, Canada

Weaning is one of the most stressful events in dairy calves' life, which may induce inflammatory responses; however, the existing knowledge is limited. Therefore, the objective of this study was to evaluate the effect of the weaning at two ages (early at 49 vs. late at 63 d) and two weaning paces (abrupt over 3 d vs. gradual over 14 d) on selected health measures and local inflammation status of dairy calves. Forty Holstein calves were blocked by gender (20 male and 20 female) and body weight at birth and randomly assigned in a 2 × 2 factorial arrangement of treatment (weaning age; weaning pace). The treatments consisted of early-abrupt (EA), early-gradual (EG), late-abrupt (LA), and late-gradual (LG). Milk replacer was fed twice daily (24% CP, 17% fat; up to 1,200 g/d), and water, calf starter, and alfalfa hay were fed ad libitum. Health measures were obtained before and after weaning. Twenty males were terminated one-day post-weaning. Tissues from jejunum, large intestine, as well as abdominal and perirenal adipose tissues were collected and stored at -80°C, and gene expression was determined using rt-qPCR. The target genes included Interleukin-8 (IL-8), Tumor necrosis factor- α (TNF- α), and Nuclear factor κ -B (NF- κ B). Cycle threshold (Ct) of target genes was corrected by Ct of house-keeping genes (GAPDH and RPS-9) and were used (Δ Ct) for statistical analysis using the mixed model of SAS with significance declared at $P \leq 0.05$ and the tendency at $P \leq 0.10$. Calves weaned at late-stage had a greater respiration rate ($P = 0.07$) than calves weaned at an early-stage. Calves weaned abruptly had a greater heart rate ($P = 0.01$) than those weaned gradually. Body temperature was not affected by the treatment ($P > 0.16$). Calves weaned late-stage presented a greater expression of TNF- α in the jejunum and perirenal adipose tissue ($P = 0.01$; 0.02 , respectively) when compared with that of the early-stage. In addition, large intestinal expression of IL-8 tended to reduce in calves weaned abruptly ($P = 0.10$) when compared to gradually. The expression of TNF- α and NF- κ B tended to be greater ($P = 0.08$; 0.07 , respectively) in abdominal adipose tissue of calves weaned gradually than abruptly. In summary, weaning pace and age at weaning altered some health measures and inflammation status in the jejunum, large intestine, and adipose tissues.

Key words: dairy calf, inflammation, stress

Evaluation of Forage Quality of Alfalfa from 200 Varieties Produced in the Pacific Northwest

S. Dreger¹, D.A. Llewellyn¹, O.S. Norberg², S. Fransen³, G. Wang⁴, D. Combs⁵, G. Shewmaker⁶, E. van Santen⁷, L.X. Yu⁸

¹ Department of Animal Sciences, WSU, Pullman, WA, U.S., ² Franklin County Extension Office, WSU, Pasco, WA, U.S., ³ WSU IAREC, Prosser, WA, U.S., ⁴ Eastern Oregon Agricultural and Natural Resource Program, OSU, LaGrande, OR, U. S., ⁵ Department of Dairy Science, U of W, Madison, WI, U.S., ⁶ Kimberly R&E Center, U of I, Kimberly, ID, U.S., ⁷ Department of Agronomy, U of FL Gainesville, FL, U.S., ⁸ USDA ARS, Prosser, WA, U.S.

Alfalfa forage quality is an important consideration for alfalfa growers, marketers, and plays a major role in variety selection, alfalfa breeding, livestock nutrition, and the related economic value. This study is an evaluation of 200 varieties using first cutting (bud stage) alfalfa quality data across two years (2018 and 2019), from three locations (Prosser, WA, LaGrande, OR, and Kimberly, ID). Data were subjected to a statistical cluster analysis to categorize the varieties into forage quality groupings ranging from high-quality to low-quality. The twelve measures of forage quality analyses are: Non-Fiber Components (CP, fat, and ash); Fiber (ADF, aNDF, and lignin); Calculated Values (RFQ, RFV, and TDN); Carbohydrates (starch, ESC, WSC). Analyses revealed four clusters of quality for the 200 varieties. This evaluation focused on the high-quality cluster to best serve alfalfa breeders, marketers, and agricultural production. The averages of all clusters had CP (N*6.25) contents ranging from ~24% for the high-quality cluster, to ~22.2% in the low-quality cluster. Fat concentrations were a minor component with a negative linear correlation from high to low quality. Low levels of ash and/or ash contamination with little variation were observed. The high-quality cluster had ADF concentrations ranging from 29.5% to 32% and aNDF ranged from 35% to 40%. The average concentration of lignin in the high-quality cluster was ~6.25%, compared with ~6.8% for the low-quality cluster. Starch, ESC, and WSC analysis resulted in low starch and sugar concentration with no clear separation between quality clusters. The high-quality cluster had exceptional values for RFQ and RFV, (RFQ=189 and RFV=162). TDN was greatest in the high-quality cluster ranging from 62.2% to 65%. In comparison, the low-quality cluster ranged from 59.3% to 61%. Using the high-quality cluster (34/200 varieties), the top 10 varieties for each analyte, were compared to each variety's frequency across all 12 analytes, providing the top 10 varieties across all parameters. The top two varieties (AFX142001 and CW104014) each had the highest frequency across analytes (9/12, 75%). In addition, Amina, Gold Finch, Mariner V, DT-3044 and D2645 all had frequencies of greater than 50%. Nine varieties out of the top 10 included ADF and aNDF, eight for lignin. The top 10 alfalfa varieties had lower ADF, aNDF, and lignin in comparison to the remaining 190 entries. RFQ, RFV, and TDN values were consistent with the fiber results. Only about half of the top 10 alfalfa varieties were associated with CP or fat, and none were associated with ash. In addition, data suggests that the highest quality alfalfa varieties in this evaluation were in Fall Dormancy 3 and 4, (1 for FD 2 and 6). Results suggest that cluster analysis paired with frequency can be used to identify high-quality alfalfa varieties from large data sets which may provide useful information to alfalfa growers, breeders, marketers, and nutritionists. Further investigations that incorporate yield along with forage quality is indicated.

Key words: alfalfa, forage quality, fall dormancy, fiber, protein

Management Strategies to Reduce Negative Health Outcomes in Transported Pre-weaned Calves

Kylee K. Elmore*, Pedram Rezamand*, Denise Konetchy*, Mireille Chahin, Bruna C. Agostinho*, Anne H. Laarman³, and Gwinyai E. Chibisa*

*Department of Animal, Veterinary, and Food Science, University of Idaho, Moscow ID

² Department of Animal, Veterinary, and Food, Twin Falls Research and Extension Center University of Idaho, Twin Falls, ID

³ Department of Agriculture, Food, and Nutrition Science, University of Alberta, Edmonton, AB

Poor colostrum management and subsequent transportation increases mortality and morbidity rates in pre-weaned calves. However, pre-transport administration of Meloxicam, a non-steroidal anti-inflammatory drug (NSAID), may reduce the negative health outcomes. Therefore, the objective of this study was to determine the effects and potential interaction of colostrum management and NSAID administration on markers of stress and inflammation, and health measures in transported pre-weaned calves. Forty-eight (24 Jersey and 24 Holstein) male calves were collected at birth from a commercial farm and randomly assigned to treatments; either colostrum feeding according to recommendations or no colostrum/milk replacer, and administration of either an NSAID (1 mg of Meloxicam/kg of body weight) or a placebo prior to transportation. Blood samples were collected, and rectal temperature (RT) and body weight (BW) were measured (<2 d old) prior to transportation (300 miles), on arrival, 12 h post transport, and at harvest (36 h post transport). Harvested plasma was analyzed for cortisol and thiobarbituric acid-reacting substances (TBARS). Adipose, muscle, and gastrointestinal tissue were harvested to quantify transcript abundance of markers of inflammation, including tumor necrosis factor alpha (TNF α), interleukin (IL) 6, IL-8, IL-1 β , intercellular adhesion molecule-1 (ICAM-1), and nuclear factor kappa B (NF κ B). Statistical analysis was conducted using the Mixed Procedure of SAS. There was no colostrum management \times NSAID administration ($P \geq 0.58$) for plasma cortisol and TBARS concentrations. However, feeding colostrum compared to milk replacer led to a decrease ($P < 0.01$) and a tendency for a decrease ($P = 0.06$) in plasma cortisol and TBARS concentrations, respectively. There was no colostrum management \times NSAID administration ($P \geq 0.25$) on rectal temperature and body weight. However, there was a tendency ($P = 0.06$) for colostrum management \times time on RT; it was greater at arrival than before transport and 36 h post transport in calves fed colostrum. Similarly, there was a colostrum management \times time ($P = 0.04$) for BW, which was lower at arrival and 36 h after arrival compared to prior to transportation for calves fed colostrum. There was no colostrum management \times NSAID administration interaction ($P \geq 0.15$) and NSAID administration effect ($P \geq 0.13$) on transcript abundance of markers of inflammation measured. However, feeding colostrum downregulated ($P \leq 0.02$) several markers of inflammation in the liver (TNF α , IL-6, IL-8, IL-1 β , and ICAM-1), rumen (IL-6 and ICAM-1), and jejunum (IL-6). In summary, feeding colostrum resulted in a decrease in the plasma concentration of indicators of stress, and downregulated gene expression for markers of inflammation in different organs, including the liver. However, pre-transport NSAID administration had no detectable effect on all measurements. This suggests that proper colostrum management is key to limiting the negative impact of transport-related stress on health outcomes in pre-weaned calves.

Key words: colostrum management, non-steroidal anti-inflammatory drug administration, pre-weaned calves, transport related stress

Evaluation of Growth, Meat Quality, and Sensory Characteristics of Wool, Hair and Composite lambs

M.L. Heimbuch*, J.B. Van Buren*, B.S. Epperson*, O.F. Kayleen*, S.M. Jepson*, J.A. Nasados*, D.A. Vinci†, W.J. Price*, K.R. Vierck^, D.E. Konetchy*, P.D. Bass*, and M.J. Colle*.

*University of Idaho, Department of Animal, Veterinary & Food Sciences, Moscow, ID 83844

†University of Idaho, Palouse Research, Extension and Education Center, Moscow, ID 83844

^University of Arkansas, Department of Animal Science, Fayetteville, AR 72701

The objective of this study is to compare the growth rate, shoulder height, heart girth circumference, and carcass characteristics of wool, hair, and composite lambs. Twenty-seven lambs were purchased at weaning (~70 days of age). The wool lambs (Suffolk × Polypay/Targhee; n = 9) and composite lambs (Dorper × Polypay/Targhee; n = 9) were purchased from the UI Sheep Center, while the hair lambs (Dorper × Dorper; n = 9) were purchased from a local producer. Lambs were weighed and measurements of shoulder height (cm) and heart girth circumference (cm) were taken on day 0 and the two days prior to harvest. Hot carcass weight (HCW), back fat (BF), and rib eye area (REA) were collected at 48 h post-harvest. Data were analyzed using the MIXED procedure in SAS. Significance was determined at $P < 0.05$. There was not a significant difference among treatments for average daily gain ($P = 0.28$). In contrast, there were significant differences for finished weight ($P < 0.01$), shoulder height ($P < 0.01$), and hearth girth circumference ($P < 0.01$). Wool lambs were heavier at harvest and had greater shoulder height and heart girth circumference than composite and hair lambs whereas composite lambs had greater shoulder height than hair lambs. No difference in backfat was observed among treatments ($P = 0.13$). Wool lambs had greater HCW ($P < 0.01$) and REA ($P = 0.02$) than the composite and hair treatments. Wool lambs grew to heavier weights, were larger framed, and were also higher volume. Furthermore, wool lambs had heavier carcasses and were heavier muscled. Research will continue to evaluate sensory characteristics; we can, however, conclude the physical characteristics of the composite compare more closely to the hair treatment than the wool treatment.

The effect of feeding supplemental zeolite (clinoptilolite) of two different particle sizes on measures of nitrogen utilization and nutrient digestibility in finishing beef heifers

Cheyenne A. Myers^{*}, Mario E. de Haro Marti², Mireille Chahine³, and Gwinyai E. Chibisa^{*}

^{*}Department of Animal, Veterinary, and Food Science, University of Idaho, Moscow ID

²Department of Animal, Veterinary, and Food, University of Idaho Extension, Gooding, ID

³Department of Animal, Veterinary, and Food, Twin Falls Research and Extension Center University of Idaho, Twin Falls, ID

Clinoptilolite (CLN) could potentially improve nitrogen (N) utilization when fed to beef cattle as it can bind ruminal-ammonia-N ($\text{NH}_3\text{-N}$), limiting its loss and subsequent detoxification into urea-N, which is released into blood and is excreted in urine. However, the effectiveness of CLN is influenced by physical properties such as particle size. Although decreasing the particle size has been shown to increase the binding of ammonium *in-vitro*, this remains to be evaluated *in vivo*. Therefore, the objective of this study was to determine the effects of feeding CLN of two different particle sizes (30 and 40 μm) on ruminal $\text{NH}_3\text{-N}$ and plasma-urea-N (PUN) concentrations, ruminal pH, and nutrient intake and apparent total-tract digestibility. Six ruminally-cannulated beef heifers (mean initial BW \pm SD, 620.8 \pm 30.15) were used in a replicated 3 \times 3 Latin square design with 21 d periods (sample collection from d 15 to 21). Dietary treatments were 1) finishing ration with no supplement (CON), 2) CON +30- μm CLN (CL-30), and 3) CON + 40- μm CLN (CL-40). Clinoptilolite was top-dressed (2.5% of diet DM) during morning feeding. Intake was measured daily. Ruminal fluid was collected on d 19 for $\text{NH}_3\text{-N}$ analysis and blood was collected 3 h post-feeding on d 21 for PUN analysis. Indwelling pH loggers were used to measure ruminal pH (d 15 to 21) and grab fecal samples were collected from d 19 to 21 to determine total-tract nutrient digestibility. Statistical analysis was conducted using PROC MIXED in SAS. There was no treatment effect ($P \geq 0.13$) on ruminal $\text{NH}_3\text{-N}$ and PUN concentrations, ruminal pH, and nutrient (DM, OM, NDF, ADF and CP) intake and apparent total tract digestibility. In conclusion, feeding CLN to finishing heifers had no effect on measures of N utilization, ruminal pH and nutrient intake and apparent total-tract digestibility.

Key words: clinoptilolite, feedlot cattle, nitrogen utilization, nutrient digestibility

Impact of maternal nutrition on postnatal growth of crossbred beef steers

K.F. Oliver*, J.B. Van Buren*, J.B. Hall*†, M.L. Heimbuch*, S. Jepsen*, B. Epperson*, J.A. Nasados*, P.D. Bass*, and M.J. Colle*

*University of Idaho, Department of Animal, Veterinary & Food Sciences, Moscow, ID 83844

†University of Idaho, N.M. Cummings REC, Carmel, ID 83462

Maternal nutrition of beef cows is critical to programming the fetus for improved performance and meat quality. Cows pastured on range often have reduced forage quality compared to cows on irrigated pasture. Therefore, the objective of this study was to determine the effects of maternal nutrition on the subsequent growth and carcass characteristics of castrated male offspring from multiparous crossbred beef cows that were pastured on irrigated pasture (IRR) vs. rangeland (RAN) during early and mid-gestation. Twenty-four crossbred steers were weaned from their dams that were pastured on irrigated pasture (n = 12) or rangeland (n = 12) during early and mid-gestation. After weaning steers were placed on a backgrounding diet for four weeks, designed to gain 1.1 kg/d before being transitioned to a finishing ration. Steers remained on the finishing ration until an estimated backfat of 1.02 cm over the 12th and 13th rib was reached. Complete carcass data (skeletal and lean maturity, marbling score, quality grade, carcass weight, dressing percent, ribeye area, 12th rib fat thickness; percent kidney, pelvic, and heart fat; and yield grade) was collected and evaluated. Strip loin steaks were aged for four and fourteen days then assigned to Warner-Bratzler shear force (WBSF) for tenderness evaluation. Data were analyzed using the MIXED procedure in SAS. Significance was determined at $P < 0.05$. Hot carcass weight was heavier ($P = 0.02$) in RAN steers. Ribeye area trended towards significance ($P = 0.05$) for RAN compared to IRR steers, while yield grade ($P = 0.56$) and marbling score ($P = 0.94$) were not different between the two groups. For WBSF, there was not a treatment by aging period interaction ($P = 0.54$) or treatment difference ($P = 0.25$); however, steaks became more tender from day 4 to 14 ($P = 0.0005$). These initial findings suggest RAN steers are exhibiting compensatory growth, yielding heavier carcasses, and have comparable tenderness relative to that for IRR steers. Understanding the impact of maternal environment on steer performance will provide an opportunity for producers to improve profitability and the industry to produce more acceptable products for consumer consumption.

Key words: fetal programming, growth, carcass, beef

The effects of allyl isothiocyanate inclusion as an additive on whole-plant corn silage

Lucelia de M. Pereira¹, Pedram Rezamand², Bruna C. Agostinho², Gabriela L. D. Vigne¹, Denise Volpi¹, Natália N. de Mello¹, Queila G. Tavares¹, Patrick Schmid¹, Maity Zopollatto¹

¹Department of Animal Science, Federal University of Parana, Curitiba, Brazil

²Department of Animal, Veterinary & Food Sciences, University of Idaho, Moscow, ID

Allyl Isothiocyanate (AIT) is a natural compound used as a food additive. This additive is a well-recognized antimicrobial agent that plays an important role in mitigating the growth of microorganisms that cause food spoilage. Undesirable microorganisms can proliferate during the ensiling, storage, and aerobic phase of silage, reducing nutritional quality and increasing the fermentative losses. Therefore, the objective of this study was to determine the effect of AIT inclusion on the fermentative losses, microbiology, and aerobic stability of whole-plant corn silage. Four AIT levels were tested in a completely randomized block design: 0, 5, 10, 20 mg/kg of fresh matter of whole-plant corn, with five replicates per treatment, totaling 20 experimental units. The AIT was applied and mixed with the material at ensiling. Each experimental unit consisted of one 8-L plastic bucket with an average density of 468 kg/m³. The silos were opened 90 days after ensiling. Data were analyzed using a MIXED model procedure of SAS with significance declared at $P \leq 0.05$ and the tendency at $P < 0.10$. The gas production, total dry matter (DM) losses, and molds decreased linearly ($P < 0.01$; 0.01; 0.03, respectively) with the AIT inclusion levels. Although aerobic stability linearly increased ($P = 0.02$), DM losses showed a quadratic increase ($P = 0.02$) with the AIT levels during aerobic deterioration. Furthermore, pH and heterolactic account tended to linearly decrease ($P = 0.06$ for both), whereas DM concentration and yeast account tended to linearly increase ($P = 0.09$; 0.08, respectively) with AIT levels. Effluent production and homolactic account were not however affected by treatment. Overall, the inclusion of AIT at ensiling affected the fermentative losses, microbiology, and aerobic stability of whole-plant corn silage.

Key words: aerobic stability, essential oil, silage fermentation,

Impacts of heifer post-weaning intake classification on performance measurements of lactating and non-lactating two-, five- and eight-year-old Angus beef females

K. R. Wellnitz,[†] C. T. Parsons,* J. M. Dafoe,* S. A. Wyffels,* D. L. Boss,* T. DelCurto,[†] and M. L. Van Emon[†]

[†]Department of Animal and Range Sciences, Montana State University, Bozeman, MT 59717

*Northern Agriculture Research Center, Montana State University, Havre, MT 59501

ABSTRACT: Data used in these studies were part of a larger project as described by Parsons et al., (2021). These studies evaluated heifer postweaning intake classification on performance measurements of two-, five- and eight-year-old lactating or non-lactating Angus beef females. We analyzed the intake and production data of fifty-seven pregnant, non-lactating (Study 1) and fifty-four, lactating, non-pregnant (Study 2) females. Heifer postweaning intake was calculated over 80 test days following weaning from the dam using GrowSafe units. Heifers were categorized based on intake as either low (< -0.05 SD from the mean), average (± 0.05 SD from the mean), or high (> 0.50 SD from the mean) within year. The non-lactating females (Study 1) showed an age effect ($P \leq 0.05$) for cow body weight (BW), DMI rate (grams/minute), and time spent at the feeder (minutes/day). As cow age increased, cow body weight also increased. In addition, intake rate was greater in five- and eight-year-old cows when compared to two-year-old cows, and eight-year-old cows spent more time at the feeder than two- and five-year-old cows. Cow BW for non-lactating cows was significant for age ($P < 0.001$), intake classification ($P = 0.03$) and showed a tendency for age*intake interaction ($P = 0.10$), with older cows weighing more than younger cows. In lactating cows (Study 2), Julian birth date of calves showed an age*intake interaction ($P < 0.001$) with two-year-old cows calving earlier in the calving season than five- and eight-year-old cows. Calf birth weights differed by age classification ($P < 0.001$) and showed an age*intake classification ($P = 0.001$) with offspring from eight-year-old cows having heavier birth weights than two- and five-year-old cows, however, an intake effect was not observed ($P = 0.95$). As expected, post-partum interval was greater for 2-year-old cow when compared to five- and eight-year-old cows ($P < 0.001$). Milk production expressed as kilograms and grams per kilogram of BW of the cow had an age*intake ($P < 0.001$) effect. Two-year-old cows with low and average intake classifications had greater daily milk production and milk produced as grams per kilogram of BW than two-year-old cows with high intake classification. Additionally, five-year-old cows with average and high intake classifications had greater daily milk production and grams of milk produced per kilogram of BW compared to five-year-old cows classified as low intake. There was no effect of intake classification ($P \geq 0.56$) for lactating females on DMI/kg of BW, DMI rate (grams/minute), coefficient of variation for intake, or time spent at the feeder (minutes/day). In summary, heifer post-weaning intake classification had minor impacts on beef female performance measurements in lactating and non-lactating commercial beef females.

Key words: beef cattle, heifer, intake, lactating, non-lactating, post-weaning