# PACIFIC NORTHWEST ANIMAL NUTRITION CONFERENCE

2023

## **PROCEEDING**



## **Maximizing Microbial Protein with Branched-Chain Volatile Fatty Acids**

Daryl Kleinschmit, Mike Socha, Arturo Gomez, Adam Geiger, Jeff Weyers, Raphael Chan, and Huw McConochie

Zinpro Corporation, Eden Prairie, MN

#### Introduction

In recent years, margins across the dairy industry have evolved to a point where they are routinely tight and oftentimes negative. For example, since 2000 income over feed costs has averaged only \$5.26/cwt and farm profitability was actually -\$3.04/cwt when accounting for all costs (Brouk, 2020). Feed costs comprise approximately 60% of the operating costs (Brouk, 2020) so it is no surprise that dairy producers and nutritionists focus on ways to control this major input cost while striving to maintain production when margins are unfavorable.

Nitrogen conversion to milk protein is extremely inefficient in dairy cattle. Huhtanen and Hristov (2009) found milk N efficiency averages only 24.7% (range = 14 to 41.1%) across 1,737 research diets from North America and Northern Europe. This is worth noting because protein supplementation also tends to be the most expensive component in a typical dairy ration. Furthermore, animal performance may be impacted as energy is required to convert excess ammonia to urea in the liver (12 kcal/g of N; Tyrrell et al., 1970).

The dairy industry has been under increasing pressure to reduce N excretion in both urine and feces as they are potential pollutants for water sources and air (gaseous N emissions). Urinary N excretion is of particular concern (Hristov et al., 2019). Localized regulations, particularly near watersheds and urban populations, will continue to become more restrictive and will have an impact on nutritional practices on dairies. Huhtanen and Hristov (2009) concluded that reducing dietary protein was the most efficient tool to reduce N losses from dairy operations.

Rumen degradable protein (RDP) typically comprises 60 to 67% of dietary protein and is converted to microbial protein (MicP), a highly digestible protein with an amino acid (AA) profile that closely matches the needs for milk protein synthesis. Unfortunately, MicP typically only provides approximately 50% of the metabolizable protein (MP) supplied to the dairy cow so part of the strategy to reduce protein supplementation is to maximize MicP yield and efficiency. Mixed rumen microbes have a growth efficiency of only 1/3 to 2/3 of their theoretical maximum (Hackmann and Firkins, 2015) so there are opportunities to improve this efficiency with the right nutritional strategies.

#### Why Focus on Microbial Protein?

Even though the rumen environment has some inefficiencies, ruminants do have some advantages over monogastric animals when microbial growth is maximized. Most notable, ruminants can utilize forages as an energy source via microbial fermentation of carbohydrates and conversion to volatile fatty acids (VFAs; acetate, propionate, and butyrate). These VFAs are used for energy by the animal. Second, the rumen microbes can detoxify mycotoxins and lessen their effect on the animal (Upadhaya, et al., 2010). Next, rumen microbes increase P availability due to endogenous production of phytase (Yanke et al., 1998). The rumen microbes are also major contributors to b-vitamins supply (Schwab et al., 2006), reducing the need to supplement relative to monogastric animals. Lastly, microbes can convert lower cost non-protein nitrogen (NPN) sources, like urea, to a superior source of MP thereby reducing protein supplementation costs.

Maximizing production in dairy cattle requires one to supply the correct array of AA via MP. For example, a 1500 lb Holstein cow producing 95 lbs of milk (3.8% fat and 3.2% true protein) in her second lactation requires over 3000 grams (6.6 lbs) of MP per day to sustain this level of production (Van Amburgh et al., 2015). In order to supply this copious amount of MP, one needs to prioritize maximizing MicP yield in the rumen and then correct the remaining MP deficit with ruminally-undegradable protein (RUP).

It should be noted that meeting the MP needs of the cow does not necessarily mean that her AA needs are being met. The true MP requirement of the animal is dependent upon the AA profile. The amount of protein fed in the diet can be lowered if the MP has an AA profile that is more reflective of the animal's AA requirements. As noted previously, MicP is a highly digestible protein source that has the most favorable AA profile for meeting the needs for milk protein synthesis relative to commonly used protein sources (Schingoethe, 1996). Thus, neglecting the RDP needs of cattle in favor of over-supplementing RUP may increase the amount of dietary protein that needs to be fed, especially if the RUP source is of poor or variable quality. Santos et al. (1998) summarized 88 lactation trials with 127 direct comparisons in which soybean meal, was replaced with protein sources containing high concentrations of RUP. In that review, only 17% of the comparisons showed that added RUP increased milk yield. This finding was attributed to: 1) poor essential AA profile of the RUP source, 2) low digestibility of the RUP source in the small intestine, 3) control diets may have had sufficient RUP, and 4) a decrease in rumen MicP synthesis.

Santos et al. (1998) noted that in 29 comparisons from 15 metabolism trials, in which soybean meal was replaced by a RUP supplement, MicP synthesis decreased in 76% of the comparisons. This finding shows that supplementing RUP at the expense RDP is a further hindrance to meeting the MP needs of dairy cattle. Furthermore, several studies have demonstrated that commonly used RUP sources, including blood meal (Paz et al., 2014), distillers grains

(Kleinschmit et al., 2006; Li et al., 2012), meat and bone meal and corn gluten meal (Maiga et al., 1996) can be variable in RUP content and digestibility. In addition to having a superior AA profile, rumen microbes have been estimated to be comprised of 82.4% true protein (Sok et al., 2017) and to be 90% digestible (Sniffen et al., 1992).

Supplementing dairy diets with rumen-protected AA (RPAA) has become a widely accepted practice as nutritional models have become more accurate in predicting dietary AA requirements and supply. Balancing with RPAA have allowed nutritionists to lower CP content of diets (<16.5%) thus lowering N excretion and improving N efficiency (Broderick et al., 2008; Broderick et al., 2009). The quality of many of these products are quite good and more predictable in nutrient content relative to other RUP sources. However, these products are costly, and are often removed from lactating diets when the price for milk protein drops. Improving microbial yield and efficiency can reduce, though not necessarily eliminate, one's reliance on RUP and RPAA sources.

### **Branched-Chain Volatile Fatty Acids (BCVFA)**

Cellulolytic bacteria (fiber digesters; Fibrobacter succinogenes, Ruminococcus albus, and Ruminococcus flavefaciens) require BCVFA (isovalerate, isobutyrate, and 2-methylbutyrate; Figure 1), but these VFAs are often limiting in the rumen. BCVFA are derived from branched-chain amino acids (BCAA; valine, leucine, and isoleucine) within the RDP fraction that have been decarboxylated and deaminated in the rumen. The first function of BCVFA in cellulolytic bacteria is the formation of MicP. Cellulolytic bacteria require BCVFA and ammonia for de novo synthesis of BCAA. In vitro research has shown that supplementing BCVFA increased ruminal MicP synthesis (Cummins and Papas, 1985; Russell and Sniffen, 1984). Gorosito et al. (1985) showed that supplementing wheat straw cell wall with individual or a combination of BCVFAs in a batch culture system improved cell wall digestion and lowered ammonia -N concentration, which indicated improved N uptake by rumen microbes. More recently, Roman-Garcia et al. (2019b) found, within an in vitro batch culture system, that rumen bacteria preferentially utilized BCVFA compared to BCAA and observed a corresponding reduction in ammonia-N concentrations and improved NDF digestibility.

BCVFA are also required by cellulolytic bacteria to synthesize branched-chain long chain fatty acids (BLFA). BLFA and odd-chain fatty acids provide fluidity (Figure 2) to the lipid membrane of the bacterial cell for adaption to environmental changes (Russell, 2002). Roman-Garcia et al. (2019d) demonstrated that a higher rumen pH (6.3 to 6.8 vs. 5.7 to 6.2) increased the requirement for 2-methylbutyrate. Furthermore, Roman-Garcia et al. (2019c) showed that microbes preferentially utilized BCVFA vs BCAA for fatty acid elongation.

Dietary factors affect the need for supplemental BCVFA. Cellulolytic bacteria have a disadvantage to amylolytic microbes (starch and sugar digesters) because amylolytic bacteria

have a faster rate of growth and preferentially utilize BCVFA over other de novo methods for BCAA synthesis (Allison et al., 1984). As a result, a diet containing high levels of rumen fermentable carbohydrates are often limiting in BCVFA for cellulolytic bacteria. DeFrain et al. (2004) observed a reduction in ruminal molar proportions of BCVFA as increasing levels of lactose was added to lactating dairy diets. Similarly, Oba and Allen (2003) found that feeding a high starch diet (31.6%) versus a low starch diet (21.2%) lowered ruminal concentrations of isovalerate and isobutyrate.

Scenarios that increase ruminal passage rate may reflect a need for BCVFA supplementation. It is not uncommon to observe lactating dairy cows consume >60 lbs of dry matter each day. Increased dry matter intake corresponds with greater ruminal passage rate of feed, which lowers potential digestibility of RDP and availability of BCAA to be converted to BCVFA. Roman-Garcia et al. (2019d) showed a greater need for 2-methylbutyrate to support NDF digestibility when passage rate in a continuous culture system was increased from 2.5 to 5%/h.

Diets with adequate to excessive RDP may not limit the supply of BCVFA. Mir et al. (1986) evaluated the effect of each individual BCAA, the corresponding BCVFA + urea, combination of all three BCAA, or all three BCVFA + urea on the DM digestibility of barley straw or alfalfa hay with in vitro batch culture. Interestingly, DM digestibility of alfalfa hay was not improved with any of the treatments. In contrast, all treatments, with the exceptions of valine and leucine, improved the DM digestibility of barley straw. The authors concluded that the lack of response in alfalfa hay was due to this substrate providing adequate RDP (BCAA) to rumen microbes. More recently, Roman-Garcia (2019a) demonstrated that supplementing various combinations of BCVFA did not improve in vitro NDF digestibility when the substrate contained ground corn and alfa lfa hay (high protein). However, NDF digestibility was improved when these same treatments were applied to ground corn and orchard grass (low protein).

### **Supplementation of Branched-Chain Volatile Fatty Acids**

BCVFA have been shown to increase MicP synthesis (Cummins and Papas, 1985; Russell and Sniffen, 1984) and reduce rumen ammonia -N concentrations (Gorosito et al., 1985; Roman-Garcia et al., 2019b). Thus, supplementing BCVFA in conjunction with an NPN source, like urea, may potentially replace a portion of the supplemental protein (soybean meal, canola meal, etc.), reducing feed costs. This strategy also shows promise at lowering dietary CP levels and improving N efficiency. Felix et al. (1980) observed a reduction in milk yield when urea replaced soybean meal in a lactating dairy cow diet. However, a portion of the milk was recovered when BCVFA were added to the urea -based diet.

Several lactating studies have shown the positive benefits of supplementing lactating dairy cattle diets with BCVFA. Felix et al. (1980) found a 5.9% improvement in milk yield when supplementing a 14% CP diet with BCVFA. In addition, this study showed a 11.3% reduction in plasma urea -N and a 22.7% reduction in rumen ammonia-N. Similarly, a three-university trial demonstrated a 12.7% increase in milk yield across the entire 305 -d lactation period (Papas et al., 1984). Peirce-Sandner et al. (1985) also showed that providing BCVFA increased yields of milk and 4% fat-corrected milk by 6.8 and 7.1%, respectively.

Best responses may be dependent on dietary factors, as previously discussed. Otterby et al. (1990) summarized a four-university trial that evaluated BCVFA being fed throughout the entire lactation. An interesting finding with this summary was that the best response in yield of milk, fat, and protein was observed in cows during the late lactation (225 – 305 days in milk) when corn silage-based diets contained less than 14% CP. Similar findings were observed by Peirce-Sandner et al. (1985), in which the best response was observed when diets contained 15% (midlactation) vs. 16.8% CP (early-lactation).

Nutritional models, such as the NRC (2001) and the CNCPS (Van Amburgh et al., 2015) have improved the precision of ration balancing. As we look to the next step of ration balancing, perhaps balancing for the needs of BCVFA by the rumen microbes is logical. Roman-Garcia et al. (2019e) conducted a meta-analysis on milk production responses to BCAA and found isoleucine to be the most important BCAA for milk production. Isoleucine is the corresponding BCAA to 2-methylbutyrate. Interestingly, Roman-Garcia (2019d) found this BCVFA to be most critical for supporting NDF digestibility in high passage-rate scenarios, similar to what is seen with high producing dairy cattle and is utilized more heavily in higher pH environments. In dairy diets, one of the objectives is to maintain a rumen pH above 6 to maximize NDF digestibility.

Roman-Garcia (2019a) concluded that best responses in NDF digestibility was achieved with isobutyrate and 2-methylbutyrate together or when all three BCVFA were fed. Isovalerate, which is derived from leucine, may not need to be supplemented in many of the dairy diets fed in North America. Dairy cattle diets in North America are largely based on corn grain, silage, and coproducts containing high concentrations of leucine. If one can better predict the requirements of individual BCVFA, supplementation may be achieved more economically, and allows one to better predict likelihood of response.

#### Conclusion

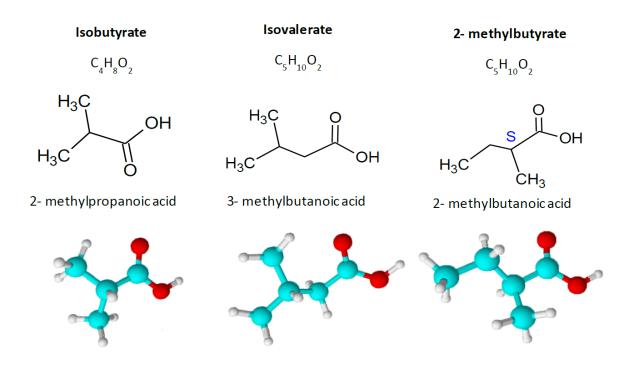
In conclusion, dairy producers have both an economical and environmental incentive to improve N efficiency. Much of the dairy industry's focus on improving N efficiency has been on providing higher quality RUP and RPAA. This strategy has been effective a s it is common to have diets with < 16.5% CP and it wasn't that long ago, that 18% CP was more common. The next stage in improving N efficiency is to maximize rumen microbial efficiency. The supplementation of BCVFA is an effective tool to achieve this objective that, together with the adoption of sophisticated dairy ration software, shows promise in meeting the needs of predicted BCVFA for rumen microbes, making this strategy profitable for dairy producers.

#### References

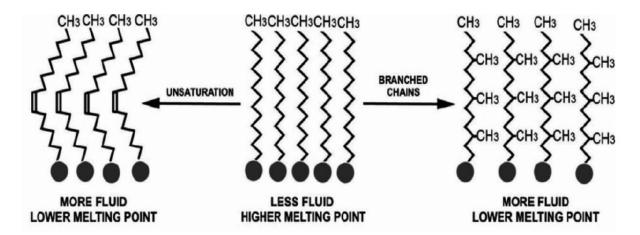
- Allison, M. J., A. L. Baetz, and J. Wiegel. 1984. Alternative pathways for biosynthesis of leucine and other amino acids in Bacteroides ruminicola and Bacteroides fragilis. Appl. Environ. Microbiol. 48:1111-1117
- Broderick, G. A., M. J. Stevenson, R. A. Patton, N. E. Lobos, and J. J. Olmos Colmenero. 2008. Effect of supplementing rumen-protected methionine on production and nitrogen excretion in lactating dairy cows. J. Dairy Sci. 91:1092-1102
- Broderick, G. A., M J. Stevenson, and R. A. Patton. 2009. Effect of dietary protein concentration and degradability on response to rumen-protected methionine in lactating dairy cows. J. Dairy Sci. 92:2719-2728
- Brouk, M. J. 2000. Using whole farm efficiencies to discover increased milk margin despite challenging economic times. Pages 38 43 in Proc. 2020 Southwest Nutrition Conference. Chandler, AZ
- Cummins, K. A. and A. H. Papas. 1985. Effect of isocarbon-4 and isocarbon-5 volatile fatty acids on microbial protein synthesis and dry matter digestibility in vitro. J. Dairy Sci. 68:2588-2595
- DeFrain, J. M., A. R. Hippen, K. F. Kalscheur, and D. J. Schingoethe. 2004. Feeding lactose increases ruminal butyrate and plasma β-hydroxybutyrate in lactating dairy cows. J. Dairy Sci. 87:2486-2494
- Devi Upadhaya, S., M. A. Park, and J. K. Ha. 2010. Mycotoxins and their biotransformation in the rumen: A review. Asian-Aust. J. Anim. Sci. 23:1250-1260
- Felix, A., R. M. Cook, and J. T. Huber. 1980. Isoacids and urea as a protein supplement for lactating cows fed corn silage. J. Dairy Sci. 63:1098-1103.
- Gorosito, A. R., J. B. Russell, and P. J. Van Soest. 1985. Effect of carbon-4 and carbon-5 volatile fatty acids on digestion of plant cell wall in vitro. J. Dairy Sci. 68:840-847.
- Hackmann, T. J. and J. L. Firkins. 2015. Maximizing efficiency of rumen microbial protein production. Frontiers in Microbiology. 6:1-17
- Hristov, A. N., A. Bannink, L. A. Crompton, P. Huhtanen, M. Kreuzer, M. McGee, P. Nozièr, C. K. Reynolds, A. R. Bayat, D. R. Yáñez-Ruiz, J. Dijkstra, E. Kebreab, A. Schwarm, K. J. Shingfield, and Z. Yu. 2019.
  Invited review. Nitrogen in ruminant nutrition: A review of measurement techniques. J. Dairy Sci. 102:5811-5852
- Huhtanen, P. and A. N. Hristov. 2009. A meta -analysis of the effects of dietary protein concentration and degradability on milk protein yield and milk N efficiency in dairy cows. J. Dairy Sci. 93:3222-3232
- Kleinschmit, D. H., J. L. Anderson, D. J. Schingoethe, K. F. Kalscheur, and A. R. Hippen. 2007. Ruminal and intestinal degradability of distillers grains plus solubles varies by source. J. Dairy Sci. 90:2909 2918
- Li, C. J. Q. Li, W. Z. Yang, and K. A. Beauchemin. 2012. Ruminal and intestinal amino acid digestion of distiller's grain vary with grain source and milling process. Anim Feed Sci Tech. 175:121-130
- Maiga, H. A, D. J. Schingoethe, and J. Ellison Henson. 1996. Ruminal degradation, amino acid composition, and intestinal digestibility of the residual components of five protein supplements. J. Dairy Sci. J. Dairy Sci. 79:1647 -1653
- Mir, P. S., Mir, Z., and J. A. Robertson. 1986. Effect of branched-chain amino acids or fatty acid supplementation on in vitro digestibility of barley straw or alfalfa hay. Can. J. Anim Sci. 66:151-156.
- National Research Council. 2001. Nutrient Requirements of Dairy Cattle. 7th rev. ed. Natl. Acad. Sci., Washington, D.C.
- Oba, M. and M. S. Allen. 2003. Effects of corn grain conservation method on feeding behavior and productivity of lactating dairy cows at two dietary starch concentrations. J. Dairy Sci. 86:174-183.
- Otterby, D. E., D. G. Johnson, R. Towns, R. M. Cook, R. A. Erdman, H. H. Van Horn, J. A. Rogers, and W. A. Clark. 1990. Dose response of dairy cows to ammonium salts of volatile fatty acids. J. Dairy Sci. 73:2168-2178.

- Papas, A. M., S. R. Ames, R. M. Cook, C. J. Sniffen, C. E. Polan, and L. Chase. 1984. Production responses of dairy cows fed diets supplemented with ammonium salts of iso C-4 and C-5 acids. J. Dairy Sci. 67:276-293.
- Paz, H. A., T. J. Klopfenstein, D. Hostetler, S. C. Fernando, E. Castillo-Lopez, and P. J. Kononoff. 2014. Ruminal degradation and intestinal digestibility of protein and amino acids in high-protein feedstuffs commonly used in dairy diets. J. Dairy Sci. 97:6485-6498
- Peirce-Sandner, S. B., A. M. Papas, J. A. Rogers, T. F. Sweeney, K. A. Cummins, H. R. Conrad, and L. D. Muller. 1985. Supplementation of dairy cow diets with ammonium salts of volatile fatty acids. J. Dairy Sci. 68:2895-2907.
- Roman-Garcia, Y., B. L. Denton, C. Lee, M. Socha, and J. L. Firkins. 2019a. Assessing different branched chain VFA combinations on NDF degradation and VFA production in vitro. J. Dairy Sci. 102 (Suppl. 1):409
- Roman-Garcia, Y., B. L. Denton, C. Lee, M. Socha, and J. L. Firkins. 2019b. Effects of branched-chain VFA and branched-chain AA supplementation on NDF degradation and VFA production in vitro. J. Dairy Sci. 102 (Suppl. 1):409
- Roman-Garcia, Y., B. L. Denton, K. E. Mitchell, C. Lee, M. Socha, and J. L. Firkins. 2019c. Assessing recovery of 13C-enriched branched-chain VFA and AA (BCAA) into rumen bacterial fatty acids. J. Dairy Sci. 102 (Suppl. 1):366
- Roman-Garcia, Y., B. L. Denton, K. E. Mitchell, C. Lee, M. Socha, and J. L. Firkins. 2019d. Relation of branched-chain VFA supplementation with solids passage rate and pH on NDF degradation and microbial function in continuous culture. J. Dairy Sci. 102 (Suppl. 1):410
- Roman-Garcia, Y., L. E. Moraes, M. Socha, and J. L. Firkins. 2019e. Quantifying the relation between diet branched-chain AA and production responses. A meta-analysis. J. Dairy Sci. 102 (Suppl. 1):164
- Russell, J. B. 2002. Page 37 in Rumen Microbiology and Its Role in Ruminant Nutrition.
- Russell, J. B. and C. J. Sniffen. 1984. Effect of carbon-4 and carbon-5 volatile fatty acids on growth of mixed rumen bacteria in vitro. J. Dairy Sci. 67:987-994
- Schingoethe, D. J. 1996. Balancing the amino acid needs of the dairy cow. Anim Feed Tech. 60:153 -160 Schwab, E. C., C. G. Schwab, R. D. Shaver, C. L. Girard, D. E. Putnam, and N. L. Whitehouse. 2006. Dietary forage and nonfiber carbohydrate contents influence b-vitamin intake, duodenal flow, and apparent ruminal synthesis in lactating dairy cows. J. Da iry Sci. 89:174-187
- Sniffen, C. J., J. D. O'Connor, P. J. Van Soest, D. G. Fox, and J. B. Russell. 1992. A net carbohydrate and protein system for evaluating cattle diets: II. carbohydrate and protein availability. J. Anim. Sci. 70:3562 -3577
- Sok, M., D. R. Ouellet, J. L. Firkins, D. Pellerin, and H. Lapierre. 2017. Amino acid composition of rumen bacteria and protozoa in cattle. J. Dairy Sci. 100:5241-5249
- Tyrrell, H. F., P. W. Moe, and W. P. Flatt. 1970. Influence of excess protein intake on energy metabolism of the dairy cow. Pages 69-72 in Proc. 5th Symp. on Energy Metabolism, Vitznau, Switzerland.
- Van Amburgh, M. E., E. A. Collao-Saenz, R. J. Higgs, D. A. Ross, E. B. Recktenwald, E. Raffrenato, L. E. Chase, T. R. Overton, J. K. Mills, and A. Foskolos. 2015. The Cornell Net Carbohydrate and Protein System: Updates to the model and evaluation of version 6.5. J. Dairy Sci. 98:6361-6380

Figure 1. Molecular structure of branched-chain volatile fatty acids.



**Figure 2.** Diagram showing change of membrane fluidity by incorporation of unsaturated and branched-chain long chain fatty acids.



## **Zinpro: Challenging Trace Mineral Dogma**

Chris Reinhardt

Zinpro Corporation

#### Abstract

#### Zinc: Immune secrets revealed

Zinc is emerging as a giant among trace minerals with specific regard to immune function. Zinc has been linked as an essential co-factor in over 400 enzyme systems throughout the mammalian body; many of these are key to fighting viral and bacterial pathogenic infection. The immune system can be thought of like a medieval castle: the moat and curtain wall are analogous to the physical barriers to penetration such as the skin, epithelium, and muco-cilliary apparatus; the archers atop the curtain wall are the Th1 lymphocytes and the downstream cytokines they signal for the destruction of invading virus; the central keep of the castle is analogous to the antibody response to the invading pathogens. The central keep is vital to a sustained defense; however, if the invaders have made it to the keep, a great deal of damage has already been done to the castle and its defenders. One key effective immunity is to maximize "the archers on the wall"---the Th1 lymphocytes. Getting more zinc into the cells of the tissues important to the immune system will dramatically increase the ability of the animal to defend itself in the immediate term. The Zinpro® trace mineral-amino acid complex has been shown, in all livestock species, to enhance immune function and improve animal performance.

## The chemistry of Zinpro. Why it matters

Peter Stark, PhD

VP of Product Development at Zinpro

#### Zinpro Advantage

My presentation will discuss three areas, what is unique about the Zinpro complex, how we evaluate organic trace minerals from a chemical standpoint and review various OTM's using some of this methodology. The overall theme is from a medicinal chemistry standpoint. I will be delving into chemistry properties, uptake, absorption and metabolism. Zinpro forms a complex with amino acids. This complex achieves all the key features required for a successful OTM. It is soluble, stable at stomach pH and taken up from the intestine as the complex. Once our mineral is absorbed into circulation our minerals are still in a different form that allows for better utilization and a longer time in circulation. All of these factors lead to Zinpro performance minerals superior animal performance. I will also explore what is important in the design of an OTM to give the audience better tools to differentiate what is real and what in not in trying to differentiate the various options. Key factors are solubility of the mineral and ligand (together), the stoichiometry of the metal and ligand, the stability of the metal and ligand and the route of uptake, absorption and utilization of the metal. I will then use these criteria to discuss the problems associated with some of the OTM's on the market. For example, proteinates are mainly inorganic metal and what little metal is bound to protein/peptide will dissociate during the digestion process. Acids such as propionic and hydroxy acids have too weak of a bonding to survive the stomach acid as will become ionic inorganic metal during digestion. Glycinates form a good bond and are soluble but since there is no side chain for bonding recognition with the amino acid transporters it has poor uptake in a competitive environment. These concepts will be illustrated with experimental data. Taken in totality it will demonstrate clearly the Zinpro advantage.

## **Trace Mineral Needs of Modern Beef and Dairy Cattle**

Stephanie L. Hansen

Department of Animal Science Iowa State University Ames, IA

#### **Summary**

Genetic selection, growth promoting technologies and improved animal husbandry practices have led to cattle that have tremendous capacity to produce meat and milk. Many of the trace mineral recommendations are based on data decades old, conducted with cattle of much lower production potential. This may explain why some nutritionists ignore recommendations and feed two to three times as much trace mineral. We will consider Zn and Cu as examples of why this may, or may not be, a good practice. Trace minerals are involved in nearly every biological process in the body and optimizing trace mineral requirements may support cattle immune function, recovery from stressors such as transit, and help maintain a positive eating experience for the consumer by maintaining or improving meat quality. Examples of each will be discussed.

## Health challenges of beef-on-dairy calves

Kip Lukasiewicz, DVM

#### Abstract

The use of beef sires in dairy herds has almost doubled from 2015 to 2019. Geiger (2020) reported a 128% increase in domestic semen sales of beef breeds in just 2 years, which has been accompanied by an improvement in beef genetics and provided the potential for better consistency across the population. However, creating a beef animal originating from a dedicated dairy operation has provided some significant challenges to managing animal health. The first 24-hour management of the calf at the dairy is critically important, as is management throughout the stages of calf development. Management of colostrum intake, hygiene, calf comfort, bedding, air quality, hydration, nutrition, appropriate vaccination, forage quantity and quality, optimal rumen development, transportation, and socialization all play a role in optimizing future feedlot health outcomes and ultimately, feedlot performance. The industry has learned a great deal from costly mistakes made early in this shift to beef-on-dairy calves. Some producers have made huge strides in calf management and have benefited from the improved health of their calves; other producers still have a ways to go in order to fully capitalize on the opportunities this new population can provide.

## Labor issues in animal agriculture

Corey Geiger
Managing Editor, Hoard's Dairyman
28 Milwaukee Avenue West
Fort Atkinson, WI 53538
cgeiger@hoards.com
(920) 650-0294

Animal agriculture had been facing labor challenges for years. Then the COVID-19 pandemic entered the scene, and labor pitfalls proliferated to levels not seen since the days of World War II. The options to overcome these challenges are relatively few: encourage more Americans to enter the workforce, encourage the entry of more seasonal workers, or open up immigration. In lieu of action in those areas, employers either must invest in technology or budget for higher wages to lure workers from another place of employment.

#### Let's call it "Food Security"

In recent years, a bill that has come to be known as the Farm Workforce Modernization Act has made its way through portions of Congress and then stalled out. That' because immigration reform has become a four letter word. To gain legislative traction on seasonal or immigrant workers, it might be better to call this matter, "Food Security."

Election year rhetoric in 2022, both on the campaign trail and political news channels, has only compounded the problem associated with the seasonal work and immigration matter. However, no amount of political posturing can remedy this national security matter — America's food security depends on a reliable supply of workers on our farms and in our processing plants. This growing crisis needs a solution.

Worker challenges loom large throughout all of agriculture. That's because declining fertility rates and aging populations leave many rural areas with a shortage of workers. Retirements in the Baby Boomer generation only compound the labor woes.

While all of agriculture has been impacted, ground zero definitely would be the dairy and meat sectors. That's because seasonal agricultural operations throughout the vegetable and fruit sector have legal access to H2-A visa workers. However, there isn't a "dairy" or "beef" season when compared to the likes of strawberries or cucumbers. Hence, dairy and livestock remain on the sidelines because they employ staff who must work throughout the year. With limited native-born options, the only logical answer is immigrants. And the legal pathways remain limited.

While immigration reform may be vilified among some politicians and their supporters, food security generally is not. At a recent town hall meeting, ranking House Committee member G.T. Thompson (R-Pa.), who will become the Chair of the House Agriculture Committee in the new 118<sup>th</sup> Congress, shared that he wants food security listed everywhere in future verbiage on the worker topic, and the word "immigration" should be struck from future legislation.

Those in animal agriculture would tend to agree with Congressman Thompson. That's because many of Thompson's conservative colleagues support legal immigration and fortifying

America's borders. However, those same conservatives know that America's food independence has been a major factor in this country's liberty and ability to lift up the cause of freedom across the globe for the past 247 years.

Aside from the worker shortage, some liberal-leaning politicians lay claim that agriculture's jobs underpay. That's simply not true. In the past three years, pay rates in the meat and dairy industries have outpaced wage growth when compared to all hourly workers, reported the American Immigration Council.

If Americans really want to rein in inflation, an ample supply of workers is a major part of the solution. That's the reality. It's also the reason wages in dairy and meat sectors have climbed nearly 34% in the past three years.

#### Is labor a national crisis?

This issue cascades far beyond the farm and has reached consumers in the form of food inflation. According to the report "Tending to America's Food Supply" by the American Immigration Council, meat and dairy prices have risen between 4.5% and 7% due to a lack of employees.

Immigrant workers have played an important and growing role in filling jobs in the meat and dairy industries. Of the 90,000 people the report classified as livestock workers – individuals who breed, raise, and care for animals – 20% were born outside of the U.S. This is higher than the 17.4% of foreign-born workers recorded when looking at all industries combined.

For the meat packing industry, the reliance on immigrant labor is even more pronounced. In 2020, 45.4% of workers in these facilities were foreign-born. The trend for using immigrant labor in the meat packing business traces back to the late 1800s, when the opening of new stockyards and packing plants attracted workers from Eastern Europe, the report shared.

When it comes to meat transportation, 26% of workers were born outside of the U.S. This number is higher than the 19.1% of immigrant transportation workers across all industries.

The need for foreign-born employees will only continue to grow, as the effect of an aging workforce is particularly noticeable in agriculture. In the next 10 years, it is estimated that nearly 30% of people working with livestock will reach the age of 65. According to the report, close to 90% of these workers are U.S. born. In contrast, of the livestock workers not reaching 65 years of age in the next decade, almost a quarter (24%) are foreign born.

Many sectors in agriculture utilize the H-2A and H-2B visa programs to hire seasonal foreign labor. In fiscal year 2021, the report noted that the US. Department of Labor certified 32,071 H-2A and H-2B workers for meat and dairy employers. These visas are only temporary, though, and often don't benefit farms that need year-long employees.

The "Tending to America's Food Supply" report reiterated that the labor shortage has led to rising production costs for farmers and employers and higher food prices for shoppers. "If the United States is to stabilize its food workforce – and thus stabilize prices – it must consider expanding temporary work visa programs and implementing other long-term reforms –

including providing a path to citizenship for many of the undocumented farmworkers in the U.S. today – to address the labor needs of the meat and dairy industries," the authors concluded.

#### The COVID-19 impact

While inflation began to ebb in November 2022, moving up just 0.1% when compared to October 2022, the labor market remains red hot. In short, strong job and wage growth indicates demand for workers still exceed supply. Employers added 263,000 jobs in November.

The worker matter is a participation problem. The participation rate actually ticked down by 0.1% from October to November 2022. That means that while job listings rose by 263,000 people, the number of Americans who want to work fell by 186,000.

The number of Baby Boomers in the workforce continues to tick downward, and this number was accelerated due to the pandemic. The bigger problem, however, is the share of men ages 25 to 54 in the workforce stands at 88.4%. These individuals typically represent prime workers. The employment rate in this category stood at 89.3% prior to COVID-19. Remember, one percentage point can represent hundreds of thousands of workers in a country with the world's third largest population. Certainly, the generous COVID-19 transfer payments coaxed some men ages 25 to 54 onto the sidelines. What will coax them back to the workforce?

#### For some dairy farms, overtime pay is closer than you think

Dairy farming is not a 40-hour per week job. With labor laws becoming more restrictive, though, employers may have to treat it as such. That will compound animal agriculture's labor woes.

During a Hoard's Dairyman DairyLivestream webcast focused on agricultural labor, agricultural workforce specialist Richard Stup of Cornell Cooperative Extension pointed out that although the vast majority of state laws mirror the federal laws regarding work week and minimum wage, if an individual state decides to enact stricter standards, that is what must be followed. Three top-10 dairy states are among those leading the charge with more protection for agricultural workers: California, New York, and Minnesota.

For example, as of last year, New York farm workers receive overtime pay (defined as time and a half) after 60 hours in a week. While Stup says that most farmers have found a reasonable way to handle that constraint, wage officials considered reducing that number even more. Due to the pandemic, that argument on changing the 60-hour-a-week metric was dropped for 2021.

"All of these changes in New York are the result of a 40-year political battle in the state, and they've really left the industry in an uncertain position with clearly higher labor costs than most other states and Canada, which is right next door," Stup added.

California dairy farmers must already deal with further limitations. In the effort to get agricultural workers down to a 40-hour work week by 2022, as of January 1, 2021, businesses with more than 26 employees had to pay overtime if an employee works more than 8.5 hours a day or 45 hours a week. For farms with fewer than 26 employees, the 40-hour work week will be implemented in 2025, and the phase-in period began in 2022 at 55 weekly hours.

Additionally, in November 2020, Washington state established overtime pay for agricultural employees past 40 hours in a week. "Washington state dairy is a special case. Their state supreme court ruled that the state's exemptions for farm labor were unconstitutional because dairy farm jobs are hazardous," Stup described. Farmers there are currently fighting against having to pay retroactive overtime pay going back years, he continued.

#### Handling the regulations

Overtime pay has its roots in the Great Depression. "The purpose of overtime as a government policy was to force employers to create more jobs and hire more of the people who had no work. It was a policy designed to create more jobs during a time of high unemployment," Stup explained. "This is in contrast with recent state actions to implement overtime in farm labor when unemployment was very low, and employees are hard to find."

Echoing that contradiction was Frank Cardoza, who manages a dairy in California and works with other farmers as a consultant. "Every January 1, you have to decide what changes you're going to make to be successful in this industry," he said. "We've looked at different options and we've tried different options as far as hiring more people and reducing hours, but in our case, it's better for us to just pay the overtime. The employee makes more money, so he seems to be happier."

Providing good jobs is clearly a priority at Cardoza's dairy. The labor advances that have been made have been good ones, he said, just as improvements have been made in animal welfare over the last decades.

However, overtime pay can create a financial burden and more challenges for farmers. There's an obvious cost to having to keep more employees.

"What's really complex about it is the dairyman is in the middle. We've got to take care of the cows, but we also have laws to protect people," Cardoza said. "It's a very difficult situation for a dairyman because he's got so many regulations, which we should have, but at the end of the day, he doesn't make any money, and it's tough."

Stup added that in the labor battles in New York, worker advocacy groups don't consider the economic argument as a legitimate part of the discussion "It's simply a justice issue," he said. "For them, economics simply do not matter.

"The challenge, of course, is that economics do matter," he emphasized. "If you can't run a business properly, you're not going to be there very long. The other piece of that is that the employment is tied to that business. If you can't run a profitable business, you won't have those jobs, and those jobs will disappear."

#### How do we move milk in this "expensive age"?

Just as producing quality milk requires attention to many different details, so does moving that milk from the farm to the customers that will process it. Between changes in client needs and capabilities as well as farm production fluctuations, it's the responsibility of the milk handler to add or cut loads, often on a daily basis and typically on short notice.

"While this has always been challenging, it's become increasingly difficult to manage over the last few years," described Corey Gillins, whose responsibilities as chief operating officer of Dairy Farmers of America's (DFA) Mountain Area Council include overseeing the co-op's transportation fleet.

Trucking costs and labor shortages have affected milk hauling in recent months and years like they have nearly every other farm and life input. The lack of truck drivers was just becoming an issue prior to the pandemic and has since intensified, Gillins noted, making moving perishable milk within and between markets limiting. Additionally, costs for new milk tankers, fuel, tires, labor, and parts are all up.

#### Some solutions

What is being done to combat these factors and ensure milk gets to the customers that need it? Gillins outlined the strategies his team and groups around the country are using in a Hoard's Dairyman DairyLivestream webcast.

At the top of the list is maintaining relationships and daily communication with customers and their broader network, Gillins said. This keeps clients aware of any transportation issues that arise and allows DFA to see where milk may need to be moved to or from. "We've found there is some inherent balancing capacity in just working within our network," he explained.

Another way they are able to balance milk and pivot to different customers is by using satellite transportation locations near a pocket of farms. Gillins shared an example where they have three large farms within 3 miles of each other that are 35 miles from three plants in one direction and 50 miles from two more plants in the other direction. They have begun basing trucks at the largest of these farms so milk can easily be transported in either direction based on needs.

"This approach also gives us the ability to stairstep milk to more distant markets using our own transportation when over-the-road haulers are unavailable," Gillins added. That's not an uncommon situation, and even though this region is using about 70% DFA-owned transportation, Gillins said contract hauler pay has increased as they have had to raise wages for their own drivers five times in the last couple of years to stay competitive. For their drivers, they also manage schedules to reach 80% of their legal hours so that the remaining hours can be used when routes change.

To reduce the total number of trucks and tankers needed, DFA has begun using 90,000-pound trailers in Colorado and part of Idaho. "Those larger units have allowed us to reduce our equipment needs and our driver needs by about 20%," Gillins said.

When those new trucks are added, though, others aren't sold off, as was standard before. Instead, they are kept as mobile storage capacity to hold milk through plant issues, holidays, or even weekends and then be delivered locally as space allows. Gillins said these tankers have given them storage for 15% to 18% of daily member production.

These practices help alleviate some of the transportation issues of moving milk to markets and offer some flexibility for milk handlers and buyers. Still, it's clear that as the processes continue to be bottlenecked and expensive, it will be a challenge to reach customers. "The lack of available transportation actually puts milk at risk of being dumped at times, leaving some available sales opportunities unfulfilled," Gillins recognized.

### How can we keep milk trucks moving?

They've slowed down since the height of the pandemic, but social media pictures of sparse grocery store dairy displays are still popping up, and they are all the evidence needed to know that, while milk is still flowing on the farm, transportation issues are sometimes preventing milk from reaching customers.

The reasons for these issues are generally the same as the reasons everything from farm supplies to exports are moving slower these days: labor shortages (of truck drivers and even at plants) and higher costs (for labor, fuel, parts, and trucks).

Truck driver shortages were becoming a concern even before the pandemic and have now exacerbated throughout the country. Roger Nordtvedt described that Southeast Milk Inc. has had many experienced drivers retiring without much availability to replace them. Working conditions play a part in that shortage as milk haulers must run 365 days a year.

"We see a lot of young drivers coming into us for maybe 6 months to a year to get some miles under them, then they leave us to go to the cross docks that work Monday through Friday with holidays off," said Nordtvedt, who recently retired from directing logistics and transportation for the Belleview, Fla.-based co-op. He noted driver pay has also shot up; it's not unusual for top drivers to be making six figures. A \$5,000 signing bonus, like Southeast Milk employed, looked enticing until another trucking company offered \$10,000.

Like DFA, Lone Star Milk Producers has also moved to using mostly its own trucks and only asking long haulers to supply the power unit. Jeff Sims said this has worked well for the co-op to move most of its milk with its own trailers. "It's extremely expensive, but it gives you some more flexibility," he said.

Another strategy that requires more investment from milk handlers but has been implemented to ease transportation challenges is drop and hook systems. This involves dropping off a full load near the processing plant and heading back out with an empty trailer without waiting for the first trailer to be unloaded. "It lets over-the-road guys do over-the-road stuff and lets short haul guys go between the drop yard and the plant," explained Sims.

Especially in the Southeast, drop and hook has helped reduce plant detention times. Having handlers carry a bigger inventory of trailers is more expensive, and trailers still need time to be washed, but it also allows for greater flexibility to keep trucks on the road. "Our experience has been good with drop and hook as a logistical tool," said Sims. "You almost start thinking about those drop yards as inventory management spots."

In Gillins' case, scheduled delivery times and slip seeding help manage inventory. They are also able to track delivery time accuracy and unloading efficiency and share that information with their plants to monitor performance.

"We feel like it's easier today to have access to equipment than it is to have access to drivers," he said. "So, we're trying to utilize our drivers the most efficiently we can and utilize equipment to be what's sitting at a plant."

#### We added technology instead of labor

Given the labor shortages and cost scenarios, dairy farmers have turned to technology to help bridge the gap. In a recent *Hoard's Dairyman* Round Table discussion, four dairy producers expounded on that situation.

"The No. 1 reason for making this decision was the difficulty in finding quality labor," shared one dairyman. "We were fighting to find reliable labor to work in our parlor," added another. Both of these farms faced this situation prior to the pandemic and turned to robots as a remedy.

"We are fortunate to have a very strong and stable team of people, but adding extra labor to the team was simply unpalatable," added a third dairyman. By incorporating robots, a fourth Round Table participant said, "We expanded by nearly 400 cows and did not change labor needs over a two-year period."

In the end, each of us must look at our own individual businesses and determine where we can get the most return on our hard-earned dollars. As inflation continues to run rampant in the near term, that may include paying more for labor or making a long-term investment in new technology. One thing is certain — neither decision will be cheap.

At the founding of *Hoard's Dairyman* in 1885, its founder who became Wisconsin's 16<sup>th</sup> governor just three years later wrote, "If a dairy farmer is going to get rich in this world, he is going to employ a whole lot of pairs of hands."

That situation really hasn't changed all the much as there is a tremendous amount of work that takes place throughout animal agriculture.

Then there's the even more pressing matter of food security. To that end, Hoard wrote, "The prosperity of the city is bound up in the prosperity of the farm."

# Understanding the host-microbiome interactions in dairy calves: intervention opportunities to improve calf performance

Yang Song<sup>1, 2,3</sup> and Le Luo Guan<sup>1</sup>

<sup>1</sup>Department of Agricultural, Food and Nutritional Science, University of Alberta, Edmonton, Alberta, Canada; <sup>2</sup>Animal Nutrition and Feed Science, College of Animal Science and Technology, Inner Mongolia Minzu University, Tongliao, China; <sup>3</sup>Key Laboratory of Zoonose Prevention and Control at Universities of Inner Mongolia Autonomous Region, Tongliao, China

#### **Abstract**

The evidence of the role of gastrointestinal microbiota in gut health of neonatal dairy calves is accumulating and has revealed that early life microbiome plays important roles in calves' development, growth and health. However, the comprehensive understanding of the gut microbiome at composition and function levels in pre-weaned calves is limited, especially for the lower gut. This paper will include to date knowledge about the initial colonization of the microbiota in the lower gut of newborn calves and how they establish in the gut from birth to pre-weaning stages, and what are the key functions of the gut microbiome, and how they may affect the calf productivity in the long term. This mini review aims to provide some insights and opportunities for novel strategies to improve calf health through manipulation of their gut microbiome.

#### Introduction:

Calves have an undeveloped gastrointestinal tract (GIT) when they are born. The maturation of the gut is modulated by many factors, including external ones such as nutrients, rearing management and environment and internal ones such as genetics, and the colonization and establishment of the microbiome. The mammalian gut microbiome consists of diverse groups of prokaryotic and eukaryotic microbes, which colonize the gut immediately after birth with different succession orders. Microbial colonization during early life is a dynamic process and the initial colonization is largely affected by the delivery methods, environment and material transmissions. Early life microbiota plays an important role in influencing host animal's development and growth. The commensal microbiota has many functions, including polysaccharide digestion, protecting the host against pathogens, stimulating host immune system development, detoxifying toxic metabolites (Tanaka and Nakayama, 2017), and impacting intestinal barrier development (Petersson et al., 2011), which can all affect intestinal barrier function, immune system development, metabolism, and health. To date, the comprehensive understanding of the gut microbiome and its functions are still limited.

#### Gut microbiome in pre-weaned dairy calves:

A recent study detected the labeled *Enterococcus faecium* admitted to the pregnant mice in the amniotic fluid and meconium of newborn mice after sterile cesarean section (C-section), suggesting that maternal microbiota could be transmitted to newborn mice in the utero (Jiménez et al., 2005). Although some evidences have suggested the microbial colonization starting *in utero*, it is still debatable. For neonatal calves, Mayer et al. (2012) reported that *Citrobacter*, *Lactococcus*, *Leuconostoc* and *Lactobacillus* are the first gut colonizers and changing patterns of several bacterial genera. *Citrobacter* spp., appeared in all calves after birth

2023

and disappeared after 24 h, and Clostridium spp. is one of the dominant bacteria between 24 h and 48 h of life based on the microbial colonization process in feces of dairy calves from birth to 42 days of life. In addition, E. coli is the dominant genus from 24 h to day seven after birth, and its abundance starts to decrease from day 3 to day 7, whereas the population of Bacteroides spp. including B. fragilis and B. vulgatus began to increase at this time (Mayer et al., 2012). This study indicated that gut microbial colonization process of pre-weaned ruminant is similar to human, which was firstly colonized by facultative bacteria, following by obligating anaerobic. Additionally, Malmuthuge et al. (2014) described the intestinal microbiota composition of three-week old dairy calves in terms of using different sample types (Mucosa vs. Digesta). Bacteroides, Prevotella, Lactobacillus, Sharpea, Faecalibacterium and Burkholderia genera are predominant in mucosa-attached bacterial community, whereas, Bacteroides, Prevotella, Lactobacillus, Clostridium, Sharpea and Faecalibacterium are predominant genera in digestaassociated community (Malmuthuge et al., 2014). Moreover, Bacteroides-Prevotella and Clostridium coccoides-Eubacterium rectale groups have higher relative abundance during the first 12 weeks after birth based on sequence-specific rRNA cleavage analysis (Uyeno et al., 2010).

There are dynamic changes in the intestinal microbiota of ruminants during the pre-weaning period since the pre-ruminants are considered as monogastric before their rumen are developed. There is a conflict about the most predominant phylum in the ruminant gut based on previous publications (Table 1). Firmicutes is reported to be the most abundant phylum during the first seven weeks of life in feces of dairy calves (Oikonomou et al., 2013; Foditsch et al., 2015), while others suggest that Bacteroidetes is the most predominant phylum in fecal samples of pre-weaned calves (Uyeno et al., 2010; Klein-Jöbstl et al., 2014). Such difference may be due to different breed, calf management strategy, and the sampling method. Malmuthuge et al. (2014) indicated that Firmicutes (57.6%) was the predominant phylum in the digesta, while Bacteroidetes phylum dominates the mucosa-associated microbiota in the small intestine when studying the microbial community using lumen and tissue samples separately. However, most of studies on gut microbiota in dairy calves are based on fecal samples since sample collection process is not invasive (Uyeno et al., 2010; Oikonomou et al., 2013; Klein-Jöbstl et al., 2014). A few researches have used local intestinal tissue and content samples for microbial profile analysis (Malmuthuge et al., 2012, 2014, 2015). In dairy calves, the supplement of calf starter during weaning period does not affect small intestinal bacterial density and lactic acid bacterial populations, however, it tends to increase the number of bacterial phylotypes (Malmuthuge et al., 2013). Additionally, the effect of antimicrobial bacitracin methylene disalicylate (BMD) on dairy calf intestinal microbial community has been reported. The abundance of potential pathogenic Escherichia, Enterococcus and Shigella increased, while the abundance of beneficial Roseburia, Faecalibacterium and Eubacterium decreased (Xie et al., 2013). In the meantime, neonatal calves fed with milk replacer supplemented with prebiotics tend to have more Lactobacilli in their feces than calves fed with milk replacer and antibiotics (Heinrichs et al., 2009). Moreover, the host genetic effect on gut microbiota in dairy calf has been reported that the rectal microbiota of the calf is more similar with its twin sister compared to other calves who are raised in the same place (Mayer et al., 2012). Therefore, selecting the representative intestinal samples as well as taking the host genetics into account are need to assess the microbial profiles in the gut of neonatal and preweaned calves and how they can be affected by different farm management.

#### Functions of the gut microbiota

Lower gut microbiota plays an important role in microbial fermentation indigestible or unabsorbed of carbohydrates including the indigestible polysaccharides (resistant starch, nonstarch polysaccharides), and some monosaccharides and disaccharides (oligosaccharide, lactose, fructose) (Chassard et al., 2010; Schwab and Gänzle, 2011; Wei et al., 2012; Ze et al., 2012). For example, Bacteroides is mainly responsible for resistant starch and xylan fermentation, and Roseburia utilize resistant starch, xylan and oligosaccharides. Additionally, Ruminococcus consume resistant starch and cellulose, and Bifidobacterium utilize oligosaccharide (Chassard and Lacroix, 2013). Short chain fatty acids (SCFAs) are the main microbial fermentation products, mainly including acetate, propionate and butyrate. Butyrate is the major energy source for colon epithelium cells (Rowe and Bayless, 1992) and is related to host immune function and inflammatory modulation by inhibiting NF-κB activation (Lührs et al., 2002) as well as affecting in gut integrity by regulating expression of genes encoding tight junction proteins (e.g., claudin-1 and Zonula Occludens-1) (Wang et al., 2012). Acetate serves as an energy source to be circulated from the blood to the peripheral tissues, such as the liver, where acetate participates lipogenesis and cholesterol synthesis (Bergman, 1990; Bäckhed et al., 2004). Propionate also is circulated into the liver, and is used for gluconeogenesis and cholesterol synthesis regulation (Venter et al., 1990; Reilly and Rombeau, 1993). These suggest that SCFAs are crucial to the host, not only that they are used as energy source, but also they are related to host metabolism and health.

The intestinal microbiota is important in shaping development of innate immune system in neonatal animals. Compared to germ free mice, gnotobiotic piglets colonized with *Lactobacillus* spp., *Colostridium, Roseburia Intestinalis* have been reported to have higher concentration of IgA and IgM in the serum compared to germ-free piglets (Laycock et al., 2012). suggesting that commensal microbiota stimulates the IgA and IgM's secretion in the gut. Gut microbiota is also associated with adaptive immune system homeostasis in neonatal animals, mainly related to T and B cells development. Previous studies also found that the introduction of *Bacteroides fragilis* to germ-free mice is related to Th1 response and regulation of the imbalance of TH1 and TH2 cells (Mazmanian et al., 2005), suggesting that specific bacteria are inducers of T cells. Similar to T cells, decreased number of plasma B cells was reported in the gut of germ-free animals (Crabbé et al., 1968). All the studies mentioned above imply the importance of commensal microbiota in the immune system development of the animals. However, such aspect has not been well studied in the ruminants.

Maintaining a healthy intestinal barrier is of significant importance to the host. The intestinal barrier has many defense mechanisms against pathogens, including mucus layer, epithelial integrity, and epithelial cell turnover (Kim et al., 2010). Mucus layer contains mucin, digestive enzymes and antimicrobial peptides which inhibit bacteria to penetrate into the inner layer (Ashida et al., 2011). Additionally, intestinal integrity is important to inhibit the translocation of pathogens to subepithelial layer. For example, strains of *Bifidobacterium* have been proven to affect gut integrity by strengthening tight junctions *in vitro* (Hsieh et al., 2015). The healthy intestinal barrier is of significant important to the neonates, and the breakdown of the barrier function predisposes the gut to the risks from intestinal pathogens and toxins (Bjarnason et al., 1994). Early life microbiota has been proven to be closely related to intestinal barrier

development. The immature gut barrier in preterm infants is closely related to necrotizing enterocolitis (NEC) (Grave et al., 2007). Through the mechanism for the NEC is still not clear, the administration of probiotic Bifidobacterium infantis protects the intestinal barrier by strengthening the tight junctions in neonatal mice model with NEC (Bergmann et al., 2013), indicating that probiotics may help to reduce NEC in premature infants by improving intestinal barrier function.

Commensal bacteria serve as a major luminal barrier to compete against the pathogens by the following mechanisms, competing for nutrients, producing metabolites (antibiotics such as bacteriocins), stimulating host immune defense, and accelerating gut motility to prevent pathogens colonization (Abt and Pamer, 2014). For example, Bifidobacterium inhibits Enterohemorrhagic Escherichia coli infection and Shiga toxin release by producing acetate in the murine gut (Fukuda et al., 2011). Meanwhile, commensal bacteria drive intestinal epithelial cells to secrete anti-microbial peptides, inhibiting pathogens expansion in mice (Cash et al., 2006). Moreover, commensal Bifidobacteria breve stimulates an immunoregulatory response by generating exopolysaccharide, which inhibits Citrobacter Rodentium expansion (inducing colonic crypt hyperplasia) in mice (Fanning et al., 2012). Overall, the above findings from mice and human suggest that commensal bacteria are crucial to prevent pathogens colonization in the gut, however, However, the mechanisms behind the how the microbiota contribute to host intestinal barrier function, immune system development, metabolism, and health are not well defined in dairy calves.

#### Effect of early life microbial shift on lifelong host health

Microbiota dysbiosis during early life has been reported to affect long-term performance on their mammalian host. In humans, the disturbance of the early life microbial colonization leads to a variety of disease in adults such as food allergies, atopic dermatitis and asthma (Stiemsma and Turvey, 2017). One case study that investigated the relationship between shifts in intestinal microbiota and the development of atopic dermatitis in Sweden and Estonia children showed that the children with atopic dermatitis have lower Enterococcus at one month old, lower Bifidobacterium at one year old, and higher Clostridium at three months of age when compared to healthy ones (Björkstén et al., 2001). In piglet, the diversity and composition of postnatal microbiota at day seven is suggested to be an indicator of post-weaning (day 35 after birth) diarrhea. Healthy piglets usually have a higher abundance of Prevotellaceae, Lachnospiraceae, Ruminococcaceae and Lactobacillaceae compared to those with diarrhea at day seven after birth (Dou et al., 2016). In addition, the higher relative abundance of Faecalibacterium. prausnitzii is reported to be related with higher weight gain and lower diarrhea rate in dairy calves at three weeks of life (Oikonomou et al., 2013). Therefore, the shifts in early life microbiota is important which can affect the long-term performance in human and animals, and those studies support that there is an opportunity in manipulating of early life intestinal microbiota to promote long-term animal health.

#### Future direction to intervene early life microbiota

Comparing to human, the research on gut microbial colonization in ruminants is scarce, preventing the effective and long last manipulation approaches. With the development of next generation sequencing method, we are getting more knowledge about the microbiota composition and functions, as well as the importance of early life microbiota on host life-long

health. One of the immediate implications is to discover effective technologies to intervene calf gut microbiome for their resilience to calf scours. Calf scours is one of the major causes of neonatal deaths and a major contributor of the therapeutic and preventative use of antimicrobials in cattle industry. In 2017, USDA reported that digestive problems in pre-weaned calves account for 7.7-11.6% in the US dairies and the calves are often treated with therapeutic (85.7% of dairy farms) or prophylactic (37.6% of dairy farms) antimicrobials [USDA. 2017 Death loss in US cattle and calves due to predator and nonpredator causes, 2015. USDA-APHIS-VS-CEAH. Fort Collins, CO.]. Enteric infection accounts for about 50% of the total death of neonatal dairy calves, which is usually caused by enteric pathogens infection (Cho and Yoon, 2014), suggesting maintaining and improving gut health during early life is essential for preventing infection and reducing mortality and morbidity of neonatal calves. With the federal regulations to control in-feed antimicrobials use in livestock and growing consumer awareness of antimicrobial resistance, the industry must adopt sustainable alternatives to increase host resilience to early life enteric infections in calves. Prebiotics and probiotics are at the forefront of the potential alternatives of antimicrobials to improve resilience to enteric infections in other livestock species, yet knowledge is limited on its efficacy as early life alternatives in cattle. Therefore, it is necessary to develop and apply practical methods to alter the gut microbiome during early life.

It has been reported that feeding heat treated (at 60 °C, 60 min) colostrum enhanced the abundance of small intestinal beneficial bacteria (Bifidobacterium) and reduced the abundance of potential pathogenic bacteria (E. coli) colonization in calves compared to fresh colostrum (Malmuthuge et al., 2014), suggesting that feeding heat treated colostrum shape the bacterial composition towards a "good" direction. Meanwhile, delayed colostrum feeding to 12 h after birth has been shown to reduce the proportion of mucosa-attached Bifidobacterium spp., Lactobacillus spp., and ileum mucosa-attached E. coli in the colon of 2 day old calves, when compared to fed colostrum within one hour after birth (Fischer et al., 2018), implying that delaying colostrum feeding after birth may postpone bacterial colonization in the gut. Moreover, previous study found significant compositional differences at genus level when compared the effect of milk supplemented with antibiotics on fecal microbial profile with milk only, suggesting that antibiotics residues in the milk disrupt the fecal microbiota (Van Vleck Pereira et al., 2016). All the findings above indicate that early life microbial profile can be manipulated through nutritional management strategies, which may affect the long-term health of the dairy calves. Therefore, more research on the influence of nutritional management on early life microbiota are needed to determine the best approaches to alter the gut microbiome for the improved lifelong productivity.

#### References:

Tanaka M, Nakayama J. Development of the gut microbiota in infancy and its impact. on health in later life. Allergol Int 2017;66:515-22.

Petersson J, Schreiber O, Hansson GC et al. Importance and regulation of the colonic. mucus barrier in a mouse model of colitis. AJP: Gastrointestinal and Liver Physiology 2010;300:G327–33.

Jiménez E, Delgado S, Maldonado A et al. Staphylococcus epidermidis: a differential trait of the fecal microbiota of breast-fed infants. BMC Microbiol 2008;8:143.

Mayer M, Abenthum A, Matthes JM et al. Development and genetic influence of the rectal bacterial flora of newborn calves. Vet Microbiol 2012;161:179-85.

- Malmuthuge N, Griebel PJ, Guan LL. Taxonomic identification of commensal bacteria associated with the mucosa and digesta throughout the gastrointestinal tracts of preweaned calves. Appl Environ Microbiol 2014;80:2021-8.
- Uyeno Y, Sekiguchi Y, Kamagata Y. rRNA-based analysis to monitor succession of faecal bacterial communities in Holstein calves. Lett Appl Microbiol 2010;51:570-7.
- Oikonomou G, Teixeira AGV, Foditsch C et al. Fecal microbial diversity in pre-weaned dairy calves as described by pyrosequencing of metagenomic 16S rDNA. Associations of Faecalibacterium species with health and growth. PLoS One 2013;8:e63157.
- Foditsch C, Pereira RVV, Ganda EK et al. Oral administration of Faecalibacterium. prausnitzii decreased the incidence of severe diarrhea and related mortality rate and increased weight gain in preweaned dairy heifers. PLoS One 2015;10:e0145485.
- Klein-Jöbstl D, Schornsteiner E, Mann E et al. Pyrosequencing reveals diverse fecal. microbiota in Simmental calves during early development. Front Microbiol 2014;5:622.
- Malmuthuge N, Li M, Chen Y et al. Distinct commensal bacteria associated with. ingesta and mucosal epithelium in the gastrointestinal tracts of calves and chickens. FEMS Microbiol Ecol 2012;79:337-
- Malmuthuge N, Chen Y, Liang G et al. Heat-treated colostrum feeding promotes. beneficial bacteria colonization in the small intestine of neonatal calves. J Dairy Sci 2015;98:8044-53.
- Malmuthuge N, Li M, Goonewardene LA et al. Effect of calf starter feeding on gut. microbialdiversity and expression of genes involved in host immune responses and tight junctions in dairy calves during weaning transition. J Dairy Sci 2013;96:3189-200.
- Xie G, Duff GC, Hall LW et al. Alteration of digestive tract microbiome in neonatal Holstein bull calves by bacitracin methylene disalicylate treatment and scours. J Anim Sci 2013;91:4984-90.
- Heinrichs AJ, Jones CM, Elizondo-Salazar JA et al. Effects of a prebiotic supplement on health of neonatal dairy calves. Livest Sci 2009;125:149-54.
- Chassard C, Delmas E, Robert C et al. The cellulose-degrading microbial community. of the human gut varies according to the presence or absence of methanogens. FEMS Microbiol Ecol 2010;74:205-13.
- Schwab C, Gänzle M. Lactic acid bacteria fermentation of human milk oligosaccharide components, human milk oligosaccharides and galactooligosaccharides. FEMS Microbiol Lett 2011;315:141-8.
- Wei X, Guo Y, Shao C et al. Fructose uptake in Bifidobacterium longum NCC2705 is mediated by an ATPbinding cassette transporter. J Biol Chem 2012;287:357–67.
- Ze X, Duncan SH, Louis P et al. Ruminococcus bromii is a keystone species for the degradation of resistant starch in the human colon. ISME J 2012;6:1535–43.Chem Biol 2011;8:36–45.
- Chassard C, Lacroix C. Carbohydrates and the human gut microbiota. Curr Opin Clin. Nutr Metab Care 2013;16:453-60.
- Rowe WA, Bayless TM. Colonic short-chain fatty acids: fuel from the lumen? Gastroenterology 1992;103:336-8.
- Lührs H, Gerke T, Müller JG et al. Butyrate inhibits NF-kappaB activation in lamina. propria macrophages of patients with ulcerative colitis. Scand J Gastroenterol 2002;37:458-66.
- Wang H-B, Wang P-Y, Wang X et al. Butyrate enhances intestinal epithelial barrier. function via upregulation of tight junction protein Claudin-1 transcription. Dig Dis Sci 2012;57:3126-35.
- Bergman EN. Energy contributions of volatile fatty acids from the gastrointestinal. tract in various species. Physiol Rev 1990;70:567-90.
- Bäckhed F, Ding H, Wang T et al. The gut microbiota as an environmental factor that. regulates fat storage. Proc Natl Acad Sci U S A 2004;101:15718-23.
- Venter CS, Vorster HH, Cummings JH. Effects of dietary propionate on carbohydrate. and lipid metabolism in healthy volunteers. Am J Gastroenterol 1990;85:549-53.
- Reilly KJ, Rombeau JL. Metabolism and potential clinical applications of short-chain. fatty acids. Clin Nutr 1993;12:S97-105.

- Laycock G, Sait L, Inman C et al. A defined intestinal colonization microbiota for gnotobiotic pigs. Vet Immunol Immunopathol 2012;149:216-24.
- Mazmanian SK, Liu CH, Tzianabos AO et al. An immunomodulatory molecule of. symbiotic bacteria directs maturation of the host immune system. Cell 2005;122:107–18.
- Crabbé PA, Bazin H, Eyssen H et al. The normal microbial flora as a major stimulus. for proliferation of plasma cells synthesizing IgA in the gut. Int Arch Allergy Immunol 1968;34:362–75.
- Kim M, Ashida H, Ogawa M et al. Bacterial interactions with the host epithelium. Cell Host Microbe 2010;8:20-35.
- Ashida H, Ogawa M, Kim M et al. Bacteria and host interactions in the gut epithelial. barrier. Nat Chem Biol 2011;8:36-45.
- Hsieh C-Y, Osaka T, Moriyama E et al. Strengthening of the intestinal epithelial tight. junction by Bifidobacterium bifidum. Physiol Rep 2015;3.
- Bjarnason I. Intestinal permeability. Gut. 1994; 35(1 Suppl):S18-22.
- Grave GD, Nelson SA, Walker WA et al. New therapies and preventive approaches for necrotizing enterocolitis: report of a research planning workshop. Pediatr Res 2007;62:510-4.
- Bergmann KR, Liu SXL, Tian R et al. Bifidobacteria stabilize claudins at tight. junctions and prevent intestinal barrier dysfunction in mouse necrotizing enterocolitis. Am J Pathol 2013;182:1595-606.
- Abt MC, Pamer EG. Commensal bacteria mediated defenses against pathogens. Curr. Opin Immunol 2014;29:16-22.
- Fukuda S, Toh H, Hase K et al. Bifidobacteria can protect from enteropathogenic. infection through production of acetate. Nature 2011;469:543-7.
- Cash HL, Whitham CV, Behrendt CL et al. Symbiotic bacteria direct expression of an intestinal bactericidal lectin. Science 2006;313:1126-30.
- Fanning S, Hall LJ, Cronin M et al. Bifidobacterial surface-exopolysaccharide facilitates commensal-host interaction through immune modulation and pathogen protection. Proc Natl Acad Sci U S A 2012;109:2108-13.
- Stiemsma LT, Turvey SE. Asthma and the microbiome: defining the critical window in early life. Allergy Asthma Clin Immunol 2017;13:3.
- Björkstén B, Sepp E, Julge K et al. Allergy development and the intestinal microflora during the first year of life. J Allergy Clin Immunol 2001;108:516-20.
- Dou S, Gadonna-Widehem P, Rome V et al. Characterisation of early-life fecal microbiota in susceptible and healthy pigs to post-weaning diarrhoea. PLoS One 2017;12:e0169851.
- Fanning S, Hall LJ, Cronin M et al. Bifidobacterial surface-exopolysaccharide. facilitates commensal-host interaction through immune modulation and pathogen protection. Proc Natl Acad Sci U S A 2012;109:2108-13.
- Stiemsma LT, Turvey SE. Asthma and the microbiome: defining the critical window. in early life. Allergy Asthma Clin Immunol 2017;13:3.
- Björkstén B, Sepp E, Julge K et al. Allergy development and the intestinal microflora. during the first year of life. J Allergy Clin Immunol 2001;108:516-20.
- Fischer AJ, Song Y, He Z et al. Effect of delaying colostrum feeding on passive. transfer and intestinal bacterial colonization in neonatal male Holstein calves. J Dairy Sci 2018;101(4):3099-3109.
- Van Vleck Pereira R, Lima S, Siler JD et al. Ingestion of milk containing very low. concentration of antimicrobials: longitudinal effect on fecal microbiota composition in preweaned calves. PLoS One 2016;11:e0147525.

## **Transforming Livestock Grazing with Virtual Fencing**

Karen Launchbaugh, Professor, and Hope de Avila, Graduate Research Assistant

University of Idaho Rangeland Center, MS 1135, Moscow, ID 83844, (208) 885-4934, range@uidaho.edu

#### Introduction

Controlling where animals graze, obtain water, or rest is an age-old challenge of livestock husbandry. For more than 10,000 years this issue was addressed by intensive herding or with barriers made of sticks and stones. In the late 1800's as ranchers staked claim to the western lands of North America, they needed tools to set boundaries and control livestock on their lands. However, ranchers moving into the Great Plains encountered a prodigious shortage of lumber and stones. These ranchers grasped the value of a metal wire armed with sharp metal points and many styles of "barbed wire" were patented in the 1860's. Today, millions of miles of barbed wire fencing exist on rangelands across the globe.

The idea of an electrified fence was proposed in the 1800's, and a reliable electric wire was developed in the 1930's in New Zealand by Bill Gallagher, who later founded Gallagher Group Ltd© and began manufacturing electric fence equipment (https://am.gallagher.com/en-US/About-Us). Electric fence systems began to appear in the United Sates in the 1950-60s and have steadily grown in popularity. A significant advantage of electric over traditional wire fence is that it can allow for relatively rapid changes in pasture configuration facilitating adaptive, intensive, and targeted grazing methods.

In 1973, a new electronic approach to fencing was proposed to contain dogs (US Patent No. 3,753,421 Aug. 1973). This became the Invisible Fence© system, which is now used across the globe (https://www.invisiblefence.com/). This fence system involves an electronic device worn by the animal that delivers an electric shock when the animal approaches a boundary delineated by a radio frequency. Though designed for dogs, this system has been successfully used to contain livestock in a targeted grazing context (Fay et al. 1989).

When Global Positioning Systems (GPS) became widely available, Dr. Dean Anderson with the USDA Agricultural Research Service collaborated with a private electronics firm to patent a system where animal location was determined with satellite GPS signals (US Patent No. 6,232,880 May 2001) and controlled within a perimeter set on a virtual map. Based on this initial idea, GPS-based systems have been developed that allow ranchers to draw a pasture on a computer or mobile device to set virtual pasture boundaries. Livestock wear an electronic band/chain/collar around their neck and a sound is emitted when the animal approaches a virtual boundary based on their GPS location and an electric shock is delivered through the collar if the animal does not turn away from the boundary. Several companies offer GPS-based virtual fence systems including: Vence (http://vence.io), eShepherd (https://am.gallagher.com/en-US/new-products/eShepherd), NoFence (http://nofence.no/en/), Halter (https://halterhq.com), and Corral (https://www.corraltech.com). In the United States, Vence is the most widely available system, with on-ranch testing that started in 2019.

#### The Attraction of Virtual Fence

The idea that animals could be contained in a pasture or excluded from a specific area without wire and posts is alluring for several reasons. The development of an electronic containment

system on grazing lands has been significantly motivated by the potential to reduce the cost of fence construction and maintenance. The cost of wire fence varies considerably depending on topography and access but expected costs to construct a multi-strand barbed wire fence range from \$10,000 to \$20,000/mile (NRCS-USDA 2020) and can cost considerably more in steep, inaccessible areas. The time and expense to build new and maintain existing fences is a significant enterprise expense and has fueled interest in electronic virtual alternatives.

Virtual fence will not replace the need for physical fences along property or unit boundaries. However, the ability to easily revise and move a virtual fence boundary will provide unlimited opportunities to develop cross fencing within a unit or pasture. Adaptable cross fences can facilitate a host of grazing options. These include management-intensive or holistic grazing methods, which rely on high stock density, small paddocks, and rapid pasture rotations. Virtual fence technologies could also facilitate grazing of crop residue and failed crops for forage (Lardy 2017). Many targeted grazing goals can also be accomplished with virtual fence. Livestock grazing can temporarily be concentrated in specific areas to facilitate weed control (Fay et al. 1989) or create fuel breaks (Boyd et al. 2022a).

Alternatively, virtual fence is a promising technology to reduce or eliminate livestock grazing in ecologically important areas such as riparian zones (Campbell et al. 2018). Virtual fence has also been applied to facilitate restoration and revegetation in forest regeneration sites (Campbell et al. 2020), and recently burned areas (Boyd et al. 2022b). It has also been suggested that virtual fence could reduce human-livestock conflicts by keeping livestock out of areas with high value to humans such as recreation sites (Wolf et al. 2017) and historically important or archaeological sites. Virtual fences would also reduce loss of livestock and impact on grazing management plans when gates are left open by cyclists, off-road vehicles, hunters, or other recreationists (Wolf et al. 2017).

Virtual fence also offers solutions for wildlife friendly livestock containment. The removal of physical fences would reduce habitat fragmentation and eliminate fence-related stress, injury, and mortality for many wildlife species (Jakes et al. 2018). Virtual fences may also benefit wildlife by reducing livestock activity in nesting or brooding areas (Bleho et al. 2014).

Several of the currently available virtual fence systems offer additional benefits beyond animal containment or exclusion. Some systems allow ranchers to view animal locations within a pasture, making it easier for them to check the herd. Systems such as Vence also collect health attributes that allow ranchers to monitor and locate injured or ill animals.

#### **Effectiveness of Virtual Fence**

The idea of containing livestock with an auditory warning followed by a small electric shock instead of a physical fence may sound unattainable or ridiculous. However, in the last several decades dozens of studies have been conducted to demonstrate that the premise of a virtual fence is feasible. One of the first attempts to electronically contain livestock was accomplished by Fay and colleagues (1989) using electric shock collars designed for dogs to successfully contain a group of six goats for 12 days. Quigley and colleagues (1990) also showed early success by training four steers to stay within a virtual boundary in just four days also using remote dog training collars.

In more recent research, a virtual fence was used to exclude ten angus heifers from entering a riparian area in a small pasture for ten days (Campbell et al. 2018). Likewise, 20 angus cows were contained in a pasture and restricted from grazing a recently burned area in a 14-day trial (Boyd et al. 2022a). Another group of 20 cattle were excluded from an area of regenerating saplings 99.8% of the time during a 44-day trial (Campbell et al. 2020) and 30 lactating dairy cows were contained in virtual pastures >99% of the time in a 10-day trial (Langworthy et al. 2021). Though research has revealed general success in containing animals, non-compliance is also observed and 100% containment after training is seldom demonstrated.

#### **Virtual Fence Failures and Challenges**

Beyond carefully controlled experiments, virtual fence systems have demonstrated varying success in active ranching operations across the globe. The results of on-the-ground applications of virtual fence reveal that the greatest source of failure appears to be the loss of the neck collar devices. There are several reasons why collars may come off the animals after they are applied. Some collars may have been initially fitted and mounted too loosely, allowing the animal to rub or shake the collar off. Other collars may have been appropriately fitted, but a loss in animal weight caused the collar to loosen and the collars were subsequently lost. Certain collar designs have electrical contacts that can be inverted to point away from the animal if the collar is rubbed, so the animal no longer receives the electrical stimulus resulting in noncompliance. Finally, some collars simply sustain damage from the animal, causing them to disengage and fall off.

Other challenges voiced by livestock producers include the time required to install and manage a virtual fence system. Some producers have noted that significant time is required to learn and become proficient with the computer user interface needed to set virtual fences and track animal locations. Producers also noted that it takes considerable time to prepare the devices for deployment and get collars attached appropriately to animals. Some devices also require an occasional battery change, another substantial time commitment especially in larger herds of animals. Finally, it has been noted in the early field applications that recovery efforts to find lost or damaged collars and replace them on the animal are another significant time investment. Aside from the time for the producer to learn and launch the system, another potential challenge to implementing a virtual fence is the time required to train animals to understand and respect a virtual boundary signaled by an audio cue and modify behavior to avoid a potential electric shock. Fortunately, animals tend to require only a few electric stimuli before learning the relevance of the audio cue warning and to turn away to avoid the aversive electric stimulus (Umsatter 2011). Protocols can 0:31 be implemented to train animals in groups rather than individually, making the application of virtual fence to a herd or flock more feasible.

Related to animal training, another challenge that will affect the application of virtual fence is the portion of animals that appear "untrainable" and do not respond appropriately to the audio cue or electric shock. While research has quantified this non-compliance as low, ranchers testing virtual fence systems have confirmed that a few animals seem to be unresponsive to the sound or electrical stimulus. Even among well-trained and responsive animals, a virtual boundary may become less effective as forage availability becomes limited (Langworthy 2021), through social facilitation of peers (Keshavarz et al. 2020), or by uncollared calves who have left the virtual pasture (Boyd et al. 2022a). Ultimately, all animals in a herd may need to be collared, and non-compliance with virtual fence may become a culling criterion for producers.

Potential stress or weight loss by animals in a virtual fence system have been a concern. Several studies have examined potential acute and chronic stress effects of containment by a virtual fence system. Few studies have detected any changes in stress indicators over short- or longer-term use. The lack of negative physiological or neurological effects, assessed in a variety of ways across several studies, indicate that the welfare impacts of virtual fencing on cattle and sheep are minimal. However, continued assessment of animals in virtual fence systems over a longer term should be conducted to confirm these initial conclusions.

The current cost of virtual fence is highly dependent on the system being used, and the number of communication base stations required. It is difficult to generalize, but recent costs for the Vence system would include \$10,000-\$12,000 for each radio gateway (or communication base station) and an additional \$35 subscription fee for each cow collar. The number of radio gateways required varies depending on topography, but generally one to three are needed. Costs of other virtual fence systems were not readily available at the time of this writing. Though the cost of virtual fence will undoubtedly decrease with technological advances, it is currently not a low-cost technology.

#### **Future Developments and Necessary Understanding.**

After decades of research and refinement, several virtual fence systems are now on the market and being tested on the range. However, these current systems are still largely in the prototype and real-world testing phases. Much is yet to be learned to hone the idea of virtual fence into a highly efficacious system for grazing animals. Virtual fence systems are clearly effective for livestock species including cattle, sheep, and goats. However, virtual fence systems have not been widely tested for horses (Janicka et al. 2022). A greater understanding of differences between breeds, ages, and sexes within species relative to their response to virtual fence is needed to develop effective virtual fence systems.

Developing effective and ethically appropriate virtual fence systems will also require deeper understanding of the role of animal experience and training. Initial training procedures for virtual fence systems have been applied but more research and understanding will lead to more efficient training protocols that require the application of fewer electrical stimuli to the animal. Several studies have examined the types of visual or audio cues that are particularly salient as warning cues (Umsatter et al. 2015). However, we still know little about how to combine these external cues to hasten animal learning.

The current virtual fence systems rely on large batteries which must be mounted on the animal in a way that can tolerate the weight. Therefore, current technology options involve devices attached to collars or bands mounted on the neck to deliver audio cues and electrical stimulation. Some systems are solar powered (i.e., Corral) while others rely solely on battery power (i.e., Vence). Though the neck can sustain devices of considerable weight, preliminary research at the University of Idaho indicates that lower levels of electrical stimulation are required to elicit an animal response when delivered to the ear compared to the neck. In 1999, an electronic ear tag device was found to be effective (Tiedemann et al. 1999). However, this device weighed 113 grams and the authors indicated that it was too heavy for an ear-borne device in rangeland settings. However, with advancement in battery technology, an electronic ear tag device may become feasible.

The path for effective virtual fence for grazinglands is being paved. As we learn more about animal behavior, we will be able to more effectively select and train animals to adapt and live within virtual pastures. Advances in electronic, battery and communication technologies will provide more effective and less expensive systems. Robust virtual fencing technology could, like barbed wire over a century ago, be a catalyst that transforms livestock operations and improves economic and environmental sustainability for ranchers across the globe.

#### References

- Bleho, B. I., Koper, N., & Machtans, C. S. (2014). Direct effects of cattle on grassland birds in Canada. *Conservation Biology*, 28(3), 724-734.
- Boyd, C. S., O'Connor, R. C., Ranches, J., Bohnert, D. W., Bates, J. D., Johnson, D. D., ... & Doherty, K. E. (2022a). Using Virtual Fencing to Create Fuel Breaks in the Sagebrush Steppe. *Rangeland Ecology & Management*. (in press)
- Boyd, C. S., O'Connor, R., Ranches, J., Bohnert, D. W., Bates, J. D., Johnson, D. D., ... & Doherty, K. E. (2022b). Virtual Fencing Effectively Excludes Cattle from Burned Sagebrush Steppe. *Rangeland Ecology & Management*, 81, 55-62.
- Campbell, D. L., Haynes, S. J., Lea, J. M., Farrer, W. J., & Lee, C. (2018). Temporary exclusion of cattle from a riparian zone using virtual fencing technology. *Animals*, *9*(1), 5.
- Campbell, D. L., Ouzman, J., Mowat, D., Lea, J. M., Lee, C., & Llewellyn, R. S. (2020). Virtual fencing technology excludes beef cattle from an environmentally sensitive area. *Animals*, *10*(6), 1069.
- Fay, P. K., McElligott, V. T., & Havstad, K. M. (1989). Containment of free-ranging goats using pulsed-radio-wave-activated shock collars. *Applied Animal Behaviour Science*, *23*, 165-171.
- Jakes, A. F., Jones, P. F., Paige, L. C., Seidler, R. G., & Huijser, M. P. (2018). A fence runs through it: A call for greater attention to the influence of fences on wildlife and ecosystems. *Biological Conservation*, 227, 310-318.
- Janicka, W., Wilk, I., Próchniak, T., & Janczarek, I. (2022). Can Sound Alone Act as a Virtual Barrier for Horses? A Preliminary Study. *Animals*, 12(22), 3151.
- Keshavarzi, H., Lee, C., Lea, J. M., & Campbell, D. L. (2020). Virtual fence responses are socially facilitated in beef cattle. *Frontiers in Veterinary Science*, *7*, 543158.
- Langworthy, A. D., Verdon, M., Freeman, M. J., Corkrey, R., Hills, J. L., & Rawnsley, R. P. (2021). Virtual fencing technology to intensively graze lactating dairy cattle. I: Technology efficacy and pasture utilization. *Journal of Dairy Science*, 104(6), 7071-7083.
- Lardy, G. (2017). Using drought stressed field crops as emergency forages. Tri-Sate Livestock News. Swift Communications, Inc.
- NRCS-USDA (2020). EQIP FY 2020 Cost List (382 Fence Barbed/Smooth Wire \$2.42/ft). Online: <a href="https://www.nrcs.usda.gov/sites/default/files/2022-09/EQIP-2020-CostList\_CB.pdf">https://www.nrcs.usda.gov/sites/default/files/2022-09/EQIP-2020-CostList\_CB.pdf</a> (retrieved 14-Dec-2022).
- Tiedemann, A.R., Quigley, A.R., White, L.D., Lauritzen, W.S., Thomas, J.W. & McInnis, M.L. (1999). *Electronic (fenceless) control of livestock* (PNW-RP-510). US Department of Agriculture, Forest Service, Pacific Northwest Research Station.
- Quigley, T. M., Sanderson, H. R., Tiedemann, A. R., & McInnis, M. L. (1990). Livestock control with electrical and audio stimulation. *Rangelands*, 12(3), 152-155.
- Umstatter, C. (2011). The evolution of virtual fences: A review. *Computers and Electronics in Agriculture*, 75(1), 10-22.
- Umstatter, C., Morgan-Davies, J., & Waterhouse, T. (2015). Cattle responses to a type of virtual fence. Rangeland Ecology & Management, 68(1), 100-107.
- Wolf, K. M., Baldwin, R. A., & Barry, S. (2017). Compatibility of livestock grazing and recreational use on coastal California public lands: importance, interactions, and management solutions. *Rangeland Ecology & Management*, 70(2), 192-201.

# A look at late-gestation and early post-partum developmental programming in beef cattle

K.W. Wood, K.S. Hare, and M.A. Steele

Department of Animal Bioscience, University of Guelph, Ontario, Canada kwood@uoquelph.ca

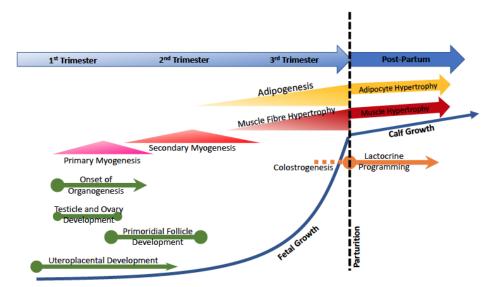
#### **Abstract**

The study of developmental programming in cattle productions has opened new possible opportunities for improved production practices through multi-generational feeding approaches. This use of nutritional stimuli during different stages of production has been shown to have influence on a variety of economically relevant traits in the beef sector, like reproduction, efficiency, and carcass traits. In beef production studying developmental programming is important not only as a proactive management approach for manipulating desirable offspring phenotype, but also as a reactive approach in attempt to overcome intentional stressors from the environment, such as drought, which may also impact offspring growth and performance potential. In late gestation in particular, adipogenesis is often the main target for developmental programming effects. However newer interests in late gestation maternal nutrition may impact colostrogenesis, resulting in lactocrine programming impacts on calf development and health. In this proceeding we will discuss our recent work on late gestation maternal nutrition impacts on developmental programming and impacts on colostrum and lactocrine programming effects. As research continues to increase the understanding of the impacts of maternal nutrition on future impacts of offspring production traits, this new management strategy may represent the next steps in improving efficiency, health, and production traits in the beef industry.

#### Introduction

Many beef producers have often noticed that the calf crop from certain years may have been a particular good year or poor year in calf performance. Although unbeknownst to those producers, they may have been noticing the impacts of fetal or developmental programming. This field of study has grown particularly from research in the 90's and 2000's from epidemiological data from health outcomes following of children of women pregnant during World War II famines (Rosebloom et al., 2001). This resulted in the general concept of developmental programming which suggests that early life stressors, such as nutritional, toxicological, environmental, etc. conditions, can alter offspring phenotypes later in life. These stressors can alter expression of a variety of genes and may even have lasting impacts that can be inherited from generation to generation. Generally, these effects on the offspring are largely a function of the timing of the stressor during fetal development.

For beef cattle some of these key developmental periods are outlined in Figure 1. There are many excellent published reviews on the potential of fetal/developmental programming as a transformative technology in the beef industry which covers these fetal programming impacts in more detail including Funston et al., (2010); Du et al., (2010); Robinson et al., (2013); Funston and Summers, (2013); Summers and Funston, (2013); Mossa et al., (2015); Du et al., (2017); Greenwood et al., (2017);



**Figure 1:** Key developmental timeline for economically relevant traits such as meat quality, reproductive development, growth and efficiency and their use as developmental programing in beef cattle. Partially adapted from Du et al., (2010)

Another key principle in the study of developmental programming is the theory of metabolic mismatch. This work is largely a result of British epidemiologist David Barker's "Thrifty Phenotype Hypothesis" where increased metabolic disease states were observed in adults born from mothers following severe nutritional restriction during mid-to-late pregnancy (Hales and Barker, 1992). This hypothesis describes a miss-match between the nutritionally restricted *in utero* environment in which the developing fetus was exposed, and the affluent nutritional environment in which offspring are raised. This mismatch primes the genetics of the fetus to expect a nutrient limiting environment, but the offspring does not encounter such hardships post-partum, resulting in improved metabolic efficiencies and increased fat deposition. Although this is a negative health outcome for humans, for the beef industry increased fat deposition and low metabolic rate are desirable characteristics. Therefore, in late gestation where adipogenesis occurs, many developmental programming approaches in beef cattle follow this metabolic missmatch approach.

#### **Late Gestation Developmental Programming**

Although there are many reasons to investigate developmental programming in early and mid gestation, from a practical management point of view for beef producers, cows are often managed more intensively as they approach parturition, meaning that a more proactive approach to developmental programing may be easier to implement during this period. During late gestation, maternal nutritional requirements increase exponentially to meet the increasing demands of the fetus and reproductive tissues (Bauman and Currie, 1980). Global nutrient restriction models or research focusing on protein or energy restriction/oversupply alone have been shown to influence developmental programming in late gestation. Many of the mechanisms controlling these developmental programming strategies are not well understood. However, previous research has indicated differential impacts on some key genes which are known to influence growth and efficiency, and adipose and muscle development. For example,

research investigating insulin-like growth factor gene families impact muscle growth in cattle both pre- and postnatally (Brameld et al., 2000; Costello, et al., 2008; Micke et al., 2011).

Some previous research in late-gestation global nutritional restriction developmental models use a severe nutritional restriction model to illicit metabolic mis-match (Hough et al., 1990; LeMaster et al., 2017; Meyer et al., 2021). However, one major concern with severe nutritional restriction is the subsequent negative outcomes on the dam, where milk production, post-partum recovery and reproductive success of low BCS cows can be negatively impacted. However, previous work by our group found that nutrient restriction does not need to be severe in order to obtain fetal differences consistent with the thrifty phenotype hypothesis (Paradis et al., 2017). In a group of cows fed either free-choice (140% of total energy requirements) or restricted (85% of total requirements) from ~150 to d 250 of gestation, fetal tissue from longissimus dorsi or semitendinosus had mRNA expression differences in IGF related genes consistent with the theory of nutritional miss-match. Similarly to the results of the studies listed above, methylation of regions of IGF-2 (DMR2) and feed restriction reduced expression of micro RNAs associated with IGF-2 (miR-1, miR-133a) in longissimus dorsi tissue, suggesting that these genes are responsive to maternal nutritional intervention. This indicates that even mild nutrition restriction can influence developmental programming effects.

Often during mid-to-late gestation cattle are managed on native range or extended grazing systems, which may be deficient in protein during this developmental period. Previous developmental programming studies suggest supplemental protein in mid-to-late gestation also improves lipogenic carcass traits in beef cattle as reported by Larson et al., (2009); Underwood et al., (2010); Shoup et al., (2015); Summers et al., (2015); Wilson et al., (2016) and reviewed by Ladeira et al., (2018). Although there are a variety of mechanisms which may be involved in the developmental programming response, peroxisome proliferator-activated receptor gamma, a nuclear receptor which plays a role in adipogenesis and cellular lipid uptake and known to increase intramuscular fat (Baik et al., 2017). Differences in PPARG mRNA expression were reported in fetal *longissimus dorsi* from restricted fed cows in late gestation (Duarte et al., 2014; Paradis et al., 2017;). Additionally, our work showed a time by treatment effect from birth to weaning of heifer calves born from cows fed at or 140% of protein in an isocaloric diet, requirements for 9 weeks prepartum, where PPARG expression in muscle biopsies increased significantly more than heifers born from cows fed at requirements (Hare et al., 2019), suggesting that oversupply may also influence developmental programming.

Although protein supplementation strategies may elicit developmental programming effects, fewer studies have investigated amino acids, such as methionine, specifically. Methionine is a nutritional strategy of interest, as maternal diets high in methyl donors have been shown to alter DNA-methylation in skeletal muscle of calves (Liu et al., 2021). Therefore, these nutritional intervention strategies show potential as an intervention strategy which influences economically important traits for both the cow-calf producer (increased weaning weights) and feedlot producers (improved carcass traits). Although more research has focused on prepartum intervention strategies, the post-partum period also represents an opportunity to influence development of the offspring. In dairy calves born from dams supplemented with remethionine had improved growth to weaning and increased the percentage of calves with acceptable passive transfer of IgG (Wang et al., 2021), suggesting that calf health status may

also benefit from supplemental methionine. Supplemental amino acids has shown to increase milk production, protein, and fat content in beef cows (Hess et al., 1998). Recently, work from our group (Collins, 2019; Leivre, 2020-cow effects; Action et al., 2020, 2020a-feedlot performance) invested the impact of late gestational supply of protein and methionine on calf heath, growth, and performance traits. Cows were fed isocaloric diets at 90, 100, or 110% of total metabolizable protein requirements, with or without rumen protected methionine for 8 weeks before parturition. An additional group of cows were fed hay ad libitum, with or without 12g/hd/d rp-methionine, and subsequent steer offspring were evaluated in the feedlot (Action et al., 2020a). In both studies, despite limited number of steers, found differences in steer offspring performance or carcass traits. In protein restricted fed cows, steer offspring had increased weaning weights, and increased grade fat and lean yield, despite no significant differences in feedlot efficiency or performance. In the applied study, steer offspring from cows supplemented with rp-methionine, did not differ in feedlot performance, but were heavier at all pre-weaning time-points and upon entering the feedlot.

Energy requirements pre-partum also greatly increase prior to parturition (Bauman and Currie, 1980). Glucose is a particularly high demand metabolic fuel during this time as the growing calf uses most of the cow's available glucose. In our previous work investigating protein supplementation during late gestation beef heifers, we observed that their capacity to digest starch decreases as they approach calving (Hare et al., 2019), raising concern as to how adequately late gestation cattle can use the starch in their rations to meet their glucose requirements prior to calving. By comparison, late gestation dairy cows are anticipated to enter into a glucose deficit immediately prior to calving when they are consuming energy in excess of their requirements (Overton, 1998). Since dairy cattle typically consume more energy dense rations during their dry period, collectively this led us to question whether beef cows are experiencing a glucose shortage before calving and whether their metabolism and colostrum production would shift when they received more dietary energy.

At the Ontario Beef Research Center, we fed primi- (n = 47) and multiparous (n = 109) cattle rations that were formulated with CNCPS 6.55 (Nutritional Dynamic System software, RUM&N Sas, Via Sant' Ambrogio, Italy) to provide 80 (LowME, n = 54), 100 (ConME, n = 51) or 120% (HighME, n = 51) of their predicted metabolizable energy requirements for 56 days prior to calving. At calving, colostrum was sampled for composition and bottle fed back to calf. Performance was similar amongst treatments prior to calving; although, HighME cattle consistently lost body weight and condition at a lesser rate than LowME cattle before calving. Feeding more energy prior increased serum glucose and cholesterol while reducing serum NEFA concentrations, indicating that markers of energy balance were altered between treatments. Part of this response is likely due to LowME cattle being in a negative energy balance and mobilizing more adipose tissue to meet energy requirements. However, we also found that the HighME cattle were less insulin responsive than the LowME cattle during glucose tolerance tests, demonstrating that alternate pathways of energy repartitioning were activated by differential energy provision. As anticipated, cattle that consumed more energy prior to calving produced more colostrum (HighME:  $2.6 \pm 0.31$  kg; ConME:  $2.1 \pm 0.25$  kg; LowME:  $1.5 \pm 0.17$  kg). Increasing energy intake increased the lactose concentration, while decreasing the crude protein, urea, and beta-hydroxybutyrate concentration. Colostrum fat concentration was greatest for the LowME treatment relative to the ConME (6.1 vs.  $4.9 \pm 0.29\%$ ), but the HighME

colostrum fat concentration ( $5.5 \pm 0.29\%$ ) did not differ from either the LowME or ConME. Colostrum immunoglobulin G concentration decreased linearly from low to high ration energy density. However, because the HighME cattle produced more colostrum yield, component yield (fat, protein, lactose, and immunoglobulin G) was uniformly increased when more energy was provided. Lastly, calf birth body weight and preweaning average daily gain were unaffected by prepartum metabolizable energy supplementation. (Croft et al., 2022; Hare et al., 2022a). Collectively these data demonstrate that beef cattle colostrum production is influenced by metabolizable energy consumption prior to calving. In addition, they demonstrate that, when beef cattle experience mild energy deficits, they will hierarchically prioritize calf development before colostrogenesis and maintaining their own body reserves; however, colostrum production is prioritized above body maintenance.

## **Post-Partum Lactocrine Programming**

Although historically colostrum has been studied more for its implications on passive transfer of immunity and subsequent impacts on calf health, there are numerous bioactive components contained in colostrum, which may have implications for calf growth. Many of these bioactive components have functions for not only immune development, but also have roles as hormones and growth factors, which may have play important signaling rolls in early growth and development (Tacoma et al., 2017). Although little is known about beef colostrum specifically, changes bioactive concentrations between colostrum and the next subsequent milkings in dairy cattle are highlighted in Blum and Hammon., (1999). Although this work and others (reviewed by Fischer-Tlustos et al., 2020) illustrates the shifting composition as colostrum matures into milk, less known is on how pre-partum maternal nutrition impacts the profile of the colostrum and abundance of other bioactives in colostrum, particularly in beef cattle.

Our previous work on late gestation protein supplementation where crossbred Hereford heifers fed isocaloric diets either 100% of predicted metabolizable protein (MP) requirements (n=7; CNCPS) or 133% predicted metabolizable protein requirements (n=6) for 55 ± 3 d prior to parturition. Protein supplementation decreased colostrum fat % (3.4 vs 7.0  $\pm$ 0.8; P=0.003) and tended to decrease net energy content (1.4 vs.1.7  $\pm$ 0.1; P=0.052) of colostrum (Hare et al., 2019). Progeny were followed until 112 d of age, however, no treatment differences in heifer calf performance was observed, although age x treatment interactions were observed for PPARg expression in muscle, as discussed above. From this study we also investigated shifts in the proteome (Radford, et al., 2018) and lipidome (Wood, unpublished). Colostrum analysis identified 213 distinct proteins, of which 11 were enriched and 13 were depleted in cows fed a high MP diet vs controls. Enriched colostrum proteins were associated with gut and immune system development (5.48E-08 < P < 2.49E-4) and depleted colostrum proteins were significantly associated with growth regulation (5.48E-08 < P < 2.49E-4). To investigate if bioactives were transferred into calf serum, serum samples from progeny 6 h post-colostrum consumption identified 179 distinct proteins, of which 60 were common with colostrum. In calf serum, maternal dietary protein treatment enriched 28 and depleted 19 proteins compared to progeny from control fed dams. These proteins were distributed across 27 interdependant interaction networks. However, generally maternal protein supplementation decreased generalized inflammatory proteins and non-specific macrophage stimulation and promoted a precision response immune system. Lipidomic analysis determined 1629 lipids in colostrum, which differences in maternal protein supplementation resulted in > 2-fold difference in 246 phospholipids, 103 sphingolipids, 69 storage lipids within colostrum. Although this study has a limited number of animals, it clearly demonstrates the major impact of pre-partum nutrition on colostrum composition and passive transfer of bioactive compounds can be impacted by maternal diet in beef cattle. Further research is needed in the area to elucidate if these differences in bioactives influence longer-term offspring efficiency, health, and carcass traits

One colostrum bioactive of particular interest in lactocrine programming is insulin, as colostrum insulin concentrations can be over 10-fold greater in colostrum that in milk (Fischer-Tlustos et al., 2020) and 50 to 100 times greater than circulating blood concentration in dairy cows (Mann et al., 2016) and can be in influenced by late gestation nutrition in dairy (Mann et al., 2016) and beef (Hare et al., 2022) cows. These greater colostrum insulin concentrations are hypothesized to influence gut development in calves (Shamir et al., 2005). In a recent study (Hare et al., 2023) in dairy bull calves supplemented with a colostrum replacer at either 5x or 10x basal (control) colostrum insulin concentrations, and slaughtered 30 h post-initial colostrum consumption. Dry rumen mass decreased and duodenal dry tissue weight increased with increasing insulin concentration, where ileal villi height and surface area and lactose activity in jejunum also increased with increasing insulin concentration. However, ileal isomaltase activity decreased with increasing colostrum insulin. After the second meal increased colostrum insulin supplementation, increased glucose absorption and NEFA clearance rates, and insulin clearance rates. Although this study is short term, it does demonstrate that changes in colostrum bioactives can have impacts on gut development, which can lead to small changes in postprandial metabolism. Further research is needed to determine if these short-term changes can lead to longer term implications for increase absorptive capacity, improved efficiency and growth and if these same results are observed in beef cattle.

In conclusion, there are clear impacts of late gestation nutrition, induced by global nutrient restriction, protein supplementation, and energy supply which can impact offspring performance, and gene expression of key genes. New investigations into the impact of late gestation nutrition on colostrum composition may also impact offspring development and growth. As new findings in this field of study continue, these results may open up new management regimes for beef producers in late-gestation, which may have positive impacts on offspring growth, health, carcass traits and production efficiency.

#### References

- Acton et al. 2020. PSIX-4 Fetal programming—Maternal plane of nutrition effects on progeny performance, feed efficiency, and carcass quality for feedlot steers. J. Anim. Sci, 98(Suppl 4), 411.
- Acton et al., . 2020a. PSIX-5 Fetal programming in an industry applied setting—Effects of feeding methionine during late gestation on progeny performance, feed efficiency, and carcass quality for feedlot steers. J. Anim. Sci, 98(Supplement\_4), 411-412.
- Baik et al., 2017. Triennial growth and development symposium: molecular mechanisms related to bovine intramuscular fat deposition in the longissimus muscle. J. Anim. Sci. 95:2284-2303.
- Bauman and Currie. 1980. Partitioning of nutrients during pregnancy and lactation: a review of mechanisms involving homeostasis and homeorhesis. J.Dairy Sci., 63(9), 1514-1529.

- Blum and Hammon. 2000. Colostrum effects on the gastrointestinal tract, and on nutritional, endocrine and metabolic parameters in neonatal calves. Livest. Prod. Sci. 66:151-159.
- Brameld et al., 2000. Maternal nutrition alters expression of insulin-like growth factors in fetal sheep liver and skeletal muscle. Journal of Endocrinology 167(3), 429–437
- Croft et al., 2022, September. Supplementation of Late Gestation Metabolizable Energy in Beef Cows Reduces Mobilization of Body Reserves Prepartum. J. Anim. Sci. 100: 65.
- Collins et al., 2019. Does supplemental protein and rumen-protected methionine improve performance and digestibility during late-gestation in beef cows?. J. Anim. Sci., 97(Supplement\_3), 75-75.
- Costello et al.,. 2008. Peri-implantation and late gestation maternal undernutrition differentially affect fetal sheep skeletal muscle development. J. Physiol. 586: 2371-2379.
- Duarte et al., 2014. Maternal overnutrition enhances mRNA expression of adipogenic markers and collagen deposition in skeletal muscle of beef cattle fetuses. J. Anim. Sci. 92:3846-3854.
- Fischer-Tlustos et al., 2020. Invited Review: Effects of colostrum management on transfer of passive immunity and the potential role of colostral bioactive components on neonatal calf development and metabolism. *Can. J. Anim. Sci.*, 101(3), 405-426.
- Du et al., 2010. Fetal programming of skeletal muscle development in ruminant animals. J. Anim. Sci. 88:E51-E60.
- Du et al., 2017. Optimizing livestock production efficiency through maternal nutritional management and fetal developmental programming. Animal Frontiers: 7:5-11.
- Funston et al., 2010. Effects of maternal nutrition on conceptus growth and offspring performance: implication for beef cattle production. J Anim. Sci.88:E205-E215.
- Funston and Summers. 2013. Epigenetics: Setting up lifetime production of beef cows by manipulating nutrition. Annu. Rev. Anim. Biosci. 1:339-363.
- Greenwood et al., 2017. Developmental Programming and beef production. Animal Frontiers. Doi:10.2527/af.2017-0127
- Hales and Barker. 1992. Type 2 )non-insulin-dependent diabetes mellitus : the thrifty phenotype hypothesis. Diabetologia. 35 : 595-601.
- Hare et al., 2023. Colostrum insulin supplementation to neonatal Holstein bulls impacts small intestinal histomorphology, mRNA expression, and enzymatic activity with minor influences on peripheral metabolism. J. Dairy. Sci. In Press.
- Hare et al., 2022. Oversupplying metabolizable protein during late gestation to beef cattle does not influence ante- or postpartum glucose insulin kinetics but does affect prepartum insulin resistance indices and colostrum insulin content. J. Anim. Sci. 100:1-14. https://doi.org/10.1093/jas/skac101.
- Hare et al., 2022a. Differential Late Gestation Metabolizable Energy Intake Alters Adipose-Specific Insulin Responsiveness in Antepartum Beef Cattle. J. Anim. Sci. 100: 72-72.
- Hare et al., 2019. Oversupplying metabolizable protein in late gestation for beef cattle: effects on postpartum ruminal fermentation, blood metabolites, skeletal muscle catabolism, colostrum composition, milk yield and composition, and calf growth performance. J Anim. Sci.97:437-455.
- Hess et al., 1998. Supplemental protein plus ruminally protected methionine and lysine for primiparous beef cattle consuming annual rye hay. *Journal of animal science*, 76(7), 1767-1777.
- Hough et al., 1990. Influence of nutritional restriction during late gestation on production measures and passive immunity in beef cattle. *Journal of animal science*, 68(9), 2622-2627.
- Ladeira, et al., 2018. Review: Nutrigenomics of marbling and fatty acid profile in ruminant meat. Animal. doi:10.1017/s1751731118001933
- Larson, et al., 2009. Winter grazing system and supplementation during late gestation influence performance of beef cows and steers progeny. J. Anim. Sci. 87:1147-1155.
- LeMaster, et al., 2017. The effects of late gestation maternal nutrient restriction with or

- without protein supplementation on endocrine regulation of newborn and postnatal beef calves. Theriogenology, 87, 64-71.
- Lievre et al., 2019. 398 Effects of protein level and supplemental methionine in late-gestation on colostrum quality and passive immunity transfer in beef cattle. J. Anim. Sci., 97(Supplement\_3), 160-161.
- Liu et al., 2021. Maternal methionine supplementation during gestation alters alternative splicing and DNA methylation in bovine skeletal muscle. BMC genomics, 22(1), 1-11.
- Mann et al., 2016. Effect of dry period dietary energy level in dairy cattle on volume, 806 concentration of immunoglobulin G, insulin, and fatty acid composition of colostrum. J. Dairy 807 Sci. 99:1515-1526. https://doi.org/10.2168/jds.2015-9926.
- Meyer et al., 2021. Late-Breaking: Late Gestational Nutrient Restriction of Primiparous Beef Heifers Decreases Milk Yield and Pre-weaning Calf Growth. J. Anim. Sci. 99:150.
- Micke et al., 2011. Protein intake during gestation affects postnatal bovine skeletal muscle growth and relative expression of IGF1, IGF1R, IGF2, and IGF2R. Molecular and Cellular Endocrinology. 332:234-241.
- Mossa et al., 2015. Early nutritional programming and progeny performance: Is reproductive success already set at birth? Animal Frontiers. 4:18-24.
- Overton et al., 1998. Metabolic adaptation to experimentally increased glucose demand in ruminants. J.Anim. Sci., 76(11), 2938-2946.
- Paradis et al., 2017. Maternal nutrient restriction in mid-to-late gestation influences fetal mRNA expression in muscles tissues in beef cattle. BMC Genomics. 18:632.
- Radford et al., 2018. Providing excess metabolizable protein prior to calving shifts the protein composition of colostrum and early post-colostrum serum proteomic profiles in neonatal beef calves. J. Anim. Sci. 96:264
- Rosebloom et al., 2001. Effects of prenatal exposure to the Dutch Famine on adult disease in later life: an overview. Twin Research and Human Genetics. 4: 293-298.
- Shamir et al., 2005. Intestinal and systemic effects of oral insulin supplementation in rats after weaning. Dig. Dis. Sci. 50:1239-1244. https://doi.org/10.1007/s10620-005-2766-7.
- Shoup et al.,2015. Beef cow prepartum supplement level and age at weaning: II. Effects of developmental programming on performance and carcass composition of steer progeny. J. Anim. Sci. 93:4936-4947.
- Summers and Funston. 2013. Fetal programming: Implications for beef cattle production.

  Proceeding of the The Range Beef Cow Symposium XXIII, Dec 3-5<sup>th</sup>, 2013. Rapid City, SD.
- Summers et al., 2015. Impact of supplemental protein source offered to primiparous heifers during gestation on II. Progeny performance and carcass characteristics. J Anim. Sci. 93:1971-1880.
- Tacoma et al., 2017. Exploration of the bovine colostrum proteome and effects of heat treatment on colostrum protein profile. J. Dairy. Sci. 100:9392
- Underwood et al., 2010. Nutrition during mid to late gestation affects growth, adipose tissue deposition, and tenderness in crossbred beef steers. Meat Sci. 86:588-593.
- Wang et al., 2021. Maternal supply of ruminally-protected lysine and methionine during closeup period enhances immunity and growth rate of neonatal calves. *Frontiers in Veterinary Science*, 1456.
- Wilson et al., 2016. Influence of excessive dietary protein intake during late gestation on drylot beef cow performance and progeny growth, carcass characteristics, and plasma glucose and insulin concentrations. J. Anim. Sci. 94: 2035-2046



Contents lists available at ScienceDirect

## **Animal**

# The international journal of animal biosciences



# Animal board invited review: Animal agriculture and alternative meats – learning from past science communication failures



A.L. Van Eenennaam\*, S.J. Werth

Department of Animal Science, University of California, 1 Shields Ave, Davis, CA 95616, USA

#### ARTICLE INFO

Article history: Received 6 June 2021 Revised 18 August 2021 Accepted 23 August 2021

Keywords:
Cultured meat
Environment
Life cycle assessment
Plant-based meat
Sustainable diet

#### ABSTRACT

Sustainability discussions bring in multiple competing goals, and the outcomes are often conflicting depending upon which goal is being given credence. The role of livestock in supporting human wellbeing is especially contentious in discourses around sustainable diets. There is considerable variation in which environmental metrics are measured when describing sustainable diets, although some estimate of the greenhouse gas (GHG) emissions of different diets based on varying assumptions is commonplace. A market for animal-free and manufactured food items to substitute for animal source food (ASF) has emerged, driven by the high GHG emissions of ASF. Ingredients sourced from plants, and animal cells grown in culture are two approaches employed to produce alternative meats. These can be complemented with ingredients produced using synthetic biology. Alternative meat companies promise to reduce GHG, the land and water used for food production, and reduce or eliminate animal agriculture. Some CEOs have even claimed alternative meats will 'end world hunger'. Rarely do such selfproclamations emanate from scientists, but rather from companies in their efforts to attract venture capital investment and market share. Such declarations are reminiscent of the early days of the biotechnology industry. At that time, special interest groups employed fear-based tactics to effectively turn public opinion against the use of genetic engineering to introduce sustainability traits, like disease resistance and nutrient fortification, into global genetic improvement programs. These same groups have recently turned their sights on the 'unnaturalness' and use of synthetic biology in the production of meat alternatives, leaving agriculturists in a quandary. Much of the rationale behind alternative meats invokes a simplistic narrative, with a primary focus on GHG emissions, ignoring the nutritional attributes and dietary importance of ASF, and livelihoods that are supported by grazing ruminant production systems. Diets with low GHG emissions are often described as sustainable, even though the nutritional, social and economic pillars of sustainability are not considered. Nutritionists, geneticists, and veterinarians have been extremely successful at developing new technologies to reduce the environmental footprint of ASF. Further technological developments are going to be requisite to continuously improve the efficiency of animal source, plant source, and cultured meat production. Perhaps there is an opportunity to collectively communicate how innovations are enabling both alternative- and conventional-meat producers to more sustainably meet future demand. This could counteract the possibility that special interest groups who promulgate misinformation, fear and uncertainty, will hinder the adoption of technological innovations to the ultimate detriment of global food security.

© 2021 The Author(s). Published by Elsevier B.V. on behalf of The Animal Consortium. This is an open access article under the CC BY license (http://creativecommons.org/licenses/by/4.0/).

#### Contents

Introduction		2 3
Greenhouse gasses	Resource use	9
	Human health and alternative meats	
* Corresponding author.	Wizards, prophets and magicians	. 11

E-mail address: alvaneenennaam@ucdavis.edu (A.L. Van Eenennaam).

MOVING TOTWARD	. 12
Communicating complexity	. 16
Ethics approval	
Data and model availability statement	
Author ORCIDs	. 17
Author contributions	
Declaration of interest	. 17
Acknowledgements	
Financial Support Statement	. 17
References	. 17

#### **Implications**

Demand for animal source food is rising with increased population and income levels. Animal-free alternatives and manufactured food items that aim to substitute for meat, milk and eggs in the diet are emerging markets. Ingredients sourced from plants and animal cells grown in culture are two approaches employed to produce alternative meats. The relative merits of these products compared to animal products depend upon the comparator metric, the manufacturing system, and the reference animal species. Technological innovations to continuously improve the efficiency of both conventional and alternative food production systems will be requisite to sustainably address global food security demands.

#### Introduction

The growth in the human population from around three billion in 1960 to 6.8 billion in 2010 was coupled with a four-fold increase in meat production (Smith, 2013). Innovations in production practices (breeding, feeding, and animal care) have contributed to sustained increases in the availability of animal source food (ASF) in many countries. Perhaps this is most famously illustrated by the modern broiler industry. Before the discovery of vitamins A and D between 1915 and 1925, it was not possible to grow chickens year-round. Vaccines and biosecurity protocols, and moving animals into secure housing facilities reduced disease exposure and predation. Breeding advances, including hybridization, increased yield and feed efficiency, were spurred by the 'Chicken of Tomorrow Contest' of the late 1940s (Sunde, 2003). In 1957, a 42-d-old broiler weighed 586 g and had a feed conversion ratio (g of feed/ g of BW gain) of 2.8; whereas in 2016, a broiler of the same age weighed 2 900 g with a feed conversion ratio under 1.70. Evidence from feeding studies involving heritage-style chicken breeds suggests that although nutrition and management have played a significant role in these changes, it is estimated that improved genetics and breeding accounted for approximately 80-90% of these efficiency gains (Tavárez and Solis de los Santos, 2016). These improvements have dramatically decreased the environmental footprint of a kilogram of chicken protein. Chicken consumption has increased globally since the mid-twentieth century, and in 2019, a staggering 72.1 billion chickens were slaughtered for food (FAO, 2020). Interestingly, consuming chicken was not perceived as 'manly' in the United States, and so in the late 1960s, Frank Perdue and Don Tyson, the two largest poultry producers in the United States, developed a marketing campaign to alter that perception which included television commercials with the slogan 'It takes a tough man to make a tender chicken.' In conjunction with studies from the American Heart Association suggesting negative health effects of red meat, chicken replaced beef as a menu item to become the most consumed terrestrial meat globally, at 132 MMt in 2020. Large-scale shifts in the consumption of ASF, as evidenced by chicken, milk and tilapia, occurred when publicly funded technological innovation was scaled-up by the private sector under supportive policy regimes (Moberg et al., 2021).

There have been many ambiguities and contradictory findings about the health impacts of ASF over the years, especially in dietary recommendations for consumers in middle to high-income countries (MHIC), where typical diets often exceed recommended levels of dietary energy and protein. These include conflicting recommendations regarding the healthfulness of eggs (Drouin-Chartier et al., 2020), dairy products in general (Dehghan et al., 2018; Soedamah-Muthu and de Goede, 2018), margarine versus butter (Pimpin et al., 2016), and red meat (Micha et al., 2010; Chen et al., 2013; Abete et al., 2014; Wang et al., 2016; Schwingshackl et al., 2017; Leroy and Cofnas, 2020). In many cases, observational nutritional epidemiology studies suggested a negative impact of ASF, but further studies did not always confirm that association leading to sometimes contradictory messages, and often fierce disagreements even among subject matter experts in the public health scientific literature (Godfray et al., 2018; Klurfeld, 2018; Leroy and Cofnas, 2020). Additionally, since the publication of Livestock's Long Shadow (Steinfeld et al., 2006), there has been an increasing focus on the negative environmental impacts of livestock production. These discussions tend to focus on a few dimensions of intensive livestock systems in the developed world, notably their environmental impacts and the harm to human health that can be caused by high rates of consumption of ASF and zoonotic diseases (Godfray et al., 2018). The other functions of livestock systems such as converting by-products from the food system, crop residues and grass resources into nutrient-dense food providing a valuable source of essential micronutrients, zinc, vitamin A, iron, vitamin B<sub>12</sub>, riboflavin, and calcium; supporting crop production with manure and draft animal power; providing a regular income, insurance and savings; in addition to fulfilling important cultural, religious and social roles, are often absent or overlooked (Salmon et al., 2020).

These issues have resulted in the development of a market for animal-free alternatives and manufactured food items that aim to be a substitute for ASF in the diet. Ingredients sourced from plants; and animal cells grown in culture are two approaches employed to produce alternative meats. This latter group encompasses products commonly referred to as 'cultured' meat, milk and other animal products. These products can be complemented with ingredients produced using synthetic biology to genetically modify microbes to manufacture specific products, typically by fermentation. To date, alternative meat companies have mostly been located in MHIC (Fig. 1). The framing employed by leading alternative protein stakeholders revolves around five key promissory narratives namely, (1) the promise of being healthier than animal foods by being higher in protein and free from antibiotics; (2) the promise to feed the projected growing global population using less planetary resources; (3) the promise of offering more environmentally efficient production without the need for intensive livestock production or animal slaughter; (4) the promise of increased food safety and traceability via techno-science; and (5) the promise that not only will the alternatives be better for

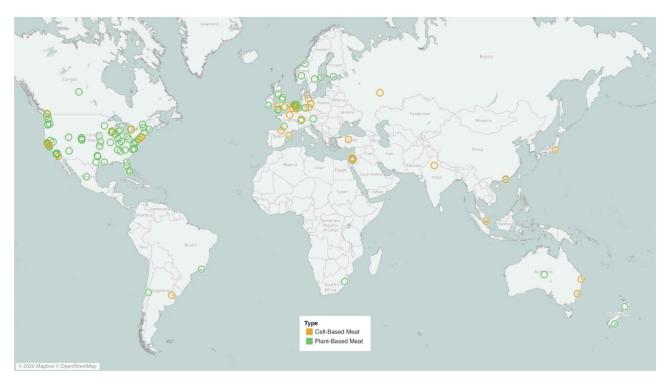


Fig. 1. Geographical distribution of plant-based (green circles) and cell-based (orange circles) alternative meat companies. Companies were listed in the Good Food Institute alternative protein company database (August 2020). Reproduced from Rubio et al. (2020).

humans and the planet but they will also be indistinguishable in taste (Sexton et al., 2019). California-based Impossible Foods even goes so far as to have a mission of completely replacing animals in the food system by 2035, and according to CNN, the Silicon Valley EAT JUST CEO, 'wants to end world hunger' (Mohorčich and Reese, 2019).

Rarely do such hubristic claims emanate from scientists familiar with the complexities of the global food system, but rather from companies, in their efforts to attract venture capital investment and market share (Sexton et al., 2019). Such bold pronouncements are reminiscent of the early days of the biotechnology industry. There too, finance followed aspirations that genetically engineered food 'would alleviate world hunger, create a more sustainable food supply, and create healthier, cheaper food for consumers' (Mohorčich and Reese, 2019). These promises opened the biotechnology industry to attack by activist groups, who effectively created fear around genetically modified organisms (GMOs) by framing GMOs as 'unnatural' and therefore unsafe to eat and grow. And while the target of these campaigns was ostensibly multinational companies, the impact was to preclude global access of academic researchers, and developing countries, to the use of agricultural biotechnology. As a result, virtually none of the promises of GMOs to solve major problems in agriculture, nutrition, sustainability, and food security came to pass. Many of these same special interest groups have since moved their 'Frankenfood' sights, a pejorative term for genetically modified food whether it be derived from genetically engineered plants or animals, toward the unnaturalness of cell-cultured meat, and the processed nature of many plant-based meat alternatives. It bears contemplating whether amplifying misinformation or creating fear about any food production method is in the long-term best interests of global food security. Perhaps now is an opportune time to communicate how producers of both alternative and conventional meats are using science and innovation to try to improve the sustainability of their products. And that jointly, rather than vilifying alternative systems, we need to tell compelling stories around how the adoption of innovation in culturally appropriate food production systems worldwide is crucial to global food security. Failure to do so may increase the chances that misinformation, fear, and uncertainty will ultimately preclude access to useful innovations in agriculture and food production globally.

#### The problem

Currently, plant sources of protein provide the majority of the global protein supply (57%), with meat (18%), dairy (10%), fish and shellfish (6%) and other animal products (9%) making up the remainder. Livestock supply chains are associated with 14.5% of all human-induced greenhouse gas (GHG) emissions (Gerber et al., 2013). The emission intensity (amount of GHGs emitted per unit of output produced) of livestock products varies depending upon product, species and environmental factors (Herrero et al., 2013). Protein-based livestock emission intensities range from a high of 404 kg CO<sub>2</sub>eq/kg of protein for buffalo to a low of 31 kg CO<sub>2</sub>eq/kg protein for eggs (MacLeod et al., 2018). The emission intensities of ASF are higher than protein-rich plant products such as nuts, peas, pulses and groundnut which average 2.6, 4.4, 8.4 and 12.3 CO<sub>2</sub>eq/kg protein, respectively (Poore and Nemecek, 2018). Demand for ASF is rising in conjunction with increased population and income levels. These are commonly accepted facts. What to do about this projected ASF demand, and whether this is a good or a bad thing, are where there are major disagreements. The scientific literature reveals a breathtaking array of different metrics being discussed, and varying perspectives. Recognizing that livestock provide multiple benefits in addition to the protein found in milk, meat and eggs adds significant complexity to already complicated and impassioned discussions. There are so many proposed solutions to this increasing demand, and counter narratives being promoted, that this topic has become something of an infodemic, even among scientists. Wikipedia, an online encyclopedia defines infodemic as a blend 'of "information' and 'epidemic' that typically refers to a rapid and far-reaching spread of

both accurate and inaccurate information about something, such as a disease. As facts, rumors, and fears mix and disperse, it becomes difficult to learn essential information about an issue.' And when it comes to the role of ASF on human health and climate change, especially as it relates to GHG emissions, it becomes increasingly murky. Because animal agriculture is immensely varied in its regional practice, impacts, and nutritional importance, it does not lend itself to a simple 'eat this, not that' dichotomous framing. The role of livestock in supporting human well-being is contentious in discourses around sustainable diets. It is close to impossible for non-experts to decipher the nuances of the various metrics being used by different groups, so people seem to be using motivated reasoning to pick the metric that agrees with their belief system and worldview and ignoring the rest. It is recognized that individuals are more likely to accept facts if they either align with the values they hold or reinforce a predisposition (Kahan et al., 2011).

In 2010, a group of international experts proposed the following definition of sustainable diets: 'Sustainable diets are protective and respectful of biodiversity and ecosystems, culturally acceptable, accessible, economically fair and affordable; nutritionally adequate, safe and healthy; while optimizing natural and human resources' (Burlingame and Dernini, 2012). While hard to contest this definition, it is inherently problematic as it involves multiple components that have potentially antagonistic interactions. Which is more important, affordability, nutrition, safety, human or natural resources? Further, it is unclear what metrics should be measured as indicators for some of these components. Reviews of the sustainable diet literature reveal that there is considerable variation in which metrics are measured, although the estimated GHG emissions per unit of food for different diets were by far the most common metric measured; with land, energy, and water use also being frequently assessed (Jones et al., 2016).

A systematic review of the literature about the relative health impacts of diets with reduced GHG emissions revealed highly heterogeneous outcomes. Across all indicators of 'healthiness', 64% of lower GHG emission diets were linked to worse nutritional and health indicators. Reduced saturated fat and salt were often associated with diets low in animal products, but these diets were often also high in sugar and low in essential micronutrients (Payne et al., 2016). Additionally, almost all of the research on sustainable diets has been centered in high-income countries. This is relevant because low- and middle-income countries (LMICs) experience very different challenges in terms of malnutrition and food insecurity, as compared to high-income countries. In fact, the diets of most poor households in LMIC are still predominantly plantbased, not necessarily by choice, but because of the high price of nutrient-dense ASF. As such, making sustainability comparisons between meat-based and vegetarian or vegan diets in terms of GHG in the developed world makes little sense in the context of LMIC. For the almost 800 million extremely poor people who live on less than \$2/day and subsist on a diet heavily based on starchy plant-based foods, more ASF will be required for sustainable development (Zhang et al., 2016), as ASF provides not only calories but, almost more importantly, the nutrients required for achievement of human development potential (Adesogan et al., 2020).

In 2015, the United Nations proposed a set of 17 global Sustainable Development Goals (SDGs) which comprises 169 targets (UN 2015). There are 14 discrete environmental areas of concern identified in the SDGs including (1) water scarcity, (2) natural resource depletion, (3) urban air quality, (4) ozone depletion, (5) human and ecotoxicity, (6) climate change, (7) marine debris, (8) marine eutrophication, (9) freshwater ecosystem quality, (10) depletion of fish stocks, (11) deforestation, (12) land degradation and desertification, (13) biodiversity loss, and (14) invasive species. In a review of 93 journal articles that reported on the environmental assessments of diets, certain areas of concern, especially GHG

emissions, were frequently reported on, but there was less focus on many of the other environmental areas outlined in the SDGs (Ridoutt et al., 2017). These authors noted that there was a disturbing tendency for sustainable diets with lower GHG emissions to be described as healthy diets in the literature. They argue that this framing is inappropriate as the social and economic aspects of sustainability were not evaluated, and further the authors argue that GHG emissions represent only one of many environmental concerns, and that in the context of a complete food system, this metric may not even be the most important environmental concern.

#### Life cycle assessments

Life cycle assessment (**LCA**) is a widely utilized methodology for both benchmarking and comparing food products and production systems (de Vries and de Boer, 2010; Gerber et al., 2015; Warner, 2019). Modern LCAs follow standards produced by the International Standards Organization (i.e., ISO 14040, 14044, 14046) which set out general principles, framework, and guidelines for life cycle practitioners which helps to standardize LCAs (Sieverding et al., 2020). However, there remains a substantial amount of variability among LCAs, especially those analyzing food products and production systems. Typically, variability comes from the system boundaries set for each LCA, the characterization methods used for each impact category, and emission factors applied to life cycle inventories to determine the final life cycle impacts.

In LCA, the functional unit (FU) is the reference base which describes the function of the studied object, thus enabling comparisons between different systems. In comparisons between plantbased and animal-based foods, the environmental impact is often expressed per kilogram of food. This approach has been criticized for favoring foods with a higher water content over nutrientdense products. Clearly, a kilogram of lettuce is not nutritionally equivalent to a kilogram of meat. Using a FU involving only GHG per kilogram of a food item, or even per kilogram of protein, may lead to the conclusion that plant alternatives are always better than those of animal origin as they do not appropriately account for protein quality, the nutritional density of ASF, or the relative availability of micronutrients. Functional units that relate to the energy content or, more recently, nutritional quality of foods (amounts and shares of various macronutrients and micronutrients) per unit of energy have been proposed to provide more nutritionally relevant comparisons (Doran-Browne et al., 2015). The use of emissions/unit nutrient density allows food products with very different nutritional profiles and water content to be more easily and equitably compared. It may also be more beneficial to consider different protein sources in terms of the additional macro- and micronutrients they provide to humans. Considering nutritional elements may also provide a better estimate of the amount of plant and ASF needed to meet the nutritional requirements of a growing global population (White and Hall, 2017; Liebe et al., 2020). When LCAs are calculated to consider aminoacid composition and nutrient density (e.g., iron, vitamin B12, zinc, retinol, and aminoacids), the footprint of animal foods becomes more similar to plantbased foods because animal foods contain highlevels of essential amino acids and micronutrients (Drewnowski et al., 2015; Tessari et al., 2016).

As with all novel meat and milk alternatives, cell-cultured meat has found a place in the conversation surrounding sustainable diets and their environmental impacts. While cell-cultured meat production has yet to be achieved at scale, there have been a few anticipatory LCAs performed to determine the potential environmental impacts of cell-cultured meat and compare them to other sources of protein (Tuomisto and de Mattos, 2011; Tuomisto et al., 2014; Mattick et al., 2015; Smetana et al., 2015). These stud-

**Table 1** Environmental impacts of cell-cultured meat from life cycle assessments. <sup>1</sup>

	GHG Emissions(kg CO <sub>2</sub> eq)	Energy Use (MJ)	Water Use (L)	Land Use (m <sup>2</sup> )	EP (g PO <sub>4</sub> eq)	AP (g SO <sub>2</sub> eq)	ODP (µg CFC11eq)
Mattick et al. (2015)	7.5	106	217.02	5.5	7.9	70.2	309
(Best case-worst case)	(3.2-22.3)	(44-316)					
Smetana et al. (2015)	23.9-24.64	290.7-373	_	0.39-0.77	-	_	<del>-</del> .
Tuomisto and de Mattos (2011)	1.69-2.66	22.8-38.3	282-651	0.19-0.23	_	_	
Tuomisto et al. (2014)	2.27-4.38	34.5-60.9	332.5-843.8	0.46 - 2.82	_	_	_
Sinke and Odegard (2021)	2.5-13.5	147-264	42-56	1.7-1.8	_	_	_
(Best case-worst case)	(2.1-22.6)	(124-445)					

Abbreviations: GHG = greenhouse gas; EP = eutrophication potential; AP = acidification potential; ODP = ozone depletion potential; CFC11 = chlorofluorocarbon-11

ies assessed GHG emissions, energy, water, and land use. In addition to these impacts, Mattick et al., 2015 assessed eutrophication, acidification and ozone depletion potentials. They present a wide range in results for each life cycle impact category (Table 1), highlighting the variability in the assumptions that were made as to how cultured meat will actually be produced when it is at commercial scale, as well as inherent variability in LCAs as a whole. A recent prospective LCA on cultivated meat was conducted by the Dutch research and consultancy firm CE Delft and commissioned by the Global Action in the Interest of Animals and The Good Food Institute, which is a non-profit advocacy group working internationally to accelerate alternative protein innovation (Sinke and Odegard, 2021). Although it is not peer-reviewed, it is included here in the interests of completeness (Table 1). The FU in that study was 1 kg of high-protein product (meat cells) and the system boundaries were from cradle to facility gate.

System boundaries set in each study, and assumptions around how cultured meats will be grown (e.g. requirement for growth factors in the culture media) were a primary driver for variability observed in the results. Smetana et al. (2015) performed a 'cradle-to-plate' analysis, accounting for not only the production of cell-cultured meat but also consumer preparation of the meat. The other analyses performed a 'cradle-to-factory gate' analysis in which transportation from factory to consumer and cooking of the product are not included in the analysis. The additional transportation accounted for in Smetana et al. (2015) is in part why greater GHG emissions were reported. While cooking will add to energy use associated with the product, this extra step in the LCA does not fully account for the reported increased energy needs. This is in part because Smetana et al. (2015) included the production of growth factors in their system boundaries. Sinke and Odegard (2021) concluded that the recombinant proteins and growth factors required to grow 1 kg of cultured meat were the main contributors to its carbon footprint. Other studies state that the carbon footprint of this step is insignificant in relation to the entire supply chain and did not include it in their analysis (Tuomisto & de Mattos, 2011). Differences in land use across studies can primarily be attributed to the feedstock and culture inputs that were assumed to have been used to successfully culture meat. Those with higher land-use values utilized corn and soybean as their base feedstock inputs, while lower values utilized cyanobacteria as the base feedstock.

Moving forward with the scaling up of cultured meat production facilities, the source of energy will be a primary area of concern. The environmental impacts of cell-cultured meat will be highly dependent on the source of energy utilized for the production processes and making the ingredients used to feed the cells, and whether these energy sources can be decarbonized (Lynch and Pierrehumbert, 2019). The prospective anticipatory cultured meat LCA reported that 'if renewable energy is used to power cell-cultured meat production, this could reduce global warming impacts by 17%, 52%, and 85–92% versus conventional chicken, pork, and beef production, respectively' (Sinke and Odegard, 2021).

To demonstrate the challenges with comparing results across studies, we performed a literature review of LCAs (and other similar assessments) on the environmental impacts of producing various food protein sources. We present the system boundaries used in 54 analyses (5 cell culture, 11 beef, 3 ground beef, 3 dairy beef, 4 pork, 8 plant, 3 insect, 7 dairy, 3 chicken, 4 egg, and 3 lamb; Table 2) and the impacts calculated per kg of product FU (Fig. 2) for: (A) GHG emissions; (B) land use; (C) water use; (D) energy use; (E) eutrophication potential; and (F) acidification potential. These studies are by no means exhaustive of all the LCAs that have been performed on ASF products, rather they serve as an example of the breadth of LCA related research performed on different protein sources, and the variability across all studies that may result from differing system boundaries. Some trends are obvious and biologically based, for example ruminants produce more GHG than monogastrics due to rumination and require more land as a result of being grazing herbivores. Emission intensities of ruminant milk are typically lower than beef, although the latter's emissions are minimized when fed a high-quality ruminant diet, making it more comparable with milk (Herrero et al., 2013). Production systems in the developed world typically have lower emission intensities than those in developing regions. The results demonstrate wide ranges to some degree for all forms of protein, and illustrate the variation that can occur depending upon the LCA system boundaries and assumptions. On average, cultured meat production appears to produce similar GHG emissions to most other ASF protein sources, with the exception of ruminant meats from extensive systems utilizing low quality forage. Plant products generally have a lower GHG than ASF, which is expected given they are at a lower trophic level. However, this framing is highly influenced by a specific definition of the global warming potential (GWP), one that has been increasingly questioned by several authors (Allen et al., 2016; Allen et al., 2018; Cain et al., 2019).

## Greenhouse gasses

When considering GHG emissions, LCAs typically use the GWP<sub>100</sub> metric. This metric is assessed over a 100-year time horizon. By this approach, the global warming potentials of methane  $(CH_4)$  and nitrous oxide  $(N_2O)$  relative to carbon dioxide  $(CO_2)$ are multiplied by 28 and 265, respectively (Myhre et al., 2014). The authors of the Intergovernmental Panel on Climate Change Fifth Assessment Report themselves state that the GWP<sub>100</sub> climate metric should not be considered to have any special significance. However, GHGs vary in their atmospheric lifetime, and importantly, for ruminant production systems, CH4 is a short-lived climate pollutant (SLCP) with an atmospheric lifetime in the order of only 12 years (Myhre et al., 2014). This has led some researchers to suggest that a new expression of the global warming potential metric, known as GWP\* (Allen et al., 2018), should be used to compare the temperature response from a change in rate of emission of SLCPs to the temperature response from a pulse emission of carbon dioxide. The very long-term climate impact of CO<sub>2</sub> is the reason

<sup>&</sup>lt;sup>1</sup> Results reported using a functional unit of kg of cell-cultured meat produced.

**Table 2**The system boundaries of the studies investigating environmental impacts of various protein sources depicted in Fig. 2.

Study by Protein Source	System Boundaries	Study by Protein Source	System Boundaries
Cell Cultured Meat		Plant-Based Meat	
Mattick et al. (2015)	Cradle-to-factory gate (excl. growth factors)	Dettling et al. (2016) Fresán et al. (2019)	Cradle-to-grave Factory gate-to-factory gate (excl. crop production
Sinke and Odegard (2021)	Cradle-to-factory gate		before factory)
Smetana et al. (2015)	Cradle-to-consumption		
Γuomisto and de Mattos (2011)	Cradle-to-factory gate (excl. growth factors, vitamins, and cell culture)	Goldstein et al. (2017)	Cradle-to-farm gate (excl. transport and packaging preparation, and disposal)
Γuomisto et al. (2014)	Cradle-to-factory gate (excl. growth factors and vitamins)	Heller and Keoleian (2018)	Cradle-to-distribution
n c1		Khan et al. (2019)	Cradle-to-factory gate
Beef <sup>1</sup> Asem-Hiablie et al. (2019)	Cradle-to-consumption	Mejia et al. (2020)	Factory gate-to-factory gate (excl. crop production before factory)
Broom (2019)	Cradle-to-slaughter		
Goldstein et al. (2017)	Cradle-to-farm gate (excl. transport and packaging,	Smetana et al. (2015)	Cradle-to-consumption
	preparation, and disposal)	Van Mierlo et al. (2017) Insect-Based Meat	Cradle-to-consumption
M	Conflicts from outs (seed about to a)	Smetana et al. (2015)	Cradle-to-consumption
Murphy et al. (2017)	Cradle-to-farm gate (excl. slaughter)	Smetana et al. (2019) Van Mierlo et al. (2017)	Cradle-to-factory gate Cradle-to-consumption
Nieto et al. (2018)	Cradle-to-farm gate (excl. finish phase)	Milk	Cradic-to-consumption
(2010)	g ( prinor)	Cederberg et al. (2009)	Cradle-to-retail
Dettling et al. (2016)	Cradle-to-grave	Eide (2002)	Cradle-to-end of life
Khan et al. (2019)	Cradle-to-slaughter	Gerber et al. (2010)	Cradle-to-retail
Rotz et al. (2019)	Cradle-to-farm gate (excl. slaughter)	Guinard et al. (2009)	Cradle-to-end of life
Stackhouse-Lawson	Cradle-to-farm gate (excl. slaughter)	Naranjo et al. (2020) Thoma et al. (2013)	Cradle-to-farm gate Cradle-to-farm gate (excl. transportation and
et al. (2012)	cradic-to-larin gate (exci. staughter)	moma et al. (2015)	processing)
Stanley et al. (2018)	Gate-to-gate (finish phase only)		
Tichenor et al. (2017)	Cradle-to-farm gate (excl. slaughter)	Wirsenius et al. (2020)	Cradle-to-farm gate (excl. transportation and processing)
Ground Beef¹		Chicken	
Goldstein et al. (2017)	Cradle-to-farm gate (excl. transport and packaging,	Goldstein et al. (2017)	Cradle-to-farm gate (excl. transport and packaging
	preparation, and disposal)		preparation, and disposal)
Khan et al. (2019)	Cradle-to-factory gate	Dettling et al. (2016)	Cradle-to-grave
Dettling et al. (2016)  Dairy Beef <sup>1</sup>	Cradle-to-grave	Smetana et al. (2015) Eggs	Cradle-to-consumption
Murphy et al. (2017)	Cradle-to-farm gate (excl. slaughter)	Cederberg et al. (2009) Leinonen et al. (2012)	Cradle-to-retail Cradle-to-farm gate (egg production)
Stackhouse-Lawson	Cradle-to-farm gate (excl. slaughter)	•	
et al. (2012)		Mollenhorst et al. (2006)	Cradle-to-farm gate (egg production)
Tichenor et al. (2017)	Cradle-to-farm gate (excl. slaughter)	Pelletier et al. (2014)	Cradle-to-farm gate (egg production)
Pork			
Dettling et al. (2016)	Cradle-to-grave	Wiedemann and McGahan (2011)	Cradle-to-farm gate (egg production)
Goldstein et al. (2017)	Cradle-to-farm gate (excl. transport and packaging, preparation, and disposal)	Verge et al. (2009)	Cradle-to-farm gate (egg production)
Rudolph et al. (2018) Wirsenius et al. (2020)	Farrow-to-finish (excl. slaughter) Cradle-to-farm gate (excl. transportation and processing	Lamb Dougherty et al. (2019)	Cradle-to-factory gate (excl. consumer)

<sup>&</sup>lt;sup>1</sup> Beef production includes both ground beef and dairy beef within respective studies; however, both dairy and ground beef have been separated in order to provide further analysis.

why climate stabilization depends on actions to achieve net zero emissions of  $CO_2$ . As an example, GHGs from Australian livestock production - beef cattle, sheep meat, chicken meat, pig meat, eggs and milk – were assessed using both  $GWP_{100}$  and  $GWP^*$  metrics from 1990 to 2018. In the case of sheep meat production, the industry was assessed as emitting 10.3 MMt  $CO_2$ eq in 2018 using the  $GWP_{100}$  metric. However, using the  $GWP^*$  climate metric, the GHG emission footprint was equivalent to the removal of 2.85 Mt  $CO_2$  in part because of the degradation of historical  $CH_4$  emissions from a larger national sheep flock in the past (Ridoutt, 2021).

These authors make the point that avoiding ASF based on the GWP<sub>100</sub> metric may result in trading a short-term climate benefit

from reducing short-lived  $CH_4$  emissions, with a longer-term problem of increased  $CO_2$  and  $N_2O$  emissions, making climate stabilization even more difficult. This has implications for sustainable intensification approaches that decrease the emission intensity of ASF by substituting  $CO_2$  emissions for SLCP. Interventions such as providing supplemental crop-based feed rations may appear to lower GHG using the GWP<sub>100</sub> metric, but they may actually be substituting a long-lived GHG for a SLCP. Perhaps even more paradoxical with prevailing thought, is that red meats from ruminants may actually outperform meat from monogastric animals (pigs and poultry) when using the GWP\* metric due to the latter's reliance on crop-based feed rations. These findings emphasize the impor-

A.L. Van Eenennaam and S.J. Werth

Animal 15 (2021) 100360

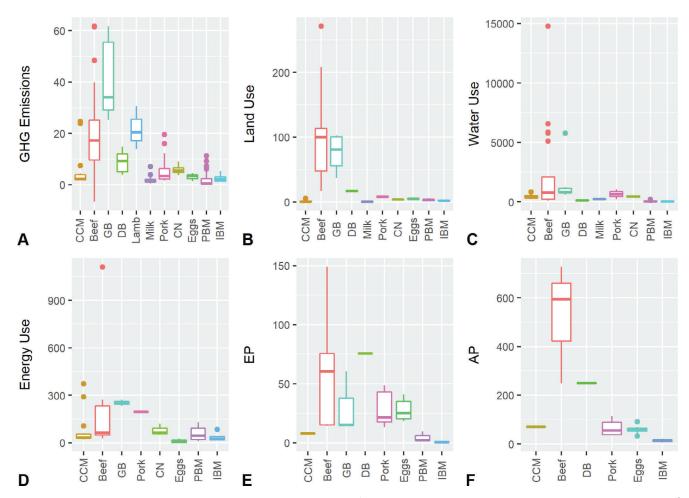


Fig. 2. Life cycle assessment results per kilogram of product for various protein sources<sup>1</sup>. (A) greenhouse gas (GHG) emissions (kg  $CO_2$ eq/kg product); (B) land use ( $m^2/kg$  product); (C) water use (L/kg product); (D) energy use (MJ/kg product); (E) eutrophication potential (EP; g  $PO_4$ e/kg product); and (F) acidification potential (AP; g  $SO_2$ e/kg product). All values have been adjusted to a 'per kg product' basis, but data have not been altered to account for other variables (e.g. system boundaries).  $^1CCM$  = cell-cultured meat; GB = ground beef; DB = dairy beef; CN = chicken; PBM = plant-based meat; IBM = insect-based meat.

tance of the choice that even a single climate metric can have on the outcomes and implications of LCA studies.

Additionally, LCAs typically do not consider changes in carbon stocks as it can be a difficult value to accurately obtain and characterize. Land use and land-use change is a large contributor to the GHG balance and within it, soil organic carbon is a major contributor. Soil carbon sequestration has the potential to be a valued sink for GHG emissions. Agricultural practices that influence soil organic carbon by reducing losses or increasing sequestration can play an important role in GHG mitigation. Accumulation rates vary with both climate and agronomic management. The amount of carbon that could be stored by the world's grazing lands is considerable, with estimates ranging from 0.04 to 1.1 Gt CO2eq/yr (Lal, 2004; Henderson et al., 2015), as a result of improved grazing management (0.148 Gt CO<sub>2</sub>eq/yr), and legume sowing (0.147 Gt CO<sub>2</sub>eq/ yr). These authors warn that the additional ruminant GHG emissions associated with higher forage output are likely to substantially reduce the mitigation potential of these practices, but could contribute to more GHG-efficient livestock production (Henderson et al., 2015). There are several factors that influence whether grazing impacts soil organic carbon including moisture, soil type and carbon saturation levels, and plant species composition (Buckley Biggs and Huntsinger, 2021). Researchers in the soil and range science communities have found the impacts of grazing systems on soil organic carbon to be highly variable. The 2016 technical standard for soil carbon, ISO 14067, prescribes that emissions and removals due to changes of soil organic carbon under ongoing land use should be included in carbon footprints (Sevenster et al., 2020).

Stanley et al. (2018) found that cattle finished on pasture using an adaptive multi-paddock grazing strategy lead to improved soil organic carbon, thereby presenting a situation where beef production produced net negative GHG emissions in a gate to gate finish phase analysis. This means that within the system boundaries set for this analysis, more carbon was sequestered than was emitted. The importance of characterizing carbon sequestration in beef systems was further highlighted in Rowntree et al. (2020), where inclusion of soil organic carbon resulted in a 113% reduction in  $GWP_{100}$  (33.55 kg  $CO_2eq/kg$  carcass weight to -4.4 kg  $CO_2eq/kg$ carcass weight). Similarly, improved soil carbon sequestration was observed for cattle raised in a pasture-based system (Thorbecke and Dettling, 2019) and bison under an adaptive multi-paddock grazing strategy (Hillenbrand et al., 2019). While enteric CH<sub>4</sub> emissions may be increased from pasture raised animals compared to feedlot finished cattle, if carbon sequestration from pasture systems are accounted for, then animal emissions can be offset, resulting in an overall net negative GWP<sub>100</sub> for specific situations. This offset would be even greater if GWP\* was used in the place of GWP<sub>100</sub>. Recently, the GWP\* methodology was used in combination with consideration of the soil organic carbon from associated pastures to examine the 1990-2018 contribution of European dairy small ruminant systems to additional atmosphere

A.L. Van Eenennaam and S.J. Werth

Animal 15 (2021) 100360

warming levels. By this metric, from 1990-2018, the whole European sheep and goat dairy sector did not contribute at all to global warming (Del Prado et al., 2021). In comparison with other protein sources, ruminant production systems present a unique opportunity to have a climate impact consistent with CO<sub>2</sub> removal if herd sizes are decreasing (which degrades historical methane emissions), and soil organic carbon is sequestered. However, it is important to note that carbon sequestration rates can be variable over time depending on factors such as historic use of land, depth, clay content and mineralogy, soil type, water availability, nutrient reserves, landscape position, and the antecedent stock of soil organic carbon (Machmuller et al., 2015; Lal, 2018). It should also be noted that some have criticized the GWP\* metric as being unfair because it gives advantages to countries that have had historically high CH<sub>4</sub> emissions (Rogelj and Schleussner, 2019). These authors make an argument that 'applying novel metrics to a predefined policy context is problematic if no appropriate measures are taken to ensure internal consistency with the earlier use of other metrics in that same policy context. In absence of such appropriate measures, policy targets can be re-interpreted without clear scientific or moral reasoning.' This critique can be applied to many of the metrics that are currently being employed to classify sustainable diets.

The Global Livestock Environmental Assessment Model is a modeling framework developed within the Animal Production and Health Division of the Food and Agriculture Organization of the United Nations (FAO) (MacLeod et al., 2018). The Global Livestock Environmental Assessment Model uses an LCA approach following guidelines issued by the Intergovernmental Panel on Climate Change, which means that the assessment includes both direct emissions from animals and indirect emissions both upstream and downstream. This approach differentiates key stages within livestock agrifood systems, such as feed production, processing and transport; animal production, animal feeding and manure management; and the processing and transport of products. The Global Livestock Environmental Assessment Model 2 has a base year of 2010 and uses CO2eq/kg protein calculated as GWP<sub>100</sub> (Fig. 3). These numbers hide large variations across different production systems. For example, the 295 kg CO<sub>2</sub>eq/kg protein average for beef ranges from 93 in feedlot systems to 434 in grassland systems. This reflects different agro-ecological conditions, farming practices and supply chain's management. It is within this gap between high and low emission intensities where opportunities for mitigation can be found. The estimation for mitigation is around 33 percent, or about 2.5 Gt CO<sub>2</sub>eq, with respect to the baseline scenario. This figure arises from the assumption that producers in a given system, region and agro-ecological zone would apply the practices of the 10th percentile of producers with the lowest emission intensities, while maintaining constant output. The Global Livestock Environmental Assessment Model 3, currently under development by the FAO with a base year of 2015,

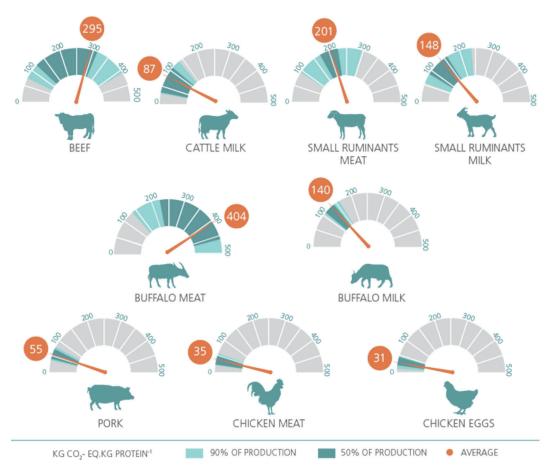
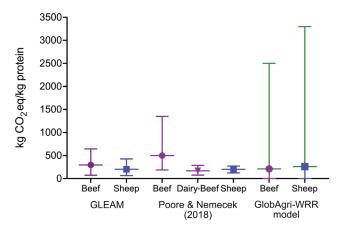


Fig. 3. Global greenhouse gas emission intensities by commodity expressed on a per kg protein basis. Averages (orange) are calculated at global scale and represent an aggregated value across different production systems and agro-ecological zones. Emission intensities vary greatly among producers with 90% of production occurring within the blue-shaded region, and 50% of production occurring within the dark blue bounds. This reflects different agro-ecological conditions, farming practices and supply chain management. It is within this gap between high and low emission intensities that opportunities for mitigation can be found. The estimation for mitigation is around 30%, or about 2.5 Gt CO<sub>2</sub>eq if producers in a given system, region and agro-ecological zone were to apply the practices of the 10th percentile of producers with the lowest emission intensities, while maintaining constant output, with respect to the baseline scenario. (GLEAM http://www.fao.org/gleam/results/en; Accessed August 8 2021)

A.L. Van Eenennaam and S.J. Werth

Animal 15 (2021) 100360



**Fig. 4.** Greenhouse gas emissions per kg protein for beef (purple) and sheep (blue) meat obtained using three different models. The average and range are shown for the FAO's GLEAM 2.0 life cycle assessment (MacLeod et al., 2018) and for Poore and Nemecek (2018). The GlobAgri-WRR model (Searchinger et al., 2019) provides a single global figure which includes agricultural production (purple circle, blue square for beef and sheep, respectively) plus the opportunity cost of agricultural land-use change (green). Inclusion of this land-use opportunity cost increases this metric by a factor of at least 5-fold relative to the average value of the other two sources for these ruminant sources of protein.

plans to include modules on enhanced accounting of nutrient use and carbon sequestration in livestock systems.

Other groups have developed their own models to arrive at environmental metric estimates. Poore and Nemecek (2018) consolidated data on the multiple environmental impacts of ~38 000 farms producing 40 different agricultural goods around the world in a meta-analysis comparing various types of food production systems. They found that impacts varied 50-fold among producers of the same product, creating substantial mitigation opportunities. They estimated that CO<sub>2</sub>eq/kg protein from a beef herd ranges from 202 (10th percentile) to 1 052 (90th percentile), with an average of 499 CO<sub>2</sub>eq/kg protein (Fig. 4) which is higher than the numbers calculated by the Global Livestock Environmental Assessment Model 2 (Figs. 3 and 4). Almost all of the variation in this estimate was due to differences in production systems, with the major driver of variance in these numbers being whether arable land was part of the production system. Feeding ruminants with highenergy, low-cellulose feed produced on arable land decreases the emission intensities associated with their milk and meat. The GlobAgri-WRR model from the World Resources Institute (Searchinger et al., 2019) has an estimate of 2 500 kg CO<sub>2</sub>eq/kg edible protein for beef, and a whopping 3 300 kg CO<sub>2</sub>eq/kg edible protein for lamb (Fig. 4). This model, which is not described in the peer-reviewed literature, includes a statement 'we believe that all or virtually all dry grazing land available in a country is used today, so that increases in grassland areas must come from wetter systems (humid or temperate). We also believe that because dry grazing lands have little alternative use, they would continue to be used even if demand for milk or ruminant meat declined. We therefore program the model so that changes in supply of milk or ruminant meat do not come from increases or decreases in arid grazing systems and instead result in changes in humid and temperate production systems.' It is therefore assumed that for each additional kg of ruminant protein produced, there is land-use change occurring either directly or indirectly through deforestation elsewhere to replace pasture/ cropland. This assumption particularly impacts sheep production, as small ruminants tend to survive in the most arid and least productive landscapes, resulting in large areas of land being required to produce one kg sheep meat. Poore and Nemecek (2018) estimated this number to be an average of 185 ha/ton protein with a range from 24 (5th percentile) to 362 (95th percentile). The assumptions of the GlobAgri-WRR model dramatically increase the  ${\rm GWP}_{100}$  of ruminant source protein as ruminants uniquely occupy grasslands. In this model, ruminant systems are not credited with any soil organic carbon occurring on grazing lands, and irrespective of actual location, they are assigned the opportunity cost of increased emissions from land-use change in humid and temperate production systems. Amortizing projected land-use change to ruminant products violates the system boundaries of the other GHG emission inventories.

These competing metrics are difficult for a non-expert audience to disentangle. If we consider sheep meat production in Australia, as an example, in 2018, the industry produced 1.62 Mt of live weight which generated 784 000 t CO<sub>2</sub>; 299 000 t CH<sub>4</sub>; and 3 810 t N<sub>2</sub>O (Ridoutt, 2021). Using the GWP<sub>100</sub> metric, the industry therefore produced 10.3 Mt CO<sub>2</sub>eq (17.4 kg CO<sub>2</sub>eq/kg edible product or ~67 kg CO₂eq/kg protein), equating to a little less than 2% of the country's 2018 emissions. If however, the GWP\* metric is used, which considers the degradation of historical methane emissions (Lynch et al., 2020), the industry resulted in a net decrease of 2.85 Mt CO2eq (-4.80 kg CO2eq/kg edible product or −18.5 kg CO<sub>2</sub>eq/kg protein) (Ridoutt, 2021). Conversely, using the GlobAgri-WRR model that considers the opportunity costs of the sheep being on non-arable grazing land through 2050, the industry produced 151 632 t edible protein (1.62 Mt \*0.36 yield of edible product \* 0.26 kg protein/kg edible product) which would result in 500 Mt  $CO_2$ eq (151 632 t × 3 300 t  $CO_2$ eq) (Fig. 4). This equates to around 90% of the entire country's 2018 CO<sub>2</sub>eq emissions of 558.4 Mt CO<sub>2</sub>eq (GWP<sub>100</sub>)! Other groups have developed estimates of GHG attributable to livestock that include respired CO2 in GHG emissions (Goodland and Anhang, 2009), an approach that has been criticized as a major deviation from international protocols because the amount of C in feed consumed and CO2 emitted by livestock are broadly equivalent (Herrero et al., 2011). When there is such variation in the assumptions and system boundaries driving these varying GHG metrics and the time frames they consider, value judgements will be embedded into which metric should be used. This will undoubtedly increase distrust in the figures, and potentially alienate landholders and livestock keepers whose cooperation is needed to adopt GHG mitigation interventions.

#### Resource use

Total land use is another category where meat alternatives have a lower number than most ASF protein sources (Fig. 2). The total land-use metric does not differentiate between arable and nonarable land. The production of global animal feed requires 2.5 billion ha of land, which is about half of the global agricultural land area. Most of this area, 2 billion ha, is grassland, of which about 1.3 billion ha cannot be converted to cropland. This means that 57% of the land used for feed production is being grazed by ruminant production systems (Mottet et al., 2017). If ruminants were removed from this land, it would produce no human food, and this would impact the livelihoods of millions of smallholder livestock keepers (Mapiye et al., 2020). There is no reason to conclude that food production on non-arable land is less sustainable than food production on well-managed arable cropland, simply because the former achieves lower yields and therefore requires more land use per unit of production. If minimizing total land use is equated to improved sustainability, it leads to the conclusion that food should be intensively produced on the smallest amount of arable land possible. This is actually the model for intensive monogastric animal agriculture systems (poultry and pigs), which are not seen as sustainable by many due to their animal welfare and environmental externalities, and a reliance on the provision of feed grown on arable land that could have been used to grow human food. While it is undoubtedly true that cultured meat facilities will

A.L. Van Eenennaam and S.J. Werth

Animal 15 (2021) 100360

occupy less land than grazing ruminants, the actual environmental and biodiversity benefits that result from that will depend on how the land 'released' from livestock production will be utilized (Stephens et al., 2019; Tuomisto, 2019b).

There are ecosystem services resulting from grazing ruminants that maintain various habitats and species and which are therefore beneficial for biodiversity. The biophysical capacity of land to supply ecosystem services is not considered in LCAs. In the United States, beef cattle ranching actively grazed over 186 million ha in 2017, approximately half of the 363 million ha of total farmland. It is estimated the cumulative economic value of this grazed land use was \$24.5 billion; comprised of \$17.5 billion for wildlife recreation, \$3.8 billion for forage production, and \$3.2 billion for other ecosystem services related to the conservation of biodiversity (Maher et al., 2021). Similarly, beef cattle ranching was found to have a positive influence on biodiversity, habitat maintenance, cultural heritage, recreation and tourism in the Canadian prairie provinces of Alberta, Saskatchewan and Manitoba, which collectively support just over 80% of the Canadian beef herd (Pogue et al., 2018). Integrity and productivity of ecosystem goods and services from rangelands are critical to the livelihoods of over a billion people worldwide (Niamir-Fuller and Huber-Sannwald, 2020). Total abandonment of grazing in natural rangelands is likely to be an ineffective climate change policy (Manzano and White, 2019). Food is not the only output of agricultural systems, so life cycle impacts should perhaps be allocated to broader functional units than simply kg of food product to more fully account for the other outputs including rural livelihoods, cultures and landscape services associated with food systems.

Among LCAs, water is commonly characterized as 'green' or 'blue' or a combination of the two, where green water is rainwater and blue water is groundwater and surface water resources (Hoekstra, 2019). While ruminant meat utilizes substantially more water than cell-cultured meat and other proteins, the vast majority of this is green water (Fig. 2). Many studies characterize consumptive water use, where any water, green or blue, removed from stores will not return to the system. Considering water in this regard can become problematic as it does not take into consideration that green water is not in direct competition with water needed for other anthropogenic activities. Green water is inseparable from land, meaning water that falls on one pasture cannot fall on another, and as such it is a proxy land-use indicator. By definition, extensive systems on arid grazing lands will have a large green water footprint, as they occur on large acreages (Damerau et al., 2019). The water is largely returned to the very area where the precipitation fell, through urination, defecation, and respiration. The only green water 'leaving' the system is what is captured in weaned calves when they leave the ranch. Meanwhile, water needed for alternative meat manufacturing systems is blue water. This consumptive water use metric is not ISO compliant (International Organization for Standardization, 2014), as it does not differentiate between water use in regions of water scarcity from that in regions of abundance. It is therefore important to calculate a water scarcity footprint, where each instance of water use in the life cycle of a food product or a diet is multiplied by the relevant local Water Scarcity Index. Recently, the FAO LEAP partnership published their recommendations on water use assessment of livestock production and supply chains (Boulay et al., 2021)

Energy use is variable across all forms of protein sources (Fig. 2). A major factor affecting energy use when comparing these studies is whether or not cooking by the consumer is considered. Many studies end their system boundaries at factory or field gate, thus eliminating any energy needed to transport, store, or cook the protein. While these factors should be relatively similar across all protein sources, it is pertinent to note these distinctions when working to draw conclusions on energy use across studies. While

outliers for cell-cultured meat, about half of the data demonstrate that cell-cultured meat has the potential to be the most energy intensive protein. This is in part due to the large amount of energy required to run the bioreactors used to multiply cells. Studies to date only consider the creation of muscle tissue and do not consider additional energy required to create fat cells. Cell-cultured fat manufacturing platforms will require considerable optimization to identify appropriate cell lines, bioprocess strategies, and tissue engineering techniques to achieve simple systems that can cost-effectively scale (Fish et al., 2020).

Eutrophication potential and acidification potential are two LCA characterization factors that are studied less frequently than other factors. In the case of cell-cultured meat, Mattick et al. (2015) is the only study to consider eutrophication potential or acidification potential, making it difficult to make any accurate comparisons to other proteins. In general, animal sourced proteins have greater eutrophication potential and acidification potential than cellcultured meat or plant-based proteins. Emissions contributing to eutrophication potential and acidification potential in animal protein systems are primarily related to crop production for animal feeds and management of animal manure, respectively (Tichenor et al., 2017). As cell-cultured meat production will not directly require these inputs, it is likely that eutrophication potential and acidification potential will remain lower than animal proteins as production of cell-cultured meat is upscaled. Compared to all meat products, both cultivated and conventional, the environmental metrics and the carbon footprint of plant-based protein products are lower.

#### Human health and alternative meats

Human health is not typically a metric formally considered in LCAs, and yet this topic is perhaps the most contested literature that comes up in the discussion around alternative meats. Are ASF foods part of a healthy diet, and if so how much is too little or too much? As with the other metrics discussed, motivated reasoning can be used to pick a segment of the scientific literature that supports a particular world view. The EAT-Lancet Commission suggested that 'healthy diets have an appropriate caloric intake and consist of a diversity of plant-based foods, low amounts of animal source foods, unsaturated rather than saturated fats, and small amounts of refined grains, highly processed foods, and added sugars' (Willett et al., 2019). Review papers that identify red and processed meat as an intrinsic cause of chronic diseases based on observational studies (Micha et al., 2010; Chen et al., 2013; Abete et al., 2014; Wang et al., 2016; Schwingshackl et al., 2017) can be countered by review papers that do not find this association (Han et al., 2019; Zeraatkar et al., 2019a; Zeraatkar et al., 2019b), and randomized controlled trials that do not find an association between reduced meat consumption and adverse health outcomes (Thomson et al., 2014). These citations are by no means an exhaustive list of the extensive nutrition literature, but are meant to be illustrative of an unsettled scientific field, and the fact that much nutritional epidemiologic research often posits implausible estimates of benefits or risks associated with diet (Ioannidis, 2018). The strongest evidence of a specific adverse effect is the increased risk of colorectal cancer with high intakes of processed meat. Conversely, various forms of micronutrient deficiencies affect some two billion people globally, particularly in developing countries. The greatest health burdens of this 'hidden hunger' are caused by deficiencies in zinc, vitamin A and iron, which lead to impaired growth, compromised immune functions and, in the case of iron, impaired cognitive development and reduced work capacity. An important factor contributing to these deficiencies is the consumption of mainly plant-based diets that are low in micronutrients, and in such situations, ASF can help reduce childhood stunting

and malnutrition (Headey et al., 2018; Pimpin et al., 2019; Adesogan et al., 2020).

With regard to nutrition, a study of 137 plant-based meat substitutes (50 burgers, 10 ground, 29 sausages, 24 chicken, 9 seafood, 15 other) in Australia reported that the plant-based options were generally lower in kilojoules, total and saturated fat; but higher in carbohydrate, sugars, and dietary fiber as compared to meat. Less than a quarter of products were fortified with vitamin B12 (24%), iron (20%), and zinc (18%) (Curtain and Grafenauer, 2019). Consumers perceived that plant-based meat substitutes were healthier, but the wide variation in nutritional levels lends some support to the concern that consumers might run into nutritional deficiencies if assuming product equivalence when replacing ASF with plant-based products. Similarly, non-dairy milk beverages differ in their nutritional profiles (Clegg et al., 2021), and although most are fortified with calcium and vitamin D, the bioavailability of these substances after fortification has not been established (Singhal et al., 2017). In Spain, 54 soy beverages, 24 rice beverages, 22 almond beverages, 31 oat beverages, 6 coconut beverages, 12 miscellaneous beverages and 15 mixed beverages were analyzed, and the nutritional quality was found to be inferior to that of cow's milk and infant formula (Vitoria, 2017). There have been instances of nutritional disorders such as rickets in infants and toddlers fed predominantly or exclusively plant-based beverages (Le Louer et al., 2014; Vitoria, 2017). Baseline nutrition data for cell-based meat are not yet publicly available (Rubio et al., 2020).

An interesting summary of an Oxford-style debate outlining opposing views on the issue 'Children and adults should avoid consuming ASF to reduce the risk for chronic disease' was published in The American Journal of Clinical Nutrition (Barnard and Leroy, 2020). After framing this issue as a binary choice, the most common YES and NO arguments were outlined. One way forward, according to both sides, was to perform research studies comparing various formulations of omnivorous and vegan diets, while controlling for confounders as much as possible. It was agreed that such studies should involve participants at a variety of stages of life, and from a variety of demographic and cultural groups. Further, it was agreed that metrics beyond BW, lipids, and other cardiometabolic endpoints were needed to examine other healthrelated conditions, particularly cognitive, digestive, hormonal, and autoimmune diseases. However, as acknowledged in the article, 'such research may not resolve discordant worldviews, ethical frameworks, and philosophical investments that have marked this debate.' And that is really the challenge in discussions around meats and alternative meats. When different parties are coming at this issue with conflicting worldviews, no amount of data is going to reconcile these differences. So if science cannot help address ongoing points of disagreement, what are the underlying influences of these disparate world views, and are there any points of agreement?

#### Wizards, prophets and magicians

Garnett (2013) argues that three main framings can be applied to the challenge of how to reduce the environmental impact of feeding people better; namely a production challenge (wizard), a consumption or demand-side challenge (prophet), or a socioeconomic challenge, which I will term the magician. These terms in parenthesis reference Charles Mann's (2018) book 'The Wizard and the Prophet', which examines historical debates about agriculture and ecology through two distinct framings – pro-growth, prodevelopment, pro-technology wizards as exemplified by Norman Borlaug on one side; versus tradition-oriented, techno-skeptical, limits to growth-minded prophets as exemplified by William Vogt on the other. Briefly, the wizard is the sustainable intensification lens, the prophet envisions changing the dietary drivers of food

production; and the magician, absent from Mann's book, sees more localized, diverse systems as better able to deliver the full range of micronutrients needed for good health – especially for women and children. This latter perspective tends to invoke a romanticized vision around smallholder production that can include some overly optimistic prophesying, hence the magician framing, and advocates for changes to the socioeconomic governance of the food system. None of these worldviews are necessarily mutually exclusive, and binary framings that pit them against each other tend to needlessly back proponents into artificially constructed corners. They each have their strengths and weaknesses; however, it is often difficult to productively engage people with alternative worldviews into considering how all three framings might be required to address future protein needs.

The promissory narrative associated with alternative meats is that this field provides 'kinder, healthier, fairer, tastier, safer and more sustainable approaches to conventional livestock products thus collectively work to make the ultimate promise of a better food system for all, and in turn a better food future for all' (Sexton et al., 2019). In some ways, it employs a wizard framing to solve the prophet's problem. This view tends to paint conventional livestock systems as outdated and primitive (Sexton et al., 2019). The response from some in the livestock industry has been to label alternative meats as 'artificial' and 'unnatural' in comparison with conventional ASF, due to the techno-scientific nature of their production. However, denigrating techno-scientific innovations in food production may backfire on the livestock sector by reinforcing the 'artisanal reaction'. This is a term used to describe the trend where consumers turn toward products that are apparently delivered by simpler and more natural processes in response to food scares alleged to be associated with overly industrialized production processes (Murdoch and Miele, 2004). If proponents of conventional meats demonize the use of GMOs in alternative meat production systems, they should not be surprised when consumers then fear the use of genetic engineering in agricultural production systems (Sexton et al., 2019). If cultured meats are framed as 'unnatural' to invoke unfounded health implications, then it is increasingly likely modern food production and processing methods will be feared, to the detriment of innovation in all production systems. Likewise, when alternative meat companies demonize animal agriculture by greatly overstating the impacts of livestock production based on widely debunked estimates in their marketing pitches [e.g. '51% of GHG emissions driven by livestock rearing and processing' whereas the number according to the FAO is 14.5% (Sexton et al., 2019)], they needlessly create antagonists. They should therefore not be surprised when the livestock sector, a potential ally in delivering on the mitigation efforts necessary to accomplish the shared goal of reduced GHG emissions and the primary caretakers of the land that will be 'released' from livestock production, become disenfranchised adversaries.

Sexton et al. (2019) noted that some alternative protein companies are suggesting they can actually provide both nutritional salvation and economic development for the hungry poor by providing low-cost, nutritionally rich and culturally tailored protein products in local Southern hemisphere contexts. These authors warn that alternative protein visions of feeding the world require critical reflection given the history of how productivist interpretations of global food security and single-sector economic development approaches have led to loss of livelihoods, increased inequality and land degradation for many pastoral communities in developing countries. They also note that advertising around alternative meats is designed to appeal to male carnivorous Western consumers who can continue to experience the taste and sensory pleasures of ASF by switching to alternative meats. The metaphoric link between meat and maleness in Western cultures has been noted by a number of scholars in the social sciences

(Sobal, 2005). Marketers use this to target those who subscribe to the metaphor and are therefore likely to be predisposed toward trying an alternative meat product (Rozin et al., 2012). It is perhaps no accident that a number of professional athletes have been employed to serve as spokesmen for alternative meat companies, alongside celebrity carnivore male chefs. This is reminiscent of the aforementioned 1960s advertising campaign to convince American men that consuming chicken was 'manly'. Ironically, this framing omits the very demographics – women and children in LMIC – who could most benefit from ASF and who have particular difficulty in obtaining adequate energy and essential micronutrients solely from bulky, plant-based diets (Dror and Allen, 2011).

There are other complex issues around conventional and alternative meat discussions that are often boiled down to an overly simplistic framing, or not even considered in the discussion. These include animal welfare, use of antibiotics, zoonotic disease, microbial contamination, food safety, biodiversity, ecosystem services, social justice, the slaughter of animals, the religious role of animals, the cultural appropriateness of food, patents, food sovereignty, and food choice. The current dichotomous framing of plant versus animal; synthetic versus natural; extensive versus intensive; clean versus dirty; GMO versus organic; alternative versus real; tradition versus progress is not helpful for discussions relating to food systems. These discussions are not unidimensional. And the framing will ultimately impact which stakeholders are willing to participate in proposed solutions. It is possible to simultaneously work to improve the efficiency of animal source foods, plant source foods, and cultured meat production systems without denigrating any of them. There is value in seeking to move beyond 'us versus them' framings, focusing on shared values around sustainable meat futures (Sexton et al., 2019).

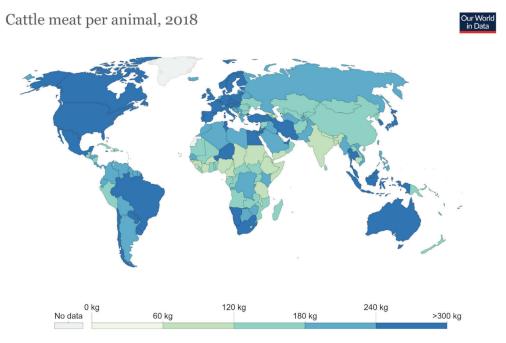
## **Moving forward**

The FAO (FAO, 2019) outlines five practical actions that can be widely implemented for measurable and rapid impacts on livestock emissions. These include (1) boosting efficiency of livestock

production and resource use; (2) intensifying recycling efforts and minimizing losses for a circular bioeconomy; (3) capitalizing on nature-based solutions to ramp up carbon offsets; (4) striving for healthy, sustainable diets and accounting for protein alternatives; and (5) developing policy measures to drive change.

Boosting efficiency sounds very much like the production challenge framing. This could include technological innovations in feeding, breeding, genetics, animal health, management, and information technology to reduce environmental impacts relative to the amount of livestock product. It is this approach that has already dramatically reduced the emission intensity of ASF in MHIC, especially in so-called 'high-input' or intensive systems in which external inputs such as supplementary feeds, veterinary medicines and advanced breeding and reproductive technologies are relatively easily obtainable and widely used. This is reflected in the global yield per animal maps for beef (Fig. 5) and milk (Fig. 6).

Cattle, as large ruminants, hold an iconic position in many climate and sustainability discussions. The FAO estimated cattle numbers at 1.511 billion head in 2019 (FAOSTAT, 2020) and Fig. 7 shows global cattle numbers versus beef production for some of the major cattle producing countries and regions of the world. The United States is the largest single beef-producing country followed by Brazil. These two countries, along with Europe, collectively produced approximately 50% of the world's beef in 2020. However, this number does not reflect the distribution of global cattle populations. Brazil is the country with the largest number of cattle at 215 million head, with India coming in second at 193 million head. It should be noted that these figures are only for cattle. It does not include the world buffalo stocks of 204 million animals, of which 110 million head live in India. Likewise, Pakistan has almost the same number of buffalo (40 million) as cattle (48 million) totaling 88 million head. Likewise, beef production numbers are usually referring to cattle. If buffalo meat and edible offal from both are included in the production numbers, the values for India and Pakistan more than double from 0.9 and 1.1, to 2.9 and 2.4 MMt, respectively.



Source: UN Food and Agricultural Organization (FAO)

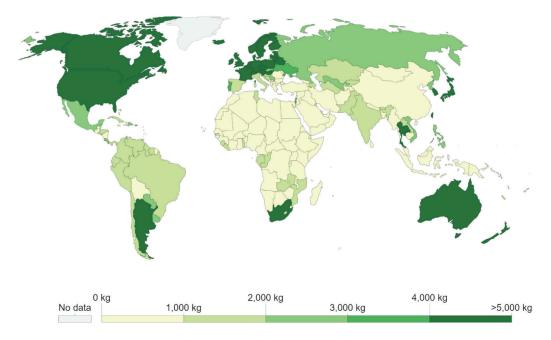
OurWorldInData.org/meat-production • CC BY

Fig. 5. 2018 global beef productivity (kg of beef per animal). Reproduced from Ritchie and Roser (2019) with data from FAO (2020).

## Milk per animal, 2018

Milk yields are measured as the quantity of milk produced per animal.





Source: UN Food and Agricultural Organization (FAO)

Note: Data is measured as the weighted-average of production across all milk-bearing livestock

OurWorldInData.org/meat-production • CC BY

Fig. 6. 2018 global milk productivity (kg of milk yield per animal). Reproduced from Ritchie and Roser (2019) with data from FAO (2020).

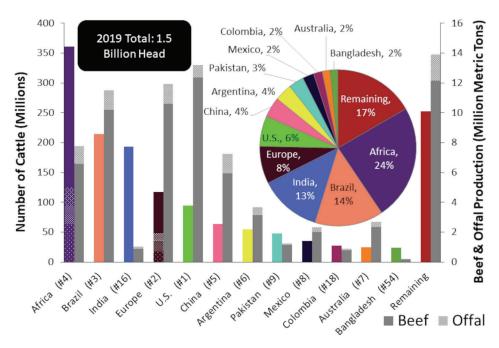


Fig. 7. Comparison of percentage of cattle population (pie chart), cattle numbers and beef/offal production for countries and regions of the world. World beef production rank is listed next to country/region name. The cattle number bars representing regions (i.e., Africa and Europe) are ordered by the top 3 countries (patterns) and then the remaining countries of a region (solid). Data are from FAO (2020).

Africa collectively is home to 361 million cattle, 24% of the global population, and 3.5 million buffalo located mainly in Egypt. In Africa, the agricultural sector is the largest sector of the domestic economy, and livestock are a crucial component of that sector and account for more than 70% of African agricultural GHG emis-

sions. Ethiopia has 63 million cattle, the most of any African country, followed by Sudan and Chad each at 31 million head, Tanzania with 28 million head, Kenya and Nigeria each with 21 million head, Uganda with 16 million head, and Niger, South Sudan, South Africa, Mali and Burkina Faso each with 10–15 million head. Over 250 mil-

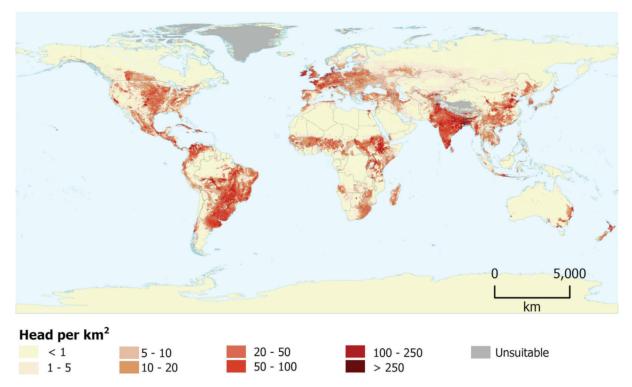


Fig. 8. Global distribution of cattle. Reproduced from Robinson et al. (2014).

lion cattle live outside of the top beef producing countries and the African continent, reflecting the fact that cattle are found throughout the world, in almost all climatic zones, with the exception of high elevations. They live in more than 200 countries around the world, and they have been bred for adaptations to heat, cold, humidity, extreme diet, water scarcity, mountainous terrain, dry environments, and for general hardiness. Cattle also produce milk, and in 2019, the world production of fresh cow milk was almost 715 MMt, of which 90 MMt was produced in India, and 36 MMt

was produced in Africa. As with meat, a lot of milk is also produced by buffalo in India, 92 MMt according to the FAO, making India the largest dairy country, producing an estimated 21% (188 MMt) of the world's 883 MMt of milk from all species in 2019. The highest cattle densities are found in India, the East African highlands (particularly in Ethiopia), Northern Europe and in South America (Fig. 8).

Only 7% of beef (2% cattle population) is produced in intensive systems. Likewise, approximately 88% of milk production occurs

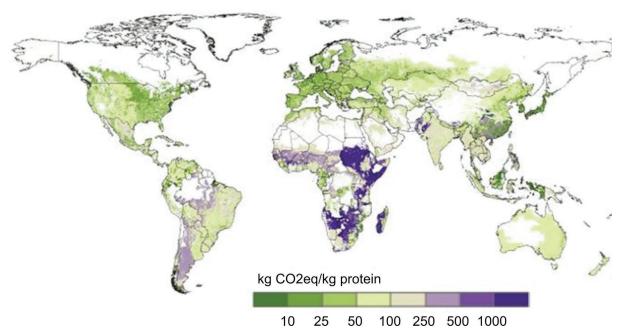


Fig. 9. Global greenhouse gas emissions from beef production in 2010 expressed as kg of CO<sub>2</sub> equivalents per kilogram of protein. Reproduced from Herrero et al. (2013).

within mixed crop and livestock systems, with only 12% being produced in intensive systems (Gerber et al., 2015). The majority of beef, 59% (63% cattle population), is produced in mixed crop and livestock systems, with the remaining 34% of total beef (35% cattle population) being produced on grazing systems. This later group can be further divided into intensive grazing systems that are found in tropical and temperate zones where high-quality grasslands and fodder production can support larger numbers of highly productive animals. These systems are mostly focused on food production, based on individual landownership, and supply about 20% of global beef production. The second category represents pastoral livestock systems that have developed in harsh environments, such as dry lands and cold areas, and which account for less than 15% of total beef production, but which support the livelihoods of 200 million households. These are driven by low animal productivity across large areas of arid lands, feed scarcity, and animals with low productive potential that are often used by smallholder farmers for other services such as draft power, manure and to manage household risk. It should be noted emissions attributable to animals used for draft power are typically excluded from the calculation of meat and milk emission estimates for that species (Gerber et al., 2013). These authors note the efficiency improvements based solely on saleable ASF products that result in herd size reductions could harm traditional farm household livelihoods due to the loss of the non-food goods and services provided by livestock.

The developing world currently contributes 75% of global GHG emissions from ruminants and 56% of emissions from monogastrics (Herrero et al., 2013). There is a wide gap in emission intensities that exist on a global and regional scale (Fig. 9), and considerable variation between producers. It has been found that the environmental impact of producing the same product can vary by 50-fold (Poore and Nemecek, 2018). It is estimated that the potential for mitigating livestock emissions - CH<sub>4</sub> in particular by applying the practices of the top 10th percentile of producers with the lowest emission intensities in a given system, region and agro-ecological zone is between 30 and 35% (FAO, 2018). Chang et al. (2021) reported that 88-91% of the livestock CH<sub>4</sub> emissions come from enteric fermentation by ruminants (i.e. cattle, sheep, goats, and buffaloes). They predicted that if there are no improvements in CH<sub>4</sub> emissions per kg protein, then global livestock CH<sub>4</sub> emissions will increase by 51-54% from 2012 to 2050. They estimated that improving production efficiencies in 10 countries (Brazil, China, India, Iran, Madagascar, Morocco, Niger, South Africa, Tanzania, Turkey) where a large increase in livestock production is projected and the current CH<sub>4</sub> emissions per kg protein are high could contribute 60-65% of the global reduction in livestock emissions by 2050 (compared to a baseline where emission intensities are held constant in the future). They further found that efforts to improve production efficiency have a much greater potential for GHG mitigating effects than would demand-side efforts to promote balanced, healthy, and environmentally sustain-

In this increased efficiency strategic perspective, the livestock sector (and its sectoral organizations) is identified as an important stakeholder in delivering on the mitigation efforts necessary to reduce GHG emissions and to improve its environmental footprint (Gerber et al., 2013). However, the efficiency improvements need to be undertaken with careful consideration of livelihood concerns. Cattle produce meat, milk, fibers, hides, skins, fertilizer and fuel, and are used for transportation and draft power. They also serve socioeconomic, cultural and ecological roles other than food and income, such as asset building in the form of stock accumulation, particularly in Africa and parts of Asia, and religious worship in India. Any proposed strategies for boosting the efficiency of cattle production need to consider these broader concerns, and also the fact that access to technologies may more be limited in some set-

tings, often because of factors such as inaccessibility, unaffordability, lack of relevant knowledge, and/or of organizational capacity. This boosting efficiency framing could equally include the need for improved efficiencies of systems for producing alternative meats to bring these products to market at scale and cost parity, and with a comparable nutritional profile to conventional ASF, which could include innovations in facility design, optimized culture media for cell growth, cell line selection and differentiation, synthetic biology, and microbial contamination control strategies (Post et al., 2020).

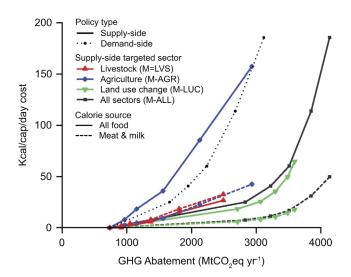
Globally, approximately 86% of the feed DM ingested by livestock is inedible by humans (Mottet et al., 2017), and likely an even high proportion in several developing countries where ruminant livestock subsist mainly on pastures and crop residues (Adesogan et al., 2020). Crop production, processing and the agrifood chains produce large amounts of residues as well as co- and byproducts, which constitute nearly 30% of global livestock feed intake. These products will be produced in ever increasing amounts as the human population grows and consumes more processed food. Livestock play, and will continue to play, a critical role in adding value to these residual products, a large share of which could otherwise be an environmental burden. Intensifying recycling efforts and minimizing losses for a circular bioeconomy include measures such as recycling clean sources of food waste as livestock feed, and adopting innovations in resource re-use. Unused crop residues, food waste, and agro-industrial byproducts are lost opportunities to recycle and optimize resource use efficiency and can be repurposed for animal feed. Additionally, manure and slaughterhouse waste can be used to generate fertilizer and biogas as a source of renewable energy. Similarly recycling of spent culture media, and water from bioreactor cleaning will be an important requirement for scaling up of cultured meats. Ironically livestock are already an important source of food processing waste, and perhaps livestock, particularly monogastrics, could potentially play a role in recycling spent culture media.

The GHG mitigation potential of the livestock sector could represent up to 50% of the global mitigation potential of the agriculture, forestry and land-use sector; however, the share that could be achieved at a reasonable economic cost is likely smaller (Herrero et al., 2016). The livestock sector is uniquely positioned to help mitigate its own emissions if accounting of net carbon sequestration is included in LCAs. For measures targeting soil carbon sequestration in grazing lands, mitigation potentials for animal GHG emissions at unit costs of US\$20, US\$50 and US\$100 per t CO<sub>2</sub> were estimated at 250, 375 and 750 MMt CO<sub>2</sub>eq annually (Metz et al., 2007). Emissions pricing could push up global food prices and reduce consumption in low-income regions, with negative impacts on food security (Herrero et al., 2016). It may be that well-managed grazing on degraded rangelands can help to capture soil organic carbon. Additionally, silvo-pastoralism offers further potential benefits (Buckley Biggs and Huntsinger, 2021; Sales-Baptista and Ferraz-de-Oliveira, 2021). There is also an opportunity to use manure and other waste from livestock farms to generate biogas, and to place solar panels in a way that not only captures sunlight for energy but also provides shade for livestock (Maia et al., 2020; Sharpe et al., 2021). Other opportunities include use of feed additives, improved feed digestibility, improved manure and animal management, and better feeds (Caro et al., 2016). For example, it could be envisioned that green renewable natural gas produced from biomethane captured from California's dairy industry could be used to provide a decarbonized source of power for colocalized alternative meat manufacturing facilities.

Striving for healthy, nutritious diets for all requires a simultaneous understanding that while consumers in MHIC would benefit from reduced consumption of calories and ASF, consumers in LMIC would benefit from improved access to ASF. While changes in diets

A.L. Van Eenennaam and S.J. Werth

Animal 15 (2021) 100360



**Fig. 10.** Total abatement calorie cost curves for mitigation policies targeting different sectors on the supply-side through carbon price or targeting the demandside through consumption reduction. Plain thick lines indicate the loss of total food calories and dashed thick lines the loss of animal calories only. Greenhouse gas (GHG) abatement is calculated as the difference between emissions under scenario DYN (2030 dynamic livestock sector capable of responding to economic drivers by adapting the structure of production systems to the changing environment) with a climate policy and emissions from the reference scenario FIX (counterfactual scenario in which the relative distribution of ruminants across the different livestock production systems was kept as it was in 2000, without climate policy). Data points correspond to carbon prices of US\$0, US\$5, US\$10, US\$20, US\$50, and US\$100 per tCO<sub>2</sub>eq. The demand-side curve (thin dotted line) represents the abatement resulting from restricting consumption to levels calculated under the ALL mitigation policy, M—ALL, but without a carbon price. Reproduced from Havlík et al. (2014).

might be a GHG mitigation option in developed countries, GHG mitigation policies designed to reduce ASF demand were found to be less effective than supply-side policies, and to come at a higher calorie cost (i.e. less food available) which could lead to further malnutrition or undernutrition in the developing world (Havlík et al., 2014). These authors also found that GHG mitigation policies targeted to prevent land-use change were 5-10 times more efficient than policies targeting direct livestock emissions. The emission reductions achieved under a demand-side policy were, depending on the level of calorie availability decrease, 30-80% less effective in reducing emissions than its supply-side policy equivalent (Fig. 10). This means that the demand-side policies modeled in that study resulted in substantially higher calorie reductions to achieve the same GHG mitigation levels as the supply-side policies were able to achieve by directly targeting emissions from agriculture and land-use change. There is also potential for innovations in biotechnology to produce alternative meats with an improved nutritional profile, and improved feedstuffs for both cultured animal cells and livestock. This might include algal, fungal, insect or microbial protein, or synthetic aminoacids being added to livestock feed, or cell culture media. There are also some promising feed additives that work to reduce CH<sub>4</sub> emissions (Roque et al., 2019). Consumer acceptance of these innovations is going to be critical (Bryant and Barnett, 2018).

Policy measures to drive change are perhaps the area where it might be most difficult to find agreement among varying stakeholders. Options span market-based instruments (e.g. carbon pricing, meat taxes, incentives etc.), investments in infrastructure and support for research and development in both conventional and alternative meats, and direct regulatory interventions. Some policies, such as incentivizing farmers to adopt better practices to reduce emissions without lowering production (e.g. mitigation

subsidies), are likely to be easier to implement than negative incentives such as a carbon tax, or demand-side interventions (e.g. taxing meat (Springmann et al., 2018)) to cause a shift to low-emissions food. Modeling carbon taxation against ruminant production systems at US\$52/t CO<sub>2</sub>eq in the UK and France was found to result in socioeconomic losses that far outweighed the value of the environmental benefits (Lee et al., 2021). Win-win outcomes that reduce emissions such as recycling by-products, producing biogas from methane, or capturing soil organic carbon through restoring degraded grasslands with well-managed grazing systems are likely to be more popular than those that result in forced reallocation of resources and large macroeconomic welfare losses, or diminished food security. It is likely that various stakeholders will weigh these trade-offs differently, but top-down interventions that ignore or dismiss these trade-offs are likely to encounter fierce stakeholder opposition. People generally do not like to be told what to think, how to act, or what to eat.

Sustainability discussions bring in multiple competing goals, and often-conflicting outcomes emerge depending how antagonistic goals are balanced. The most environmentally friendly diet might be the least healthy option, or the least palatable, or nutritionally inadequate, or the most expensive, or culturally unacceptable. The trade-offs among production, environmental protection, food and nutrition security, food affordability, livelihoods, human and animal health and welfare are all part of sustainability discussions, and they must be explicitly included in discussions around policy options.

#### **Communicating complexity**

The public debate surrounding the global livestock sector is becoming increasingly polarized, with advocates of reductions in meat consumption being challenged by counter narratives that seek to defend ASF, the livestock industry and the rural livelihoods associated with it (Maye et al., 2021). There is very diverse literature related to the likely impact of alternative meats on a number of different metrics written by subject matter experts in very disparate fields, ranging from technical production aspects, nutritional and human health impacts, behavioral economics and behavior change, policy implications, and environmental and sustainability ramifications. As with all disciplines, there is a minority of experts among a majority population of non-experts. It can be difficult for those with expertise in livestock production systems to make themselves heard. In the words of Salmon et al. (2020), 'The application of scientific information outside the science community is to some extent uncontrollable; nevertheless, the livestock community must remain broadly objective and balanced in presenting information about global livestock production and both its future role in sustainable diets and impacts on broader sustainable development goals.'

The livestock sector has complex interactions with more than half of the UN sustainable development goals (FAO, 2018). Numerous narratives in favor of alternative proteins have emphasized the ability for these novel foods to 'disrupt', and thus overcome the negative impacts associated with conventional livestock production. Aspirational rhetoric calling for an end to animals in the global food system by 2035 might play well with Silicon Valley venture capitalists (Stephens et al., 2018), but it is breathtakingly naïve given the importance of animals in global food systems. Much of the discussion around alternative meats is taking place in high-income countries (Fig. 1), where malnutrition takes the form of excessive food intake resulting in obesity and associated non-communicable diseases. In LMIC, malnutrition takes the form of undernourishment and micronutrient deficiencies. Companies pledging to eliminate livestock apparently overlook the hundreds of millions of pastoralists in South Asia and sub-Saharan Africa, two-thirds of whom are women, and many of whom depend on

A.L. Van Eenennaam and S.J. Werth

Animal 15 (2021) 100360

livestock for food and livelihoods (Niamir-Fuller and Huber-Sannwald, 2020). Steve Myrick, Vice President of operations at Silicon Valley-based Memphis Meats, recently renamed Upside Foods, stated that his company wants to 'augment, not disrupt' the mix of food production methods in the next five to 10 years. He continued, 'we have this philosophy of a big tent. We want to partner with existing industry, coexist, respect consumer traditions', demonstrating a promising move away from the unhelpful binary 'us versus them' framing.

Productive interactions among relevant subject matter experts explicitly identifying the multitude of socio-economic and environmental considerations and trade-offs associated with proposed changes in a complex system such as the livestock sector, and as compared to alternative meat options, will be essential given the complexity of sustainable diets (Tuomisto, 2019a). The global food system is far too diverse and driven by unique environmental and socioeconomic circumstances to allow for a one-size-fits-all policy recommendation (van Vliet et al., 2020). Undoubtedly addressing future food demands will require efforts and investments to increase the environmental efficiency of all food production systems. However, this approach on its own will not deliver a sustainable food system. We need a diversity of voices including wizards, prophets and magicians, farmers, men and women to be involved in developing nutritionally and culturally appropriate food production systems using plants, animals, and cellular agriculture to sustainably address future demand. Proposing major changes in agricultural systems such as replacing ASF requires interdisciplinary and multi-sectorial collaborations, and a nuanced understanding of the impacts of such changes on the multiple interconnected pillars of sustainability (Lee et al., 2021). Furthermore, it is important that the ability to employ innovative technologies in food production systems be preserved. Spreading misinformation or fear around food innovations jeopardizes access to these tools for all food-producing sectors, reducing future opportunities for the co-delivery of nutritious food with a reduced environmental impact.

#### **Ethics approval**

Not applicable.

#### Data and model availability statement

None of the data were deposited in an official repository.

#### **Author ORCIDs**

Alison Van Eenennaam: https://orcid.org/0000-0003-1562-162X

**Samantha Werth** https://orcid.org/0000-0002-9129-6665.

#### **Author contributions**

**Alison Van Eenennaam:** Conceptualization, Writing-Original draft preparation, reviewing and editing. **Samantha Werth:** Formal LCA analysis, Visualization, Investigation, Writing-reviewing and editing.

#### **Declaration of interest**

None.

#### Acknowledgements

We acknowledge Maci Mueller for the preparation of Fig. 7.

## **Financial Support Statement**

This research received no specific grant from any funding agency, commercial or not-for-profit section.

#### References

- Abete, I., Romaguera, D., Vieira, A.R., Lopez de Munain, A., Norat, T., 2014. Association between total, processed, red and white meat consumption and all-cause, CVD and IHD mortality: a meta-analysis of cohort studies. British Journal of Nutrition 112, 762–775. https://doi.org/10.1017/S000711451400124X.
- Adesogan, A.T., Havelaar, A.H., McKune, S.L., Eilittä, M., Dahl, G.E., 2020. Animal source foods: sustainability problem or malnutrition and sustainability solution? Perspective matters. Global Food Security 25,. https://doi.org/10.1016/j.gfs.2019.100325 100325.
- Allen, M.R., Fuglestvedt, 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, 773–776. https://doi.org/10.1038/nclimate2998.
- Allen, M.R., Shine, K.P., Fuglestvedt, J.S., Millar, R.J., Cain, M., Frame, D.J., Macey, A.H., 2018. A solution to the misrepresentations of CO 2-equivalent emissions of short-lived climate pollutants under ambitious mitigation. Npj Climate and Atmospheric Science 1, 1–8. https://doi.org/10.1038/s41612-018-0026-8.
- Asem-Hiablie, S., Battagliese, T., Stackhouse-Lawson, K.R., Rotz, C.A., 2019. A life cycle assessment of the environmental impacts of a beef system in the USA. International Journal of Life Cycle Assessment 24, 441–455. https://doi.org/10.1007/s11367-018-1464-6.
- Barnard, N.D., Leroy, F., 2020. Children and adults should avoid consuming animal products to reduce the risk for chronic disease: Debate Consensus. The American Journal of Clinical Nutrition 112, 937–940. https://doi.org/10.1093/ajcn/nqaa237.
- Boulay, A.-M., Drastig, K., Amanullah, Chapagain, A., Charlon, V., Civit, B., DeCamillis, C., De Souza, M., Hess, T., Hoekstra, A.Y., Ibidhi, R., Lathuillière, M.J., Manzardo, A., McAllister, T., Morales, R.A., Motoshita, M., Palhares, J.C.P., Pirlo, G., Ridoutt, B., Russo, V., Salmoral, G., Singh, R., Vanham, D., Wiedemann, S., Zheng, W., Pfister, S., 2021. Building consensus on water use assessment of livestock production systems and supply chains: Outcome and recommendations from the FAO LEAP Partnership. Ecological Indicators 124, 2. https://doi.org/10.1016/j.ecolind.2021.107391.
- Broom, D.M., 2019. Land and water usage in beef production systems. Animals 9, 13. https://doi.org/10.3390/ani9060286.
- Bryant, C., Barnett, J., 2018. Consumer acceptance of cultured meat: a systematic review. Meat science 143, 8–17. https://doi.org/10.1016/j.meatsci.2018.04.008.
- Buckley Biggs, N., Huntsinger, L., 2021. Managed grazing on California annual rangelands in the context of state climate policy. Rangeland Ecology & Management 76, 56–68. https://doi.org/10.1016/j.rama.2021.01.007.
- Burlingame, B., Dernini, S., 2012. Sustainable diets and biodiversity directions and solutions for policy, research and action. Food and Agriculture Organization of the United Nations (FAO), Rome, Italy.
- Cain, M., Lynch, J., Allen, M.R., Fuglestvedt, J.S., Frame, D.J., Macey, A.H., 2019. Improved calculation of warming-equivalent emissions for short-lived climate pollutants. Npj Climate and Atmospheric Science 2, 1–7. https://doi.org/ 10.1038/s41612-019-0086-4.
- Caro, D., Kebreab, E., Mitloehner, F.M., 2016. Mitigation of enteric methane emissions from global livestock systems through nutrition strategies. Climatic Change 137, 467–480. https://doi.org/10.1007/s10584-016-1686-1.
- Cederberg, C., Flysjo, A., Sonesson, U., Sund, V., Davis, J., 2009. Greenhouse gas emissions from Swedish consumption of meat, milk, and eggs 1990 and 2005. The Swedish Institute for Food and Biotechnology, Göteborg, Sweden.
- Chang, J., Peng, S., Yin, Y., Havlik, P., Herrero, M., 2021. The Key role of production efficiency changes in livestock methane emission mitigation. AGU Advances 2. e2021AV000391. https://doi.org/10.1029/2021AV000391.
- Chen, G.C., Lv, D.B., Pang, Z., Liu, Q.F., 2013. Red and processed meat consumption and risk of stroke: a meta-analysis of prospective cohort studies. European Journal of Clinical Nutrition 67, 91–95. https://doi.org/10.1038/ejcn.2012.180.
- Clegg, M.E., Tarrado Ribes, A., Reynolds, R., Kliem, K., Stergiadis, S., 2021. A comparative assessment of the nutritional composition of dairy and plant-based dairy alternatives available for sale in the UK and the implications for consumers' dietary intakes. Food Research International 148, https://doi.org/10.1016/j.foodres.2021.110586 110586.
- Curtain, F., Grafenauer, S., 2019. Plant-based meat substitutes in the flexitarian age: an audit of products on supermarket shelves. Nutrients 11, 2603. https://doi. org/10.3390/nu11112603.
- Damerau, K., Waha, K., Herrero, M., 2019. The impact of nutrient-rich food choices on agricultural water-use efficiency. Nature Sustainability 2, 233–241. https:// doi.org/10.1038/s41893-019-0242-1.

de Vries, M., de Boer, I.J.M., 2010. Comparing environmental impacts for livestock products: A review of life cycle assessments. Livestock Science 128, 1–11. https://doi.org/10.1016/j.livsci.2009.11.007.

- Dehghan, M., Mente, A., Rangarajan, S., Sheridan, P., Mohan, V., Iqbal, R., Gupta, R., Lear, S., Wentzel-Viljoen, E., Avezum, A., 2018. Association of dairy intake with cardiovascular disease and mortality in 21 countries from five continents (PURE): a prospective cohort study. The Lancet 392, 2288–2297. https://doi.org/10.1016/S0140-6736(18)31812-9.
- Del Prado, A., Manzano, P., Pardo, G., 2021. The role of the European small ruminant dairy sector in stabilising global temperatures: lessons from GWP\* warming-equivalent emission metrics. Journal of Dairy Research 88, 8–15. https://doi.org/10.1017/S0022029921000157.
- Dettling, J., Tu, Q., Faist, M., DelDuce, A., Mandlebaum, S., 2016. A comparative Life Cycle Assessment of plant-based foods and meat foods. Quantis USA, Boston, MA. USA.
- Doran-Browne, N.A., Eckard, R.J., Behrendt, R., Kingwell, R.S., 2015. Nutrient density as a metric for comparing greenhouse gas emissions from food production. Climatic Change 129, 73–87. https://doi.org/10.1007/s10584-014-1316-8.
- Dougherty, H.C., Oltjen, J.W., Mitloehner, F.M., DePeters, E.J., Pettey, L.A., Macon, D., Finzel, J., Rodrigues, K., Kebreab, E., 2019. Carbon and blue water footprints of California sheep production. Journal of Animal Science 97, 945–961. https://doi. org/10.1093/jas/sky442.
- Drewnowski, A., Rehm, C.D., Martin, A., Verger, E.O., Voinnesson, M., Imbert, P., 2015. Energy and nutrient density of foods in relation to their carbon footprint. The American Journal of Clinical Nutrition 101, 184–191. https://doi.org/10.3945/ajcn.114.092486.
- Dror, D.K., Allen, L.H., 2011. The importance of milk and other animal-source foods for children in low-income countries. Food and Nutrition Bulletin 32, 227–243. https://doi.org/10.1177/156482651103200307.
- Drouin-Chartier, J.-P., Chen, S., Li, Y., Schwab, A.L., Stampfer, M.J., Sacks, F.M., Rosner, B., Willett, W.C., Hu, F.B., Bhupathiraju, S.N., 2020. Egg consumption and risk of cardiovascular disease: three large prospective US cohort studies, systematic review, and updated meta-analysis. BMJ 368, https://doi.org/10.1136/bmj. m513 m513.
- Eide, M.H., 2002. Life Cycle Assessment (LCA) of industrial milk production. International Journal of Life Cycle Assessment 7, 115–126. https://doi.org/ 10.1007/Bf02978855
- FAO, 2018. World Livestock: Transforming the livestock sector through the Sustainable Development Goals. Licence: CC BY-NC-SA 3.0 IGO. Food and Agriculture Organization of the United Nations (FAO), Rome, Italy.
- FAO, 2019. Five practical actions towards low-carbon livestock, Food and Agriculture Organization of the United Nations (FAO), Rome, Italy.
- FAO, 2020. FAOSTAT In Food and Agricultural Organization. Food and Agriculture Organization of the United Nations (FAO), Rome, Italy.
- Fish, K.D., Rubio, N.R., Stout, A.J., Yuen, J.S.K., Kaplan, D.L., 2020. Prospects and challenges for cell-cultured fat as a novel food ingredient. Trends in Food Science & Technology 98, 53–67. https://doi.org/10.1016/j.tifs.2020.02.005.
- Fresán, U., Mejia, M.A., Craig, W.J., Jaceldo-Siegl, K., Sabate, J., 2019. Meat analogs from different protein sources: a comparison of their sustainability and nutritional content. Sustainability 11, 3231. https://doi.org/10.3390/su11123231.
- Garnett, T., 2013. Food sustainability: problems, perspectives and solutions. Proceedings of the Nutrition Society 72, 29–39. https://doi.org/10.1017/S0029665112002947.
- Gerber, P., Vellinga, T., Opio, C., Henderson, B., Steinfeld, H., 2010. Greenhouse Gas Emissions from the Dairy Sector A Life Cycle Assessment. Food and Agricultural Organization of the United Nations (FAO) Animal Production and Health Division. Rome. Italy.
- Gerber, P.J., Mottet, A., Opio, C.I., Falcucci, A., Teillard, F., 2015. Environmental impacts of beef production: review of challenges and perspectives for durability. Meat Sci 109, 2–12. https://doi.org/10.1016/j.meatsci.2015.05.013.
- Gerber, P.J., Steinfeld, H., Henderson, B., Mottet, A., Opio, C., Dijkman, J., Falcucci, A., Tempio, G., 2013. Tackling climate change through livestock: a global assessment of emissions and mitigation opportunities. Food and Agriculture Organization of the United Nations (FAO), Rome, Italy.
- Godfray, H.C.J., Aveyard, P., Garnett, T., Hall, J.W., Key, T.J., Lorimer, J., Pierrehumbert, R.T., Scarborough, P., Springmann, M., Jebb, S.A., 2018. Meat consumption, health, and the environment. Science 361, eaam5324. https://doi.org/10.1126/science.aam5324.
- Goldstein, B., Moses, R., Sammons, N., Birkved, M., 2017. Potential to curb the environmental burdens of American beef consumption using a novel plantbased beef substitute. PLoS One 12., https://doi.org/10.1371/journal. pone.0189029 e0189029.
- Goodland, R., Anhang, J., 2009. Livestock and climate change: What if the key actors in climate change are... cows, pigs, and chickens? World Watch Institute, Washington, USA. pp. 10-19. Retrieved on 8 August 2021 from https://awellfedworld.org/wp-content/uploads/Livestock-Climate-Change-Anhang-Goodland.pdf.
- Guinard, C., Verones, F., Loerincik, Y., 2009. Environmental/ecological impact of the dairy sector: Literature review on dairy products for an inventory of key issues, list of environmental initiative and influences on the dairy sector. Bulletin of the International Dairy Federation, 436. International Dairy Federation, Brussels, Belgium.
- Han, M.A., Zeraatkar, D., Guyatt, G.H., Vernooij, R.W.M., El Dib, R., Zhang, Y., Algarni, A., Leung, G., Storman, D., Valli, C., Rabassa, M., Rehman, N., Parvizian, M.K., Zworth, M., Bartoszko, J.J., Lopes, L.C., Sit, D., Bala, M.M., Alonso-Coello, P.,

Johnston, 2019. Reduction of red and processed meat intake and cancer mortality and incidence. Annals of Internal Medicine 171, 711–720. https://doi.org/10.7326/M19-0699.

- Havlík, P., Valin, H., Herrero, M., Obersteiner, M., Schmid, E., Rufino, M.C., Mosnier, A., Thornton, P.K., Böttcher, H., Conant, R.T., Frank, S., Fritz, S., Fuss, S., Kraxner, F., Notenbaert, A., 2014. Climate change mitigation through livestock system transitions. Proceedings of the National Academy of Sciences 111, 3709–3714. https://doi.org/10.1073/pnas.1308044111.
- Headey, D., Hirvonen, K., Hoddinott, J., 2018. Animal Sourced Foods and Child Stunting. American Journal of Agricultural Economics 100, 1302–1319. https:// doi.org/10.1093/ajae/aay053.
- Heller, M.C., Keoleian, G.A., 2018. Beyond Meat's Beyond Burger Life Cycle Assessment: A detailed comparison between a plant-based and an animalbased protein source. CSS Report 18-10. pp. 1-38. University of Michigan, Ann Arbor. MI. USA.
- Henderson, B.B., Gerber, P.J., Hilinski, T.E., Falcucci, A., Ojima, D.S., Salvatore, M., Conant, R.T., 2015. Greenhouse gas mitigation potential of the world's grazing lands: Modeling soil carbon and nitrogen fluxes of mitigation practices. Agriculture, Ecosystems & Environment 207, 91–100. https://doi.org/10.1016/ j.agee.2015.03.029.
- Herrero, M., Gerber, P., Vellinga, T., Garnett, T., Leip, A., Opio, C., Westhoek, H.J., Thornton, P.K., Olesen, J., Hutchings, N., Montgomery, H., Soussana, J.F., Steinfeld, H., McAllister, T.A., 2011. Livestock and greenhouse gas emissions: The importance of getting the numbers right. Animal Feed Science and Technology 166–167, 779–782. https://doi.org/10.1016/j.anifeedsci.2011.04.083.
- Herrero, M., Havlík, P., Valin, H., Notenbaert, A., Rufino, M.C., Thornton, P.K., Blümmel, M., Weiss, F., Grace, D., Obersteiner, M., 2013. Biomass use, production, feed efficiencies, and greenhouse gas emissions from global livestock systems. Proceedings of the National Academy of Sciences 110, 20888–20893. https://doi.org/10.1073/pnas.1308149110.
- Herrero, M., Henderson, B., Havlík, P., Thornton, P.K., Conant, R.T., Smith, P., Wirsenius, S., Hristov, A.N., Gerber, P., Gill, M., Butterbach-Bahl, K., Valin, H., Garnett, T., Stehfest, E., 2016. Greenhouse gas mitigation potentials in the livestock sector. Nature Climate Change 6, 452–461. https://doi.org/10.1038/pclimate2025
- Hillenbrand, M., Thompson, R., Wang, F., Apfelbaum, S., Teague, R., 2019. Impacts of holistic planned grazing with bison compared to continuous grazing with cattle in South Dakota shortgrass prairie. Agriculture, Ecosystems & Environment 279, 156–168. https://doi.org/10.1016/j.agee.2019.02.005.
- Hoekstra, A.Y., 2019. Green-blue water accounting in a soil water balance. Advances in Water Resources 129, 112–117. https://doi.org/10.1016/j.advwatres.2019.05.012.
- International Organization for Standardization, 2014. ISO 14046:2014. Environmental Management: Water Footprint-Principles, Requirements and Guidelines. Geneva, Switzerland, International Organization for Standardization.
- Ioannidis, J.P.A., 2018. The challenge of reforming nutritional epidemiologic research. Journal of the American Medical Association 320, 969–970. https:// doi.org/10.1001/jama.2018.11025.
- Jones, A.D., Hoey, L., Blesh, J., Miller, L., Green, A., Shapiro, L.F., 2016. A systematic review of the measurement of sustainable diets. Advances in Nutrition 7, 641– 664. https://doi.org/10.3945/an.115.011015.
- Kahan, D.M., Jenkins-Smith, H., Braman, D., 2011. Cultural cognition of scientific consensus. Journal of Risk Research 14, 147–174. https://doi.org/10.1080/ 13669877.2010.511246.
- Khan, S., Loyola, C., Dettling, J., Hester, J., Moses, R., 2019. Comparative environmental LCA of the Impossible Burger® with conventional ground beef burger. Quantis, Boston, MA, USA.
- Klurfeld, D.M., 2018. What is the role of meat in a healthy diet? Animal Frontiers 8, 5–10. https://doi.org/10.1093/af/vfy009.
- Lal, R., 2004. Soil carbon sequestration impacts on global climate change and food security. Science 304, 1623–1627. https://doi.org/10.1126/science.1097396.
   Lal, R., 2018. Digging deeper: a holistic perspective of factors affecting soil organic
- Lal, R., 2018. Digging deeper: a holistic perspective of factors affecting soil organic carbon sequestration in agroecosystems. Global Change Biology 24, 3285–3301. https://doi.org/10.1111/gcb.14054.
   Le Louer, B., Lemale, J., Garcette, K., Orzechowski, C., Chalvon, A., Girardet, J.P.,
- Le Louer, B., Lemale, J., Garcette, K., Orzechowski, C., Chalvon, A., Girardet, J.P., Tounian, P., 2014. Severe nutritional deficiencies in young infants with inappropriate plant milk consumption. Archives de pediatrie: organe officiel de la Societe francaise de pediatrie 21, 483–488. https://doi.org/10.1016/j. arcped.2014.02.027.
- Lee, M.R.F., Domingues, J.P., McAuliffe, G.A., Tichit, M., Accatino, F., Takahashi, T., 2021. Nutrient provision capacity of alternative livestock farming systems per area of arable farmland required. Scientific Reports 11, 14975. https://doi.org/ 10.1038/s41598-021-93782-9.
- Leinonen, I., Williams, A.G., Wiseman, J., Guy, J., Kyriazakis, I., 2012. Predicting the environmental impacts of chicken systems in the United Kingdom through a life cycle assessment: egg production systems. Poultry Science 91, 26–40. https://doi.org/10.3382/ps.2011-01635.
- Leroy, F., Cofnas, N., 2020. Should dietary guidelines recommend low red meat intake? Critical Reviews in Food Science and Nutrition 60, 2763–2772. https://doi.org/10.1080/10408398.2019.1657063.
- Liebe, D.L., Hall, M.B., White, R.R., 2020. Contributions of dairy products to environmental impacts and nutritional supplies from United States agriculture. Journal of Dairy Science 103, 10867–10881. https://doi.org/10.3168/jds.2020-18570.

- Lynch, J., Cain, M., Pierrehumbert, R., Allen, M., 2020. Demonstrating GWP\*: a means of reporting warming-equivalent emissions that captures the contrasting impacts of short-and long-lived climate pollutants. Environmental Research Letters 15,. https://doi.org/10.1088/1748-9326/ab6d7e 044023.
- Lynch, J., Pierrehumbert, R., 2019. Climate impacts of cultured meat and beef cattle. Frontiers in Sustainable Food Systems 3, 5. https://doi.org/10.3389/fsufs.2019.00005.
- Machmuller, M.B., Kramer, M.G., Cyle, T.K., Hill, N., Hancock, D., Thompson, A., 2015.
  Emerging land use practices rapidly increase soil organic matter. Nature Communications 6, 6995. https://doi.org/10.1038/ncomms7995.
- MacLeod, M., Vellinga, T., Opio, C., Falcucci, A., Tempio, G., Henderson, B., Makkar, H., Mottet, A., Robinson, T., Steinfeld, H., 2018. Invited review: a position on the global livestock environmental assessment model (GLEAM). Animal 12, 383–397. https://doi.org/10.1017/S1751731117001847.
- Maher, A., Quintana Ashwell, N., Maczko, K., Taylor, D., Tanaka, J., Reeves, M., 2021. Valuation of beef cattle ecosystem services An economic valuation of federal and private grazing lands ecosystem services supported by beef cattle ranching in the US. Translational Animal Science 5, txab054. https://doi.org/10.1093/tas/ txab054.
- Maia, A.S.C., Culhari, E.D.A., Fonsêca, V.D.F.C., Milan, H.F.M., Gebremedhin, K.G., 2020. Photovoltaic panels as shading resources for livestock. J. Clean. Prod. 258, 120551. https://doi.org/10.1016/j.jclepro.2020.120551.
- Manzano, P., White, S.R., 2019. Intensifying pastoralism may not reduce greenhouse gas emissions: wildlife-dominated landscape scenarios as a baseline in life-cycle analysis. Climate Research 77, 91–97. https://doi.org/ 10.3354/cr01555.
- Mapiye, O., Chikwanha, O.C., Makombe, G., Dzama, K., Mapiye, C., 2020. Livelihood, food and nutrition security in Southern Africa: what role do indigenous cattle genetic resources play? Diversity 12, 74. https://doi.org/10.3390/d12020074.
- Mattick, C.S., Landis, A.E., Allenby, B.R., Genovese, N.J., 2015. Anticipatory life cycle analysis of in vitro biomass cultivation for cultured meat production in the United States. Environmental Science and Technology 49, 11941–11949. https://doi.org/10.1021/acs.est.5b01614.
- Maye, D., Fellenor, J., Potter, C., Urquhart, J., Barnett, J., 2021. What's the beef?: Debating meat, matters of concern and the emergence of online issue publics. Journal of Rural Studies 84, 134–146. https://doi.org/10.1016/j.jrurstud.2021.03.008.
- Mejia, M., Fresan, U., Harwatt, H., Oda, K., Uriegas-Mejia, G., Sabate, J., 2020. Life cycle assessment of the production of a large variety of meat analogs by three diverse factories. Journal of Hunger & Enviornmental Nutrition 15, 699–711. https://doi.org/10.1080/19320248.2019.1595251.
- Metz, B., Davidson, O., Bosch, P., Dave, R., Meyer, L., 2007. Climate change 2007:
  Mitigation of climate change. Contribution of Working Group III to the Fourth
  Assessment Report of the Intergovernmental Panel on. Climate Change
  Cambridge University Press, Cambridge, UK.
- Micha, R., Wallace, S.K., Mozaffarian, D., 2010. Red and processed meat consumption and risk of incident coronary heart disease, stroke, and diabetes mellitus. Circulation 121, 2271–2283. https://doi.org/10.1161/ CIRCULATIONAHA.109.924977.
- Moberg, E., Allison, E.H., Harl, H.K., Arbow, T., Almaraz, M., Dixon, J., Scarborough, C., Skinner, T., Rasmussen, L.V., Salter, A., Lei, X.G., Halpern, B.S., 2021. Combined innovations in public policy, the private sector and culture can drive sustainability transitions in food systems. Nature Food 2, 282–290. https://doi.org/10.1038/s43016-021-00261-5.
- Mohorčich, J., Reese, J., 2019. Cell-cultured meat: Lessons from GMO adoption and resistance. Appetite 143,. https://doi.org/10.1016/j.appet.2019.104408 104408.
- Mollenhorst, H., Berentsen, P.B., De Boer, I.J., 2006. On-farm quantification of sustainability indicators: an application to egg production systems. British Poultry Science 47, 405–417. https://doi.org/10.1080/00071660600829282.
- Mottet, A., de Haan, C., Falcucci, A., Tempio, G., Opio, C., Gerber, P., 2017. Livestock: On our plates or eating at our table? A new analysis of the feed/food debate. Global Food Security 14, 1–8. https://doi.org/10.1016/j.gfs.2017.01.001.
- Murdoch, J., Miele, M., 2004. A new aesthetic of food? Relational reflexivity in the 'alternative' food movement. Qualities of food. Manchester University Press, Manchester and New York, pp. 157–175.
- Murphy, B., Crosson, P., Kelly, A.K., Prendiville, R., 2017. An economic and greenhouse gas emissions evaluation of pasture-based dairy calf-to-beef production systems. Agricultural Systems 154, 124–132. https://doi.org/ 10.1016/j.agsy.2017.03.007.
- Myhre, G., Shindell, D., Bréon, F.-M., Collins, W., Fuglestvedt, J., Huang, J., Koch, D.,
   Lamarque, J.-F., Lee, D., Mendoza, B., Nakajima, T., Robock, A., Stephens, G.,
   Takemura, T., Zhang, H., 2014. Anthropogenic and Natural Radiative Forcing. In
   Climate Change 2013 The Physical Science Basis: Working Group I
   Contribution to the Fifth Assessment Report of the Intergovernmental Panel
   on Climate Change (ed. Intergovernmental Panel on Climate, C.), Cambridge
   University Press, Cambridge, UK, pp. 659–740. https://doi.org/10.1017/CB09781107415324.018.
- Naranjo, A., Johnson, A., Rossow, H., Kebreab, E., 2020. Greenhouse gas, water, and land footprint per unit of production of the California dairy Indust over 50 years. Journal of Dairy Science 103, 3760–3773. https://doi.org/10.3168/jds.2019-16576.
- Niamir-Fuller, M., Huber-Sannwald, E., 2020. In: Lucatello, S., Huber-Sannwald, E., Espejel, I., Martínez-Tagüeña, N. (Eds.), Stewardship of Future Drylands and Climate Change in the Global South: Challenges and Opportunities for the

Agenda 2030. Springer International Publishing, Cham, Switzerland, pp. 41–55.  $\label{eq:https://doi.org/10.1007/978-3-030-22464-6} https://doi.org/10.1007/978-3-030-22464-6\_3.$ 

- Nieto, M.I., Barrantes, O., Privitello, L., Reine, R., 2018. Greenhouse Gas Emissions from Beef Grazing Systems in Semi-Arid Rangelands of Central Argentina. Sustainability 10, 4228. https://doi.org/10.3390/su10114228.
- Payne, C.L., Scarborough, P., Cobiac, L., 2016. Do low-carbon-emission diets lead to higher nutritional quality and positive health outcomes? A systematic review of the literature. Public Health Nutrition 19, 2654–2661. https://doi.org/10.1017/ s1368980016000495.
- Pelletier, N., Ibarburu, M., Xin, H.W., 2014. Comparison of the environmental footprint of the egg industry in the United States in 1960 and 2010. Poultry Science 93, 241–255. https://doi.org/10.3382/ps.2013-03390.
- Pimpin, L., Kranz, S., Liu, E., Shulkin, M., Karageorgou, D., Miller, V., Fawzi, W., Duggan, C., Webb, P., Mozaffarian, D., 2019. Effects of animal protein supplementation of mothers, preterm infants, and term infants on growth outcomes in childhood: a systematic review and meta-analysis of randomized trials. The American Journal of Clinical Nutrition 110, 410–429. https://doi.org/10.1093/ajcn/nqy348.
- Pimpin, L., Wu, J.H., Haskelberg, H., Del Gobbo, L., Mozaffarian, D., 2016. Is butter back? A systematic review and meta-analysis of butter consumption and risk of cardiovascular disease, diabetes, and total mortality. PLoS One 11, https://doi. org/10.1371/journal.pone.0158118 e0158118.
- Pogue, S.J., Kröbel, R., Janzen, H.H., Beauchemin, K.A., Legesse, G., de Souza, D.M., Iravani, M., Selin, C., Byrne, J., McAllister, T.A., 2018. Beef production and ecosystem services in Canada's prairie provinces: a review. Agricultural Systems 166, 152–172. https://doi.org/10.1016/j.agsy.2018.06.011.
- Poore, J., Nemecek, T., 2018. Reducing food's environmental impacts through producers and consumers. Science 360, 987–992. https://doi.org/10.1126/science.aaq0216.
- Post, M.J., Levenberg, S., Kaplan, D.L., Genovese, N., Fu, J., Bryant, C.J., Negowetti, N., Verzijden, K., Moutsatsou, P., 2020. Scientific, sustainability and regulatory challenges of cultured meat. Nature Food 1, 403–415. https://doi.org/10.1038/ s43016-020-0112-z.
- Ridoutt, B., 2021. Short communication: climate impact of Australian livestock production assessed using the GWP\* climate metric. Livestock Science 246, https://doi.org/10.1016/j.livsci.2021.104459 104459.
- Ridoutt, B.G., Hendrie, G.A., Noakes, M., 2017. Dietary strategies to reduce environmental impact: a critical review of the evidence base. Advances in Nutrition 8, 933–946. https://doi.org/10.3945/an.117.016691.
- Ritchie, H., Roser, M., 2019. Meat and Dairy Production. Retrieved on 8 August 2021 from https://ourworldindata.org/meat-production.
- Robinson, T.P., Wint, G.R.W., Conchedda, G., Van Boeckel, T.P., Ercoli, V., Palamara, E., Cinardi, G., D'Aietti, L., Hay, S.I., Gilbert, M., 2014. Mapping the global distribution of livestock. PLoS ONE 9, https://doi.org/10.1371/journal.pone.0096084 e96084.
- Rogelj, J., Schleussner, C.-F., 2019. Unintentional unfairness when applying new greenhouse gas emissions metrics at country level. Environmental Research Letters 14, https://doi.org/10.1088/1748-9326/ab4928 114039.
- Roque, B.M., Salwen, J.K., Kinley, R., Kebreab, E., 2019. Inclusion of Asparagopsis armata in lactating dairy cows' diet reduces enteric methane emission by over 50 percent. Journal of Cleaner Production 234, 132–138. https://doi.org/ 10.1016/j.iclepro.2019.06.193.
- Rotz, C.A., Asem-Hiablie, S., Place, S., Thoma, G., 2019. Environmental footprints of beef cattle production in the United States. Agricultural Systems 169, 1–13. https://doi.org/10.1016/j.agsy.2018.11.005.
- Rowntree, J.E., Stanley, P.L., Maciel, I.C., Thorbecke, M., Rosenzweig, S.T., Hancock, D. W., Guzman, A., Raven, M.R., 2020. Ecosystem impacts and productive capacity of a multi-species pastured livestock system. Frontiers in Sustainable Food Systems 4, https://doi.org/10.3389/fsufs.2020.544984 544984.
- Rozin, P., Hormes, J.M., Faith, M.S., Wansink, B., 2012. Is meat male? A quantitative multimethod framework to establish metaphoric relationships. Journal of Consumer Research 39, 629–643. https://doi.org/10.1086/664970.
- Rubio, N.R., Xiang, N., Kaplan, D.L., 2020. Plant-based and cell-based approaches to meat production. Nature Communications 11, 6276. https://doi.org/10.1038/ s41467-020-20061-y.
- Rudolph, G., Hortenhuber, S., Bochicchio, D., Butler, G., Brandhofer, R., Dippel, S., Dourmad, J.Y., Edwards, S., Fruh, B., Meier, M., Prunier, A., Winckler, C., Zollitsch, W., Leeb, C., 2018. Effect of three husbandry systems on environmental impact of organic pigs. Sustainability 10, 3796. https://doi.org/10.3390/su10103796.
- Sales-Baptista, E., Ferraz-de-Oliveira, M.I., 2021. Grazing in silvopastoral systems: multiple solutions for diversified benefits. Agroforestry Systems 95, 1–6. https://doi.org/10.1007/s10457-020-00581-8.
- Salmon, G.R., MacLeod, M., Claxton, J.R., Pica Ciamarra, U., Robinson, T., Duncan, A., Peters, A.R., 2020. Exploring the landscape of livestock 'Facts'. Global Food Security 25, https://doi.org/10.1016/j.gfs.2019.100329 100329.
- Schwingshackl, L., Schwedhelm, C., Hoffmann, G., Lampousi, A.-M., Knüppel, S., Iqbal, K., Bechthold, A., Schlesinger, S., Boeing, H., 2017. Food groups and risk of all-cause mortality: a systematic review and meta-analysis of prospective studies. The American Journal of Clinical Nutrition 105, 1462–1473. https://doi.org/10.3945/ajcn.117.153148.
- Searchinger, T., Waite, R., Hanson, C., Ranganathan, J., Dumas, P., Matthews, E., Klirs, C., 2019. Creating a sustainable food future: a menu of solutions to feed nearly 10 billion people by 2050. Final report. Retrieved on 8 August 2021 from https://research.wri.org/wrr-food.
- Sevenster, M., Luo, Z., Eady, S., Grant, T., 2020. Including long-term soil organic carbon changes in life cycle assessment of agricultural products. The

International Journal of Life Cycle Assessment 25, 1231–1241. https://doi.org/10.1007/s11367-019-01660-4.

- Sexton, A.E., Garnett, T., Lorimer, J., 2019. Framing the future of food: the contested promises of alternative proteins. Environment and Planning E: Nature and Space 2, 47–72. https://doi.org/10.1177/2514848619827009.
- Sharpe, K.T., Heins, B.J., Buchanan, E.S., Reese, M.H., 2021. Evaluation of solar photovoltaic systems to shade cows in a pasture-based dairy herd. Journal of Dairy Science 104, 2794–2806. https://doi.org/10.3168/jds.2020-18821.
- Sieverding, H., Kebreab, E., Johnson, J.M.F., Xu, H., Wang, M., Grosso, S.J.D., Bruggeman, S., Stewart, C.E., Westhoff, S., Ristau, J., Kumar, S., Stone, J.J., 2020. A life cycle analysis (LCA) primer for the agricultural community. Agronomy Journal 112, 3788–3807. https://doi.org/10.1002/agj2.20279.
- Singhal, S., Baker, R.D., Baker, S.S., 2017. A comparison of the nutritional value of cow's milk and nondairy beverages. Journal of Pediatric Gastroenterology and Nutrition 64, 799–805. https://doi.org/10.1097/MPG.000000000001380.
- Sinke, P., Odegard, I., 2021. LCA of cultivated meat. Future projections for different scenarios. Retrieved on 8 August 2021 from https://cedelft.eu/wp-content/ uploads/sites/2/2021/04/CE\_Delft\_190107\_LCA\_of\_cultivated\_meat\_Def.pdf.
- Smetana, S., Mathys, A., Knoch, A., Heinz, V., 2015. Meat alternatives: life cycle assessment of most known meat substitutes. The International Journal of Life Cycle Assessment 20, 1254–1267. https://doi.org/10.1007/s11367-015-0931-6.
- Smetana, S., Schmitt, E., Mathys, A., 2019. Sustainable use of Hermetia illucens insect biomass for feed and food: Attributional and consequential life cycle assessment. Resources Conservation and Recycling 144, 285–296. https://doi. org/10.1016/j.resconrec.2019.01.042.
- Smith, P., 2013. Delivering food security without increasing pressure on land. Global Food Security 2, 18–23. https://doi.org/10.1016/j.gfs.2012.11.008.
- Sobal, J., 2005. Men, meat, and marriage: Models of masculinity. Food and Foodways 13, 135–158. https://doi.org/10.1080/07409710590915409.
- Soedamah-Muthu, S.S., de Goede, J., 2018. Dairy consumption and cardiometabolic diseases: systematic review and updated meta-analyses of prospective cohort studies. Current Nutrition Reports 7, 171–182. https://doi.org/10.1007/s13668-018-0253-v.
- Springmann, M., Mason-D'Croz, D., Robinson, S., Wiebe, K., Godfray, H.C.J., Rayner, M., Scarborough, P., 2018. Health-motivated taxes on red and processed meat: a modelling study on optimal tax levels and associated health impacts. PLoS One 13,. https://doi.org/10.1371/journal.pone.0204139 e0204139.
- Stackhouse-Lawson, K.R., Rotz, C.A., Oltjen, J.W., Mitloehner, F.M., 2012. Carbon footprint and ammonia emissions of California beef production systems. Journal of Animal Science 90, 4641–4655. https://doi.org/10.2527/jas.2011-4653.
- Stanley, P.L., Rowntree, J.E., Beede, D.K., DeLonge, M.S., Hamm, M.W., 2018. Impacts of soil carbon sequestration on life cycle greenhouse gas emissions in Midwestern USA beef finishing systems. Agricultural Systems 162, 249–258. https://doi.org/10.1016/j.agsy.2018.02.003.
- Steinfeld, H., Gerber, P., Wassenaar, T., Castel, V., Rosales, M., de Haan, C., 2006. Livestock's long shadow: environmental issues and options. Food & Agriculture Organization of the United Nations, Rome, Italy.
- Stephens, N., Di Silvio, L., Dunsford, I., Ellis, M., Glencross, A., Sexton, A., 2018. Bringing cultured meat to market: Technical, socio-political, and regulatory challenges in cellular agriculture. Trends in Food Science & Technology 78, 155– 166. https://doi.org/10.1016/j.tifs.2018.04.010.
- Stephens, N., Sexton, A.E., Driessen, C., 2019. Making sense of making meat: key moments in the first 20 years of tissue engineering muscle to make food. Frontiers in Sustainable Food Systems 3, 45. https://doi.org/10.3389/fsufs.2019.00045.
- Sunde, M., 2003. Seventy-five years of rising American poultry consumption: was it due to the chicken of tomorrow contest? Nutrition Today 38, 60–62. https://doi.org/10.1097/00017285-200303000-00009.
- Tavárez, M.A., Solis de los Santos, F., 2016. Impact of genetics and breeding on broiler production performance: a look into the past, present, and future of the industry. Animal Frontiers 6, 37–41. https://doi.org/10.2527/af.2016-0042.
- Tessari, P., Lante, A., Mosca, G., 2016. Essential amino acids: master regulators of nutrition and environmental footprint? Scientific Reports 6, 26074. https://doi. org/10.1038/srep26074.
- Thoma, G., Popp, J., Nutter, D., Shonnard, D., Ulrich, R., Matlock, M., Kim, D.S., Neiderman, Z., Kemper, N., East, C., Adom, F., 2013. Greenhouse gas emissions from milk production and consumption in the United States: a cradle-to-grave life cycle assessment circa 2008. International Dairy Journal 31, S3–S14. https://doi.org/10.1016/j.idairyj.2012.08.013.
- Thomson, C.A., Van Horn, L., Caan, B.J., Aragaki, A.K., Chlebowski, R.T., Manson, J.E., Rohan, T.E., Tinker, L.F., Kuller, L.H., Hou, L., Lane, D.S., Johnson, K.C., Vitolins, M. Z., Prentice, R.L., 2014. Cancer Incidence and Mortality during the Intervention and Postintervention Periods of the Women's Health Initiative Dietary

- Modification Trial. Cancer Epidemiology Biomarkers & Prevention 23, 2924–2935. https://doi.org/10.1158/1055-9965.Epi-14-0922.
- Thorbecke, M., Dettling, J., 2019. Carbon footprint evaulation of regenerative grazing at White Oak Pastures: Results presentation. Quantis USA, Boston, MA, ISSA
- Tichenor, N.E., Peters, C.J., Norris, G.A., Thoma, G., Griffin, T.S., 2017. Life cycle environmental consequences of grass-fed and dairy beef production systems in the Northeastern United States. Journal of Cleaner Production 142, 1619–1628. https://doi.org/10.1016/j.jclepro.2016.11.138.
- Tuomisto, H.L., 2019a. The complexity of sustainable diets. Nature Ecology & Evolution 3, 720–721. https://doi.org/10.1038/s41559-019-0875-5.
- Tuomisto, H.L., 2019b. The eco-friendly burger: Could cultured meat improve the environmental sustainability of meat products? EMBO Reports 20. https://doi.org/10.15252/embr.201847395.
- Tuomisto, H.L., de Mattos, M.J.T., 2011. Environmental Impacts of Cultured Meat Production. Environmental Science & Technology 45, 6117–6123. https://doi. org/10.1021/es200130u.
- Tuomisto, H.L., Ellis, M.J., Haastrup, P., 2014. Environmental impacts of cultured meat: alternative production scenarios. In: The 9th International Conference on Life Cycle Assessment in the Agri-Food Sector, San Francisco, CA, USA (eds. Schenck, R. and Huizenga, D.), pp. 1360-1366.
- Van Mierlo, K., Rohmer, S., Gerdessen, J.C., 2017. A model for composing meat replacers: Reducing the environmental impact of our food consumption pattern while retaining its nutritional value. Journal of Cleaner Production 165, 930– 950. https://doi.org/10.1016/j.jclepro.2017.07.098.
- van Vliet, S., Kronberg, S.L., Provenza, F.D., 2020. Plant-based meats, human health, and climate change. Frontiers in Sustainable Food Systems 4. https://doi.org/10.3389/fsufs.2020.00128.
- Verge, X.P.C., Dyer, J.A., Desjardins, R.L., Worth, D., 2009. Long-term trends in greenhouse gas emissions from the Canadian poultry industry. Journal of Applied Poultry Research 18, 210–222. https://doi.org/10.3382/japr.2008-00091.
- Vitoria, I., 2017. The nutritional limitations of plant-based beverages in infancy and childhood. Nutrición Hospitalaria 34, 1205–1214.
- Wang, X., Lin, X., Ouyang, Y.Y., Liu, J., Zhao, G., Pan, A., Hu, F.B., 2016. Red and processed meat consumption and mortality: dose-response meta-analysis of prospective cohort studies. Public Health Nutrition 19, 893–905. https://doi.org/10.1017/S1368980015002062.
- Warner, R.D., 2019. Review: Analysis of the process and drivers for cellular meat production. Animal 13, 3041–3058. https://doi.org/10.1017/S1751731119001897.
- White, R.R., Hall, M.B., 2017. Nutritional and greenhouse gas impacts of removing animals from US agriculture. Proceedings of the National Academy of Sciences 114, E10301–E10308. https://doi.org/10.1073/pnas.1707322114.
- Wiedemann, S.G., McGahan, E.J., 2011. Environmental Assessment of an Egg Production Supply Chain using Life Cycle Assessment. Australian Egg Corporation Limited, North Sydney, NSW, Australia.
- Willett, W., Rockström, J., Loken, B., Springmann, M., Lang, T., Vermeulen, S., Garnett, T., Tilman, D., DeClerck, F., Wood, A., 2019. Food in the Anthropocene: the EAT–Lancet Commission on healthy diets from sustainable food systems. The Lancet 393, 447–492.
- Wirsenius, S., Searchinger, T., Zionts, J., Peng, L., Beringer, T., Dumas, P., 2020. Comparing the Life Cycle Greenhouse Gas Emissions of Dairy and Pork Systems across Countries Using Land-Use Opportunity Costs. Working Paper. Retrieved on 8 August 2021 from https://www.wri.org/research/comparing-life-cycle-greenhouse-gas-emissions-dairy-and-pork-systems-across-countries.
- Zeraatkar, D., Han, M.A., Guyatt, G.H., Vernooij, R.W.M., El Dib, R., Cheung, K., Milio, K., Zworth, M., Bartoszko, J.J., Valli, C., Rabassa, M., Lee, Y., Zajac, J., Prokop-Dorner, A., Lo, C., Bala, M.M., Alonso-Coello, P., Hanna, S.E., Johnston, B.C., 2019a. Red and Processed Meat Consumption and Risk for All-Cause Mortality and Cardiometabolic Outcomes: A Systematic Review and Meta-analysis of Cohort Studies. Annals of Internal Medicine 171, 703–710. https://doi.org/10.7326/m19-0655.
- Zeraatkar, D., Johnston, B.C., Bartoszko, J., Cheung, K., Bala, M.M., Valli, C., Rabassa, M., Sit, D., Milio, K., Sadeghirad, B., Agarwal, A., Zea, A.M., Lee, Y., Han, M.A., Vernooij, R.W.M., Alonso-Coello, P., Guyatt, G.H., El Dib, R., 2019b. Effect of Lower Versus Higher Red Meat Intake on Cardiometabolic and Cancer Outcomes: A Systematic Review of Randomized Trials. Annals of Internal Medicine 171, 721–731. https://doi.org/10.7326/m19-0622.
- Zhang, Z., Goldsmith, P.D., Winter-Nelson, A., 2016. The Importance of Animal Source Foods for Nutrient Sufficiency in the Developing World: The Zambia Scenario. Food and Nutrition Bulletin 37, 303–316. https://doi.org/10.1177/ 0379572116647823.

# Effects of wildfire smoke exposure in cattle: a review of the current state of knowledge and future directions

A. L. Skibiel, A. Anderson, and P. Rezamand

Department of Animal, Veterinary and Food Sciences, University of Idaho, Moscow, ID 83844-2330, USA

Corresponding author: Amy L. Skibiel. Address: 875 Perimeter Dr., Moscow, ID 83844, Email: askibiel@uidaho.edu, Telephone: 208-885-1161

#### **Abstract**

Wildfires burn millions of acres of land annually in the United States and experts predict the number and intensity of wildfires to continue to increase. Beyond destruction of land, vegetation, and structures in the immediate path of the fire, wildfires emit a variety of particulates and other pollutants that are hazardous to breathe. Associations between exposure to wildfire particulates and many negative health outcomes, such as hospital admissions, respiratory disease, cardiovascular morbidity, and premature mortality, have been reported in humans. The western U.S. is particularly prone to wildfire breakouts and is home to more than 15 million beef and dairy cattle that are also exposed to wildfire smoke each year. Health and production issues related to wildfire smoke inhalation in cattle have not been thoroughly researched despite the susceptibility of cattle to respiratory disease and greater potential health risk from inhaled pollutants relative to other mammals. An emerging body of literature is, however, unequivocally demonstrating that wildfire smoke exposure is a threat to cattle health and performance. This review summarizes the current state of knowledge regarding the effects of exposure to wildfire particulate matter on cow and calf health and production. Implications of wildfire smoke exposure on the industry are also discussed.

**Keywords**: air quality, air pollution, pulmonary disease, reproduction, dairy cattle

## Introduction

Wildfires have become more frequent and severe over the past several decades, as exemplified by a number of recent high-profile conflagrations, such as the 2022 Calf Canyon blaze in New Mexico that burned an area larger than New York City and the 2021 Dixie fire that burned almost 1 million acres, becoming the second largest wildfire in California's history. Since the beginning of 2022, there have been more than 61,390 wildfire events in the U.S. resulting in 7.25 million acres burned (NICC, 2022). In the U.S., further increases in the number and intensity of large wildfires and a 10-30% extension in the duration of the wildfire season is expected (IPCC, 2014). Annual area burned by wildfires is also expected to increase by 76-152% by the end of the 21st century (Flannigan et al., 2006). Warmer temperatures combined with lower precipitation and earlier spring snow melts contribute to dry vegetation, providing ample fuel for wildfires to ignite and spread (IPCC, 2014).

In the U.S., wildfires occur often in the western states, home to more than 12 million beef cattle and 3.5 million dairy cattle that produce approximately 40% of the nation's milk (NASS, 2019). It is predicted that wildfires will disrupt the U.S. agricultural industry because of declines in crop and forage harvest and quality, and livestock health and production (USGCRP, 2016), although the economic consequences of wildfires on agriculture are currently unknown because of a lack

of research. It is estimated that economic losses just in the Utah cattle industry attributable to wildfires are over \$1.4 million for an average fire year, related to premature cattle sale and necessity of feed purchasing because of scorched rangeland (Jakus et al., 2017). However, the actual costs are likely much higher as this estimate was for a single state and does not account for adverse effects of wildfire smoke on cattle productivity and health.

The detrimental effects of wildfires extend beyond the direct area burned and include the release of toxic gases and particulates into the atmosphere that are hazardous to breathe. Fine particulate matter (PM<sub>2.5</sub>) is thought to be one of the greatest contributors to adverse health outcomes after wildfire smoke exposure (Black et al., 2017a). According to the U.S. Environmental Protection Agency, the total annual amount of PM<sub>2.5</sub> produced by wildfires was more than 3.3 million tons (U.S. EPA, 2020). Liu et al., (2015) estimated that exposure to PM from wildfire smoke will increase by 160% by the year 2051 in the U.S. As the number of wildfire occurrences and intensity increases, the likelihood of adverse health impacts is also expected to increase (USGCRP, 2016).

Inhalation of wildfire PM is associated with pulmonary disease and mortality in humans (Liu et al., 2015; Reid et al., 2016; Stowell et al., 2019), largely attributed to inflammation (Nakayama Wong et al., 2011). Cattle may be especially vulnerable to air pollutants owing to several unique characteristics of their pulmonary system (Veit and Farrell, 1978) and the inability of cattle to seek refugia from smoke. Indeed, ambient (i.e., non-wildfire) air pollution is associated with an increased risk of mortality in mature dairy cows and calves (Cox et al., 2016; Egberts et al., 2019). Further, research by our group found an increase in cow disease and calf mortality, and reduced milk yield when exposed to wildfire smoke PM (Anderson et al., 2020; Anderson et al., 2022).

This review will summarize the literature to date of the known consequences of wildfire smoke exposure on health, production, and mortality of cattle. This topic is particularly timely as large, catastrophic wildfires continue to increase in frequency in the U.S. and as animal producers are becoming more aware of the threats of wildfires to their agricultural operations (O'Hara et al., 2021). Because of the limited data available in cattle, we will also present research in other domesticated and free-ranging animals as well as studies rooted in human epidemiology to highlight remaining questions and areas of focus for future research endeavors involving cattle. While the focus of the review is specifically on wildfire-derived PM, relevant studies of non-wildfire PM sources and other air toxics are also discussed.

## Wildfire smoke composition, dispersion, and toxicity

Wildfire smoke is a mixture of gaseous chemical compounds and particulate matter (PM) that reduce air quality (Michel et al., 2005; Wentworth et al., 2018). Chemical components of wildfire smoke include water vapor, carbon monoxide and dioxide, nitrogen oxides, sulfur dioxide, polycyclic aromatic hydrocarbons (PAH; e.g., formaldehyde, acenaphthene, napthelene, pyrene) and volatile organic compounds (VOC; e.g., benzene, ethylbenzene, toluene, xylenes), which are known neuromuscular toxins, teratogens, and carcinogens in humans (Urbanski et al., 2009; Wentworth et al., 2018; Sokolik et al., 2019; Miller et al., 2022). Particulate matter consists of liquid and solid particles that are categorized based on size. The

coarse particulates (PM $_{10}$ ) are smaller than 10  $\mu m$  in diameter and fine particulates (PM $_{2.5}$ ) are smaller than 2.5  $\mu m$  in diameter (Wilson and Suh, 1997). Wildfire emissions contain both PM $_{10}$  and PM $_{2.5}$ , but typically emit more PM $_{2.5}$  (Groß et al., 2013; Vicente et al., 2013).

Particulate matter is one of the most harmful pollutants in wildfire smoke. PM is inhaled into the respiratory tract where it can cause tissue inflammation and damage (Löndahl et al., 2007; Huttunen et al., 2012). PM<sub>2.5</sub> is particularly hazardous because it can penetrate deep into the respiratory tract and deposit into the lower airways and lungs (Carvalho et al., 2011). Additionally, PM<sub>2.5</sub> is capable of crossing the lungs to enter the blood (Fu et al., 2011; Schulze et al., 2017). PM in wildfire emissions can reach daily average concentrations that far exceed the established standards for ambient air quality (24-hour average of 35  $\mu$ g/m³ for PM<sub>2.5</sub> and 150  $\mu$ g/m³ for PM<sub>10</sub> for human outdoor exposure; (U.S. EPA., 2013; Landis et al., 2018).

Wildfire smoke can travel great distances via air plumes, thus a single wildfire can reduce air quality in geographical locations far removed from the area burned (Stowell et al., 2019). The area affected by wildfire smoke can be 50 times larger than the areas that were directly burned by the fires (Knowlton, 2013). PM<sub>2.5</sub>, because it is lighter than other particles, spends the longest amount of time in the atmosphere (~100 days), can travel farther distances, and therefore has a larger geographic distribution relative to other pollutants in wildfire smoke (U.S. EPA, 2010). Because of the capacity of wildfire smoke to disperse across vast distances, populations of humans and animals far from the wildfire may suffer from adverse health consequences (USGCRP, 2016).

## Effects of wildfire smoke on mortality

Associations between wildfire smoke exposure and increased mortality risk in humans, including cardiovascular, pulmonary, and all-cause mortality, are well established. In a systematic review of 63 human epidemiological studies, 13 studies quantified mortality risk from wildfire smoke exposure, and of those, 9 studies reported elevated mortality rates upon exposure to wildfire smoke (Liu et al., 2015). Similarly, in a critical assessment of more than 300 studies on human health effects from wildfire smoke, positive associations between wildfire smoke exposure and mortality were found in most studies (Reid et al., 2016). Analyzing over 13 years of air quality data in Australia, including a total of 48 days affected by wildfire smoke, Johnston et al., (2011) found a 5% increase in human mortality on smoke-affected days.

Related data on mortality risk in cattle exposed to wildfire smoke are limited. In a preliminary study by our group, we collated data from two dairy farms in the Pacific Northwest across a five-year period, and found that mortality of dairy calves, but not cows, was greater on days when PM<sub>2.5</sub> concentration from wildfire smoke was elevated (Anderson et al., 2020). Similarly, specific human subpopulations, such as children, are more vulnerable to negative health outcomes from wildfire smoke than are adults (Shaposhnikov et al., 2014; Liu et al., 2015). Air pollution from non-wildfire sources is also associated with an increased risk of mortality in dairy cows and calves (Cox et al., 2016; Egberts et al., 2019). In mature dairy cows, ambient, non-wildfire-PM<sub>10</sub>, ozone, and nitrogen dioxide concentrations were positively correlated with risk of mortality on the day of exposure and up to two weeks later, especially in the warm summer months (Cox et al., 2016). In a more recent analysis, Egberts et al., (2019) found acute and

cumulative effects of ambient, non-wildfire sources of ozone on risk of mortality in beef and dairy calves, young stock, and lactating cows in the warm season. As the chemical composition of wildfire smoke can be radically different from other sources of air pollution, and wildfire emissions can vary spacially and temporally within and across wildfire seasons, further research is warranted in cattle to assess links between mortality risk and wildfire smoke exposure.

#### Health effects from wildfire smoke inhalation

Exposure to wildfire smoke has been linked to impaired lung function, pulmonary disease, and pulmonary-specific hospitalizations in humans (Reid et al., 2016; DeFlorio-Barker et al., 2019; Stowell et al., 2019). In Washington state, increases in wildfire smoke PM<sub>2.5</sub> concentrations were positively correlated with increased risk of adverse respiratory outcomes, specifically in asthma hospitalization, chronic obstructive pulmonary disease hospitalization, and pneumonia cases (Gan et al., 2017). Assessing three years of health records in Colorado, Stowell et al., (2019) found that for every 1  $\mu$ g/m³ increase in wildfire-PM<sub>2.5</sub> there was a 10% increase in the risk of asthma and combined respiratory disease. Further, there was a rise in the number of cases of acute bronchitis and pneumonia after exposure to smoke from brushfires in Sydney, Australia between 1994 and 2014 (Morgan et al., 2010). These and other studies clearly point to an elevated risk of respiratory morbidity from wildfire smoke inhalation.

Respiratory illness is a leading cause of cow and calf deaths in the U.S. (USDA, 2017; Dubrovsky et al., 2019). In 2014, 12% of pre-weaned and 5% of weaned dairy calves were diagnosed with respiratory disease in the U.S. Among adult dairy cattle, on average 2.8% of cows (257,600 cows) were diagnosed with respiratory disease in 2014, 10.5% of which subsequently died and respiratory disease was cited for the removal of an average 2.1% of dairy cows across herds (USDA, 2017). Cattle are prone to respiratory disorders, such as Bovine Respiratory Disease, in part because of unique anatomical and physiological characteristics of the respiratory tract relative to other mammals (Veit and Farrell, 1978). The same characteristics that predispose cattle to respiratory infection may also contribute to greater susceptibility to inhaled PM. For example, cattle also longer trachea and bronchi, increasing pathogen (and presumably PM) retention time in the respiratory tract, which increases the likelihood of deposition (Kirschvink, 2008). Furthermore, the bovine respiratory system contains lower levels of lysozyme and fewer alveolar macrophages, which play a role clearing and breaking down inhaled pathogens and PM (Mariassy et al., 1975; Veit and Farrell, 1978; Lohmann-Matthes et al., 1994).

In a recent review of articles related to wildfires and health in cattle, no relevant peer-reviewed studies were found, leading the authors to conclude that wildfire smoke inhalation was not likely to have much impact on cattle (Eid et al., 2021). However, across two commercial farms in the U.S. Pacific Northwest, there was an increase in the number of mastitis and general illness cases among mature dairy cows on days when PM<sub>2.5</sub> from wildfires was elevated (Table 1; Anderson et al., 2020). In a further empirical study by our group, we found that immune cell populations, such as basophils and eosinophils in systemic circulation were higher, whereas neutrophils were lower, when cattle were exposed to wildfire PM<sub>2.5</sub> along with elevated air temperature and humidity (Anderson et al., 2022). Similarly, captive bottlenose dolphins housed in the San Diego Bay exposed to wildfire smoke in 2003 and 2007 had lower circulating neutrophils and elevated eosinophil counts during and 1 month following the fires (Venn-

Watson et al., 2013). However, these studies did not focus specifically on respiratory illness and thus, research is critically needed to characterize the pulmonary response to PM exposure in cattle.

Table 1. Summary of health and productive outcomes in cattle associated with exposure to gaseous and particulate matter pollutants

gaseous and particulate matter politicalits				
Exposure	Observed effect	Reference		
Naturally occurring	Increased incidence of mastitis and general illness	Anderson et al., 2020		
wildfire-PM <sub>2.5</sub>	in cows			
	Increased calf mortality			
Naturally occurring wildfire-PM <sub>2.5</sub>	Decreased milk yield and milk protein content	Anderson et al., 2022		
Naturally occurring	Fewer circulating neutrophils, increased circulating	Anderson et al., 2022		
wildfire-PM <sub>2.5</sub> and elevated	eosinophils and basophils			
THI	Initial decrease in blood urea nitrogen and increase			
	in plasma non-esterified fatty acids. Opposite			
	effects with subsequent exposure days			
Ambient (non-wildfire)	Increased relative risk of mortality in mature dairy	Cox et al., 2016		
PM <sub>10</sub>	cows			
Ambient (non-wildfire) and wildfire PM <sub>2.5</sub>	Decreased milk yield and increased SCC	Beaupied et al., 2022		
Ambient (non-wildfire)	Increased relative risk of mortality in beef and dairy	Egberts et al., 2019		
ozone	calves, heifers, and mature cows			
PM <sub>10</sub> in dust	Increased neutrophil count in lung lavage, increased	van Leenen et al.,		
	risk of pneumonia	2021		
Naturally occurring	Decreased circulating concentration of total	Pace et al., 2022		
wildfire-PM <sub>2.5</sub> and elevated	leukocytes, neutrophils, and eosinophils			
THI	Increased eye discharge and coughing			

 $PM_{2.5}$  = fine particulate matter less than  $2.5\mu m$  in diameter,  $PM_{10}$  = course particulate matter between 2.5 and  $10~\mu m$  in diameter, THI = temperature-humidity index, SCC = somatic cell count

## Effects of wildfire smoke exposure on inflammation

Accumulating evidence suggests that the pulmonary disease associated with exposure to wildfire-PM derive from systemic and local inflammatory responses. Production and release of leukocytes from bone marrow was elevated in healthy participants during the 1997 Southeast Asia wildfires compared to a month later after the smoke had dissipated (Tan et al., 2000). Healthy wildland firefighters had an increase in white blood cell count, peripheral blood mononuclear cell (PBMC) count, and serum concentrations of the pro-inflammatory cytokines interleukin (IL)-6 (IL-6) and IL-8, one day after fighting wildland fires compared to the day before exposure (Swiston et al., 2008). Similarly, cytokines, such as IL-1 $\beta$  and IL-6, which induce white blood cell production in bone marrow and production of acute phase proteins in liver, were higher in the blood of healthy individuals following acute exposure to wildfire-PM (van Eeden et al., 2001). Cytokine release plays an important role in initiating and regulating the inflammatory response and inducing neutrophil migration to the site of injury or infection (Ferreira et al., 2018).

A plethora of research also implicates local inflammation in respiratory disease following wildfire smoke exposure. In wildland firefighters, there was an increase in granulocytes, specifically neutrophils, in sputum samples collected the day after compared to the day before

the firefighters were combating an active wildfire (Swiston et al., 2008). Intratracheal instillation of wildfire-PM in mice resulted in inflammatory cell influx, including monocytes and neutrophils, to the lungs and evidence of lung damage within 24 h (Wegesser et al., 2009). A follow-up study by the same group reported an increase in the concentration of proinflammatory cytokines, such as TNF- $\alpha$  and IL-8, in lung lavage samples collected 6 h after instillation, indicating induction of a local inflammatory response (Wegesser et al., 2010). These studies indicate that exposure to wildfire emissions is associated with both local and systemic inflammation.

In cattle, there is currently little evidence to indicate that wildfire smoke induces an inflammatory response, but further research is needed in this area. Before and after exposure to wildfire smoke in dairy cattle, there were no detectable changes in circulating acute phase proteins, such as serum amyloid A and haptoglobin, which play a role in the inflammatory response (Anderson et al., 2022). Although a drop in neutrophil concentration in the blood of dairy cows and calves have been documented after wildfire smoke exposure, the mechanisms contributing to the decline are unknown (Anderson et al., 2022; Pace et al., 2022). Given that neutrophils migrate to the site of tissue injury and infection, it is possible that neutrophil declines in the blood after wildfire smoke inhalation are associated with migration to the lungs and local inflammation, as observed in humans and rodents (Swiston et al., 2008; Wegesser et al. 2009), but this hypothesis has yet to be tested in cattle.

## Effects of wildfire smoke exposure on pathogen susceptibility

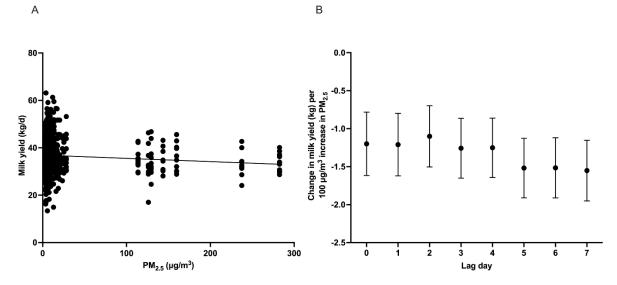
Inhalation of wildfire smoke increases the susceptibility of the respiratory system to microorganisms that contribute to disease. A retrospective study found that the number of influenza cases diagnosed during the winter was positively correlated with PM<sub>2.5</sub> concentrations during the previous summer wildfire season in western Montana (Landguth et al., 2020). Furthermore, Migliaccio et al., (2013) showed that the lungs of mice pre-exposed to wood smoke had a greater pathogen burden when inoculated with *Streptococcus pneumoniae* compared to mice that were not pre-exposed to smoke. Although specific effects of wildfire smoke exposure on pathogen invasion in cattle have not yet been explored, inadequate ventilation and poor air quality on farm, including elevated aerosolized bacterial counts and ammonia, are known risk factors for BRD development in cattle (Lago et al., 2006; Peek et al., 2018; Zhao et al., 2021). It is plausible that inhalation of toxic gases or PM in wildfire smoke increases cattle vulnerability to contracting viruses that contribute to BRD (Fulton, 2020). It is also worth noting that researchers recently discovered the transport of viable microbes through wildfire-smoke plumes; however, the impacts of inhaling smoke-microbes on human and animal health are largely unknown (Kobziar et al., 2018; Kobziar et al., 2019).

## Effects of particulate matter on lactation performance

A few studies have investigated the impacts of PM and other air pollutants on milk production. Beaupied et al (2022) reported reduced milk yield and higher somatic cell count (SCC) in dairy cattle experiencing elevated ambient- (i.e., non-wildfire) and wildfire-PM concurrent with elevated air temperature and humidity index (THI). Our group found that for every 100 ug/m³ increase in wildfire-PM, milk yield decreased by 1.2-1.5 kg/cow/day and milk protein concentration decreased by 0.14% independent of THI, and these effects persisted for at least 7

days after the last day of exposure to wildfire smoke (Figure 1; Anderson et al., 2022). Similarly, lactating ewes housed in a moderately ventilated showed a reduced exposure to PM and increased milk yield compared to that of a poorly ventilated barn (Sevi et al., 2003).

Figure 1. Relationship between PM<sub>2.5</sub> from wildfire smoke and milk production in early-lactation Holstein cows. Cows experienced 7 consecutive days of elevated PM<sub>2.5</sub> from wildfires. A) Milk yield was lower with increasing daily PM<sub>2.5</sub> concentration (P = 0.004). B) Change in average milk yield per cow per day for every 100 µg/m3 increase in PM<sub>2.5</sub> across lag d-0 (day of exposure) through lag d-7 (7 days after last exposure). From Anderson et. al., 2022.



The causal linkages between poor air quality and lactation performance are unclear but may be associated with interacting changes in metabolism, immune status, and feed intake. The immediate metabolic response to wildfire-PM exposure in lactating dairy cattle includes a decrease in blood urea nitrogen and an increase in plasma non-esterified fatty acids, which are reversed with continued exposure to elevated PM (Anderson et al., 2022). As the induction of an immune response is energetically demanding, it is also possible that immune activation in response to inhaled PM or secondary pathogen infection following PM exposure occurs at the expense of other energetically-demanding physiological processes such as milk production (Bird, 2019). Previous work showing positive correlations between wildfire-PM concentrations in the atmosphere and incidences of mastitis in dairy cows (Anderson et al., 2020) and increases in milk SCC with elevated PM (Beaupied et al., 2022) suggest that air pollutants contribute to or worsen mammary infection. Finally, changes in feed intake may precipitate or exacerbate metabolic shifts during exposure to wildfire-PM with a consequent reduction in milk yield, such as occurs in response to other environmental stressors (e.g., heat stress; Rhoads et al., 2009; Wheelock et al., 2010). Although loss of body condition and body weight occur during and following wildfire-PM exposure in dairy cows, feed intake has yet to be directly measured. Further research is warranted to investigate the physiological mechanisms underlying changes in milk synthesis during and following episodes of poor air quality in cattle.

## Effects of wildfire smoke exposure during pregnancy and early postnatal life

Adverse environmental conditions experienced during critical windows of early development,

such as during the prenatal and postnatal period, can alter the developmental trajectory of the offspring (i.e., developmental programming), leading to lifelong or permanent impacts on physiology and health (Lucas, 1991; Barker et al., 2002). Developmental programming of offspring performance associated with prenatal exposure to PM has not been studied in cattle, but there are multiple reports in humans of premature birth and low birth weight in babies gestated under conditions of poor air quality (O'Donnell and Behie, 2013; Abdo et al., 2019; Zhu et al., 2019). For example, Abdo et al., (2019) reported a 13.2% increase in the likelihood of preterm birth with every 1  $\mu$ g/m³ increase in average wildfire smoke PM<sub>2.5</sub> experienced during the second trimester. Further, although there is evidence that low birthweight is a risk factor for the development of metabolic dysfunction and disease in adulthood (Barker et al., 2005), investigations of the long-term consequences of prenatal exposure to wildfire-PM are lacking both in cattle and humans.

Postnatal exposure to wildfire-PM impacts offspring physiology and health. In rhesus macaques, exposure to wildfire-PM<sub>2.5</sub> for 10 consecutive days during infancy decreased immune function and lung capacity at adolescence and adulthood relative to an unexposed group (Miller et al., 2013; Black et al., 2017b; Bassein et al., 2019). Furthermore, white blood cells collected from adolescent monkeys exposed to wildfire smoke early in life produced fewer cytokines, such as IL-6 and IL-8, in response to a lipopolysaccharide challenge relative to control monkeys, indicating long-term immune dysfunction (Miller et al., 2013; Black et al., 2014; Black et al., 2017b). In dairy calves, elevated wildfire-PM<sub>2.5</sub> concentrations in combination with elevated THI during the pre-weaning period were associated with changes in immune cell populations in systemic circulation including reduced total white blood cell, neutrophil, and eosinophil counts, indicative of infection or immune suppression (Pace et al., 2022). The calves also exhibited signs of respiratory and ocular irritation including increased ocular discharge and coughing. Furthermore, across a 5-year period and two dairy farms in the Pacific Northwest, dairy calf mortality was higher on days when PM2.5 was elevated from wildfires. Other air pollutants, such as ozone from non-wildfire sources, also contribute to mortality risk in preweaned and weaned calves on the day of exposure and up to several weeks later (Egberts et al., 2019). Van Leenen et al., (2021) also documented increased neutrophil influx to the lungs and greater incidence of pneumonia in beef and dairy calves associated with elevated PM concentrations in dust in the calf barns. Future research is needed to assess long-term or permanent impacts of early life exposure to wildfire smoke on productivity and health in calves.

## **Conclusions**

As wildfires continue to burn more intensely and across larger areas, the numbers of cattle exposed to, and affected by, wildfire smoke will continue to expand. This review summarized the current state of knowledge regarding the health and production impacts of wildfire-PM exposure in cattle and the many gaps in knowledge. Specific areas worthy of further exploration in cattle are pulmonary immune responses to inhaled wildfire-PM, opportunistic infections following wildfire-PM inhalation, and short and long-term phenotypic consequences from wildfire-PM exposure in utero and in early postnatal life. Armed with this information, preventative and responsive measures can be developed to aid producers and protect livestock herds.

## **Acknowledgments**

Funding for this work was provided by the University of Idaho College of Agricultural and Life Sciences and the Department of Animal, Veterinary, and Food Sciences (Moscow, ID). The authors declare no conflicts of interest.

## **REFERENCES**

- Abdo, M., I. Ward, K. O'Dell, B. Ford, J. R. Pierce, E. V. Fischer, and J. L. Crooks. 2019. Impact of wildfire smoke on adverse pregnancy outcomes in Colorado, 2007–2015. Int. J. Environ.l Res. 16: 3720.
- Anderson, A., P. Rezamand, and A. L. Skibiel. 2022. Effects of wildfire smoke exposure on innate immunity, metabolism, and milk production in lactating dairy cows. J. Dairy Sci. 105:7047-7060.
- Anderson, A. A., P. Rezamand, A. Ahmadzadeh, and A. L. Skibiel. 2020. Effects of particulate matter on health and production of dairy cattle. J. Dairy Sci. 103 (Suppl. 1):283 (Abstr).
- Barker, B. J. P., J. G. Eriksson, T. Forsén, and C. Osmond. 2002. Fetal origins of adult disease: strength of effects and biological basis. Int. J. Epidemiol. 31:1235-1239.
- Barker, D. J., C. Osmond, T. J. Forsén, E. Kajantie, and J. G. Eriksson. 2005. Trajectories of growth among children who have coronary events as adults. N Engl J Med 353:1802-1809.
- Bassein, J., S. Ganesh, M. Dela Pena-Ponce, J. De Backer, M. Lanclus, D. Belmans, C. Van Holsbeke, and L. Miller. 2019. Wildfire smoke exposure during infancy results in impaired lung function. Eur. Respir. J.I 54:PA2830.
- Beaupied, B. L., H. Martinez, S. Martenies, C. S. McConnel, I. B. Pollack, D. Giardina, E. V. Fischer, S. Jathar, C. G. Duncan, and S. Magzamen. 2022. Cows as canaries: The effects of ambient air pollution exposure on milk production and somatic cell count in dairy cows. Environ. Res. 207:112197.
- Bird, L. 2019. Getting enough energy for immunity. Nat. Rev. Immunol. 19:269-269.
- Black, C., Y. Tesfaigzi, J. A. Bassein, and L. A. Miller. 2017a. Wildfire smoke exposure and human health: Significant gaps in research for a growing public health issue. Environ. Toxicol. Pharmacol. 55:186-195.
- Black, C., J. E. Gerriets, J. H. Fontaine, E. S. Schelegle, F. Tablin, and L. A. Miller. 2014. Wildfire smoke exposure during infancy results in constitutive attenuation of transcription factor and signaling genes associated with the toll like receptor pathway in adults. Am. J. Respir. Crit. 189:A3837.
- Black, C., J. E. Gerriets, J. H. Fontaine, R. W. Harper, N. J. Kenyon, F. Tablin, E. S. Schelegle, and L. A. Miller. 2017b. Early life wildfire smoke exposure is associated with immune dysregulation and lung function decrements in adolescence. Am. J. Respir. Cell Mol. 56:657-666.
- Carvalho, T. C., J. I. Peters, and R. O. Williams. 2011. Influence of particle size on regional lung deposition What evidence is there? Int. J. Pharm. 406:1-10.
- Cox, B., A. Gasparrini, B. Catry, F. Fierens, J. Vangronsveld, and T. S. Nawrot. 2016. Ambient air pollution-related mortality in dairy cattle: does it corroborate human findings? Epidemiology 27:779-786.
- DeFlorio-Barker, S., J. Crooks, J. Reyes, and A. G. Rappold. 2019. Cardiopulmonary effects of fine particulate matter exposure among older adults, during wildfire and non-wildfire periods, in the United States 2008-2010. Environ. Health Perspect. 127:37006.

- Dubrovsky, S. A., A. L. Van Eenennaam, B. M. Karle, P. V. Rossitto, T. W. Lehenbauer, and S. S. Aly. 2019. Bovine respiratory disease (BRD) cause-specific and overall mortality in preweaned calves on California dairies: The BRD 10K study. J. Dairy Sci. 102:7320-7328.
- Egberts, V., G. van Schaik, B. Brunekreef, and G. Hoek. 2019. Short-term effects of air pollution and temperature on cattle mortality in the Netherlands. Prev. Vet. Med. 168:1-8.
- Eid, B., D. Beggs, and P. Mansell. 2021. The Impact of Bushfire Smoke on Cattle—A Review. Animals 11:848.
- Ferreira, V. L., H. H. L. Borba, A. d. F. Bonetti, L. P. Leonart, and R. Pontarolo. 2018. Cytokines and Interferons: Types and Functions. Page Ch. 4 in Autoantibodies and Cytokines. K. Wahid Ali, ed. IntechOpen, Rijeka.
- Flannigan, M. D., B. D. Amiro, K. A. Logan, B. J. Stocks, and B. M. Wotton. 2006. Forest fires and climate change in the 21st century. Mitig. Adapt. Strateg. Glob. Change 11:847-859.
- Fu, M., F. Zheng, X. Xu, and L. Niu. 2011. Advances of study on monitoring and evaluation of PM2.5 pollution. Meteorol. Disaster Reduct. Res. 34:1-6.
- Fulton, R. W. 2020. Viruses in bovine respiratory disease in North America: knowledge advances using genomic testing. Vet. Clin. North Am. Food. Anim. Pract. 36:321-332.
- Gan, R. W., B. Ford, W. Lassman, G. Pfister, A. Vaidyanathan, E. Fischer, J. Volckens, J. R. Pierce, and S. Magzamen. 2017. Comparison of wildfire smoke estimation methods and associations with cardiopulmonary-related hospital admissions. GeoHealth 1:122-136.
- Groß, S., M. Esselborn, B. Weinzierl, M. Wirth, A. Fix, and A. Petzold. 2013. Aerosol classification by airborne high spectral resolution lidar observations. Atmos. Chem. Phys. 13:2487-2505.
- Huttunen, K., T. Siponen, I. Salonen, T. Yli-Tuomi, M. Aurela, H. Dufva, R. Hillamo, E. Linkola, J. Pekkanen, A. Pennanen, A. Peters, R. O. Salonen, A. Schneider, P. Tiittanen, M.-R. Hirvonen, and T. Lanki. 2012. Low-level exposure to ambient particulate matter is associated with systemic inflammation in ischemic heart disease patients. Environ. Res. 116:44-51.
- IPCC. 2014. Climate change 2014: synthesis report. Page 151 in Contribution of working groups I, II and III of the fifth assessment report of the intergovernmental panel on climate change. T. c. w. team., R. K. Pachauri, and L. A. Meyer, ed, Geneva, Switzerland.
- Jakus, P. M., M. Kim, R. C. Martin, J. Stout, I. Hammond, E. Hammill, and N. Mesner. 2017. Wildfire in Utah: The Physical and Economic Consequences of Wildfire. Utah State University Logan, UT, USA.
- Johnston, F., I. Hanigan, S. Henderson, G. Morgan, and D. Bowman. 2011. Extreme air pollution events from bushfires and dust storms and their association with mortality in Sydney, Australia 1994–2007. Environ. Res. 111:811-816.
- Kirschvink, N. 2008. Respiratory function in cattle: impact of breed, heritability and external factors. Dtsch. Tierarztl. Wochenschr. 115:265-270.
- Kobziar, L. N., M. R. Pingree, H. Larson, T. J. Dreaden, S. Green, and J. A. Smith. 2018.

  Pyroaerobiology: the aerosolization and transport of viable microbial life by wildland fire.

  Ecosphere 9:e02507.
- Kobziar, L. N., M. R. A. Pingree, A. C. Watts, K. N. Nelson, T. J. Dreaden, and M. Ridout. 2019. Accessing the life in smoke: A new application of unmanned aircraft systems (UAS) to sample wildland fire bioaerosol emissions and their environment. Fire 2:56.
- Lago, A., S. McGuirk, T. Bennett, N. Cook, and K. Nordlund. 2006. Calf respiratory disease and pen microenvironments in naturally ventilated calf barns in winter. J. Dairy Sci. 89:4014-4025.
- Landguth, E. L., Z. A. Holden, J. Graham, B. Stark, E. B. Mokhtari, E. Kaleczyc, S. Anderson, S. Urbanski, M. Jolly, E. O. Semmens, D. A. Warren, A. Swanson, E. Stone, and C. Noonan.

- 2020. The delayed effect of wildfire season particulate matter on subsequent influenza season in a mountain west region of the USA. Environment International 139:105668.
- Landis, M. S., E. S. Edgerton, E. M. White, G. R. Wentworth, A. P. Sullivan, and A. M. Dillner. 2018.

  The impact of the 2016 Fort McMurray Horse River Wildfire on ambient air pollution levels in the Athabasca Oil Sands Region, Alberta, Canada. Sci. Total Environ. 618:1665-1676.
- Liu, J. C., G. Pereira, S. A. Uhl, M. A. Bravo, and M. L. Bell. 2015. A systematic review of the physical health impacts from non-occupational exposure to wildfire smoke. Environ. Res. 136:120-132.
- Lohmann-Matthes, M. L., C. Steinmuller, and G. Franke-Ullmann. 1994. Pulmonary macrophages. Eur. Respir. J. 7:1678.
- Löndahl, J., A. Massling, J. Pagels, E. Swietlicki, E. Vaclavik, and S. Loft. 2007. Size-Resolved Respiratory-Tract Deposition of Fine and Ultrafine Hydrophobic and Hygroscopic Aerosol Particles During Rest and Exercise. Inhal. Toxicol. 19:109-116.
- Lucas, A. 1991. Programming by early nutrition in man. Ciba Found. Symp. 156:38-55.
- Mariassy, A. T., C. G. Plopper, and D. L. Dungworth. 1975. Characteristics of bovine lung as observed by scanning electron microscopy. Anat. Rec. 183:13-25.
- Michel, C., C. Liousse, J. M. Gregoire, K. Tansey, G. R. Carmichael, and J. H. Woo. 2005. Biomass burning emission inventory from burnt area data given by the
- SPOT-VEGETATION system in the frame of TRACE-P and ACE-Asia campaigns. J. Geophys. Res. 110:D09304.
- Migliaccio, C. T., E. Kobos, Q. O. King, V. Porter, F. Jessop, and T. Ward. 2013. Adverse effects of wood smoke PM2.5 exposure on macrophage functions. Inhal. Toxicol. 25:67-76.
- Miller, D. D., A. Bajracharya, G. N. Dickinson, T. A. Durbin, J. K. P. McGarry, E. P. Moser, L. A. Nuñez, E. J. Pukkila, P. S. Scott, P. J. Sutton, and N. A. C. Johnston. 2022. Diffusive uptake rates for passive air sampling: Application to volatile organic compound exposure during FIREX-AQ campaign. Chemosphere 287:131808.
- Miller, L. A., E. S. Schelegle, J. P. Capitanio, C. C. Clay, and W. F. Walby. 2013. Persistent immune effects of wildfire PM exposure during childhood development. California Air Resources Board.
- Morgan, G., V. Sheppeard, B. Khalaj, A. Ayyar, D. Lincoln, B. Jalaludin, J. Beard, S. Corbett, and T. Lumley. 2010. Effects of bushfire smoke on daily mortality and hospital admissions in Sydney, Australia. Epidemiology 21:47-55.
- Nakayama Wong, L. S., H. H. Aung, M. W. Lamé, T. C. Wegesser, and D. W. Wilson. 2011. Fine particulate matter from urban ambient and wildfire sources from California's San Joaquin Valley initiate differential inflammatory, oxidative stress, and xenobiotic responses in human bronchial epithelial cells. Toxicol. in Vitro 25:1895-1905.
- NASS, U. 2019. 2017 Census of Agriculture. in Geographic Area Series. Vol. 1. USDA. National Agricultural Statistics Service (NASS). Accessed Sep. 14, 2020.

  <a href="https://www.nass.usda.gov/Publications/AgCensus/2017/Full\_Report/Volume\_1">https://www.nass.usda.gov/Publications/AgCensus/2017/Full\_Report/Volume\_1</a>, Chapter 1 US/usv1.pdf
- NICC. 2022. Incident Management Situation Report. Thursday May 5, 2022. National Interagency Coordination Center.
- O'Donnell, M. H. and A. M. Behie. 2013. Effects of bushfire stress on birth outcomes: A cohort study of the 2009 Victorian Black Saturday bushfires. Int. J. Disaster Risk Reduct. 5:98-106.
- O'Hara, K. C., J. Ranches, L. M. Roche, T. K. Schohr, R. C. Busch, and G. U. Maier. 2021. Impacts from wildfires on livestock health and production: producer perspectives. Animals 11:3230.

- Pace, A., P. Rezamand, and A. L. Skibiel. 2022. Effects of wildfire smoke PM2.5 on preweaned Holstein dairy calves. J. Dairy Sci. 105 (Suppl. 1):113 (Abstr).
- Peek, S. F., O. T.L., and D. T.J. 2018. Respiratory diseases. Rebhun's Diseases of Dairy Cattle 2018:94–167.
- Reid, C. E., M. Brauer, F. H. Johnston, M. Jerrett, J. R. Balmes, and C. T. Elliott. 2016. Critical review of health impacts of wildfire smoke exposure. Environ. Health Perspect. 124:1334-1343.
- Rhoads, M. L., R. P. Rhoads, M. J. VanBaale, R. J. Collier, S. R. Sanders, W. J. Weber, B. A. Crooker, and L. H. Baumgard. 2009. Effects of heat stress and plane of nutrition on lactating Holstein cows: Production, metabolism, and aspects of circulating somatotropin. J. Dairy Sci. 92:1986-1997.
- Schulze, F., X. Gao, D. Virzonis, S. Damiati, M. R. Schneider, and R. Kodzius. 2017. Air Quality Effects on Human Health and Approaches for Its Assessment through Microfluidic Chips. Genes 8:244.
- Sevi, A., L. Taibi, M. Albenzio, M. Caroprese, R. Marino, and A. Muscio. 2003. Ventilation effects on air quality and on the yield and quality of ewe milk in winter. J Dairy Sci 86:3881-3890.
- Shaposhnikov, D., B. Revich, T. Bellander, G. B. Bedada, M. Bottai, T. Kharkova, E. Kvasha, E. Lezina, T. Lind, E. Semutnikova, and G. Pershagen. 2014. Mortality related to air pollution with the moscow heat wave and wildfire of 2010. Epidemiology 25:359-364.
- Sokolik, I. N., A. J. Soja, P. J. DeMott, and D. Winker. 2019. Progress and Challenges in Quantifying Wildfire Smoke Emissions, Their Properties, Transport, and Atmospheric Impacts. Journal of Geophysical Research: Atmospheres 124:13005-13025.
- Stowell, J. D., G. Geng, E. Saikawa, H. H. Chang, J. Fu, C.-E. Yang, Q. Zhu, Y. Liu, and M. J. Strickland. 2019. Associations of wildfire smoke PM2.5 exposure with cardiorespiratory events in Colorado 2011–2014. Environ. Internat. 133:105151.
- Swiston, J. R., W. Davidson, S. Attridge, G. T. Li, M. Brauer, and S. F. van Eeden. 2008. Wood smoke exposure induces a pulmonary and systemic inflammatory response in firefighters. Eur. Respir. J. 32:129-138.
- Tan, W. C., D. Qiu, B. L. Liam, T. P. Ng, S. H. Lee, S. F. van Eeden, Y. D'Yachkova, and J. C. Hogg.2000. The Human Bone Marrow Response to Acute Air Pollution Caused by Forest Fires. Am.J. Respir. Crit. 161:1213-1217.
- Urbanski, S. P., W. M. Hao, and S. Baker. 2009. Chapter 4 Chemical Composition of Wildland Fire Emissions. Pages 79-107 in Developments in Environmental Science. Vol. 8. A. Bytnerowicz, M. J. Arbaugh, A. R. Riebau, and C. Andersen, ed. Elsevier.
- USDA. 2017. Death loss in U. S. cattle and calves due to predator and nonpredator causes, 2015. United States Department of Agriculture.
- U. S. EPA 2010. Overview of Airborne Metals Regulations, Exposure Limits, Health Effects, and Contemporary Research. United States Environmental Protection Agency.
- U. S. EPA. 2013. National ambient air quality standards for particulate matter, final rule. Pages 3085- 3287 in Federal Register. Vol. 78.
- U. S. EPA. 2020. 2017 National Emissions Inventory (NEI). United States Environmental Protection Agency.
- USGCRP. 2016. The impacts of climate change on human health in the United States: A scientific assessment. Page 312 in U.S. Global Change Research Program. A. J. Balbus, J. L. Gamble, C. B. Beard, J. E. Bell, D. Dodgen, R. J. Eisen, N. Fann, M. D. Hawkins, S. C. Herring, L. Jantarasami, D. M. Mills, S. Saha, M. C. Sarofim, J. Trtanj, and L. Ziska, ed, Washington, DC.

- van Eeden, S. F., W. C. Tan, T. Suwa, H. Mukae, T. Terashima, T. Fujii, D. Qui, R. Vincent, and J. C. Hogg. 2001. Cytokines involved in the systemic inflammatory response induced by exposure to particulate matter air pollutants (PM10). Am. J. Respir. Crit. 164:826-830.
- van Leenen, K., J. Jouret, P. Demeyer, P. Vermeir, D. Leenknecht, L. Van Driessche, L. De Cremer, C. Masmeijer, F. Boyen, P. Deprez, E. Cox, B. Devriendt, and B. Pardon. 2021. Particulate matter and airborne endotoxin concentration in calf barns and their association with lung consolidation, inflammation, and infection. J. Dairy Sci. 104:5932-5947.
- Veit, H. P. and R. L. Farrell. 1978. The anatomy and physiology of the bovine respiratory system relating to pulmonary disease. Cornell Vet. 68:555-581.
- Venn-Watson, S., C. R. Smith, E. D. Jensen, and T. Rowles. 2013. Assessing the potential health impacts of the 2003 and 2007 firestorms on bottlenose dolphins (Tursiops trucatus) in San Diego Bay. Inhal. Toxicol. 25:481-491.
- Vicente, A., C. Alves, A. I. Calvo, A. P. Fernandes, T. Nunes, C. Monteiro, S. M. Almeida, and C. Pio. 2013. Emission factors and detailed chemical composition of smoke particles from the 2010 wildfire season. Atmos. Environ. 71:295-303.
- Wegesser, T. C., L. M. Franzi, F. M. Mitloehner, A. Eiguren-Fernandez, and J. A. Last. 2010. Lung antioxidant and cytokine responses to coarse and fine particulate matter from the great California wildfires of 2008. Inhal. Toxicol. 22:561-570.
- Wegesser, T. C., K. E. Pinkerton, and J. A. Last. 2009. California wildfires of 2008: coarse and fine particulate matter toxicity. Environ. Health Perspect. 117:893-897.
- Wentworth, G. R., Y.-a. Aklilu, M. S. Landis, and Y.-M. Hsu. 2018. Impacts of a large boreal wildfire on ground level atmospheric concentrations of PAHs, VOCs and ozone. Atmos. Environ. 178:19-30.
- Wheelock, J. B., R. P. Rhoads, M. J. VanBaale, S. R. Sanders, and L. H. Baumgard. 2010. Effects of heat stress on energetic metabolism in lactating Holstein cows. J. Dairy Sci. 93:644-655.
- Wilson, W. E. and H. H. Suh. 1997. Fine particles and coarse particles: concentration relationships relevant to epidemiologic studies. J.. Air Waste Manag Assoc. 47:1238-1249.
- Zhao, W., C. Y. Choi, G. Li, H. Li, and Z. Shi. 2021. Pre-weaned dairy calf management practices, morbidity and mortality of bovine respiratory disease and diarrhea in China. Livest. Sci. 251:104608.
- Zhu, J., R. W. Lee, C. Twum, and Y. Wei. 2019. Exposure to ambient PM2.5 during pregnancy and preterm birth in metropolitan areas of the state of Georgia. Environ. Sci. Pollut. Res. 26:2492–2500.

## What Really is the Role of Nutrition(ists) in Lameness?

Gerard Cramer, DVM, DVSc. Department of Veterinary Population Medicine University of Minnesota Saint Paul, MN, USA 55108 gcramer@umn.edu

Lameness is a continues to be an animal welfare and impacts the economics of the dairy industry (Ventura et al., 2015; Dolecheck and Bewley, 2018). Most of the dairy cow lameness originates in the hoof (Murray et al., 1996) and in North America the most common hoof lesions are digital dermatitis (DD), sole ulcers (SU), and white line lesions (WLD) (Cramer et al., 2008; DeFrain et al., 2013; Solano et al., 2016). Although infectious lesions such as DD are the most common type of lesions, in most herds hoof horn lesions such as SU and WLD are costlier due to their effects on milk production and culling (Dolecheck and Bewley, 2018). Economic losses due to hoof horn lesions are difficult to quantify, yet it is becoming apparent that cows affected with hoof horn lesions are usually cows with higher production potential and production losses start prior to a lameness diagnosis (Bicalho et al., 2008). Typical production losses for cows with hoof horn lesions range from 200-500 kg plus these cows are also at increased risk of culling (Cramer et al., 2009; Huxley, 2013).

Given the importance of lameness to a dairy farm it is no surprise that there are a wide variety of stakeholders that can impact how a farm addresses lameness. Hoof trimmers are typically considered the main person by farms for both technical and herd level advice (Wynands et al., 2021). Veterinarians and nutritionists also have a role however their role is less technical and considered more advisory by farmers (Wynands et al., 2021). These differing roles can lead do some discomfort of their role and distrust of other advisors (Wynands et al., 2022).

The role of nutrition in lameness has traditionally been considered large, as acidosis was considered to play a large role (Nocek, 1997). This resulted in nutritionists becoming an easy target to blame when a herd had a lameness problem. Recently the focus of the link between acidosis and laminitis has been come under scrutiny and evidence for this link is weak (Lean et al., 2013; Randall et al., 2018).

The aim of this paper is to describe the role of nutrition and nutritional advisors in lameness. Specifically, this paper will briefly review the pathogenesis of the most common causes of lameness and conclude with opportunities for nutritionists to get more involved with lameness.

#### **Digital Dermatitis**

Digital Dermatitis is an infectious bacterial infection of the skin typically located in the interdigital cleft of the foot (ICAR Working Group on Functional Traits and International Claw Health Experts, 2020). The presentation of DD varies ranging from acute painful and ulcerated skin (M2 lesions) to hyperkeratotic or granulomatous lesions (M4 lesions) that are not as painful and more chronic in nature (Döpfer et al., 1997; Berry et al., 2012).

The development of DD requires a breakdown of the natural skin barrier (Gomez et al., 2014; Krull et al., 2016). Once this skin barrier has been broken down a synergistic group of bacteria invade the initial layers of the skin and create an inflammatory process. Several types of bacteria have been isolated from DD lesions, but Treponema species are considered a necessary component of the disease (Krull et al., 2014). Treponema species are microaerophilic, gram-negative spirochaetes that encyst to protect itself. As they invade the epidermis and damage the different skin layers, an immune response occurs that results in hyperkeratosis and proliferative lesions(Döpfer et al., 1997).

Prevention of DD typically focused on maintaining a clean environment and the use of properly designed and used footbaths containing copper sulfate or formalin (Cook, 2017; Jacobs et al., 2019). Skin has a physical, chemical, and immunological role in preventing damage and DD infections. Specific nutrients can contribute to ensuring optimum functioning of these barriers. Zinc has a role in cellular repair and replacement. Copper, Manganese and Zinc also contribute to the immune system through the regulation of specific cells, factors, and antioxidant pathways (Lean et al., 2013; Gomez et al., 2014). Similarly iodine affects the local inflammatory response and can prevent foot rot (Berg et al., 1984). A combination product containing Zn, Cu, Mn and I has been shown to reduce DD development (Gomez et al., 2014). The use a Saccharomyces cerevisiae fermentation product has also shown some promise as potential nutritional aid in DD control (Anklam et al., 2022).

#### **Hoof horn lesions**

Sole ulcers and WLD are both lesions of the horn due to damage to the corium. Visually this can present in clinical signs as mild as hemorrhage to necrotic tissue if the lesion was not treated in an appropriate time frame.

Hoof horn lesions such as SU and WLD are different lesions, but both are thought to be caused by trauma within the hoof and damage to the internal anatomy of the hoof through internal and external concussive forces (Newsome et al., 2016). The lesion that results from these forces is thought to depend are oriented toward in the hoof (Le Fevre et al., 2001; Lischer et al., 2002; Newsome et al., 2016). The first time a cow develops a SU or WLD changes in the hoof anatomy occur, primarily the suspensory apparatus (Lischer et al., 2002; Tarlton et al., 2002) and the supporting structures under the third phalanx or pedal bone (Bicalho et al., 2009; Newsome et al., 2017). Once the function of these structures is impaired, extra concussive force is exerted on the horn-producing tissue, leading to the disruption of horn production, inflammation,

exostosis, and, eventually, the formation of SU and WLD (Newsome et al., 2016). Once these lesion are present the pedal bone continues to change due to the inflammation in the corium resulting in exostosis, increasing the chance of chronic lameness due to a cycle of excessive concussive forces (Newsome et al., 2016). Therefore, it is important to consider previous lameness history when evaluating a dairy's records.

Prevention of hoof horn lesion is multifactorial with hoof trimming, standing time and the transition period thought to play a key role (Stoddard and Cramer, 2017; Randall et al., 2018). Nutritionally excessive carbohydrates and protein have been suggested as causes of hoof horn lesions. Evidence for this hypothesis is limited (Randall et al., 2018) and in some studies the environment (housing/flooring) has a larger impact than diet (Bergsten and Frank, 1996). However is has been suggested that glucose availability (Lübbe, 2015) at the cellular level and insulin concentration and peripheral tissue insulin sensitivity during the period of negative energy balance in early lactation are important factor in SU development (Wilhelm et al., 2017). This supports the epidemiological evidence that the transition period is associate with hoof horn lesion (Proudfoot et al., 2010; Omontese et al., 2020).

Like DD development trace minerals play a role in promoting optimum hoof health. The development of keratin requires adequate supply of both macro and trace minerals. For example calcium is required for an enzyme required in the final differentiation step of keratinocytes (Tomlinson et al., 2004). Similarly, sulfur containing amino acids (Methionine and cysteine) provides cell wall rigidity(Tomlinson et al., 2004; Lean et al., 2013). Zinc has various roles in the keratinization process and mixing of Zn sources can improve hoof health (Nocek et al., 2000; Lean et al., 2013). Like zinc, copper is involved in various enzymes that provide strength to the cell matrix and intracellular cementing substance(Tomlinson et al., 2004; Lean et al., 2013). When it comes to vitamins biotin is the vitamin that is the most important as it is a cofactor for various enzymes and is essential for the formation of the lipids in the intracellular cementing substance (Mülling et al., 1999). Biotin has been show to improve hoof health (Hedges et al., 2001; Bergsten et al., 2003) however it requires long term supplementation (Pötzsch et al., 2003).

#### **Roles for nutritionists**

It is clear nutritionists have a role to play in formulating diets that meet or exceed the requirements for optimum hoof health. There are likely herds in which specific minerals, vitamins or nutritional additives will be effective and other herds where they will not reduce lameness (Nocek et al., 2000; Hedges et al., 2001). What causes this difference between herds is still unclear and hence it provides an opportunity for advisors to evaluate suitability. One tool that is exists is the use of a risk assessment (van Huyssteen et al., 2020; Wynands et al., 2022). A recent study found that although the risk assessment was long it allowed team members to focus on important areas (Wynands et al., 2022). This risk assessment can be found at <a href="https://conservancy.umn.edu/handle/11299/226886">https://conservancy.umn.edu/handle/11299/226886</a>. This same study used a team based collaborative approach between veterinarians, hoof trimmers and nutritionists to work with

farms that resulted in increased collaboration and alignment of goals for common farms (Wynands et al., 2022). These findings suggest that there is a role for nutritionists to become involved with other stakeholders to address lameness in herds where it is a farm priority.

To properly improve communication and collaboration between on-farm stakeholders it is imperative that there is good data collected related to lesions found by the hoof trimming. Specifics of setting up a lameness recording system can be found at https://z.umn.edu/lamenessmanagerwebinar. Nutritionists, due to their regular interactions and data analysis of dairy records, are well suited to either assisting dairies in setting up the system or monitoring the data to ensure performance is meeting the farm's goals. To properly develop and use a recording system will require hoof trimmers and nutritionists to work together and use common language for lesions and causes. Of note is that records should be evaluated considering the lameness history of cows and the effectiveness of interventions should be evaluated on the number of new lesions not all lesions.

Finally, since nutritionists play a large role in all aspects of feeding management it is important to understand the relationship between errors in feeding management and lameness. The biggest impact is likely going to come from ensuring cows have consistent access to feed. Cows waiting for feed induces excessive standing. Standing time plays a key role in the development and recovery of lameness and anything that causes a cow to stand excessively is problematic (Cook, 2020; Tucker et al., 2021). Next since the transition period is a key source of inflammatory process in a cow's life (Bradford et al., 2015) it is important to ensure a cow's transition period is as uneventful as possible. Finally, the low inclusion nature of minerals and vitamins make proper mixing and delivery of diets critically important (Oelberg and Stone, 2014).

#### Conclusion

There are various opportunities for nutritionists to play a role in improving lameness across the dairy industry. This will require moving beyond solely ensuring the diet contains the proper amount of nutrients and feeding management is adequate. Seeking out opportunities for collaboration with other on-farm stakeholders is key to truly create lasting change on dairy farms.

#### References

- Anklam, K., P. Cernek, I. Yoon, J. Wheeler, T. Birkle, and D. Döpfer. 2022. Effects of supplementing a Saccharomyces cerevisiae fermentation product on the prevention and control of digital dermatitis in lactating dairy cows. Applied Animal Science 38:98–109. doi:10.15232/aas.2021-02229.
- Berg, J.N., J.P. Maas, J.A. Paterson, G.F. Krause, and L.E. Davis. 1984. Efficacy of ethylenediamine dihydriodide as an agent to prevent experimentally induced bovine foot rot. Am. J. Vet. Res. 45:1073–1078.
- Bergsten, C., and B. Frank. 1996. Sole haemorrhages in tied primiparous cows as an indicator of periparturient laminitis: effects of diet, flooring and season. Acta Vet. Scand. 37:383–394.
- Bergsten, C., P.R. Greenough, J.M. Gay, W.M. Seymour, and C.C. Gay. 2003. Effects of biotin supplementation on performance and claw lesions on a commercial dairy farm. Journal of dairy science 86:3953–3962. doi:S0022-0302(03)74005-3 [pii].
- Berry, S.L., D.H. Read, T.R. Famula, A. Mongini, and D. Döpfer. 2012. Long-term observations on the dynamics of bovine digital dermatitis lesions on a California dairy after topical treatment with lincomycin HCl. The Veterinary Journal 193:654–658. doi:10.1016/j.tvjl.2012.06.048.
- Bicalho, R.C., V.S. Machado, and L.S. Caixeta. 2009. Lameness in dairy cattle: A debilitating disease or a disease of debilitated cattle? A cross-sectional study of lameness prevalence and thickness of the digital cushion. Journal of Dairy Science 92:3175–3184. doi:10.3168/jds.2008-1827.
- Bicalho, R.C., L.D. Warnick, and C.L. Guard. 2008. Strategies to Analyze Milk Losses Caused by Diseases with Potential Incidence Throughout the Lactation: A Lameness Example. Journal of Dairy Science 91:2653–2661. doi:10.3168/jds.2007-0744.
- Bradford, B.J., K. Yuan, J.K. Farney, L.K. Mamedova, and A.J. Carpenter. 2015. Invited review: Inflammation during the transition to lactation: New adventures with an old flame. Journal of Dairy Science 98:6631–6650. doi:10.3168/jds.2015-9683.
- Cook, N.B. 2017. A Review of the Design and Management of Footbaths for Dairy Cattle. Veterinary Clinics of North America: Food Animal Practice 33:195–225. doi:10.1016/j.cvfa.2017.02.004.
- Cook, N.B. 2020. Symposium review: The impact of management and facilities on cow culling rates. Journal of Dairy Science 103:3846–3855. doi:10.3168/jds.2019-17140.
- Cramer, G., K.D. Lissemore, C.L. Guard, K.E. Leslie, and D.F. Kelton. 2008. Herd- and Cow-Level Prevalence of Foot Lesions in Ontario Dairy Cattle. Journal of Dairy Science 91:3888–3895. doi:10.3168/jds.2008-1135.
- Cramer, G., K.D. Lissemore, C.L. Guard, K.E. Leslie, and D.F. Kelton. 2009. The association between foot lesions and culling risk in Ontario Holstein cows. Journal of Dairy Science 92:2572–2579. doi:10.3168/jds.2008-1532.
- DeFrain, J.M., M.T. Socha, and D.J. Tomlinson. 2013. Analysis of foot health records from 17 confinement dairies. Journal of dairy science 96:7329–7339. doi:10.3168/jds.2012-6017.
- Dolecheck, K., and J. Bewley. 2018. Animal board invited review: Dairy cow lameness expenditures, losses and total cost. animal 12:1462–1474. doi:10.1017/S1751731118000575.
- Döpfer, D., A. Koopmans, F.A. Meijer, I. Szakáll, Y.H. Schukken, W. Klee, R.B. Bosma, J.L. Cornelisse, A.J. van Asten, and A.A. ter Huurne. 1997. Histological and bacteriological evaluation of digital dermatitis in cattle, with special reference to spirochaetes and Campylobacter faecalis. Vet. Rec. 140:620–623.
- Gomez, A., N. Bernardoni, J. Rieman, A. Dusick, R. Hartshorn, D.H. Read, M.T. Socha, N.B. Cook, and D. Döpfer. 2014. A randomized trial to evaluate the effect of a trace mineral premix on the incidence

- of active digital dermatitis lesions in cattle. Journal of Dairy Science 97:6211–6222. doi:10.3168/jds.2013-7879.
- Hedges, J., R.W. Blowey, A.J. Packington, C.J. O'Callaghan, and L.E. Green. 2001. A Longitudinal Field Trial of the Effect of Biotin on Lameness in Dairy Cows. Journal of Dairy Science 84:1969–1975. doi:10.3168/jds.S0022-0302(01)74639-5.
- Huxley, J.N. 2013. Impact of lameness and claw lesions in cows on health and production. Livestock Science 156:64–70. doi:10.1016/j.livsci.2013.06.012.
- van Huyssteen, M., H.W. Barkema, S. Mason, and K. Orsel. 2020. Association between lameness risk assessment and lameness and foot lesion prevalence on dairy farms in Alberta, Canada. Journal of Dairy Science 103:11750–11761. doi:10.3168/jds.2019-17819.
- ICAR Working Group on Functional Traits and International Claw Health Experts ed. . 2020. The ICAR Claw Health Atlas. 2nd ed. ICAR.
- Jacobs, C., C. Beninger, G.S. Hazlewood, K. Orsel, and H.W. Barkema. 2019. Effect of footbath protocols for prevention and treatment of digital dermatitis in dairy cattle: A systematic review and network meta-analysis. Preventive Veterinary Medicine 164:56–71. doi:10.1016/j.prevetmed.2019.01.011.
- Krull, A.C., V.L. Cooper, J.W. Coatney, J.K. Shearer, P.J. Gorden, and P.J. Plummer. 2016. A Highly Effective Protocol for the Rapid and Consistent Induction of Digital Dermatitis in Holstein Calves. PLOS ONE 11:e0154481. doi:10.1371/journal.pone.0154481.
- Krull, A.C., J.K. Shearer, P.J. Gorden, V.L. Cooper, G.J. Phillips, and P.J. Plummer. 2014. Deep Sequencing Analysis Reveals Temporal Microbiota Changes Associated with Development of Bovine Digital Dermatitis. Infection and Immunity 82:3359–3373. doi:10.1128/IAI.02077-14.
- Le Fevre, A.M., D.N. Logue, J.E. Offer, I. McKendrick, and G. Gettinby. 2001. Correlations of measurements of subclinical claw horn lesions in dairy cattle. Veterinary Record 148:135–138. doi:10.1136/vr.148.5.135.
- Lean, I.J., C.T. Westwood, H.M. Golder, and J.J. Vermunt. 2013. Impact of nutrition on lameness and claw health in cattle. Livestock Science 156:71–87. doi:10.1016/j.livsci.2013.06.006.
- Lischer, C.J., P. Ossent, M. Räber, and H. Geyer. 2002. Suspensory structures and supporting tissues of the third phalanx of cows and their relevance to the development of typical sole ulcers (Rusterholz ulcers). Veterinary Record 151:694–698. doi:10.1136/vr.151.23.694.
- Lübbe, K. 2015. Entwicklung und Einsatz eines In-vitro-Ischämiemodels zur Untersuchung zellulärer Pathomechanismen der Klauenrehe des Rindes.
- Mülling, C.K., H.H. Bragulla, S. Reese, K.D. Budras, and W. Steinberg. 1999. How structures in bovine hoof epidermis are influenced by nutritional factors.. Anatomia, histologia, embryologia 28:103–108.
- Murray, R.D., D.Y. Downham, M.J. Clarkson, W.B. Faull, J.W. Hughes, F.J. Manson, J.B. Merritt, W.B. Russell, J.E. Sutherst, and W.R. Ward. 1996. Epidemiology of lameness in dairy cattle: description and analysis of foot lesions. Veterinary Record 138:586–591. doi:10.1136/vr.138.24.586.
- Newsome, R., M.J. Green, N.J. Bell, M.G.G. Chagunda, C.S. Mason, C.S. Rutland, C.J. Sturrock, H.R. Whay, and J.N. Huxley. 2016. Linking bone development on the caudal aspect of the distal phalanx with lameness during life. Journal of Dairy Science 99:4512–4525. doi:10.3168/jds.2015-10202.
- Newsome, R.F., M.J. Green, N.J. Bell, N.J. Bollard, C.S. Mason, H.R. Whay, and J.N. Huxley. 2017. A prospective cohort study of digital cushion and corium thickness. Part 1: Associations with body condition, lesion incidence, and proximity to calving. Journal of Dairy Science 100:4745–4758. doi:10.3168/jds.2016-12012.

- Nocek, J.E. 1997. Bovine Acidosis: Implications on Laminitis. Journal of Dairy Science 80:1005–1028. doi:10.3168/jds.S0022-0302(97)76026-0.
- Nocek, J.E., A.B. Johnson, and M.T. Socha. 2000. Digital characteristics in commercial dairy herds fed metal-specific amino acid complexes. J. Dairy Sci. 83:1553–1572. doi:10.3168/jds.S0022-0302(00)75028-4.
- Oelberg, T.J., and W. Stone. 2014. Monitoring Total Mixed Rations and Feed Delivery Systems. Veterinary Clinics: Food Animal Practice 30:721–744. doi:10.1016/j.cvfa.2014.08.003.
- Omontese, B.O., R.S. Bisinotto, and G. Cramer. 2020. Evaluating the association between early-lactation lying behavior and hoof lesion development in lactating Jersey cows. Journal of Dairy Science 103:10494–10505. doi:10.3168/jds.2020-18254.
- Pötzsch, C.J., V.J. Hedges, R.W. Blowey, A.J. Packington, and L.E. Green. 2003. The impact of parity and duration of biotin supplementation on white line disease lameness in dairy cattle. J. Dairy Sci. 86:2577–2582.
- Proudfoot, K.L., D.M. Weary, and M.A.G. von Keyserlingk. 2010. Behavior during transition differs for cows diagnosed with claw horn lesions in mid lactation. Journal of Dairy Science 93:3970–3978. doi:10.3168/jds.2009-2767.
- Randall, L.V., M.J. Green, and J.N. Huxley. 2018. Use of statistical modelling to investigate the pathogenesis of claw horn disruption lesions in dairy cattle. The Veterinary Journal 238:41–48. doi:10.1016/j.tvjl.2018.07.002.
- Solano, L., H.W. Barkema, S. Mason, E.A. Pajor, S.J. LeBlanc, and K. Orsel. 2016. Prevalence and distribution of foot lesions in dairy cattle in Alberta, Canada. Journal of Dairy Science 99:6828–6841. doi:10.3168/jds.2016-10941.
- Stoddard, G.C., and G. Cramer. 2017. A Review of the Relationship Between Hoof Trimming and Dairy Cattle Welfare. Veterinary Clinics of North America: Food Animal Practice 33:365–375. doi:10.1016/j.cvfa.2017.02.012.
- Tarlton, J.F., D.E. Holah, K.M. Evans, S. Jones, G.R. Pearson, and A.J.F. Webster. 2002. Biomechanical and Histopathological Changes in the Support Structures of Bovine Hooves around the Time of First Calving. The Veterinary Journal 163:196–204. doi:10.1053/tvjl.2001.0651.
- Tomlinson, D.J., C.H. Mülling, and T.M. Fakler. 2004. Invited review: formation of keratins in the bovine claw: roles of hormones, minerals, and vitamins in functional claw integrity. Journal of dairy science 87:797–809.
- Tucker, C.B., M.B. Jensen, A.M. de Passillé, L. Hänninen, and J. Rushen. 2021. Invited review: Lying time and the welfare of dairy cows. Journal of Dairy Science 104:20–46. doi:10.3168/jds.2019-18074.
- Ventura, B.A., M.A.G. von Keyserlingk, and D.M. Weary. 2015. Animal Welfare Concerns and Values of Stakeholders Within the Dairy Industry. Journal of Agricultural and Environmental Ethics 28:109–126. doi:10.1007/s10806-014-9523-x.
- Wilhelm, K., J. Wilhelm, and M. Fürll. 2017. Claw disorders in dairy cattle an unexpected association between energy metabolism and sole haemorrhages. Journal of Dairy Research 84:54–60. doi:10.1017/S0022029916000510.
- Wynands, E.M., S.M. Roche, G. Cramer, and B.A. Ventura. 2021. Dairy farmer, hoof trimmer, and veterinarian perceptions of barriers and roles in lameness management. Journal of Dairy Science 104:11889–11903. doi:10.3168/jds.2021-20603.
- Wynands, E.M., S.M. Roche, G. Cramer, and B.A. Ventura. 2022. Promoting farm advisor engagement and action toward the improvement of dairy cattle lameness. Journal of Dairy Science 105:6364–6377. doi:10.3168/jds.2021-21745.

# Forage cover crops and their potential benefits and unknowns to western cattle operations

Jillian D. Bainard Agriculture & Agri-Food Canada jillian.bainard@agr.gc.ca

Integrated crop-livestock systems are increasing in popularity across the Pacific Northwest. Annual crops can provide nutritious forage for livestock at various times in the growing season, and the animals in turn provide nutrient additions to the soil. Incorporating grazing livestock onto annual crop land also often involves the use of diverse annual crop mixtures (also known as polycrops, polycultures, or cocktail mixtures) that can contribute to a range of other ecosystem services. Polycultures can provide several benefits compared to monocultures including increased yield and yield stability, reduced inputs and weed suppression benefits, improved soil fertility, and increased yield of subsequent crops (e.g., Lithourgidis et al. 2011; Szumigalski and Van Acker 2006).

In our research, we have explored the utility of using diverse annual forage crops as forage in the Canadian Prairies. There appears to be the potential improvement of forage dry matter production, but production is impacted by site and seasonal growing conditions (Bainard et al. 2020a). Under poor growing conditions, biomass production can be particularly limited by weeds (Bainard, unpublished). In other cases (and under good growing conditions) there is some evidence of improved weed control with the inclusion of crops such as barley and radish (Bainard et al. 2020a).

In regards to forage quality, the inclusion of diverse annual crops can improve forage nutrition. In particular, mixtures had higher organic matter digestibility, lower acid detergent fiber and neutral detergent fiber, and higher crude protein as compared to an oats monoculture (Bainard et al. 2020b). Other nutrients were also found to increase in the mixtures including calcium, copper, potassium, and phosphorus (Bainard et al. 2020b). These factors point to the importance of feed tests to ensure livestock are receiving the appropriate balance of nutrients when grazing diverse annual crops. There are also concerns related to potential toxicity of grazing brassicas that are high in nitrates and sulfur (e.g., Barry 2013).

While there appears to be several benefits to grazing diverse annual crops, there are still many unknowns and some possible limitations linked to productivity and quality under difficult growing conditions. Continued research into forage potential across different production practices and geographic regions will be important.

#### Literature Cited:

Bainard JD, M Serajchi, L Bainard, MP Schellenberg, EG Lamb. 2020a. Impact of Diverse Annual Forage Mixtures on Weed Control in a Semiarid Environment. Frontiers in Sustainable Food Systems https://doi.org/10.3389/fsufs.2020.00092

Bainard LD, B Evans, E Malis, T Yang, LD Bainard. 2020b. Influence of Annual Plant Diversity on Forage Productivity and Nutrition, Soil Chemistry, and Soil Microbial Communities Frontiers in Sustainable Food Systems https://doi.org/10.3389/fsufs.2020.00092

Barry TN. 2013. The feeding value of forage brassica plants for grazing ruminant livestock. Animal Feed Science and Technology 181(1–4): 15-25.

Lithourgidis AS, CA Dordas, CA Damalas, DN Vlachostergios. 2011. Annual intercrops: an alternative pathway for sustainable agriculture. Australian Journal of Crop Science 5(4): 396-410.

Szumigalski AR, and RC Van Acker. 2006. The agronomic value of annual plant diversity in cropweed systems. Canadian Journal of Plant Science 86(3): 865-874. https://doi.org/10.4141/P05-074

# Mineral and Vitamin Supplementation to Dairy Cows Under Different Situations

Bill Weiss
Department of Animal Sciences
OARDC/The Ohio State University
Weiss.6@osu.edu

#### **Summary**

The new NASEM (2021) is an excellent starting point when formulating diets for dairy cows; however, adjustments to mineral and vitamin recommendations are often needed. In most situations and for most minerals and vitamins, nutritionists should formulate diets that exceed the needs of the average cow in the pen. Based on expected variation, a safety factor of about 20% (i.e., NASEM x 1.2) is probably adequate in normal situations. Vitamin A supplementation may need to be increased when high starch diets or high straw diets are fed and reduced when cows are grazing good pasture. Vitamin D supplementation can be reduced when cows are exposed to direct, intense sunlight. Increased supply of magnesium, copper, and perhaps manganese and zinc is often warranted because of reduced absorption caused by potassium (for magnesium) and sulfur (for trace minerals) antagonism. Feeding hydroxy or organic trace minerals may not reduce the amount of mineral needed but may have positive effects on health and production when they replace sulfate trace minerals.

#### Introduction

Cows need to absorb adequate, but not excessive amounts of minerals and vitamins to maintain good health and obtain high milk yields. The amounts of minerals and vitamins a cow needs to consume depends on the requirement for absorbed mineral and vitamins, the absorbability of the nutrients, and environmental conditions. Absorbed requirements depend on body weight, dry matter intake (DMI), milk yield, stage of gestation, and rate of growth. Absorbed requirements are generally positively correlated with all those factors. Some of these baseline requirements are affected by environmental conditions such as heat stress but these effects are poorly quantified. The amount of mineral or vitamin needed in the diet is calculated as the absorbed requirement divided by the absorption coefficient (AC). Absorption depends on the source of mineral (i.e., the specific feedstuff), concentrations of other minerals and vitamins in the diet and water, and concentrations of some macronutrients such as fiber and starch. Baseline requirements and AC for minerals and vitamins are from NASEM (2021); this paper will discuss situational adjustments to those requirements and AC.

#### **Vitamins**

We have little quantitative data on vitamins requirements, and essentially no information on vitamin absorption by cows. In addition, nutritionists rarely, if ever obtain assayed concentrations of vitamins in feedstuff. Therefore, NASEM recommendations are based on supplemental vitamins without considering absorption. For most situations, those recommendations are adequate, but nutritionists should include a safety factor to account for

normal biological variation in supply and requirements. Based on data from laboratory animals and humans, a safety factor of about 20% should be adequate. In other words, for most situations feeding 1.2X NASEM recommendations should be adequate.

Supplementation of vitamins A, D and E, should be modified in certain situations. NASEM vitamin recommendations assume typical supplies of basal vitamins. Diets that contain more fresh forage provide more B-carotene (vitamin A precursor) and more tocopherol (vitamin E) than typical should require less supplemental vitamins A and E. Conversely, diets that contain substantial amounts of bleached hay or straw provide less basal vitamins A and E. Supplemental vitamin A is destroyed in the rumen and destruction increases as dietary starch increases. NASEM assumes cows are fed diets with about 25% starch; therefore, additional supplementation is needed with higher starch diets. Lastly, cows with direct exposure to sunshine can synthesize substantial amounts of vitamin D and will require less supplementation.

Suggested adjustments to NASEM vitamins A, D, and E

- 30-40% of diet DM is pasture: -500 IU/d of vitamin E (this adjustment is included in NASEM software)
- 30 to 40% of diet DM is pasture: -50,000 IU/d of vitamin A
- Majority of forage is hay rather than silage: +5000 IU of vitamin A/d
- ~8 lbs of straw in prefresh diet: +15,000 IU of vitamin A/d
- Diet starch >25% of DM: +2000 IU of vitamin A/d per percentage unit of starch >25%
- Cows have 2 to 3 hours of exposure to direct intense sunlight: -20,000 IU of vitamin
   D/day (intense sunlight probably only occurs in late spring, summer, and early fall)

Choline (rumen-protected), biotin, niacin, and B-carotene are vitamins that can have positive effects on cow health and milk production, but NASEM did not establish recommendations. The committee considered those effects as responses rather than requirements. In other words, similar responses can occur without supplementing those vitamins. Rumen protected choline (~15 g/d of choline) usually increases milk yield in early lactation and can reduce ketosis and fatty liver. Biotin (20 mg/d) usually increases milk production and improves hoof health. Niacin, at high enough inclusion rates (12 g/d) has increased milk yields but responses are not as consistent as those from biotin and choline. B-carotene can improve immune function and reproductive efficiency.

#### **Minerals**

NASEM mineral requirements are for total, not supplemental, mineral intake which means for some minerals no supplementation is needed. As discussed above, for most minerals, total dietary supply should be about 1.2 times NASEM requirements. Iodine (excess I in milk), phosphorus (normal recycling and high confidence in the equations), selenium (US FDA regulation) and sulfur (modest excess can cause significant problems) should be fed at NASEM recommendations without a safety factor. In addition to the standard safety factor, increased supplementation may be needed for some minerals in some situations. If the mineral is not discussed, NASEM recommendations with safety factor should be adequate.

#### Magnesium

Magnesium absorption is affected by numerous commonly occurring conditions which justify adjusting its safety factor.

- Dietary K has a strong negative effect on Mg absorption and the antagonism starts at very low K concentrations (Figure 1). The NASEM software includes an adjustment of the AC for Mg as dietary K changes but because of the prediction error associated with the equations, users may want to consider reducing the AC more than the model does.
- Long chain fatty acids can reduce Mg absorption. Diets with supplemental fat may need 10 to 20% more Mg.
- Availability of Mg from magnesium oxide is variable depending on particle size and manufacturing conditions. Some MgO may provide almost no absorbable Mg. We lack laboratory methods that accurately quantify these differences. Nutritionists should buy MgO from known, reputable sources. This uncertainty increases the risk of inadequate dietary Mg. The average AC for Mg from MgO is 0.23, you may want to reduce this by about 30% (i.e., AC of about 0.16)
- Feeding monensin at standard rates increases absorption of Mg by about 30% when MgO is the source of supplemental Mg (Tebbe et al., 2018). If you are feeding monensin, reducing the AC for MgO is probably not necessary (i.e., monensin is the safety factor)

#### **Electrolytes and DCAD**

Absorption of the electrolytes (sodium, potassium, chloride is high and consistent; therefore, uncertainty in supply does not justify increasing dietary concentrations above NASEM plus a 20% safety factor. However, cows often respond positively to feeding more than requirement. The NASEM committee considered these as responses rather than requirements (e.g., cows might produce more milk fat with increasing potassium, but increased potassium is not required to obtain additional milk fat). The response to additional electrolytes can be caused by the specific element or more often by a change in DCAD caused by changing concentrations of the electrolytes or sulfur. Feeding excess potassium or sodium increases water intake and urine output. This may be beneficial in hot conditions by increasing heat loss from the cow. If requirements for sodium, potassium, sulfur, and chloride are met, the diet will have a DCAD of about 175 mEq/kg. Increasing dietary DCAD by feeding additional potassium or sodium without increased chloride or sulfur can increase DM intake, milk yield, fat yield, fiber digestibility and dry matter digestibility. Maximum responses typically occur around 380 mEq/kg but the optimal concentration depends on cost of the supplements and the price of milk components and feed. Decreasing DCAD by feeding excess chloride or sulfur reduces risk of hypocalcemia after calving. Improved calcium metabolism usually requires DCAD less than about -100 mEq/kg.

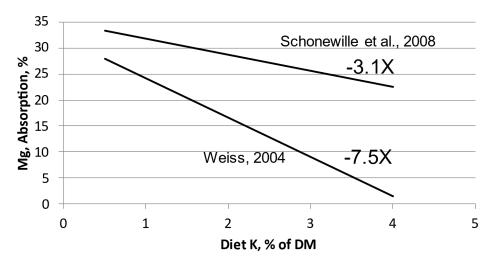


Figure 1. Relationship between dietary potassium concentration and magnesium absorption by dairy cows from two different studies. Slopes are shown in the figure. Data from (Schonewille et al., 2008) was derived mainly from dry cows consuming an average of 0.45% Mg and data from (Weiss, 2004) was derived mainly from lactating cows fed diets that averaged 0.25% Mg. The main reason potassium has a less negative effect on Mg absorption in the Schonewille et al study was because feeding extra Mg reduces the antagonism.

#### **Trace minerals**

Absorption of many trace minerals can be reduced by many 'real world' conditions. Excess dietary sulfur (including sulfur in drinking water) reduces the absorption of copper, manganese, and zinc although the effects have not been quantified with great accuracy. For copper, and perhaps the other metals, absorption can be reduced with as little as 0.3% sulfur in the diet. Based on limited data, absorption of copper in diets with about 0.2 to 0.25% sulfur averages 5% but with 0.3% sulfur it may drop to 4.2% and with 0.4% S it may be only 3.5%. Molybdenum interacts with sulfur to further reduce copper absorption. For example, with 4 mg/kg Mo, copper absorption may decrease those AC by another 25% (NASEM, 2021). Although data are lacking with dairy cows, apparent absorption of Mn was reduced about 65% when growing steers were fed diets with 0.7% S compared with feeding a 0.24% S diet (Pogge et al., 2014). A similar reduction in apparent zinc absorption was found. This suggests that antagonism of Mn and Zn by S may follow a similar pattern as that for Cu.

Source of trace mineral can affect absorption but because measuring true absorption of trace minerals is exceedingly difficult almost no direct absorption data are available. Based on relative availability assays, high quality organic copper and hydroxy copper chloride are more available than copper sulfate especially in the presence of antagonists. Selenium from selenized yeast is probably about 50% more absorbable than selenium from selenite but because of the way selenium methionine is used in the body, 'bioactivity' is probably only about 20% higher (Weiss, 2003). Some forms of organic zinc are more absorbable in a cell culture model than is zinc from zinc sulfate (Sauer et al., 2017) but whether this occurs in cows is not known. Apparent absorption of Mn by dairy cows did not differ between manganese sulfate and organic manganese (Weiss and Socha, 2005). Although data showing organic or hydroxy trace minerals are actually more absorbable than sulfate trace minerals is very limited, clinical

responses such as milk production, digestibility, immune function, and health are often positive when specialty trace minerals replace some of the sulfate minerals (sampling of papers: (Rabiee et al., 2010, Osorio et al., 2016, Faulkner and Weiss, 2017).

Some data suggests that at least some of the responses observed when specialty trace minerals are fed is not caused by improved absorption but rather by effects on the ruminal and intestinal microbiome ((Faulkner et al., 2017). If this is the case, then AC would not change when using specialty trace minerals, rather various positive responses would be expected when specialty trace minerals replaced sulfate minerals at similar dietary concentrations (i.e., NASEM x 1.2). Experiments quantifying actual, not relative, absorption are needed to confirm this.

Chromium is a required nutrient but similar to biotin and choline, the NASEM committee did not establish a requirement. This was because a clinical chromium deficiency has never been reported (indicating basal chromium supply may be adequate). Although no deficiencies have been reported, supplementing chromium (at about 0.5 mg/kg diet) often increases milk production in early lactation.

#### References

- Faulkner, M. J. and W. P. Weiss. 2017. Effect of source of trace minerals in either forage- or by-product—based diets fed to dairy cows: 1. Production and macronutrient digestibility. J Dairy Sci 100:5358-5367.
- Faulkner, M. J., B. A. Wenner, L. M. Solden, and W. P. Weiss. 2017. Source of supplemental dietary copper, zinc, and manganese affects fecal microbial relative abundance in lactating dairy cows. J Dairy Sci 100:1037-1044.
- NASEM. 2021. Nutrient Requirements of Dairy Cattle, 8th rev. ed. National Acad Press, Washington DC. Osorio, J. S., E. Trevisi, C. Li, J. K. Drackley, M. T. Socha, and J. J. Loor. 2016. Supplementing Zn, Mn, and Cu from amino acid complexes and Co from cobalt glucoheptonate during the peripartal period benefits postpartal cow performance and blood neutrophil function. J Dairy Sci 99:1868-1883.
- Pogge, D. J., M. E. Drewnoski, and S. L. Hansen. 2014. High dietary sulfur decreases the retention of copper, manganese, and zinc in steers. J Anim Sci 92:2182-2191.
- Rabiee, A. R., I. J. Lean, M. A. Stevenson, and M. T. Socha. 2010. Effects of feeding organic trace minerals on milk production and reproductive performance in lactating dairy cows: A meta-analysis. J. Dairy Sci. 93:4239-4251.
- Sauer, A. K., S. Pfaender, S. Hagmeyer, L. Tarana, A.-K. Mattes, F. Briel, S. Küry, T. M. Boeckers, and A. M. Grabrucker. 2017. Characterization of zinc amino acid complexes for zinc delivery in vitro using Caco-2 cells and enterocytes from hiPSC. BioMetals 30:643-661.
- Schonewille, J. T., H. Everts, S. Jittakhot, and A. C. Beynen. 2008. Quantitative Prediction of Magnesium Absorption in Dairy Cows. J. Dairy Sci. 91(1):271-278.
- Tebbe, A. W., D. J. Wyatt, and W. P. Weiss. 2018. Effects of magnesium source and monensin on nutrient digestibility and mineral balance in lactating dairy cows. J Dairy Sci 101:1152-1163.
- Weiss, W. P. 2003. Selenium nutrition of dairy cows: comparing responses to organic and inorganic selenium forms. Pages 333-343 in Proc. Nutritional Biotechnology in the Feed and Food Industries. Alltech, Inc., Lexington, KY.
- Weiss, W. P. 2004. Macromineral digestion by lactating dairy cows: Factors affecting digestibility of magnesium. J. Dairy Sci. 87:2167-2171.
- Weiss, W. P. and M. T. Socha. 2005. Dietary manganese for dry and lactating Holstein cows. J. Dairy Sci. 88:2517-2523.

### NASEM 2021 – Carbohydrates

Mary Beth Hall, PhD U.S. Dairy Forage Research Center, USDA-ARS 1925 Linden Drive, Madison, WI 53706 marybeth.hall@usda.gov

#### Introduction

Carbohydrates are key players in dairy cattle diets: they make up 70 to 80% of the diet dry matter and provide the nutrition and chewable fiber that keep cows healthy and productive. The 2021 Nutrient Requirements of Dairy Cattle, 8<sup>th</sup> edition from the National Academies of Sciences, Engineering and Medicine (NASEM, 2021) keeps much of the familiar landscape of carbohydrates we've been using, but provides some new additions, revisions, and applications (Figure 1).

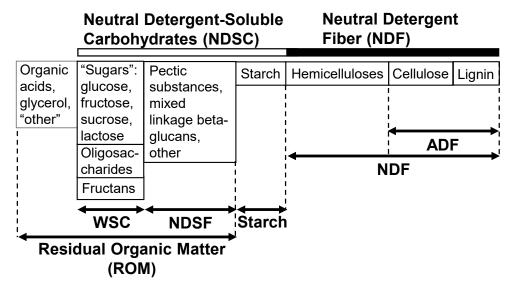


Figure 1. Carbohydrate fractions in feeds. ADF = acid detergent fiber, NDSF = neutral detergent-soluble fiber, WSC = water-soluble carbohydrates.

#### **Carbohydrate Fractions**

The neutral detergent soluble carbohydrates fraction (NDSC) replaces nonfiber carbohydrates (NFC) as a more accurate description of this fraction: these are the carbohydrates soluble in neutral detergent. It includes the more readily fermentable carbohydrates: starch, water-soluble carbohydrates (WSC), and neutral detergent-soluble fiber. Starch consists of chains of glucose that can be digested by microbes or the animal. The WSC include sugars, fructans from cool season grasses, and oligosaccharides (short chains of sugars). Total sugars as invert can be used as the WSC value for molasses. Soluble fiber is a kind of fiber not in NDF. It includes pectins and other polysaccharides extracted by neutral detergent and not digestible by mammalian enzymes. Neither WSC nor soluble fiber as such are used to calculate nutrient

supply in NASEM 2021 because there was not enough published research to define their specific impact. Of the NDSC, starch receives the most focus. Not surprising since most NDSC research has centered on starch. Why? Besides accounting for a substantial portion of many dairy cattle diets, we have good, relatively easy methods to measure starch in feeds, diets, and feces to assess its digestibility, something we lack for soluble fiber and WSC. Recommended methods of analysis to get the inputs needed for the NASEM 2021 equations and model are listed in the Feed Analysis chapter.

The 2001 NRC recommendations relied on NFC calculated by-difference to describe the pool of readily available carbohydrates. NASEM 2021 omitted NFC, made starch its own fraction, and came up with a new by-difference fraction: residual organic matter (ROM) that is used in energy supply calculations. ROM covers carbohydrates not accounted for by starch and NDF. It is calculated as a percentage of dry matter as: 100 - ash - NDF - starch - (fatty acids/fatty acid factor) – (crude protein – 0.64 x supplemental nonprotein nitrogen). The fatty acid factor = 1 if the source is fatty acids or fatty acid soaps, or 1.06 for all other feeds. The ROM contains WSC, soluble fiber, fermentation acids like those in silage, glycerol, and other feed components not measured in the main nutrient fractions. Similar to NFC, ROM is estimated to be 96% truly digestible. Nutritionists: Before you get bothered that a variety of fractions we can measure were combined into ROM, remember two things: 1) using ROM and starch reduces the size of the "nutritional black box" that NFC was, and 2) the NASEM committee didn't find enough published data to describe the impact of the ROM carbohydrates. The only way to find out whether parsing NDSC further will improve how well we can predict nutrient supply and cow performance is to do the research to explore those questions.

Neutral detergent fiber (NDF), acid detergent fiber (ADF), and sulfuric acid lignin – the "insoluble" fiber fractions - stay as they have been in previous editions. The NDF is analyzed for using heat-stable alpha-amylase (the "a" in aNDF or aNDFom) and sodium sulfite to remove starch and protein. But, there can be 2 NDF options to choose from on feed analyses: which should be used with NASEM 2021? On feed analyses, you may see "NDF" and "NDFom". The "om" stands for "organic matter" or "ash-free" meaning the ash in NDF was subtracted out of NDFom, but not from NDF. The NASEM 2021 nutritional model uses NDF values that include ash that was not extracted with neutral detergent because that was the version of NDF used in the research studies used to develop the recommendations. Typically, ash is a minor part of NDF. However, if a feed sample and NDF show heavy ash/soil contamination, resampled feed should be analyzed or NDFom should be used to avoid counting the excess ash as carbohydrate.

#### **Application of Carbohydrate Values**

What you find in NASEM 2021 is that the Carbohydrate chapter gives guidelines related to feeding carbohydrates that maintain desired rumen function, and the Energy and Protein chapters use various carbohydrate fractions in equations for predicting nutrient supply.

NASEM 2021 took the approach of allowing energy supply values from starch and NDF to be affected by the diets they're in. This is handled by calculating energy predictions from a base or starting point total tract digestibility (TTD) of a nutrient in a feed. For starch, the base TTD for a feed is assigned in the feed library, ranging from 96% for finely ground high moisture corn to 77% for coarsely ground dry corn. If you have information on starch digestibility specific to your feeds, it can be entered into the feed analyses in the NASEM 2021 model. For NDF, the base TTD of a feed is calculated from either a laboratory-measured 48 hour NDF in vitro fermentation digestibility, or from NDF and lignin analyses. Next, the diet TTD of that nutrient is modified based on the dietary factors that affect it. Both starch and NDF TTD, and so the energy available from them, decrease with increasing dry matter intake as a percentage of body weight. The NDF TTD also decreases as dietary starch concentration increases. NASEM 2021 mentioned that a having a factor for starch fermentability as it affects NDF digestibility would have been useful, but there was not enough data to define it. Lastly for the carbohydrate contributions to energy, TTD of ROM is estimated to be 96% and is unaffected by other factors.

Microbial protein production, an important source of protein to the cow, is calculated from predictions of how much starch and NDF are fermented in the rumen. A number of factors affect these predictions. The amounts of fermented starch and NDF are both affected by dry matter intake. The fermented NDF value is also affected by dietary concentrations of crude protein, wet forage, and ADF/NDF. The fermented starch value is influenced by dietary concentrations of forage NDF, wet forage, and starch (yes, starch). The forage NDF likely affects passage and how long feeds are retained in the rumen to be fermented. Crude protein may relate to meeting the needs of the fiber digesters, whereas starch concentration may relate to maintaining a population of microbes that ferment starch.

Physical form and NDF are crucial to maintaining proper rumen function and health. The NDF in forages is particularly important. Its typically larger particle size encourages rumination, and the slower fermentation and breakdown of NDF maintains that form and makes it available to be chewed for a longer time. It also affects the ruminal retention of feeds. Factors affecting a cow's fiber needs are complex and we do not have all the needed measurements to pinpoint precise relationships with rumen function. However, as a starting point for maintaining good rumen function, NASEM 2021 has a table for recommended dietary minimum total NDF and minimum forage NDF and maximum starch concentrations with forage NDF increasing to balance as starch increases and total NDF decreases. Additionally, recommended directional changes in forage NDF inclusion are intended to counterbalance diet, management, and behavioral effects that could reduce ruminal pH and disturb rumen function (Table 1).

Another approach in NASEM 2021 is physically adjusted NDF (paNDF) which works with the interaction of forage NDF, starch, particle sizes on the Penn State Particle Separator (PSPS), and other elements in the diet that affect ruminal pH. It integrates these to give recommendations for the proportion of the diet dry matter that should be on the PSPS 8 mm / 0.315 inch sieve. There was a free paNDF app (MUNCH for Dairy Cows) that was available from the App Store or

Google Play Store – but through some glitch it was taken down. We're working to get it back up and available. University of Nebraska extension bulletin G2316 gives more information on the app (<a href="https://extensionpubs.unl.edu/">https://extensionpubs.unl.edu/</a>). For now, we suggest not changing diet protein or cow bodyweight in the app (once the app is available again).

Table 1. Based on NASEM 2021 Figure 5-2

Suggested direction for adjusting for optimal forage NDF
17% <> 27%
←Higher dry matter intakes
←Added buffers in diet
Forage chop length – Finer
Higher starch% of diet→
Starch degradability – Higher
Bunk space is limited→
Slug feeding Yes→
Daily variation is high in diet and mixing

#### References

NRC. 2001. Nutrient Requirements of Dairy Cattle. 7th rev. ed. Natl. Acad. Sci., National Academy Press, Washington, DC.

NASEM. 2021. Nutrient Requirements of Dairy Cattle. 8th rev. ed. Natl. Acad. Sci., National Academy Press, Washington, DC.

### **Mineral Toxicoses Most Commonly Encountered in Cattle**

Jeffery O. Hall, D.V.M., Ph.D. Huvepharma, Inc. jeffery.hall@huvepharma.us

With over thirty years of veterinary diagnostic experience, numerous cases of mineral toxicosis have been encountered. Some mineral poisoning cases occur relatively infrequently, but others are more commonly encountered in cattle. During the previous 5 years, the most commonly encountered mineral toxicoses cases in cattle include sulfur and selenium, but cases involving copper and lead are also relatively common. This presentation will focus on these four most frequently encountered mineral toxicosis etiologies.

#### SULFUR<sup>1</sup>

In my career, sulfur poisoning has been the most commonly identified type of mineral toxicosis. During the previous five years, 6-14 cases per year were handled. Even though sulfur is a needed dietary mineral, excesses can be toxic. Sulfur is an essential dietary nutrient in ruminants that is recommended to be in the diet at no more than 0.4%. The sulfur is utilized by microbes for production of sulfur containing amino acids. But, excessive sulfur intake in the form of organic (sulfur containing proteins or amino acids) or inorganic sulfur (sulfates, etc.) can results in adverse health effects. Sulfur from either organic or inorganic sources is converted into sulfide by the ruminal microbes prior to being used for incorporation into sulfur containing amino acids and proteins. The adverse health effects of excessive sulfur can be broken down into two primary types, PEM (polioencephalomalasia) and alterations in trace mineral balance. The conversion to sulfide is responsible for the adverse neurologic health effects and most of the effects on mineral balance.

Systemic absorption of toxic amounts of sulfide is predominantly via respiratory absorption. This occurs via eructation, then inhalation of hydrogen sulfide gas. The ruminal microbial conversion of excess sulfur to hydrogen sulfide gas is a gradual adaptive process, which results in a delay from the start of excess sulfur in the diet to the onset of adverse clinical effects. This delay can be a couple of weeks.

Sulfide appears to affect neurologic tissues by either blocking cytochrome c oxidase or interfering with thiamine utilization. The most recent literature suggests that there is no thiamine or transketolase effect in sulfate poisoning, but some literature has shown a beneficial clinical effects when animals are treated with thiamine. This has occurred even when serum concentrations of thiamine are normal.

The outcome of the neurologic effects of excess sulfur is development of polioencephalomalasia (PEM). This necrosis of the gray matter of the brain results in clinical signs of lethargy, anorexia, facial muscle twitching, head pressing, recumbency, seizures, and death. In field cases the most common clinical presentation is "found dead".

Since both the organic and inorganic forms of sulfur can be metabolized to sulfide, one must account for both water and forage sources of sulfur. For instance, 0.35% dietary sulfur should not be a problem, but the addition of 500 mg/L sulfate in the water would push the total intake over the maximum recommended. Since ruminants consume 2-3X the weight of water per day as compared to daily dry matter intake, water can be a critical component of mineral intake.

Gross and histologic lesions are primarily in the brain, but ruminal changes can be observed. Gross pathologic lesions include a darkening of the rumen contents from precipitated metallic sulfide salts, swelling of the cerebral hemispheres, softening of the cerebral hemispheres, yellow discoloration of the cortical gray matter. Histological lesions include necrosis of the cortical gray matter and occasional areas of necrosis in the thalamus or midbrain.

The first component of treatment is removal of the source of high sulfate/sulfur. There is evidence that therapeutic doses of thiamine can be beneficial effects on the outcome for PEM cases, even though these animals can have normal plasma thiamine content. This would indicate that the sulfide is competitively interfering with the tissue thiamine utilization or thiamine may be causing the release of the oxidase bound sulfide in some way. Use of corticosteroids to decrease the cerebral edema has also been suggested. Other than the aforementioned therapies, good supportive care and dietary management is the only other treatment.

Excessive sulfate can also interfere with systemic mineral balance of copper, selenium, and zinc. One means by which this occurs is the precipitation of copper sulfide and zinc sulfide salts, rendering them non-bioavailable from the diet. High sulfur in the form of sulfate in the water and high dietary sulfur caused severe liver depletion of liver copper stores in as little as a few weeks, which indicates that the effects with copper are systemic as well as from the standpoint of bioavailability. In addition, sulfate can directly compete with selenium for digestive absorption sites, competitively inhibiting the bioavailability of selenium.

Clinical signs of copper and selenium deficiency are common with excessive sulfur/sulfate intake. These herds present with poor growth rates, poor immune function (high incidence of infectious disease), and poor reproductive function. In addition, white muscle disease from selenium deficiency can occur. Treatment of sulfur/sulfate induced mineral deficiency is by removal of the source and adequate supplementation. In cases where removal of the source is not an option, use of chelated mineral supplements can be beneficial.

Slow adaptation to increasing dietary sulfur/sulfate occurs. This is likely due to microbial adaptation that results in less sulfide being released and absorbed systemically. In addition, with adaptation, less adverse effects occur with respect to mineral balance, but deficiencies can still develop.

Diagnosis of sulfur toxicosis involves analysis of the diet, analysis of water sources, and histologic evaluation of brain tissue from animals that die. Histologic analysis of brain tissue can identify polio lesions, but there are other causes of this specific lesions, including lead poisoning, true clinical thiamine deficiency, and water deprivation/salt poisoning. Tissue sulfur

is not a good indicator of poisoning, as the body has relatively high natural tissue sulfur concentrations. Total dietary sulfur needs to be evaluated to include both water and feed.

#### SELENIUM<sup>2,3</sup>

Selenium is an essential trace element in cattle that is key in the function of several selenoproteins. These selenoproteins function in free radical clearance, reductases, deiodinases, and other key enzymes. The selenium containing enzymes are essential for normal immune function, reproductive function, biotransformation reactions, neurotransmitter turnover, and anticarcinogenic actions. But, as with many essential minerals, excesses can result in adverse effects. During the previous 5 years, 3-8 cases of selenium poisoning were investigated per year.

Selenium excess can be a result of excess intake of natural diets high in selenium that occur in certain geographic regions of North America, be a result of errors in diet inclusion of selenium supplements, or by excessive dosage of injectable products. In North America the easter slopes of the northern Rocky Mountains and the western areas of the north central great plains have areas of high selenium soils that can result in excessive forage selenium accumulation. Within the areas of high selenium soils, alkaline soils tend to hold selenium in a chemical form that is readily absorbed by plants, selenates. Cases where plant selenium content is over hundreds to thousands of parts per million have been investigated. Some "indicator" or "obligate accumulator" plants that have developed a requirement for high selenium can occur in these areas and can have several thousand PPM of selenium.

Selenium toxicoses cases can be acute, sub-acute, or chronic in nature, dependent on the exposure dose. Acute and sub-acute poisoning are associated with higher exposure rates. With very high exposures, clinical signs can start in less than 24 hours. Clinical presentation is generally associated with respiratory distress, a garlic smell to the breath, lethargy, anorexia, diarrhea, tachycardia, weakness, teeth grinding, and deaths. Tissue lesions include pulmonary edema, systemic congestions, and heart/skeletal muscle necrosis. Chronic selenium poisoning is generally associated with excessive selenium intake over longer periods of time. Clinical signs of chronic selenosis are of weight loss, emaciation, hair loss (bobtail disease), hoof growth abnormalities, lameness, reproductive failure, and deaths.

The mechanism of action for selenium poisoning in cattle is thought to be multifactorial. With acute poisoning, depletion of intermediate metabolic substrates and development of free radical damage are likely mechanisms. With chronic poisoning, those mechanisms may also play a role, but incorporation of seleno-amino acids, like selenomethionine or selenocysteine, in place of their respective sulfur containing amino acids may alter a broader array of proteins. The incorporation of selenium in place of sulfur in sulfur containing amino acids could result in loss of key disulfide bridges and structural integrity of key enzymes/proteins.

The amount of selenium that will result in either acute or chronic selenosis can be variable. Different chemical form of the selenium have different relative toxic potentials. In general, selenate is slightly more toxic than selenite. Similarly, different organic selenium compounds differ in toxic potential. It has been recommended that total dietary selenium of less than 5 ppm is safe. Some have suggested that this value is inappropriately too low, but this author has

been involved with dosing studies where 10 ppm total dietary selenium caused over 50% reproductive failure in sheep.

Some adaptation to higher dietary selenium can occur. This is predominantly a result of rumen microbial adaptations, which can convert some of the dietary selenium to elemental. Elemental selenium is insoluble which prevents it from being available to be absorbed systemically.

Post-mortem diagnosis of selenium poisoning can be accomplished by liver selenium analysis. Cases of acute and sub-acute selenium poisoning in cattle will generally have liver concentrations greater than 7 ppm on a wet weight basis. Chronic selenosis can start to occur at liver selenium concentrations of greater than 1.5 ppm, but most cases have concentrations greater than 2 ppm on a wet weight basis. Care must be taken in evaluation of liver selenium content, as recent use of an injectable product containing selenium can cause liver selenium concentrations to be higher than normal. It is common for liver selenium content to increase to concentrations of up to a non-toxic 3-4 ppm after an injection of a selenium containing product. This increase will gradually decrease over a period of up to 10-14 days. Without an appropriate history, an injected animal could be erroneously identified as one with chronic selenosis.

The only treatment for selenium poisoning is removal of the dietary over-exposure, supportive care and time. Animals will gradually eliminate the excessive accumulated tissue selenium.

#### Lead<sup>4</sup>

Lead poisoning cases in cattle have historically been relatively common, but case numbers have diminished over the years due to decreased lead use in many products. However, this author has investigated 2-7 cases per year of lead poisoning in cattle over the past 5 years. The cases involved ingestion of lead from batteries, ingestion of old lead-based paint materials, ingestion of lead caulking materials, ingestion of lead shot, and cases where the source was not identified. Almost half of the diagnosed lead poisoning cases were identified when very high liver lead concentrations were identified during routine liver mineral analyses. In one case, lead shot was identified in the rumen and omasum of dead yearlings. It was later found that silage being fed to the yearlings had lead shot in it. The source of the lead shot was a shooting range next to the corn field. Apparently, the shot got lodged in the corn stalks and was present when the corn field was chopped for silage.

Lead poisoning cases in cattle most commonly present as neurologic disease with ataxia, blindness, weakness, muscle tremors, and seizures or just as animals that are found dead. Lead can also be a reproductive and developmental toxin. Gross lesions of lead poisoning are minimal, but pieces of lead plates from batteries or lead shot can sometimes be identified in the rumen or omasum.

Metallic lead that gets caught in the rumen or omasum can slowly dissolve, allowing for some delay from the time of absorption to the onset of clinical disease. Lead material that reaches the abomasum will more rapidly dissolve in the much lower pH environment, allowing for a more rapid systemic absorption. Thus, when multiple animals are exposed from the same source, animals may have onset of clinical signs over a wide time frame.

Diagnosis of lead poisoning is based on high liver or whole blood lead content. Normal background liver lead content is less than 1 ppm, while normal whole blood lead content is less than 0.1 ppm. Liver lead concentrations greater than 5 ppm or whole blood lead greater than 0.35 ppm can be diagnostic of lead poisoning. Lead concentrations between normal and that which is diagnostic of lead poisoning indicate excessive exposure to lead. In some states, diagnosis of lead poisoning in a food producing animal is reportable disease. In several cases where lead poisoning was diagnosed, other animals in the herd had higher than normal whole blood lead, even though they were not showing clinical disease. These animals were deemed not to be suitable for sale/slaughter (quarantined) until blood lead concentrations returned to normal background values. Cases where systemic lead remained high for months have occurred, likely due to slow systemic absorption of lead that was retained in the digestive tract.

Treatment for lead poisoning is possible, but in most cases cost prohibitive. For particulate lead (plates, shot, etc.), rumenotomy to remove any remaining lead from the rumen and omasal folds can prevent further lead absorption. Use of chelating agents, such as EDTA, BAL, or Succimer, to aid in the removal of tissue lead can also help eliminate lead.

#### Copper<sup>5</sup>

Copper is an essential trace element for cattle. Copper toxicosis in cattle is not very common, as they are much more resistant to excess accumulation than sheep. In fact, copper deficiency is much more commonly encountered in cattle than toxicosis. However, higher than normal, but less than toxic, liver copper is routinely identified in some mature dairy cattle. Even though copper toxicosis is relatively uncommon in cattle, an increasing number of cases have presented over the past 5 years.

Prior to 2017, this author averaged less than one case per year of copper poisoning in cattle. However, during the past 5 years 3-5 cases per year have been investigated. Almost all of the cases have been in younger dairy calves, with most being calves sent to feedlots from calf raising operations. However, colleagues have discussed cases in heifer development operations.

Cases of copper poisoning in calves primarily are associated with hepatic accumulation followed some time later by liver failure and deaths. Copper poisoning in cattle generally presents as an acute development of liver failure, jaundice, weakness, anorexia, and deaths. The inciting cause of the sudden hepatic release of copper stores is generally unknown. Several cases investigated by this author involved 250 to 300+ pound calves that had been moved to a feed yard from a calf raising operation. Many of the calves were beef-dairy cross calves that were being pushed harder for growth. Almost all the cases occurred within the first 30 days after arrival.

In discussions with colleagues, several factors many be at play in the increasing occurrence of copper poisoning in these dairy calves. Many dairy cows are over-supplemented with copper, as indicated by frequent findings of higher than normal liver copper concentrations. Movement of copper to the fetus may give these calves higher liver copper at birth. In the past 10 years, many calf raising operations have increased the amount of milk replacer being fed to each calf per day, some by as much as double what was used historically. But, the trace mineral pack in the milk replacer has stayed the same, resulting in the calves getting double the amount of trace mineral supplementation as would have historically occurred. Many calf starter rations

contain high concentrations of trace minerals and young animals tend to have higher absorption capability for trace minerals than adult animals. And, many feedlots that are accustomed to feeding beef calves use higher trace mineral content in their starter rations to counteract potential deficiencies often encountered in typical beef calves. One or several of these factors may be at play in the increased occurrence of copper poisoning cases.

Diagnosis of copper poisoning cases involves analysis of the copper content in both liver and kidney. High liver values alone only prove that the liver has accumulated excess. High kidney copper values verify that excessive copper was dumped into systemic circulation. High liver copper with appropriate histologic pathology can also be suggestive of copper poisoning.

Treatment is limited to supportive care, as use of chelation therapy is cost prohibitive in most cases. Removal of excessive copper from the diet plus animal growth will gradually dilute the excessive liver copper stores in younger animals. In more mature animals, use of increased sulfur and molybdenum in the diet can aid in the reduction of liver copper, but how much and how long are difficult to answer.

Normal liver copper values in cattle tend to vary depending on the source of the information. This author uses the following for copper: Liver normal = 25 to 100 ppm on a wet weight basis (normal for early neonates is 65 to 150 ppm), Toxic concentrations tend to be greater than 250 ppm, but if significant systemic dump occurs then values may be lower; Normal kidney copper = 4-6 ppm on a wet weight basis, Toxic concentrations are greater than 10 ppm but most are much higher.

#### References

- <sup>1</sup> Hall JO. Sulfur. In: Veterinary Toxicology, 3rd edition. By R Gupta (ed). Elsevier Publishing. pp. 483-487. 2018.
- <sup>2</sup> Hall JO. Selenium. In: Veterinary Toxicology, 3rd edition. By R Gupta (ed). Elsevier Publishing. pp. 469-477. 2018.
- <sup>3</sup> Hall JO and Davis TZ. Selenium. In: Reproductive and Developmental Toxicology, 2<sup>nd</sup> Edition. By R Gupta (ed). Elsevier Publishing. 2017. pp. 595-605.
- <sup>4</sup> Thompson LJ. Lead. In: Veterinary Toxicology, 3rd edition. By R Gupta (ed). Elsevier Publishing. pp. 439-443. 2018.
- <sup>5</sup> Thompson LJ. Copper. In: Veterinary Toxicology, 3rd edition. By R Gupta (ed). Elsevier Publishing. pp. 425-427. 2018.

# 56th Annual Pacific Northwest **Animal Nutrition Conference**

January 16-17, 2023 Boise, Idaho

### Graduate Student Abstracts

#### **GRADUATE STUDENT POSTER ABSTRACTS**

000 - 000

#### Effects of the physical form of starter feed on intake, performance and health of young Holstein calves

Í.R.R. Castro, G.B.C. Leite, J.C.C. Chagas, G. Fields, A. Bartelheimer, A. Harder, D. Landin, and M.I. Marcondes.

#### Effect of iodine source on dairy cow colostrum production and growth and health of their calves

K.R. Johnston, D.C. Reyes, K.N. Klobucher, T.C. Stahl, P.S. Erickson, and A.F. Brito.

#### Botanical Composition and Quality of Beef Cattle Diets on a Burned and/or Unburned Rangeland

J.A. Kluth, N.G. Davis, S.A. Wyffels, C.B. Marlow, L.T. Vermeire, M.L. Van Emon, M.F. Nack, T.E. Sitz, T.G. Hamilton, J.J. Peterson, G.J. Collins, A.V. Rutledge, M.A. Whitehurst, and T. DelCurto.

### Impact of improving the heat detection method on farm dietary costs, production, and profitability in Washington and Florida dairies

G.B.C. Leite, and M.I. Marcondes.

#### Effects of wildfire smoke PM<sub>2.5</sub> exposure on health and performance of dairy heifer calves

A. Pace, P. Rezamand, and A.L. Skibiel.

#### Performance of grazing beef cattle supplemented with increasing energy levels in a tropical environment

Luiza de Nazaré Carneiro da Silva, Robert Emilio Mora-Luna, Daniel Henrique Sousa Tavares, Mirelle Magalhães Souza, José Neuman Miranda Neiva, Fabrícia Rocha Chaves Miotto, and M.I. Marcondes.

### Does transition milk and colostrum feedings provide benefits to dairy calves' gastrointestinal tract after feed restriction and fasting? Fecal immune and microbiota parameters assessment.

M. Tortadès, S. Martí, M. Devant, F. Fàbregas, and M. Terré.

#### Effects of weaning strategies on health, hematology, and productivity in Holstein dairy calves

A. Wolfe, P. Rezamand, B.C. Agustinho, D.E. Konetchy, and A.H. Laarman.

#### Effects of the physical form of starter feed on intake, performance and health of young **Holstein calves**

Ícaro R. R. Castro<sup>1,2</sup>, Giulia B. C. Leite², Juana C. C. Chagas³, Grace Fields², Andrew Bartelheimer², Alynn Harder², Dinorha Landin<sup>2</sup> and Marcos I. Marcondes<sup>2</sup>

Commercial starters are formulated containing coarsely rolled or ground grains, whole grains, protein, mineral, and vitamin supplements in their composition. The processing and physical form of starter feed (PFSF) can affect the palatability, acceptability, intake, and/or performance of dairy calves, which is critical to the rumen development and play a crucial role in easing the transition from pre-ruminant to mature ruminant state. Despite the relevance of this topic, the literature on the effects of different PFSF on calf performance is inconclusive. Thus, this study aimed to investigate the effects of the PFSF on feed intake, growth performance, and the health of dairy calves. Twenty-four female Holstein calves (5 days of age; 40.42 ± 3.86 of body weight; mean  $\pm$  sd) were used in a completely randomized block design (n = 12 calves per treatment). Individually housed calves were randomly assigned to one of the two dietary treatments, including (1) texturized feed starter (TFS; a mix of a protein pellet with whole kernel corn, and (2) pelleted feed starter (PFS; protein pellet). Both starters had the same ingredients and nutrient compositions but differed in their physical forms. All calves were offered the same milk replacer (MR; 26% to 28% CP and 16% to 24% lipid, 6 L/d of MR until 30 d of age, 4 L/d of MR from 31 to 60 d, 3 L/d of MR from 61 to 62 d, 2 L/d of MR from 63 to 64 d and 1 L/d of MR from 65 to 66 d of age). Calves were weaned on d 67. All calves had free access to drinking water and the starter throughout the experiment, and the feed and water intakes were measured daily. Animals were weighed weekly during the trial and at 30 days (97 d of age) and 90 days (157 d of age) after weaning for performance evaluation. Data were analyzed as a randomized block design in a repeated measurements scheme using the GLIMMIX procedure of SAS (SAS Institute Inc., Cary, NC). Initial body weight (iBW) was included as a covariate. For health variables, as they usually do not follow a normal distribution, we tested all distributions available in the GLIMMIX procedure of SAS and used the best distribution according to the Bayesian Information Criterion (BIC). No interactions between time and treatment were observed on all variables (P > 0.05). There was no effect of the physical form of the starter on starter (P > 0.05) and water (P > 0.05) intakes. PFSF did not affect body measurements of body weight (BW), average daily gain (ADG), and withers height (WH) (P > 0.05). Clinical signs of disease, such as ear position, and cough incidence were not affected by PFSF (P > 0.05). Whereas general attitude (P = 0.010), presence of eye discharge (P = 0.004), total respiratory score (P = 0.019), fecal score (P = 0.04), and nonrespiratory score (trend, P = 0.087) of animals receiving the texturized feed starter were superior to those fed the protein pellet. Overall, our results indicate that starter diets containing whole kernel corn as a texturizer cannot improve the intake and performance of young dairy calves compared with diets containing pelleted feed starter; however, we observed that the health of the group receiving the texturized feed starter was improved.

<sup>&</sup>lt;sup>1</sup>Department of Animal Science, Universidade Federal de Viçosa (UFV), Av. Peter Henry Rolfs, s/n - Campus Universitário, Viçosa, MG, 36570-900, Brazil.

<sup>&</sup>lt;sup>2</sup>Department of Animal Sciences, Washington State University, Pullman, WA, USA.

<sup>&</sup>lt;sup>3</sup>Swedish University of Agricultural Sciences, Umeå, Sweden.

#### Effect of iodine source on dairy cow colostrum production and growth and health of their calves

K.R. Johnston<sup>a</sup>, D.C. Reyes<sup>a</sup>, K.N. Klobucher<sup>a</sup>, T.C. Stahl<sup>a</sup>, P.S. Erickson<sup>a</sup>, and A.F. Brito<sup>a</sup>

<sup>a</sup>Department of Agriculture, Nutrition, and Food Systems, University of New Hampshire, Durham, New Hampshire, **United States** 

Limited research exists on how the high iodine (I) concentration of seaweed supplements affects prepartum cow colostrum production and development of their calves. The objectives of this study were: (1) evaluate the effects of incremental amounts of Ascophyllum nodosum (ASCO) meal supplementation to prepartum cows on colostrum production and the growth and blood metabolite concentrations of their offspring, and (2) compare ASCO meal versus a common I source [ethylenediamine dihydroiodide (EDDI)] on the same variables under objective 1. Forty Holstein cows were blocked by lactation number and expected calving date and assigned to 1 of 4 treatments 28 d prior to parturition: (1) EDDI supplemented to meet recommended I intake [0.5 mg I/kg of dry matter intake (CON)], CON plus 57 g/d of ASCO meal (LO), CON plus 113 g/d of ASCO meal (HI), and CON plus EDDI (124.8 mg/d) supplemented to match the amount of I provided by HI (EDDI). Within 1 h of calving, colostrum was harvested and weighed. Colostrum was analyzed for fat, protein, total solids, I, and IgG. Forty-one calves were blocked based on their dams' treatments. At birth, calves were weighed and fed 300 g lgG via colostrum replacer. At 24 h old, calves were offered 676 g dry matter daily of milk replacer (MR) (25% crude protein, 16% fat) until 49 d, where they were offered 338 g of MR. Free choice textured starter (28% CP) and water were offered ad libitum at 24 h of life until study completion. Blood samples were collected at 0 h and 24 h of age for IgG and thyroid hormone (TH), at d 14, 28, and 56 for TH, and weekly for beta-hydroxybutyrate (BHB) analyses. On d 5 of life, a xylose challenge was conducted by supplementing 0.5 g/kg body weight (BW) of D-xylose in the MR, with plasma samples taken over a 12 h period. Plasma samples were analyzed for xylose and glucose concentrations to serve as a proxy for intestinal absorption. Weekly skeletal and BW measurements were recorded. Fat concentration of colostrum was greater in HI than EDDI cows, and there was a tendency for fat concentration to decrease linearly with ASCO meal supplementation. Plasma concentration of weekly total T<sub>4</sub>, weekly BHB, and final BHB responded quadratically to ASCO meal supplementation, with the lowest concentration occurring with the LO treatment. There was a tendency for IgG apparent efficiency of absorption to be lower in EDDI versus HI calves. Overall, these data indicate that addition of ASCO meal to the dams' diet did not negatively impact calf growth and metabolism or colostrum production and composition. Additionally, ASCO may benefit colostrum composition and calf passive transfer when used as an I source compared to EDDI.

Keywords: immunoglobulin G, iodine, seaweed, colostrum production, calf development

Author Contact: Kayla Johnston: krjohns@ualberta.ca

#### Botanical Composition and Quality of Beef Cattle Diets on a Burned and/or Unburned Rangeland

JA Kluth, NG Davis, SA Wyffels, CB Marlow, LT Vermeire, ML Van Emon, MF Nack, TE Sitz, TG Hamilton, JJ Peterson, GJ Collins, AV Rutledge, MA Whitehurst, T DelCurto.

Montana State University

Current management paradigms suggest deferring grazing rangeland for two years after a wildfire to avoid additional stress on native plant species, but there is little to no research supporting these recommendations. This experiment was conducted within and adjacent to the burn area of an August 2020 wildfire to evaluate the differences in diet quality, botanical composition, and foraging efficiency of beef cattle on both burned and unburned rangeland. A randomized complete block design with a 2x2 factorial arrangement of treatments contrasting burn versus no burn and June versus September grazing periods was used. Pastures (4 ha) within the burn area were grazed in either June 2021 or deferred to September 2021. Grazing occurred for two days by 20 cow/calf pairs in June and 16 cow/calf pairs in September. Cattle diet composition and masticate samples were collected during 20-minute bite-count periods using six ruminally cannulated cows in each pasture prior to and after two-day grazing periods. Cannulated cows grazed a pasture adjacent to the burn area to compare diet quality and composition between burned and unburned rangeland. Nutrient value of key grass species in the study area were 30% higher in crude protein in the burned area as compared to the unburned area (12.47 vs. 8.53%, respectively) for the June grazing period. Difference of diet quality of standing vegetation was similar in the September grazing period (6.53% CP). Foraging behavior was influenced by the previous year's fire for the June grazing period. Beef cattle grams per minute ( $\bar{x} \pm SD$ ; 23.86 vs 7.22 ± 6.78), bites per minute (15.83 vs 9.4 ± 5.95), and grams per bite  $(1.53 \text{ vs } 1.02 \pm 0.85)$  were higher in unburned sites versus burned sites during the June grazing period. However, foraging behavior is similar in the burned and unburned areas during the September grazing period.

Keywords: diet quality, foraging behavior, grazing, season of use, wildfire

#### Impact of improving the heat detection method on farm dietary costs, production, and profitability in Washington and Florida dairies

Giulia B. C. Leite, Marcos I. Marcondes

Department of Animal Sciences, Washington State University, Pullman, WA, USA

The herd's reproductive performance vastly impacts the dairy's efficiency, not just because of the costs directly associated with the elected protocols but also indirectly on the performance, the permanence of the animals in the herd, and feed costs. Throughout the years, authors gathered data about the chosen heat detection method and economic and productive indexes of dairy farms, but they never linked those results with their consequences on diet costs. We aimed to evaluate the effects of the heat detection method (and reproduction efficiency) on the productivity and feed expenses of FL and WA dairy operations. A discrete Markov Chain model was used, simulating the 22-d period corresponding to one reproductive cycle, assessing the probabilistic performance of dairy cows and heifers over 10 years. We used the solver non-linear tool of Excel subjected to constraints of a minimum of 900 and a maximum of 1000 cows, and a minimum of 0% and a maximum of 40% voluntary culling rate of 3 or more lactation cows. The model was evolved over 10 years by changing (using the Solver function) the culling rate of 3 or more lactating cows and heifer calves sold at birth, aiming to reach a steady state herd and maximizing annual profit. The computed data of the last year was compiled to compare scenarios. Milk (class III) and other prices were obtained from the USDA (12/2022). The scenarios were created by computing differences in breeding detection using visual observation (VO), rump/tail markers (RM), or electronic detectors (ED, collars, or pedometers). We considered a 6% improvement in heat detection when changing from VO to RM and an improvement of 12% when changing from RM to ED. Lastly, we ran scenarios for two weather conditions (WA and FL states), totaling 6 scenarios for this study. The milk yield/cow increased by 0.8% and 2.3% using RM or ED, respectively, compared to VO in FL. In WA, this increase was 1.6 and 4.7%, respectively. Using RM in FL, we obtained a 0.6% increase in pregnant heifers and 15.3% pregnant cows, while using ED, these numbers go up to 3.3% and 34.9%. In WA, using RM, we observed 0.7% and 13.4% more pregnant heifers and cows, respectively; and using ED, 11.5% and 25.1%. The calving interval decreased from 13.1 mo using VO to 12.8 and 12.5 using RM and ED in FL. The same trend is observed for WA, showing a decrease from 12.7 to 12.4 (VO vs. RM) and down to 12.4 with ED and follows the same decreasing pattern in WA, with 12.7 (VO), 12.4 (RM), and 12.4 (ED) mo. When analyzing expenses with forage for diets, we observed that the more modern the heat identification method, the fewer expenses with forages, regardless of the region. Dairy operations in FL saved up to 2.9% from changing from VO to ED, and that number raised to 8.6% in WA. However, this relationship is inverse when we analyze expenses with concentrate, where FL producers increased their costs by 8.2% and WA producers increased by 22.6%. Overall, farms increased diet costs by 2.4% when changing from VO to RM; however, profit increased by 1.2%. In WA, farms increased diet costs by 3.5% when changing from VO to RM; however, profit increased by 3.3%. When switching from VO to ED, diet costs increased by 5.8 and 22% for WA and FL, respectively, but profit also improved by 8 and 38.7%, respectively. We concluded that the more specialized the heat detection system, the higher the dietary costs. Still, these costs are followed by increased milk production, improved reproduction, and higher profit.

#### Effects of wildfire smoke PM<sub>2.5</sub> exposure on health and performance of dairy heifer calves

Alexandra Pace, Pedram Rezamand, Amy L. Skibiel

University of Idaho

The western United States experiences wildfires that are increasing in severity and emit dangerous pollutants into the atmosphere. Fine particulate matter (PM<sub>2.5</sub>) in smoke is thought to be especially hazardous. Fine particulates can deposit in the lower airways and lungs, and enter circulation through the pulmonary system in humans, where it causes local and systemic inflammatory responses, resulting in disease. Yet, there is limited knowledge on how wildfire smoke PM<sub>2.5</sub> affects dairy calves. The present study aims to understand the effect of wildfire smoke exposure on dairy calf metabolic physiology and inflammatory response. Holstein heifer calves (n = 15) at the University of Idaho Dairy Center were followed from birth in July through weaning in September 2021, concurrent with the Pacific Northwest wildfire season. Blood samples were collected weekly to analyze hematology, blood metabolites [glucose,  $\beta$ hydroxybutyrate (BHB), nonesterified fatty acids (NEFA)], and acute phase proteins [haptoglobin (Hp), serum amyloid A (SAA)]. Health scores, respiratory rates (RR), heart rates (HR), and rectal temperatures (RT) were recorded weekly. Hourly PM<sub>2.5</sub> concentrations and meteorology data were recorded from a monitoring station in close proximity to the farm. AirNowTech Navigator with HYSPLIT modeling was used to map active wildfires, wind trajectory, and PM<sub>2.5</sub> to assess if the detected PM<sub>2.5</sub> was derived from wildfires. Statistical analysis was conducted using mixed models with calf as a random effect and fixed effects of average daily PM<sub>2.5</sub>, temperature-humidity index (THI), and their interactions, with separate models for lags (delayed responses) of up to seven days. Elevated PM<sub>2.5</sub> was concurrent with wildfires and ranged from 2.0  $\mu$ g/m<sup>3</sup> to 118.8  $\mu$ g/m<sup>3</sup>, while THI ranged from 48 to 73. Initially, there was a positive interacting effect of PM<sub>2.5</sub> and THI on glucose and NEFA on lag day 1 (P < 0.01, P = 0.05, respectively) and a negative interacting effect on BHB at lag day 1. However, these relationships were reversed with subsequent lag days. Higher PM<sub>2.5</sub> and THI together increased SAA on lag days 1 and 7 (both P < 0.01), and increased Hp on lag days 0, 2, 3, and 4 (all P < 0.03). Together, higher PM<sub>2.5</sub> and THI increased RR (P = 0.04), HR (P < 0.01), and RT (P = 0.04). 0.04) at lag day 0. Additionally, greater PM<sub>2.5</sub> and THI together elevated eosinophil count (P =0.02) on lag day 0, decreased total white blood cell (P = 0.01) and neutrophil counts (P < 0.01) on lag day 3, and decreased lymphocyte count on lag day 2 (P = 0.03). Cough score (P < 0.01) and eye score (P = 0.02) increased with greater PM<sub>2.5</sub> and THI after a 3-day lag. A number of these effects persisted at subsequent days. Our findings indicate that exposure of calves to wildfire smoke PM<sub>2.5</sub>, concomitant with higher THI, causes alterations in metabolism and the innate immune response, which could potentially compromise calf health and performance.

Keywords: air pollution, inflammation, calf health

Performance of grazing beef cattle supplemented with increasing energy levels in a tropical environment

Luiza de Nazaré Carneiro da Silva<sup>1,2</sup>, Robert Emilio Mora-Luna<sup>1</sup>, Daniel Henrique Sousa Tavares<sup>1</sup>, Mirelle Magalhães Souza<sup>1</sup>, José Neuman Miranda Neiva<sup>1</sup>, Fabrícia Rocha Chaves Miotto<sup>1</sup>, Marcos Inacio Marcondes<sup>2</sup>

The correct synchronization between nitrogen and carbohydrates favors the high efficiency of the ruminal microbiota, promoting improvements in digestibility and feed efficiency. Thus, we aimed to evaluate the effects of increasing energy supplementation on the performance of grazing beef cattle in a tropical pasture. The project was developed at the Tocantins Federal University, in the Araguaína - TO, in northern Brazil, between February and May 2021, totaling 73 days. Twenty-four zebuine calves with a mean age of 12 months and an initial average weight of 150 kg were used. The treatments consisted of the evaluation of supplements composed of whole grain corn and protein pellet with increasing energy supply while maintaining protein intake from the supplement: Treatment 1 - Concentrated feed composed of 100% of protein pellet and supply to 0.25% of body weight (BW); Treatment 2 - Concentrate composed of 50% protein pellet and 50% corn, at 0.50% BW; Treatment 3 - Concentrate composed of 34% protein pellet and 66% corn, at 0.75% BW; Treatment 4 – Concentrated diet in the proportion of 25% of protein pellet and 75% of corn, at 1% BW. The animals were run in a rotating stocking system with seven days of occupation and twenty-one days of rest. A completely randomized design was used with four treatments and two sets of paddocks (each replication represented by a paddock with three animals). The experimental grazing area comprised 4 hectares of pasture formed with Megathyrsus maximus grass cv. Mombasa. Stocking management was variable, in which the putand-take technique composed of testing and regulatory animals was used (Mott; Luke, 1952). Before entering and after the animals left the paddocks, pasture height and availability of total dry matter were measured. Supplements were provided once a day at 8:00 a.m. The animals were weighed at the beginning and end of the experiment and each 24-day cycle to monitor weight development. The data were submitted to normality (Shapiro-Wilks) and homoscedasticity (Levene) tests, and once the assumptions were met, they were submitted to analysis of variance following a completely randomized design. The treatment effects were evaluated with polynomial orthogonal contrast for linear and quadratic effects using a 5% probability level. Performance and DM intake increased linearly as energy supplementation increased (P < 0.050). The same behavior was observed for Final weight (P = 0.041), total weight gain (P = 0.021), average daily gain (P = 0.021), stocking density (animals/hectare; P< 0.001), and total weight gain per ha/day (P = 0.001). Based on these results, we speculate that the increase in the energy supplementation allowed better efficiency of the ruminal microbiota, minimizing nitrogen loss and consequently maximizing animal production. Furthermore, since carbohydrates are the primary energy source for microbial growth, possibly the greater energy intake promoted an increased supply of microbial protein to the small intestine and, consequently, animal performance. Therefore, providing supplementation with a higher proportion of corn can be helpful as a strategy to reduce the duration of the rearing phase.

<sup>&</sup>lt;sup>1</sup>Universidade Federal do Tocantins, Araguaína, Tocantins, Brazil.

<sup>&</sup>lt;sup>2</sup>Washington State University

Does transition milk and colostrum feedings provide benefits to dairy calves' gastrointestinal tract after feed restriction and fasting? Fecal immune and microbiota parameters assessment.

M.Tortadès, S. Martí, M. Devant, F. Fàbregas and M. Terré

<sup>1</sup>Department of Ruminant Production, IRTA (Institut de Recerca i Tecnologia Agroalimentàries), 08140 Caldes de Montbui, Spain

Bovine colostrum (BC) and Transition milk (TM) are rich in nutrients and bioactive molecules, which may help calves to recover digestive tract functionality from stressful periods such us marketing and transport. The objective of our study was to evaluate the digestive tract recovery of feeding either BC, TM, or milk replacer (MR) after an episode of feed restriction (FR) and fasting. In this conference, we will assess our objective through fecal biomarkers of immune response and microbiota. The FR and fasting was simulated by feeding 2L of an oral rehydration solution twice daily for 3 days and 19 h of fasting to 35 male calves (22 ± 4.8 days old) and then randomly assigned the calves to one of 5 treatments (n=7; day 1 of study): feeding either pooled BC during four (C4) or ten (C10) days, pooled TM during four (TM4) or ten (TM10) days, or MR for ten days (CTRL) at the rate of 720 g/d DM content in a total volume of 3L. After, all calves were fed the same feeding program, decreasing MR gradually from 3L twice daily to 2L once daily at 12.5% DM until weaning (d 42 of study). Concentrate feed, water, and straw were offered ad libitum. Feed, MR, and straw intake were recorded daily, and body weight on d -3, 1, 2, 5 and 11, and weekly afterwards. Volatile fatty acids, lactoferrin, IgA and microbiota (Firmicutes to Bacteroidetes ratio and Faecalis prausnitzii) were analyzed in feces from samples obtained on d 5 and 11 before the morning feeding. Data were analyzed with a mixed-effect model accounting for the random effects of animal and the fixed effect of treatment, time, and its interaction. Calf performance, intake, fecal lactoferrin concentrations, and microbiota quantification were similar among treatments throughout the study. Fecal IgA concentrations were greater (P < 0.05) in C10 than in CTRL, TM4, and TM10 calves, and in C4 and TM10 than in CTRL animals. Fecal propionate proportion was less abundant in C10 calves than in CTRL, TM4 and TM10 ones, while butyrate was greater in C4 and C10 calves than in TM4 and CTRL ones. Results showed that TM and BC provide gut immune protection and BC may promote butyrate-producing bacteria in calves after a FR and fasting episode.

#### Effects of weaning strategies on health, hematology, and productivity in Holstein dairy calves

A. Wolfe<sup>1,2</sup>, P. Rezamand<sup>1</sup>, B. C. Agustinho<sup>1</sup>, D. E. Konetchy<sup>1</sup>, A. H. Laarman<sup>2</sup>

University of Idaho<sup>1</sup>, University of Alberta<sup>2</sup>

Weaning strategies in dairy calves vary considerably, though the impact on animal health is unclear. This study examined the effects of calf weaning age (6 vs. 8 wk) and pace (abrupt vs. gradual) on health parameters in dairy calves. Holstein calves (n = 72), blocked by sex and birth weight, were randomly assigned to one of four treatments (n = 18 per): Early-Abrupt (EA), Early-Gradual (EG), Late-Abrupt (LA), and Late-Gradual (LG). Milk replacer (24% CP, 17% fat; up to 1200 g/d) was fed twice daily; water, calf starter (18% CP), and chopped alfalfa hay were fed ad libitum. Daily intakes of milk replacer, calf starter, and forage were recorded. Body weight, health measures, blood hematology, and fecal scores were obtained prior to, and after weaning. Calves were orally bolused with a rumen pH logger for the last three days of the weaning transition and rumen pH was measured continuously. Calves also had fresh blood analyzed using an HM5 hematology unit during weaning to determine immune function. Data was analyzed with age and pace as fixed effects. Age at weaning increased respiration (P = 0.02), while gradual weaning groups had lower respiration rate (P = 0.01). Heartrate was lower in gradual than in abrupt groups (P = 0.01). Fecal score tended to increase in late-weaned groups (P = 0.06) and gradually-weaned groups (P = 0.04). No difference was detected in body core temperature by age or pace. During the weaning transition, average daily gain was lower in LA than EA (0.62 vs. 0.11 kg/d, P < 0.01) and gradually-weaned groups had increased ADG (0.65 kg/d, P = 0.02). Change in grain intake, but not forage intake, was greater in gradually-weaned groups (P < 0.01). Mean rumen pH tended to increase from EG to LG (7.65 vs. 8.84, P = 0.1) and from LA to LG (7.89 vs. 8.84, P = 0.1). No difference was detected among treatments in red or white blood cell counts, and hemoglobin. Procalcitonin tended to be different depending on the age x pace interaction with the LA group having the highest percentage of procalcitonin (0.18%, P = 0.07). Blood hematocrit increased in abruptly-weaned groups (41.3 %, P = 0.01). Overall, calf health is affected by both age and pace of weaning, though the health parameters impacted by age and pace differ.

**Keywords:** calf health, weaning pace, weaning age

Author Contact: arwolfe@ualberta.ca

## **Ecological Costs of the Status Quo on Grazing Lands in the Intermountain West**

Barry L Perryman, Brad Schultz, and Bob Alverts University of Nevada-Reno

perryman@unr.edu 775-784-6644

Perryman et al. (2003) coined the phrase pristine-management-paradigm to describe the widely held management paradigm that ecological systems are static entities that can be held in a static condition if they are protected from burning, grazing, and other disturbances. The authors argued it was impossible to achieve societal objectives today based on returning landscape conditions to those perceived to exist prior to the 20<sup>th</sup> century and European settlement of the Intermountain West. Processes that created the landscape conditions of 1800 A.D. or any other previous time period have changed or been altered, making their replication impossible. For example: Little Ice Age weather conditions have ended; uncontrolled grazing by wild ungulates presumably influenced by codependent predators is no longer possible or desirable; widespread burning by Native Americans is no longer practiced; and annual grasses have colonized many sagebrush and salt desert shrub communities, permanently altering plant community compositions. Instead, objectives for ecosystem management should focus upon specific measurable goals that society has determined are valuable under current ecological conditions (e.g., soil stability, biodiversity, wildlife habitat, forage production, etc.). Today's landscapes are not those described by Smith, Ogden, and Simpson (Dale 1918, Cline 1974, Petersen 2008). With over 400,000 km<sup>2</sup> colonized by cheatgrass (Bromus tectorum L.) and other annual grasses (Mack 1981), It is time to declare: The pristine-management-paradigm has failed. Continued, wholesale application of this concept is misguided, and managing for this status quo has unacceptable consequences.

Management of the status quo includes the exclusive use of perennial grass grazing systems on ranges where cheatgrass and other invasive annual species are already entrenched, AUM reductions since the early 1980s, and more fire frequency and extent. Although a healthy, resilient perennial grass understory is likely the single most important long-term assurance against invasive annual grass dominance, rangeland ecologists and managers have long applied science-based management practices that exclude consideration of the biology, ecology, and probable management effects these perennial grass grazing systems would have on the non-native annual grass component of modern landscapes. For instance, the two major grazing systems employed in the Great Basin are deferred-rotation and rest-rotation. Both focus on meeting the physiological needs of grazed perennial grasses (Sampson 1913; 1951), but their implementation throughout the region failed to address how annual grasses would respond. Authorized grazing of animal unit months (AUM) on public lands in the Great Basin focuses on allotment carrying capacities provided by only native perennial species (CFR 4110.2-2 Specifying grazing preference). Non-native annual grasses generally are not recognized, authorized, allocated, or normally considered in the development of district wide or allotment management plans. In fact, almost all management planning efforts and implementations are designed to manage perennial grass or palatable shrub species. The allocation of forage derived from annual grasses requires a separate Record of Decision based on an Environmental Assessment (CFR 4130.6–2 Nonrenewable grazing permits and leases) and is seldom granted. Grazing fuel breaks have received considerable attention for several decades, exclusively for reducing fuel and fire risks in and around annual grass-dominated plant communities. At best, this management tool, especially when applied as a stand-alone action, is only a stopgap measure to postpone the fire effects of annual grasses near areas still dominated by desired native species. All the while, annual grasses have become the ecologically dominant life form on upwards of 20,000 km² in the Great Basin (Young and Clements 2009).

Over the past decade or so, a related movement toward an ecologically based weed management approach has spawned the development of potential new tools for the management of invasive annual grasses. Scientists are currently developing delivery methods for newly identified biological control agents. Undoubtedly, these tools will find useful and appropriate applications for yet undetermined situations and scales. The precise combination of chemical fallow and seeding with both native and non-native, deep-rooted perennial grasses and half-shrubs like forage kochia (Bassia prostrata L.) has provided success on many ecological sites and topographic settings, but only for a relatively small percentage of the entire affected area (Young and Clements 2009). Likewise, grazing cheatgrass in the fall and early winter months, when perennial grasses are dormant, has demonstrated that managed livestock grazing can reduce carryover fuels going into the next year's fire season, while simultaneously reducing the ability of cheatgrass to dominate areas with a remnant perennial grass component (Schmelzer et al. 2014, Perryman et al. 2020). Managing cheatgrass with dormant season grazing has been successful on demonstration and research projects at a scale of thousands of acres in southeastern Oregon, on winter dominated precipitation sites (W. Dragt, B. Wilber, and S. Davies, personal communication, August, 2017; Davies et al. 2021).

Given the advances and successes in the management tools available, the rangeland ecology and management community must recognize the requirement that annual invasive grasses must be managed as a permanent component of the Great Basin and Intermountain West. For the past 50 years, perhaps longer, most of our collective management objectives, goals, and practices have focused on only the perennial grass component, or toward palatable shrubs in the case of salt desert shrub communities. Rest-rotation and deferred rotation grazing systems (and their various combinations) focus management on the perennial grass component of the plant community while ignoring the annual grasses. Both grazing systems actually favor the proliferation and dominance of annual invasive grasses, especially on warmer and drier sites (Chambers et al. 2016), by essentially maximizing the standing dead biomass left at the end of the traditional grazing season (Trowbridge et al. 2013, Schmelzer et al. 2014). The antigrazing sentiment (Beschta et al. 2013) that led to a general reduction of annual and temporary grazing authorizations over the past several decades has also played a significant role in annual grass proliferation by providing an increase in safe sites for annual grass establishment, as well as creating larger, more contiguous fuel loads. Through our management activities that foster standing dead litter, we have inadvertently exacerbated invasive annual grass dominance in the Great Basin and Intermountain West. Most standing litter eventually becomes surface litter, creating the "safe site" for the germination of seed from annual grasses. Fall grazing of cheatgrass also directly controls seed bank volumes, the higher the grazing intensity, the lower the volume (Perryman et al. 2020). Research-based science has been applied toward the

management of perennial grasses on many landscapes (Launchbaugh et al. 2008), but not toward the ecologically dominant annual grasses that often occur with remnant populations of native perennial species.

The first step for dealing with this issue is recognition of the almost ubiquitous presence of invasive annual grasses across the Intermountain West, particularly at lower and drier elevations. Cheatgrass, medusahead (*Taeniatherum caput-medusae* (L.) Nevski), and North Africa grass (*Ventenata dubia* [Leers] Coss.; a relative newcomer) are here to stay. Not only are annual grasses present, they have become one of, if not the primary driver of the ecological changes occurring in many lower elevation big sagebrush and salt desert shrub communities. It is time that scientists, managers, and policy makers begin to develop and implement research, planning objectives, policies, and management actions that allow and provide for the active landscape-scale management of annual grasses, instead of continuously lamenting of being their victim. We must admit that many of the shrub dominated communities in the Great Basin and Intermountain West now have diminished perennial grass understories, and have become mixed-communities of annual and perennial grasses. They should be recognized and managed first and foremost as annual grasslands, just as the California annual grasslands have been recognized for decades, despite many having some perennial grasses in the plant community.

Current management paradigm practices were often implemented for rational reasons (improve the perennial herbaceous plant community) but failed to fully understand and/or include the ecology of the invasive annual component. The result is an unacceptable large-scale ecological situation for almost all users of sagebrush and salt desert rangelands. For landscapes where annual grasses are an ecologically dominant lifeform, a step in the right direction would be to address both the annual and perennial grass components simultaneously, with all the necessary management flexibility and situationally available tools (Svejcar et al. 2008). This approach requires the recognition and management of mixed annual-perennial grass understories for what they are, but also for what we want them to be in the future rather than what they may have been in the past. The ecological costs are too high to ignore.

#### References

- BESCHTA, R.L., D.L. DONAHUE, D.A. DELLASALA, J.J. RHODES, J.R. KARR, M.H. O'BRIEN, T.L. LEISCHNER, AND C.D. WILLIAMS. 2013. Adapting to climate change on western public lands: addressing the ecological effects of domestic, wild, and feral ungulates. Environmental Management 51:474-491.
- CHAMBERS, J.C., M.J. GERMINO, J. BELNAP, C.S. BROWN, E.U. SCHUPP, S.B. ST. CLAIR, M.J. GERMINO, J.C. CHAMBERS, AND C.S. BROWN. 2016. Plant Community Resistance to Invasion by Bromus Species: The Roles of Community Attributes, Bromus Interactions with Plant Communities and Bromus Traits. Exotic Brome-Grasses in Arid and Semiarid Ecosystems of the Western U.S. p. 275-306.
- CLINE, G.G. 1974. Peter Skene Ogden and the Hudson's Bay Company. University of Oklahoma Press. 279 pp.

- DALE, H.C. 1918. The Ashley-Smith Explorations and the Discovery of a Central Route to the Pacific 1822-1829 with the Original Journals. Cleveland, OH, USA: Arthur H. Clarke Company. 352 pp.
- DAVIES, K.W., J.D. BATES, B.L. PERRYMAN, S. ARISPE. 2021. Fall-winter grazing after fire in annual grass-invaded sagebrush steppe reduced annuals and increased a native bunchgrass. Rangeland Ecology and Management. 77:1-8.
- LAUNCHBAUGH, K.L., B. BRAMMER, M.L. BROOKS, S.C. BUNTING, P. CLARK, J. DAVISON, M. FLEMMING, R. KAY, M. PELLANT, AND D. PYKE. 2008. Interactions among livestock grazing, vegetation type, and fire behavior in the Murphy Wildland Fire Complex in Idaho and Nevada, July 2007. United States Geological Survey, Open-File Report 2008-1214.
- MACK, R. 1981. Invasion of Bromus tectorum L. into western North America; an ecological chronical. Agro-Ecosystems 7:145-165.
- PERRYMAN, B.L., B.W. SCHULTZ, M. BURROWS, T. SHENKORU, and J. WILKER. 2020. Fall-grazing and grazing exclusion effects on cheatgrass (Bromus tectorum) seed bank assays in Nevada, USA. Rangeland Ecology and Management. 73:343-347.
- PERRYMAN, B.L., R.E. WILSON, AND W.I. MORRILL. 2003. Viewpoint: Eastern Nevada Landscape Coalition position: There are consequences of doing nothing in natural resource management. What are they? Rangelands 25:30-34.
- PETERSEN, J.G. 2008. Route for the Overland Stage: James H. Simpson's 1859 Trail Across the Great Basin. Utah State University Press. 256 pp.
- SAMPSON, A.W. 1913. Range improvement by deferred and rotation grazing. U.S. Department of Agriculture Bulletin 34. p. I-16.
- SAMPSON, A.W. 1951. A symposium on rotation grazing in North America. Journal of Range Management 4:19-23.
- SCHMELZER, L., B. PERRYMAN, B. BRUCE, B. SCHULTZ, K. MCADOO, G. MCCUIN, S. SWANSON, J. WILKER, AND K. CONLEY. 2014. Case Study: Reducing cheatgrass (Bromus tectorum L.) fuel loads using fall cattle grazing. The Professional Animal Scientists 30:270-278.
- SHELEY, C. MARLOW, D. BOHNERT, M. BORMAN, R.MATAGONZÀLEZ, J. BUCKHOUSE, T. STRINGHAM, B. PERRYMAN, S. SWANSON, K. TATE, M. GEORGE, G. RUYLE, B. ROUNDY, C. CALL, K. JENSEN, K. LAUNCHBAUGH, A. GEARHART, L. VERMEIRE, J. TANAKA, J. DERNER, G. FRASIER, AND K. HAVSTAD. 2014. Western land managers will need all available tools for adapting to climate change, including grazing: a critique of Beschta et al. Environmental Management 53:1035-1038.
- SVEJCAR, T., C. BOYD, K. DAVIES, M.MADSEN, J. BATES, R. TROWBRIDGE, W., T. ALBRIGHT, S. FERGUSON, J. LI, B.L. PERRYMAN, AND R.S. NOWAK. 2013. Explaining patterns of species dominance in the shrub steppe systems of the Junggar Basin (China) and Great Basin (USA). Journal of Arid Land 5:415-427.
- YOUNG, J.A., AND C.D. CLEMENTS. 2009. Cheatgrass: Fire and Forage on the Range. Reno, NV, USA: University of Nevada Press. 348 pp.