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FATTY ACID COMPOSITION OF FORAGES AND THEIR EFFECT ON THE

FATTY ACID COMPOSITION IN BEEF CATTLE

By

Britney G. Allen

A dissertation submitted in partial fulfillment of the requirements for the degree

of

MASTER OF SCIENCE

in

Plant Science

Approved:

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Logan, Utah

2021

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ABSTRACT

Fatty Acid Composition of Forages and Their Effect on the Fatty Acid Composition in Beef Cattle, and Extension of Research Results to the Deaf Agricultural Community

by

Britney G. Allen, Master of Science

Utah State University, 2021

Major Professor: Dr. Jennifer MacAdam Department: Plants, Soils and Climate

The goal of the present study was to understand the transformations of the longchain fatty acids found in pasture and feedlot diets that occur as fatty acids are processed by microbes in the rumen of cattle, through transport in blood plasma and during deposition in a subcutaneous fat depot. The impacts of diet on the rumen bacterial microbiome and the short-chain fatty acids these microbes synthesize from feed carbohydrates was also assessed.

In the present study, short-chain fatty acids in the rumen and the rumen microbome, and long-chain fatty acids in the diet, blood plasma and subcutaneous fat were compared for a tannin-containing (birdsfoot trefoil) and a non-tannin legume (cicer milkvetch) pasture, a grass (meadow bromegrass) pasture, and a feedlot (concentrate) diet. The study demonstrated that rumen microbial diversity was reduced by feedlot diets compared with all three pasture diets. The rumen bacterial phylum *Tenericutes* was highly correlated with the rumen concentration of acetate. The ratio of acetic to propionic acid was greater in the rumens of cattle grazing grass than birdsfoot trefoil pasture, and least in the rumen of feedlot cattle. Feedlot diets contained a much higher ratio of omega-6 to omega-3 fatty acids than all pasture diets, but concentrations of long-chain fatty acids in blood plasma suggest these differences were reduced by rumen biohydrogenation of alpha-linolenic acid. In blood plasma, cattle fed concentrates and birdsfoot trefoil had more omega-6 fatty acid than cattle fed cicer milkvetch, and cattle fed both cicer milkvetch and birdsfoot trefoil had more blood omega-6 fatty acid than grass-fed cattle. Plasma of cattle grazing birdsfoot trefoil had more omega-3 fatty acid than the other pasture diets, and blood of all pasture-fed cattle had more omega-3 fatty acid than feedlot-fed cattle.

We concluded that even a relatively low concentration of tannin in the birdsfoot trefoil diet may have provided some protection to the unsaturated fatty acids in birdsfoot trefoil, inhibiting rumen biohydrogenation. Another aspect of this master's study was to demonstrate the effective extension of data, such as the potential benefits of legumefinishing compared with grass- or concentrate-finishing, to the community of deaf agricultural producers.

(105 Pages)

PUBLIC ABSTRACT

Fatty Acid Composition of Forages and Their Effect on the Fatty Acid Composition in Beef Cattle, and Extension of Research Results to the Deaf Agricultural Community Britney G. Allen

This study focused on how diet changes the rumen microbiome in the cattle and the effects of that on the long chain fatty acids (LCFA) by microbes in the rumen, and on the short chain fatty acids (SCFA) these microbes produce from feed carbohydrates like fiber and starch. The abundance of bacteria belonging to the phyla *Tenericutes* and *Proteobacteria* increased in response to high-fiber or high-starch diets, respectively. The production of two SCFA was positively correlated with the presence of increased *Tenericutes* (acetate) and *Proteobacteria* (propionate). A greater acetate to propionate ratio is associated with elevated production of enteric methane in the rumen, a lower ratio is more desirable. For LCFA, there are negative implications of omega-6 fatty acids and positive implications of omega-3 fatty acids for human health, so a lower omega-6 to omega-3 fatty acid ratio is considered more desirable.

Compared with grass-fed cattle, ecological concerns with raising beef can be mitigated in pasture systems by reducing methane emissions and improving soil health by using legumes that supply their own nitrogen. Compared with feedlot-finished cattle, the meat produced on birdsfoot trefoil pastures is healthier but better-tasting than the meat from grass-finished cattle. These benefits of non-bloating perennial legume pastures gives cattle producers an option for raising and marketing their own cattle that can increase beef profitability while reducing methane emissions and improving soil health. A further result of this study was the communication of the results of a relatively complex scientific study through an extension video accessible to all interested farmers and ranchers, and a second video demonstrating the elements of making an accessible video for the Deaf and hard of hearing communities.

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I would like to thank my husband, Ryan Allen, for being by my side as I've pursued this goal. My infinite appreciation is extended to the many neighbors and friends who stepped up when my plans changed unexpectedly. I especially appreciated Chris and Myia Barton for being able to help such an incredible amount my first semester, and Krystal Uresk for her continued help and support. I could not have accomplished this feat without your help. I would like to thank my daughter, Olivia, for being my reason to continue when discouragement felt overpowering.

I would like to thank my major professor, Dr. Jennifer MacAdam, for her countless hours in helping me with every aspect of my graduate school experience. I would especially like to thank her for her support and encouragement to integrate Deaf agriculture communication with the rest of my thesis work. I would also like to thank Drs. Robert Ward, Juan Villalba, Korrey Hintz, and Freeman King for their insight and help in preparing for and defending my thesis. Lastly, I would like to thank the many mentors, especially Dr. Fee Busby, that I had in my undergraduate work who listened to my abstract thoughts and helped me turn them into goals that I could work toward.

Britney G. Allen

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CHAPTER I

GENERAL INTRODUCTION

Birdsfoot trefoil (Lotus corniculatus) is a nitrogen-fixing legume that accumulates a limited amount of a condensed tannin. In previous studies, the ratio of omega-6 to omega-3 fatty acids in steaks of beef cattle finished on birdsfoot trefoil was less (more favorable) than in steaks of cattle finished on concentrates in a feedlot (Chail et al., 2016), and cheese made from the milk of dairy cows grazing birdsfoot trefoil had a greater concentration of omega-3 fatty acid than cheese made from the milk of cows grazing a mixed grass pasture, which in turn had a greater concentration than cheese made from the milk of cows fed a conventional total mixed ration in a dry lot (MacAdam et al., 2015). A cattle production system based on tannin-containing perennial legumes such as birdsfoot trefoil results in additional ecological benefits, such as increased nitrogen retention by beef cattle (Stewart et al., 2019) and increased rate of gain in beef cattle (MacAdam and Villalba, 2015). The goal of the work described in this thesis was to better understand the interactions between diet and the rumen microbiome resulting in the synthesis of some fatty acids and the transformation of others that can influence both environmental outcomes and the quality and quantity of meat.

Grazed pastures increase the sequestration of soil organic matter compared with fields used for grain crop production, the inclusion of nitrogen-fixing legumes in pastures eliminates the need for nitrogen fertilization, and the products and production methods of pasture-finished beef are more satisfactory to consumers concerned about the use of hormones and antibiotics in feedlot-finished beef as well as the impacts on air, soil and water quality. Finishing beef cattle on non-bloating legume pastures could become a profitable niche cattle production and marketing opportunity in the western United States where irrigated perennial legumes are more persistent and productive than irrigated grass pastures in mid-summer.

While it's important to design and carry out applied research, the results need to be effectively communicated to producers and other stakeholders. The technical study described in Chapter III was designed to investigate fatty acid synthesis and transformations during ruminant digestion as these processes are affected by diet. The results, however, have practical relevance to the selection of forage species that can improve the production of beef on western irrigated pastures. For example, there is likely to be a subset of ranchers in Utah or the northern Mountain West who could benefit from integrating tannin-containing perennial legumes such as birdsfoot trefoil into their beef production systems. Most state extension services routinely use YouTube channels to communicate, but personal experience made the author aware of a need to create extension videos that are accessible not only to hearing but also to members of the Deaf and hard of hearing community who are also ranchers. Therefore, Chapter IV of this thesis addresses this need, and includes two extension videos, one that was created to serve as a guide to making videos that are accessible to the Deaf and hard of hearing community, and the other to serve as an example of a video that makes research accessible to producers, and that also incorporates the elements that make the video accessible to the Deaf and hard of hearing community.

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CHAPTER II

LITERATURE REVIEW

1 | CATTLE MANAGEMENT CRITERIA

The beef cattle industry is an enormous asset to the American economy. In 2018, the cattle industry in the United States resulted in 18% of all agricultural cash receipts, a total of \$67.1 billion (US Department of Agriculture, 2019). Beef cattle and other ruminants are excellent at grazing forages on agricultural land that is unsuitable for annual crop cultivation. (USDA National Organic Program, 2013). Within the beef industry there are two niche markets: one is natural and organic beef, making up 3% (Drouillard, 2018). The other niche market that is important in the context of this study is grass-finished beef, which constitutes 1% of the US beef market (Hayek, 2018).

The difference between natural, organic, and grassfed must be considered from a legal point of view to clearly distinguish differences in the markets. For a product to be certified natural by the USDA it must meet three criteria: 1) the product must be minimally processed, 2) the product cannot contain any artificial ingredients, and 3) the product cannot contain any preservatives (Troxel, 2005). USDA certified organic meat must meet five criteria: 1) be allowed year round access to the outdoors, except in inclement weather, 2) be raised on certified organic land meeting all organic crop production standards, 3) be raised by specified animal health and welfare standards, 4) be fed 100% certified organic feed, and 5) be managed without feeding antibiotics, growth hormones, mammalian or avian byproducts (USDA, 2013). Grass-finished is an

expanding area of the beef market and is perceived as an environmentally friendly and humane way to raise beef. Grass-finished beef is not certified by the USDA so there are no federal regulations governing claims of grass fed status, although the main USDA grassfed criterion is that cattle have been fed solely forage after weaning off the mother's milk (FSIS USDA, 2019).

The American Grassfed Association, a nonprofit organization, does have detailed requirements for grassfed certification. There are four main criteria: 1) a forage diet free of grain or other concentrates, 2) access to pasture, 3) no antibiotics or hormones, and 4) a family farm origin (AGA, 2019). Cattle fed hay and silage and finished on birdsfoot trefoil pasture would qualify for grassfed certification within AGA specifications.

2 | EFFECTS OF PASTURE PLANT SPECIES

Certified grassfed beef has a more healthful ratio of omega-6 to omega-3 fatty acids than feedlot-finished beef, and any type of perennial pasture is better for soil health than the cultivation of annual crops such as cereal grains. The daily rate of cattle gain, however, is greater in a feedlot than on grass pastures, and this is cited by Capper (2012) as a more efficient model of beef production by the feedlot industry. To produce a billion pounds of red meat finishd on grass requires approximately 3.6 million animals. Feedlot production, by comparison, only requires 2.7 million animals to produce the same amount of red meat (Capper, 2012). In addition, while all cattle spend their first six to 12 months on grasslands, and mother cows live their entire lives on rangeland or pastures, the three to six months cattle spend in feedlots with a total lifespan of 14.6 months was compared by Capper (2012) to a total of lifespan of 22.6 months for cattle finished on grass. It has been calculated that birdsfoot-finished beef has a productivity closer to feedlot beef (approximately 2.9 million cattle to produce a billion pounds of beef) than to grass-finished beef (MacAdam & Villalba, 2015). Legume pastures also qualify as "grass-fed" under both the AGA and USDA organic certification specifications.

Birdsfoot trefoil can be planted in irrigated pastures and grazed by cattle as an option for either feeding or finishing in cattle operations. It is a nitrogen-fixing legume that does not require chemical nitrogen fertilizer. Perennial forage legumes are productive over multiple years without cultivation, contributing organic matter to the soil and therefore increasing soil health. Because tannins bind to proteins in the rumen and eliminate the threat of bloat, cows grazing the tannin-containing legume birdsfoot trefoil have produced milk with an increased omega-3 fatty acid concentration (MacAdam et al., 2015). Consumers liked the tenderness and juiciness of steaks from cattle finished on birdsfoot trefoil equally to steaks from grain-finished beef (Chail et al., 2016). Furthermore, the methane emissions from cattle grazing birdsfoot trefoil was found to be less than the methane emissions from cattle grazing the grass meadow brome (MacAdam et al., 2016). These benefits could lead to the creation of a profitable niche market for irrigated legume pasture-finishing in the beef industry unique to cattle finished on tannin-containing legumes such as birdsfoot trefoil and sainfoin.

Methane emissions from cattle constitute one-third of agricultural methane emissions (EPA 2013). Finishing cattle on a lower quality, higher fiber grass diet results in greater amounts of methane production than finishing cattle on concentrates. Methanogens are microorganisms in the rumen of cattle and other ruminants that generate hydrogen and carbon dioxide, ultimately producing methane. This methane production can be interrupted by either directly interfering with methanogens, or it can be interrupted indirectly by altering substrate availability. The condensed tannins in birdsfoot trefoil can precipitate proteins, causing them to leave the rumen before digestion, or they can inhibit methanogens directly or indirectly limit methanogenesis by reducing available hydrogen in the rumen (Hook et al., 2010).

Compared to grasses, average daily gains were greater on birdsfoot trefoil and other legumes, and approached feedlot levels of average daily gains in calves grazing in summer on irrigated pastures (MacAdam et al. 2011). Birdsfoot trefoil is high in protein and non-fiber carbohydrates, and low in neutral detergent fiber (NDF) relative to grasses (Chail et al., 2016). The utilization of nitrogen in rumen digestion is most efficient when the availability of easily digestible carbohydrates and proteins is synchronized in the rumen to maximize rumen microbe proliferation. Less-digestible fiber needs to be available to keep microbial growth in check and prevent excessive fermentation gases and bloat, which can cause ruminants to endure periods of discomfort and can lead to death if left unchecked (Howarth et al., 1991; Dado & Allen 1995).

Birdsfoot trefoil is like alfalfa in that both contain higher protein than is needed by cattle. Because it contains a sufficient concentration of tannins, birdsfoot trefoil does not cause bloat, but alfalfa can. The chemical nature of the tannins synthesized in birdsfoot trefoil results in release of precipitated plant proteins at the lower pH of the abomasum so they can be digested and their amino acids absorbed in the small intestine. When more ingested protein is directly used by the ruminant, nitrogen that might be used to generate energy in the rumen, resulting in the generation of ammonia that is absorbed into the blood through the rumen wall and lost to the environment in urine or milk, is used instead for microbial colonization, increasing ruminant retention of nitrogen compared with a non-tannin forage such as alfalfa (Stewart et al., 2019).

3 | OMEGA-3 FATTY ACIDS

In 2020, the primary source of protein in the American diet is the meat of chicken and beef. The second most consumed form of protein is dairy products, almost entirely from dairy cows (Pasiakos et al., 2015). There is a growing market in plant-based beverages, but only soy and pea milks have protein concentrations comparable to cow's milk (Bridges, 2018). As with beef, a low ratio of omega-6 to omega-3 fatty acids in dairy products is considered more healthful and is a signature of pasture-fed dairy cows (Benbrook et al., 2013). The ratio of omega-6 to omega-3 fatty acids in foods can predict implications for human health. In western culture it is typical for the ratio of omega-6 to omega-3 fatty acids to be close to 16:1, while our evolutionary history suggests that humans evolved with a diet that had an omega-6 to omega-3 ratio closer to 1:1 (Simopoulus, 2002a). Increasing omega-3 fatty acids has been shown to have benefits such as relieving symptoms of autoimmune disorders and inflammation, reducing risks of cardiovascular disease, and as a breast and colon cancer prevention treatment (Rose and Connolly, 1999; Kris-Etherton et al., 2002; Simopoulos, 2002a; Simopoulos, 2002b). Alpha-linolenic acid (C18:3), an omega-3 fatty acid, and linoleic acid (C18:2), an omega-6 fatty acid, are essential fatty acids and must be consumed in the diet. With the importance of including omega-3 fatty acids in diets and the popularity of beef cattle as a

protein source, increasing the omega-3 fatty acids in beef could improve the health of beef consumers in America.

Milk and cheese produced by cattle grazing pasture have an increased concentration of omega-3 fatty acids, making high omega-3 fatty acid concentrations a characteristic of organic milk (Benbrook et al. 2013; Dewhurst, 2006). Organic cattle generally graze grass pastures during the summer, but we see a similar or greater elevation of omega-3 fatty acids when cattle are grazed on birdsfoot trefoil. Cheese made from birdsfoot trefoil-fed organic cattle had significantly higher levels of omega-3 fatty acids than cheese made from the milk of grass-fed organic cows. Cheese made from the milk of both grass- and birdsfoot trefoil-grazed dairy cows were higher in omega-3 fatty acids than cheese made from the milk of cows from a conventional dairy fed a total mixed ration (MacAdam et al., 2015).

4 | FATTY ACID TRANSFORMATIONS

The composition of the fatty acids and the digestibility of forages fed to or grazed by ruminants depends on multiple factors. These include the forage species grown, the time of year it is grazed or harvested, the degree of maturity, and whether the forage is grazed or made into hay or silage. Perennial temperate legumes typically have more protein and less fiber than grasses and are more digestible (Dewhurst, 2013). The concentration of fatty acids in forages is lower in the summer, but the concentrations can be maintained if flowering is prevented (Clapham et al., 2005). There are substantial losses of omega-3 fatty acids when forages are made into hay or when they wilt prior to being made as silage. Red clover, (a legume) silage had increased levels of omega-3 fatty acids compared to grass silage (Dewhurst et al., 2003).

Fats undergo two major steps when they are metabolized in the rumen: the hydrolysis of ester linkages and the biohydrogenation of unsaturated fatty acids. Hydrolysis is the initial step and separates fatty acids from the glycerol backbone of diglycerides and triglycerides. The separated fatty acids undergo biohydrogenation, which adds hydrogens to two carbon atoms linked by double bonds, increasing the saturation of the fatty acid. Fatty acids can also be isomerized in the rumen, typically from a *cis* bond, where functional groups on both sides of a double bond are on the same side of the carbon chain, to a *trans* double bond where functional groups are on opposite sides of the carbon chain. An example of these processes is the transition of alphalinolenic acid, an 18:3 omega-3 fatty acid with cis double bonds at carbons 9, 12 and 15, to an 18:0 fatty acid (stearic acid). The first step is the migration and isomerization of the *cis*-12 bond to a *trans*-11 bond. Then there are successive reductions (addition of hydrogen atoms) of the cis-9 double bond, creating an 18:2 fatty acid and the cis-15 double bond, creating vaccinic acid (18:1). The last step reduces (hydrogenates) the trans-11 bond and produces the saturated fatty acid stearic acid (Bauman, 2003).

5 | IMPORTANCE OF EXTENSION AND FARMER COMMUNICATION

The Land Grant University system was established through the Morrill Act of 1862 to teach agriculture and agricultural mechanics. An agricultural research function was added by the Hatch Act of 1887, and the Cooperative Extension Service was established by the Smith-Lever act in 1914 (NRC, 1995). The role of extension has evolved along with cultural changes in the United States because of its local support (National Research Council, 1996). The essential activity of Extension since its establishment is bringing the most current research to the people who could use it. This information is provided to practitioners without the bias introduced by monetary incentive and potentially ulterior motives of private businesses. The Cooperative Extension Service was established to support rural communities in the United States, primarily by educating farmers, but has expanded to provide education in many different fields since its inception, including family resource management, human nutrition, 4-H, community economic development, and natural resource and environmental management (Al-Kaisi et al., 2015).

Effective scientific communication requires an educated population to think critically and understand the principles underlying their approach to farming or ranching, and scientists willing and able to communicate on the level of the audience. Scientific communication is vital for the general population to make informed decisions. It is useful and important for scientists to be sensitive and aware of how different cultures interact with scientific material, including the culture of communities that are considered disabled. Deaf agriculturists are a diverse group and should have the same access as other people to materials that will benefit their operations. There are many materials that address the prevention of hearing loss in agricultural settings (McBride et al., 2003; Ehlers and Grayden 2011; Couth et al., 2019), but there are no materials that are specifically for and about the Deaf agricultural community and outreach to this group.

6 | OUTREACH TO DEAF AGRICULTURAL PRODUCERS

Diversity and inclusion of the full range of viewpoints and experiences is needed to optimize decision-making in every area of society. It is important to extend input and decision-making to the deaf community to avoid "groupthink." Groupthink describes a group being stuck in believing that it is more important to reach a unanimous decision than to consider alternative courses of action. There is a hypothesis that groupthink contributed to the Watergate scandal and the Bay of Pigs fiasco (Janis, 1971). There are also hypotheses that increasing diversity can discourage groupthink in varying professions, and efforts to increase diversity for this purpose are being undertaken (Bernile, Bhagwat, & Yonker, 2018; Smith, 2016).

While most people tend to think of diversity as inclusion of various ethnic groups and genders, my goal is to extend diversity to inclusion of the Deaf and hard-of-hearing community. Preventing groupthink situations is different in the agricultural profession due to the rural and semi-isolated nature of agricultural work. It is important to ensure that adequate representation is available in agricultural spaces where it matters. I classify these spaces as political spaces (i.e. Farm Bureau, AgrAbility, and specialized crop societies) and learning spaces (schools and agricultural extension materials). These spaces are not just important for the Deaf agricultural community; they are important for the hearing community to avoid groupthink. Increasingly, video outlets such as the USU Extension YouTube channel are used to communicate with agricultural producers, while the support for fact sheets and bulletins that can be printed is decreasing. The goal of this portion of my master's program is to learn the best way to produce material to communicate complex ideas, such as the benefits of a decrease in the ratio of beef omega-6 to omega-3 fatty acids resulting from cattle grazing legume forages, in a format that would cater to the strengths of the Deaf agriculturist. Access to these materials would help the agriculturist make an informed decision in regard to their business. Knowing how the fatty acid ratio changes, the reduction in methane emissions, improved soil health from pasture systems, and high levels of animal productivity could give agriculturists a unique marketing strategy that could help their operation stand out in the beef cattle market. However, since the video created to illustrate the elements necessary for accessibility had to be recorded while microbiome and fatty acid data were still being created, the video included with this thesis is on a related subject from the same project.

Current outreach materials in agricultural production fields accessible to Deaf individuals are limited to written publications, but written extension publications do not need modifications to be accessible to the Deaf community. There are a substantial number of publications that specifically address situations that cause hearing loss in agricultural workers (Getts & Ploss, 1995). Creating accessible materials for all agriculturists, including the promotion of safety in all operations, and providing materials that can promote responsible agricultural practices, is an important but separate area of outreach. While using written publications and web sites with written descriptions are great, the Deaf community misses out on many aspects of the visual learning that can be gained from extension videos unless the videos have been created with Deaf and hard of hearing accessibility in mind.

Typically, extension videos rely on automated captioning systems. These systems can be inaccurate to the point that they are useless or even misleading to someone who is Deaf or hard of hearing. The frequency of incomplete or inaccurate captions is not solely an agricultural issue. The frequency and variety of faulty captions inspired Twitter users to create an entire Twitter thread labeled #craptions. What is available for deaf agriculturists on YouTube is stories of Deaf farmers located at

(https://www.youtube.com/watch?v=8kr8G6TzCbM). Most of these videos have signing narrators and accurate captions available or are crafted to support lip reading. Extension videos are geared toward a hearing audience that does not need captions or American Sign Language, but accurate captioning is a relatively simple solution that should be applied consistently to educational videos.

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CHAPTER III.

FATTY ACID TRANSFORMATIONS AND THE RUMEN MICROBIOME

Abstract

It has been established that dairy cattle grazing birdsfoot trefoil pastures have greater concentrations of omega-3 fatty acids in their milk than cattle on conventional total mixed ration diets. It has also been shown that the omega-6 to omega-3 fatty acid ratio of steaks from cattle finished on birdsfoot trefoil is less (more healthful) than that of steaks from cattle finished in a feedlot, and comparable to cattle finished on grass pastures. The objective of the present study was to gain a better understanding of changes in fatty acids that occur in the rumen microbiome of cattle grazing pure stands of legumes, and relate those changes to differences in fatty acids found in the blood and subcutaneous fat of cattle grazing a tannin-containing and a non-tannin legume compared with cattle grazing grass or fed a feedlot (concentrate) diet. This study demonstrated that rumen microbial diversity was reduced by feedlot diets compared with both legume and grass diets. The phylum *Tenericutes*, which was more dominant in pastured than feedlotfed cattle, was highly correlated with the synthesis of acetate, a short-chain fatty acid (SCFA) synthesized in greater amounts by rumen microbes of cattle on diets higher in fiber. The ratio of acetic to propionic acid, a SCFA associated with higher-starch diets, was greater for grass- than for birdsfoot trefoil-fed cattle. Feedlot diets contain a much higher ratio of omega-6 to omega-3 fatty acids, but these differences were reduced by rumen biohydrogenation of the omega-3 fatty acid alpha-linolenic acid, based on

concentrations of these long-chain fatty acids (LCFA) in blood plasma. However, it was still possible to detect a lower omega-6 to omega-3 fatty acid ratio in the subcutaneous fat of birdsfoot trefoil-fed cattle backfat compared with cattle fed the legume cicer milkvetch.

1 | INTRODUCTION

The balance of omega-6 to omega-3 fatty acids was more healthful in ribeye steaks from cattle finished on grazed pasture compared to cattle finished in a feedlot (Chail et al., 2016). The omega-3 fatty acids of cheese made from the milk of organic cows grazing birdsfoot trefoil pastures was greater than that of cheese made from the milk of cows grazing grass pastures (MacAdam et al., 2015) and both were greater than cheese made from the milk of total mixed ration-fed (conventional) dairy cows. Shortchain fatty acids are generated by rumen microbes from the fermentation of dietary carbohydrates, while dietary long-chain fatty acids are metabolized by rumen microbes and by enzymatic activity, they are transported in the blood, and deposited in the fat of the animal being fed. The present study was undertaken to determine the interaction of diet with the rumen bacterial microbiome and resulting synthesis of rumen SCFA, and to determine transformations of dietary LCFA as they move from the rumen to the blood and into subcutaneous fat. When fiber (amylase-treated neutral detergent fiber; aNDF) concentrations are elevated, more of the SCFA acetate is synthesized, and when non-fiber carbohydrates (NFC) are elevated, more of the SCFA propionate is synthesized. Propionate is converted to glucose by the liver, and elevated propionate is thought to result in greater deposition of intramuscular fat (Smith and Johnson, 2014), resulting in meat that is more tender and juicier. Diets enriched in starch (i.e., concentrate diets) result in increased fat deposition and greater intramuscular fat relative to cattle finished on grass. Previous work from the MacAdam lab demonstrated that finishing cattle on birdsfoot trefoil, a tannin-containing forage legume, resulted in steaks that were as tender and juicy as steaks from feedlot-finished cattle (Chail et al., 2016). The present study was undertaken to investigate the fatty acid transformations leading to differences among cattle fed legume, grass, and feedlot diets.

Dietary polyunsaturated long chain fatty acids such as the omega-6 fatty acid linoleic (C18:2) or the omega-3 fatty acid alpha-linolenic (C18:3) are hydrogenated in the rumen during microbial fermentation of feeds, transforming them into mono-unsaturated or saturated fatty acids (Nguyen et al., 2019). In forages, both linoleic and alpha-linolenic acids are present, but alpha-linolenic acid is present in higher concentrations, while in concentrates such as corn grain, linoleic acid is dominant and there is little alphalinolenic acid (Rego, 2016). In a typical feedlot ration containing grain and sufficient forage to support rumen function, the ratio of linoleic to alpha-linolenic acid is 10:1 (Rego, 2016). Studies with condensed and hydrolysable tannins has demonstrated that tannins suppress rumen biohydrogenation; however, in both forage and concentrate diets, linoleic acid is protected more effectively than alpha-linolenic acid, and tannins do not necessarily decrease the ratio of omega-6 to omega-3 fatty acids in intramuscular fat (Vasta et al., 2009).

This study was undertaken to gain insight into the metabolism of fatty acids in cattle grazing legume pastures compared with cattle on grass pastures as well as in comparison to cattle on a feedlot diet. The pasture treatments employed were birdsfoot trefoil (*Lotus corniculatus* L.; BFT), cicer milkvetch (*Astragalus cicer* L.; CMV), and meadow bromegrass (*Bromus biebersteinii* Roem. & Schult.; MBG).

2 | MATERIALS AND METHODS

3.1 | Cattle and diets

All animal interactions were conducted according to procedures approved by the Utah State University Institutional Animal Care and Use Committee as protocols 2733 and 2858. Five replications of three monoculture pasture species were established in August of 2012 at the Utah State University (USU) Intermountain Irrigated Pasture Project farm in Lewiston, Utah (41.95°N; 111.87°W; altitude 1370 m.a.s.l.) and were rotationally stocked each summer for approximately 12 weeks beginning in 2013. See Fig. 1 for the pasture treatment plot plan.

In March of the two study years, 20 two-year-old Angus heifers (*Bos taurus*) were selected from the USU beef herd for use in this study. Heifers were held in a drylot pen and fed a 1:1 mixture of alfalfa hay and corn silage until the initiation of the study. During this period, heifers were halter-trained and introduced to electric fencing. Heifers were sorted by weight into five groups, and individuals from each weight group were randomly assigned to the same replication of one of the four treatments (BFT, CMV, MBG or Feedlot). In 2018, three poorly trained heifers were removed from the study, reducing the number of replications of the grazing treatments to four. Each spring, a few days before heifers began grazing or adjusting to feedlot diets, and again in August on the day the study was terminated, samples of rumen fluid, blood and intramuscular fat were collected.

2.2 | Pasture treatments

Each experimental unit in the grazing treatments consisted of a 0.365-ha pasture and the assigned heifer. Paddocks consisted of one-twelfth pasture, defined by portable electric fencing, and each paddock was supplied with fresh water and a trace mineralized salt block (Morton iOFIXT T-M). Pastures were rotationally stocked, with each paddock grazed for 3.5 days. Legume monocultures could be grazed because BFT and CMV are both non-bloating legumes; birdsfoot trefoil because it contains a low concentration of a condensed tannin (MacAdam et al., 2013), and cicer milkvetch because of the structure of its leaves (Lees et al., 1982). Meadow bromegrass is a high-quality cool-season bunchgrass that is commonly used in high-elevations pastures in the western U.S. Heifers were rotated through paddocks in the same order in all pastures to facilitate handline irrigation and avoid compaction of wet soil.

2.3 | Pasture sampling

Pasture samples and dry matter (DM) accumulation data were collected three times during each grazing season, on 7 June, 5 July and 2 August in 2017 and 20 June, 19 July, and 9 August in 2018. Intake on pastures was estimated as the difference between pre- and post-grazing forage DM and expressed as intake ha⁻¹ d⁻¹. Pasture intake was also estimated from near infrared spectroscopy (NIRS) values for DM intake (DRYMI) expressed as a function of body weight and used to calculate intake ha⁻¹ d⁻¹. A rising plate meter was calibrated in kg ha⁻¹ for each of the three pasture species by taking single measurements and cutting the forage beneath the plate to the soil surface and drying the sample to constant weight at 60°C (MacAdam and Hunt, 2015). Calibration samples of each forage species were taken from five replications in both pre- and post-grazed paddocks. The rising plate meter was used predict the DM in pre-grazed paddocks just before cattle were turned in or in post-grazed paddocks just after cattle were moved to a new paddock, by taking at least 30 measurements while walking through the paddock in a "lazy W" pattern. The mean of these measurements from each paddock was used to create a linear relationship between calibration sample DM and the respective rising plate meter measurements. Forage in pastures was removed only by grazing, not clipping, and grazing pressure was moderate, so all forages grazed during this study were relatively mature.

For the tannin assay, samples of the seeded species were collected by walking across the paddock from corner to corner clipping whole stems to a 10-cm stubble. Forage nutritive value samples were collected similarly, but all grazable pasture plant species were included; thistles were not sampled and were spot sprayed with Milestone VM (a.i. triisopropanolammonium salt of 2-pyridine carboxylic acid, 4-amino-3,6dichloro-) at a rate of 0.365 L ha⁻¹ following grazing. Tannin and forage nutritive value samples were frozen in the field under dry ice and stored at -20°C until they were freezedried.

2.4 | Feedlot treatment

The feedlot treatment of the study was located at the USU Animal Science Farm in Wellsville, UT (41.67°N 111.89°W; altitude 1369 m.a.s.l.). The five heifers allocated to the feedlot treatment were randomly assigned to individual adjacent 5- x 10-m pens in a covered barn and received a total mixed ration (TMR) composed of 25% alfalfa hay, 25% corn silage and 50% hammer-milled barley. Heifers had free access to water and trace mineralized salt blocks (Morton iOFIXT T-M).

The TMR was offered each day at 0900, and the amounts offered were 27 kg head⁻¹ in both years. In 2017, the feed was split between two 79-L feeders inside each pen and in 2018, feed was offered in one larger 378-L feeder inside each pen. Refused feed was collected at 0850 the following morning and weighed; fresh feed was offered immediately upon refusal collection. The difference between offered and refused feed was recorded as feed intake.

2.5 | Feed and forage analysis

Nutritive value of the TMR used for the feedlot treatment was determined by a commercial lab (Cumberland Valley Analytical Services, Waynesboro, PA). Freeze-dried forage samples were ground to pass the 1 mm screen of a Wiley mill, and forage nutritive value was determined via NIRS with a FOSS 2500 spectrophotometer (FOSS Analytics, Hilleroed Denmark). A mixed hay equation (NIRS Forage and Feed Testing Consortium, Hillsboro, WI) developed according to procedures of Shenk and Westerhaus (1991) from a calibration set containing multiple species was used to predict forage nutritive value. The distribution and boundaries of BFT, CMV and MB sample spectra were well-represented by the population structure of spectra in the calibration set, so no additional wet chemistry was required. Determination of ADF, CP, aNDF, and acid detergent lignin of NIRS calibration samples were made according to AOAC International (2012) methods 973.18, 984.13, 2002.04, and 942.05, respectively. Total digestible nutrients and

non-fiber carbohydrate concentrations were calculated according to the equations of Undersander et al. (2010).

Forage samples were analyzed for condensed tannins based on the butanol-HClacetone method of Grabber et al. (2013). Tannin assay solution contained 0.15% w/v ammonium iron (III) sulfate dodecahydrate, 3.3% v/v water, 5% v/v concentrated HCl, 41.7% v/v butyl alcohol, and 50% v/v acetone. Briefly, triplicate 0.030 g DM of ground plant tissue was suspended in 15 mL of tannin assay solution and heated for 2.5 h in a 70 °C water bath. Samples were mixed periodically during heating. Standard, blank solutions and check samples were included in each run. After tubes cooled, they were centrifuged at 5,000 *x g* for 10 minutes and the absorbance of the supernatant was determined at 554 nm.

Tannin standards for the spectrophotometric assay were isolated from samples of tannin-containing plant material based on Hageman (2011). Briefly, a suspension of 10% w/v finely ground plant material in 1% v/v acetic acid, 24% v/v water and 75% v/v acetone was sonicated for 30 minutes with periodic mixing. Mixtures were centrifuged for 10 minutes at 3000 x g and the supernatant filtered through a coarse fritted disk; plant material was extracted a total of three times and supernatants combined. The supernatant was mixed with an equal volume of ethyl ether and the aqueous layer was retained; the supernatant was extracted a total of three times with equal volumes of ethyl ether. The acetone and ethyl ether remaining in the aqueous solution were removed by rotary evaporation. The aqueous solution was mixed with Sephadex LH 20 resin equilibrated in a 4:1 v/v ethyl alcohol;water solution, rinsed with 95% ethyl alcohol, and extracted with a

3:1 v/v acetone:water solution. The acetone was removed by rotary evaporation, and the aqueous solution was frozen and freeze-dried.

2.6 | Collection and analysis of animal samples

Rumen liquor of each animal was extracted using an orally administered Geishauser probe connected to a suction pump with flexible thermoplastic tubing (Tygon; Saint-Gobain Corporation). The first 100 mL of rumen fluid was discarded to avoid contamination from saliva, and 300 mL was collected in sterile containers for analysis. Rumen fluid was cooled on crushed ice following collection then transported to the lab where samples were stirred, pH measured, and 15-mL aliquots were stored at -80°C until fatty acid and rumen microbiome analyses were run.

Blood was obtained from the caudal vein of each animal using sterile 18-gauge, 2.5-cm-long needles, and 7 mL glass whole blood (lavender cap) Vacutainers lined with K+EDTA (Becton, Dickinson and Company, Franklin Lakes, NJ). Filled blood containers were inverted to mix blood with EDTA, cooled on crushed ice, transported to the lab, and centrifuged at 2100 x g for 15 minutes at 4°C. Two 0.5-mL aliquots of plasma were stored at -80°C until fatty acid composition was determined.

Under local anesthesia, a 1-cubic-centimeter sample of subcutaneous fat was removed from the loin of each animal. Before surgery, 8 to 10 cc of 2% lidocaine was administered for pain relief. Sampled tissue was wrapped in aluminum foil, labeled, and frozen in liquid nitrogen for transportation to the lab where it was stored at -80°C until tissue was ground and analyzed for fatty acid composition. Wounds were sutured, 750 mg of ampicillin was injected at the surgical site and the wound was covered with AluShield aerosol bandage and fly spray.

2.7 | Fatty acid analysis

Short chain fatty acids (SCFA) were extracted from the rumen fluid and vortexed with 0.9 mL of distilled water. Next, 0.2 mL acid solution (250 g/L of metaphosphoric acid containing 2 g/L of ethyl butyric acid as an internal standard) was added, centrifuged at 10 $000 \times g$ for 20 min and the supernatant was filtered and stored at 4°C. Gas chromatograph analysis was performed using a Shimadzu GC2010 (Shimadzu Scientific Instruments, Columbia, MD). Calibration was performed using individual SCFA standards, and SCFA concentrations were normalized to sample nucleic acid content by measuring absorbance at 260 nm (Ward et al., 2017).

Fatty acid methyl ester (FAME) analysis was done on the feed treatments and cattle tissue. These samples were prepared by the method described by O'Fallon et al. (2007). One gram of raw homogenate was weighed into a screw-cap glass vial along with an internal standard solution of tridecanoic acid (0.5 mg/mL in methanol; T-135; Nu-Chek Prep, Inc., Elysian, MN), and the vial was sealed with a polypropylene-lined cap (ThermoFisher Scientific, Waltham, MA). Vials were placed in a water bath (catalog number 67120; Precision Scientific, Chicago, IL) for incubation at 55°C.

Hexane was used to extract FAME before analysis by gas chromatography (GC). Separation of FAME was performed by a Shimadzu GC-2010 (Shimadzu Corporation; Kyoto, Japan) equipped with a HP-88 capillary column (100 m by 0.25 mm by 0.20 μm; Agilent Technologies, Palo Alto, CA) and a flame ionization detector (FID). The gas chromatograph was operated based on conditions described by Tansawat et al. (2013). The column head pressure was 195.6 kPa and the total flow rate was 129.1 mL/min (column flow: 2.47 mL/min; purge flow: 3.0 mL/min). One microliter of sample was injected with a split ratio of 50:1. The oven was held at 35°C for 2 min, then increased to a temperature of 170°C at a rate of 4°C/min, then held for 4 min, then increased to a temperature of 240°C at a rate of 3.5° C/min, and then held for 7 min. Hydrogen was used as the carrier gas. The injector and FID were operated at 250°C. Fatty acids were identified based on the similarity of retention times with gas chromotography reference standards (Nu-Chek Prep, Inc.). Short-chain fatty acid concentrations were calculated relative to initial wet sample weight (mg g⁻¹) (Chail et al. 2016); LCFA composition is reported as percent of total fat.

2.8 | Rumen microbiome analysis

Rumen microbiome composition was determined by isolating DNA from the rumen fluid using the QIAamp DNA stool minikit (Qiagen) (Hintze et al., 2014; Rodriguez et al., 2019). Rumen fluid samples were thawed and immediately centrifuged at 10,000 × g for 10 min. Supernatant was discarded, and bacterial DNA was extracted from the pellets. The taxonomic abundance (α -diversity) and species diversity (β -diversity) of rumen bacteria were determined via 16S sequencing using primers that amplify the V3-V4 hypervariable region (Klindworth et al., 2013). Libraries were prepared using MiSeq v3 reagents using paired 300 bp reads, including 5% PhiX to serve as an internal control. Equal amounts of the PCR products with different barcodes were combined, amplified, and sequenced using protocols adapted by the Center for Integrated BioSystems at Utah State University. Microbiota sequences were processed through the latest version of QIIME2 and DADA2 software (Callahan et al., 2016). To assign taxonomy, the qiime feature-classifier classifysklearn command was used with a classifier pre-trained for the V3-V4 region and the most recent release of the Silva database (Quast et al. 2013).

Alpha-diversity measures include the total number of amplicon sequence variant (ASV) sequenced, Chao1 richness (number of species represented), Faith's phylogenetic diversity (phylogenetic distance of species present), and Shannon index (weighted abundance of species present). β -diversity was determined using unweighted (qualitative measure that is sensitive to low abundance features) and weighted (accounts or abundance of species) unifrac distance measures and is represented as principal coordinates plots (PCoA) of the first two coordinates. β -diversity values among test groups were analyzed by the nonparametric permanova test in Qiime2, which partitions a distance matrix among sources of variation to describe the strength and significance that a categorical variable has in determining variation of distances. A permanova p value <0.01 for this test is considered statistically significant.

2.9 | Statistical Analysis

The long-chain fatty acids of interest in the diet were analyzed with a mixed model in which species, period and their interaction were fixed effects, while year, replication block, and sampling plot within replication were random effects. The repeated periodic measurements on the same plot were modeled with unrestricted variance-covariance error structure (UNR) based on information criteria selection to account for heterogenous variations and correlations of the three periods. Model residuals were checked and no violation of assumptions of the statistical tests were found. Simple effects of species for each period and of period for each species were compared using Tukey-Kramer's method to adjust for multiplicity.

A mixed model was also formulated to analyze long-chain and short-chain fatty acids in dietary, animal blood plasma and subcutaneous fat samples. Species, a covariate value comprising the fatty acid value of diets, rumen fluid, blood plasma or subcutaneous fat in May, and their interactions were fixed effects. Although the May fatty acid composition significantly affected final measurements in some responses, the interaction was insignificant in all analyzed responses. Therefore, main effect of species adjusted at the average level of the May fatty acid composition were compared by the Tukey-Kramer's method. All analyses were performed using PROC GLIMMIX (SAS/STAT 15.1, SAS Institute Inc., Cary, NC). Significance was defined at the 0.05 level.

3 | RESULTS AND DISCUSSION

3.1 | Diet and intake

There were heifers on pasture (BFT, CMV and MBG) and feedlot treatments in both 2017 and 2018, but feedlot animals were only assayed for rumen microbiome and fatty acid data in 2018. In 2017, grazing began 22 May and ended 7 August 2017 and in 2018, grazing and feedlot feeding began 29 May and ended 20 Aug. In 2017, feedlot heifers were confined in feedlot pens from 30 June to 8 August and intake was assessed twice, from 3 to 13 July and 27 July to 6 Aug. In 2018, feedlot heifers were confined from 21 May to 6 August and intake was assessed three times, from 1 to 9 June, 28 June to 7 July and 26 July to 4 August. In 2017, initial weights of heifers in all treatments was 504 ± 7 kg and final weights were 563 ± 9 , 522 ± 12 , 540 ± 15 , and 559 ± 8 kg for BFT, CMV, MBG and feedlot treatments, respectively. In 2018, initial heifer weight was 587 ± 13 kg and final weights were 607 ± 21 , 597 ± 16 , 612 ± 18 , and 634 ± 41 kg for BFT, CMV, MBG and feedlot treatments, respectively. All heifer average daily gains (ADG) were less than 1 kg/d and the only ADG treatment difference was between BFT and CMV in 2017.

Intake was more variable through the season for cattle on pasture diets than in the feedlot (Fig. 2). Data for diet composition (Table 1) show that protein was greater, and fat was less in the two legumes, BFT and CMV, than the grass (MBG). The fiber (aNDF) concentration of BFT is similar to that of the feedlot treatment; the aNDF of the MBG treatment is greater than that of the other treatments, and CMV has the least aNDF. The calculated values (total digestible nutrients, TDN; non-fiber carbohydrates, NFC) suggest that BFT and CMV contain similar energy. Intake calculated as pre- minus post-grazing dry matter (Fig. 2A) suggests greater intake for CMV than BFT, but intake calculated as DRYMI by NIRS (Fig. 2B) predicted greater intake for BFT than CMV, which is more likely to be correct based on ADG. The NIRS TDN concentration of both legumes is similar to that of the feedlot treatment and greater than for the grass (Table 1).

3.2 | Rumen microbiome

The enzymatic pathways, and the byproducts of those pathways, change depending on the composition of the bacteria and archaea communities in the rumen (Song et al., 2018). Pairwise comparisons of the family-level within-treatment α -diversity of the rumen bacterial microbiome demonstrates that rumen microbiome diversity of heifers on the pasture treatments (BFT, CMV and MBG) was statistically similar in both 2017 (Fig. 3A) and 2018 (Fig. 3B), while feedlot treatments had significantly less diversity than pasture treatments (Fig. 3C). The greater diversity of the pasture treatments is also apparent from a comparison of the number of small-population phyla in the three pasture treatments with the feedlot treatment (Fig. 4C). These differences in microbiome ecosystems can account for differences in short chain fatty acid (SCFA) composition among the treatments (Buitenhuis et al., 2019).

Pairwise comparisons of the relative abundance of rumen bacterial phyla (Fig. 4) demonstrate similarities in abundance of phyla as well as phylum diversity among the three pasture treatments in 2017 (Fig. 4A) and 2018 (Fig. 4B). In published studies of the bovine rumen microbiome, the dominant bacterial phyla are commonly *Firmicutes*, *Bacteroidetes*, and *Proteobacteria* (Vasta et al., 2019) and this was also the case for the feedlot treatment in the present study where those three phyla represented 95% of the rumen bacterial microbiome (Fig. 4C). More than 90% of the bacterial microbiome comprised *Firmicutes* and *Bacteroidetes* in all treatments, but *Proteobacteria* increased from between 1 and 2.5% of the microbiome in pasture treatments to 25% in the feedlot treatment, while *Bacteroidetes* decreased from 66% in pasture treatments to 48% in the feedlot treatment (Fig. 4C).

It is notable was that the phylum *Tenericutes* was third in abundance in all pasture treatments, even though it only represented between 2 and 5.5% of the bacterial population in all treatments (Fig. 4A and 4B). The fourth most abundant phylum in pasture treatments was *Proteobacteria* or *Kiritimatiellaeota*. A few other studies have also identified the phylum *Tenericutes* as elevated in the microbiome of ruminants on

high-forage diets compared with the same animals on high-starch rations: in beef cattle fed grass hay (Petri et al., 2013), in Tibetan sheep (*Ovis aries*) grazing native pasture (Cui et al. 2019) and in Chinese Tan sheep grazing a mixture of 90% legumes and 10% grass (Fu et al., 2020).

Fernando et al. (2010) compared rumen bacterial phyla of steers fed prairie hay with those transitioning to a high-grain diet and found that the ratio of *Firmicutes* to *Bacteroidetes* (F:B) was similar between the two groups until the 3rd step (40:60 fiber to grain) of the transition to a feedlot diet, when the F:B was less for cattle fed more grain. In the present study, after 12 weeks on either pasture or feedlot diets, the F:B was 0.36 for combined pasture treatments and 0.44 for the feedlot treatment, the opposite result. In the present study, the ratios of *Bacteroidetes* to *Firmicutes* were 2.8 and 1.9 for the pasture and feedlot treatments, respectively.

3.3 | Short-chain fatty acids

Rumen SCFA result primarily from microbial digestion of dietary carbohydrates, and feed with a greater concentration of fiber, such as MBG relative to the two legumes, usually results in the synthesis of more acetic and less propionic acid (Table 2); differences among treatments in these two SCFA and their ratio by year can be seen in Fig. 5. In the summary statistical analysis across both years (Table 2), acetic acid is greater for MBG than for CMV or the feedlot treatment (Fig. 5A), while propionic acid is greater for the feedlot treatment than any pasture treatment (Fig. 5B). The ratio of acetic to propionic acid (A:P; Table 2 and Fig. 5C) is greater for MBG than for BFT, and BFT is greater than the feedlot treatment; CMV is intermediate to MBG and BFT. Although we expect diet to affect acetic and propionic acids more than other SCFA, many phyla were either positively or negatively correlated with SCFA synthesis. Figure 6 illustrates the significant positive and negative correlations between rumen SCFA and bacterial phlya; both are listed in alphabetical order in the figure. Across all treatments, there is only one strongly positive correlation, and that is between acetic acid and the phylum *Tenericutes*, which was more abundant in the BFT and MBG treatments than *Proteobacteria* (Fig. 4A and B) and was the fourth most abundant phylum in the feedlot treatment (Fig. 3C).

The focus of this study is on acetic and propionic acids due to the relationship those fatty acids have with milk production and animal performance. Propionate is glucogenic, which supports the deposition of intramuscular fat (Olafadehan, 2016; Rodriguez et al., 1985). An important aspect of this study was to determine if SCFA synthesis in the rumen was affected differently by a legume compared with a grass or feedlot diet.

Maintaining an appropriate acetate to propionate ratio (A:P) is important for preventing rumen acidosis, a condition that can affect the health of the cow and change her productivity (Sauvant et al., 1998). The most beneficial A:P is 2.2:1 or greater (Hutjens, 1998); all treatments in this study met or exceeded that criterion including the feedlot treatment. The A:P is also considered to be a predictor of methane emissions, which are greater when the A:P is elevated and which occurs on higher-fiber, lowerquality forage diets. The largest component of the rumen microbiome is the bacteria, and their composition is influenced by the structural carbohydrate (aNDF) and the non-fibrous carbohydrate (NFC) composition of feed. In turn, rumen bacteria alter feed fatty acids and synthesize new proteins and vitamins (Vasta et al., 2019). The dominant fatty acids in forages including BFT, CMV and MBG are the omega-6 fatty acid linoleic (C18:2 n6) and the omega-3 fatty acid alpha-linolenic (C18-3 n3; Table 3). Alpha-linolenic is the dominant fatty acid in forages, but in the feedlot diet, where half the diet comprised the forages alfalfa hay and corn silage and the other half comprised the concentrate barley, the proportion of linoleic acid was greater than that of alpha-linolenic acid by an order of magnitude. This difference is illustrated as the omega-6 to omega-3 (n-6:n-3) fatty acid ratio (Table 3). A lower n-6:n-3 in red meat is desirable and was achieved in BFT-finished cattle along with tenderness and juiciness equal to that of grain-finished beef (Chail et al., 2016).

Differences in the concentrations of dietary LCFA in blood plasma at the end of the 3-month grazing period (Table 4) compared with the fatty acid composition of the diet (Table 3) suggest the degree of biohydrogenation (loss of double-bonds) that occurred due to microbial activity in the rumen. Alpha-linolenic, with 3 double bonds, constituted about 50% of LCFA in pasture diets and only about 2.5% of the feedlot diet. In the blood of pastured cattle, alpha-linolenic is reduced to about 10% of the LCFA although there is significantly more in the blood of cattle grazing BFT than cattle grazing CMV or MBG (Table 4), and alpha-linolenic is reduced to approximately 2% in the blood of feedlot-fed cattle. Linoleic acid, with 2 double bonds, constituted about 20% of the LCFA concentration of pasture diets and about 30% of the feedlot diet. In the blood plasma of BFT and feedlot-fed cattle, the linoleic acid concentration by the end of the study was 30%, while it constituted less for CMV, and even less in the blood of grass-fed cattle, suggesting that condensed tannins were protective of linoleic acid in the rumen. The n-6:n-3, however, did not differ for the three pasture diets, but was significantly greater (which is considered less healthy) in the blood of feedlot-fed cattle (Table 4).

The subcutaneous fat of cattle, along with intramuscular fat, accumulates after bone and muscle growth have slowed or stopped (Owens et al., 1995). Although we were using 2-year-old heifers in this study, we had reason to think that turnover or further deposition in the subcutaneous fat of animals 24-30 months of age might reflect the influence of different diets (Okumura et al., 2007). Both the linoleic acid (n-6) and the alpha-linolenic acid (n-3) concentrations of the subcutaneous fat of BFT-fed cattle was significantly greater than that of the CMV- and MBG-fed cattle, and the n-3 concentration was greater than that of the feedlot-fed cattle (Table 5). These differences were sufficient to result in a lower n-6:n-3 for BFT-fed cattle than for CMV-fed cattle, although the BFT n-6:n-3 was not significantly different from the MB- and feedlot-fed cattle. In Fig. 7, the statistical differences among treatments are provided separately for 2017 and 2018 for both blood plasma and subcutaneous fat concentrations of linoleic and alpha-linolenic acids and for their ratio.

In Fig. 8, pie charts of the five most abundant LCFA for dietary, blood plasma, and subcutaneous fat are presented side-by-side to illustrate the transitions in fatty acids that occur in the rumen and during subcutaneous fat deposition. Pasture diets were dominated by alpha-linolenic, linoleic, and palmitic fatty acids, while the feedlot diet contained linoleic, palmitic (C16:0) and oleic (C18:1) fatty acids in about equal proportions. It can be seen that much of the alpha-linolenic acid (n-3) in the pasture diets was hydrogenated by rumen microbes, increasing the proportions of linoleic (n-2), oleic (n-1) and stearic (C18:0) fatty acids absorbed into the blood following gastric digestion. For heifers on feedlot diets, biohydrogenation in the rumen increased the proportion of stearic acid while the proportions of oleic and palmitic acids both decreased. In the subcutaneous fat, the proportions of LCFA are nearly identical regardless of diet, with both alpha-linolenic and linoleic acids at less than 1%, while oleic constitutes approximately half the fat, and the balance of subcutaneous fat consists primarily of the saturated fatty acids palmitic and stearic.

4 | CONCLUSION

The present study demonstrated that rumen microbial diversity was suppressed by feedlot diets compared with pasture diets. The phylum *Tenericutes*, which was more dominant in pastured than feedlot cattle, was highly correlated with acetic acid, a short-chain fatty acid synthesized in greater amounts by rumen microbes of cattle on higher-fiber pasture diets. The ratio of acetic to propionic acid, a short-chain fatty acid associated with lower methane emissions, was greater for cattle on grass- than on birdsfoot trefoil-pasture diets and greater for all pasture diets than for cattle on feedlot diets. Feedlot diets contain a much higher ratio of omega-6 to omega-3 fatty acid alpha-linolenic acid, based on concentrations of the long-chain fatty acids in blood plasma. However, it was still possible to detect a lower omega-6 to omega-3 fatty acid

ratio in the subcutaneous fat of birdsfoot trefoil-fed cattle compared with cattle grazing the legume cicer milkvetch. A further aspect of this study is the effective extension of data, such as the potential benefits of legume-finishing compared with grass- or concentrate-finishing, to Deaf and hard-of-hearing agricultural producers.

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TABLES

Diet:	BFT§	BFT SEM	CMV	CMV SEM	MBG	MBG SEM	Feedlot [†]
2017							
Protein	20.70	1.15	25.39	0.92	13.40	0.88	14.70
Fat	2.04	0.09	1.78	0.13	2.77	0.16	2.40^{\ddagger}
aNDF	33.74	2.31	25.46	1.55	57.42	2.27	32.85
ADF	23.87	1.36	21.23	1.15	35.14	1.29	20.60
Lignin	4.97	0.28	4.56	0.24	5.41	0.26	3.94
TDN	74.58	1.52	77.53	1.28	61.99	1.46	72.35
NFC	38.94	1.36	41.01	1.35	23.84	1.60	42.70 [‡]
CT	19.55	0.82	1.90	0.07	1.26	0.12	0.71
2018							
Protein	20.65	0.92	22.67	1.01	7.51	0.51	12.07
Fat	2.41	0.13	1.52	0.32	3.88	0.35	2.40^{\ddagger}
aNDF	31.91	2.26	26.43	1.99	64.15	1.64	35.87
ADF	23.69	1.42	22.78	1.49	40.85	1.18	24.80
Lignin	5.86	0.52	6.43	0.31	4.96	0.31	4.14
TDN	75.54	1.62	76.59	1.70	55.97	1.35	68.00
NFC	40.67	1.71	43.05	1.01	20.78	1.12	42.70 [‡]
CT	12.96	0.28	0.86	0.10	-0.61	0.08	0.68

TABLE 1	Diet composition (%)
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Note. Analysis carried out by Cumberland Valley Analytical Services. NFC and fat values were determined for a sample from the same feedlot analyzed by Dairy One Forage Testing Laboratory in 2015. BFT, birdsfoot trefoil; CMV, cicer milkvetch; MBG, meadow bromegrass; SEM, standard error of the mean; aNDF, neutral detergent fiber assayed with the addition of heat-stable alpha amylase; ADF, acid detergent fiber; TDN, total digestible nutrients; NFC, non-fibrous carbohydrates; CT, condensed tannins.

Fatty acid	Diet	Mean		SEM
Acetic	BFT	39.33	AB	3.68
	CMV	38.13	В	3.47
	MBG	47.78	А	4.36
	Feedlot	35.59	В	4.23
Propionic	BFT	8.50	В	0.86
	CMV	7.80	В	0.74
	MBG	9.11	В	0.88
	Feedlot	14.51	А	2.20
A:P	BFT	4.59	В	0.22
	CMV	4.91	AB	0.22
	MBG	5.24	А	0.22
	Feedlot	2.77	С	0.29

TABLE 2Rumen short-chain fatty acids (mM).

Note. BFT, birdsfoot trefoil; CMV, cicer milkvetch; MBG, meadow bromegrass; SEM,
standard error of the mean; A:P, ratio of acetic to propionic acid.

Fatty Acid	Period	Diet	Mean		SEM
C18:2 n6 linoleic	1	BFT	18.71	А	1.12
	1	CMV	18.04	А	1.08
	1	MBG	17.32	А	1.04
	1	Feedlot	31.94		
	2	BFT	21.51	А	1.80
	2	CMV	18.58	AB	1.55
	2	MBG	15.99	В	1.34
	2	Feedlot	31.42		
	3	BFT	21.36	А	1.47
	3	CMV	18.63	AB	1.29
	3	MBG	17.32	В	1.20
	3	Feedlot	31.72		
C18:3 n3 alpha-linolenic	1	BFT	47.17	В	1.72
	1	CMV	51.03	А	1.72
	1	MBG	49.91	AB	1.72
	1	Feedlot	2.43		
	2	BFT	46.91	В	2.01
	2	CMV	50.35	AB	2.01
	2	MBG	52.04	А	2.01
	2	Feedlot	2.47		
	3	BFT	45.84	В	2.02
	3	CMV	51.01	А	2.02
	3	MBG	45.83	В	2.02
	3	Feedlot	2.47		
n-6:n-3	1	BFT	0.41	А	0.04
	1	CMV	0.36	А	0.03
	1	MBG	0.37	А	0.03
	1	Feedlot	13.16		
	2	BFT	0.47	А	0.06
	2	CMV	0.38	AB	0.05
	2	MBG	0.32	В	0.04
	2	Feedlot	12.75		
	3	BFT	0.47	А	0.05
	3	CMV	0.37	А	0.04
	3	MBG	0.40	А	0.04
	3	Feedlot	12.87		

TABLE 3Dietary long-chain fatty acids

Fatty acid	Diet	Mean		SEM
C18:2 n6 linoleic	BFT	31.58	А	2.57
	CMV	23.48	В	1.91
	MB	18.46	С	1.48
	Feedlot	32.50	А	3.29
C18:3 n3 alpha-linolenic	BFT	11.52	А	1.25
	CMV	9.69	В	1.05
	MB	9.26	В	0.99
	Feedlot	1.97	С	0.24
n-6:n-3	BFT	2.69	В	1.05
	CMV	2.72	В	1.06
	MB	1.86	В	0.72
	Feedlot	9.39	А	3.92

TABLE 4 Blood plasma long-chain fatty acids

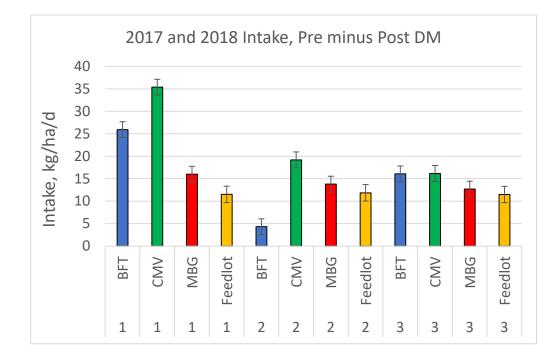
Fatty acid	Diet	Mean		SEM
C18:2 n6 linoleic	BFT	1.14	А	0.05
	CMV	0.99	В	0.04
	MB	0.89	В	0.03
	Feedlot	1.00	AB	0.06
C18:3 n3 alpha-linolenic	BFT	0.85	А	0.11
	CMV	0.58	В	0.08
	MB	0.54	В	0.07
	Feedlot	0.54	В	0.08
n-6:n-3	BFT	1.36	В	0.27
	CMV	1.81	А	0.36
	MB	1.68	AB	0.33
	Feedlot	1.77	AB	0.38

TABLE 5Subcutaneous fat long-chain fatty acids

FIGURES

Rep1	Rep 2	Rep 3	Rep 4	Rep 5
CM∨	BFT	CMV	MB	BTF
МВ	CM∨	BFT	BFT	CM∨
BFT	МВ	MB	CM∨	МВ

FIGURE 1 Plot design for pasture treatments.



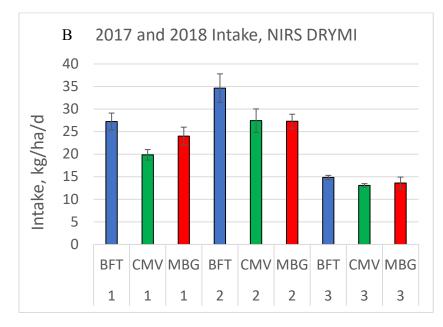
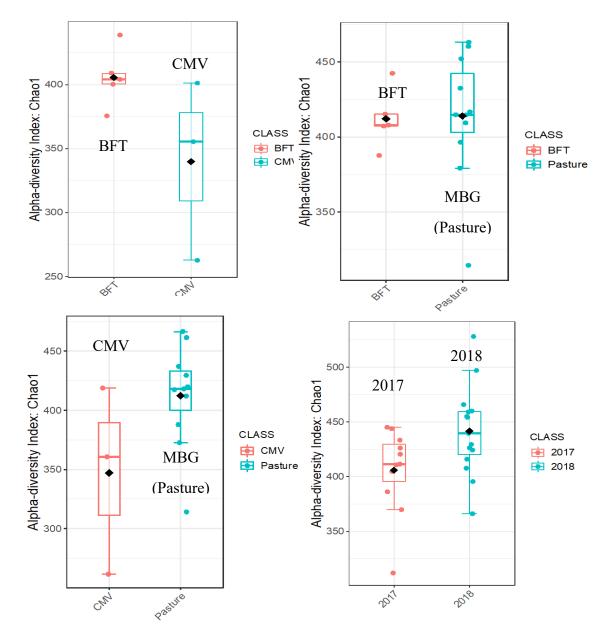
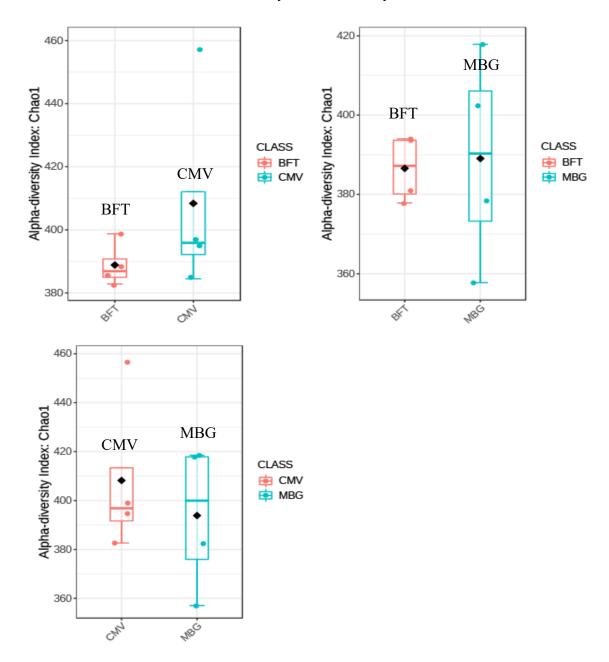


FIGURE 2 The intake of 2-year-old heifers determined from the difference between pre- and post-grazing forage DM for three different periods during the grazing season.



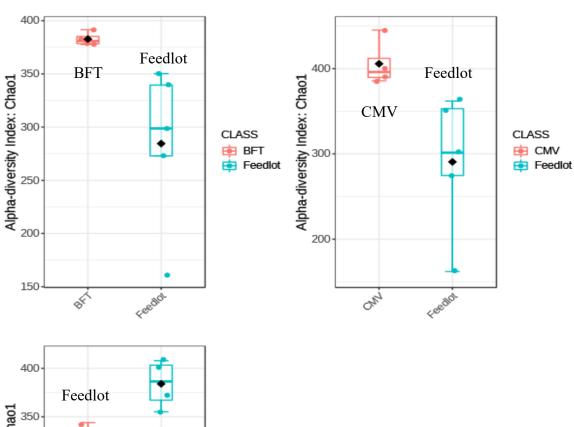
2017 Family-Level Diversity

FIGURE 3A Family-level within-treatment, α -diversity of the rumen bacterial microbiome. 2017 pairwise comparisons among pasture treatments BFT, CMV and MBG.



2018 Family-Level Diversity

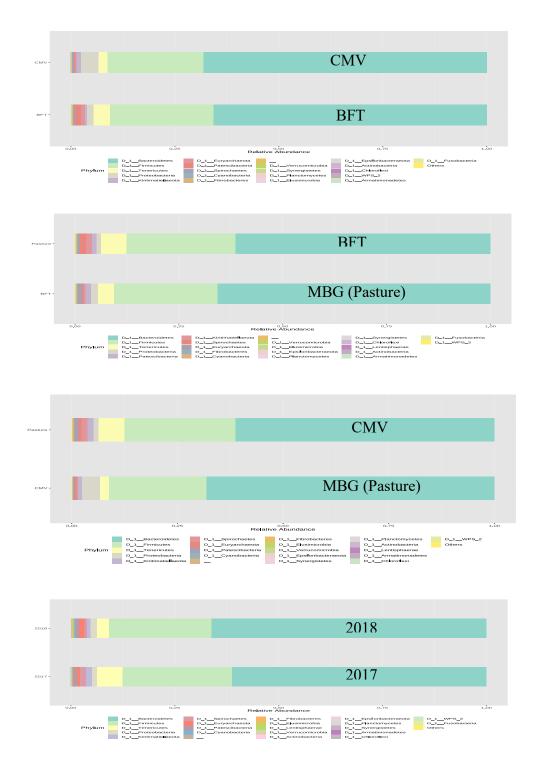
FIGURE 3B 2018 pairwise comparisons of pasture treatments.



2018 Family-Level Diversity

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FIGURE 3C Comparisons of each pasture treatment with the feedlot treatment for 2018.



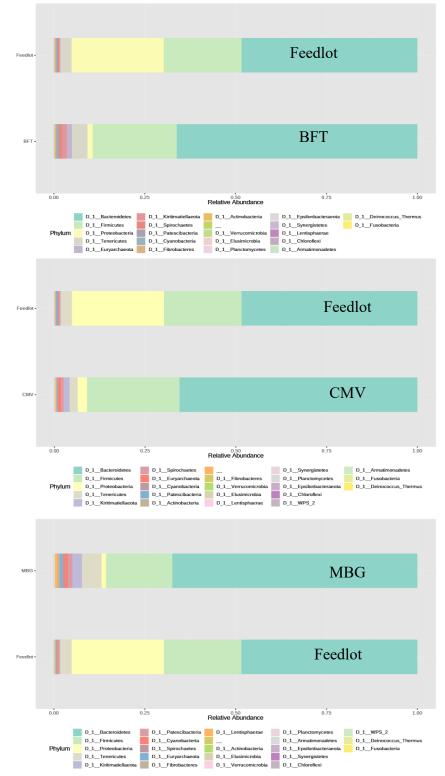
2017 Bacterial Phylum Abundance

FIGURE 4A Phylum abundance of rumen bacterial microbiome. 2017 pairwise comparisons among pasture treatments BFT, CMV and MBG.

2018 Bacterial Phylum Abundance



FIGURE 4B 2018 pairwise comparisons of pasture treatments.



2018 Bacterial Phylum Abundance

FIGURE 4C Comparisons of each pasture treatment with the feedlot treatment for 2018.

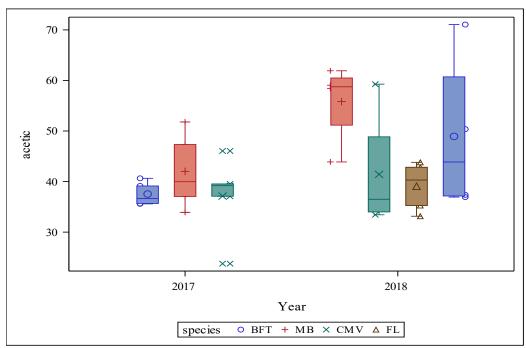
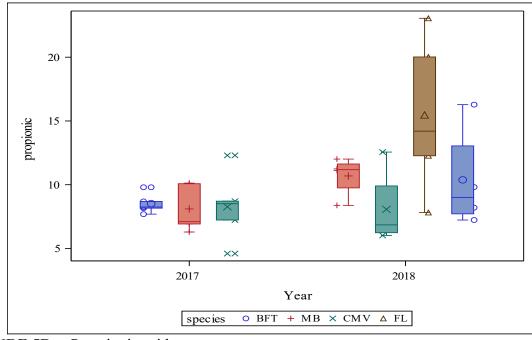


FIGURE 5A Statistical comparison of rumen production of select short-chain fatty acids by treatment and year. Acetic acid.





Rumen Short-Chain Fatty Acids

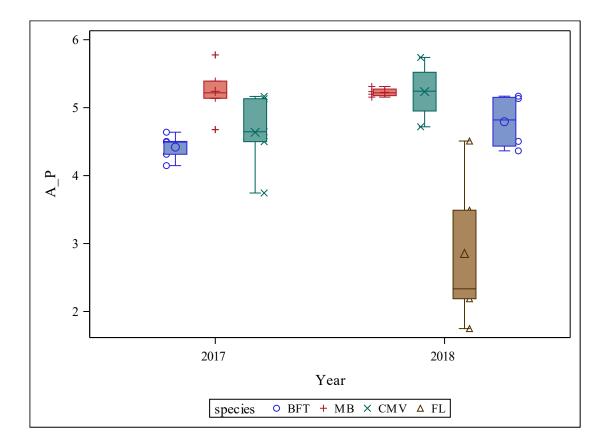


FIGURE 5C The ratio of acetic: propionic acid.

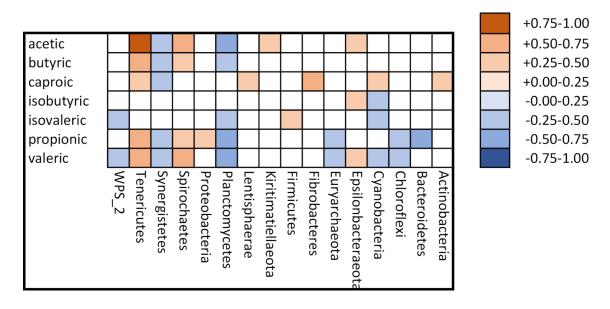


FIGURE 6 Significant positive and negative correlations between all rumen SCFA and bacterial phlya.

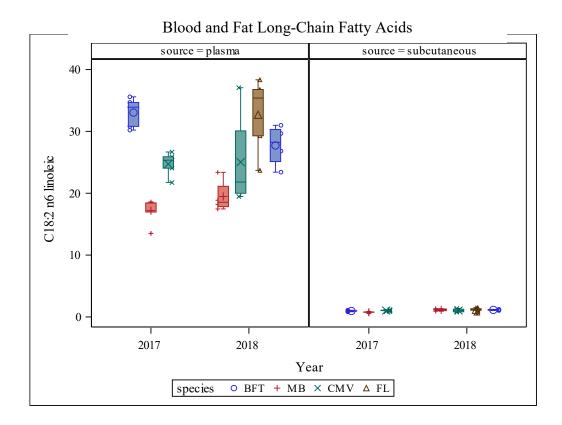


FIGURE 7A Statistical comparison of selected blood plasma and subcutaneous fatty acids. Linoleic acid.

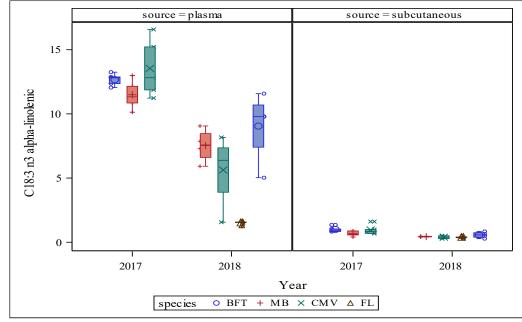


FIGURE 7B Alpha-linolenic acid.

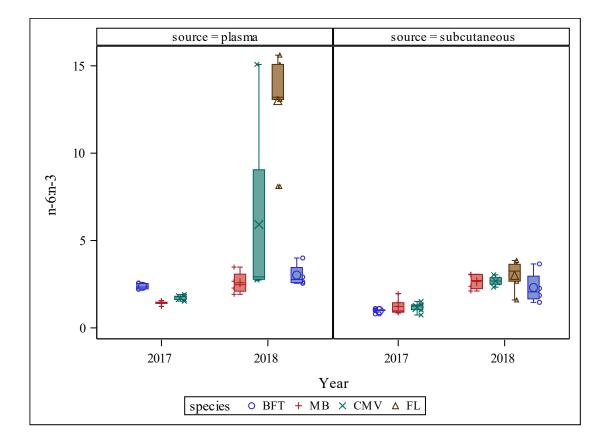


FIGURE 7C n-6: n-3, the ratio of the sum of omega-6 with the sum of omega-3 fatty acids.

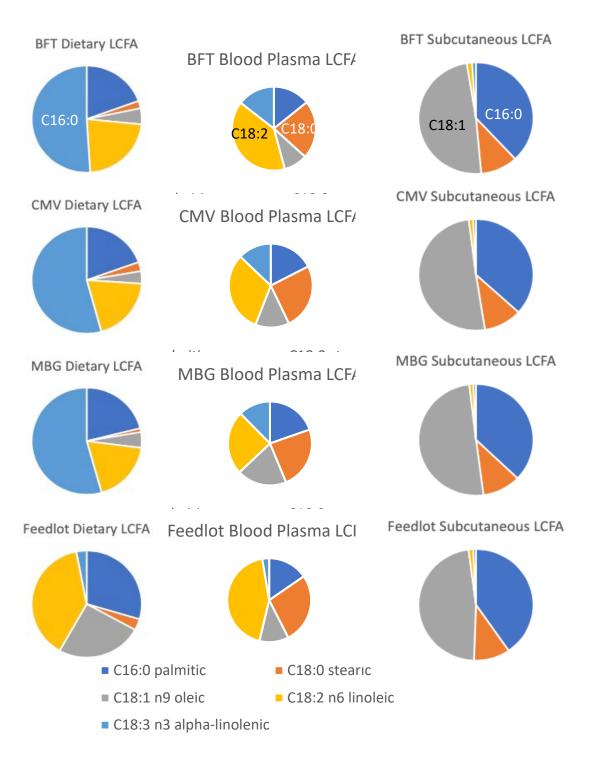


FIGURE 8 Comparison of the five most abundant LCFA in dietary, blood plasma, and subcutaneous fat samples to illustrate the transitions in fatty acids that occur in the rumen and during body fat deposition.

CHAPTER IV.

DEAF AGRICULTURE

Abstract

As noted in Chapters I and II, this chapter of the thesis explores accessibility of Extension videos for the Deaf and hard of hearing community. Culture varies from location to location, across ethnicities, and with similarly abled people. In the United State there are urban, suburban, and rural settings that add nuance to rich and diverse cultures. The Deaf community is an excellent example of these nuances. The difference between Deaf and deaf is that Deaf refers to identification within a cultural community, while deaf refers to an audiological status. Both terms will be used as appropriate throughout this chapter. Deaf culture is as much a part of rural America as urban and suburban cultures, but is undervalued, and as a result understudied. There is a lack of understanding when it comes to what Deaf culture looks like in the countryside of the United States. The goal of this chapter is to explore the available factual information and discuss how those facts influence the deaf and Deaf in agricultural settings day to day. This includes the probability and lifestyle of being deaf and Deaf in agricultural professions, what living an agricultural lifestyle looks like within this culture, resources available to Deaf children and adults in agricultural settings, and what can be improved.

It is important for rural communities, regardless of different cultural identities, to be able to work together. Production agriculture workers make up 2% of the United States workforce, and the entire United States is dependent on that 2% to survive and thrive in all fields of work and labor (USDA, 2020). Having all agricultural voices be heard is vitally important for the future of agriculture and promoting ecologically sound practices. It is the responsibility of all agricultural organizations to be willing to work with their deaf and Deaf cohorts for the present and future benefits of the agricultural community.

1 | INTRODUCTION

There have been a few studies on hearing diversity in U.S. childhood education, but nothing on Deaf adults in these settings (Luetke-Stahlman, 1995). Where no research has been done on Deaf demographics in rural areas, there are no accurate measurements of how many Deaf people are in the production agriculture profession. This is a problem caused in part by a lack of representation within agricultural agencies that are responsible for lobbying on behalf of production agriculture in legislative circles.

Conducting these studies is important because the data and understanding that would be gained from a study geared toward Deaf agriculturists would represent people who exist and who are not currently being represented in their agricultural organizations. The Deaf community should be represented by agricultural organizations from which they have been excluded, possibly unintentionally. These organizations would give them a political voice and lobbying power on their behalf. This power is an easily available resource to a hearing agriculturist but is only as accessible to a Deaf person as the Americans with Disabilities Act allows (University of Minnesota Duluth, 2020). While the Americans with Disabilities Act gives a lot of power to Deaf individuals to assert their rights, there are only two agricultural organizations in the United States that recognize Deaf farmers and ranchers even though there are hundreds of organizations in agriculture that would benefit from including Deaf agriculturists and would be able to offer tools and services to Deaf agriculturists (American Farm Bureau, 2018; National Institute of Food and Agriculture, 2019). As state and federally supported institutions, Cooperative Extension should be a model for inclusion but common resources such as Extension YouTube videos commonly are not accessible in any meaningful way to Deaf agriculturists.

People living in rural areas of the United States tend to struggle to find resources. One in five Americans live in a rural area, or about sixty million people (Kreis 2010; Ratcliffe et al., 2016). There are benefits and disadvantages to living relatively isolated from other people. Some disadvantages are a lack of employment opportunities, inadequate health care availability, and increased instances of drug abuse (Hamel et al., 2017). Ironically, living in a rural area increases the probability that an individual will live an overweight and inactive life, but this is because it is unrealistic or unsafe to walk or bike to shops or restaurants (Patterson et al., 2004; Paul, 2019). Benefits to living in rural America includes a slower lifestyle, cleaner air, a quieter environment and generally a lower cost of living.

Living in a rural area is not the same as living in an agricultural area. While the two factors tend to go together, many individuals in fly-over states are employed in local businesses and industries, Alternatively, urban agriculturists may carve out a small organic farm in more densely populated areas. Despite the variety of approaches to agriculture, there are gaps in the research done on rural demographics. In fact, sociologists debate whether "rural" is enough of a distinction to warrant specialized research (Tickamyer et al., 1990). Those in favor of studying rural areas separately from urban areas argue that specialized information about these areas would increase understanding of poverty dynamics because there are more people living in poverty in rural areas (Tickamyer et al., 1990).

Rural communities are diverse and it is outside the scope of this paper to discuss their characteristics, but it is important to understand the difficulties that are inherent to rural areas including lack of class mobility, lack of healthcare, and isolation. Feelings of community must be purposely sought in church and other community gatherings. It is not hard to imagine that if it is difficult for hearing people to build a sense of community in rural areas that it requires much more effort for a deaf agriculturist to feel a sense of community. However, Deaf people have been building their communities and culture for centuries despite adversity in doing so (Longmore et al., 1990). This shows the resilience and perseverance of individuals and the power and importance of community.

2 | DEAF CHILDREN IN RURAL AREAS

Although childhood education is not the focus point of this paper it is an important aspect of the Deaf experience, and childhood is an important time in the shaping of the identities of people who become agriculturists. Agricultural education and access for Deaf children would also enrich their experiences, including accessible outreach via 4H Extension. Having a mental picture and understanding of the lack of resources at an early stage of life can help Extension and 4H educators understand the need for additional research and resources for the adults in this niche group. In rural areas educational resources can be minimal, even for hearing people. There are additional factors that make obtaining an education as a Deaf child in a rural area even more challenging.

The only way that agricultural education, based on a Career Technical Education (CTE) program that includes Future Farmers of America (FFA), would be offered to Deaf children is through a mainstream setting. There are only three states that have an FFA and CTE program in the residential Deaf schools and those are Louisiana, Kentucky, and North Carolina (Career & Technical Education Center; Kentucky School for the Deaf; Sign of the Times). 4-H programs are an after-school program that are part of Cooperative Extension. There is a lot of flexibility within 4H programs for kids to discover their own interests and expound on those interests. As a result, there are a lot of 4-H programs that teach American Sign Language or topics related to agriculture. However, there are very few that are accessible to ASL users in agricultural topics. These accessibility hurdles can be mitigated with curriculum materials and interpreters if a mostly hearing culture make the effort to integrate Deaf children into the program.

For the well-being and success of the children in a mostly hearing setting it is important for them to feel socially included (Kersting, 1997). Where more than 90% of Deaf children are born to hearing parents, Deaf children tend to feel isolated from the world and their families (National Institute on Deafness and Other Communication Disorders, 2016). Children's success in any setting, including 4H and agricultural education settings, depends on feeling included with their peers and having Deaf role models that they can look up to (Abram & Gallegos, 2011). 4H programs could be a wonderful outlet for Deaf children and give them an agricultural education as well. In addition to making current 4H materials accessible, there is an opportunity to create a Deaf-centered curriculum in subjects other than communications.

These factors are important to consider because having exposure to agricultural careers and education could influence the level of consideration a Deaf person has for agricultural professions. Resources that are available at the childhood level will affect career options and preferences. While academics are important and necessary for occupational success, having exposure and education about various career options is important. Children learning that careers are just as diverse as themselves and seeing the potential that is available is important in establishing an identity and being successful enough for life satisfaction. It is the mission of 4H, FFA and Cooperative Extension to provide outreach and education. Because they are well-established as institutions across rural America, these are ideal organizations to model accessibility and inclusion to the individuals they interact with, setting the tone for private organizations like the Farm Bureau, state Cattlemen's Associations and other groups that are needed to lift Deaf agriculturalists' voices.

3 | DEAF INTERACTIONS WITH AGRICULTURAL ORGANIZATIONS

Combatting isolation and other challenges in rural agricultural communities for Deaf people is aided by organizations like the Farming Association for the Deaf, Hard of Hearing. This is an activist group that works with federal programs, mainly Agrability, which is a federal program that helps disabled agriculturists continue working on their farms (National AgrAbility Project, 2012). The National Agrability Project mission statement is 'to enable a high-quality lifestyle for farmers, ranchers, and other agricultural workers with disabilities, so that they, their families, and their communities continue to succeed in rural America.' While Agrability has resources for hearing impaired farmers such as hearing aids, they do not address the needs of farmers who are Deaf, and their approach is limited to altering or "improving" the state of deafness rather than increasing acceptance and accommodation of the Deaf community.

In an interview with the Farming Association for the Deaf, Hard of Hearing President, David Galyean, he expresses his frustration with Agrability and their failure to advocate for Deaf agriculturists, and he notes there are no Deaf agriculturists in Agrability leadership. When the author expressed to Mr. Galyean that the Utah Agrability chapter had no information on Deaf agriculturists in Utah, and asked if there were other Agrability agencies that the author could contact about Deaf agriculturists in different states he responded with the following (edited for clarity),

"Unfortunately, they [Agrability] do not help us as much as we wanted them to, but hopefully one day they realize our perspective like I told them one day. What if a big, bad war comes and everyone is sent off to war. Is then a good time to train all of us deaffies to work in farm fields feeding both civilians and soldiers. They said I have a great idea, but no money to support my system... Bull!!!! But I am not going to waste my breath, until the time it does happen then they will wake up realizing what I had been trying to tell them in the first place." (D. Galyean, personal communication, November 1, 2018) Mr. Galyean's frustration is understandable. Basic demands that should be put into action are being ignored. Just the simple request to allocate funds to study how many deaf agriculturists there are has been ignored. The Farming Association for the Deaf, Hard of Hearing has three main purposes in their agenda, which will be directly quoted from Mr. Galyean.

"We need them to 1) Gather all folks to write up a grant mainly for deaf farmers to receive land either through the homestead act, or a land grant, and providing extensive training programs. That way a majority of us would be employed instead of relying on government handouts. 2) Gather a few deaf people to be selected as board member on Agrability's diversity panel to make several recommendations, etc. 3) Allowing us, board members, to travel around and interview deaf and hard of hearing farmers that are out there. We can gather information and data proving to Agrability that these are useful resources."

Listening to people like Mr. Galyean is important because his perspective can change things positively for all agriculturists. Late onset hearing loss is common in agriculturists (Ehlers 2011). Loud equipment, the increasing median age of farmers, and high likelihood of accidents means 92% of agriculturists will experience a loss of hearing (McBride et al., 2003). It is incredibly common, but not accommodated for. Political groups such as crop coalitions, the Farm Bureau, and others do not make accommodations for hard of hearing or deaf persons. There are no interpreters, no captions, and the only aid is a microphone. There needs to be a shift to explaining concepts visually and with large printed text for the aging population of agriculturists regardless of whether the organization wants to be inclusive of Deaf people or not. To meet the needs of the constituents of their groups they need to be accommodating and understanding of the needs of all the people that make up their organizations. Even though the niche group of Deaf agriculturists is small, agriculture in general is getting older and working in environments that can result in hearing loss (Ehlers, 2011). Adjusting the emphasis of presentation to be visually based would help others in addition to Deaf agriculturists, such as the aging and late-deafened agriculturist population that includes many farmers and ranchers today.

While other groups of agriculturists would benefit from Deaf accessibility it should not distract from how essential accessibility is to Deaf individuals. Accessibility to information changes everything. It allows an individual agriculturist to make informed and educated changes on their operations and to contribute their experience to the larger agricultural community. Information and education for adults in agriculture can change watershed quality, economic prosperity, and ecological soundness. When these aspects of agricultural production are similarly accessible for all agriculturists, it not only affects the agriculture community, it affects the entire population. It affects quality of life for every single person in the United States. The problem has become acute as more educational materials are disseminated through YouTube.

Resources are the primary element needed for success and equal opportunity. In today's farming economy it is more challenging to be a primary owner and operator of a family farm. Land is expensive, so it is difficult to start a farm without an inheritance or a loan. Agricultural loans can be issued at a reduced interest rate compared to a normal loan. Starting a farm is hard and economies of scale increasingly favor large farms. In most situations this means most agricultural workers are hired by large farming companies. There is no way to know for sure how Deaf agriculture interacts with these dynamics because no studies have been done to quantify the proportion of hired Deaf agriculturists to self-employed Deaf agriculturists (USDA ERS, 2019).

Resources available to farmers differ depending on economics, geographic location, types of crop planted, climate conditions, policy, and market shifts. Some examples are crop failure insurance, crop subsidies, Agrability, and water conservation programs. While all these organizations are compelled by the ADA to provide interpreters at events where they know a Deaf person will be present, there is the question of how Deaf agriculturists can find out about these programs in order to attend and make their presence known beforehand. For Farm Bureau programs, people often find out about offered programs by word of mouth. Farm Bureau prides itself on its grassroots focus, but this focus can create an exclusive club that limits membership to friends and people who interact with each other regularly. Farm Bureau offers a lot of political power and tools for agriculturists, but those tools would be more beneficial if more people could access and learn about them. Farm Bureau would benefit by having all their programs spelled out on their website. Conferences are another location where Farm Bureau members can learn about programs like estate planning, but conference presentations usually include few or no visual aids.

Agrability is a federal program that provides practical assistance to farmers with varying types and levels of disability. It would make sense for Agrability to have a minimum of one Deaf agriculturist on its advisory board but there is no representation for deaf farmers in Agrability. (D. Galyean, personal communication, November 1, 2018) A person could assume that this would mean that there are no Deaf agriculturists, or so few that representation is not important. While there may not be many Deaf agriculturist, the exact number is unknown due to lack of research and research funding. There are enough Deaf agriculturists to validate at least a minimal amount of investigation and there is no excuse for Agrability not recognizing and incorporating Deaf agriculturists as stakeholders even though advocates have made the request.

To counter the lack of support from Agragility, 4H, FFA or Cooperative Extension, there are four resources for Deaf agriculturists. First is the Farming Association for the Deaf, Hard of Hearing, an organization with two hundred and sixtyseven members. Second is YouTube and other social media videos created for Deaf agriculturalists, including the stories of six vastly different agricultural operations in six different parts of the United States. Third is the three different Future Farmers of America (FFA) chapters that are associated with schools for the deaf in North Carolina, Louisiana, and Kentucky (Career & Technical Education Center, Kentucky School for Deaf, Sign of the Times, respectively). Agrability is another resource that is designed to aid disabled agriculturists so they can continue farming. Agrability has a lot of resources for deaf agriculturists who would like to receive hearing aids and similar resources. They leave a gap for Deaf agriculturists who see their Deafness as a part of their identity and not as a disability. More resources need to be developed for this niche group of people, and to encourage more talented, creative people to become agriculturists. The small minority group that already exists deserves to be adequately represented in farming organizations and provided with accessible resources.

Land Grant universities are mandated to make educational materials available to agriculturists (National Resources Council, 1995). Extension resources created across all types of media disperse important continuing education material that can increase land and livestock productivity and sustainability in a world being driven by anthropomorphic changes. Most Land Grant extension publications are readily available to people who can read and understand English because they are written out in the form of peer reviewed bulletins and fact sheets, and all written resources have been accessible on the internet. However, extension educational videos that rely on automated captioning are not welldesigned to be accessible except to hearing people because auto captioning is not reliable in conveying the message of the video. There are several ways to make extension videos and presentations more accessible to a larger variety of people, many of which do not take a lot of time.

4 | MEDIA ACCESSIBILITY

Being able to access information from credible sources is vital for effective Extension education. There are several resources on how to create media that is accessible to everyone, but they differ depending on the media being used. Some typical resources for disseminating information are videos, face to face presentations, and websites. While accessibility looks different for each medium there is a consistent element underlying all of them, which is to put oneself into the shoes of the audience the presenter is trying to reach. If materials produced are meant to reach Deaf people, the creator can interact with those materials like they are a Deaf individual by reviewing the material without audio. This goes for all messages regardless of the audience that the educator is wishing to reach.

At the very least, accessible videos need to include accurate captions. It is tempting to let automatic captioning services take over because the alternative is a paid captioning service while YouTube provides auto captioning for free and it requires no thought on the part of the creator. Unfortunately, those subtitles will vary in accuracy depending on the quality of the sound. At best, automatic captioning is only 70% accurate (University of Minnesota Duluth, 2020). Automatic captioning rarely includes punctuation, which is an important element for comprehension. There are also options for allowing content consumers to add subtitles in multiple languages. However, to ensure accurate messages, people creating content should not rely on others to interpret the message of their presentation. An American Sign Language (ASL) interpreter can also be helpful in some videos, especially when the message is complicated.

Face to face presentations will require an interpreter or a live captioning system. Working with an interpreter is not as simple as just plowing through the planned presentation and is benefited greatly by the preparation of a script of the expected presentation. American Sign Language has a different grammatical structure than English and some statements may take longer to translate than others. Do not over enunciate, speak slowly without overexaggerating, and take time to pause. This will create a natural flow to the presentation, while allowing time for the information being presented to be interpreted. It is also helpful to limit the amount of words and to clearly define jargon. Jargon terms will typically need to be finger spelled and taking time to define those terms will make a big difference in the level of understanding the audience can accomplish (Department of Workforce Services, 2020).

Any message worth conveying accurately should be thought through beforehand, even if it impinges on the spontaneity of the presentation. Some Deaf viewers can lipread, but only if the presenter is turned toward the camera and fully visible. Even for Deaf people who are good at lip reading, the accuracy of lip-reading is about 52%, so it is important to consider lip reading as a supplemental communication tool rather than an assumed skill on the part of the Deaf (Lavars, 2017). Accurate captions or a trained interpreter need to be included to make educational information accessible.

This chapter includes videos exemplifying elements of accessibility for the Deaf as well as scripts for the videos (below):

5 | VIDEO SCRIPTS

Britney Allen Grass-Legume Storyboard https://drive.google.com/file/d/1wsmZ4ogsRTDZ3phhFG2aEIpq4REc6Zc/view?usp=sharing

Title	Benefits of Grass-Legume mixtures
Audio	Visual
Irrigated pastures	Video of pasture
typically have plenty of	
nitrogen to encourage	
beef cattle gain, but not	
enough energy. This can	
be due to limited intake	
because of a high	
proportion of fiber	

relative to available	
nutrients. ²	
Grasses have more fiber	Define legumes
than legumes (a	Define legunies
flowering plant in the	
pea family. Legumes	
that are	
Cultivated (grown with	Define cultivated
human care) in the	
Intermountain West have	
more available energy	
and less fiber than the	
same plant species	
cultivated in other parts	
of the U.S. We think this	
is due to long, hot sunny	
days and cool night	
temperatures that	
maximize the	
accumulation of sugars	
and slow the rate of loss	
of these sugars at night.	
Legumes are high in	
protein, but Mountain	
West legumes are also	
high in non-fiber	
carbohydrates that can	
be used for energy.	
Grazing livestock on	
grass-legume mixtures in	
the Intermountain West	
can have multiple	
benefits. ¹	
Some of those benefits	Overlayed over previous pasture footage
are	
1. Legumes can	
supply nitrogen	
to pasture	
grasses.	
2. Legumes	
increase ruminant	Define ruminant then refer to previous slide.
	1

(an animal with a 4 chambered stomach) intake.

3. Legumes provide additional energy to support daily gain or milk production.

Legumes are nitrogen fixing. While N₂ is abundant in the atmosphere, plants are not able to use it in that form. Legumes have a relationship with soil bacteria that take up residence inside nodules on the root. Protected and fed by the plant, the bacteria change atmospheric nitrogen into a form that can be used by the plant to make protein.^{3,4}

Some nitrogen is transferred to nonnitrogen fixating plants, such as from decomposing leaves and roots, and more is transferred in the form of waste from grazing ruminants. This nitrogen increases soil health and pasture production.⁵



https://biology.anu.edu.au/news-events/news/nodulationlegumes

82

Legumes have more energy and less fiber, or bulk, than grasses. ⁸ Ruminants prefer legumes over grass by 70%. ⁹ Mixed legume- grass pastures help increase the amount that a ruminant can eat.	Photo has Salar Carder
XX71 1 '	Photo by Sabra Gerdes
When some dairy cows grazed birdsfoot trefoil pastures and others grazed grass pastures, cheese from the milk of cows grazing legume pastures gave 20% more milk ⁶ , and their milk contained more omega-3 fatty acids than cheese from the milk of cows grazing grass pastures, and both were higher than cheese from the milk of cows fed a	This Photo by Unknown Author is licensed under <u>CC BY-SA</u>
conventional	
total mixed ration (a mixture of forages, grains, protein supplements, byproducts, vitamins, and minerals).	Define total mixed ration
Grazing legumes or	Video of Britney talking
mixed legume-grass	
pastures have been found	
to improve the	
productivity of	
ruminants and to	
improve the nutritive	

value of milk and milk	
products.	

Citations for description

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Britney Allen Extension Video Storyboard https://drive.google.com/file/d/1GzGM5_hQmpm58OEW4G0u2-Lus9134IH5/view?usp=sharing

Title	Increasing Accessibility in Extension Materials
Audio	Video
This is a video on how to make extension materials accessible for audiences with a variety of hearing levels.	Title screen
We will be covering three different points	Organization of video
on how to make your presentation straight forward and understandable. Those three	Visual presentation aids
topics address visual presentation aids,	Captions
captions, and interpreters.	Interpreters
There are several different ways that you can have good visual aids in your extension materials. One is an actual demonstration. Here is a good example of that.	Kevin Heaton Demonstration Video
Another way to incorporate visuals is with pictures. This is a visual showing the utilization of a plant and the text below each drawing provides the response of the roots to different amounts of shoot utilization. Make sure that there is time, for the viewer to read the text on the slide, then add the captions and leave time for additional discussion.	USU Range Extension Picture
Another important factor when creating a presentation is to avoid complicated tables or charts, unless you point out exactly what	Picture of flow chart.

you're using from the table by highlighting	
and explaining it.	
This is an example of a visual that's impossible to understand without a visual aid as well as captioning to provide context.	
One way that this table could be made easier to understand is by taking the parts that are important to the discussion and adding an order to them. The order could be chronological, hierarchy of importance, or organizational.	
Difficult to read tables and charts have no place in your presentation and will distract everyone in the audience from your overall message. If you're going to use a complicated table, take the time to make the information you're using stand out so the caption can be brief.	
1	
In some cases, it may be desirable to use an interpreter for your presentation.	Interpreters
In some cases, it may be desirable to use an	Interpreters Gallaudet video

It's an important thing when presenting with an interpreter to remember that American Sign Language (ASL) and English are two different languages with different grammatical formats. The interpreter for your presentation may need time to finish interpreting what you've said due to differences in ASL grammatical structure or the need to spell out a name or technical term.	
Another important part of videos for deaf and hard of hearing individuals is the captions. This next video, unfortunately, does not have the best captions. Right off the bat, we see that there is no punctuation. We also see that instead of 'hive' the caption says 'hi' creating a distraction. The captions only make sense if you can hear the speaker as well as read the captions. This is a common issue when auto captioning is applied to a YouTube video.	USU Bee video
If we were to mute this video and continue from this point, what would we be able to understand? So, Monday's no good for some reason. A movie is going pull one of the frames. It isn't clear what we're supposed to be noticing on top. When a presenter isn't pointing at something but there are no captions that is hard to understand. Right now, a lot is being said that we're just not getting.	
The most helpful tip I can give you when it comes to making your video is to watch your video without the sound.	Helpful Tip slide.
I hope this video was helpful and I think the most important take away is that to be inclusive is not a hard thing it just requires a little extra thought.	Head shot of Britney talking

Video Description information

Citations

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More Resources

Interpreters (Utah only)

https://jobs.utah.gov/usor/uip/directory.html

Presentation suggestions

https://disabilitynavigator.org/article/12329/tips-communicating-deaf-and-hard-hearing-people%20%20

https://www.washington.edu/doit/how-can-you-make-your-presentation-accessible

6 | CONCLUSION

The lack of availability of research on Deaf and hard of hearing farmers should be addressed in support of all the services needed by this community, but in particular to increase awareness of the need for accessible video design, the subject of this chapter. The three main needed for the accommodation of the Deaf community by agriculture are 1) have adequate representation in all organizations, both public and private; 2) have material that is universally accessible; and 3) provide funding to better understand what embodies Deaf agriculture. Overall, agricultural organizations have not done what needs to be done to be inclusive of Deaf perspectives, and it shows in the presentation of their materials or lack of materials.

Agrability should provide funding to determine how many deaf agriculturists there are, how many are self-employed, and what crops and livestock they raise. Agricultural organizations should make meetings and presentations accessible for all members, including members of the Deaf community. Land Grant universities should make all media as accessible to members of the Deaf community as to hearing agriculturists.

The implications of fatty acid ratios and the changes that happen in regard to feed type could help all agriculturists in the cattle industry in having data to support a marketing change that would be better for human health and the environment. While there was not enough time during the span of this project to make a video that directly addresses the content material of this thesis, a video specifically addressing the differences between BFT, grass, and feedlot finished beef can serve as an example of accessible media. Ensuring that this information is available to all agriculturists is vital in creating widespread change throughout the industry. Extension's role in distributing educational material for adults will be foundational in this endeavor.

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CHAPTER V.

SUMMARY

The implications of this study give agriculturists more options to manage their beef operations. The human health implications and environmental benefits of finishing beef cattle on BFT pastures gives producers a unique way to market their beef that potentially falls under American Grassfed Association's grassfed certification and the USDA organic certification. Providing opportunities for agriculturists to make choices that could benefit the overall health of people, reduce the environmental impacts of raising cattle, and still benefit the agriculturist's economic situation is an important aspect of the mission of Land Grant Universities and the Cooperative Extension Service. Information that can have an impact on agriculturists needs to be communicated to all agriculturists, regardless of their hearing status. Good scientific communication includes understanding and valuing all stakeholders. If audience accessibility to information is not being considered, then it is not good scientific communication.

Because of the importance of this study and the necessity to reach all farmers and ranchers, results associated with this study were used to create Extension media accessible to Deaf and hard-of-hearing producers whose operations could benefit from the results. This is information that could be quickly implemented by agriculturists who already operate using irrigated pasture systems in the Intermountain West. Other ways to initially communicate the results of the study would be as a written Extension publication, but video can be more compelling. In areas where the information could be useful to a larger group, Extension agents could organize and carry out local workshops,

but videos achieve the inherently visual aspects of such demonstrations. For all parts of this educational outreach, tools improving accessibility should and can easily be implemented.