HERBAGE CHARACTERISTICS AFFECTING INTAKE BY DAIRY HEIFERS GRAZING GRASS-MONOCULTURE AND GRASS-BIRDSFOOT TREFOIL PASTURES

by

Marcus F. Rose

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Approved:

J. Earl Creech, Ph.D. Major Professor	Blair L. Waldron, Ph.D. Committee Member
Michael D. Peel, Ph.D. Committee Member	S. Clay Isom, Ph.D. Committee Member
Kara Thornton-Kurth, Ph.D. Committee Member	Richard S. Inouye, Ph.D. Vice Provost for Graduate Studies

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ABSTRACT

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by

Marcus F. Rose, Master of Science

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Major Professor: Dr. J. Earl Creech
Department: Plant Soils and Climate

Performance of dairy cattle on pasture is often reduced when compared to conventional dairy operations. The reduced performance in grazing dairy animals is often due to reduced dry matter intake and energy, which are the major limiting factors in grazing animal performance. We hypothesized that high-energy grasses coupled with the low levels of condensed tannins in birdsfoot trefoil would complement one another to improve heifer dry matter intake and performance. Jersey heifers were rotationally grazed each year for 105 days in 2017 and 2018 on eight different pasture treatments, which included perennial ryegrass (*Lolium perenne* L.; PR), orchardgrass (*Dactylis glomerata* L.; OG), meadow bromegrass (*Bromus biebersteinii* Roem. & Schult.; MB), and tall fescue (Schedonorus arundinaceus Schreb.; TF), with each respective grass also mixed with birdsfoot trefoil (*Lotus corniculatus* L; BFT). Apparent dry matter intake was measured as the difference between pre- and post-grazing herbage mass for each seven-day grazing period, and analyzed as a randomized complete block design. Dry matter intake was from greatest to least as follows: MB+BFT ≥ OG+BFT ≥ OG ≥ MB ≥ PR+BFT > TF+BFT = PR = TF ($p=0.05$). Principal component analysis showed that physical herbage characteristics such as bulk density, height, herbage allowance, leaf pubescence, leaf softness, and birdsfoot trefoil content as well as nutritive properties such as fat, non-fibrous carbohydrates (NFC), neutral detergent fiber (NDF), acid detergent fiber (ADF), metabolizable energy and net energy for gain had important associations with intake. Crude protein and ash were also somewhat associated with intake. PR+BFT, the treatment with the most energy and tannins, had increased intake over PR in all analyses that were performed, suggesting that high energy in the grass interacted with tannins to improve heifer intake. However, other treatments had greater overall intake, and many herbage characteristics were associated with intake. The fact that both physical and chemical herbage characteristics were associated with intake shows the importance of planting the right species in pasture as well as making proper management decisions to maximize nutritive value and herbage intake.

(110 pages)

PUBLIC ABSTRACT

HERBAGE CHARACTERISTICS AFFECTING INTAKE BY DAIRY HEIFERS GRAZING GRASS-MONOCULTURE AND GRASS-BIRDSFOOT TREFOIL PASTURES

Marcus F. Rose

Pasture-based dairies have become more prevalent in recent years due to a higher proportion of organic milk demand and production. Organic certification requires that animals must graze at least 120 days in each growing season. However, dry matter intake is often limited when dairy animals receive most of their herbage from pasture, resulting in lower animal performance and milk production. The purpose of this study was to analyze the complimentary effect of high energy grasses with birdsfoot trefoil (BFT) tannins to improve intake of dairy heifers. Jersey heifers were rotationally grazed for 105 days in 2017 and 2018 on eight different pasture treatments, which included monocultures of perennial ryegrass (PR), orchardgrass (OG), meadow bromegrass (MB), and tall fescue (TF), with each respective grass also planted in mixture with BFT. Intake was measured by sampling herbage before and after each seven-day grazing period and was from greatest to least as follows: MB+BFT, OG+BFT, OG, MB, PR+BFT, TF+BFT, PR, TF. Physical characteristics such as pasture bulk density, herbage height, herbage allowance, leaf pubescence, leaf softness, and birdsfoot trefoil content as well as nutritional properties such as fat, non-fibrous carbohydrates, fiber, and energy were all

associated with intake. Crude protein and ash were also associated with intake. While PR+BFT did not have the greatest overall intake, it was the only treatment that consistently had greater intake than its respective grass monoculture (PR). Since it had more energy and tannins than all other grasses, a complimentary effect between energy and tannins to increase intake was likely. The fact that both physical and chemical herbage characteristics were associated with intake shows the importance of planting the right species in pasture as well as making proper management decisions to maximize nutritive value and herbage intake.

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INTRODUCTION AND OBJECTIVES

Introduction

With over 3.5 million milk cows in the Western United States, dairy is an important sector of the region's agriculture (USDA-NASS, 2017). Organic milk production has grown over the last 10-20 years and some producers have transitioned to organic milk production to take advantage of higher milk prices and better economic returns (USDA-ERS, 2018). Along with organic milk production come requirements that producers must allow their animals to graze at least 120 days per year, making them largely pasture-based operations during the growing season (USDA-AMS). However, pasture-based organic dairy production is not without its challenges. Dairies in which 75-100% of forage intake is pasture-based experienced a 32% decrease in milk production and a $10.36 decrease in net return when compared to those that used 0-24% grazing (William D McBride, 2010).

Senft et al., (1987) stated that two opposing problems must be resolved to maximize large herbivore diet selection: maximizing forage quality while maintaining adequate forage quantity. Related to these problems, dry matter intake (DMI) and/or dietary energy are two of the most important, and often the most limiting factors in high producing milk cows and beef steers on pasture (M. S. Allen, 2000; F. Bargo, Muller, Kolver, & Delahoy, 2003; Kolver & Muller, 1998; Leaver, 1985; Blair L. Waldron et al., 2019). By increasing energy and DMI on pasture, animal performance could be increased

A possible tool to increase animal DMI and performance is the use of high sugar grasses. In recent years, interest in breeding grasses with elevated water soluble carbohydrates (WSC) or "high sugar" grasses has grown (Smith, Stewart, & Spangenberg, 2007). Some of the biggest advantages of these grasses is that the increased WSC content leads to more efficient digestibility and increased metabolizable energy (ME) levels (Edwards, Parsons, Rasmussen, & Bryant, 2007; Miller et al., 2001; Smith et al., 2007; Waghorn, 2007). When compared to conventional cultivars, some high sugar perennial ryegrass varieties have also shown increased DMI in dairy and beef cattle (Lee et al., 2002; Moorby, Evans, Scollan, MacRae, & Theodorou, 2006). The increase in DMI in these cases could be at least partially explained by an increased rumen degradation rate, leading to reduced feed retention time and less limitations on intake (Miller et al., 2001). Another partial explanation could be a slightly higher dry matter content in high sugar grasses, suggesting that rumen fill would be less limited by moisture content in feed (Lee et al., 2002; Miller et al., 2001; Moorby et al., 2006). Water soluble carbohydrate levels of perennial ryegrass varieties have shown large fluctuations depending on the geographic location, time of year, soil moisture content, night temperatures, and/or day length and temperature (G. Cosgrove et al., 2007; G. P. Cosgrove, Mapp, Taylor, Harvey, & Knowler, 2014; Parsons et al., 2004; J. G. Robins & Lovatt, 2016). However, with few exceptions, high sugar grasses, especially high sugar orchardgrass varieties, have not been extensively studied in the irrigated pastures of the intermountain United States (J. G. Robins & Lovatt, 2016).

Another possible way to improve pasture nutritive value, herbage production and DMI is to utilize grass-legume mixtures. Previous research has shown that grass-legume mixtures often have higher crude protein (CP) and digestion (IVDMD) as well as lower neutral detergent fiber (NDF) and acid detergent fiber (ADF) in comparison to grass monocultures (Sleugh, Moore, George, & Brummer, 2000). These more favorable nutritive characteristics of grass-legume mixtures can potentially increase intake and performance of grazing animals.

Birdsfoot trefoil (BFT); *Lotus corniculatus*) can be a valuable legume in mixed pasture to improve animal intake and performance. Birdsfoot trefoil is a tannin-containing, non-bloating legume. Bloat is prevented when tannins bind to proteins released from plant mesophyll cells during mastication and thus reduce the activity of bacteria that create bloat-causing froth in the rumen (Min, Attwood, McNabb, Molan, & Barry, 2005; B. Min, T. Barry, G. Attwood, & W. McNabb, 2003). The binding of condensed tannins to plant proteins has also been shown to increase passage of undegraded dietary protein (UDP) in the rumen, leading to increased protein uptake in the small intestine of the ruminant and increased animal performance (Piluzza, Sulas, & Bullitta, 2014). Although tannins are often considered a forage anti-quality, low concentrations (5-40 g kg^{-1}) like those found in BFT have been shown to increase animal performance without affecting voluntary intake (Barry & McNabb, 1999; Hoveland et al., 1981; Ramírez-Restrepo & Barry, 2005).

Cows that graze BFT monoculture pastures have shown higher intake and milk production when compared to animals on grass pastures (Harris, Clark, & Laboyrie, 1998; Macadam et al., 2015; Woodward, Laboyrie, & Jansen, 2000) and BFT silage and hay in dairy feed rations increased milk production (Christensen et al., 2015; Hymes-Fecht, Broderick, Muck, & Grabber, 2013). Other studies reported increased beef steer growth performance from tall fescue-BFT mixtures in comparison to tall fescue monocultures (L Wen et al., 2002). In an initial study leading to this research, Waldron et al., (2019) reported that while tall fescue-BFT pastures did not always have the most herbage mass, they did result in the greatest average daily gains (ADG) of beef steers when compared to tall fescue monocultures and tall fescue-alfalfa mixtures.

Research Objectives

Previous research shows that high sugar grasses and BFT monocultures have potential to increase DMI and/or animal performance. However, these high energy grasses planted *in mixture* with BFT have not been extensively studied. Therefore, this study looked at the potential to increase DMI and performance of dairy heifers by grazing mixtures of high energy grasses and the tannin-containing legume BFT.

Specific objectives were to:

1) Analyze the complimentary effect of high grass-energy concentrations combined with the low levels of condensed tannins in BFT to improve dairy heifer DMI when grazing mixed grass-BFT pastures.

2) Compare herbage production and nutritive value of grass monocultures and grass-BFT mixtures.

3) Determine which herbage nutritive value traits have the biggest influence on dairy heifer DMI.

LITERATURE REVIEW

Grass-Legume Mixtures

Grass-legume mixtures can be a valuable component in a grazing system. Sleugh et al., (2000) report that mixing cool-season grasses with legumes resulted in greater herbage mass, crude protein (CP), and in vitro dry matter digestion (IVDMD) when compared to grass monocultures alone. Additional research has shown that grass-legume mixtures can also have less neutral detergent fiber (NDF) than grass monocultures, more consistent herbage mass during hot summer months, and as much forage as a grass monoculture fertilized with N at 134 kg ha^{-1} (Cox et al., 2017). In a mechanically harvested study conducted in Wisconsin, birdsfoot trefoil and Kura clover mixtures had greater potential milk production per kilogram of dry matter than all N-fertilized grass monocultures and matched potential milk production per hectare of orchardgrass fertilized with 336 kg N ha^{-1} (Zemenchik, Albrecht, & Shaver, 2002). A previous Utah study found that tall fescue-legume mixture herbage mass was less than N fertilized tall fescue monocultures, but livestock gains were greater (Blair L. Waldron et al., 2019).

Soder et al., (2006) found that complex mixtures made up of more than two species do not necessarily increase intake and performance of dairy cows compared to simple grass-legume mixtures. Pembleton et al., (2016) suggest that the nutritive density of the ingested forages, rather than the forage diversity, ultimately determines dairy cow production and milk yield.

One challenge associated with grass legume mixtures is that one species is often preferred over the other (Rutter, 2006; Lian Wen et al., 2004). Planting compatible species and careful grazing management must be implemented in order to maintain a desirable mixture in these situations.

High Sugar Grasses

In recent years, interest in breeding grasses with elevated water soluble carbohydrates (WSC) or "high sugar" grasses has grown (Smith et al., 2007). One of the biggest advantages of these grasses is that the increased WSC content leads to higher digestibility and higher metabolizable energy (ME) levels (Smith et al., 2007; Waghorn, 2007). When compared to conventional cultivars, some high sugar perennial ryegrass varieties have also shown increased DMI (Lee et al., 2002; Moorby et al., 2006). The increase in DMI in these cases could be at least partially explained by an increased rumen degradation rate, leading to reduced feed retention time and increased intake (Miller et al., 2001). Another explanation could be the slightly greater dry matter content in high sugar grasses, which could lead to less distention in the rumen due to moisture content (Lee et al., 2002; Miller et al., 2001; Moorby et al., 2006). Research also shows that high sugar grasses have improved ruminal balance of carbon and nitrogen supply, leading to more efficient digestion in ruminants (Edwards et al., 2007; Miller et al., 2001).

Water soluble carbohydrate levels of perennial ryegrass varieties have shown large fluctuations depending on the geographic location, time of year, night temperatures, and/or day length and temperature (G. Cosgrove et al., 2007; G. P. Cosgrove et al., 2014; Parsons et al., 2004). High sugar grasses, especially high sugar orchardgrass varieties, have not been extensively studied in irrigated intermountain U.S grazing studies.

Pasture Grasses

Tall fescue (*Schedonorus arundinaceus* Schreb.) is a very important and well-known pasture grass in much of the United States due to high forage mass potential, responsiveness to irrigation and fertilizer, persistence under heavy grazing, and broad adaptation to different soil types and climates (Kevin Jensen, Horton, Reed, & Whitesides, 2001; Smeal, O'Neill, & Arnold, 2005; Blair L Waldron, Asay, & Jensen, 2002). It grows best receiving at least 45 centimeters of annual moisture and has been shown to produce over 21 Mg ha^{-1} in ideal conditions in the intermountain west (Asay, Jensen, & Waldron, 2001; K. Jensen et al., 2001; Blair L Waldron et al., 2002). Nutritive value of tall fescue can be good depending on management, but plants quickly become coarse and much less palatable with reproductive maturity, which can result in lower animal preference (Collins & Casler, 1990; K. Jensen et al., 2001)

Tall fescue can form relationships with the naturally occurring endophyte *Acremonium coenophialum*, which results in increased herbage growth, drought tolerance, and faster regrowth after harvest (Arachevaleta, Bacon, Hoveland, & Radcliffe, 1989; Camp, 1986). The problem with most endophyte infected tall fescues is that they often have adverse effects on animal growth and production (Camp, 1986; Liebe & White, 2018; Schmidt & Osborn, 1993). However, recently discovered novel endophyte varieties deliver many of the same plant benefits as the wild type endophyte, but without detrimental animal effects (Nihsen et al., 2004). Endophyte infected tall fescue is mostly used in the South Eastern United States and is generally not recommended for the west (Hannaway et al., 1999).

Meadow Bromegrass (*Bromus biebersteinii* Roem. & Schult.) is an early maturing, rapid regrowing, cool season perennial that is very compatible with legumes such as birdsfoot trefoil and alfalfa (Briscoe, 2018; Cox et al., 2017; St.John, Tilley, & Jensen, 2012). It is well adapted to slightly acidic to mildly alkaline soils and can be grown in dryland settings that receive over 38 cm of precipitation per year (K. Jensen et al., 2001). In mechanically harvested studies conducted in Utah and Montana, annual herbage mass of various meadow bromegrass cultivars averaged 16-19.8 Mg ha^{-1} under optimum irrigation and fertility (Anonymous, 2001; K. B. Jensen, Asay, & Waldron, 2001; Blair L Waldron et al., 2002). Compared to smooth brome, meadow brome has shorter rhizomes, better forage yield and fall growth, and faster regrowth after cutting (KB Jensen, Waldron, Larson, & Peel, 2004; Knowles, Baron, & McCartney, 1993). ‘Cache’ meadow brome, the cultivar used in this study, was developed in Logan Utah for irrigated and semi-irrigated pastures from ‘Regar’, ‘Fleet’, and ‘Paddock’ varieties. In a line-source irrigation study it produced significantly more herbage mass than ‘Fleet’ at all irrigation levels and significantly more herbage mass than orchardgrass cultivars under repeated defoliation (KB Jensen et al., 2004).

Orchardgrass (*Dactylis glomerata* L.) is a perennial bunchgrass that is native to Europe that has been grown in North America for over 200 years. (Casler, Undersander, Fredericks, Combs, & Reed, 1998). It is a widely used species for hay, pasture, or silage and is compatible with alfalfa, birdsfoot trefoil, and various clovers and grasses (Bush, Ogle, St. John, Stannard, & Jensen, 2012; Sulivan, 1992). Orchardgrass is a popular species to plant with alfalfa because the life cycles of orchardgrass and alfalfa match up well, making these mixtures easy to manage (Bush et al., 2012). Orchardgrass can be grown in areas receiving over 46 cm of effective precipitation or irrigation, but requires moderately high moisture soils, making it more drought tolerant than perennial ryegrass, but less-so than meadow brome or tall fescue (K. Jensen et al., 2001). In mechanically harvested Utah studies, orchardgrass produced slightly more forage than meadow brome at higher irrigation levels but less at reduced irrigation levels, leading to equivalent overall forage mass between the two grasses (K. B. Jensen et al., 2001; Blair L Waldron et al., 2002).

Perennial ryegrass (*Lolium perenne* L.) is a short-lived, cool-season perennial often desired for its high nutritive value (E. Allen, Sheaffer, & Martinson, 2013). Because of its high nutritive value characteristics, it is an important grass in Western Europe, New Zealand, and the Northeastern and Northwestern United States (K. J. Moore, 2003). However, it does not produce as much herbage as tall fescue, meadow brome, or orchardgrass, or persist in highly productive stands for more than a few years in the Intermountain Western U.S (K. B. Jensen et al., 2001; Blair L Waldron et al., 2002). Perennial ryegrass tolerates wet soils well, but does not tolerate many common conditions in the intermountain west such as drought, low fertility, heat stress, or severe winters (K. J. Moore, 2003). It has been shown to have elevated non-structural carbohydrates and excellent digestibility in comparison to other grasses, making it a desirable species for dairy pasture production (E. Allen et al., 2013; K. J. Moore, 2003; Terry & Tilley, 1964).

Birdsfoot Trefoil and Condensed Tannins

Birdsfoot trefoil (BFT) is a tannin-containing, non-bloating legume. Bloat is prevented when tannins bind to proteins released from plant mesophyll cells during mastication and thus reduce the activity of bacteria that create bloat-causing froth in the rumen (Min et al., 2005; B. Min et al., 2003). The binding of condensed tannins to plant proteins has also been shown to increase undegraded dietary protein (UDP) from the rumen, leading to increased protein uptake in the small intestine of the ruminant and increased performance (Piluzza et al., 2014). Ramírez-Restrepo & Barry (2005) and Barry & McNabb (1999) suggest that plants must contain at least 5g condensed tannins/kg dry matter (0.5%) in order to reduce bloat in cattle. They also suggest that condensed tannin content of 30-40 g/kg dry matter (3-4%) is optimum to increase amino acid absorption from the small intestine and milk secretion in cattle without suppressing voluntary intake.

In a lead-up study conducted in Lewiston, Utah, tall fescue-BFT treatments did not always have the greatest herbage mass, but resulted in the greatest average daily gains (ADG) of beef steers when compared to tall fescue monocultures and tall fescue-alfalfa mixtures (Blair L. Waldron et al., 2019). Other studies also show similar herbage mass and livestock production results (Hoveland et al., 1981; L Wen et al., 2002).

Studies have been conducted that show there is potential to increase milk production and quality by using birdsfoot trefoil silage or hay in dairy feed rations (Christensen et al., 2015; Hymes-Fecht et al., 2013). In dairy grazing studies, cows that grazed BFT monoculture pastures showed greater intake and milk production when compared to animals on grass pastures (Harris et al., 1998; Macadam et al., 2015; Woodward et al., 2000).

MATERIALS AND METHODS

Pasture Treatments and Pastures

Grazing terminology in this paper is written according to definitions by Allen et al., (2011). This experiment was conducted at the Utah State University Intermountain Pasture Research Farm (41°57'01.85" N, 111°52'15.75" W, elev. 1,369 m, 46 cm annual precipitation and 56.1 precipitation days per year) located near Lewiston, UT, USA. The soils at the site are a Kidman fine sandy loam (Coarse-loamy, mixed, superactive, mesic Calcic Haploxerolls) and Lewiston Fine Sandy Loam (Coarse-loamy, mixed, superactive, mesic Calcic Haploxerolls). The site is within the semiarid Central Great Basin region of the western USA, characterized by hot, dry summers, and a majority of the annual precipitation as snowfall (Figure 1). In this particular area (Cache Valley, Utah, USA), the precipitation from winter-time snowfall is stored in reservoirs and used in the summer for irrigated crop production (Utah Climate Center, 2018). Pasture treatments were endophyte-free tall fescue (‘Fawn’, TF), meadow bromegrass (‘Cache’, MB), high-sugar orchardgrass (‘Quickdraw’, OG), and high-sugar perennial ryegrass (‘Amazon’, PR) in monoculture and as binary mixtures with birdsfoot trefoil (‘Pardee’, BFT). Treatments were arranged in a strip-plot design with three replicates. Seeding occurred in June 2015 with a Great Plains drill (Great Plains Ag, Salina, KS, USA) with double disk openers spaced 15.3 cm apart. Prior to planting, the pastures were prepared with conventional tillage equipment. For grass monocultures, TF, MB, and PR were seeded at 16.8 kg pure live seed (PLS) ha^{-1} and OG at 15.1 kg PLS ha^{-1}. In binary mixtures, TF, MB, and PR were seeded at 10.1 kg PLS ha^{-1}, and OG was seeded at 9 kg PLS ha^{-1}, whereas, the BFT was seeded at 6.7 kg PLS ha^{-1} in all the grass-legume treatments. The BFT was seeded separately from the grasses to ensure proper depth.

Within each replication, pastures of each treatment were considered the experimental unit and consisted of 0.45 ha (i.e., 24 experimental units of 0.45 ha each, totaling 10.7 ha for the entire experimental area) divided evenly into five 0.09 ha paddocks with a single strand of poly-wire charged with a battery-powered fence energizer (Gallagher USA, Riverside, MO). The study was conducted using organic dairy grazing protocols, so no treatments received commercial fertilizer. However, in 2017 and 2018, approved organic sources of nitrogen were applied to the treatments. Chilean nitrate (sodium nitrate, 15-0-2, N-P-K) (SQM, Santiago, Chile) was applied at 28 kg N ha^{-1} in April to all treatments (both monoculture and mixtures). In addition, grass monocultures also received a second application of 28 kg N ha^{-1} of Chilean nitrate in July, and further received 35 kg N ha^{-1} in the form of hydrolyzed poultry feathers in June 2017 and March 2018 (80% CP/6.25=12.8% N) as a slow-release source of N. Pastures were irrigated regularly from mid-May to mid-September of each year. Irrigation was applied in 12 h applications every 14 to 20 days, occurring within 5 days before and 5 days after moving heifers to a new paddock. In 2016, pastures were mechanically harvested in June, and then a preliminary grazing study was conducted throughout the rest of the growing season. Due to differences in how the forage sampling and grazing was conducted, including timing of such events, data from 2016 were not included in the analyses.

Livestock Grazing and Growth Performance Evaluation

Livestock used in the study were 81 (per year) post-puberty Jersey dairy heifers, with mean initial body weights (BW) of 209±47 kg and 183±72 kg in 2017 and 2018, respectively. Animals were cared for with the approval, and in accordance with the guidelines of the Institutional Animal Care and Use Committee at Utah State University under protocol # 2777 and #10063. Three heifers (testers) were randomly allocated to each of the eight pasture treatments (TF, MB, OG, PR, TF+BFT, MB+BFT, OG+BFT, and PR+BFT) within each of the three replications. In addition, three replicates of three control feedlot heifers were fed a mixed ration formulated to meet the nutritional needs of an average daily gain (ADG) target of 0.8 kg day^{-1}.

Rotational stocking was used with a stocking period of 7 days, followed by a rest period of 28 days for each of the five paddocks, such that the entire rotation cycle was 35 days. There were three rotation cycles each year, thus, heifers were on pasture for a total of 105 days (17 May to 30 August, 2017 and 16 May to 29 August, 2018). In a few instances, a tester was removed due to illness, and that heifer's growth performance was no longer used in the analyses. In such cases, a spare heifer was placed in the treatment in order to keep herbage allowance similar for that rotation, but its growth performance was not used in analyses. The total BW of heifers in each pasture were recorded, and later converted to standard animal units (AU) to equalize all treatments over the grazing season. The standard animal unit was defined as a 250 kg post-puberty Jersey dairy heifer (i.e., mean final heifer BW), thus AU was calculated as the total observed metabolic live BW (i.e., BW $kg^{0.75}$) divided by the metabolic live BW for a 250 kg dairy heifer (i.e., 62.87 kg) (V.G. Allen et al., 2011). Paddocks were mowed to a uniform stubble height of

15 cm with a rotary mower at the end of each 7-day stocking period to reduce confounding of remaining residue on herbage mass and nutritive value in subsequent grazing rotations. All heifers had access to water and trace mineral supplement. Heifers were weighed at the beginning of the study, and after each 35-day rotation cycle to determine BW. Cumulative ADG were calculated for each 35-day rotation cycle by dividing the BW gain observed at each weighing by the cumulative number of days on pasture (e.g., BW gain at day 35, 70, and 105). Heifers were gathered from pastures at 20:00 h and held/fasted for 12 hours prior to weighing the next morning.

Herbage Evaluation

Pre-grazed and post-grazed herbage samples were collected weekly throughout the experiment 24 hours prior to (pre-) and immediately after (post-) heifer rotation to the next paddock, by hand-clipping four random quadrats (0.25 m^2) per paddock to a stubble height of 7.6 or 3.8 cm, in 2017 and 2018, respectively. Stubble height was lowered in 2018 to reduce sampling inconsistencies. Post-graze samples were taken immediately adjacent to the pre-grazing samples, unless it was in an area that the heifers had defecated or lain. Herbage samples were placed into a paper bag and dried to a constant weight at 60°C and weighed to determine herbage mass (as dry matter). Pre- and post-grazing compressed sward heights (cm) were measured each time herbage was clipped using a rising plate meter (RPM) (Jenquip, Fielding NZ). Sward height was measured with the RPM directly over each pre- and post-graze clipped quadrat and as the mean of 30 measurements taken in a ‘w’ pattern throughout each paddock. Individual quadrat herbage mass measurements were regressed against the respective RPM measurements, forcing a zero intercept as described by Dillard et al. (2016), to develop an equation that permitted prediction of herbage mass. Separate equations were developed within each year and treatment with resulting R^2 ranging from 0.78 to 0.97 (Appendix A – table 13). Pre- and post-grazing herbage mass were then predicted by converting the 30-measurement RPM mean height to herbage mass using these regression equations. Because of the tall height of the herbage in the first rotation cycle, rising plate meter measurements were not accurate for paddocks 3, 4 and 5 in 2017 and paddocks 4 and 5 in 2018 and not used in the calibration equations.

Pre- and post-grazing herbage mass was converted to average herbage allowance as described by Sollenberger et al., (2005) for rotational stocking. Briefly, for each paddock, average herbage allowance was calculated as ((pre-graze herbage mass/heifer BW) + (post-graze herbage mass/heifer BW)/2), where heifer BW was that obtained from the beginning of each rotation cycle. This method of calculation addresses the questions concerning point-in-time requirements for herbage allowance, and accounts for changes in herbage mass during the 7-day stocking period (L. E. Sollenberger, J. E. Moore, V. G. Allen, & C. G. S. Pedreira, 2005). For inclusion in multivariate analysis (see below), herbage mass was also converted to herbage bulk density (kg m^{-3}) using the mean pre- and post-grazing herbage mass (kg ha^{-1}) and compressed herbage height (cm) following Mayne et al (1997).

[i.e., herbage bulk density = herbage mass in kg ha^{-1} / ((herbage height in cm/100 cm m^{-1}) × 10000 m^2 ha^{-1}))]

Dried herbage samples were ground to pass through a 1-mm screen using a Thomas Wiley Laboratory Model 4 mill (Arthur H Thomas Co, Swedesboro, NJ, USA), and were scanned with a Foss XDS near-infrared reflectance spectroscopy (NIRS) instrument (Foss, Eden Prairie, MN, USA) to determine herbage nutritive value of the feed on offer. The most recent NIRS equations, developed by the NIRS Forage and Feed Testing Consortium (Hillsboro WI, USA), were used to predict nutritive values of the forages. Samples were analyzed with the appropriate equation for each treatment (i.e., grass hay-18gh50 for monocultures, and mixed hay-18mh50 for the grass-BFT mixtures), resulting in estimates of crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF), acid detergent lignin (ADL), in vitro true digestibility (IVTD), 48-hour NDF digestibility (NDFD), fatty acid (FA), and ash. Metabolizable energy (ME) was then calculated as Total digestible nutrients × 0.04409 × 0.82 (National Research Council, 2000); and Net energy for gain (NEg) was estimated from ME using the equation, NEg = $1.42ME - 0.17ME^2 + 0.0122ME^3 - 1.65$ (National Research Council, 2000). Total digestible nutrients (TDN) were calculated using the appropriate formulas for grass monocultures or grass/legume mixtures:

$TDN_{grass} = (NFC \times 0.98) + (CP \times 0.87) + (FA \times 0.97 \times 2.25) + [NDFn \times (NDFDp \div 100)] - 10)$;

$TDN_{grasslegume} = (CP \times 0.93) + (FA \times 0.97\times 2.25) + [NDFn \times (NDFD \div 100)] + (NFC \times 0.98) - 7)$;

where non fibrous carbohydrates (NFC) = 100 – (NDFn + CP + (FA+1) + ash), nitrogen free NDF (NDFn) = NDF × 0.93, and NDFDp = 22.7 + 0.664 × NDFD (Saha et al., 2010).

In addition, proportion of legume in each clipped sample was determined with NIRS. NIRSystem software was used to calibrate an existing grass-legume NIRS equation developed by Waldron et al., (2019) such that it was appropriate for this study. One-half of all clipped grass-BFT samples were hand separated of which 50% were used for additional equation development and 50% were used for equation validation. Following hand separation, grass and legume components were dried and weighed to determine actual percent legume in the herbage mass. Components were then ground separately, and a sub-sample recombined at the original ratio in preparation for NIRS scanning and analysis. The recombined subsample and both individual components (grass and BFT) were each individually scanned for NIRS analysis. The validation for percent legume was R^2 = 0.94, and standard error of prediction (SEP) was 6.20. Tannin concentrations were predicted for the pre-grazed birdsfoot trefoil portion of the separated grass-BFT samples using an NIRS equation that was developed by Grabber et al. (2015; 2014). The equation resulted in prediction statistics of R^2 = 0.88, and SEP = 3.79 (not validated with an independent sampling). Concentration of tannins in the total herbage were calculated as: Herbage tannin (%) = BFT tannin (%) × %BFT in herbage. Pre-grazed BFT tannin concentration was used to calculate both pre-grazed and post-grazed forage tannins, under the assumption that the tannin content did not change significantly between the pre-grazed and post-grazed BFT samples. All herbage nutritive value data, percent legume, and percent tannin are presented on a dry matter basis, and like herbage allowance, calculated as the average between pre- and post-grazing for each 7-day grazing period.

Apparent Herbage Intake

Estimates of herbage intake (on a dry matter basis, kg ha^{-1}) were based upon herbage disappearance and calculated as the difference between pre-grazing and post-grazing herbage mass (HM) (Macoon et al., 2003). Mean herbage intake for a pasture treatment represented measurements made in each of the five 7-day grazing periods for each rotation. Herbage intake on a kg ha^{-1} basis was also converted to kg $heifer^{-1}$ day^{-1}, and additionally as kg AU^{-1} day^{-1} to account for any differences in heifer growth performance among pasture treatments, where an AU was defined as a 250 kg dairy heifer (see description under livestock performance). The percent of the total herbage that was utilized (i.e., disappeared) was also calculated and reported.

Statistical Analysis

Pastures were defined as the experimental units, and the five paddocks and three tester heifers within each experimental unit were observational/sampling units. Therefore, the means of the four herbage samples from each paddock and of the three tester heifers from each experimental unit, within a rotation cycle, were used for statistical analysis. Livestock and herbage data were analyzed across years using the MIXED procedure of SAS (SAS Institute Inc., Cary, NC, USA). Pasture treatment type (monoculture vs mixture), pasture treatment within type, and rotation cycle were considered fixed effects, whereas year and replication were considered random. Rotation cycle was considered a repeated measure and the best covariance models for each trait (most often heterogeneous compound symmetry) were determined and used in the analysis (Littell, Milliken, Stroup, & Wolfinger, 2006). Mean comparisons were made between treatments using Fisher's protected least significant difference (LSD) test at the $p = 0.05$ level of probability. When pasture treatment × rotation interactions were significant, the treatment × rotation interaction means and standard errors were plotted and presented.

Multivariate analyses were conducted to determine which herbage traits were largely associated with differences in herbage intake, following the principal component (PCA) and canonical discriminant (CDA) analyses and procedures outlined by Yeater & Villamil, (2017), using the PRINCOMP and DISCRIM procedures of SAS. In addition to the measured herbage traits, the multivariate analysis also included the traits of leaf softness and plant pubescence as previously characterized for these species in a separate study at the same location (Waldron, unpublished). For leaf softness, OG, PR, and BFT were given the score of '5' (softest), whereas MB and TF received scores of '4.8' and 2.0', respectively. Meadow bromegrass was the only species with pubescence on its leaves and stems and thus assigned a score of '1' for pubescence while the rest of the treatments were scored as '0' for no pubescence. Leaf softness (LSOFT) and pubescence (LPUB) scores for mixtures were adjusted according to the amount of BFT present. PCA analysis, using the correlation matrix, was used to identify independent patterns of association between herbage variables, without any prior assumptions. Use of the correlation matrix rather than the covariance matrix ensured that results were not biased by numerically large variables. Principal components (PC) scores were then examined to determine which herbage traits were associated with the observed patterns of herbage intake by the heifers. Herbage variables directly calculated from another herbage variable were not included in the analysis. Principal components having eigenvalues greater than '0.8' (i.e., PC 1-6), and their corresponding loadings, were retained and further analyzed with multiple regression (SAS Regression procedure with the 'stepwise' option) to investigate the relationship among the herbage variables within principal components and the heifer DMI. For CDA analyses, the first five PC were used to discriminate and classify the eight pasture treatments. In CDA, linear discriminants are still linear combinations of the original variables, but rather than explain as much variation as possible, they maximize the difference between treatments (Yeater & Villamil, 2017). The CDA functions were examined to determine if the herbage trait model could effectively distinguish among pasture treatments, and which PCs and subsequently herbage traits (from absolute loading scores) largely contributed to the functions' ability to discriminate and classify treatments.

RESULTS

Heifer growth performance was reported in a companion paper by Hadfield et al. (2019) and will not be reported herein. Mean values of pasture treatments and rotation cycles for all herbage traits measured are reported (Table 1-3, and 9), whereas, those traits that exhibited significant pasture treatment by rotation cycle interactions (Appendix A) are also graphed to show changes across the grazing season (Figure 2-16). Many of the pasture treatment × rotation interactions were due to the rapid spring-growth and flowering head development of the cool-season grasses in the first rotation which affected most herbage traits. All grass species reached reproductive growth by the third paddock of the first rotation, with meadow bromegrass heading first, followed closely by orchardgrass, tall fescue, and perennial ryegrass. Thereafter, the grass species remained in a vegetative growth stage through the second and third stocking rotations.

Herbage Intake

The pasture-type (mixture vs monoculture) × rotation interaction was not significant ($p>0.05$) for herbage intake (Appendix A), with herbage intake of grass-BFT mixtures greater ($p<0.03$) than grass monocultures (Table 1). Herbage intake was greater ($p<0.05$) in rotations 1 and 3 than rotation 2 (Table 1). Herbage mass did not limit intake, with only 23 to 40% of total herbage utilized (Table 1).

Pasture treatment also differed ($p<0.0001$) for herbage intake, and the pasture treatment × rotation interaction was significant ($p=0.0259$) for percent herbage utilized (Appendix A). This interaction was largely due to greater intake (%) during rotation 3, but also a dramatic increase in % herbage disappearance of MB and MB+BFT from rotation 2 to 3 contributed to the interaction (Figure 2). The MB+BFT, OG+BFT, and OG pastures had the greatest ($p<0.05$) herbage intake (kg AU^{-1} day^{-1}), whereas, PR and TF consistently had the least (Table 1). Also notable was that the addition of BFT consistently increased ($p<0.05$) herbage intake for PR, but not so for the other grasses (Table 1).

Herbage Traits

Pasture-type differed ($p<0.01$) for all herbage mass, morphological and nutritive traits except digestibility (IVTD), and exhibited significant ($p<0.05$) interactions with rotation for herbage allowance, CP, fat, ADL, and minerals (Appendix A). These interactions were primarily due to magnitude differences, with grass-BFT mixtures having greater ($p<0.05$) herbage allowance, CP, ADL and minerals, but less fat, than monocultures at each rotation (Figures 3, 4, and 5; Table 3). In addition, on average grass-BFT mixtures also had greater ($p<0.0001$) pre-graze herbage mass, herbage height and bulk density, more favorable ($p<0.0001$) (less) NDF, ADF, (more) NFC, ME, and NEg, and less favorable (less) WSC, ESC, fructans, and NDFD than the grass monocultures (Tables 2 and 3).

In a PCA of all herbage mass, morphological, and nutritive characteristics, the first six principal components (PC) had eigenvalues greater than 0.8 and respectively explained 34.9, 22.3, 14.0, 9.4, 6.4, and 4.0% (cumulative 91%) of the variation observed for herbage data (Table 4). In PC1, the highly correlated fiber and energy traits of ADF, NDF, NFC, ESC, WSC, and ME were of most importance (Table 4). NDFD, ADL, BFT proportion in herbage, and % tannin in the herbage were the most important variables in PC2, whereas herbage allowance and compressed height, both highly correlated with herbage mass, primarily contributed to PC3 (Table 4). Interestingly, leaf traits of softness and pubescence, plus ash were most important in PC4 (Table 4). Finally, herbage traits not previously listed that were important in PC5 and PC6 were CP and bulk density, respectively (Table 4). Stepwise multiple regression using the loading scores for these six PCs and regressing against measures of herbage intake resulted in models with significance of $p<0.0001$ and fits (R^2) of 0.32 to 0.42 (Table 5). Of these PC variables, PC6 explained the highest percent of the variation, whereas, PC1 was eliminated ($p>0.15$) in all models (Table 5).

Principal component scores were used in canonical discriminant analysis (CDA) and resulted in the first three canonicals (CAN) explaining 58.3, 28.4, and 7.3% (cumulative 94.1%) of the differences among pasture treatments (Table 6). Furthermore, these canonicals were able to effectively discriminate among pasture treatments 77.3 to 98.7% of the time, with an overall error rate of only 9.3% (Table 7). Interestingly, PC4 dominated CAN1, whereas, PC1 and PC2 largely contributed to CAN2, indicating that leaf texture and herbage fiber traits effectively discriminated among the pasture treatments (Table 8). However, PC6 dominated CAN3, indicating that herbage bulk density and height (i.e., herbage mass related traits) also contributed to the differences among pasture treatments (Table 8).

The main effects of pasture treatment, rotation, and the pasture treatment × rotation interactions were highly significant ($p<0.0001$) for those herbage traits identified in PC1, namely ADF, NDF, NFC, ESC, WSC, and ME (Appendix A). The pasture treatment × rotation interaction for ADF and NDF primarily resulted from an increase in these fiber traits in rotation 2, followed by a decrease in rotation 3 (Figures 6 and 7). In addition, grass monocultures and their respective BFT mixtures did not differ ($p>0.05$)

for ADF or NDF in rotation 1, but most BFT mixtures had more favorable (less) ($p<0.05$) ADF and NDF than their respective monocultures in rotation 2 and 3 (Figures 6 and 7). On average, PR+BFT had the least ($p<0.05$) ADF and NDF, followed closely by PR, whereas, ADF and NDF were greatest ($p<0.05$) in MB (Table 3).

Like fiber, the significant pasture treatment × rotation interactions for carbohydrate traits of NFC, ESC, and WSC were primarily due to differences between rotations with carbohydrates decreasing from rotation 1 to 2 as days became hotter, and then leveling off between rotations 2 to 3 (Figures 8, 9, and 10). However, a continued decline between rotations 2 and 3 for NFC in PR also contributed to the interaction (Figure 8). Interestingly, the PR and PR+BFT treatments exhibited the greatest ($p<0.05$) concentrations of NFC, ESC, and WSC possibly validating the claim of the high-sugar perennial ryegrass cultivar used (Table 3 and Figures 8, 9, and 10). In contrast, carbohydrate concentrations in OG and OG+BFT were the least (i.e., NFC; $p<0.05$) or not different (i.e., WSC) compared to the remaining pasture treatments, thereby not supporting the putative high-sugar OG cultivar used. Metabolizable energy followed a similar pattern as carbohydrate concentrations, however, ME in MB+BFT and OG+BFT was equivalent to PR and PR+BFT in rotations 2 and 3 (Figure 11).

Traits in PC2 included NDFD, ADL, and BFT proportion, which were all significant ($p<0.0001$) at the pasture treatment, rotation, and pasture treatment × rotation interaction levels (Appendix A). These three traits were highly correlated (absolute values r=0.59 to 0.83), and as BFT and lignin increased in rotation 2, NDFD declined (Figures 5, 12 and 13). On average, PR had the greatest ($p<0.05$) NDFD and least ($p<0.05$) lignin (e.g., most favorable values) (Table 3), whereas, PR+BFT had the least favorable levels of both these traits (Table 3) corresponding to the greatest ($p<0.05$) BFT proportion (Table 2).

Herbage allowance was the most prominent variable in PC3, and in general declined in each successive rotation (Figure 3), but more so between rotation 1 and 2 resulting in a significant ($p=0.0009$) treatment × rotation interaction (Appendix A). Overall, herbage allowance was greatest ($p<0.05$) for MB+BFT and more than double the least found in PR (Table 2). Crude protein was the predominant herbage variable in PC5 and except for MB+BFT and OG+BFT, declined from rotation 1 to 2 and then increased in rotation 3 (Figure 4). Overall, grass-BFT mixtures had 45% greater ($p<0.05$) CP than their respective monocultures, and PR+BFT had the greatest ($p<0.05$) individual CP level (Table 3). Principal component 6 was primarily comprised of herbage bulk density and height. Mean herbage bulk density was greater ($p=0.05$) in mixtures than in monocultures, with respective measurements of 0.97 and 0.82 kg m^{-3}. Pasture treatment ($p<0.0001$), rotation ($p=0.0006$), also had an effect ($p<0.0001$) on bulk density. Ranking of individual pasture treatments was not as expected with bulk density of PR+BFT greatest ($p<0.05$), but the PR monoculture exhibiting the least ($p<0.05$) bulk density (Table 2). This was likely due to the large proportion of BFT in the PR+BFT treatment. In contrast to bulk density, herbage height decreased from rotation 1 to 2, but more so for MB, TF, and OG and their mixtures than the shorter statured, PR, and PR+BFT (Figure 14).

DISCUSSION

Herbage Intake

Pasture-based milk production is the fastest growing segment of U.S. organic agriculture; but such dairies experience up to 32% decrease in milk production (William D. McBride & Greene, 2009), due to reduced herbage intake by grazing dairy cows (F. Bargo et al., 2003). Thus, determining the herbage variables that are highly associated with herbage intake by dairy breeds will be useful in putting together the most optimum pasture mixtures. Multiple regression using the first six principal components from PCA only explained 32% of the variation in herbage intake by Jersey dairy heifers. Thus, there were obviously still other unidentified variables associated with the variation in herbage intake, possibly including environmental conditions, heifer breeding and background, and errors associated with measuring herbage intake. Nevertheless, we found significant variation among pasture treatments for herbage intake, and discriminant analysis indicated that these differences were largely associated with the variation in prominent herbage variables.

On average, grass-BFT mixtures had greater ($p<0.05$) herbage intake than grass monocultures (4.5 and 3.8 kg heifer^{-1} day^{-1}, respectively) (Table 1). These levels of herbage intake equate to 2.0 and 1.7% of heifer BW for grass-BFT mixtures and grass monocultures, respectively, and are within norms expected for heifers within this weight class (National Research Council, 2000). The greater herbage intake of grass-BFT mixtures, compared to grass monoculture, coincides with many previous studies that have concluded that legumes increase forage intake. For instance, Woodward et al., (2000) found that cows fed freshly harvested BFT in a feed bunk had increased forage intake compared to cows fed freshly cut perennial ryegrass, and Macadam et al., (2015) reported that dairy cows grazing BFT monocultures had greater herbage intake than those grazing grass monocultures. Ribeiro-Filho et al., (2003, 2005) found that grass-clover swards with average clover contents of 42% increased herbage intake, but swards with 27% clover did not significantly increase intake over the grass monocultures. Like their findings, our pasture treatments with the most BFT proportion in the herbage (41% in PR+BFT and 21% in MB+BFT) had significantly greater ($p<0.05$) herbage intake over respective grass monocultures, as compared to no difference ($p>0.05$) between monoculture and mixtures for treatments with less than 20% BFT (orchardgrass and tall fescue) (Table 1). Thus, it appears that grass-BFT mixtures with greater than 20% BFT proportion increases herbage intake of grazing dairy heifers.

Herbage Traits that Most Characterized Pastures and Intake Differences

In this study we observed variation among pasture treatments in both herbage intake, as well as in herbage quantity and quality. It is often difficult to obtain significant differences in grazing studies given the limited replication and spatial and biological variability (Bransby, 1989; Giesbrecht, 1989). However, multivariate analysis utilizes highly correlated traits, such as herbage characteristics, and given the response data, can point to which variables drive the differences among the treatments (Yeater & Villamil, 2017). As such, canonical discriminant analysis (CDA) was highly efficient in identifying individual pasture treatments based upon the measured herbage traits (77 to 99% accuracy), with the most discriminating herbage traits being: leaf softness and pubescence (PC4); ADF, NDF, NFC, ESC, WSC, and ME (and to a lesser extent, IVTD and CP) (PC1); NDFD, ADL, BFT proportion, and % tannin in herbage (PC2); and herbage bulk density and compressed height (PC6). In addition, regression analysis using PC loading scores indicated that herbage allowance (PC3) was also associated with herbage intake differences.

Leaf Texture Characteristics (PC4)

Leaf softness and pubescence were associated with both the ability to distinguish among pasture treatments and variation for herbage intake. Leaf pubescence has often been considered a plant defense mechanism to reduce herbivory (Briske, 1996; Tarazona, Ceballos, Naranjo, & Cuartas, 2012), however, much less so for vertebrate herbivores as compared to invertebrates (Briske, 1996). As such, meadow bromegrass, the only species classified as having pubescent leaves and stems, had moderate and high herbage intake in monoculture and BFT mixtures, respectively, compared to other treatments (Table 1). Thus, pubescence was likely more associated with treatment differentiation than herbage intake.

In contrast, tall fescue was classified as having the least soft leaves of all species, and tall fescue monocultures and BFT mixtures also consistently had the lowest herbage intake (Table 1). 'Leaf harshness' has been reported to be negatively correlated with sheep preference (Cougnon, De Koker, Fievez, & Reheul, 2014). However, Cougnan et al., (2018) recently found that leaf softness becomes more difficult to characterize after several cycles of plant breeding, and as such, the correlation between leaf softness and sheep grazing preference was low in elite tall fescue breeding populations. In this study, we used 'Fawn' tall fescue, an old variety with coarse leaves, and as such it is probable that course leaf texture was negatively associated with herbage intake. The use of dairy heifers probably exacerbated this effect as the dairy breeds can be finicky grazers (F. Bargo et al., 2003).

Ash content was also a dominant herbage characteristic in this PC. Ash content increased as the grazing season progressed, likely due to hoof action and other normal grazing activities, and heifers probably avoided grazing areas with notable amounts of soil on the leaves. However, ash would also be highly associated with leaf texture as it would represent the silica, and other non-organic compounds contained on tall fescue leaves (Shewmaker Glenn E., 1989).

Fiber and Carbohydrates (PC1)

The concentrations of fiber in forage diets has been reported to be the best single nutritive predictor of intake (Waldo, 1986) and as the main source of energy for ruminants (Wilson, 1994). In this study, fiber and energy herbage traits within PC1 were highly important in differentiating pasture treatments. Fiber traits (i.e., NDF and ADF) were highly negatively correlated with PC1 (Pearson correlation; r=-0.96 and -0.88, respectively), whereas carbohydrate and digestibility traits (i.e., NFC, ESC, WSC, and IVTD) were highly positively correlated with PC1 (Pearson correlation; r= 0.67 to 0.89) indicating that the pasture treatments differed in rapidly available energy. Overall, these results confirm that we were successful in choosing pasture treatments with a range of inherent energy levels (see objectives), however the effect of energy on herbage intake was not straight forward.

The importance of cell wall fiber components in our analysis is not surprising, given the large amount of research showing that NDF and ADF are negatively correlated to intake and digestibility. As a general rule, animals will not consume more than 1.3% of their BW in NDF, but research indicates that animals on pastures with high herbage allowance often consume greater than 1.3% BW of NDF (Vazquez & Smith, 2000). However, even though our herbage allowance greatly exceeded metabolic need (e.g., ~2.0 to 2.5 % of BW), average apparent NDF intake was 1.0 and 1.1% of BW, for grass monocultures and BFT mixtures, respectively (based upon herbage intake and NDF estimates, Table 1 and 3). Herbage intake in the OG monoculture pastures was not different than the most consumed pasture treatment and had the greatest apparent NDF intake at 1.4% BW. Whereas, the tall fescue monoculture was one of the least consumed treatments, but only had NDF intake of 0.75% BW. Thus, these counter-intuitive results help explain why PC1 (e.g., NDF and ADF) was more highly associated with pasture treatment differentiation than in the regression analysis of herbage intake.

In comparison to other research, our orchardgrass monoculture NDF concentration (61%) is within the range of 51-61% NDF observed in their two mechanically harvested orchardgrass studies near the site of our grazing study (J. Robins, Bushman, Feuerstein, & Blaser, 2016; 2015). In contrast, our NDF concentrations of 59 and 55% for tall fescue monocultures and mixtures, respectively, is 4-5 percentage points greater than the 55 and 50% reported by Waldron et al., (2019) in a grazing study at the same location. This may be because we used 35-day rotation cycles, resulting in more stem and leaf growth and greater NDF concentrations, compared to their 28-day rotation cycles. Furthermore, Jensen et al., (2016) conducted a mechanically harvested study in northern Utah that included all of the grass species in our study and found mean NDF concentrations at least 9 percentage points less than our grass monocultures. This large discrepancy was likely due to differences in harvest frequency (every 30 days) and clipping versus grazing regrowth response.

Non-structural carbohydrates (NSC) (e.g., NFC and WSC) were the other predominate part of PC1. Mayland et al., (2000) examined the effects of different types of NSC on livestock preference in tall fescue, and though no specific sugar fraction increased preference, the livestock did prefer grass varieties with greater total NSC. Likewise, Cougnan et al., (2018) reported that sheep preferred tall fescue with high WSC and low NDF. However, given our 7-day grazing periods, as opposed to short-duration periods, preference based upon NSC is probably not directly related to our herbage intake. This is particularly true for NFC, the third most important variable in PC1. Baudracco et al., (2010) reviewed available research and came to the conclusion that feeding high NFC supplements to grazing animals usually reduced intake of pasture due to a substitution of digestible energy sources. They attributed the lower herbage intake to reduced ruminal pH, and a lower rate of fiber digestion. Vasquez & Smith, (2000) also found an inverse relationship between pasture intake and NFC supplementation. Grazing studies have consistently reported that increased concentrate/NFC supplementation reduced grazing time (Arriaga-Jordan & Holmes, 1986; F Bargo, Muller, Delahoy, & Cassidy, 2002; Gibb, Huckle, & Nuthall, 2002; Kibon & Holmes, 1987; Rook, Huckle, & Penning, 1994). However, interestingly, Stakelum & Dillon, (2003) found that fibrous concentrates, like those in our study, have a less depressing effect on grass intake than cereal or starch-based concentrates. Our study results mostly concur with these NFC-supplemented grazing trials and uniquely indicate that substitution of inherently greater herbage NFC in place of more bulky fibrous energy sources probably reduces herbage intake by grazing livestock.

Metabolizable energy of the herbage was also an important variable in PC1 and as expected was associated with fiber and non-structural carbohydrates (Table 5). Mean grass-BFT mixture ME was greater ($p<0.05$) than mean grass-monoculture, and every individual grass-BFT pasture ME was greater than its corresponding grass-monoculture (Table 3). Given these differences, and the fact that energy is often the most limiting nutrient on pasture (F. Bargo et al., 2003; Kolver & Muller, 1998), it is not surprising that ME was associated with treatment differences.

NDFD, Lignin, BFT, and Tannins (PC2)

NDFD was highly positively correlated with PC2 (Pearson correlation; $r=0.90$, $p<0.0001$), whereas, BFT%, ADL, and % tannins in the herbage were all highly negatively correlated with PC2 (Pearson correlation; $r=-0.83$ to -0.63, $p<0.0001$). Given the effect that BFT had on NDFD, lignin, and tannins (i.e., mean mixture vs monoculture data), it can be concluded that this PC was primarily BFT related. Inclusion of BFT in pasture treatments resulted in lesser ($p<0.05$) NDFD (less favorable) than grass-monocultures, but only PR+BFT and TF+BFT were less ($p<0.05$) than their respective grass-monocultures (Table 3). Because of its effect on passage rate, Brink & Soder, (2011) hypothesized that superior cell wall digestibility (NDFD) would increase herbage intake, but they were unable to validate this using several cool-season grasses varying in NDFD, including meadow fescue and orchardgrass. On average, our grass-BFT mixtures also had greater ($p<0.05$) amounts of highly indigestible lignin, and in contrast to NDFD, all individual grass-BFT mixtures had up to 64% greater ($p<0.05$) lignin (Table 3).

It has been suggested that low levels of condensed tannins (CT) improve herbage intake, however there is little research to validate this hypothesis (Piluzza et al., 2014). We hypothesized that CT in the birdsfoot trefoil would interact in a complimentary way with inherently highly-accessible energy (i.e., WSC) to increase herbage intake, and at least in the case of perennial ryegrass (greatest CT and WSC) this proved to be the case (Tables 1-3). Low levels of CT from forage legumes have been shown to improve protein use efficiency and livestock performance (B. R. Min, T. N. Barry, G. T. Attwood, & W. C. McNabb, 2003). However, these beneficial effects are usually realized at CT concentrations of 1 to 2.5% (MacAdam, 2019), far above our highest level of 0.5% in the PR+BFT herbage. This would support our conclusion of a synergistic effect between CT from BFT and a high energy grass, however it is also possible that selective grazing of BFT resulted in dietary CT levels above 1%. Nevertheless, CT levels of 0.16 to 0.23 in the herbage of the other BFT mixtures (Table 2) was probably not sufficient to have much effect on herbage intake.

Herbage Bulk Density, Height, and Allowance (PC6 and PC3)

Herbage bulk density has been cited as an important factor influencing herbage intake (Brink & Soder, 2011), and in our study was one of the most influential variables in the regression equation for intake. The influence of bulk density was especially notable in perennial ryegrass and tall fescue monocultures, which had less bulk density in comparison to many other treatments and had the least herbage intake (Table 1). Casey et al., (2004) noted that bite mass of cows increased significantly as sward bulk density of perennial ryegrass increased under uniform sward height, whereas, McGilloway et al., (1999) concluded that bulk density became increasingly influential on intake as sward height of perennial ryegrass was reduced. These relationships appear to hold true in other cool-season grass species as Brink & Soder, (2011) found that herbage intake of meadow fescue, orchardgrass, quackgrass and reed canarygrass was positively related to leaf bulk density and negatively related to stem bulk density.

Herbage height, as a closely related trait to bulk density was also an important factor of intake in this study. McGilloway et al., (1999) found that sward height was the principal factor influencing intake per bite in perennial ryegrass swards, but that sward bulk density became increasingly more important as the sward height was reduced due to grazing. Furthermore, Tharmaraj et al., (2003) reported that perennial ryegrass swards with pre-grazed sward heights of 28 cm and herbage allowance of 70 kg HM cow^{-1} day^{-1} resulted in greater herbage intake than those with 14 cm height and allowance of 35 kg HM cow^{-1} day^{-1} (based on a 525 kg cow). In comparison, with the exception of tall fescue, our pasture treatments where herbage height exceeded 28 cm also had the greatest $p<0.05$) herbage intake (Table 2), providing further evidence for the importance of this trait.

Vazquez & Smith, (2000) highlighted the importance of herbage allowance, concluding that it influenced intake more than herbage nutritive value. Bargo et al., (2002) showed that as herbage allowance increased from 20 to 40 kg DM cow^{-1} day^{-1} , herbage intake also increased from 2.9% to 3.4% of BW (based on a 631 kg cow), but their herbage allowances were much lower than ours. Our mean grass-BFT mixture herbage allowance of 0.44 kg HM kg^{-1} live weight (LW) was 29% greater ($p<0.05$) than mean grass monoculture (Table 2), and very close to that of a study using Holstein heifers in Wisconsin by Brink and Soder (2011) (i.e., 0.43 kg HM kg^{-1} LW). They noted that there was no relationship between herbage intake and herbage allowance in their study, suggesting that their herbage allowance allowed for ad libitum intake. We also allowed ad libitum intake with only 29 and 33% utilization (as measured by disappearance) of the grass-BFT and grass monoculture pastures, respectively (Table 1), which may have reduced the effect of herbage allowance on intake. However, in our study every grass-BFT mixture had greater herbage allowance than their respective grass monoculture (Table 2), and except for tall fescue, had greater corresponding herbage intake (Table 1).

At the beginning of this study, stocking rates were the same among treatments and remained similar throughout, thus differences in herbage allowance were primarily driven by herbage mass. As such, mean herbage mass of grass-BFT mixtures was also 29% greater than that of grass monocultures (Table 2). Multiple studies have reported that tall fescue-, meadow bromegrass-, and orchardgrass-BFT mixtures had equal forage mass to their comparative fertilized (134 kg N ha^{-1} yr^{-1}) grass monocultures (Cox et al., 2017; Guldan, Lauriault, & Martin, 2000; Lauriault, Guldan, Martin, & VanLeeuwen, 2006). However, comparable to our results, Sleugh et al., (2000) reported that orchardgrass-BFT mixtures increased forage mass by 20% over fertilized (67 kg N ha^{-1} yr^{-1}) orchardgrass monocultures. Cox et al., (2017) also reported that PR+BFT forage mass was 9% greater than fertilized (134 kg N ha^{-1} yr^{-1}) PR monocultures, whereas we found that PR+BFT pastures had 78% greater herbage allowance than the PR monoculture. Major differences between these studies and ours was that most previous reports were mechanically harvested, compared to grazed, and differences in fertilizer rates. We applied 91 kg N ha^{-1} yr^{-1} on grass monocultures, and 28 kg N ha^{-1} yr^{-1} on grass-BFT mixtures in the early spring, whereas the previous studies did not apply any fertilizer to grass-birdsfoot trefoil mixtures.

Other Factors Possibly Influencing Herbage Intake

Fat content of the herbage was observed in PCs 2, 3, 4, 5, and 6 with slightly lower loading scores than the traits previously discussed. Mean grass-monoculture treatments had greater ($p<0.05$) fat than grass-BFT mixtures, and orchardgrass exhibited greater ($p<0.05$) fat in both monoculture and BFT mixture than most other pasture treatments (Table 3). Bargo et al., (2003) conducted an extensive review and concluded that fat-supplemented dairy cows on pasture generally do not significantly differ in dry matter intake compared to non-supplemented animals. Schroeder et al., (2004) was in agreement with the findings of Bargo et al., but did find some studies that had reported that fat greater than 8-9% in a TMR diet for dairy cows reduced dry matter intake due to slower fiber digestion in the rumen. They also hypothesized that since typical pasture diets are relatively low in fatty acid content, a growth response from additional fat may be expected. Both papers mainly reviewed fat-supplemented grazing studies, whereas, heifers in our study received all dietary fat from grazed herbage, which ranged from 2-3% (Table 3). Inasmuch as fat appeared in every PC, except the ‘energy’ PC (PC1), it is not clear how fat affected herbage intake or differences among the pasture treatments. But given the low levels of fat in these treatments, perhaps as Schroeder et al., (2004) hypothesized, even the minimal differences had an effect on herbage intake.

Crude protein was an important secondary variable in PC5 and PC1. Moore et al., (1999) stated that crude protein has been shown to increase ruminant intake when TDN:CP ratio is >7 (deficient in N). Our treatments never exceeded a TDN:CP ratio of 7, but the TDN:CP ratio of PR and TF monocultures was between 6.8-6.9 in rotation two. Similarly, Fisher, (1996) reported that when CP is less than 6-9% it is closely associated with intake, but that digestibility and NDF have a greater influence on intake when protein is over 9%. Crude protein of OG, PR, and TF grass monocultures fell between 7.5 and 9% at times during the growing season, suggesting that CP might have influenced intake during those periods.

Preliminary multivariate analyses included the mineral content of the herbage and P, Ca, and Mg appeared to be important variables. There is minimal literature on minerals and intake, but in one case, a confined feeding trial showed that heifer intake increased quadratically as dietary phosphorus increased from 0.10 to 0.38% (Geisert et al., 2010). However, our herbage phosphorus content was moderately high (2.1 to 3.0 g kg^{-1}) (Table 9) and our heifers had free access to mineral supplements throughout the study. Therefore, our mean phosphorus levels would have been well within heifer requirements of 0.30-0.34% (3.0-3.4 g kg^{-1}) established by the National Research Council (Council, 2001). Overall, given that we had free-choice mineral supplementation, minerals probably appeared in PCA analysis due to differences in intake as opposed to being associated/influencing intake.

CONCLUSION

We found that heifer herbage intake in our study was largely influenced by both physical and nutritive herbage characteristics. Some of the most important physical characteristics influencing intake included herbage bulk density, herbage height, herbage allowance, plant pubescence, leaf softness, and birdsfoot trefoil content. The most important nutritive characteristics included energy and fiber related traits such as fat, NFC, NDF and ADF, metabolizable energy and net energy gain. Crude protein and ash were also associated with intake.

While we hypothesized that birdsfoot tannins would interact with grass energy to increase intake, they did not come up as an important characteristic in the multivariate analysis, possibly due to the small amount that we observed in the herbage. However, the two treatments with the most BFT (PR+BFT and MB+BFT) generally showed an increase in intake over their respective monocultures while the other two mixtures did not. Furthermore, PR+BFT had the greatest concentration of energy and tannins compared to all other treatments, suggesting that a complimentary effect between energy and tannins occurred to increase herbage intake in this treatment.

The results of this study show the importance of not only planting the right type and composition of grasses and legumes but using best management practices as well. Planting highly nutritious grasses and BFT in the right proportions (at least 20% BFT) and managing animals in a way that maintains moderately tall, dense herbage with favorable leaf texture characteristics must both be taken into account if managers are to optimize intake on pasture.

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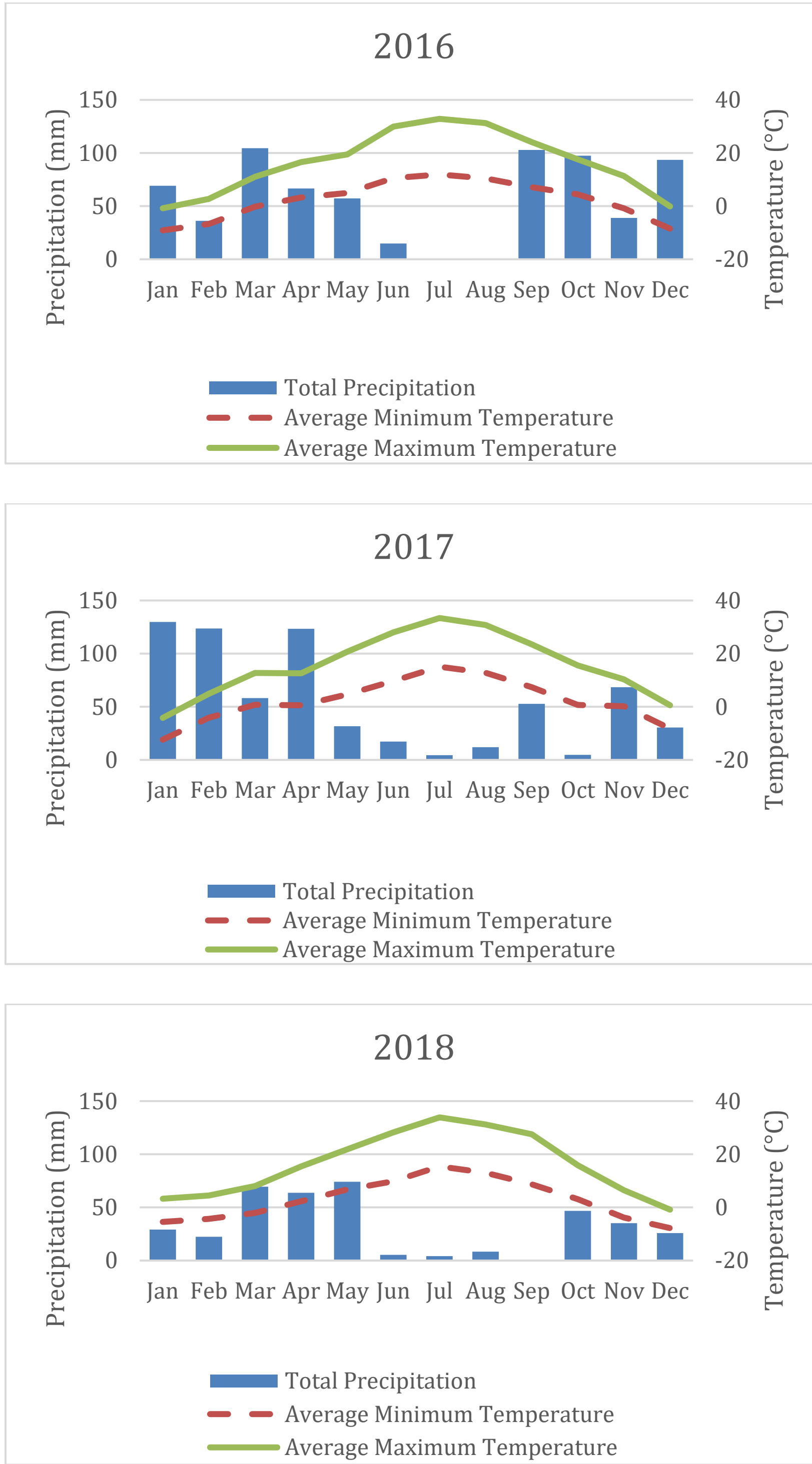


Figure 1: Total monthly precipitation, and average minimum and maximum monthly temperatures in 2016, 2017, and 2018. Data is from a dairy heifer grazing study in Lewiston, Utah. (Utah Climate Center, Station Name: Richmond, Station ID: USC00427271)

Table 1 Apparent intake per hectare, per animal unit (AU) and per heifer (heif), and percent disappearance of herbage. Data is from a dairy heifer grazing study in Lewiston, Utah in 2017 and 2018. Pasture treatments included monocultures of meadow brome (MB), orchardgrass (OG), perennial ryegrass (PR) and tall fescue (TF), and each grass in binary mixture with birdsfoot trefoil (BFT) Metabolic liveweights were converted to AU (animal units) based on a 250 kg heifer.

Treatment type	---------------------Intake---------------------						Utilization	
	kg AU^{-1} day^{-1}		kg $heif.^{-1}$ day^{-1}		kg ha^{-1}		%	
Mixture	4.9	a	4.5	a	1031	a	29.4	b
Mono	4.3	b	3.8	b	870	b	33.0	a
Mean S.E	0.2		0.04		75		5.5	
Treatment								
MB+BFT	5.9	a	5.3	a	1241	a	31.9	b
OG+BFT	5.7	a	5.1	ab	1191	ab	35.6	b
OG	5.6	a	4.8	ab	1126	bc	40.1	a
MB	5.0	ab	4.3	bc	1022	cd	33.4	b
PR+BFT	4.3	bc	3.9	cd	913	d	26.6	c
TF+BFT	3.7	cd	3.3	de	780	e	23.6	c
TF	3.2	d	2.8	e	668	e	24.6	c
PR	3.3	d	2.8	e	664	e	34.0	b
Mean S.E	0.3		0.4		90		5.6	
Rotation								
1, 0-35 days	5.2	x	4.3	x	1018	x	28.2	y
2, 35-70 days	3.7	y	3.3	y	775	y	28.9	y
3, 70-105 days	4.8	x	4.5	x	1059	x	36.6	x
Mean S.E	0.2		0.3		73		5.5	

Pasture treatments followed by different letters (a,b,c,d,e) are significantly different (p = 0.05).

Rotation cycle followed by different letters (x,y,z) are significantly different (p = 0.05).

†The number and body weight of heifers in each paddock were recorded and converted to animal units (AU) where for this study one AU = a 250 kg jersey heifer (Vivien Gore Allen et al., 2011).

Table 2 Midpoint herbage height, herbage bulk density, birdsfoot trefoil (BFT) proportion of herbage, BFT and total tannin content in the forage, herbage allowance, and pre-graze herbage mass. Data is from a dairy heifer grazing study in Lewiston, Utah in 2017 and 2018. Treatments included monocultures of meadow brome (MB), orchardgrass (OG), perennial ryegrass (PR), tall fescue (TF), and each grass in binary mixture with birdsfoot trefoil (BFT).

	Herbage height	Herbage bulk density	BFT proportion	BFT tannins	Forage tannins	Herbage allowance	Pre-graze herbage mass
Mixture	cm	kg m^{-3}	%	g kg^{-1}	g kg^{-1}	kg HM kg^{-1} BW	kg ha^{-1}
Mono	31.4 a	1.0 a	22.9	10.9	2.5	0.44 A	3423 a
Mean S.E	27.9 b	0.8 b	-	-	-	0.34 B	2656 b
	0.6	0.1	-	-	-	0.07	252
Treatment							
TF+BFT	34.4 a	0.91 d	13.8 c	11.4 ab	1.8 bc	0.45 b	3443 bc
MB+BFT	33.1 bc	1.02 b	20.7 bc	10.8 ab	2.3 b	0.48 a	3962 a
OG+BFT	32.3 bc	0.86 e	16.1 bc	9.6 b	1.6 c	0.39 cd	3373 bc
TF	31.9 c	0.80 f	-	-	-	0.38 d	2855 d
MB	30.0 d	0.93 c	-	-	-	0.40 cd	3190 c
OG	29.6 d	0.78 f	-	-	-	0.34 e	2875 d
PR+BFT	25.8 e	1.12 a	41.0 a	11.8 a	4.8 a	0.41 c	3315 bc
PR	21.0 f	0.75 g	-	-	-	0.23 f	1913 e
Mean S.E	0.7	0.14	4.4	1.2	0.5	0.08	345
Rotation							
1, 0-35 days	36.0 x	0.898 a	14.6 y	8.4 y	0.6 z	0.51 x	3774 x
2, 35-70 days	26.3 y	0.898 a	26.4 x	11.1 x	1.3 y	0.33 y	2717 z
3, 70-105 days	26.5 y	0.896 b	27.7 x	13.1 x	1.8 x	0.31 y	2897 y
Mean S.E	0.5	0.136	7.1	1.2	0.3	0.08	341

Pasture treatments and treatment types followed by different letters (a,b,c,d,e,f) are significantly different (p = 0.05).

Rotation cycle followed by different letters (x,y,z) are significantly different (p = 0.05).

Table 3 Forage nutritive values. Included are values for crude protein (CP), neutral detergent fiber (ADF), acid detergent fiber (ADF), lignin, neutral detergent fiber digestibility (NDFD), in-vitro true digestibility (IVTD), ethanol-soluble carbohydrates (ESC), non-fibrous carbohydrates (NFC), water soluble carbohydrates (WSC), fructan, fat, metabolizable energy (ME), and net energy gain (NEg) from a dairy heifer grazing study in Lewiston, Utah in 2017 and 2018. Treatments included monocultures of meadow brome (MB), orchardgrass (OG), perennial ryegrass (PR), tall fescue (TF), and each grass in binary mixture with birdsfoot trefoil (BFT).

Treatment type	CP	NDF	ADF	Lignin	NDFD	IVTD	ESC	NFC	WSC	Fructan	Fat	ME	NE_g
	---------------g kg^{-1} herbage---------------				g kg^{-1} NDF	----------------------------------g kg^{-1} herbage----------------------------------						Mcal kg^{-1}	Mcal kg^{-1}
Mixture	147 a	525 b	344 b	46 a	585 b	768 a	57 b	220 a	69 b	14.6 a	23 b	2.74 a	1.2 a
Mono	101 b	575 a	366 a	36 b	621 a	765 a	62 a	212 b	76 a	14.4 b	27 a	2.61 b	1.1 b
Mean S.E	3	20	9	1	6	6	6	20	7	0.4	0.05	0.03	0.02
Treatment													
PR+BFT	176 a	421 f	302 e	54 a	554 d	790 b	72 b	295 a	85 b	17.0 b	21.2 g	2.85 a	1.3 a
MB+BFT	153 b	559 d	362 c	44 b	599 b	764 c	51 d	194 cd	64 c	13.6 d	23.5 e	2.76 b	1.2 b
OG+BFT	131 c	573 c	359 c	41 cd	610 b	757 cd	50 d	191 d	65 c	14.3 c	27.3 c	2.75 b	1.2 b
TF+BFT	130 c	547 d	354 c	43 bc	579 c	762 cd	56 c	200 c	61 c	13.6 d	21.9 fg	2.60 c	1.1 c
PR	108 d	495 e	311 d	33 f	662 a	815 a	92 a	277 b	112 a	17.7 a	28.6 b	2.78 b	1.2 b
MB	105 d	605 a	403 a	38 e	606 b	762 cd	49 d	194 cd	64 c	13.1 d	25.4 d	2.60 c	1.1 c
OG	98 e	612 a	373 b	32 f	606 b	730 e	51 d	181 e	64 c	13.5 de	30.3 a	2.58 c	1.1 c
TF	92 f	589 b	377 b	40 de	611 b	753 d	56 c	194 cd	64 c	13.2 ef	22.7 ef	2.49 d	1.0 d
Mean S.E	3	21	9	1	7	7	7	20	7	0.5	0.4	0.03	0.02
Rotation													
1, 0-35 days	118 y	530 z	328 z	39 z	643 x	802 x	79 x	267 x	97 x	12 z	23 z	2.84 x	1.3 x
2, 35-70 days	115 y	582 x	382 x	43 x	572 z	736 z	48 z	192 y	58 z	15 y	24 y	2.55 z	1.0 z
3, 70-105 days	139 x	547 y	356 y	40 y	595 y	762 y	52 y	190 y	63 y	16 x	28 x	2.64 y	1.1 y
Mean S.E	2	19	9	1	4	6	6	20	5	0.5	0.3	0.03	0.02

Pasture treatments followed by different letters (a,b,c,d,e,f) are significantly different (p = 0.05).

Rotation cycle followed by different letters (x,y,z) are significantly different (p = 0.05).

Table 4 Principal component analysis (PCA) results. Data is from a dairy heifer grazing study in Lewiston, Utah in 2017 and 2018. Treatments included monocultures of meadow brome (MB), orchardgrass (OG), perennial ryegrass (PR), tall fescue (TF), and each grass in binary mixture with birdsfoot trefoil (BFT). Principal components were examined to determine which herbage traits were associated with the observed pattern of herbage intake. Principal components, and their corresponding scores, having eigenvalues equal to or greater than 0.8 are shown. Individual loading scores in each principal component that are greater than plus or minus 0.21 are shown.

Herbage Trait	PC 1	PC 2	PC 3	PC 4	PC 5	PC 6
NDF	-0.354					
ADF	-0.325				-0.210	
DNDF	-0.306					
ASH			-0.288	-0.353	0.433	
FAT		0.276	-0.236	0.266	0.290	-0.291
Leaf pubescence			0.211	0.444		0.337
Herbage height			0.459	-0.214		-0.459
Herbage allowance			0.515		0.272	
NDFD		0.415			0.256	
Bulk density			0.224	0.246	0.308	0.496
Leaf softness				0.578		-0.304
Fructan			-0.456			
Lignin		-0.383				
Forage Tannins		-0.293				
BFT percent		-0.311				-0.229
CP	0.252				0.369	
IVTD	0.254	0.271				
WSC	0.263	0.287				
ESC	0.274	0.249				
ME	0.290					
NFC	0.323				-0.266	
Eigenvalue	7.336	4.677	2.944	1.970	1.352	0.844
Proportion of variance	0.349	0.223	0.140	0.094	0.064	0.040
Cumulative proportion of variance	0.349	0.572	0.712	0.806	0.870	0.911

Table 5 Summary of stepwise selection for apparent intake and utilization. Data is from a dairy heifer grazing study in Lewiston, Utah in 2017 and 2018. Treatments included monocultures of meadow brome (MB), orchardgrass (OG), perennial ryegrass (PR), tall fescue (TF), and each grass in binary mixture with birdsfoot trefoil (BFT).

	-------------kg ha^{-1}-------------			----------kg AU^{-1} day^{-1}----------			--------kg $heifer^{-1}$ day^{-1}--------			----------Utilization %----------		
Variable	Estimate	Partial R^2	Pr > F	Estimate	Partial R^2	Pr > F	Estimate	Partial R^2	Pr > F	Estimate	Partial R^2	Pr > F
Intercept	939.04		<.0001	4.46		<.0001	3.98		<.0001	31.68		<.0001
PC 1	-	-	NS	-	-	NS	-	-	NS	-	-	NS
PC 2	-29.76	0.01	0.0470	-	-	NS	-0.13	0.005	0.0473	1.96	0.03	<.0001
PC 3	84.96	0.04	<.0001	0.62	0.08	<.0001	0.36	0.04	<.0001	-3.86	0.10	<.0001
PC 4	122.46	0.08	<.0001	0.60	0.08	<.0001	0.52	0.08	<.0001	3.08	0.06	<.0001
PC 5	26.09	0.004	0.0814	0.12	0.003	0.0862	0.11	0.004	0.0814	-1.98	0.03	<.0001
PC 6	-196.22	0.20	<.0001	-0.84	0.16	<.0001	-0.83	0.20	<.0001	-5.51	0.20	<.0001
Model R^2	0.32			0.32			0.32			0.42		

Table 6 Canonical correlation, adjusted canonical correlation, standard error, squared canonical correlation, eigenvalues, likelihood ratio, and approximate f values from discriminant analysis.Analysis is from a dairy heifer grazing study in Lewiston, Utah in 2017 and 2018.

Canonicals	Canonical Corr.	Adj. Canonical Corr.	Approx. S.E.	Squared Canonical Corr.	Eigenvalues of Inv(E)*H'= CanRsq/(1-CanRsq)				Test of H0: The canonical correlations in the current row and all that follow are zero				
					Value	Diff.	R^2	Cum. R^2	Likelihood Ratio	Approx. F Value	Num DF	Den DF	Pr > F
1	0.96	0.96	0.00	0.92	12.02	6.17	0.60	0.583	0.00194511	182.76	42	2756.7	<.0001
2	0.92	0.92	0.01	0.85	5.85	4.35	0.28	0.867	0.02532233	118.24	30	2354	<.0001
3	0.78	0.77	0.02	0.60	1.50	0.34	0.07	0.941	0.17348789	67.99	20	1954.4	<.0001
4	0.73	.	0.02	0.54	1.16	1.10	0.06	0.997	0.43449752	48.18	12	1561.3	<.0001
5	0.25	0.24	0.04	0.06	0.06	0.06	0.003	1.000	0.93920848	6.28	6	1182	<.0001
6	0.03	-0.01	0.04	0.00	0.00		0.00	1.000	0.9993607	0.19	2	592	0.8275

Table 7 Cross validation summary using discriminant function of canonical discriminant analysis. Analysis is from a dairy heifer grazing study in Lewiston, Utah in 2017 and 2018. Number of observations and percent classified into treatment (TRMT).

TRMT	MB	MB+BFT	OG	OG+BFT	PR	PR+BFT	TF	TF+BFT	Total
MB	70†	3	2	0	0	0	0	0	75
	93.33‡	4	2.67	0	0	0	0	0	100
MB+BFT	8	58	0	0	0	9	0	0	75
	10.67	77.33	0	0	0	12	0	0	100
OG	5	0	62	7	1	0	0	0	75
	6.67	0	82.67	9.33	1.33	0	0	0	100
OG+BFT	2	0	2	71	0	0	0	0	75
	2.67	0	2.67	94.67	0	0	0	0	100
PR	0	0	1	0	74	0	0	0	75
	0	0	1.33	0	98.67	0	0	0	100
PR+BFT	0	3	0	0	0	72	0	0	75
	0	4	0	0	0	96	0	0	100
TF	0	0	0	0	0	0	68	7	75
	0	0	0	0	0	0	90.67	9.33	100
TF+BFT	0	0	0	0	0	0	6	69	75
	0	0	0	0	0	0	8	92	100
Total	85	64	67	78	75	81	74	76	600
	14.17	10.67	11.17	13	12.5	13.5	12.33	12.67	100
Priors	0.125	0.125	0.125	0.125	0.125	0.125	0.125	0.125	
Error Rate	0.0667	0.2267	0.1733	0.0533	0.0133	0.04	0.0933	0.08	0.0933

† Number of instances

‡ Percent

Table 8 Canonical discriminant analysis values. Values are from a dairy heifer grazing study in Lewiston, Utah in 2017 and 2018.

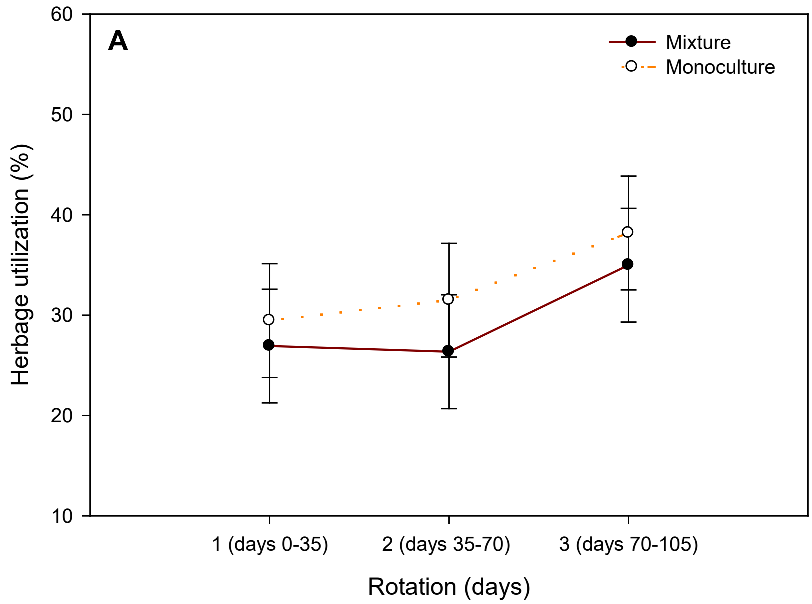
Variable	Canonical 1	Canonical 2	Canonical 3	Canonical 4	Canonical 5	Canonical 6
PC 1	0.122	0.444	0.315	-0.625	0.215	0.501
PC 2	-0.028	-0.344	0.340	-0.350	0.710	0.372
PC 3	0.034	0.053	-0.160	0.428	0.403	0.790
PC 4	0.655	-0.171	0.161	0.278	0.188	-0.635
PC 5	-0.028	0.091	-0.282	-0.057	0.791	-0.531
PC 6	-0.086	0.131	0.700	0.502	0.277	-0.395

Table 9 Herbage mineral contents including calcium (Ca), phosphorus (P), potassium (K), Magnesium (Mg), and ash. Data is from a dairy heifer grazing study in Lewiston, Utah in 2017 and 2018. Treatments included monocultures of meadow brome (MB), orchardgrass (OG), perennial ryegrass (PR), tall fescue (TF), and each grass in binary mixture with birdsfoot trefoil (BFT).

Treatment type	Ca		P		K		Mg		Ash	
	--------------------------------g kg^{-1} herbage--------------------------------									
Mixture	8.8	A	2.8	a	24.0	a	2.6	b	120.5	b
Mono	4.0	B	2.3	b	19.7	b	2.8	a	125.9	a
Mean S.E	4.6		0.06		0.6		0.05		1.7	
Treatment										
PR+BFT	12.5	a	3.0	a	23.4	b	3.1	a	115.5	de
MB+BFT	7.7	b	3.0	a	24.9	a	2.4	c	109.5	f
OG+BFT	7.6	b	2.7	b	24.6	a	3.1	c	117.8	cd
TF+BFT	7.3	b	2.7	b	23.3	b	2.4	c	139.5	a
PR	4.7	c	2.2	c	18.4	d	2.8	b	126.1	b
MB	4.3	cd	2.4	c	20.7	c	2.7	b	112.4	ef
OG	3.9	d	2.4	d	21.3	c	3.1	a	121.1	bc
TF	3.0	e	2.1	e	18.5	d	2.7	b	144.0	a
Mean S.E	0.5		0.07		0.6		0.1		2	
Rotation										
1, 0-35 days	5.5	z	2.5	y	24.1	x	2.1	z	107.7	z
2, 35-70 days	6.4	y	2.4	z	19.1	z	2.8	y	127.4	y
3, 70-105 days	7.3	x	2.8	x	22.4	y	3.3	x	134.4	x
Mean S.E	0.5		0.07		0.6		0.03		1.7	

Pasture treatments and treatment types followed by different letters (a,b,c,d,e,f) are significantly different (p = 0.05).

Rotation cycle followed by different letters (x,y,z) are significantly different (p = 0.05).



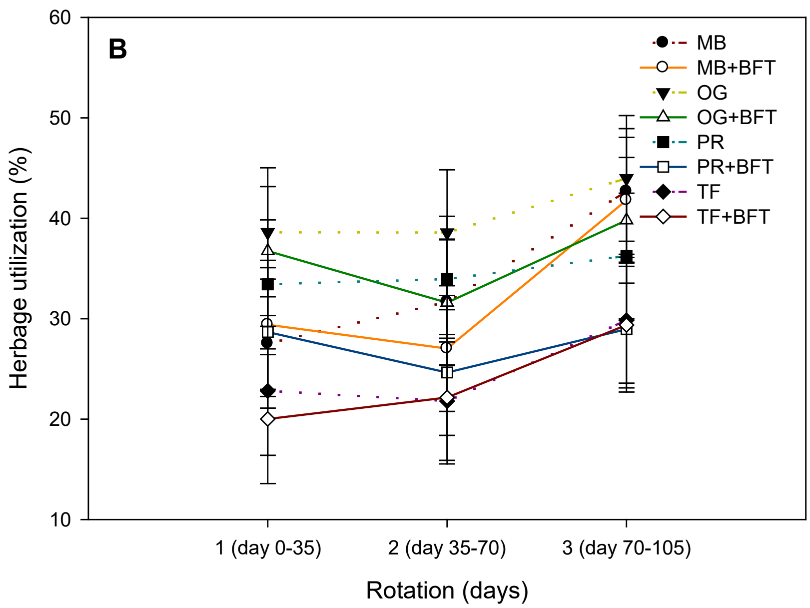
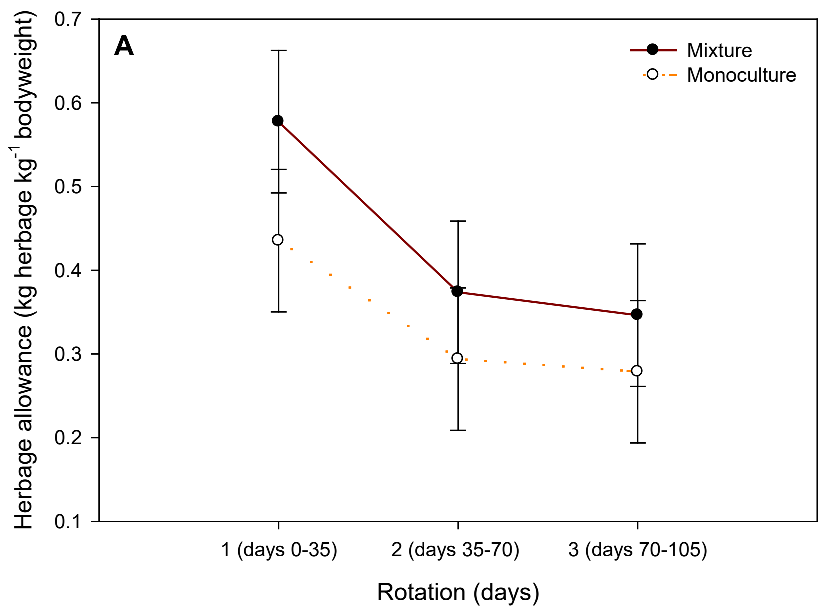


Figure 2: Percent of herbage utilized of monocultures and mixtures (A), and of individual grasses and mixtures (B). Figures are for pasture treatments from a dairy heifer grazing study in Lewiston, Utah. Pasture treatments included meadow brome(MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT). Rotational stocking was used with three 35-day cycles. Bars represent plus or minus the standard error of the means. N=4 and N=6 for treatment types (A) and treatments (B) respectively.



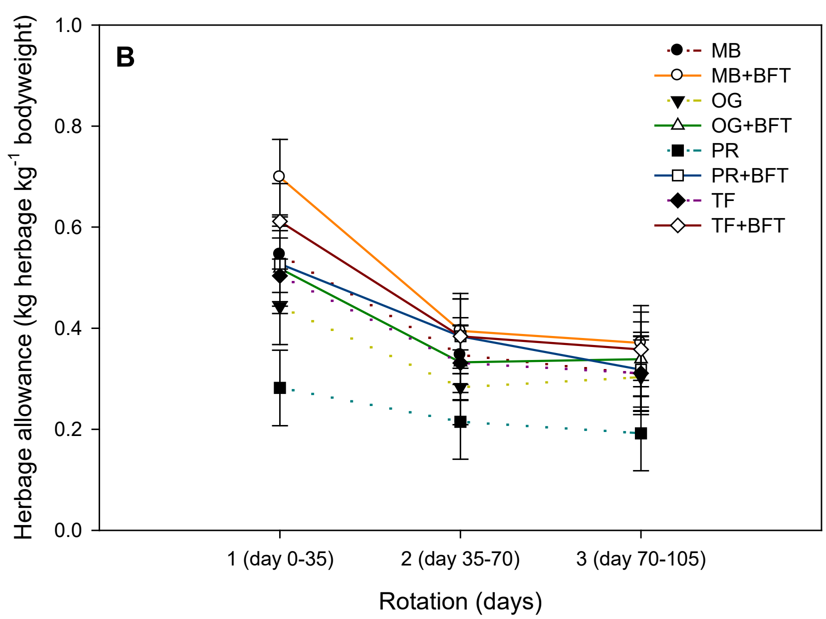
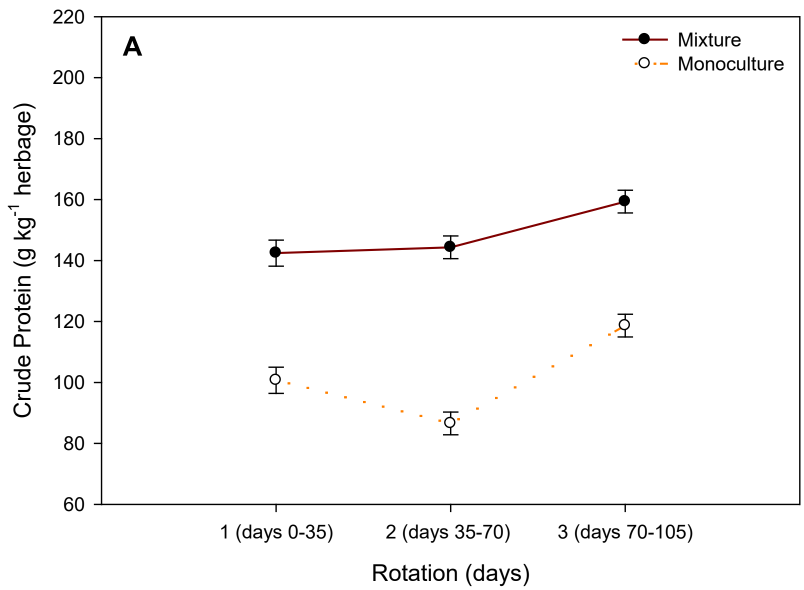


Figure 3: Herbage allowance of monocultures and mixtures (A), and herbage allowance of individual grasses and mixtures (B). Figures are for pasture treatments from a dairy heifer grazing study in Lewiston, Utah. Pasture treatments included meadow brome (MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT). Rotational stocking was used with three 35-day cycles. Bars represent plus or minus the standard error of means. N=4 and N=6 for treatment types (A) and treatments (B) respectively.



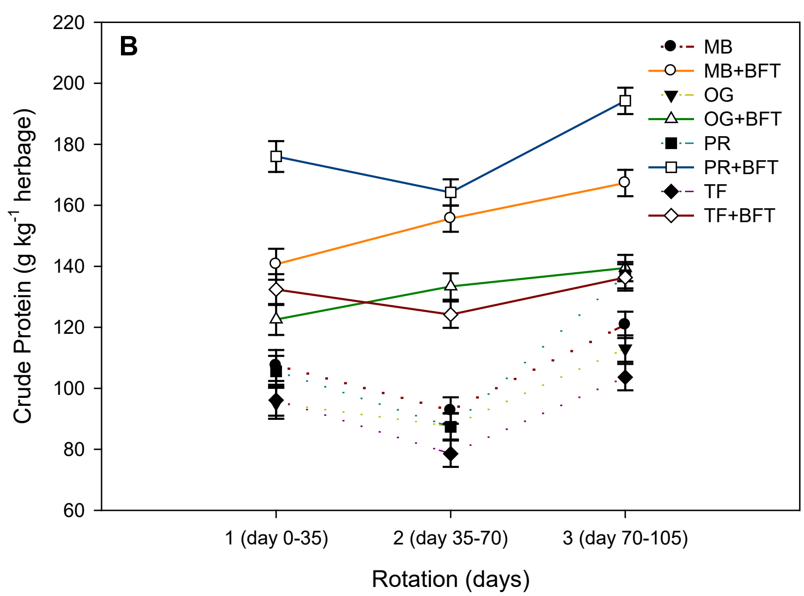


Figure 4: Crude protein content of monocultures and mixtures (A), and crude protein content of individual grasses and mixtures (B). Figures are for pasture treatments from a dairy heifer grazing study in Lewiston, Utah. Pasture treatments included meadow brome (MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT). Rotational stocking was used with three 35-day cycles. Bars represent plus or minus the standard error of mean. N=4 and N=6 for treatment types (A) and treatments (B) respectively.

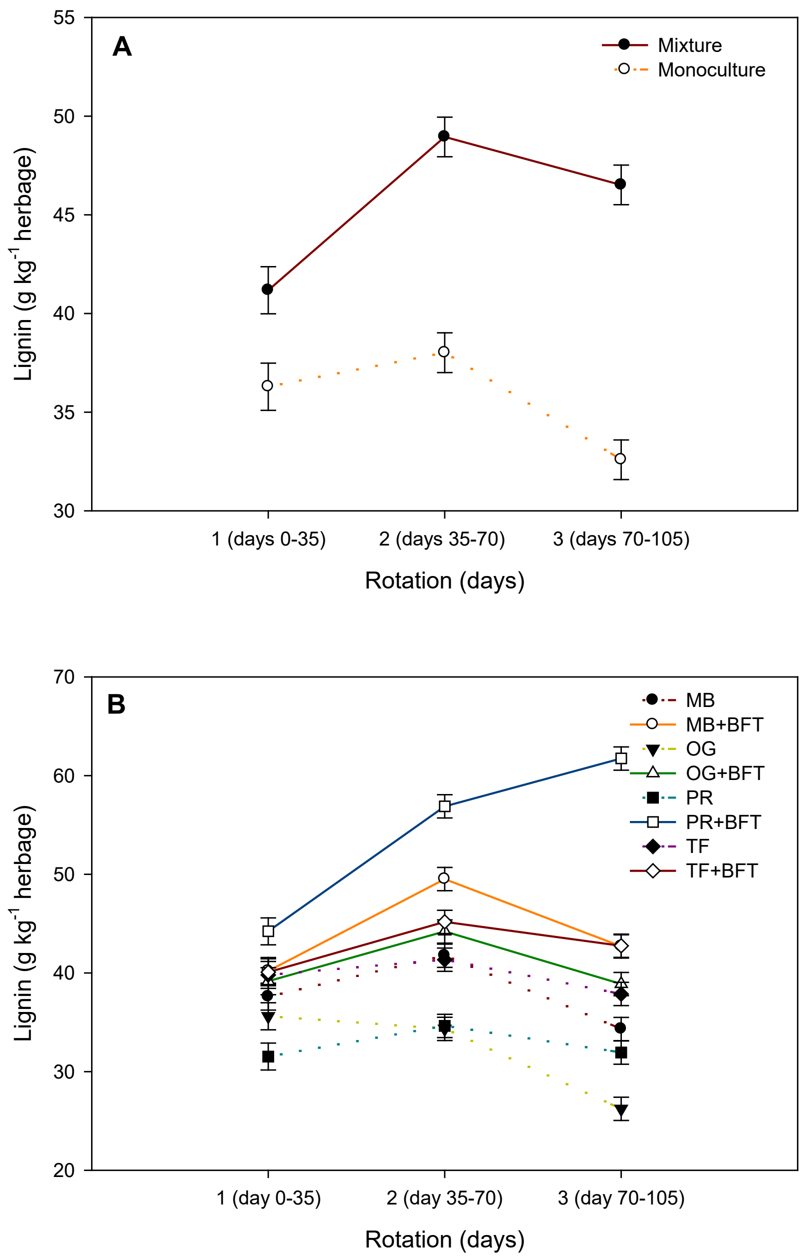


Figure 5: Lignin content of monocultures and mixtures (A), and lignin content of individual grasses and mixtures (B). Figures are for pasture treatments from a dairy heifer grazing study in Lewiston, Utah. Pasture treatments included meadow brome (MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT). Rotational stocking was used with three 35-day cycles. Bars represent plus or minus the standard error of means. N=4 and N=6 for treatment types (A) and treatments (B) respectively.

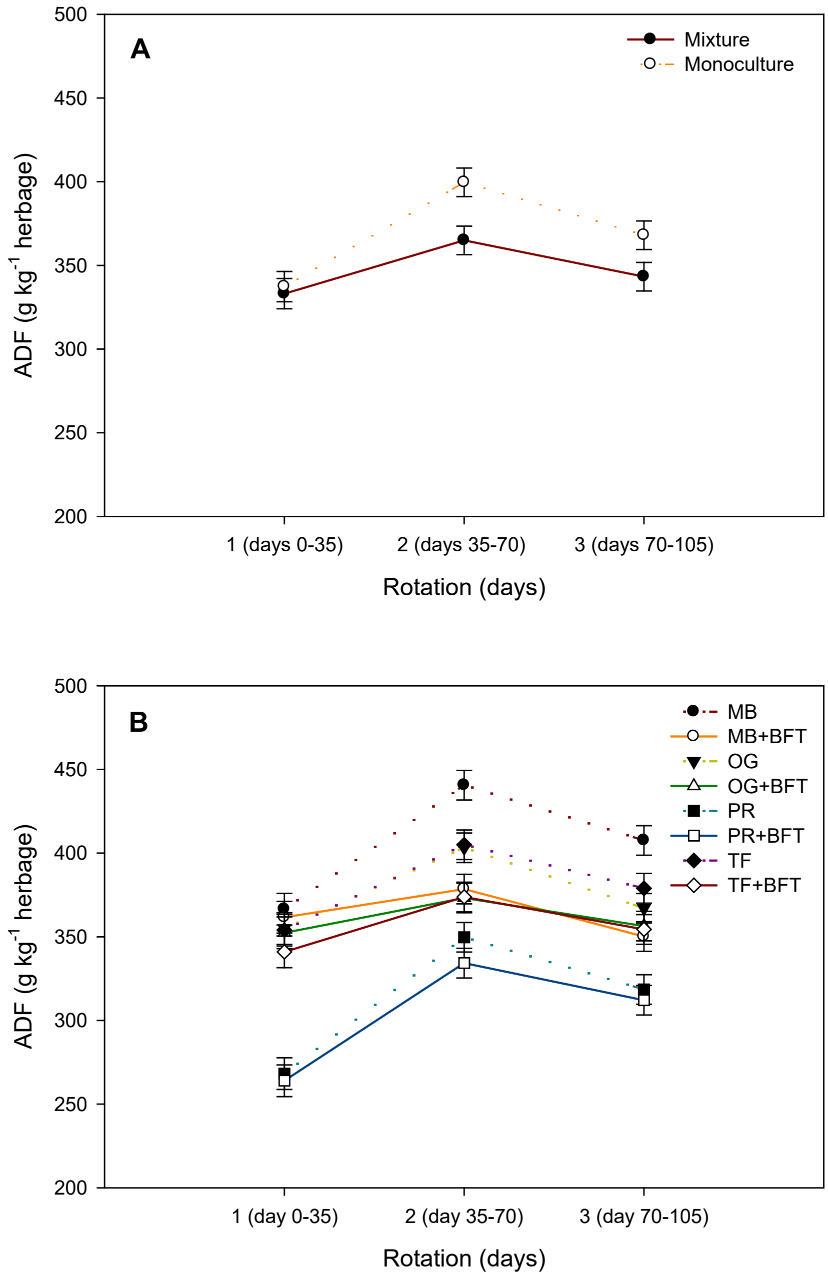
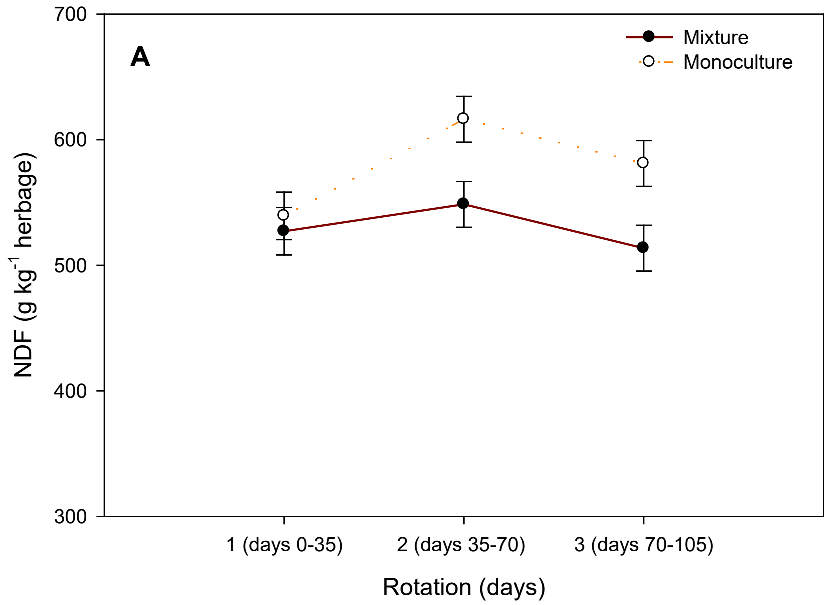


Figure 6: ADF content of monocultures and mixtures (A), and ADF content of individual grasses and mixtures (B) Figures are for pasture treatments from a dairy heifer grazing study in Lewiston, Utah. Pasture treatments included meadow brome (MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT). Rotational stocking was used with three 35-day cycles. Bars represent plus or minus the standard error of means. N=4 and N=6 for treatment types (A) and treatments (B) respectively.



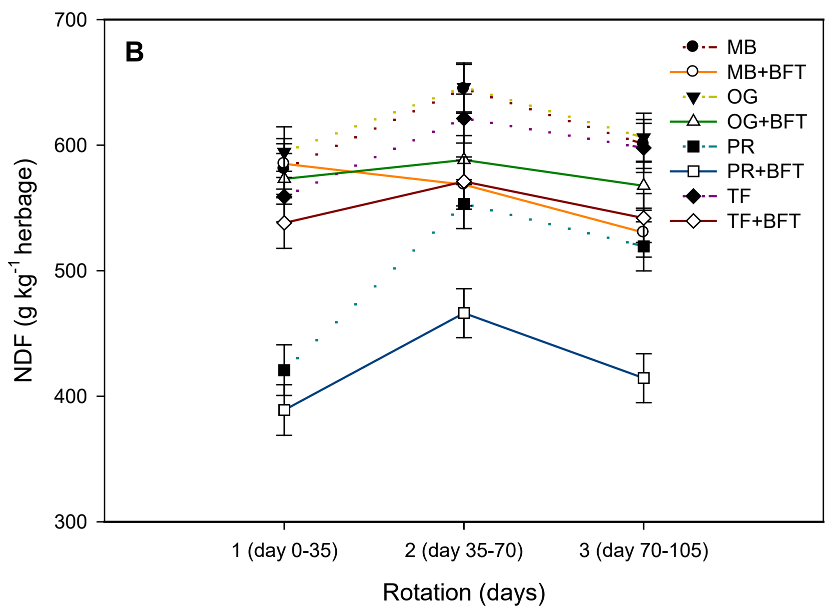


Figure 7: NDF content of monocultures and mixtures (A), and NDF content of individual grasses and mixtures (B) Figures are for pasture treatments from a dairy heifer grazing study in Lewiston, Utah. Pasture treatments included meadow brome (MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT). Rotational stocking was used with three 35-day cycles. Bars represent plus or minus the standard error of means. N=4 and N=6 for treatment types (A) and treatments (B) respectively.

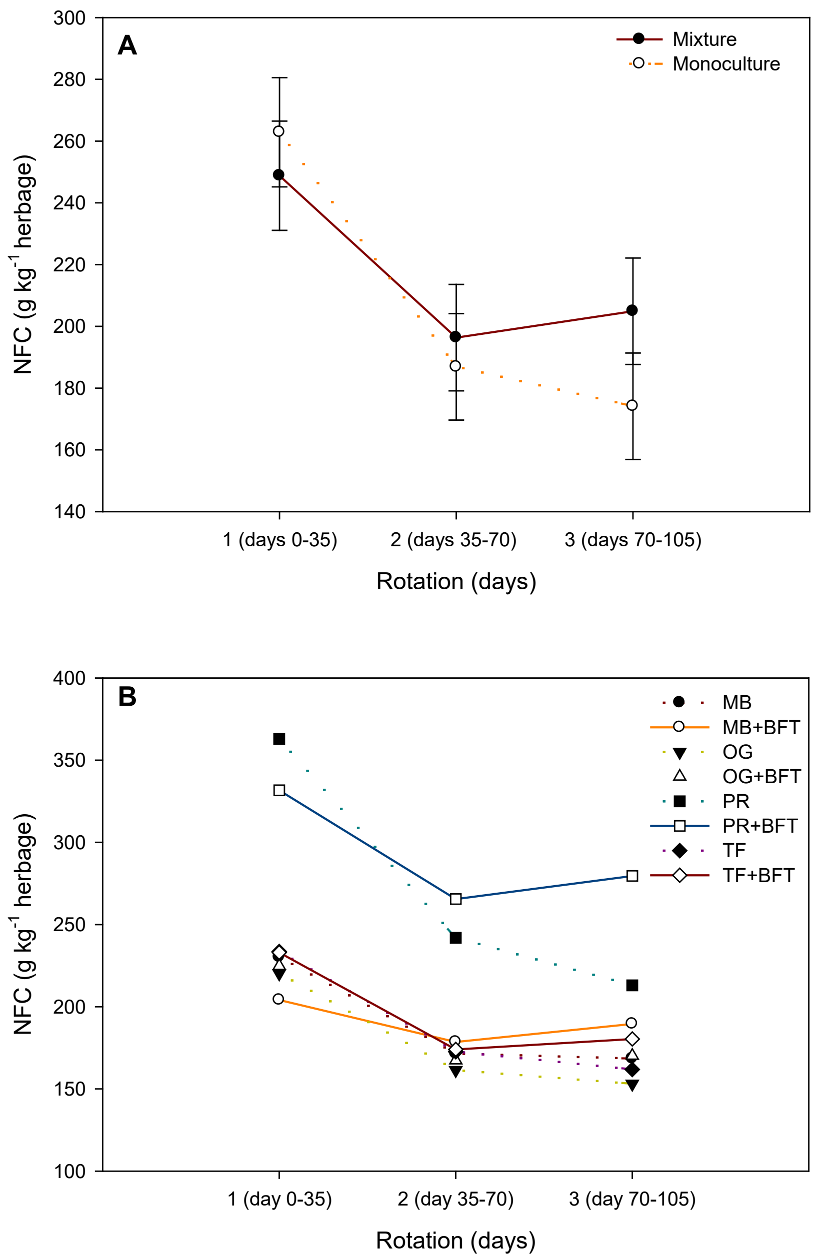


Figure 8: Non-fibrous carbohydrates of monocultures and mixtures (A), and non-fibrous carbohydrate content of individual grasses and mixtures (B) Figures are for pasture treatments from a dairy heifer grazing study in Lewiston, Utah. Pasture treatments included meadow brome (MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT). Rotational stocking was used with three 35-day cycles. Bars represent plus or minus the standard error of means. N=4 and N=6 for treatment types (A) and treatments (B) respectively.

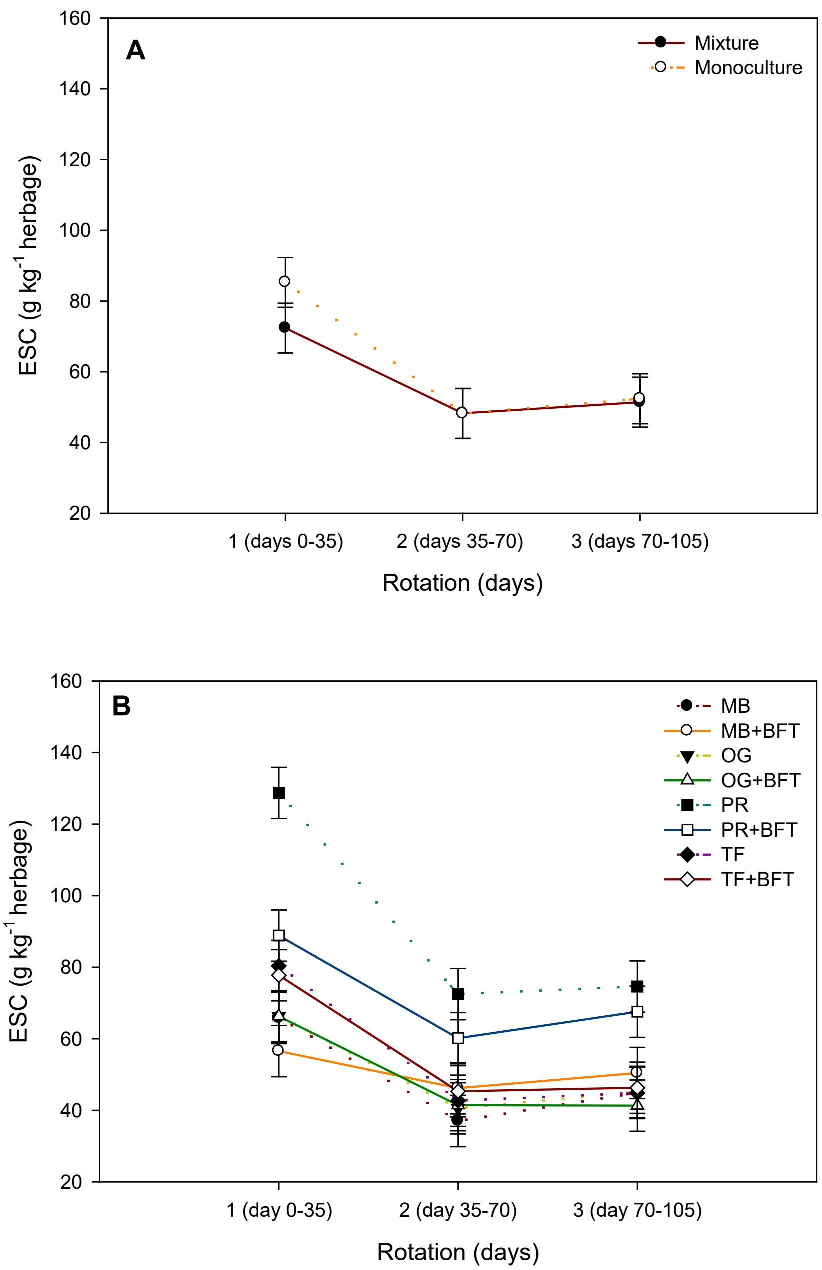


Figure 9 Ethanol soluble carbohydrates of monocultures and mixtures (A), and ethanol soluble carbohydrates of individual grasses and mixtures (B) Figures are for pasture treatments from a dairy heifer grazing study in Lewiston, Utah. Pasture treatments included meadow brome (MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT). Rotational stocking was used with three 35-day cycles. Bars represent plus or minus the standard error of means. N=4 and N=6 for treatment types (A) and treatments (B) respectively.

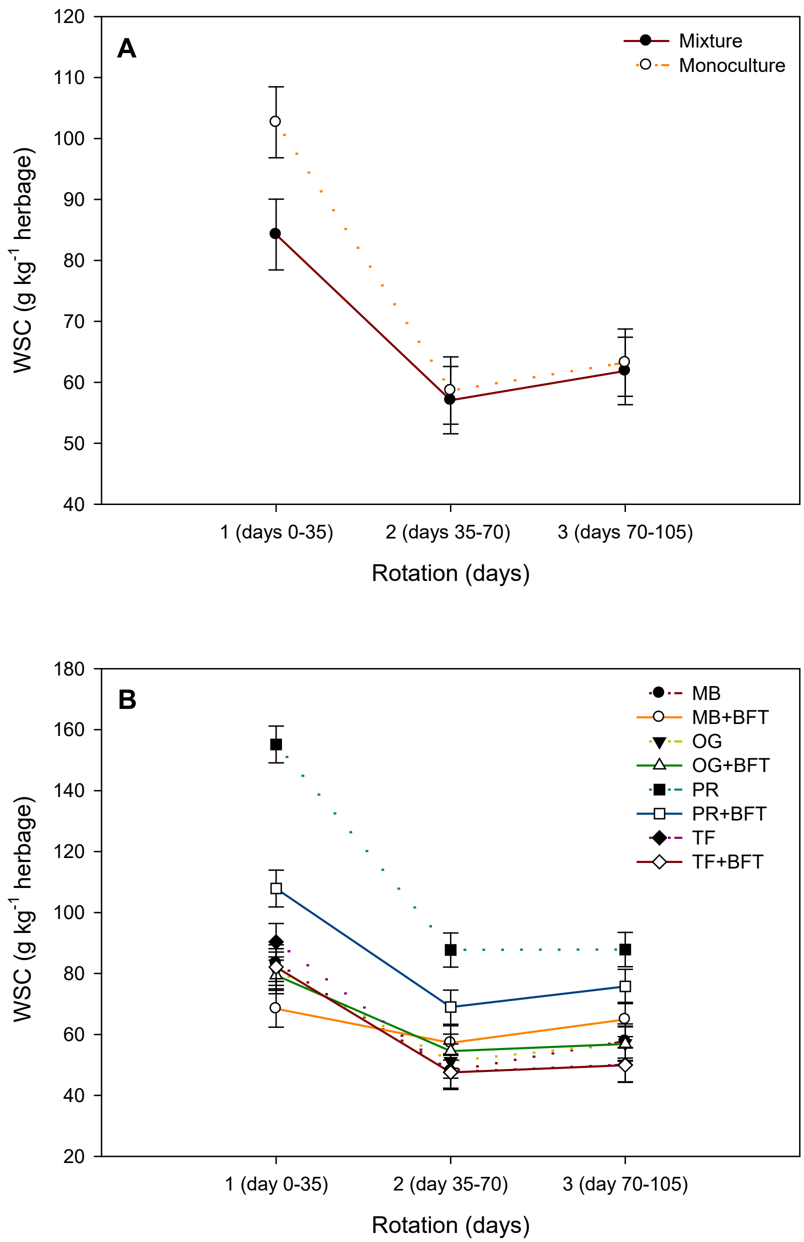
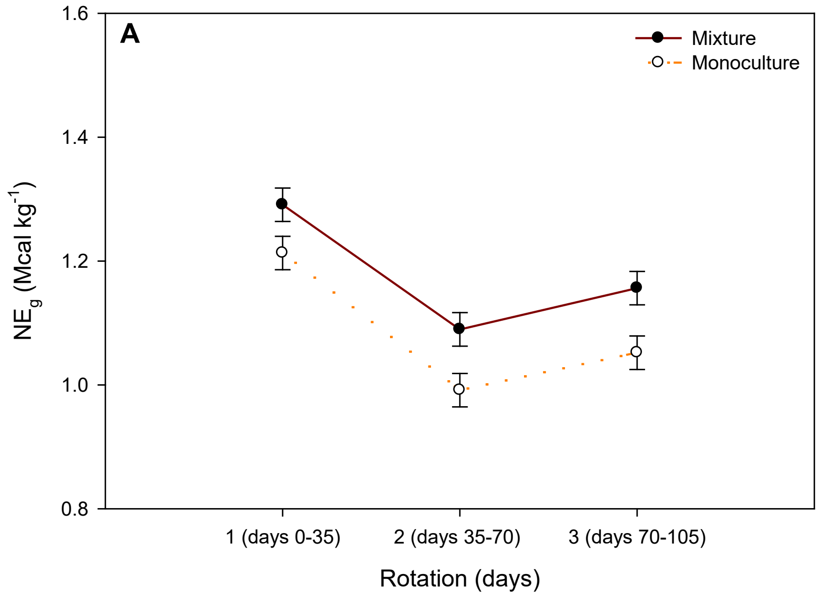


Figure 10: Water soluble carbohydrate content of monocultures and mixtures (A), and water-soluble carbohydrate content of individual grasses and mixtures (B) Figures are for pasture treatments from a dairy heifer grazing study in Lewiston, Utah. Pasture treatments included meadow brome (MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT). Rotational stocking was used with three 35-day cycles. Bars represent plus or minus the standard error of means. N=4 and N=6 for treatment types (A) and treatments (B) respectively.



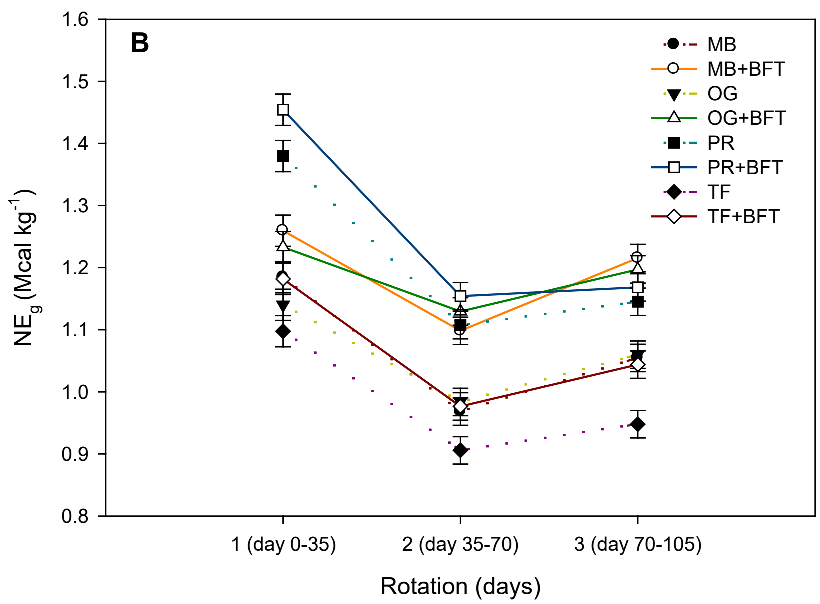


Figure 11: Net energy gain (NE_g) of monocultures and mixtures (A), and net energy gain of individual grasses and mixtures (B) Figures are for pasture treatments from a dairy heifer grazing study in Lewiston, Utah. Pasture treatments included meadow brome (MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT). Rotational stocking was used with three 35-day cycles. Bars represent plus or minus the standard error of means. N=4 and N=6 for treatment types (A) and treatments (B) respectively.

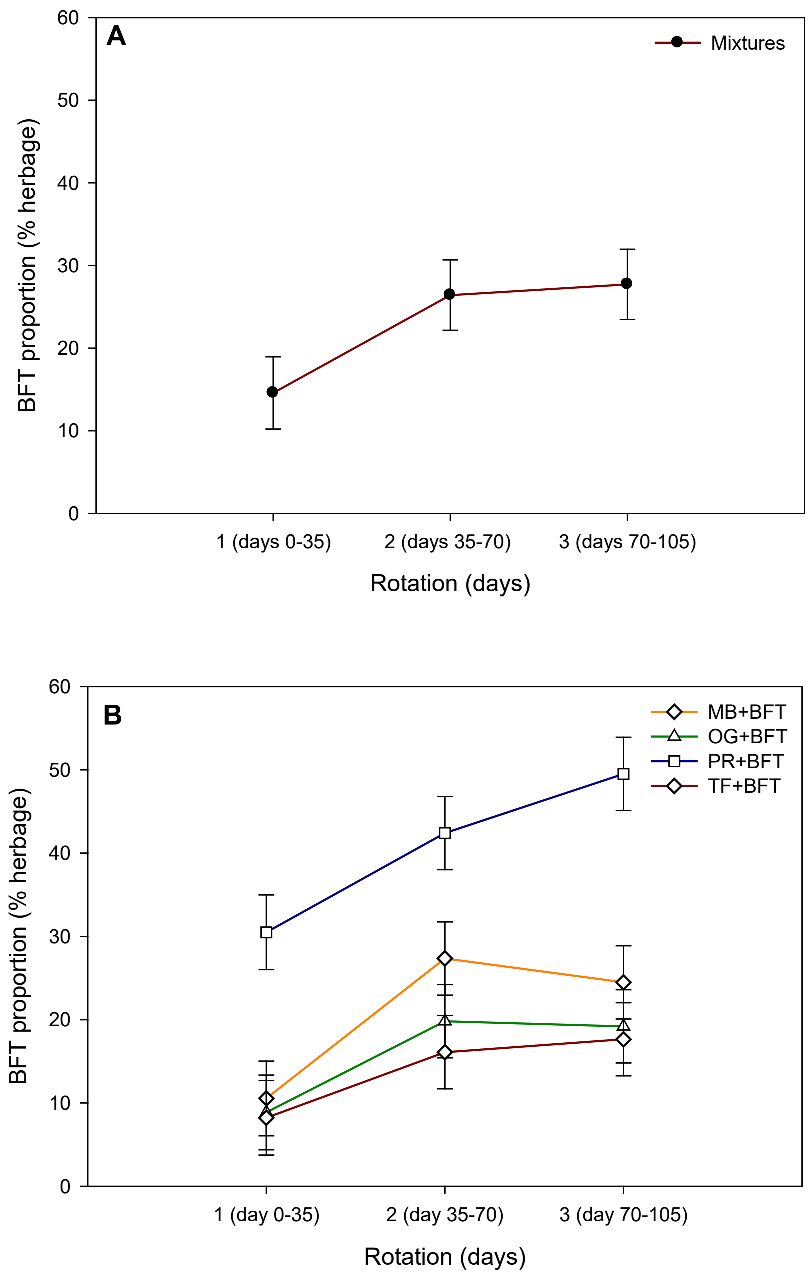
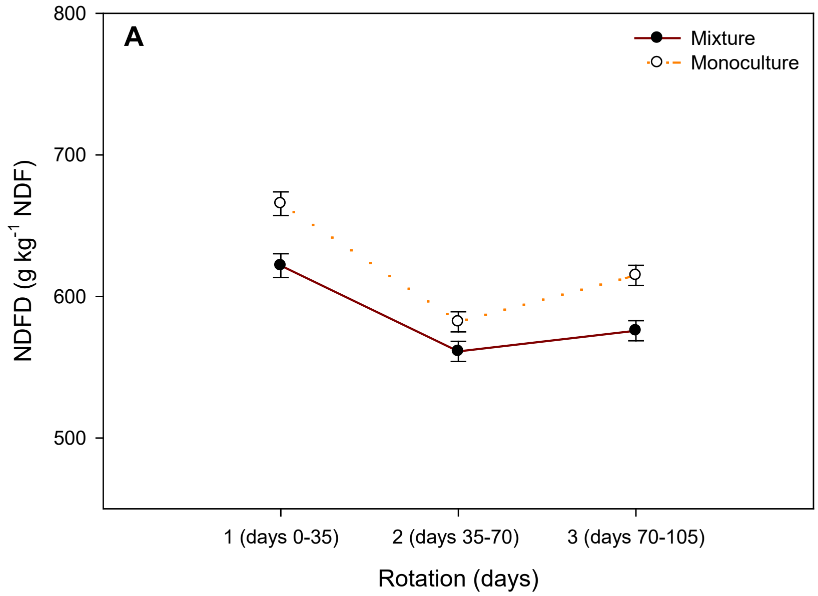


Figure 12: Birdsfoot trefoil content as a proportion of total herbage averaged across all mixtures (A) and for individual mixtures (B). Figures are for pasture treatments from a dairy heifer grazing study in Lewiston, Utah. Pasture treatments included meadow brome (MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT). Rotational stocking was used with three 35-day cycles. Bars represent plus or minus the standard error of means. N=4 and N=6 for treatment types (A) and treatments (B) respectively.



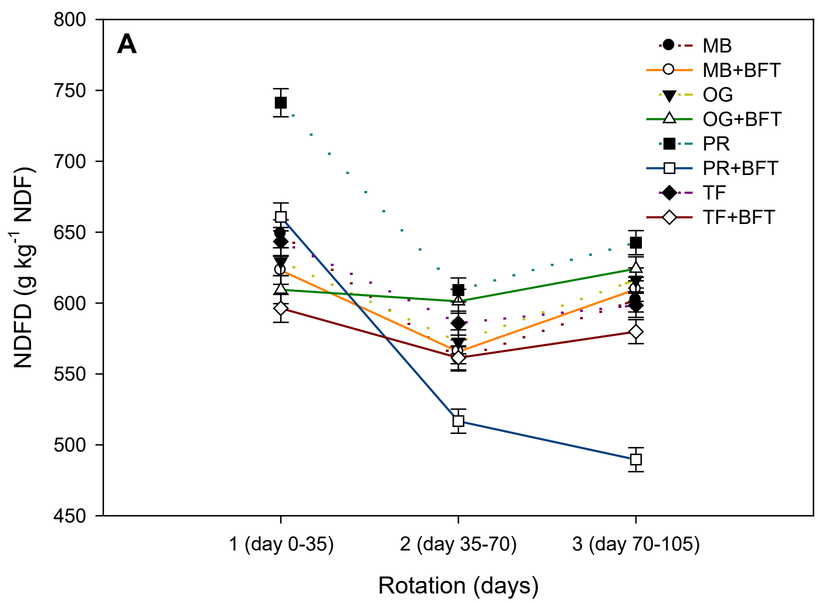


Figure 13: NDF digestibility (NDFD)of monocultures and mixtures (A), and NDF digestibility of individual grasses and mixtures (B) Figures are for pasture treatments from a dairy heifer grazing study in Lewiston, Utah. Pasture treatments included meadow brome (MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT). Rotational stocking was used with three 35-day cycles. Bars represent plus or minus the standard error of means. N=4 and N=6 for treatment types (A) and treatments (B) respectively.

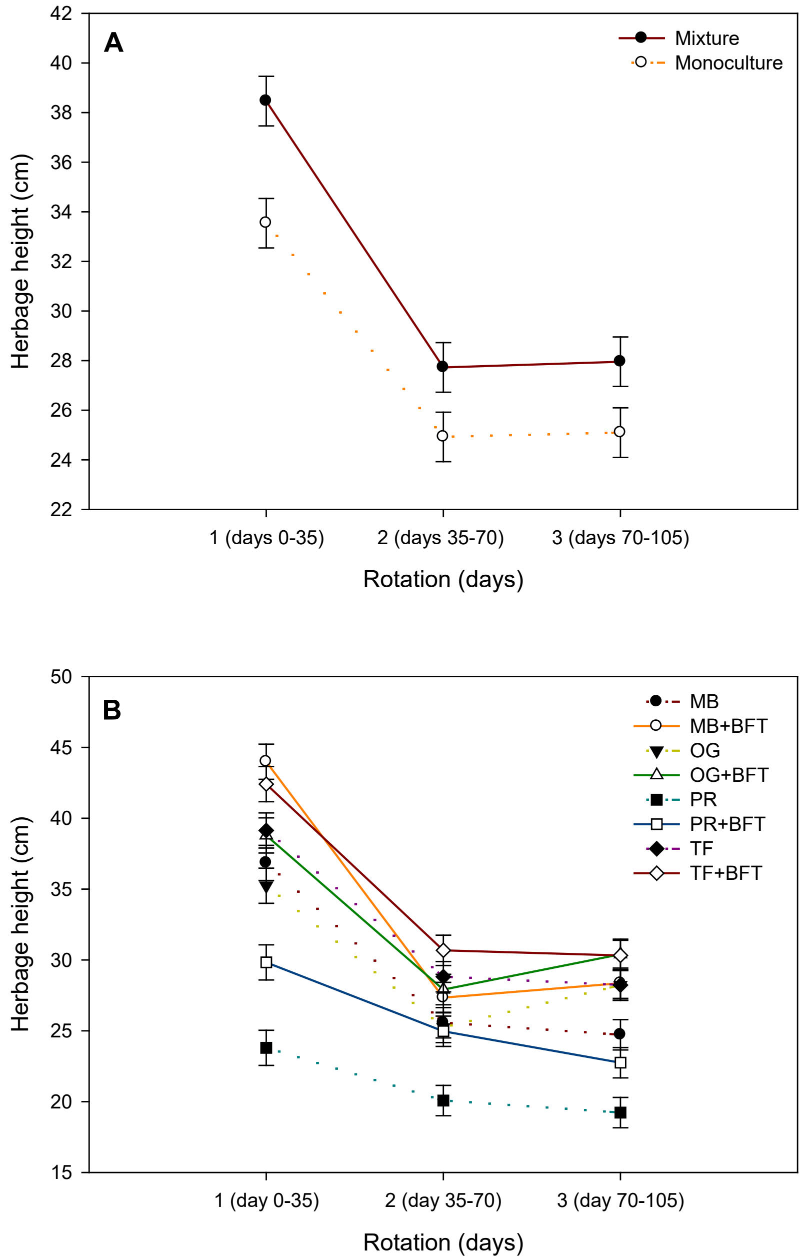


Figure 14: Pasture treatment herbage height of monocultures and mixtures (A), and herbage height of individual grasses and mixtures (B) Figures are for pasture treatments from a dairy heifer grazing study in Lewiston, Utah. Pasture treatments included meadow brome (MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT). Rotational stocking was used with three 35-day cycles. Bars represent plus or minus the standard error of means. N=4 and N=6 for treatment types (A) and treatments (B) respectively.

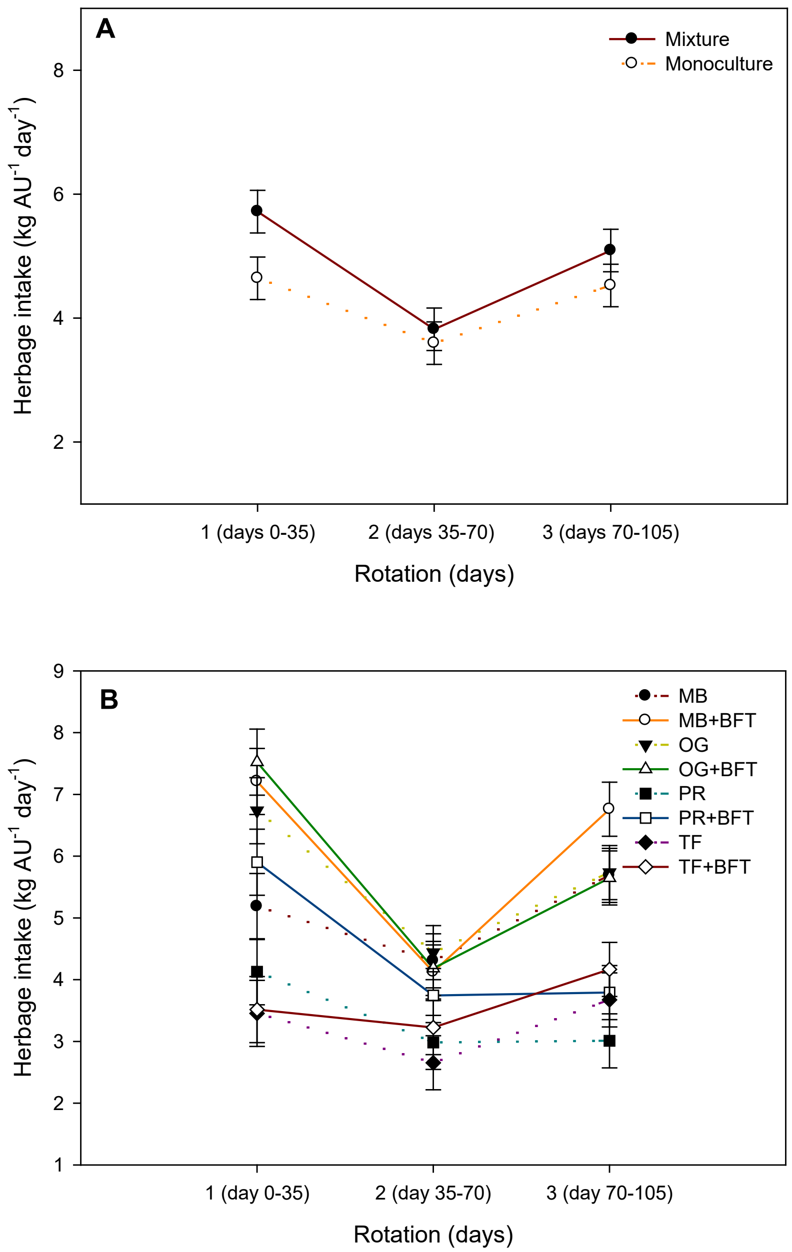


Figure 15: Herbage intake in kg AU^{-1} day^{-1} of monocultures and mixtures (A), and herbage intake of individual grasses and mixtures (B) Figures are for pasture treatments from a dairy heifer grazing study in Lewiston, Utah. Pasture treatments included meadow brome (MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT). Rotational stocking was used with three 35-day cycles. Bars represent plus or minus the standard error of means. N=4 and N=6 for treatment types (A) and treatments (B) respectively.

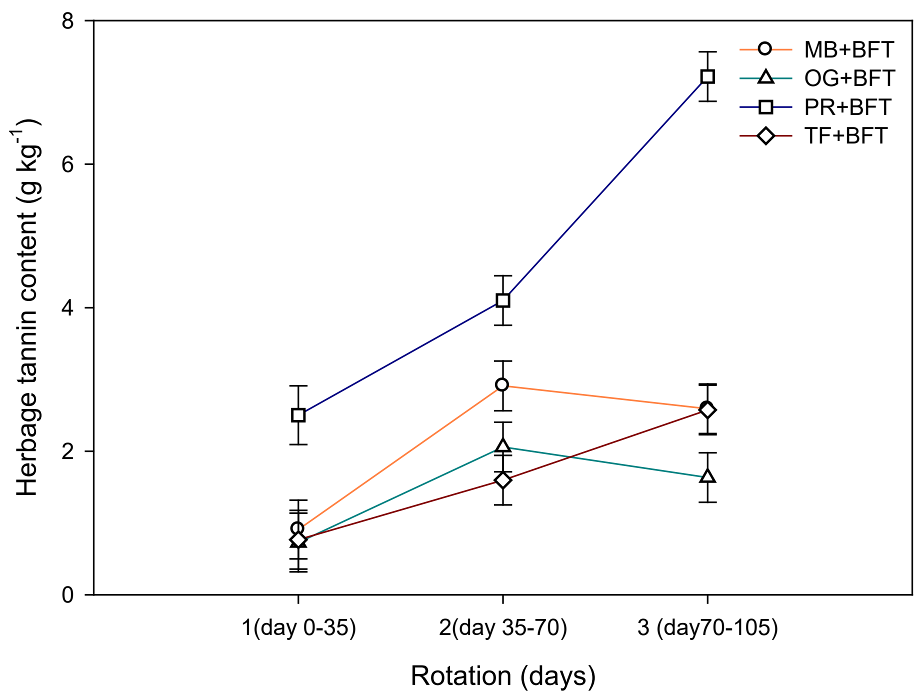


Figure 16: Total tannins per kg of herbage for pasture treatments. Pasture treatments were from a dairy heifer grazing study in Lewiston, Utah and included meadow brome (MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT). Rotational stocking was used with three 35-day cycles. Bars represent plus or minus the standard error of means. N=6

APPENDIX A

Table 10 Analysis of variance (ANOVA) significance of herbage mass and composition traits. Analysis is from a dairy heifer grazing study in Lewiston, Utah in 2017 and 2018.

Mixture vs. monoculture	Pre-graze herbage mass	Herbage allowance	BFT proportion	BFT Tannins	Forage Tannins	Height	Bulk Density
Treatment type	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
Rotation	<.0001	<.0001	0.0005	<.0001	0.0002	<.0001	0.989
Treatment type*rotation	0.9200	0.0465	0.0005	<.0001	0.0002	0.4920	0.9975
Individual treatments							
Treatment	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
Rotation	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	0.0006
Treatment*Rotation	0.0057	0.0009	<.0001	<.0001	<.0001	<.0001	0.2039

Table 11 Analysis of variance (ANOVA) significance for herbage nutritive value traits. Analysis is from a dairy heifer grazing study in Lewiston, Utah in 2017 and 2018.

Mixture vs. monoculture	Ash	CP	NDF	ADF	NDFD	IVTD	Fat	WSC	Lignin
Treatment type	<.0001	<.0001	<.0001	<.0001	<.0001	0.4175	<.0001	0.0001	<.0001
Rotation	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
Treatment type*rotation	0.1655	0.0427	0.0596	0.1261	0.2034	0.1211	0.0300	0.0651	0.0013
	NE_g	Ca	P	K	Mg	ESC	Starch	Fructan	NFC
	<.0001	<.0001	<.0001	<.0001	<.0001	0.0002	0.2433	0.0007	0.0016
	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
	0.7720	0.2246	0.0056	0.0488	<.0001	0.1307	<.0001	0.5975	0.0747

Individual treatments	Ash	CP	NDF	ADF	NDFD	IVTD	Fat	WSC	Lignin
Treatment	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
Rotation	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
Treatment*Rotation	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
	NE_g	Ca	P	K	Mg	ESC	Starch	Fructan	NFC
	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001
	<.0001	<.0001	0.0004	0.0016	<.0001	<.0001	<.0001	<.0001	<.0001

Table 12 Analysis of variance (ANOVA) significance for intake and utilization. Analysis is from a dairy heifer grazing study in Lewiston, Utah in 2017 and 2018.

Mixture vs. monoculture	Intake kg/ha	Intake kg/au/day	Intake kg $heif.^{-1}$ day^{-1}	util. %
Treatment type	0.0056	0.0325	0.0056	<.0001
Rotation	<.0001	<.0001	<.0001	<.0001
Treatment type*rotation	0.5129	0.4518	0.5129	0.6883
Individual treatments				
Treatment	<.0001	<.0001	<.0001	<.0001
Rotation	<.0001	<.0001	<.0001	<.0001
Treatment*Rotation	0.0897	0.1076	0.0897	0.0259

Table 13 Linear regression parameter estimates and associated R^2 values for rising plate meter herbage mass predictions. Data is from a dairy heifer grazing study in Lewiston, Utah. Pasture treatments included meadow brome (MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT). X-intercept was forced to zero (Dillard et al., 2016).

	2017 Pre-graze							2018 Pre-graze						
	Variable	Parameter Estimate	Standard Error	t Value	Pr > |t|	R^2	Adj. R^2	Variable	Parameter Estimate	Standard Error	t Value	Pr > |t|	R^2	Adj. R^2
MB	DAY0RPM	79.370	4.434	17.9	<.0001	0.887	0.884	DAY0RPM	101.550	3.899	26.05	<.0001	0.939	0.938
MB+BFT	DAY0RPM	89.689	4.617	19.42	<.0001	0.902	0.900	DAY0RPM	107.093	4.406	24.3	<.0001	0.931	0.929
OG	DAY0RPM	65.677	2.952	22.25	<.0001	0.924	0.922	DAY0RPM	92.734	3.864	24	<.0001	0.929	0.927
OG+BFT	DAY0RPM	75.259	3.440	21.88	<.0001	0.921	0.919	DAY0RPM	93.852	3.189	29.43	<.0001	0.952	0.951
PR	DAY0RPM	65.130	1.833	35.54	<.0001	0.969	0.969	DAY0RPM	90.114	3.208	28.09	<.0001	0.947	0.946
PR+BFT	DAY0RPM	95.755	2.972	32.22	<.0001	0.962	0.961	DAY0RPM	111.729	3.933	28.41	<.0001	0.948	0.947
TF	DAY0RPM	67.165	2.857	23.51	<.0001	0.931	0.929	DAY0RPM	90.033	3.188	28.24	<.0001	0.948	0.947
TF+BFT	DAY0RPM	77.809	4.269	18.22	<.0001	0.890	0.887	DAY0RPM	95.968	3.175	30.22	<.0001	0.954	0.953

	2017 Post-graze							2018 Post-graze						
	Variable	Parameter Estimate	Standard Error	t Value	Pr > |t|	R^2	Adj. R^2	Variable	Parameter Estimate	Standard Error	t Value	Pr > |t|	R^2	Adj. R^2
MB	DAY7RPM	74.930	2.963	25.29	<.0001	0.940	0.938	DAY7RPM	120.478	8.960	13.45	<.0001	0.804	0.800
MB+BFT	DAY7RPM	91.633	4.188	21.88	<.0001	0.921	0.919	DAY7RPM	124.720	9.875	12.63	<.0001	0.784	0.779
OG	DAY7RPM	54.922	1.789	30.7	<.0001	0.958	0.957	DAY7RPM	97.302	6.645	14.64	<.0001	0.830	0.826
OG+BFT	DAY7RPM	67.652	2.325	29.09	<.0001	0.954	0.953	DAY7RPM	110.853	7.432	14.92	<.0001	0.835	0.831
PR	DAY7RPM	57.886	3.113	18.6	<.0001	0.894	0.891	DAY7RPM	84.527	5.394	15.67	<.0001	0.848	0.845
PR+BFT	DAY7RPM	111.131	4.448	24.99	<.0001	0.938	0.937	DAY7RPM	141.295	8.630	16.37	<.0001	0.859	0.856
TF	DAY7RPM	65.301	3.123	20.91	<.0001	0.914	0.912	DAY7RPM	99.235	5.224	18.99	<.0001	0.891	0.889
TF+BFT	DAY7RPM	75.311	2.678	28.12	<.0001	0.951	0.950	DAY7RPM	115.569	5.343	21.63	<.0001	0.914	0.912

APPENDIX B

B1: Bingham on Farm Trial

Question

Can herbage nutritive value and predicted milk production be improved in organic lactating dairy cows using grasses of varying energy with grass-birdsfoot trefoil mixtures?

Objective:

Determine which grass and/or grass-birdsfoot trefoil mixture has the greatest nutritive value, predicted intake and predicted milk production in a rotationally grazed dairy operation.

Materials and Methods:

Pasture treatments were endophyte-free tall fescue (‘Fawn’, TF), meadow bromegrass (‘Cache’, MB), high-sugar orchardgrass (‘Quickdraw’, OG), and high-sugar perennial ryegrass (‘Amazon’, PR) in monoculture and as binary mixtures with birdsfoot trefoil (‘Pardee’, BFT). Treatments were arranged in a randomized complete block (RCB) design with two replicates. Seeding occurred in April 2015 with a Great Plains drill (Great Plains Ag, Salina, KS, USA). For grass monocultures, TF was seeded at 16.8 kg pure live seed (PLS) ha^{-1}, MB was seeded at 18.0 kg PLS ha^{-1}, PR was seeded at 17.5 kg PLS ha^{-1} and OG at 14.9 kg PLS ha^{-1}. In binary mixtures, grasses were seeded at 60% of monocultures whereas, the BFT was seeded at 6.4 kg PLS ha^{-1} in all the grass-legume treatments.

Animals in the study consisted of lactating, crossbred Holstein, Montbeliard, Swedish red dairy cows in an organic dairy operation in Weston, Idaho. Rotational stocking was used with a typical stocking period of 24 hours, followed by a rest period ranging from 21 to 45 days depending on herbage growth and climatic conditions (freezing dates, etc.). Cows were on pasture for at least 120 days per growing season which consisted of five and seven rotations in 2017 and 2018, respectively.

The herbage inside two randomly placed 0.25 m^2 hoops was clipped before and after grazing in 2017 and before grazing in 2018 at a height of 7.6 and 3.8 cm respectively. Samples were dried, weighed, and ground to pass through a 1-mm screen using a Thomas Wiley Laboratory Model 4 mill (Arthur H Thomas Co, Swedesboro, NJ, USA), and were scanned with a Foss XDS near-infrared reflectance spectroscopy (NIRS) instrument (Foss, Eden Prairie, MN, USA) to determine herbage nutritive value of the feed on offer.

Results:

The perennial ryegrass treatments generally had the most favorable nutritive characteristics in many respects (Table 14), but they did not persist well and were largely overtaken by weeds by the end of 2018. The tall fescue treatments were considered the benchmark in this study based on previous research. While they were not as readily consumed as the other treatments, likely due to course leaves, it consistently produced large amounts of herbage and had more predicted milk $acre^{-1}$ than any other treatment (Table 14). The meadow brome and orchardgrass were comparable in many respects. They often had intermediate nutritive values, with meadow brome treatments being slightly greater than the orchardgrass treatments (Table 14). However, since the orchardgrass treatments had the second highest herbage production, they were usually similar to the meadow brome treatments in milk production per acre (Table 14).

Table 14 Herbage production, nutritive values, and predicted intake and milk production from the Bingham on-farm trial conducted in 2017 and 2018 in Weston, Idaho. Pasture treatments included meadow brome (MB), meadow brome and birdsfoot trefoil (MB+BFT), orchardgrass (OG), orchardgrass and birdsfoot trefoil (OG+BFT), perennial ryegrass (PR), perennial ryegrass and birdsfoot trefoil (PR+BFT), tall fescue (TF), and tall fescue and birdsfoot trefoil (TF+BFT).

Trmt.	Herbage production		CP		NDF		NDFD		IVTD		Lignin		WSC	
	lb. acre^{-1}				--------------------%--------------------									
TF+BFT	2640	A	15.3	D	52.9	AB	63.5	CD	79.5	CD	3.5	CD	6.2	E
TF+N	2629	A	14.6	D	54.9	A	62.3	D	77.2	E	3.7	C	6.3	E
OG+BFT	1774	B	15.8	D	54.0	AB	68.4	AB	81.1	C	3.0	E	7.4	DE
OG+N	1445	BC	14.9	D	54.2	AB	65.6	BC	79.3	D	2.9	E	8.2	CD
MB+N	1262	BC	16.1	D	52.0	B	65.8	BC	83.4	B	3.4	D	8.9	BC
MB+BFT	1144	C	20.0	B	43.5	C	67.1	B	84.6	B	4.1	B	9.6	AB
PR+BFT	579	D	23.3	A	29.0	E	61.1	D	88.3	A	5.1	A	10.8	A
PR+N	454	D	18.1	C	40.5	D	71.0	A	87.0	A	3.7	C	10.2	AB
Mean S.E	565		1.7		2.8		2.0		1.8		0.2		0.5	

	TDN		NE_L		Pred. DMI		2017 Milk day^{-1}†		2017 Milk acre^{-1}†		2018 Milk day^{-1}†		2018 Milk acre^{-1}†	
			Mcal kg^{-1}		lb. cow^{-1} day^{-1}		-------------lb.-------------				-------------lb.-------------			
TF+BFT	62.2	E	0.6	E	2.8	CD	58	CD	2638	AB	50	D	5281	A
TF+N	57.1	F	0.6	F	2.6	D	45	E	2704	A	39	E	3997	B
OG+BFT	67.8	B	0.7	B	2.9	CD	67	C	2439	ABC	59	C	3949	B
OG+N	63.2	DE	0.6	DE	2.9	CD	54	DE	1919	C	56	CD	2763	C
MB+N	65.2	C	0.7	C	2.9	CD	60	CD	1961	C	59	C	2526	C
MB+BFT	71.0	A	0.7	A	3.5	B	90	B	2023	BC	76	B	2827	C
PR+BFT	72.7	A	0.8	A	4.8	A	141	A	1129	D	110	A	1557	D
PR+N	65.3	CD	0.7	CD	3.0	C	61	CD	576	D	59	C	1071	D
Mean S.E	1.7		0.02		0.2		4		232		3		329	

† Predicted values are intended for comparison between treatments rather than for precise numerical estimates.
Pasture treatments followed by different letters (a,b,c,d,e) are significantly different (p = 0.05).

Conclusions/Implications:

This study gives a good indication of how well these four common grasses grow and persist in a rotationally grazed dairy operation, and how well suited they are for milk production on pasture. The perennial ryegrass-birdsfoot trefoil mixture consistently had the greatest nutritive value and predicted milk production, but perennial ryegrass does not usually persist well in the climate of Utah and Idaho. Unless a producer plans on reseeding his or her pasture every 2-3 years, this species is likely not a viable option. This study was in concurrence with other studies that show that tall fescue is a very persistent and productive grass in the intermountain region, making it a popular choice for irrigated pasture production. Tall fescue can be useful in dairy grazing productions, but pastures should be grazed frequently enough to give animals young, lush growth rather than older leaves and stems, which quickly become course with age. Based on this study, meadow brome and orchardgrass are quite comparable in many respects and can be useful for dairy grazing systems. Though often not as productive as tall fescue, they generally persist and produce well in the intermountain region. Dairy animals also readily consume them and maintain reasonable milk production on pasture.

B2: Wangsgard on Farm Trial

Question:

Can herbage mass and nutritive value be increased by applying Chilean nitrate, elemental sulfur, and/or high sulfur gypsum to organic dairy grazing pastures?

Objective:

Apply soil supplements and take herbage samples before each grazing event or harvest throughout the growing season to determine the impact that these soil amendments have on herbage mass and nutritive value.

Materials and Methods:

Soil amendments were applied in April 2018. Treatments consisted of high sulfur gypsum (G), elemental sulfur (S), high sulfur gypsum+sulfur (Gyp+Sulf), nitrate (Nit), nitrate+high sulfur gypsum (Nit+Gyp), and no amendment. Gypsum was applied at a rate of 300 lb./acre, sulfur was applied at a rate of 125 lb./ acre, and nitrate was applied at a rate of 100 lb./ acre, resulting in a nitrogen rate of 15 lb./acre. Mixed amendment treatments were applied separately at the same rates as single amendment applications.

Grazing and sampling took place on existing mixed pastures of meadow bromegrass, garrison creeping foxtail, and clover in Young Ward, Utah. In addition to those samples taken on grazing pastures in Young Ward, samples were also taken on a mechanically harvested field of triticale and sorghum-sudangrass just prior to harvest in Cornish, Utah.

Animals in the Young Ward study location consisted of lactating Holstein dairy cows in an organic dairy operation. Rotational stocking was used with a typical stocking period of approximately 24 hours, followed by a rest period ranging from 21 to 45 days depending on herbage growth and climatic conditions. Cows were on pasture for at least 120 days per growing season which consisted of three rotations. Herbage within four randomly placed 0.25 m^2 hoops was clipped to a height of 3.8 cm before grazing and harvesting. Samples were dried, weighed and ground to pass through a 1-mm screen using a Thomas Wiley Laboratory Model 4 mill (Arthur H Thomas Co, Swedesboro, NJ, USA), and were scanned with a Foss XDS near-infrared reflectance spectroscopy (NIRS) instrument (Foss, Eden Prairie, MN, USA) to determine

herbage nutritive value of the feed on offer. Data was analyzed using the mixed procedure in SAS.

Results:

Results from the triticale/sorghum-sudangrass harvest in Cornish were largely not significant due to the fact that only a single harvest was taken. Since three harvests took place in Young Ward and more significance was found, those results are presented in table 15.

Although only 15 lb. $acre^{-1}$ of nitrogen was applied to the nitrate treatments, a slight nitrogen response was evident, showing the importance of nitrogen in pasture grass production (Table 15). The Nit+Gyp treatment herbage production was significantly greater than the untreated pasture and the sulfur treatments and not significant from the others (Table 15). The nitrate treatment was only greater than the sulfur treatment (Table

15). The nitrate treatments also had the greatest predicted milk production $acre^{-1}$ (Table 15).

While the pasture treated with sulfur had low herbage mass, it seemed to play a role in many of the nutritive value parameters and was usually among those treatments with the greatest nutritive value and milk production day^{-1} (Table 15). Many of the other treatments were not significant from one another and more replication would be necessary to effectively distinguish one from the other.

Table 15 Herbage production, nutritive values, and predicted milk production from the Wangsgard on-farm trial located in Young Ward, Utah. Treatments consisted of mixed pasture with applications of Chilean nitrate (Nitrate), high sulfur gypsum (Gypsum), elemental sulfur (Sulfur), and mixtures of nitrate and gypsum (Nit+Gyp), and gypsum and sulfur (Gyp+Sulf)

Trmt.	Herbage production		CP		NDF		IVTD		TDN	
	lb. acre^{-1}		-----------------------------------			%			-----------------------------------	
Nit+Gyp	4740	a	14.2	a	56.2	a	77.4	ab	62.5	d
Nitrate	4244	ab	14.2	a	55.5	ab	77.4	ab	65.5	abc
Nothing	3765	b	14.7	a	52.7	b	79.8	a	67.1	ab
Gypsum	3667	bc	13.1	a	57.0	a	75.9	b	63.5	cd
Gyp+Sulf	3595	bc	14.8	a	53.8	ab	78.2	ab	65.0	bcd
Sulfur	2983	c	15.1	a	52.5	b	78.6	a	67.9	a
Mean S.E	265		n.s		1.1		0.9		1.1	

	RFQ		NE_L		Milk day^{-1}†		Milk acre^{-1}†	
			Mcal kg^{-1}		----------------	lb.	----------------	
Nit+Gyp	141.2	b	0.6	d	53.1	b	7353.8	a
Nitrate	150.4	ab	0.7	abc	57.9	ab	6987.2	a
Nothing	163.9	a	0.7	ab	64.7	a	6455.3	ab
Gypsum	140.3	b	0.7	cd	52.8	b	5853.7	bc
Gyp+Sulf	158.4	ab	0.7	bcd	61.9	ab	5642.2	bc
Sulfur	165.6	a	0.7	a	65.6	a	5194.0	c
Mean S.E	6.6		0.01		3.3		399.1	

Pasture treatments followed by different letters (a,b,c,d,e) are significantly different (p = 0.05).

†Predicted milk values are intended for comparison between treatments rather than for precise numerical estimates

Conclusions/Implications:

More replication and sampling would be needed to determine differences between treatments with more confidence, but some treatment differences did emerge in this study. Results from the Young Ward location showed that even low rates of nitrogen can have an effect on plant growth, since the nitrate treatments were among those with the greatest herbage production. Sulfur also seemed to play a small role in nutritive value, since it was often among those treatments with the greatest nutritive value and milk production day^{-1}.