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Determining Forage Intake of Nursing Calves Grazing Native Sandhills Rangeland

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Effective management of grazing lands is important for longevity and productivity. Accurate calculations of stocking rates per animal unit will aid in long term sustainability. Calculating stocking rates for cattle can prove challenging for cows and nursing calves. Limited research has been conducted to determine forage intake of the nursing calf pre-weaning. The objective of Trial 1 was: to determine nursing calf forage intake over time until weaning. Trial 1 used 8 crossbred cow-calf pairs in experiment 1 and 12 pairs in experiment 2. Pairs were separated into two groups rotating between grazing upland range or housed in individual pens separated by pair. Pairs were fed harvested meadow hay similar in quality to the rangeland. Hay, diet, and refusal samples were collected as well as total calf fecal collection. Organic matter intake (OMI) of nursing calves on pasture increased from 1.33-1.68% of BW and decreased to 1.43% in period 6. Pen OMI increased from 0.89-1.55% of BW and then decreased to 1.42% of BW also in period 6.There was no significant difference in OMI between Pasture and Pen for kg per day or as a percent of BW (P>.10). Determining the digestibility of forages is challenging and time consuming. Using in vitro techniques to evaluate feed digestibility can expedite the process. The use of forage samples as standards with known in vivo digestibility values is important for adjusting in vitro values. Trial 2 used 6 crossbred

yearling steers in a switchback design to determine in vivo digestibility of two chopped hays (meadow and brome hay). The objective was to create two forage standards to use in in vitro laboratory procedures to estimate unknown in vivo digestibility of forages. Hays were evaluated for differences in digestibility and digestibility measured by three methods. Meadow and brome hays were found to be significantly different in digestibility ($P<.0001$). These hays can be used as part of a standard forage sample set to evaluate unknown samples for digestibility.

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INTRODUCTION

Forage is the primary source of nutrient consumption for beef cattle throughout a majority of their life span. Since forage plays a major role in the diet, determining the quantity and quality of the forage consumed is of great interest and importance. Having a good grasp of what and how much cattle are eating aids producers in feeding cattle, managing pasture production, species composition and longevity, and meeting cattle nutrient requirements.

Determining forage intake of cattle is a challenge and of great importance. Intake will vary between breed, frame size, stage of maturity, and production level. This variation provides challenging management practices for producers as they strive to meet varying classes of cattle within their system. Cattle grazing on pasture are normally not handled or monitored daily thus measuring daily intake is difficult. Several methods have been created to estimate forage intake, including internal and external markers, total fecal collection and clipping forage in pastures pre and post grazing. Each type of method has its benefits and challenges along with level of accuracy of measurement. If forage intake can be measured consistently and accurately, this information can be passed on to producers in assisting in improved management of their herds and pastures.

There has been minimal research conducted on measuring cow-calf pair forage intake; particularly with calves. The majority of ranchers have not considered the amount of forage consumed by calves to be significant or even measurable due to the primarily nutrition supply provided from milk. Many producers do not include the calf as part of their stocking rate until a minimum of three or even six months of age. Even when

included, calves' weights are combined with their mother's body weight in the total pounds grazing pasture as part of stocking rate calculations. Thus the objective of this study was to determine cow-calf pair forage intake grazing Sandhills range.

Determining the Animal Unit and related calculations

ANIMAL UNIT

Animal unit (AU) is a term commonly used to define a standard animal consuming a standard amount of forage when calculating stocking rates. From this defined basis, changes in the animal class, weight, and/or production state can be applied to the AU value accordingly in a proportional ratio from the defined standard. Meyer (2012) stated that terms associated with an AU include animal unit day (AUD), animal unit month (AUM) and animal unit year (AUY). An AUD defines the daily forage intake, an AUM measures the amount of forage required to sustain the standard animal for one month, and an AUY is the amount required for one year (Meyer, 2012). For the purpose of this paper the AU will be assumed to be used for cattle equivalence. There is a lack of consistency between publications as to what is classified as the standard animal and amount of forage consumed. Alongside this challenge are the generalizations in applying animal production status, forage production, and physiological stages to the adjustment value of an animal unit equivalent (AUE).

Definitions of an AU began to develop as early as the 1900s and the parameters surrounding them have been morphing ever since (Scarnecchia, 1985). Initially, these definitions considered only one variable, such as animal weight, into AU calculations. With time and further research, the consideration of variables has changed and began to

include multiple factors that can have an effect on intake, i.e. metabolic size, gestation, or stage of lactation (Scarnecchia, 1985). The Society of Range Management (SRM, 1974) defined an AU as "one mature 454 kg cow or its equivalent based on an average forage consumption of 12 kg of dry matter per day". An AU is also defined as a 454 kg cow with a calf up to 6 months of age, consuming 11.8 kg DM/d or 354 kg/month (SRM, 1989; Iowa State University, 1998; USDA NRCS, 2003). Waller et al. (1986) defined an AU as a mature cow weighing 454 kg with or without a calf up to 4 months, consuming 340 kg DM/month, while Stubbendieck & Reece (1976) and Reynolds et al. (2000) used the same definition except with or without a calf up to 3 months of age. Redfearn and Bidwell (2003) define an AU as a 454 kg cow with calf consuming 11.8 kg dry matter intake (DMI) daily equaling 1 AU or a bull as a 1.25 AU with 14.5 kg of DMI per day. Holechek (1988) varies slightly in definition at an average 454 kg cow eating 9.1 kg of DMI daily or approximately 2% of her body weight (BW). The percentage changes when forage is more dormant lowering consuming intake to 1.5% of BW and increases to 2.5% of BW when forage is actively growing. Dry matter intake can be affected by production demands i.e. lactation and or quality of forage consumed (Hibbard and Thrift, 1992; Meyers et al., 2012). Reynolds et al. (2000) broke down the AU definition further for a 454 kg dry cow equals 0.9 AU and a calf over 4 months of age to weaning represented by 0.3 AU. Meyer (2012) found a significant difference between a dry cow and lactating cow with calf organic matter intakes at 2.15% and 2.34% of BW respectively ($P<.01$). These diverse definitions show how the AU value has changed over time and is continually being refining itself as the industry gains further insight into defining what and how much forage the different categories of cattle consume.

A particular species of animal must have an animal unit equivalent (AUE) independent of the kind of herbage it is eating or the temperature of its environment (Scarnecchia, 1985). The animal must have the same AUE whether it is in a pasture or feedlot. An AUE calculation can only be based on animal-related factors that affect the animal's demands. Scarnecchia and Kothmann (1982) quoted that an animal which has a demand rate more or less than 12 kg DM/day rate, as stated by SRM (1974), will have an AUE which is a proportionate fraction or multiple of 1 animal unit. Using a multi-variable animal demand model allows for more precise calculation of stocking variables such as stocking rate when determining AUE's (Scarnecchia, 1985). An example of applying multiple variables to an AUE is illustrated in Figure 1.These variables for Figure 1 include potential intake, cell wall content, ambient temperature, daily animal energy expenditures and sustainability index of diet available. This figure demonstrates some of the many potential variables that can be included in determining an AUE as well as demonstrating the diversity of factors that affect an animal's system.

FACTORS AFFECTING FORAGE INTAKE

Voluntary forage intake can be highly variable due to a wide range of influencers. These factors can be summarized into four major groups. These include genetic, physiological state, environmental, and forage quality and quantity. Genetic factors can encompass the age of the animal, sex, and body composition. The physiological state of an animal can include pregnancy status, physical activity, and lactation. While the environmental aspects range from thermoneutral zones, low or high temperatures, and topography. Forage quality and quantity are rated amongst some of the greatest influencers on intake. This can also be included with the inclusion of supplementation as

a part of the feeding program as well as stage of production of the forage itself (USDA NRCS, 2003; Lalman, 2004; Allison, 1985). All of these factors create different nutritional demands on the body and require various amounts of energy and nutrients to maintain those demands. The ability of the animal to meet those needs through forage intake can have a wide range of results.

The quality of a forage can be described by its digestibility in how available the nutrients are able to be broken down and utilized by the animal. It can also be described by what nutritional components comprise that forage. Individual intake varies with individual animal ability to consume forage (Van Soest, 1994). Intake of coarse forage, however high in nutrient quality, is limited by cell wall content. Poor quality forage restricts intake levels and consequently effects expression of individual genetic potential. The variability in intake is largest for high quality forages (Van Soest, 1994). Kartchner and Campbell (1979) found that digestibility is affected by several factors including intake level, rate of passage, and particulate size. Depending on length of time the forage takes to be broken down through initial mastication and rumination as well as travel through the digestive tract can directly affect further forage intake. Dry matter intake of forage is often limited by the ease (rate) with which a diet can be prehended and the resident time of particles in the digestive tract (Burns et al., 1994). Intake limitations can be physical as well as metabolic (Burns et al., 1994).

Depending on the actual quality of the forage in combination with other factors such as physiological, for example lactation, can show a wide range of intakes. Lalman (2004) cited Hibbard and Thrift (1992) for varying forage intakes for both lactating and dry cows. They found that a lactating cow forage DMI ranged from 2.2% BW consumed

for low quality (<52% TDN) to 2.7% BW for high quality (>59% TDN) forages. Dry matter intake for a dry cow was 1.8% BW for low quality and up to 2.5% BW for high quality forages. It was assumed that animal nutrient requirements are met or supplemented if needed. Reynolds (2000) also found similar intakes for beef cows ranging from 409-500 kg. Dry cows DMI for low quality roughages (dry grass, straw, etc.) consumed 1.5% BW, for average quality (meadow, native grass, etc.) at 2.0% BW and for high quality (alfalfa) and green pasture, 2.5% BW. In comparison for lactating cows with 2.0% BW for low quality, 2.3% BW for average, 3.3% BW for high, and 2.7% BW on green pasture. These intake variances will directly affect how much forage is consumed and for the producer, the challenge is if these needs can be met through pasture alone or through supplementation as well.

STOCKING PASTURE MANAGEMENT

Pasture management is crucial for the longevity of the land as well as the production of the animals utilizing its natural resources. Knowledge of range herbivore food habits is essential for efficient range management (Holechek et al., 1982b). The number of animal units and with what concentration and time length they are managed on a piece of land will have a direct correlation on forage removal, regrowth the following season, and animal production. The Society of Range Management (1974) defined stocking density as the relationship between the number of AU's and the land area at any instant in time. This is usually measured in au/ha. Stocking rate is the number of AU's per unit of land over a period of time (ISU, 1998; SRM, 1974). Stocking rates are based on the amount of forage that is standing at the end of the growing season in an ungrazed condition (Redfearn and Bidwell, 2003). Units of stocking rate must be in terms of

amount of forage demand per unit area, typically aud/ha, aum/ha, or auy/ha (Scarnecchia and Kothmann, 1982). The range supply must meet the animal's daily forage requirements in both quality and quantity (White and Troxell, 1995). For beef cattle sustainability for beef cattle production, the number of animals and their forage demand must be balanced with forage production (Redfearn and Bidwell, 2003).

Management challenges lie in knowing the long term forage production and stocking rates for the grazing season. The ability to adjust the rate or availability to supplement cattle with other forage when there is a deficit also contributes to management considerations (ISU, 1998). The art and science of pasture management will directly be reflected in subsequent years of pasture longevity and regrowth. Heady (1975) found that the coordination of forage utilization with forage growth through control of animal numbers determines the success or failure of other range practices and the economic stability of the operation. Using a multi-variable animal demand model allows for more precision in determining the AUE's used in calculating stocking variables like stocking rates (Scarnecchia, 1985). The current stocking rate for an area is based on the average long term end-of-season crop values for an operation. Proper rates over time will directly affect if the land will remain productive and sustainable (Redfearn and Bidwell, 2003). Meyer (2012) showed significant differences ($P<0.01$) in DMI between cow-calf pairs, dry cows, and steers (2.58, 2.37, 2.24% of BW) consuming different amounts of forage. If these values are utilized in adjusting stocking rates, producers will be able to more efficiently manage their natural forage resources.

Precipitation goes hand in hand with stocking rates for forage and land management. Gillen and Sims (2002) found that cattle in various stocking rates

maintained body condition during wet years. Yet, there was a negative effect on cow weights during dry years. Management changes need to be made when there are drastic changes in precipitation levels to strive to prevent long term damage as well as provide enough forage for the animals to consume. Reece et al. (2007) discovered that failure to reduce the grazing pressure during rapid growth windows for forage during drought will have negative results on the forage. The combined stress of overgrazing and drought are likely to cause long term declines in forage production potential.

Major differences are evident in stocking rates between introduced species vs rangeland forage. The percentage of utilization of available forage is lower for rangeland forage. Introduced species can be utilized to a higher degree with adequate moisture and fertility availability (Redfearn and Bidwell, 2003). This is a key concept in knowing what type of forage is being grazed and the ability to adjust the grazing density or length of time for long term longevity.

Kothmann et al. (1971) conducted an eight year trial in the Rolling Plains region of Texas to evaluate two systematic deferred-rotation systems. They observed seasonal and annual fluctuations in the weight of cows as direct indicators of their nutritional status. The stocking rate and grazing system had significant ($P<0.05$) influence on average cow weight over the course of the trial. Calf weaning weights were significantly affected by stocking rate and grazing management system. The greatest effect was in the fall and winter months. Calf weights from April through September had minimal variation from stocking rate or grazing system. These findings reinforce the importance of year-round forage management for longevity and optimal animal productivity.

FORAGE PRODUCTION

The composition and diversity of species of plants that cover an area of land are unique to each part of the country, even within regions of each state. Knowing, in detail, what those species are, their growth patterns, and how best to utilize their potential as well as maintain for longevity are valuable tools to have. Schacht et al. (2000) reported that the Sandhills of Nebraska are predominately composed of C4 grasses and have a mixture of other C3 and C4 species as well. There is a unique combination of upland range as well as meadows that comprise the grazing lands of the Sandhills. The upland range vegetation is dominated by warm-season grasses (Bragg and Steuter, 1995). Volesky et al. (2004) observed that cool-season grasses, sedges, and rushes dominate the meadows. Cool and warm season species respond differently for growth patterns as the temperatures change in spring and summer (Reece et al., 2007). Maximum growth rates for cool-season (C3) species are when the temperatures range from 18-24°C. Warm-season (C4) species grow optimally when the temperatures are between 32-35°C. Utilizing these growth periods for high quality forage consumption can adequately meet the needs of grazing cattle.

A balance between stocking rates and range utilization is necessary for effective conversion of range forage to animal production plus maintaining future range production capabilities (White and Troxel, 1995). Optimizing utilization during these high growth periods for C3 and C4 species is ideal, yet there are compositional changes as the forage matures. With forage maturity, comes a decrease in digestibility and CP, as well as increase in fiber and lignin content (Johnson et al., 1998; Cook, 1972; Savage and Heller, 1947). Knowing pasture or land forage production can aid in maximizing usage as well as

assist in long term productivity. Standing crop measured by clipping with grazing enclosures in key areas will provide forage production information (Redfern and Bidwell, 2003). Enclosures can be moved each year in the winter. White and Troxel (1995) recommend forage inventories and analysis every March, July, and November. The more years of data collected, the better the stocking rate decisions can be made over time.

Forage management in relation to production and growth periods is intimately intertwined. There is a science and art for livestock and pasture management. Knowing what types of species of plants grow in a grazed area, when their growth periods are, and yearly forage production (ideally over multiple years), can provide crucial information for pasture utilization. For moderate utilization of pasture land, 50% of the annual peak standing crop can be removed from ecological sites without negative effects on the plant community relative to species abundance or for beef cattle production (Redfern and Bidwell, 2003; White and Troxel, 1995). There is 25% allocated for animal consumption and 25% allotted for trampling, insects, other animals and nature disappearance loss. The remaining 50% is left for future forage production and soil erosion protection. With these categories in mind, there are three possible outcomes with stocking rates for livestock: correct, over, or under. Heady (1975) stated, “The coordination of forage utilization with forage growth through the control of animal numbers usually determines the success or failure of other range practices and the economic stability of the operation.”

MEASURING FORAGE INTAKE

Forage intake can be measured in a variety of ways. It is very important that accuracy and precision in collecting information start from this point on to ensure that

future calculations and measurements are not skewed incorrectly. Information can be collected from confined animals or those grazing. There are several methods to determining intake in confinement with individually fed animals, the Calan gate system where animals are in a herd environment, yet fed individually, or the use of empirical equations. When feeding individual animals in confinement, forage that has already been harvested is offered to the animal. Animals can become selective when fed at *ad libitum* levels and determining the objective of the study can be skewed when the data collected may not be a true representation of the feed offered (Burns et al., 1994). When collecting data on intake, Cochran and Gaylean (1994) advise to feed a percentage below *ad libitum* to ensure that animals will not sort through their feed for selectivity. This is normally in the 90-95% of ad libitum and should begin at least 2 days prior to the beginning of collection. Amounts of feed provided, orts, and feces can be collected relatively easily in a confinement set up. The Calan gate system allows for animals to experience more natural influences and socializing (Burns et al., 1994). This electronic gate system is configured to allow animals to socialize, yet be fed individually. The use of empirical estimates has been developed through sets of equations using regression techniques to estimate forage intake (Burns et al., 1994). Equations are created using measurements of cattle weight pre and post grazing as well as daily gain. Burns et al. (1994) stated that there are challenges with the use of this method due to lack of accounting for animal variation and the environment.

A grazing environment provides its own set of collection options and challenges to measure forage intake. Intakes can be measured with confined or grazing animals. Direct measures are collected with animal weights pre and post grazing along with forage

mass weight samples. Indirectly, measurements include fecal sampling, empirical equations or digestibility of forage and diet. Animals should be exposed to handling and sampling prior to collecting date to reduce stress and possible intake variation due to sampling. Fecal sampling can be through total collection or sampling using markers to measure digestibility. Intake with empirical equations and digestibility can be measured through a variety of collection methods with the use of diet samples, in vitro, and in vivo work.

DIET SAMPLING

The goal with collecting diet samples is to obtain a representative sample of what the animal is actually eating. The vast range of scenarios where samples need to be collected has led to several options for obtaining these samples. Collecting material representative of forage consumed from each plant species several times throughout the trial should be conducted (Cook, 1964). One method observes grazing animals' selection of plant species and portion of plants. Samples are hand plucked similar to that of the observed consumed plants by the animal. Hand plucking is only used in areas lightly grazed. Cages can be used to collect from plants grazed by animals for larger, higher grazed areas (Cook, 1964). Cook et al. (1958) found hand plucking samples adequate for single species of forage samples, but not for complex mixtures. The extent of plucking based on operator expectations of what an animal would select of a plant (Van Soest, 1994). This method is labor intensive, but does not require animals for collection. The other core challenge is in gathering a representative sample of what the animal would select; especially in complex mixed species.

Another form of sampling is through collecting clippings from pastures. Sampling is easy to obtain and saliva free (Holechek et al., 1982a). Random quadrat samples are collected pre and post grazing from the selected pasture being evaluated. Samples are clipped from within the quadrat, weighed, dried in an oven, and weighed again. The average dry weight gives an estimate of the quantity of forage available (ISU, 1998). When performed pre and post grazing, the differences in weight and chemical composition represent what was consumed (Cook, 1964). This is an estimate of removal, yet clipping to uniform height is not realistic to selectivity of the animal (Van Soest, 1994). Cages can also be used to measure growth over a grazing time period for production. Van Soest (1994) explains that caged areas are protected and not exposed grass to other natural occurrences such as wild life, trampling, manure and urine, which all influence plant growth. Clipping can also provide available forage estimate by dry matter per unit of land, which equals the weight of the fresh forage from the quadrats times the DM content (Van Soest, 1994).

Using fistulated animals has become one of the more accurate and widely used processes for collecting forage diet samples to measure digestibility of the diet. Esophageal and rumen fistulated animals are the two types of animals used for sampling. Van Dyne and Torell (1964) reported that esophageal fistulated animals have been reported to be used since the mid 1800's by Bernard and Pavlov. This sampling method collects the most realistic samples of forage intake. Holechek et al. (1982b) found that fistulated animal samples are advantageous over other techniques to obtain naturally grazed samples, but is expensive to set up. Torell (1954) first published and developed a set of basic procedures for esophageal fistulation. Van Dyne and Torell (1964) had at

least 90% success rate with surgery and collection for esophageal fistulated animals. Longevity and functionality of the fistulated animals varies. Some loss is due to operative and post-operative complications (Van Dyne and Torell, 1964). Some animals are functional for a few years while others usually for many years. Adams et al. (1991) had success with esophageal fistulas in 34 day old calves for surgery, post recovery, and acquiring adequate diet samples without complications. However, Hollingsworth (1994) had complications with esophageally fistulated calves in collecting diets, thus decided to have ruminally fistulated calves in the second year of the trial and had successful diet collections. Arnold et al. (1964) measured production performance of esophageal fistulated and non fistulated ewes. There were no data for negative effects on lamb birth weight, growth weight or mortality from either group, reinforcing that animals with esophageal fistulas can continue to lead productive lives without major negative effects on dam or their offspring.

There are various types of cannulas for the animal with some being permanent and others removable (Cook, 1964). Removable tend to be preferred over permanent for reduced incidences of blind pockets or pouches forming anterior to the fistula for esophageal animals (Van Dyne and Torell, 1964). When using an esophageally fistulated animal, the plug is removed and a diet sample bag is attached to the animal to collect diet contents as the animal grazes (Cook, 1964; Van Dyne and Torell, 1964). Various types of bags have been used for collection including plastic, canvas, rubberized canvas and bags with screen bottoms. Holechek et al. (1982b) found the esophageally fistulated samples to be more accurate and less labor intensive than rumen collections. Challenges with

esophageal fistulated samples include the potential for opening to get plugged or sample loss form collection bags.

Cook (1964) and Van Dyne & Torell (1964) explain that the rumen must be evacuated and rinsed with water prior to diet sampling to prevent any sample contamination. After grazing, contents are removed from the rumen and dried to prepare for compositing and chemical analysis. Contents removed prior to collection must be immediately placed back in the animal as to not deprive the animal of microbial population. This method requires greater amounts of time and labor (Olson, 1991; Cook, 1964). There is greater opportunity for larger samples than with an esophageal fistulated animal (Cochran and Gaylean, 1994). Though, rumen sampling is not suited for repeated consecutive days of diet sampling (Van Dyne and Torell, 1964) and not as conducive for cold weather sampling and potentially decreased selectivity due to an empty rumen (Olson, 1991).

Sampling decisions should be based on resources, labor availability, and what samples are being collected at what time of year (Olson, 1991). There were no differences in diet nutrient content in samples collected from rumen fistulae and esophageal fistulae (Olson, 1991). Animals in this experiment were both esophageally and ruminally fistulated. The animals showed no differences with and without rumen contents as well. Number of animals required for sampling can depend on area covered for the required sample and variability of vegetation being sampled (Cook, 1964). Number of animals can range from 3-6 with one morning sampling being adequate or sometimes two groups of animals collecting once in the morning or afternoon. Length of collection time will vary based on species (sheep vs cattle), size of fistula, rate of grazing,

and type of forage. Time length ranges from 10-15 minutes for irrigated pasture to upwards of two hours for winter range pasture (Van Dyne and Torell, 1964).

There is the consideration of saliva contamination from the fistulated animal. It can pose issues when determining DM digestibility, yet saliva must not be drained off due to possible changes in DM digestibility (Burns et al., 1994). Van Soest (1994) suggests correcting for both mineral and organic compounds that can come from saliva. Wallace et al. (1972) stated that saliva increased ash content, but had no effect on other chemical constituents on an OM basis. Presenting data on an ash free basis minimizes the effects of salivary contamination of minerals as well as soil contamination (Wallace, 1972; Van Dyne and Torell, 1964; Holechek et al., 1982a).

Measuring Digestibility

FECAL COLLECTION

The use of fecal collection is a major part of the in vivo studies. It is either evaluated through total fecal collection or the use of markers to determine output. Hatfield et al. (1993) tested sheep for differences between fecal bag and external marker estimates for the effects of fecal output and stress. They found no adverse effects from using either method. The study was conducted on both range and in metabolism crates. Total collection is used to determine total tract DM or OM “apparent” digestion using DM or OM output (Cochran and Gaylean, 1994). This is collected in metabolism stalls or with fecal bags. Steers are traditionally used for fecal collection for ease of fecal gathering and not deal with urine contamination of the feces. Feces are weighed and sampled for analysis. Cochran and Gaylean (1994) state that a fixed percentage or fixed

amount is kept each day, composited by day and a sub sample from the composite is kept. Burns et al. (1994) concludes that this provides rapid results, requires only DM and ash determinations before calculations can be made. The use of fecal bags for greater than seven consecutive days of collection (Cochran and Gaylean, 1994) can lead to soreness of the withers and/or tail head in addition to potential negative effects on intake and defecation. Total collection is the preferred method, but if not possible, markers can be used (Cochran and Gaylean, 1994).

When total fecal collection is inconvenient or the direct measurement of intake is difficult (i.e. grazing studies), markers can be utilized (Van Soest, 1994). A marker is a reference compound used to monitor chemical (hydrolysis and synthesis) and physical (flow) aspects of digestion as well as fecal output (Owens and Hanson, 1992). There are several qualifications for an ideal marker to be used (Owens and Hanson, 1992; Van Soest, 1994): 1) must not be absorbed, 2) must not effect or be affected by the digestive tract or its microbial population, 3) must flow parallel with or be physically similar to or intimately associated with the material it is to mark, and 4) must have a specific and sensitive method of estimation. No single marker fulfills all of these criteria, but the tolerable degree of error differs with the variable being measured.

There are two major types of markers; internal and external. Internal markers are recoverable indigestible feed fractions that are the basis for this type (Van Soest, 1994). Grazing feed intake scenarios are difficult to control creating the need for an internal marker to be used. Internal markers are inherent dietary constituents such as NDF, ADF, and lignin are resistant to digestion (Cochran and Gaylean, 1994). Van Soest (1994) lists some of the major types of internal makers and when they can be utilized: lignin-a

challenge with recovery in immature forage. Acid-Insoluble Ash (AIA) and silica-had variable success with sources from plants as well as soil/dust. Indigestible NDF or ADF could be used when lignin and AIA are too low to measure. It requires long digestion and better to use in vitro fermentation than in vivo. Chromogens, waxes, hydrocarbon, fecal nitrogen are some other potential internal markers that can be utilized. Internal markers can be utilized similar to external marker if intake is known (Cochran and Gaylean, 1994). If intake is not known external markers may be used to estimate digestibility directly.

An external marker is a substance added to the diet that must have no effect on the animal, diet, and not occur in the diet or soil (Van Soest, 1994). It too must be recoverable, indigestible, and not absorbed to the walls or lining of the digestive tract. External markers are administered either single pulse-dose or with a constant or frequent dose (Owens and Hanson, 1992). The single pulse-dose is used to estimate digesta volume and retention time at specific parts of the gut. The constant or frequent dose is administered in attempt to reach steady state conditions and measure at specific sites at successive times. Van Soest (1994) lists a variety of external markers that can be used including: stains and dyes, plastic and rubber, metal oxides, rare earths and other mordants, isotopes, soluble markers and metal chelates. Hollingsworth et al. (1995) conducted a grazing experiment on range and sub-irrigated meadow to compare total fecal collection and an intraruminal continuous release marker. They found a correlation between marker device and fecal collection at 0.93 in July and 0.99 in October. Estimates between the two methods were similar ($P<0.10$; $r=0.93$) on range in September. Fecal output was under estimated with the marker from total collection on range in July and

meadow in October ($P<0.10$). The marker device was shown to be a viable option for estimating fecal output, but recommendations were made to use total fecal collection to correct for fecal output for each forage source.

DIGESTIBILITY CALCULATIONS

After digestion trials have been conducted, calculations can be utilized to determine digestibility. The formulas below represent some of the key formulas used when total fecal collection or markers are applied to calculate digestibility. Cochran and Gaylean (1994) use this fundamental formula to be used when solving for digestion of a nutrient when orts do not have to be accounted for:

1. $$\%\text{Nutrient Digestion} = \frac{\text{Nutrient Consumed (kg)} - \text{Nutrient in feces (kg)}}{\text{Nutrient Consumed (kg)}} \times 100$$

When there are significant levels of refusals and the orts differ in chemical composition from the feed offered, another equation can be used. This formula is utilized to determine the digestion coefficient for a particular nutrient (Cochran and Gaylean, 1994).

2. $$\%\text{Nutrient digestion} = \frac{\text{Nutrient Fed(kg)} - \text{Nutrients Refused(kg)} - \text{Nutrient in feces(kg)}}{\text{Nutrient fed (kg)} - \text{Nutrient Refused (kg)}} \times 100$$

When intake is known and an external marker is continuously or frequently fed, fecal output can be calculated as follows (Cochran and Gaylean, 1994):

3. $$\text{Fecal DM Output (g/d)} = \frac{\text{Marker Dose (g/d)}}{\text{Concentration of Marker in Feces (g/g of DM)}}$$

After fecal output has been determined using an external marker, equation 1 or 2 can be applied to calculate digestibility. If an internal marker is used where intake is

known, the internal marker can be used to determine fecal output as in equation 3 by simply determining the amount of marker consumed by each animal (concentration of marker in diet x amount of diet consumed). If intake is not known, then digestion coefficients for different nutrients can be calculated with the following equation (Cochran and Gaylean, 1994):

4. $$\% \text{ Nutrient Digestion} = 100 - 100 \times \frac{\% \text{ Marker in Feed} \times \% \text{ Nutrient in Feces}}{\% \text{ Marker in Feces} \times \% \text{ Nutrient in Feed}}$$

IN VIVO DIGESTIBILITY

The ability to measure in vivo digestibility is important for diet and supplement formulation. Knowing diet digestibility aids in determining if an animal is lacking or exceeding in its requirements and can be addressed as such. Cochran and Gaylean (1994) define digestibility as the fraction of a feed stuff or dietary constituent that is lost on the passage through the digestive tract. The process of determining in vivo digestibility is a time consuming and costly endeavor. Criteria for these digestion trials has been explored previously in this paper through intake, fecal output and/or the use of markers for determining in vivo digestibility. Another way to determine in vivo digestibility of a nutrient is through the use of nylon or Dacron bags. Kartchner and Campbell (1979) walk through the process of sample in these small bags, replicated and attached to a weight that is place in a rumen fistulated animal to be exposed to digestion. After the exposure, the bags are removed, rinsed, dried, and reweighed to calculate the amount of digestion.

IN VITRO DIGESTIBILTY

Another method that has been developed to determine digestibility is through an in vitro digestion process or mimicking an artificial rumen. To determine forage

digestibility can be through in vitro techniques to determine in vivo values (Weiss, 1994). Tilley and Terry (1963) developed a two stage process that is still used today with minor modifications. The process includes replications of samples measured out and strained rumen fluid from two donor animals is used to mimic the rumen. The tubes are placed in a temperature controlled water bath for a specific length, usually 48 hours. Then, pepsin and hydrochloric acid are added to the tubes to mimic the feed stuff's travel to the omasum and abomasum. Tube contents are then filtered, dried and weighed.

There are specific criteria when creating an in vitro system which include: 1) the maintenance of a normal microbiological population, 2) the maintenance of normal rates of digestion, and 3) the ability to predict in vivo results (Warner, 1956). McDougall (1948) analyzed sheep saliva and published data on its mineral composition. There is now a buffer added to the rumen fluid before adding to the tubes to allow for longer digestion in the in vitro process. There are many digestibilities that can be measured through using the in vitro system. Judkins et al. (1990) compared 11 digestibility techniques for predicting in vivo apparent DM digestibility. Not one single method was accurate across all six diets and feeding conditions tested. Wilson et al. (1971) compared the Tilly-Terry two stage techniques with other methods for estimating forage digestibility. They determined that the two stage process for forage low in digestibility was less reliable. Yet, for all forages the two stage technique was as reliable for other methods tested. There were no in vivo digestibilities available for the samples used in the experiment. Scales et al. (1974) used in vivo work and fed forage samples from esophageal fistulated sheep to compare in vivo digestibilities with the two stage process (Tilley and Terry, 1963) as well as the one stage in vitro (Mellenberger et al., 1970). Both processes were

shown to be successful options ($P<0.01$) with the two stage method $r^2=0.86$ and $r^2=0.83$ for the one stage.

Several advantages found by Kartchner and Campbell (1979) explained the benefits of the in vitro system. Advantages include: 1) digestibility coefficients can be determined on large numbers of samples simultaneously, 2) time required per sample is minimal in relation to conventional digestion trial technique, 3) only a small sample is needed to determine digestibility, 4) degree of accuracy in estimating in vivo digestibility can be high. With this and any evaluation process come the potential for error. Animal variation can be a large contributing factor as well as variation between labs. Cochran and Gaylean (1994) advise that researchers must be cognizant of analytical and technical error, animal variation, as well as environmental and experimental design being taken into account.

The relationship between in vitro and in vivo digestibility is not a direct one to one correlation. When performing in vitro tests, there needs to be adjustment factors for run variances (Weiss, 1994). Creating a set of standard samples that have known in vivo digestibilities can assist in correcting the in vitro digestibility values to more accurately represent their apparent digestibility. With these known values, investigators can carefully compare samples of known in vivo digestibility with the in vitro digestion run values to ensure that the technique used provides satisfactory results under the experimental conditions used (Kartchner and Campbell, 1979). Weiss (1994) developed guidelines when researchers are developing calibration equations for the relationship between in vivo and in vitro:

1) Each lab determines in vivo & in vitro digestibility coefficients for diverse population of feeds. Expensive, labor intensive, data limited & appropriate for certain feeds.

2) Diverse set of feeds have known in vivo digestibilities as a calibration set. Set is included in in vitro procedure w/ forage samples of unknown in vivo digestibility. In vivo data regressed on the in vitro data to generate equation. Then the in vitro data are entered into the equation to adjust in vitro values to in vivo.

3) Use samples of known IVDMD from one lab and are analyzed at another lab. Equation derived to convert in vitro data from second lab to estimate in vitro data from original lab. Original lab must have accurate in vitro-in vivo equation to use to convert sample in vitro to in vivo values.

Weiss (1994) also advises to create separate equations for different types of samples being analyzed. The groups include legumes and grasses, corn silage, concentrate feeds, and low quality roughages. It is also strongly suggested that each lab develop its own equations due to technique, animal and sample variation as well as technician variance. Geisert et al. (2000) used three hays and two legumes to create standards to run with in vitro runs to adjust for the difference between in vivo and in vitro. The goal was to have a wide range of digestibilities for the standards to create better accuracy in regression equations used for determining the digestibilities of feeds used for formulation and animal response prediction. The use of forage standards included in the two-stage method of in vitro analysis is necessary to aid in determining

the in vivo regression equations to determine the unknown sample digestibilities (Weiss, 1994).

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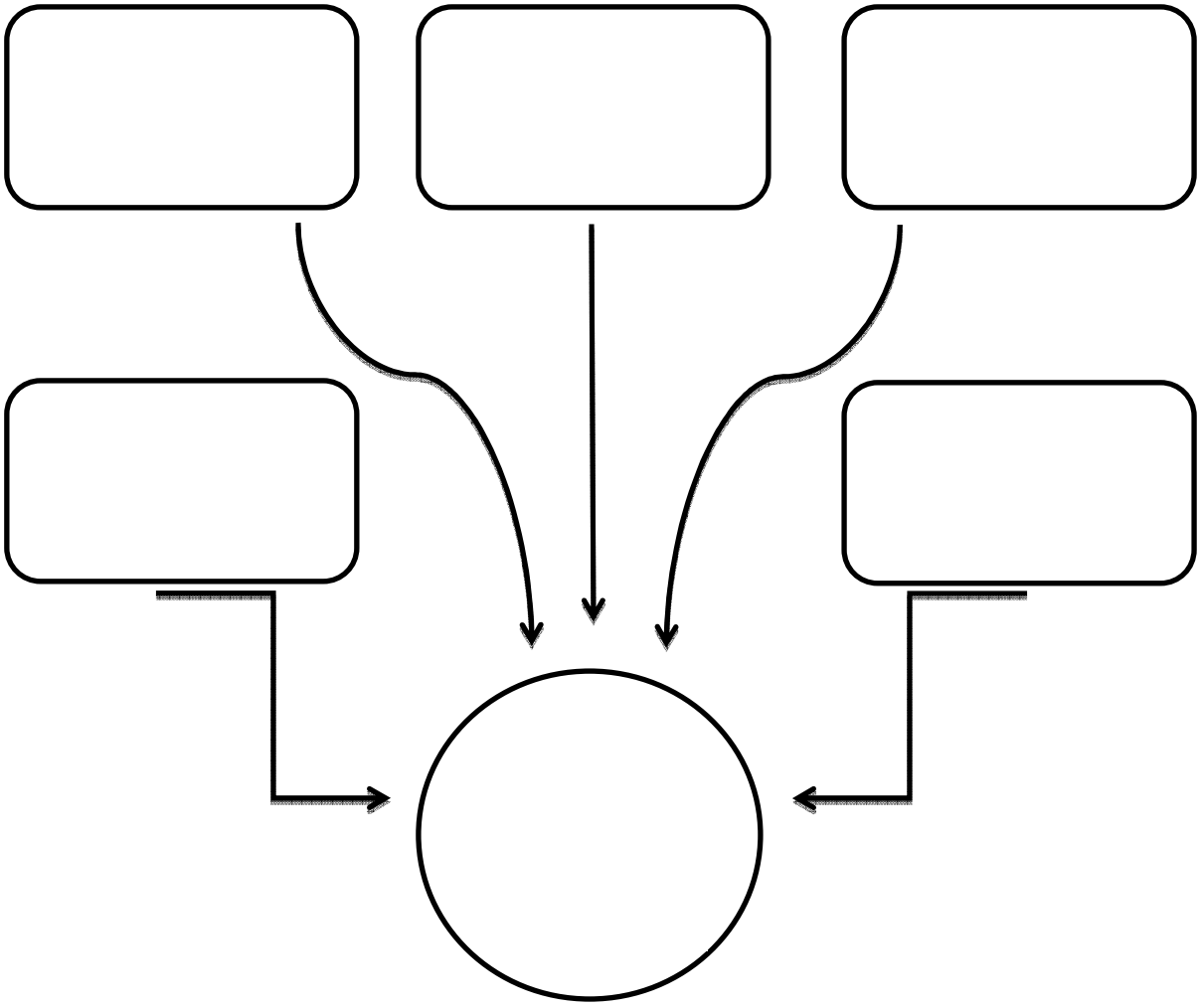
Figure 1. Simplified conceptual model of some variables affecting actual herbage intake of a grazing animal on pasture.



Scarnecchia 1985

Chapter II

Determining Forage Intake of Nursing Calves on Sandhills Rangeland

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ABSTRACT

The objective of this study was to determine nursing calf forage intake prior to weaning in the Nebraska Sandhills range. Eight crossbred cow-calf pairs in year one and 12 pairs in year two were selected for each trial. Pairs were randomly assigned to one of two rotation groups, pasture (Graze) or individual fed dry lot pens for each pair (Pen). Pen pairs were fed harvested meadow hay of similar quality to forage grazed in Graze daily. Cattle were adapted for two weeks and had one week of collection per period before switched to the other rotation. Groups were switched back and forth between Pen and Graze from June until mid-October. Diet samples were collected each period in pasture assigned to study. Total fecal collection for calves was conducted consecutively for five days and subsampled daily for analysis each period. Feed and diet samples were analyzed for DM, OM, CP, IVOMD, NDF, and indigestible ADF. Milk production was measured using weigh-suckle-weigh techniques each period. Calf organic matter intake (OMI) increased for both Pen and Graze in Year 2 from periods 1-6 for kg per day intake 0.99 to 3.04 and 1.47 to 3.17, respectively. Intakes increased as well as a percent of BW from periods 1-5 for Pen going from 0.89-1.55% and decreasing to 1.42% for period 6 and Graze rising from 1.33-1.68% for periods 1-5 and decreasing to 1.43% in period 6. There was no significant difference between calf OMI for Pen and Graze groups ($P>.10$) for either kg per day or as a percent of BW. In Year 1 milk production decreased over time from 7.5 to 4.0 kg/d and Year 2 milk production was fairly constant at an average 4.0 kg/d. Calf forage intake increased over time until weaning in the fall and range managers would be advised to include calf forage intake in pasture management and stocking rate decisions throughout grazing season.

Key Words: cattle, forage intake, grazing, nursing calf

INTRODUCTION

Raising beef cattle for consumption and managing herds on pasture land has been an evolving process over the years. The industry began to develop and define what cattle grazing requirements were over time. J.T. Jardine used the cow-day, a forerunner for the AUD, in range reconnaissance surveying methods for the U.S. Forest service between 1907 and 1911 (Scarnecchia, 1985). This basic, fundamental definition has evolved over the years as scientists and ranchers have observed and measured the variables that contribute to the forage consumption of cattle. These variables can include age, maintenance, frame size, growth, forage species composition, and production. Estimating intake of nursing calves on rangeland is challenging. Traditionally, forage intake of calves is not considered significant until 6 months of age (SRM, 1989; Iowa State University, 1998; USDA NRCS, 2003). An animal unit (AU) in Nebraska has been defined as a lactating cow with calf up to 3 months of age (Stubbendieck & Reece, 1976) or 4 (Waller et al., 1986). Even with adding nursing calves to the AU definition, their weight is accumulated with the cow's and the forage intake is lumped together for both animals. This does not account for differences in frame size, production status, or stage of maturity. All of these factors can influence intake and utilization of the forage consumed.

Cow-calf pair intake has been researched to define that unit of forage intake in a variety of locations and environments. Research has been conducted to determine forage and milk intake interactions (Ansotegui et al., 1991; Boggs et al., 1980; Holloway et al., 1982) as well as determining calf forage diet quality or limiting nutrients in the calves' diets (Grings et al., 1995; Walker et al., 1981; Hollingsworth-Jenkins, 1994). Yet, there

has been minimal research conducted to determine the change in forage intake for the grazing nursing calf pre-weaning. Our objective of this study was to measure forage intake of a nursing calf and cow/calf pair grazing Sandhills rangeland prior to weaning.

MATERIALS AND METHODS

Study Site

This study was conducted at the University of Nebraska's Gudmundsen Sandhills Laboratory (GSL), near Whitman, NE (elevation 1,073 m, lat 42°05' N, long 101°26' W). The average maximum daily temperatures range from 2 C in January to 32 C in July. Mean annual precipitation at the site is 543 mm, with 75% of which occurs during the growing season from April through September.

The upland Sandhills rangeland pasture was comprised of a combination of warm and cool-season grasses. Dominant warm season grasses included prairie sandreed [*Calamovilfa longifolia* (Hook.) Scribn.], sand bluestem [*Andropogon gerardii var. paucipilus* (Nash) Fern.], switchgrass (*Panicum virgatum L.*), and little blue stem [*Schizachyrium scoparium* (Michx.) Nash]. Major cool-season grasses were needleandthread (*Stipa comate Trin. & Rupr.*), prairie junegrass [*Koeleria pyramidata* (Lam.) Beauv.], and Kentucky bluegrass (*Poa pratensis L.*) (Volesky et al., 2005). Several sedge species (*Carex spp.*) were also present.

Harvested Sandhills sub-irrigated meadow hay was fed to individual pens daily. Previous year (2009) harvested hay from the Gudmundsen Ranch was analyzed for digestibility and CP and selected based on IVDMD values at the Ruminant Nutrition Lab at the University of Nebraska-Lincoln in Lincoln, Nebraska in the spring of 2010 for

experiment 1. Hays were selected to replicate the change in digestibility of the pasture treatment throughout the length of the experiment. Selected hays were based off of Geisert et al., (2008) work determining IVOMD for the GSL Sandhills rangeland (Table 1). Late spring, immature growth hay was also harvested in late May 2010 to capture the high digestibility forage to mimic the quality in the pasture. Immature hay was fed solely during period 1 and was mixed with the previous year's hay in proportions to simulate the digestibility in the pasture for periods 2-4. The previous year hay was fed alone for periods 5-6. Volesky et al., (2004) found that cool-season grasses, sedges, and rushes dominate the meadows of the Sandhills. The most common grasses were slender wheatgrass [*Elymus trachycaulus* (Link) Matte], red top bent (*Agrostis stolenifera L.*), and timothy (*Phleum pretense L.*). Other grasses present included Kentucky bluegrass (*Poa pratensis L.*) and smooth bromegrass (*Bromus inermus Leyss.*). Wooly sedge (*Carex lanuginose Michx.*) and several spike rush species (*Eleocharis spp.*) were also abundant. The primary legumes included white clover (*Trifolium repens L.*), alsike clover (*Trifolium hybridum L.*) and red clover (*Trifolium pretense L.*). Warm-season grasses present in minor amounts were prairie cordgrass (*Spartina pectinanta L.*) and big bluestem (*Andropogon gerardii Vitman*).

Animals, Design and Treatments

Exp. 1

Crossbred cow-calf pairs (Husker Red composite- ¾ Red Angus and ¼ Simmental or Gelbveih) were selected from the March calving herd each spring. Nine crossbred cow-calf pairs were selected for experiment 1 in 2010 and five of those calves

were selected for rumen cannulation. Cannulation surgeries were conducted at GSL in early May and procedure was approved by the University of Nebraska's Institutional Animal Care and Use Committee. Calves were monitored daily for one week and surgical sites were checked two and four weeks post operation according to animal cannulation procedures and care. All calves had a 48 hour adjustment period to fecal bags and harnesses prior to initiation of the experiment.

The experiment was a switchback design with two treatments. Groups of cow calf pairs rotated between the two. Treatments were grazing Sandhills upland range pasture as one large group (Graze) and individually fed pens separate for each cow/calf pair (Pen). Pairs grazing pasture were kept in a herd environment for the duration of the period. Each dry lot pen housed one cow/calf pair and were fed harvested sub-irrigated meadow hay daily of similar quality to that of the pasture. Eight pairs were randomly assigned with two cannulated calves per group to one of the two treatments, while having the extra cannulated pair if needed. One cannulated pair on trial was removed at the end of period one and a replacement pair was substituted into the trial at the beginning of period 2. The removed calf's data were not included in analysis and summary for period one. Pasture was 46.96 hectares in size and individual pens were 504 m^2. The experiment was 19 weeks long with 3 week periods except the first period being 4 weeks in length. Cow initial average body weight was 524 kg and calf initial weight was 116 kg. Cow and calf were treated as one unit, with calf age averaging 75 d at the start of experiment. Pens and pasture had water and salt provided ad libitum.

Exp. 2

Twelve crossbred cow-calf pairs were selected from the Gudmundsen research beef herd for experiment 2 in 2011 and no cannulation surgeries were conducted on the calves. The decision to not have cannulated calves was based on health complications in experiment 1, lack of quantity and consistency of cannulated calf samples collected, and the economical investment value for this experiment. Pairs were randomly assigned to one of two groups similar to experiment 1. Experimental design, treatments, and locations were the same as described in experiment 1. Calves were handled daily for halter breaking and desensitizing to fecal bag and harness two and half weeks prior to experiment initiation. Cow initial average BW was 410 kg and calf BW was 91 kg and 74 d of age.

Diet

Exp. 1

Hay was fed in bunks, weighed and offered daily, and fed at libitum amounts. Orts were also collected and weighed back daily. Hay and orts were subsampled three times per week and composited by week. Dry matter content was determined from composited weekly samples. Orts from each pen were composited into one sample per week. Hay fed was in round bales and processed to decrease stem length for easier handling during feeding.

Exp. 2

Pasture treatment was the same as in experiment one. Hay fed to individual pens came from sub-irrigated meadows at GSL harvested summer 2010. For the high quality, immature hay required in period 1, regrowth meadow hay was harvested in September 2010 to feed the following summer. Due to resource and hay quality constraints, the immature hay was fed for period 1 only. Two other hays were selected after lab analysis based on IVDMD. The Home Valley East Division North Side hay was fed for periods 2-4 (52.5% IVDMD) and the Home Valley West Division South Side hay was fed for periods 5-6 (50.49% IVDMD). Hay and orts were handled and sampled the same as experiment 1. Orts were kept separate by pair and composited by week by pair.

Data Collection

Exp. 1

All cattle were limit fed three days prior to the beginning and end of trial and two day weights were recorded. Cow weights and body condition scores (BCS) were recorded at the end of each period when calf weights and milk production were measured. Milk production was measured using a 12 hour weigh-suckle-weigh (WSW) procedure for periods one through four. An 18 hour WSW was used for periods five and six. Calves were separated from their dams for 5-6 hours and then allowed to nurse. Afterwards, calves were separated again for 12 or 18 hours. At 0700 h calves were weighed, allowed to nurse until complete, and re-weighed. Twenty four hour milk production was calculated by doubling the 12 hour milk production. For the 18 hour milk production the weight of the milk was divided by 0.75 to provide the 24 hour production.

Cow diet samples were collected in the pasture once per period. Previously esophageally fistulated, mature, multiparous beef cows from the GSL herd were used for collections. Cows were already well established for diet collections prior to experiment. Cows were withheld from feed, but not water for 18 hours prior to collection time. Samples were collected beginning at 0700 h each time. Cows were hauled in a trailer to collection site. Once on site, esophageal fistulae plugs were removed and bags with screened bottoms were hung on the neck of each cow. The bags were secured with a bungee cord attached to a nylon belly strap tied behind the front shoulder. The cord kept the bag in place and did not restrict cow movement. Cows grazed for 20 to 45 minutes until adequate sample (approximately 1 kg) was collected. After grazing, bags and straps were removed from the cow, plugs inserted, and masticate samples were subsampled and squeezed to remove excess saliva. Sub-samples were frozen and then freeze dried. Freeze dried samples were ground through a Wiley Mill using a 2-mm screen and half of the sample was ground through a 1-mm screen.

Calf diet samples were collected once per period. All four rumen cannulated calves (two per treatment) were used for each collection. Rumen contents were evacuated and contents held in plastic lined buckets and covered during diet sampling. Calves were allowed to graze or eat from the bunks in the individual pens for 30 to 45 minutes. Calves were caught, masticate sample removed from the rumen and original contents returned into the rumen. Samples were squeezed, frozen and then freeze dried. Freeze dried samples were ground through a Wiley Mill using a 2-mm screen and half of each sample was ground through a 1-mm screen. Diet collections were conducted at least four days

prior to the beginning of fecal collection to allow animals body to equilibrate after sampling.

Total fecal collection was conducted on all eight calves for five days each period. Harnesses and bags were attached to calves at 0700 hour on day one of collection each period. Clean, empty bags were weighed prior to placement on the calves each day. Bags were changed out every 24 hours. Bags containing fecal output from each day were weighed, sub-sampled and excess sample disposed of. If the calf lost its bag or fecal sample spillage was detected outside of the bag, that day's measurement was excluded from the average fecal output calculation. Fecal sub-samples were frozen and freeze dried, then ground through a Wiley Mill using a 2-mm and 1-mm screen. Individual, daily ground samples were composited by calf by period. Part of the sub sample from each day was also dried in duplicate in a 60°C force air oven for 72 hours to determine fecal dry matter.

Exp. 2

Procedures for measuring beginning and ending weights as well as calf weights, cow weight, and BCS were the same as experiment 1. WSW was also estimated using the same procedures at the 12 and 18 hour time periods. Four esophageally fistulated cows were used for each collection period and collections were conducted twice per period. Only two diet samples were collected in week seven because the cows escaped the holding pen and grazed the night before collection. Calf diet samples were not collected because there were no fistulated calves in this experiment year. Fecal collection and sampling was handled similarly to experiment 1, but samples were not freeze dried. The

individual dry matter samples were ground, composited, and used for analysis. In period five, one calf was not fitted with a total fecal collection bag due to a hoof injury. Fecal grab samples were collected each day to use for analysis.

Sample Analysis

Exp. 1

All hay, ort, diet, and composite fecal samples were dried in a 100° C forced air oven to determine lab corrected DM and subsequently burned in an ash oven at 600° C for 6 hrs to determine OM content. Samples were analyzed on an OM basis to correct for soil and saliva contamination. Hay, ort, and diet samples were analyzed for CP, NDF, and in-vitro organic matter disappearance (IVOMD). Crude protein was analyzed using a combustion N analyzer (Leco FP-528, St. Joseph, MI). Neutral detergent fiber analysis was conducted according to Van Soest (1991). The Tilley and Terry method (1963) was used with a 50:50 mixture of the rumen inoculum and McDougall's buffer with the addition of 1 g/L of urea to buffer solution (Weis, 1994) to determine in vitro OM digestibility. After being filtered, samples were dried for at least 5 hours in a 100° C forced air oven, weighed, and then burned in an ash oven for 6 hours at 600° C to determine the DM and OM content of the hay, ort and diet samples. The IVOMD run included 5 feed standards with known in vivo OM digestibility. The IVOMD of these standards was then regressed on their known digestibilities to develop regression equations for comparison of experimental samples between runs (Geisert et al., 2007).

Exp. 2

Indigestible acid detergent fiber (IADF) analysis was conducted on hay samples, composited diet samples by period, and the non-cannulated calf composited fecal samples. Diet and hay samples proceeded with a two-stage in vitro digestion technique of Tilley and Terry (1963) with 0.5 g samples in 50 ml centrifuge tubes inoculated with 30 ml of 50:50 mixture of the rumen inoculum and McDougall's buffer with 1 g/L urea to buffer solution (Weis, 1994). After stage one of digestion for 48 hours, tubes were centrifuged and excess inoculum was removed. All samples were then inoculated with the same type of mixture as listed previously and incubated at 39°C for 96 hours in a water bath. Contents were transferred to a 600 ml beaker and using a wash bottle containing ADF solution, rinsed all particles clinging to the tube into the beaker. Solution was added to bring total contents to 150 ml. ADF analysis was continued with the Goering and Van Soest technique (1970), filtering the hot solution through tared filters. Filters were dried for at least 5 hours in a 100°C forced air oven and after weighing, burned in a 600°C ash oven for 6 hours. Percent IADF for feed and fecal samples was calculated:

$$\textbf{\% IADF} \text{ (feed or fecal)} = 100 \times \frac{\text{ADF residue (OM) - blank}}{\text{Sample wt (OM)}}$$

Dry Matter Fecal Output from each day per calf and digestibility values analyzed from forage samples were used to determine calf forage intake. Forage values for dry lot and pasture samples were applied accordingly to each group of calves for each period. Values were inputted into the following estimation equation:

Forage Intake (kg/d): $\frac{\text{Fecal Output (OM)}}{\text{(1-IVOMD)}}$

Where Calf Fecal Output is on an OM basis (kg) and divided by [1-IVOMD]. The denominator portion of the equation is to determine the indigestible portion of the feed and divide that into the fecal output to determine how much forage was consumed by the calf.

Statistical Analysis

Calf intake data from Experiment 1 were not statistically analyzed due to lack of quality and quantity of calf samples, and ongoing health issues with calves throughout the 2010 experiment. Calf calculated intake and forage quality for Experiment 1 are in Appendix A for comparison with Experiment 2.

In Experiment 2, calf forage consumption over time on a daily and percent of body weight basis were plotted using Proc Reg of SAS (SAS Inst. Inc., Cary, NC) to determine differences in intake within period by treatment. Equal slopes analysis in Proc Glimmix of SAS was used to determine if the slopes of the different treatment lines were different.

RESULTS AND DISCUSSION

The DMI of cow-calf pairs in the pens varied between Experiment 1 and 2. In each experiment the same set of cow-calf pairs were kept in each rotation group for each experiment and either grouped in periods 1,3, & 5 (A) or 2,4, & 6 (B). When comparing

the two groups in Exp. 1, OMI of group A increased from 13.48 kg/d/pair to 15.95 kg/d/pair from periods 1 to 3 to 5. Group B increased from 12.34 kg/d to 14.27 kg/d from period 2 to 4 and back down to 12.88 kg/d in period 6 (Table 3). Group A had an intake range of 2.11-2.24% BW and group B had a slightly lower range of 1.55-1.99 % BW intake. These intakes as percent BW are slightly lower than the findings of Meyer et al. (2012) as well as Reynolds (2000) having intake at the 2.3-2.5% BW range. Reynolds (2000) reported intakes on a DM basis, while Meyers et al. (2012) reported pair intake on both a DM and OM basis. Experiments 1 & 2 measured results on an OM basis. The change between DM to OM can result in up to 7-8% difference due to variance in ash content. In Experiment 2, Group A increased from 13.69 to 17.35 kg/d/pair, while Group B increased from 12.53 to 14.69 kg/d/pair through the duration of the experiment. Group A had higher intake ranges as a percent of BW at 2.59-2.61%, while Group B had lower intake percentages beginning at 2.60 decreasing to 2.32% BW (Table 4). Variance in intake could be due to several factors such as the weather with extreme storms or heat, animal variation and selectivity, or collection period stress due to greater amounts of handling. Another major factor is the increase in calf growth and rumen development over time. As both of these elements increase, there is the potential for greater forage intake.

Calf calculated OMI increased through the periods for both the pen and graze treatments for intake in kg/d as well as on a %BW basis (Table 5). When following each group of calves (A & B) through their respective rotations (see notations of 1&2 for each period), there was an increase in intake throughout experiment 2. Graze intakes for periods 2-4 for the pasture treatment are very similar to Hollingsworth-Jenkins et al.

(1994) findings of nursing calf forage intake for July, August and September. Hollingsworth-Jenkins et al. (1994) measured calf OM forage intake starting at 1.1 to 1.5 to 1.3% BW for those respective months, while experiment 2 measured 1.12 to 1.27 to 1.22 % BW intake for that similar range of months on pasture. Experiment 2 had lower forage intake based on kg OM/d thank Hollingsworth-Jenkins et al. (1994). Intakes were not reported as a percent of BW and harder to evaluate against current findings when calf weight is not part of the equation.

For experiment 2 (Table 5), there were constant increases in intake on the kg/d as well as on a % BW for groups A & B. There was a pronounced increase in Period 5 intake for both sets of calves. This could be due to the fall cool season forage regrowth that has a small spike in the early fall causing more digestible feed available and increased intakes as seen in Table 6 diet sample OMD increasing from 52.81% in Period 4 to 57.47% in Period 5. A change was also found from September to October in Table 1 from Geisert et al. (2008) of 51.4-53.0% OMD. Calf forage OMI on a kg/d basis were not significantly different ($P>0.1$) between the Pen and Graze groups throughout the trial in Experiment 2 (Figure 1). Intakes were also not significantly different on a percent of BW either ($P>0.1$; Figure 2). Intakes were reported on an OMI basis due to correction for sand contamination of grazed diets and dry lot pair feeding. Sand was found in samples when evaluated for nutrient composition in the lab. Samples were ashed to be able to correct for the effect of sand in the calculated nutritive values of the feed, diet, ort, and fecal samples collected.

Milk production for cows in Experiment 1 decreased from an average 7.5 to 4.0 kg/d while in Experiment 2, milk production was more constant at an average of 4.0 kg/d

(Figure 3 and 4). Calf OMI increased from period 1 to 6 despite the overall consistent average milk production of the cows. Boggs et al. (1980) found that even with varying birth period, spread over two months, there was a decrease in milk intake from April through September and an inverse of forage DMI increasing throughout that same time period. Ansotegui et al. (1991) found a direct relation between two years of a study where there was significant difference in milk production between years as well as fecal output (FO). Milk intake was higher in year 2 than year 1, while FO was higher in year 1 than 2 ($P<0.05$). Holloway et al. (1982) found that calves were more efficient in converting milk and forage DE to weight gain on a less energy dense forage source than calves on a higher energy density forage. Hollingsworth-Jenkins (1994) found that with grazing nursing calves in the first year, forage intake increased from July through September (1.4 to 2.9 kg OM/d; $P<0.05$), while milk intake stayed consistent ranging from (1.0 to 1.1 kg OM/d). In the second year, forage intake also increase linearly (1.1 to 2.6 kg OM/d; $P<0.05$), while milk intake decreased linearly (1.1 to .73 kg OM/d; $P<0.05$). These researchers have shown that calves adjust forage intake based on milk intake as well as forage quality. Utilizing this information alongside the information found in this experiment, shows that the forage intake of calves is variable as well as important.

Meyers et al. (2012) found a significant ($P<0.01$) difference in intake between a cow-calf pair at 2.34% of BW on OM basis vs a non-lactating cow consuming 2.15% of BW on OM basis. Intake was also evaluated for cow-calf intake when the calf BW was not considered in part of the calculation as well as for a lactating cow's intake minus the calf BW and an assumed calf DMI of 1.3% of BW. Meyers et al concluded that when the calf BW is removed from the DMI as a percent of BW calculation it increases DMI from

2.58% with calf BW to 3.16% of BW without. The second calculation subtracts the calf BW as well as the calf DMI of 1.3% of BW or 1.5 kg DMI which was used from Hollingsworth-Jenkins et al. (1994) from the lactating cow. The calculated forage intake for the lactating cow was 14.7 kg DMI and 2.87% of BW, DMI. This value is still higher than the measured pair intake of 2.58% of BW basis. These calculations reinforce the influence that the calf has on intake rates and overall forage removal pre-weaning.

Cow-calf pair intake was shown to vary from 1.9 to 2.2% of BW on an OM basis in Experiment 1 and from 2.11 to 2.69 % of BW depending on collection period in Experiment 2. These findings reinforce work done by Meyers et al. (2012) finding the cow-calf pair to have DMI at 2.58% of BW and 2.34% of BW with OMI. Coupling with the increasing calf forage intake (Table 4) over the duration of the summer and fall months, shows that an average pair forage consumption rate should be considered for adjustment for each producers operation. Further calculations are needed to express how the calf's forage intake is affecting the traditional AUE value. Yet, with this information, producers can have a deeper understanding of managing and utilizing their resources to meet their specific operations and locations.

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Table 1. Monthly Average IVOMD and CP (%DM) values of diet samples from Sandhills upland range pastures. (Geisert et al., 2008).

Sample Date	IVOMD[1]	CP[2]
June	62.6	10.8
July	55.9	11.5
August	55.5	8.9
September	51.4	8.8
October	53.0	7.9

[1]IVOMD average values for pastures.
[2]CP (%DM) average for all pastures.

Table 2. Experiment 1 & 2 Timeline

Experiment 1[1]

Period	1	2	3	4	5	6
Dates	6/8-6/30	7/1-21	7/22-8/11	8/12-9/1	9/2-22	9/23-10/13

Experiment 2[2]

Period	1	2	3	4	5	6
Dates	6/14-7/4	7/5-25	7/26-8/15	8/16-9/5	9/6-26	9/27-10/17

[1]Experiment 1 was conducted in 2010.
[2]Experiment 2 was conducted in 2011.

Table 3. Forage OMI of cow-calf pairs in dry lot pens in Experiment 1.

	Period[4]					
	1[1]	2[2]	3[1]	4[2]	5[1]	6[2]
Cow BW, kg	476.2	513.2	483.3	536.0	508.0	579.5
Calf BW, kg	131.4	134.3	159.9	179.5	204.8	230.0
Cow BCS	5.3	5.3	4.9	5.3	5.0	5.25
Pair OMI, kg/d	13.48	12.34	13.60	14.27	15.95	12.58
Pair OMI, % of BW[3]	2.22	1.91	2.11	1.99	2.24	1.55

[1] Group A: Periods 1, 3, & 5 contain the same four cow-calf pairs.
[2] Group B: Periods 2, 4, & 6 contain the same four cow-calf pairs.
[3] BW calculation combines cow and calf weights.
[4]Period lengths for Experiment 1 are listed in Table 2.

Table 4. Forage OMI of cow-calf pairs in dry lot pens in Experiment 2.

	Period[4]					
	1[1]	2[2]	3[1]	4[2]	5[1]	6[2]
Cow BW, kg	418.7	356.2	438.7	376.7	469.8	426.5
Calf BW, kg	109.6	125.3	150.5	163.8	196.3	213.7
Cow BCS	5.0	4.8	5.0	5.0	5.1	4.9
Pair OMI, kg/d	13.69	12.53	16.63	12.78	17.35	14.69
Pair OMI, % of BW[3]	2.59	2.60	2.82	2.36	2.61	2.32

[1] Group A: Periods 1, 3, & 5 contain the same six cow-calf pairs.
[2] Group B: Periods 2, 4, & 6 contain the same six cow-calf pairs.
[3] BW calculation combines cow and calf weights.
[4]Period lengths for Experiment 2 are listed in Table 2.

Table 5. Calculated OMI of calves in Experiment 2.

	Period[3]					
Pen	1[1]	2[2]	3[1]	4[2]	5[1]	6[2]
Calf BW, kg	109.6	125.3	150.5	163.8	196.3	213.7
OMI, kg/d	0.99	1.54	1.93	2.52	3.04	3.04
OMI, % BW	0.89	1.23	1.28	1.54	1.55	1.42
Grazing	1[2]	2[1]	3[2]	4[1]	5[2]	6[1]
Calf BW, kg	109.8	127.1	148.1	171.9	184.1	221.7
OMI, kg/d	1.47	1.43	1.88	2.10	3.09	3.17
OMI, % BW	1.33	1.12	1.27	1.22	1.68	1.43

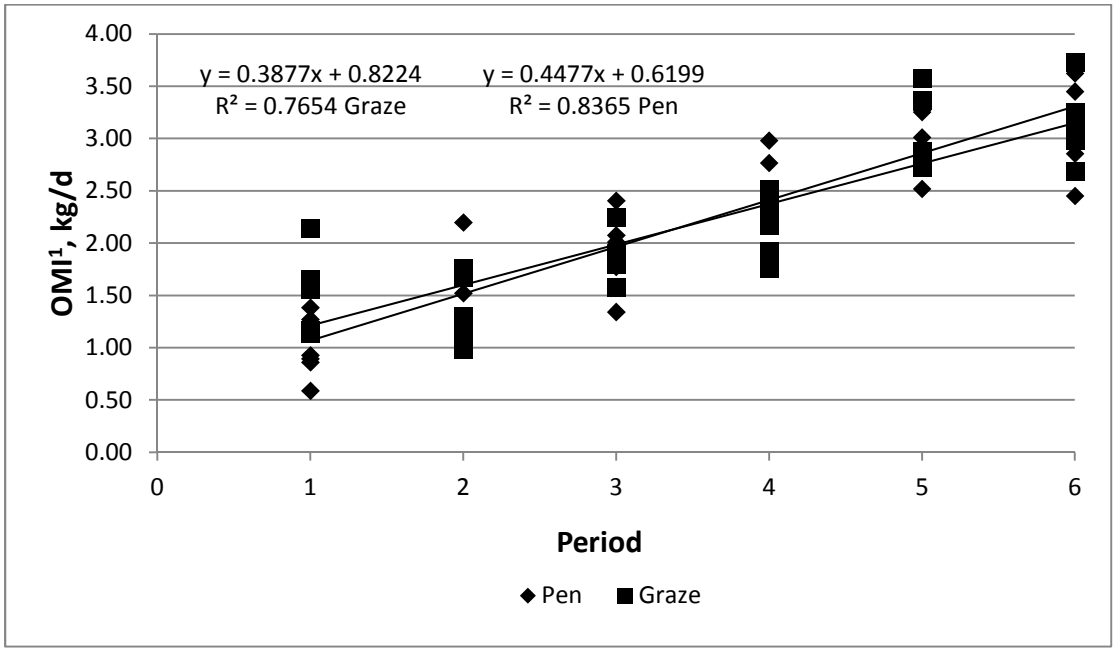
[1] Group A: Periods 1, 3, & 5 contain the same six calves.
[2] Group B: Periods 2, 4, & 6 contain the same six calves.
[3] Intakes measured for 5 days during the third week of each period listed in Table 2.

Table 6. Forage quality of hay fed to Pen animals and of diets collected from Graze pasture for Experiment 2.

Period	Forage Sample	OMD[1]	CP	NDF[1]	IADF[1]
1	Hay[2]	59.95	9.65	66.74	13.01
	Diet[3]	65.39	8.51	67.85	13.29
2	Hay	60.38	9.93	72.57	13.24
	Diet	60.55	9.23	64.08	13.17
3	Hay	60.44	8.07	72.99	14.49
	Diet	55.80	8.52	67.09	15.75
4	Hay	56.92	7.60	77.02	16.02
	Diet	52.81	6.00	53.22	16.77
5	Hay	57.38	5.19	75.49	14.84
	Diet	57.47	5.35	72.27	16.55
6	Hay	56.47	6.54	75.83	14.59
	Diet	53.98	4.77	75.78	19.38

[1]All values are on an organic matter basis.
[2]Hay represents harvested meadow hay fed to individually penned pairs.
[3]Diet represents diet samples taken in pasture of grazing pairs.

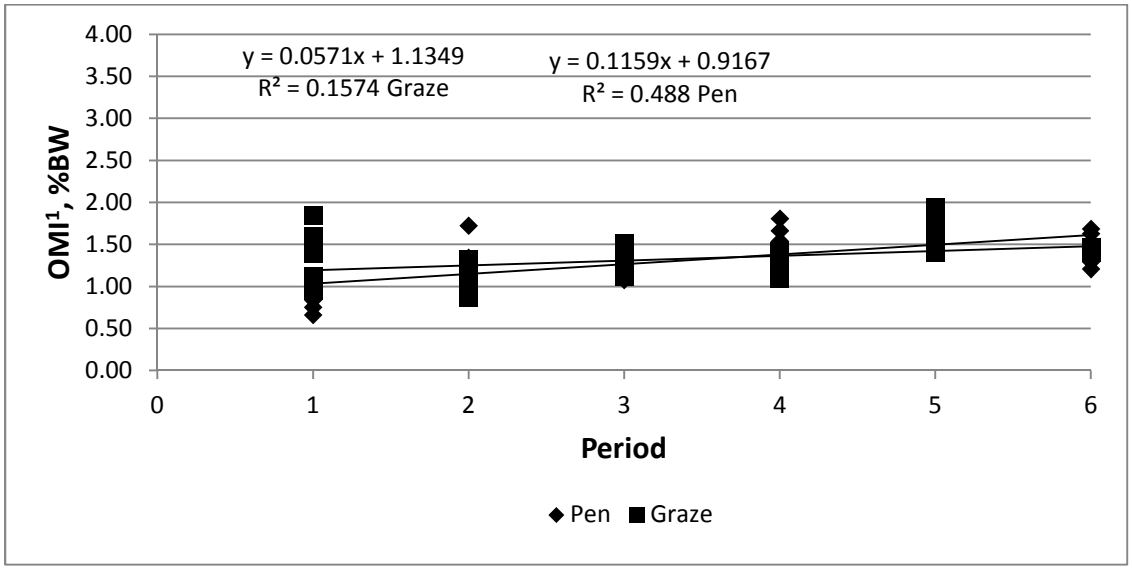
Figure 1.OMI for nursing calves in kilograms/day in Experiment 2.



[1]OMI: organic matter intake. Digestibility values used in calculating forage intake based on in vitro organic matter analysis of diet and fed forage samples.

		SEM
Environment	Slope	Intercept
Pen	0.0339	0.13202
Graze	0.03739	0.14427
	P value	
Equal Slopes	0.3347	

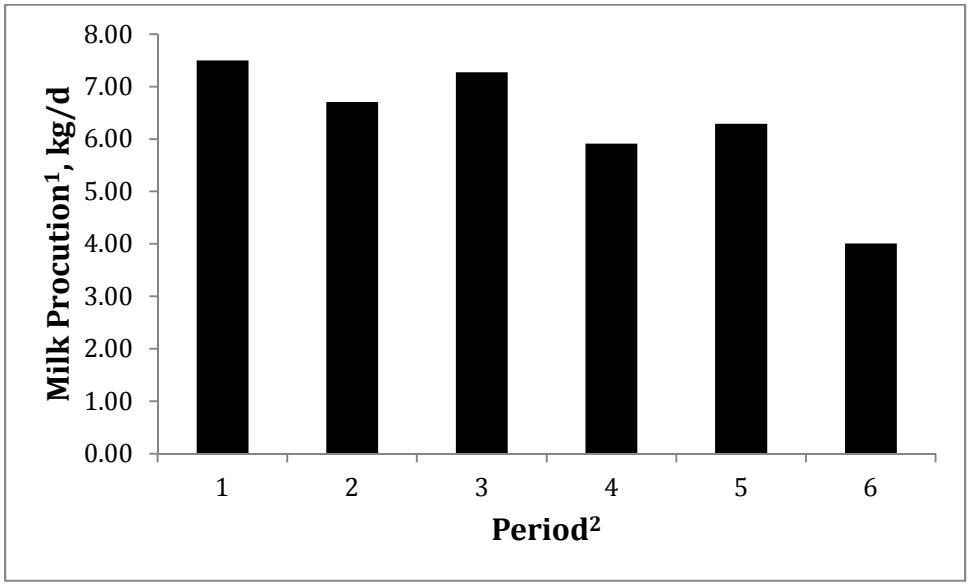
Figure 2. OMI for nursing calves as a percentage of BW in Experiment 2.



[1]OMI: organic matter intake. Digestibility values used in calculating forage intake based on in vitro organic matter analysis of diet and fed forage samples.

		SEM
Environment	Slope	Intercept
Pen	0.02035	0.07926
Graze	0.02298	0.08866
	P value	
Equal Slopes	0.1628	

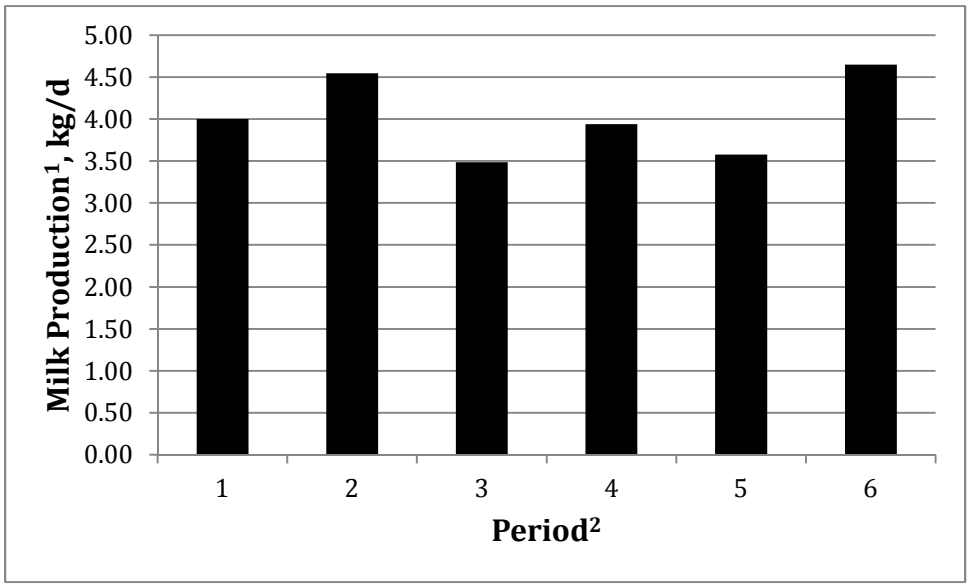
Figure 3. Cow milk production from June through mid-October weaning in Experiment 1.



[1]Weigh-suckle-weigh technique was used to determine a 24 h milk production.

[2]Periods 1-4: a 12 h WSW was used and milk weight was divided by 0.5 to determine production for 24 h time length. Periods 5-6: an 18 h WSW was used and milk weight was divided by 0.75 to determine total milk produced for a 24 h length.

Figure 4. Cow milk production from June through mid-October weaning in Experiment 2.



[1]Weigh-suckle-weigh technique was used to determine a 24 h milk production.

[2]Periods 1-4: a 12 h WSW was used and milk weight was divided by 0.5 to determine production for 24 h time length. Periods 5-6: an 18 h WSW was used and milk weight was divided by 0.75 to determine total milk produced for a 24 h length.

[3]Some individual measurements were excluded in the average in periods 1, 5, & 6 due to error in collecting weights necessary for calculations.

Appendix A: Experiment 1

Table 1. Calf calculated DMI in Experiment 1.

	Period					
Pen	1	2	3	4	5	6
DMI, kg/d[1]	0.72	1.00	1.30	2.08	2.55	2.7
DMI, % BW[1]	0.23	0.29	0.32	0.46	0.5	0.48
Graze	1	2	3	4	5	6
DMI, kg/d[1]	0.49	0.36	0.67	1.43	1.73	2.57
DMI, % BW[1]	0.35	0.26	0.44	0.77	0.79	1.19

[1]Intakes are composed of both ruminally cannulated and non cannulated calves.

Table 2. Forage quality of hay fed to Pen animals and diet samples collected from pasture animals in Experiment 1.

Period	Forage Sample	IVOMD[1]	CP	NDF[1]	IADF[1]
1	Hay[2]	58.95	11.09	71.87	13.16
	Diet[3]	54.08	8.81	70.75	16.01
2	Hay	53.44	8.88	71.52	13.94
	Diet	51.27	8.16	64.48	17.93
3	Hay	52.73	9.37	71.42	14.92
	Diet	51.67	6.56	69.79	2165
4	Hay	50.08	7.96	75.18	14.92
	Diet	37.60	10.19	56.11	26.37
5	Hay	49.99	6.73	70.59	15.80
	Diet	42.08	8.39	68.21	21.63
6	Hay	55.38	7.66	69.93	16.68
	Diet	42.68	7.20	76.50	22.05

[1]All values are on an organic matter basis.
[2]Hay represents harvested meadow hay fed to individually penned pairs.
[3]Diet represents diet samples taken in pasture of grazing pairs.

Chapter III

Comparison of In Vivo Digestibility and In Vitro Digestibility of Two Forages Fed to Steers

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ABSTRACT

Six crossbred yearling steers (IBW = 252 kg) were used in a switchback design to determine in vivo dry matter digestibility, organic matter, and indigestible ADF of two forages. Determining the in vivo digestibility of these two forages could be used as standard values in adjusting in vitro dry matter digestibility procedure to in vivo digestibility. Smooth brome grass pasture hay (Brome; *Bromus inermus)* and Nebraska Sandhills meadow hay (Meadow) were used to determine in vivo digestibility for two in vitro forage standards. Forages were supplemented with 50% urea solution to ensure degradable intake protein (DIP) was not limiting. In vitro digestibility and in vivo digestibility were compared and DM, OM, and IADF were evaluated for predicting digestibility and which method(s) could be used. There were significant differences between digestibilities of the Brome and Meadow hays (P<.0001) using in vivo techniques for dry matter digestibility (DMD), organic matter digestibility (OMD), and indigestible acid detergent fiber (IADF). In vivo values for Brome were 51.33% DMD, 54.25% OMD, and 45.47% IADF and Meadow were 55.03%, 60.30%, and 53.58% respectively. They were also significantly different (P<.0001) for the in vitro method with DMD and OMD. The in vitro values for Brome were 47.58% DMD and 47.42% OMD, while for Meadow were 56.25 and 58.83% respectively. Determining known in vivo values for these hays and finding significant in vivo digestibility differences between the two, creates standards to be used in in vitro procedures to adjust digestibility values found through the technique.

Key Words: in vivo digestibility, in vitro digestibility, forage quality, cattle

INTRODUCTION

The ability to evaluate the nutrient quality and digestibility of a forage or feed source fed to an animal can provide vast amounts of information as well as assist in formulating diets or rations to meet the animal's needs. In particular, with forages and the vast diversity of species that encompasses this classification, comes further challenges in predicting energy and nutritional content. Forages that are utilized for feed, whether harvested or by animal selection in grazing environments, can greatly vary from year to year as well as within season. Determining forage intake and if the animal's needs are being met can be evaluated through models such as the NRC (1996). To be able to use such a model, nutrient information such as TDN and protein levels must be known beforehand. Having these in vivo values for animals can be challenging, especially for grazing scenarios. Creating an evaluation process similar to in vivo digestibility and translating it to be used in a laboratory setting provides the opportunity to evaluate large numbers of samples, with less labor, time, resources, and animals.

In vitro techniques have been developed to determine forage digestibility to be able to determine in vivo values (Weiss, 1994). Mellenberger et al. (1970) created a one stage process and Tilley and Terry (1963) created a two stage process of in vitro evaluation to determine in vivo digestibility. Weiss (1994) stated that there have been several studies showing a high correlation ($R > 0.9$) between in vivo and in vitro digestibility data (e.g., Tilley and Terry, 1963; McLeod and Minson, 1974; Genizi et al., 1990). Even though they show a strong correlation, this does not mean that they are equal. Thus, to adjust for run variation, regression equations are used account for this (Weiss, 1994). In these in vitro studies, standard samples are used with known in vivo

values to utilize in creating these regression equations to adjust the in vitro digestibility values to in vivo. The objective of this experiment was to determine in vivo digestibility values of two forage hays and for them to be utilized in in vitro digestibility procedures to estimate in vivo values for unknown forage samples.

MATERIALS AND METHODS

The experiment was conducted at the University of Nebraska-Lincoln's Animal Science Complex in Lincoln, Nebraska. Animals were housed in the metabolism research area of the building in a climate controlled room at 21°C. Cattle were in individual pens with free access to water and salt blocks.

Animals, Design, & Treatments

This experiment used six crossbred (Husker Red composite- ¾ Red Angus and ¼ Simmental or Gelbveih) yearling steers (Initial BW = 242 kg) in a three period, two treatment switchback design. Periods were 21 days long and the first seven days of each period steers were fed *ad libitum* hay of assigned treatment. Days 8-21, steers were fed 95% of *ad libitum*. Two of the steers were ruminally cannulated from a previous study and distributed between treatment groups. All intact steers were assigned randomly to either group. Treatments consisted of smooth bromegrass hay (Brome) and immature meadow hay (Meadow). The smooth bromegrass hay was harvested in Eastern Nebraska and meadow hay at the Gudmundsen Sandhills Laboratory (GSL), near Whitman, NE during the summer of 2010.

Diets

Brome hay consisted of smooth bromegrass harvested hay (*Bromus inermus).* Meadow hay was harvested from sub-irrigated meadows in the Sandhills of Nebraska 11 km northeast of Whitman, NE (42° 04'N 101° 26'W, elevation = 1,075m). Volesky et al., (2004) found that cool-season grasses, sedges, and rushes dominated the meadows. The most common grasses were slender wheatgrass [*Elymus trachycaulus* (Link) Matte], redtop bent (*Agrostis stolenifera* L.), and timothy (*Phleum pretense* L.). Other grasses present included Kentucky bluegrass (*Poa pratensis* L.) and smooth bromegrass (*Bromus inermus* Leyss.). Wooly sedge (*Carex lanuginose* Michx.) and several spike rush species (*Eleocharis* spp.) were also abundant. The primary legumes included white clover (*Trifolium repens* L.), alsike clover (*Trifolium hybridum* L.) and red clover (*Trifolium pretense* L.). Warm-season grasses present in minor amounts were prairie cordgrass (*Spartina pectinanta* L.) and big bluestem (*Andropogon gerardii* Vitman).

A tub grinder with a 10 mm screen was used to chop all hay prior to initiation of the trial. Hays were stored separately in a cement building to prevent spoilage and contamination. The first7 days were adaptation to treatment diet and steers were fed *ad libitum*. During days 8-21, cattle were fed 95% of individual *ad libitum* intake. Each collection period was from days 15-21. Feed refusals (orts) were collected and weighed daily throughout the trial. Hays were top dressed with a 50% urea solution to ensure degradable intake protein (DIP) was not limiting. Feed grade urea was used in the solution. Metabolizable protein (MP) levels contributed by the two hays were calculated to determine if DIP would be limiting or not. Meadow hay values were 7.66% CP, 2.25% UIP, and 57% TDN and Brome hay values were 8.24% CP, 2.25% UIP, and 50% TDN.

These equations were used for both Meadow and Brome hay to determine if the DIP potential was great enough to meet the cattle's needs.

Meadow Hay Calculations:

MP Content of Hay

MP (g/kg) = (UIP (g/kg) * 0.8) + (TDN (g/kg) * 0.13 * 0.8 *0.8)

MP (g/kg) = (1.724 * 0.8) + (570 * 0.13 * 0.8 * 0.8)

MP = 48.3232 g/kg

DIP potential (DIPP)

DIPP (g/kg) = (0.13 * TDN (g/kg)) – DIP (g/kg)

DIPP (g/kg) = (0.13 * 570) – 74.8576

DIPP *= -0.7576 g/kg*

Total MP = 48.3232 g/kg

Brome Hay Calculations

MP Content of Hay

MP (g/kg) = (UIP (g/kg) * 0.8) + (TDN (g/kg) * 0.13 * 0.8 *0.8)

MP (g/kg) = (1.854 * 0.8) + (500 * 0.13 * 0.8 * 0.8)

MP = 43.0832 g/kg

DIP potential (DIPP)

DIPP (g/kg) = (0.13 * TDN (g/kg)) – DIP (g/kg)

DIPP (g/kg) = (0.13 * 500) – 80.546

DIPP *= -15.546 g/kg*

Total MP = 43.0832 g/kg

Based on calculations, the DIPP for Meadow Hay was negative at -0.7576 g/kg, which suggests that requirements would minimally be met. The calculated DIPP for Brome hay was also negative at -15.546 g/kg. To ensure that DIP would not be limited in the rumen

due to the small negative DIPP values to meet the animal requirements, a 50% urea solution was top dressed to the hay treatment fed cattle daily. The Meadow hay was top dressed with 6.27 mL/kg and Brome hay was dressed with 2.24 mL/kg hay fed daily.

Steers were fed at 0730 hour after orts were collected and weighed daily. Diets and orts were sampled three times per week and composited by week by steer. Ort samples for collection weeks were from day 13 to 19. This was to account for total tract travel time of diet consumed to relate feed consumed to time of fecal collection. All samples were stored in a cooler until composited and oven DM was conducted. Dried samples were stored in totes until sample analysis was conducted.

Total Fecal Collection

Total fecal collection was conducted on all six steers for seven days each period. Harnesses and bags were attached to steers at 1700 hour on day 14 of each period. Bags were weighed prior to placement on steers. Bags were weighed at 0700 hour each day. Contents were emptied into separate garbage bags for each steer and stored in labeled totes in a cooler until 1700 hr. Emptied bags were reweighed and reattached to the steer to continue total collection. At 1700 hour, bags were removed and weighed again. Sample was removed and composited by steer with the morning collection using a 22.7 kg feed mixer. Bags were reweighed and reattached to the steer. Total fecal collection per day per steer was mixed in feed mixer for 2-3 minutes. A 10% subsample was collected per steer per day, labeled, and stored in a cooler until the end of the collection period. If the calf lost their bag or fecal sample spillage was not recoverable, that day's measurement was excluded from the average daily fecal output calculation.

At the end of each collection period, daily fecal subsamples per steer were composited in the feed mixer. Three 0.9 kg samples were taken to determine DM content and saved for analysis for each steer. One 0.9 kg sample was frozen per steer per period for future analysis. Excess fecal sample was disposed of after each daily and weekly composite. Mixer was rinsed and cleaned out between each steer fecal compositing.

Sample Analysis

Diet and ort composite-samples were collected each week and dried in a 60°C forced-air oven for 48 h. Dry matters were calculated and recorded. Diet and ort samples from each collection periods were retained for lab analysis. After drying, samples were ground through a Wiley mill 2mm screen and approximately half of the 2mm ground samples were ground through a 1mm screen. Samples were analyzed in the laboratory for DM, OM, IVDMD, in vitro organic matter disappearance (IVOMD), and indigestible acid detergent fiber (IADF). Dry matter and OM were determined for all samples following the AOAC standard procedure (1996). In vitro DM and OM disappearance were determined using the Tilley and Terry method (1963) with a 50:50 mixture of the rumen inoculum and McDougall's buffer with the inclusion of 1 g/L of urea to the buffer solution (Weis, 1994). Samples were incubated in 100 mL tubes in a water bath at 39°C for 48 hours and swirled every 12 hours. After incubation, samples were filtered, dried for at least 5 hours in a 100°C forced air oven, weighed, and burned in an ash oven for 6 hours at 600°C to determine the DM and OM content of residue for the calculated IVDMD and IVOMD.

Three, weekly composited 0.9 kg fecal subsamples were taken per steer and dried in a 60 C forced air oven for 72 hour to determine DM. Dried fecal samples were ground through a Wiley Mill using a 2-mm and 1-mm screen. Indigestible acid detergent fiber (IADF) analysis was conducted on hay samples and weekly composited fecal samples. Hay samples proceeded with a two-stage in vitro digestion technique of Tilley and Terry (1963) with 0.5 g samples in 50 ml centrifuge tubes inoculated with 30 ml of 50:50 mixture of the rumen inoculum and McDougall's buffer with 1 g/L urea to buffer solution (Weis, 1994). After stage one of pre-digestion, tubes were centrifuged and excess inoculum was removed. All samples were then inoculated with the same ratio of inoculum mixture listed previously and incubated at 39°C for 96 hours in a water bath. Contents were transferred to a 600 ml beaker and using a wash bottle containing ADF solution, rinsing all particles clinging to the tube into the beaker. Solution was added to bring total contents to 150 ml. ADF analysis was continued with the Goering and Van Soest technique (1970), filtering the hot solution through tared filters. Filters were dried for at least 5 hours in a 100°C forced air oven and after weighing, burned in a 600°C ash oven for 6 hours. Percent IADF for feed and fecal samples was calculated:

$$\textbf{\% IADF} \text{ (feed or fecal)} = 100 \times \frac{\text{ADF residue (DM) - blank}}{\text{Sample wt (DM)}}$$

Statistical Analysis

The composition of in vivo DMD, OMD, and IADF data were analyzed using Glimmix procedures of SAS (SAS Inst. Inc., Cary, NC) to detect differences in digestibility by treatment. The model included treatment and period as fixed and calf as

random effect. The REG procedure of SAS was used plot data of in vivo and in vitro digestibility to detect differences in digestibility by period.

RESULTS AND DISCUSSION

One ruminally cannulated calf's data were not included in the results due to chronic health complications throughout the trial. Forages were compared for in vivo vs. in vitro digestibilities and by several methods of measurement. Methods evaluated included DMD, OMD, and IADF for in vivo digestibility and DMD and OMD for in vitro digestibility. Data from this comparison are found in Table 1 where there are significant differences in digestibility between Brome and Meadow hays in all in vivo and in vitro digestibility methods that were evaluated ($P<.0001$). Meadow hay digestibilities were higher than Brome digestibilities across the table for all methods ($P<.0001$). The in vivo digestibilities increased when comparing DMD to OMD methods for both Brome, 51.33% to 54.25% and Meadow hays, 55.03% to 60.30%. This would tend to be natural due to removing of the inorganic matter from the sample, thus increasing the digestibility of the forage sample (Weiss, 1994). Figure 1 compares the in vivo vs in vitro digestibilities for both forages using the three methods. The visual comparison between Meadow and Brome hays for each method reinforce their differences in digestibility. Having a range in digestibility values will assist in evaluating other feed samples for digestibility through in vitro techniques.

In vitro digestibilities showed a smaller change in percentage units between DMD and OMD (Table 1) for Brome and Meadow hays. In vitro digestibility using IADF was found to be significantly different between the Brome (45.47%) and Meadow (53.58%)

(P<.0001), yet values were lower than of those measured with DMD and OMD. This range in digestibility values is supported by Cochran et al. (1986) who also found that IADF had higher variability for measuring digestibility in harvested and grazed forage. With a significant difference between the two hays, by design, this provides the opportunity to utilize them as potential standards for in vitro digestion techniques.

Hays tested were measured for in vivo digestibility over three periods. With both the Brome and Meadow hay, there were changes in digestibility between periods. In Figure 2, Brome DMD saw an increase in in vivo digestibility as the periods progressed, as well as with OMD. Theoretically the digestibility of the hay would change minimally between periods. Potential causes for the increase in digestibility could be loss of fecal matter during collection periods or miscalculation in forage intake due to loss of feed from waste or ort collection. If either or both of these measurements are incorrect, it will be reflected directly in the calculations of in vivo digestibility. Animal adaptation over time to the diets consumed and increased efficiency could also affect digestibility (Cochran & Galyean, 1994). The IADF measure of digestibility showed the least change in digestibility across periods. The amount of ADF content shouldn't change over time, thus less fluctuation. The challenge in using IADF is in the recovery of the marker itself. In Figure 3, Meadow hay digestibility showed less variation over time for DMD and OMD, yet while IADF had a slightly greater range in value. Both graphs depict the variation that can be measured due to individual animal digestibility (Weiss, 1994). This reinforces the importance for number of animals, replication, and continuity in specimens selected for a trial.

The use of determining in vivo digestibility is both labor intensive and time consuming. Yet, with this technique, comes greater understanding of how the animal processes and digests its consumed diet. Defining the in vivo digestibility of the two forage samples in this trial provides numerical values for them to be utilized with the in vitro technique in the lab. The in vivo OMD for Brome hay, 54.25%, and Meadow hay, 60.30%, can be utilized in regression equations to adjust for run effect in in vitro analyzing of unknown forage samples. Also, being found to be significantly different ($P<.0001$) allows the lab technician greater confidence in evaluating new feed samples and measuring the feed values against these standard samples as part of a range in digestibilities to measure against.

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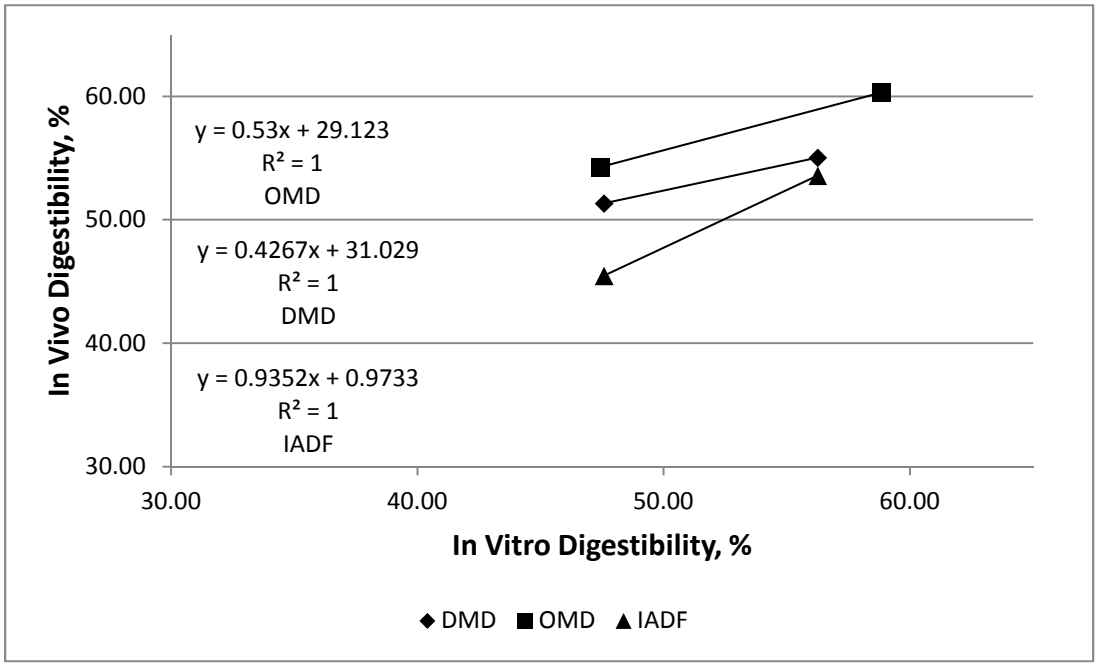
Table 1. Comparing digestibilities between in vitro vs in vivo.

	Method	Brome	Meadow	SEM	P value
In vivo (%)	DMD[1]	51.33	55.03	0.67	<0.0001
	OMD[2]	54.25	60.30	0.73	<0.0001
	IADF[3]	45.47	53.58	0.73	<0.0001
In vitro (%)	DMD	47.58	56.25	0.59	<0.0001
	OMD	47.42	58.83	0.73	<0.0001

[1] DMD: Dry Matter Digestibility
[2] OMD: Organic Matter Digestibility
[3] IADF: Digestibility determined by Indigestible Acid Detergent Fiber on DM basis

Figure 1. In vitro vs in vivo digestibility by method[1].



[1]Digestibility is by method used to determine it. DMD: dry matter digestibility, OMD: organic matter digestibility, IADF: digestibility from indigestible acid detergent fiber (1-IADF=digestibility).

[2]Brome hay is the lower digestibility values for each method and Meadow Hay is the higher per method.

Figure 2. In vivo digestibility for Brome hay by method.

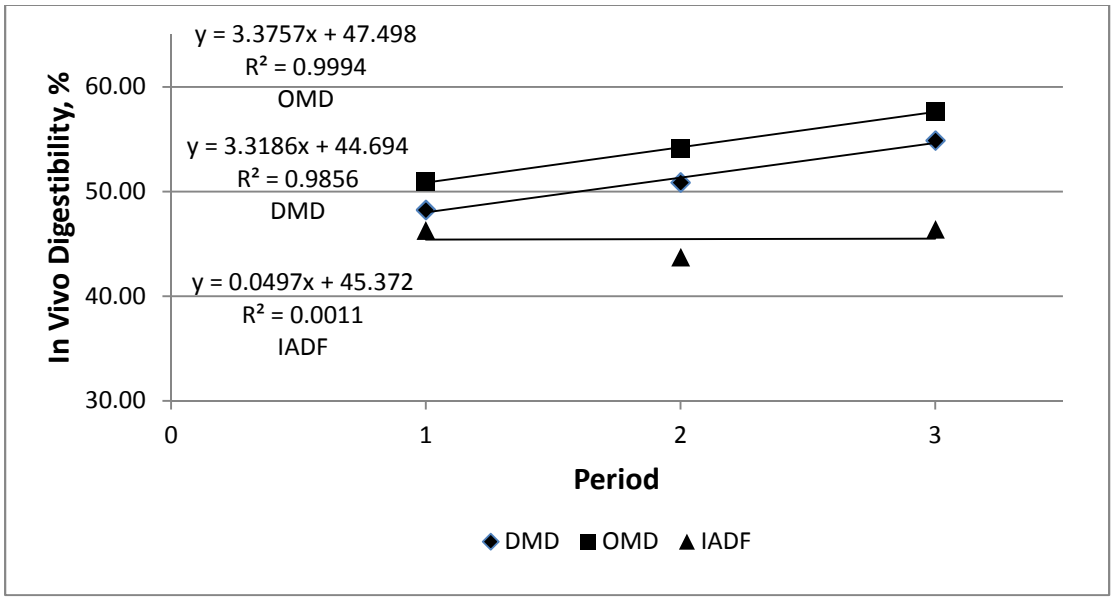


Figure 3. In vivo digestibility for Meadow hay by method.