

SUPPLEMENTATION OF CATTLE FED LOW-
QUALITY PRAIRIE HAY WITH
FIBER, STARCH OR
PROTEIN

By

TIMOTHY NOEL BODINE

Bachelor of Science

University of Idaho

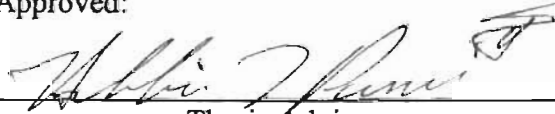
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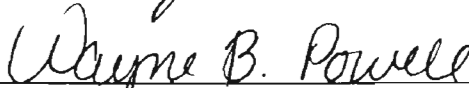
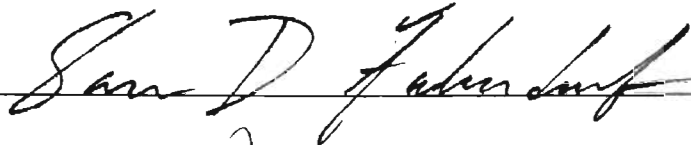
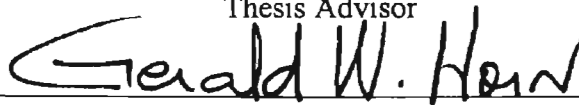
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FORMAT OF THESIS

This Thesis is presented in the Journal of Animal Science style and format, as outlined by the Oklahoma State University graduate college style manual. The use of this format allows the independent chapters to be suitable for submission to scientific journals. Two papers have been prepared from the data collected for research to partially fulfill the requirements for the M. S. degree. Each paper is complete in itself with an abstract, introduction, materials and methods, implications and literature cited section.

Chapter I

Introduction

Supplements should be formulated to provide required nutrients necessary to achieve a specific production goal. Forage consumption is not always adequate to meet a desired level of animal performance. In most production situations, protein, energy or usually both limit performance. It is imperative to understand the forage base and what nutrient(s) is(are) limiting in order to use supplementation effectively. Ideally, supplementation would be minimal due to matching nutrient requirements of the animals with nutrient supply from the forage. Yet, many systems will not support economically feasible levels of animal performance without supplementation even when animal requirements are closely matched with forage supply. Understanding the effects of various feedstuffs used to formulate supplements helps in assuring that the supplement will aid in reaching the desired production objective. However, interactions between feedstuffs, as well as the various components in those feedstuffs, reduce our ability to accurately predict animal response. Factors that influence these interactions include level of forage intake, forage digestibility, grazing time, efficiency of ME use and DOM intake which all can affect energy intake by grazing animals.

Common belief (Nickel, 1999) is that supplements formulated from grain will depress forage intake and utilization while protein supplements will improve forage utilization. Previous research (Chase and Hibberd, 1987) has indicated that feeding large amounts of an energy supplement high in starch (based on cereal grain) will decrease forage intake and utilization and may actually decrease animal performance. While this clearly seems to be a negative side effect, grain can often be the most economical source

of energy. The price and energy advantage of cereal grains has led many researchers to attempt to find methods to utilize grains effectively to increase production of forage-fed livestock. However, other research has investigated improving performance of livestock by using energy supplements based on highly digestible fiber sources. Findings from this research have been interpreted to indicate that feeding fibrous byproducts as energy supplements will not replace forage in the diet and can increase animal performance. For every trial where these beliefs are confirmed, another trial exists that suggests the opposite. Consequently, much of the research on supplementation has had varied results which can often be explained by the design of the study, the formulation of the supplement, the quality of the forage or the level of feeding.

This research was designed to determine if the negative associative effects due to grain supplementation of cattle consuming low-quality forages could be alleviated or eliminated by the addition of protein sources that are degraded in the rumen. Current research in supplementation has addressed the ratio of degradable intake protein to digestible nutrients as a method to determine protein adequacy for microbial fermentation based on the latest Nutrient Requirements for Beef Cattle (NRC, 1996).

Chapter Two provides an overview of past research involving supplementation of forage-fed ruminant livestock with feedstuffs of various sources, fed at various levels and with differing nutrient concentrations. Two trials involving supplemented beef steers fed low-quality prairie hay are detailed in chapters Three and Four. Chapter Five is a summary of the research efforts undertaken for the completion of this thesis. The Appendix contains supporting data relative to each experiment that was not included in the journal article format of the chapters.

Chapter II

Review of Literature

Response of ruminants to supplementation

Livestock producers have offered supplemental nutrients to their flocks or herds for many years. However, after years of research, clear guidelines do not exist so that the addition of any limiting nutrients to livestock diets will result in an increase the performance of domestic animals. Supplementation strategies intended to increase animal performance by increasing intake and(or) digestion may have other, less well-documented effects. Ruminants differ from other species in response to nutrient deficiencies. While many other animals increase intake when confronted with a dietary deficiency, ruminant livestock will typically reduce intake in situations when nutrient(s) deficiencies occur. This makes it imperative that in the process of formulating supplements, we do not inadvertently create a deficiency and subsequently cause intake to decrease, rather than increase, as we intended.

Supplements have often been used to increase or maintain a desired level of animal production. This may be related to economic parameters, animal well-being issues, or the need to ensure continued animal reproduction. Increasing nutrient intake from forage and supplement is typically positively related to improvements in animal performance. Supplementation strategies have often increased overall intake, while different supplements have had varied effects on forage intake. Altering forage intake may be desired in some instances but not in others, while using supplementation to increase total diet intake may not always be economically or logistically feasible. This is important from an economic point of view as well as a livestock production standpoint.

Effects of supplementation on grazing behavior

Supplementation may alter animal behavior in ways that conflict with expected results based on digestive system effects. Feeding supplements may alter bite size, bite frequency (rate of biting), grazing time, harvesting efficiency, distance traveled, time spent ruminating, or efficiency of energy utilization. If supplementation decreases grazing time, maintenance energy expenditures from walking and eating would decrease and vice versa (Caton and Dhuyvetter, 1997). Higher levels of grain supplementation has been shown to decrease livestock grazing time (Krysl and Hess, 1993) while decreases in grazing time due to protein supplementation have also occurred (Barton et al., 1992). However, Beaty et al. (1994) showed no change in grazing time as a result of increasing supplemental protein concentration. This may have been due to differences in climate, amount of supplement fed, or forage type. No difference was found due to frequency of supplementation for cows supplemented three vs seven times per week (Beaty et al., 1994). Effects of grain supplements on harvesting efficiency were variable and dependent on feeding time while protein supplements increased harvesting efficiency (Krysl and Hess, 1993). Supplementation has had inconclusive effects on the distance traveled during grazing (Krysl and Hess, 1993, DelCurto et al., 1990c, Adams, 1985). Adams (1985) suggested that corn supplementation had time dependent (am vs pm) effects on forage intake, energy intake, ADG and time spent grazing. While supplementation altered grazing patterns and grazing did not occur immediately following supplementation, afternoon feeding had less impact on grazing behavior than did cattle supplemented in the morning. Adams (1985) also found no effect of supplementation or time of feeding on other behavioral measures (lying, standing or

ruminating) or in estimated energy expenditure. However, DelCurto et al. (1990c) and Minson (1990) have shown little decrease in grazing time due to supplements being fed. Vanzant et al. (1990), DelCurto et al. (1990b) and Barton et al. (1992) showed little change in the nutrient composition of forage selected by steers supplemented with grain or protein. Owensby et al. (1995) indicated that over six years, IES stocker cattle fed increasing levels of supplemental milo had increasing residual standing biomass of tallgrass prairie at the completion of the grazing season, while forage species composition was not greatly altered. This appears to indicate reduced forage intake by supplemented cattle, while all cattle had the potential for similar diet quality. Producers supplementing cattle should be aware of potential changes in animal behavior that may alter the expected outcome of a given supplementation strategy.

Supplementation effects vary with forage quality

Many researchers have indicated that the effects of supplementation change with forage type. Caton and Dhuyvetter (1997) suggested that substitution ratios are sensitive to forage quality, especially forage CP (Minson, 1990). Horn and McCollum (1987) and Owens et al. (1991) suggested that greater substitution ratios (less forage intake as a result of supplement) occur as forage quality (digestibility) increases. However, this is not always the case, as Matejovsky and Sanson (1995) reported greater forage intake changes as level of corn supplementation increased, but changes were smaller as forage quality increased.

Intake. Supplementing high quality forages has typically reduced forage intake and increased overall diet intake. Matejovsky and Sanson (1995), Hess et al. (1996) and Elizalde et al. (1999) reported decreased forage intake due to energy supplementation of

medium and high quality grass hay, irrigated endophyte-free fescue, or fresh alfalfa, respectively. Krysl et al. (1989) reported no effect on intake of blue grama range, while Mieres (1992) found increasing levels of corn supplementation increased forage and total intake on grazed native tallgrass prairie in June, but peaked and decreased in August, forage quality declined. This points to an imbalance between degradable intake protein (DIP) and total digestible nutrients (TDN) during the late-summer grazing season. Some of the reported decreases in forage intake can be explained by the level of supplementation, specific nutrient deficiencies or cattle type.

Protein supplementation has not been extensively researched on higher quality forages. Matejovsky and Sanson (1995) found no increase in forage or total NDF, ADF or DM intake by sheep when medium and high quality grass hays were supplemented with protein. Forage and consequent total intake may be more difficult to manipulate via protein supplementation as forage quality increases.

Substitution. When supplement intake replaces forage intake, supplement has substituted for forage. Sanson and Clanton (1989) suggested that substitution rates will increase as forage quality increases and the different substitution ratios noted in their study were a result of differences in forage quality. Hess et al. (1996) reported substitution ratios of 1.97, 2.38 and 2.10 for corn or two levels of wheat bran, while Elizalde et al. (1999) found a substitution rate of .69 units of forage for each unit of corn.

Digestion. Different supplement types have had variable effects on digestion. A portion of this variation can be explained by differences in digestion of specific dietary components (forage or total DM, OM, fiber) or how they are measured (rate or extent, ruminal, *in situ* or total tract). Krysl et al. (1989), Matejovsky and Sanson (1995) and

Elizalde et al. (1999) found no effect on forage DMD, increased total DMD and no effect on fiber digestion for steers fed medium and high quality forages with protein or grain supplements. Krysl et al. (1989) suggested that low-level supplementation had little effect on forage utilization or site of digestion regardless of source, and reported no effect on rate of *in situ* NDF digestion (ISNDFD). Hess et al. (1996) found a greater rate of ISNDFD for control diets with no differences between cracked corn or two levels (.34 or .48% BW) of wheat bran while rate and extent of ISNDFD decreased as forage maturity increased. Mieres (1992) found increasing levels of corn supplementation increased forage, fiber and total diet digestibility and intake of DOM on grazed native tallgrass prairie in June, but peaked and decreased in August with declines in forage quality. This again points to imbalanced DIP and TDN. This wide range in response to supplements makes it difficult to predict expected animal response due to supplement feeding.

Ruminal fermentation. One primary mechanism targeted by supplementation is the supply of fermentation substrates for ruminal microbes. Many of the ruminal measurements commonly cited in research are an attempt by the researcher to quantitatively describe the effects on ruminal microflora. These can be divided into three major categories: pH, NH₃ and VFA concentrations.

Hess et al. (1996) suggested that pH was not a major factor in reductions in forage intake, and that improvements in animal performance were possibly a result of increased ruminal fermentation due to supplementation. Krysl et al. (1989) and Hess et al. (1996) found no effect on ruminal pH while Elizalde et al. (1999) found decreased pH as corn increased. However, average pH was never below 6.0, and neither fiber nor forage OM digestion were reduced.

Hess et al. (1996) and Elizalde et al. (1999) found supplemented steers to have greater total VFA while Krysl et al. (1989) reported that supplementation had no effect on total VFA. This may have been due to the low-level of supplementation in Krysl's study.

Krysl et al. (1989) reported increased ruminal NH_3 as a result of supplementation while Hess et al. (1996) reported wheat bran supplemented steers had greater NH_3N than corn or control fed cattle, possibly due to differences in DIP.

Passage. Much supplementation research emphasis has been given to the effects of supplements on the rate of ruminal passage. Passage rate is crucial to animal production because performance is driven by intake, and intake is regulated by outflow. Passage also exerts an effect on extent and rate of digestion, microbial turnover and removal of fermentation endproducts.

Krysl et al. (1989) and Hess et al. (1996) reported no effect on particulate or fluid passage rates. Elizalde et al. (1999) found a quadratic response in fluid passage rate to increasing corn with the greatest rate found at .8% BW corn and the least at 1.2% BW. Hess et al. (1996) reported no effect of corn or two levels of wheat bran supplementation on gastrointestinal fill or rumen volume for steers grazing high-quality fescue.

Performance. Animal production is influenced by many of the previously discussed factors. Individual animal performance can be improved by any one of these factors, but changes in one mechanism usually result in a myriad of changes, since many of these mechanisms are interdependent. Poppi and McLennan (1995) suggested that protein supplements improve ADG with a decreasing response as forage quality increases. Lake et al. (1974) and Hess et al. (1996) found corn supplementation increased ADG for steers grazing high-quality irrigated pastures while Hess et al. (1996) reported corn increased

ADG more than wheat bran supplemented steers. Differences in animal response between supplement sources suggest that a balance between protein and energy supply exists. The greatest level of animal performance will be achieved by feeding the supplement with the combination of protein and energy that most closely achieves that balance in the total diet.

Control of Intake

Many different researchers have identified diverse and complex factors in specific situations that are involved in the control of intake. However, no one has been able to identify any one factor or even a small group of factors that can consistently explain variation in feed intake. Considerable research has been conducted on intake limitations due to ruminal fill of low-quality, high-roughage diets without clear results occurring in the realm of actual physiological levels. Allen (1996) suggested the possibility that reserve ruminal capacity combined with increased ruminal outflow, could explain research that has failed to show an intake depression due to ruminal fill.

Forbes (1996) suggested that several effects controlled intake in an additive manner. Combined infusion of organic acids and inert fill decreased intake in a manner that physiologically possible levels of each individual component could not explain. Fisher (1996) also suggested the presence of many factors and used protein, digestibility and NDF to represent small intestinal protein flow feedback, chemostatic feedback from energy level and ruminal distension feedback.

Illius and Jessop (1996) suggested that intake was a physiological effect controlled by many integrated signals including perception and learning. Animal physiological

status, metabolic pathways, energy, protein and synthetic capacity all may play a role in controlling intake (Illius and Jessop, 1996).

Ketelaars and Tolkamp (1996) suggested that maximizing the efficiency of oxygen utilization may be one of the main factors working to control intake. Oxygen efficiency is expressed as the relationship between gain of net energy and cost of oxygen consumption. Intake would occur at the level where this ratio is greatest (Ketelaars and Tolkamp, 1996). Factors influencing this ratio would include feed composition, body composition, physiological status, metabolic load and environmental factors. This type of theory appears to have much promise in explaining observed responses in intake, performance and behavior and fits well from an evolutionary perspective. While ruminal fill may have an effect on intake in a few specific situations, it appears that many coordinated physiological signals based on animal requirements play a major role in determining level of intake.

Effects of supplementation on intake

Supplementation can effect intake via many mechanisms. McCollum and Horn (1990) indicated that protein deficiencies would decrease intake while adding protein to meet requirements would increase intake. Protein supplementation of low-quality forages commonly increases forage intake. Owens et al. (1991) suggested that protein supply is critical because protein is an important factor involved in controlling intake, and performance is directly related to intake. Responses to supplemental protein vary with forage CP and are usually greater with lower forage protein concentration. Supplemental protein is typically required when forage CP falls below 6-8% (McCollum and Horn, 1990). However, intake response is not solely a result of forage CP level (NRC, 1987).

Caton and Dhuyvetter (1997) stated that the effects of energy supplementation on intake could differ depending on the amount of supplement fed. Horn and McCollum (1987) suggested that concentrate intake greater than 30 g/kg BW⁷⁵ would decrease forage intake. However, increases in forage intake as a result of small amounts of grain supplementation have occurred, especially with sheep (Caton and Dhuyvetter, 1997). Feeding supplements formulated from fibrous by-products has typically resulted in very little effect on forage intake (Caton and Dhuyvetter, 1997). One possible explanation for depressed forage intake due to grain supplementation may be a result of increasing specific nutrient deficiencies (DIP or ruminal NH₃) common on low-quality, high-fiber forage diets (NRC, 1996).

Other possible factors that may play a role in controlling intake that will not be discussed in this review include dietary fiber level and(or) structure, rate of digestion, extent of feed processing and secondary or phenolic compounds. While physiological status can alter intake, many dietary and environmental factors also influence intake (NRC, 1996). Improved genetics, decreasing temperature, increasing photoperiod, increasing forage availability and growth-promoting implants all tend to increase feed intake. Increasing temperature, decreasing photoperiod and forage availability, monensin and nutrient deficiencies typically reduce feed intake (NRC, 1996).

Many studies have shown increased forage intake due to increasing amounts or concentration of supplemental protein (Rittenhouse et al., 1970, McCollum and Galyean, 1985, Guthrie and Wagner, 1988, Hannah et al., 1991, Matejovsky and Sanson, 1995, Köster et al., 1996 and Olson et al., 1999). Many of these researchers have linked increased forage and total intake due to protein supplementation to increases in passage

rate. However, Barton et al. (1992) and Hollingsworth-Jenkins et al. (1996) found little effect on forage intake of low-level protein supplementation in cattle grazing dormant forages. Possible explanations for this include animal requirements or supplement type.

Supplementation with grain or by-products has not always resulted in consistent results. Chase and Hibberd, (1987) found decreased hay and total DM intake as corn increased. They attributed the decrease to reduced rates of digestibility and passage, which were a result of a ruminal NH_3 deficiency. Ruminal NH_3 was deficiency due to decreased supplemental DIP and DIP:DOM ratios, as well as being compared against a protein supplemented diet rather than a true control. Other research involving positive controls has not always shown this magnitude of decrease. Catlett (1991) found a slight decrease (< 1 kg) in hay intake but increased total intake for whole, cracked, ground or pelleted corn mixed with cottonseed meal and fed at 2.8 kg/hd/d. Other work (Carey et al., 1993) has reported that forage and total intake did not differ between ground corn, barley or beet pulp supplements when compared to a soybean meal supplemented diet. Heldt et al. (1998) fed wheat midds or a corn/SBM mix and found similar forage and total intake vs a soybean meal control, while high levels of wheat midds depressed intake.

In trials involving low to moderate quality forages and grain or by-product supplements fed at a variety of levels, decreased forage intake and increased total intake have commonly occurred (Jones et al., 1988, Sanson and Clanton, 1989, Sanson et al., 1990, Martin and Hibberd, 1990, Galloway et al., 1991, 1993a,b, Forster et al., 1993, Matejovsky and Sanson, 1995, Marston and Lusby, 1995 and Garcés-Yépez et al., 1997). Pordomingo et al. (1991) reported low-level corn supplementation increased intake while levels greater than .2% BW decreased both forage and total intake. Feng et al. (1995)

reported hay intake decreased as level of barley increased (.73 to .90% BW) with no change in total intake and corn and barley (equal starch) had similar hay intake while Brake et al. (1989) reported grain decreased forage intake and to a greater extent for corn than for barley. In two trials, Vanzant et al. (1990) either fed increasing levels of milo or fed similar levels of corn, wheat or milo and found no effect on forage intake and increased total intake. Chan (1992) found corn or soybean hull supplementation increased forage intake and total DMI. Arelovich et al. (1983), Fleck et al. (1988) and Sunvold et al. (1991) found greater forage and total intake with protein, by-product or grain supplementation than with an unsupplemented control.

In trials involving grain fed with additional supplemental protein, DelCurto et al. (1990a,b), Hannah et al. (1991) and Beaty et al. (1994) fed milo and soybean meal in various combinations and increased forage and total intake as soybean meal increased. Guthrie and Wagner, (1988) found no decrease in forage intake and an increase in total intake from corn/SBM vs an unsupplemented control while Freeman et al. (1993) found no effect of milo/cottonseed meal supplements on total intake. Rittenhouse et al. (1970), Fick et al. (1973), and Olson et al. (1999) reported forage OMI decreased while total OMI increased as starch increased. Rittenhouse et al. (1970) found no effect of increasing protein from soybean meal. However, Fick et al. (1973), DelCurto et al. (1990a) and Olson et al. (1999) found that forage intake increased as DIP increased within each level of starch and was higher than cattle fed an unsupplemented control. This suggests the amelioration of negative associative effects of starch on forage intake when DIP was fed. When effects of supplemental starch were compared across treatments with similar supplemental DIP:TDN ratios, Olson et al. (1999) found increased forage and total OM

intake. Feeding greater amounts of fermentable OM to cattle grazing or fed harvested low-quality forages appears to be greatly impacted by the concentration of DIP in the supplement. It also appears that many of the studies previously conducted that have shown large depressions in forage intake due to supplemental starch have fed high levels, not included any additional DIP, or have not had a negative or true control for comparison.

Substitution effects

When intake of supplement replaces intake of forage, we say that the supplement has substituted for the forage. However, it is more difficult to predict what influence this will have on animal performance or economical considerations. Replacing low-quality forage (45% TDN) with corn (90% TDN) may actually increase energy intake. This will only occur if the digestibility of the forage is not greatly reduced. Horn and McCollum (1987) suggested that substitution ratios were varied by forage quality, animal physiological state, level of activity and livestock requirements.

Chase and Hibberd (1987) found a range of substitution ratios of supplement for forage from .86:1 to 1:1.83 when compared to a high level of forage intake due to a protein supplement. However, other trials with protein controls have not reported as great of ratios. Carey et al. (1993) found an average substitution rate of .56 when corn, barley, or beet pulp were compared to a protein supplemented diet. Catlett (1991) found substitution rates of .14 to .44 as processing of corn increased for supplements fed with cottonseed meal and compared to a cottonseed meal control. Martin and Hibberd (1990) found substitution rates of .21 for levels of soybean hulls

Caton and Dhuyvetter (1997) suggested average substitution ratios for barley-based supplements would range from .4 to .48. Sanson and Clanton (1989) found substitution ratios between .19 and .52 while Sanson et al. (1990) reported substitution rates of .2 when corn was added to low-quality hay. Rittenhouse et al. (1970) found average substitution rates of .35 for corn and low-quality hay. When supplementation has increased hay intake, no substitution of supplement for hay has occurred. Many studies have shown this lack of negative associative effects on intake, or even increased forage intake, indicating a positive associative effect (Arelovich et al., 1983, Guthrie and Wagner, 1988, Fleck et al., 1988, DelCurto et al., 1990a,b, Sunvold et al., 1991 and Olson et al., 1999). Crabtree and Williams, (1971b) found no substitution until dietary concentrate exceeded 50%. Substitution occurred at the 50% dietary concentrate level when the supplement was 11% CP and had no SBM, but not when the supplement was 33% CP and contained equal parts barley and SBM. Effects of supplements are often judged by substitution ratios. However, many variables other than substitution ratios may effect animal performance.

Effects of supplementation on digestibility

Increasing levels of highly digestible supplements would be expected to increase the digestibility of the diet. Many experiments have found starchy supplements to decrease forage and(or) fiber digestion. Mertens and Loften, (1980) suggested that decreased forage digestibility may be due to increased lag time or decreased extent of digestion. However, they were unable to explain *in vivo* reductions in cellulose digestion observed in literature using Mertens (1977) model to adjust either lag time or extent of digestion. This suggests that fibrolytic microbes were limited by competition for N due to increased

starch digestion. When determining the effects of a given level, type or combination of supplement sources, the digestibility of various components of the diet may be influenced differently. Owens et al. (1991) suggested that ruminal digestion was effected by animal type, forage type, feed intake, CP level and source of carbohydrate. On low-quality forages, the primary mechanisms of response may be mostly due to ruminal effects. While few researchers have seen decreased total tract DM and(or) OM digestion, many have noted similar or increased digestibility. Reasons for differences noted previously between the effects of energy supplementation on digestibility may be due to level of supplement fed or level of protein, specifically DIP. Increasing fermentable organic matter intake from grain when DIP is deficient would exacerbate the DIP deficiency, resulting in the commonly observed negative associative effects. This was suggested by McCollum and Horn in 1990, when they indicated that correcting protein deficiencies would increase rate and possibly extent of digestion. In most studies of protein supplementation, rate or extent of digestion, or both, have been increased. Research findings have suggested that smaller amounts of energy supplementation do not decrease forage utilization as drastically as do larger quantities. These findings have also suggested the possibility for improvements in forage digestion by feeding additional DIP along with grain-based supplements.

Total tract apparent supplement digestion. Many researchers (Chase and Hibberd, 1987, Sanson and Clanton, 1989) have used 90% digestibility of corn supplements to calculate forage digestion. This allows the indigestible portion of the supplement to be subtracted from total fecal output to determine forage digestibility. However, if interactions between supplement and forage, or if experimental treatment alters

supplement digestibility, fecal output from forage would be incorrect, thereby causing forage digestibility to be inaccurate. Few researchers have investigated the effects of forage(fiber) or DIP on starch digestion, however, it often is assumed that, when considered total tract, these effects would be minimal. Catlett (1991) found corn processing did effect both ruminal and total tract digestion of starch, while the range of total tract starch digestion was 81 - 96% for whole, cracked, ground or pelleted corn. Chan (1992) fed two levels of corn (1.5 or 3 kg) and found similar starch digestion ruminally, intestinally and total tract. Vanzant et al. (1990) found increasing milo decreased starch digestion slightly, while corn and milo were similar with wheat being slightly higher. These research findings suggest that assuming a constant supplement digestibility may introduce inherent error into calculations of forage digestibility.

Total tract apparent forage digestion. The primary effects of supplementation are on the digestion of the various components of the basal forage. Forage DM or OM make up the greatest portion of a ruminants diet, and the consequences of decreasing utilization of the forage portion of the diet should be carefully considered. Chase and Hibberd (1987) reported total tract OM hay digestibility was decreased by corn. However, most researchers have found no effect of corn on hay digestion (Rittenhouse et al., 1970, Sanson and Clanton, 1989, Sanson et al., 1990, Chan, 1992 and Heldt et al., 1998). Many researchers have reported increased forage DMD as a result of protein or by-product supplementation for cows grazing native range (Rittenhouse et al., 1970, Fleck et al., 1988, Marston and Lusby, 1995 and Hollingsworth-Jenkins et al., 1996).

Total tract apparent fiber digestion. Large decreases in total DM or OM digestion are often the result of decreases in fiber digestion. However, these decreases in fiber

digestion often vary with the type of fiber reported. Stern et al. (1978) suggested that increasing starch decreased ADF or cellulose digestion. Decreased total tract fiber digestibility due to grain or starch supplementation has occurred when DIP is not adequate (Fontenot et al., 1955, Chase and Hibberd, 1987, Sanson et al., 1990, Galloway et al., 1991 and Carey et al., 1993). Galloway et al. (1993b) found corn (.5% BW) or soybean hulls (.7% BW) to increase fiber digestion of steers fed either moderate quality bermudagrass or orchardgrass hay. Livestock consuming low to moderate quality forages, have often had similar or increased fiber digestion depending on amount of supplement fed, supplement source, or the type of fiber that digestion is measured on (Fick et al., 1973, Arelovich, 1983, Guthrie and Wagner, 1988, Fleck et al., 1988, Jones et al., 1988, Sanson and Clanton, 1989, Brake et al., 1989, Martin and Hibberd, 1990, Vanzant et al., 1990, Hannah et al., 1991, Sunvold et al., 1991, Chan, 1992, Galloway et al., 1993a, Forster et al., 1993, Grigsby et al., 1993, Beaty et al., 1994, Köster et al., 1996, Heldt et al., 1998 and Olson et al., 1999).

Total tract apparent diet digestion. Total tract digestion of the diet reflects the amount of the feed consumed that is available for animal use. This indicates that supplement effects on passage rate are of paramount importance. Owens et al. (1991) stated that altered retention time would result in the greatest change in extent of digestion. Köster et al. (1996) found DIP supplementation increased total OMD as well as passage rates, therefore suggesting that rate of ruminal digestion or lower tract digestion were increased.

Very few researchers have found supplementation to decrease total diet OMD. Galloway et al. (1991, 1993b) found molasses, corn (.5% BW), soybean hulls (.7% BW)

or wheat midds to decrease OMD of steers fed moderate quality grass hays. Total diet digestibility has typically been increased by increasing the level of energy supplement fed (Rittenhouse et al., 1970, Fick et al., 1973, Arelovich et al., 1983, Chase and Hibberd, 1987, Guthrie and Wagner, 1988, Fleck et al., 1988, Jones et al., 1988, Brake et al., 1989, Sanson and Clanton, 1989, Martin and Hibberd, 1990, Sanson et al., 1990, Sunvold et al., 1991, Hannah et al., 1991, Chan, 1992, Forster et al., 1993, Galloway et al., 1993a, Beaty et al., 1994, Feng et al., 1995, Heldt et al., 1998 and Olson et al., 1999). Fontenot et al. (1955) found little effect on OMD with added starch at three dietary concentrations of protein. Possibly, this was due to the animals being limit-fed, resulting in similar extent of digestion due to ruminal retention time being altered. Replacing soybean hulls with corn resulted in similar total tract OMD for all treatments and a protein supplemented control and may have been a result of limit-fed animals (Grigsby et al. 1993). Carey et al. (1993) reported similar OMD values for barley and SBM while corn and beet pulp increased OMD. The authors suggested that site of starch fermentation may explain differences. It would be expected that ground barley would be rapidly fermented in the rumen while ground corn could possibly escape to the small intestine. Fiber digestion was lowest for barley and intermediate for corn, suggesting greater ruminal starch digestion of barley diets. The SBM control was fed at a lower level, resulting in the lower total OMD for that diet. Vanzant et al. (1990) reported minimal effects on total diet digestion that varied with grain source and amount fed. Total tract fiber digestion was not altered, however, starch digestion decreased as milo increased, and varied with grain source, possibly explaining decreases in total diet OMD. Several reported trials in which various levels of grain or starch were fed with various levels of protein all found

that feeding the lowest level of protein with the highest level of starch was the only treatment combination that significantly depressed fiber, forage or total diet digestibility (Rittenhouse et al., 1970, Crabtree and Williams, 1971b, Fick et al., 1973, DelCurto et al., 1990a and Olson et al., 1999). This may help explain the large decreases found by Chase and Hibberd, (1987) and Sanson and Clanton (1989) who fed levels of corn without added protein and reported depressions in forage digestion.

In situ disappearance. Disappearance of forage from *in situ* bags has often been used to model the effects of a dietary treatment on ruminal forage digestion. Caton and Dhuyvetter (1997) and Mertens and Loften (1980) suggested that rate of digestion has not been affected by supplementation. However, Chase and Hibberd (1987) noted no effect of increasing corn on extent (96 h) of ISDMD while rate of NDF and hay DM disappearance decreased. While some researchers (Pordomingo et al., 1991, Chan, 1992, Freeman et al., 1993 and Grigsby et al., 1993) have shown no effect of increasing energy or protein on rates of *in situ* disappearance, others (Arelovich, 1983, Sanson and Clanton, 1989, Chan, 1992, Barton et al., 1992 and Freeman et al., 1993) have shown no effect on extent of *in situ* disappearance due to energy or protein supplementation. Carey et al. (1993) found no effect of energy or protein supplements on rate of degradation while beet pulp increased extent of *in situ* disappearance. Feng et al. (1995) found greater ISDMD for barley than corn supplements while Barton et al. (1992) and Heldt et al. (1998) reported greater rate of *in situ* disappearance from oilseed meals and(or) corn-SBM supplements. Carey et al. (1993) and Freeman et al. (1993) reported supplementation altered disappearance of *in situ* N and rate of forage CP degradation when grain was supplemented. However, extent of ISDMD may not always be indicative of true ruminal

effects since fixed incubation times are not reflective of changes in rate of passage due to supplementation.

Effects of supplementation on energy intake and status

A primary goal of supplementation strategies has been to increase energy intake and consequently improve livestock performance. Crabtree and Williams (1971a) reported increased DE intake for sheep consuming straw or low-quality grass hay with increasing supplemental concentrate up to 67% of diet DM. Chase and Hibberd (1987), Marston and Lusby (1995) and Olson et al. (1999) reported similar energy intakes as level of energy supplement increased while Pordomingo et al. (1991) found a peak at .2% BW corn and decreases above that level. However, many workers (Arelovich et al., 1983, Guthrie and Wagner, 1988, Fleck et al., 1988, Sanson and Clanton, 1989, Martin and Hibberd, 1990, Sanson et al., 1990, Chan, 1992, Beaty et al., 1994, Matejovsky and Sanson, 1995, Köster et al., 1996, Garcés-Yépez et al., 1997 and Olson et al., 1999) have reported increased DOM intake as a result of supplementation. Consequently, energy intake would also be expected to increase as protein or energy supplementation increased.

Effects of supplementation on efficiency of energy use

While changes in calculated DE or ME, or observed DOM, are important as a predictor of energy intake and often occur as a result of supplementation, inadequate attention has been paid to possible changes in efficiency of forage or total diet ME use due to supplementation. Caton and Dhuyvetter (1997) stated that energy increases from supplement should increase the efficiency of metabolizable energy (ME) use, since concentrate ME is used more efficiently for maintenance and(or) gain than forage ME (NRC, 1984). McCollum and Horn (1990) also indicated that animal performance could

be increased if supplementation increased ME efficiency. Little research exists on the influence of supplementation on efficiency of forage and(or) supplement ME use. Lake et al. (1974) and Garcés-Yépez et al. (1997) reported that corn supplementation decreased BUN and Lake et al. (1974) showed decreased urinary N excretion in steers, indicating greater capture of ammonia in the rumen, and possibly increased microbial flow to the duodenum. Fick et al. (1973) found improved N retention and increased BUN with increasing dietary N. Fontenot et al. (1955) found added starch resulted in varied effects on N retention, but consistently increased the biological value of N, decreased urinary N output and increased fecal N output. McCollum and Horn (1990) indicated that protein deficiencies would decrease ME efficiency. They also suggested increasing ruminal N, small intestinal NAN, specific or total amino acids, greater recycled N or supply of glucogenic precursors could improve ME efficiency. Since acetate is the primary product of ruminal forage fermentation, ruminant metabolism requires glucogenic substrates. If supplementation increases ruminal fermentation, ruminants will also have a greater need for propionate and glucogenic amino acids to supply metabolic intermediates. Supplementation of ionophores, natural protein sources, or non-structural carbohydrate sources should aid in supplying needed glucogenic precursors.

Effects of various protein to energy ratios

Calculations for DIP requirements necessitate knowledge of intake, digestibility and nutrient concentrations. As these factors change, DIP needs will vary (Cochran et al., 1998). Sanson et al. (1990) suggested that there was no interaction between protein and energy, and meeting the animals protein requirement would not prevent negative associative effects. However, they also stated that the observed pH and VFA values in

their trial were indicative of greater amounts of carbohydrate fermentation in the rumen, which would emphasize the need for additional DIP due to increased fermentable OM. Owens et al. (1991) suggested an average microbial protein production of approximately 160 g per kg of OM fermented in the rumen indicating a need for greater DIP as fermentable OM increased. Poppi and McLennan, (1995) stated that dietary CP would be transferred to the intestines when ratios were below 160 g CP/kg OM, while N loss from the GI tract would occur when the ratio exceeded 210 g CP/kg OM. They related this to a ratio of 9.3-13.3 g CP/MJ of ME. This would be due to decreased microbial fermentation when protein was insufficient for fermentable OM and from NH_3 loss when N was excessive for microbial incorporation. Stern et al. (1978) suggested that losses of NH_3 were reduced by the addition of starch. McCollum and Horn (1990) suggested that energy can be used as a supplement if dietary nitrogen was high. Microbial yield of crude protein can change as intake changes due to increased growth rate of microbes resulting in increased net efficiency of microbial protein synthesis. Various researchers and feeding systems have reported widely varying values that may be a result of differing concentrations of ruminally fermentable OM in the diets used to calculate these values. The NRC (1996) suggests 13% efficiency of microbial synthesis of TDN into bacterial crude protein and reported a value as low as 8% for 50% TDN diets with intakes of about 2% BW. Past research with TDN ranges from 50-65% averaged 7.82% and had a range from 5 to 11.4% (NRC, 1996). In 1985, the NRC suggested 12.8% while the AFRC (1992) suggested 13% and summarized a variety of international systems whose estimates ranged from 9.5-17%. Burroughs et al. (1974 and 1975) suggested 13.05 and 10.4%, Karges et al. (1990 and 1992) suggested 9.97 and 12.3%, Russell et al. (1992)

suggested 11% for 50% TDN diets, Hollingsworth-Jenkins (1996) determined a value of 7.1% and Köster et al. (1996) suggested either 11.1 or 15.8%, depending on the mathematical method used to calculate DIP requirements. Burroughs et al. (1975) suggested 80% of MCP was digested in the small intestine (a value currently used by the NRC, 1996) resulting in one kilogram of roughage or corn producing 52 or 93 g of microbial CP and net absorption of 30 or 62 g of protein in the small intestine. Burroughs et al. (1975) stated that either grain (high TDN, low CP) or fibrous feeds (low TDN, low CP) would be most benefited by additional DIP and suggested a CP:TDN ratio of 13-14.5%. One possible explanation for the range of estimates may be differences in fermentable OM concentration in TDN. Russell et al. (1992) suggested that using TDN may result in inaccurate estimates of DIP requirements due to inherent differences in the amount of fermentable OM in the TDN portion of different feeds. Stern et al. (1978) also suggested that TDN may not be the best method for determining energy supply for microbial synthesis. Cochran et al. (1998) used DOM, the combined effect of OMI and OMD coming from both supplement and forage, to calculate DIP requirements and found a positive relationship. Using differing proportions of corn and SBM, Cochran, et al. (1998) theoretically formulated supplements ranging from 10 to 40% CP and showed a 40% difference in DIP supply. These supplements had DIP:TDN ratios ranging from 5 to 30 as CP increased from 10 to 40%. This difference would be expected to create large differences in fermentation profiles and consequently, intake, digestion and performance, especially when large quantities of fermentable energy are supplemented. Supplemental DIP has to be sufficient to adequately ferment supplemental energy and have DIP remaining to aid in forage fermentation. Cochran et al. (1998) estimated that level to be

around 20% CP when grain and oil-seed meal-based supplements are fed. When effects of supplemental starch (at the .15 and .30% BW levels) were compared across treatments with equal supplement DIP:TDN ratios, Olson et al. (1999) found no difference or increased forage and total OM intake, OM digestibility and DOM intake. Research by El-Shazly et al. (1961) suggested that large numbers of amylolytic microorganisms (AM) that are faster growing than cellulolytic microorganisms (CM) may outcompete the CM for essential nutrients. Russell et al. (1992) suggested that current thinking did not adequately account for competition between microbial types. In 1961, El-Shazly and coworkers added purified starch *in vitro* and found maximum cellulose digestion when urea, as a N source, was included at levels that approximated DIP:DOM ratios ranging from 16 - 22%. These results agree with the *in vivo* data generated by El-Shazly et al. (1961) in sheep fed diets with up to 1:2 forage to concentrate ratios and up to 52 g of urea. Matejovsky and Sanson (1995) suggested that protein supplements increased intake of low-quality hay when DIP was inadequate. Poppi and McLennan (1995) suggested that supplying additional energy to the rumen could increase duodenal flow of NAN. However, Owens et al. (1991) stated that the ratio method ignores feed intake, physiological status and level of production. Consequently, it may not predict protein supply adequately.

Effects of supplementation on the ruminal environment

Ruminal pH. Many researchers have looked at pH as being responsible for many of the negative effects of starch supplementation. However, the lack of consistency in findings involving pH and digestion appear to implicate reduced pH as the primary causative agent in decreasing fiber digestion in only the most extreme cases. Horn and

McCollum (1987) concluded that maintaining ruminal pH would only partially alleviate negative associative effects and would vary with forage type and concentrate level. They also suggested that ruminal pH effects would vary with forage buffering capacity which may be linked to form, type, fragmentation, (chewing, ruminating and salivary input) as well as the inherent buffering capacity of the forage (Horn and McCollum, 1987). In a review of energy supplementation, Caton and Dhuyvetter (1997) indicated ruminal pH could not explain all negative effects of energy supplementation on intake and digestibility and that additional mechanisms do exist. El-Shazly et al. (1961) found inhibition of cellulose digestion even when pH was maintained by continuous culture. Stern et al. (1978) found decreased *in vitro* ADF and cellulose digestion with no changes in pH, indicating that depressed fiber digestion was not caused by pH reductions. Ruminal pH values have often been decreased by energy supplements or as level of supplementation increased (Chase and Hibberd, 1987, Brake et al., 1989, Martin and Hibberd, 1990, Vanzant et al., 1990, DelCurto et al., 1990a,b,c, Sunvold et al., 1991, Chan, 1992, Grigsby et al., 1993, Heldt et al., 1998 and Olson et al., 1999). However, none of these researchers found average ruminal pH measurements below 6.0 at any time point measured. Protein supplements have decreased pH as supplemental protein increased in many studies (Guthrie and Wagner, 1988, Köster et al., 1996, Heldt et al., 1998 and Olson et al., 1999). A variety of researchers (Arelovich, 1983, McCollum and Galyean, 1985, Guthrie and Wagner, 1988, Fleck et al., 1988, Jones et al., 1988, Sanson and Clanton, 1989, Barton et al., 1992, Forster et al., 1993 and Galloway et al., 1993a,b) have not found an effect on ruminal pH as energy or protein supplements increased. This appears to further indicate that ruminal pH may not play a primary role in decreasing

digestion. Carey et al. (1993) reported barley and beet pulp had the lowest pH response curve with values falling below 6.0 while corn or soybean meal did not have pH response curves below 6.1. This did not explain the decreased fiber digestion for energy supplemented diets. The barley and soybean meal diets had similar total diet OMD while corn and beet pulp had greater total diet OMD. Sanson et al. (1990) found high levels (3 kg) of corn supplementation depressed rumen pH below 6.0 for 8 hours without decreasing hay digestion. Feng et al. (1995) reported low pH (> 6.0) for steers fed corn or two levels of barley (4.3 and 5.3 kg/hd/d; .73 and .90% BW). However, total diet OMD was greater as supplement increased. These could be instances when ruminal pH may be responsible for a portion of the decrease in fiber digestion. Hoover (1986) suggested the presence of several effects of feeds on ruminal pH and depressions of forage utilization. Slight depressions (to 6.0) or short-term depressions between 5.8 and 6.2 may only mildly decrease fiber digestion while depressions below 6.0 will greatly reduce digestion and possibly result in complete inhibition of fiber degradation. This may be due to a "carbohydrate" effect, increased washout, or lag time of attachment. Possibly, pH is an effect that can be measured and correlated to digestion, but not the primary agent in decreasing fiber digestion by the ruminal microflora in many instances.

VFA concentrations and profiles. Much attention has been paid to the concentrations of VFA in ruminal fluid. This is due to the fact that they are the primary source of energy for ruminants and in some cases can be the entire dietary energy source. However, since we are not actually measuring the production of VFA, only the concentration at one point in time, we can only attempt to draw inferences between treatments. Often, total VFA concentration has not been affected by supplementation while differences due to

treatment were found for individual VFA profiles (McCollum and Galyean, 1985, Chase and Hibberd, 1987, Fleck et al., 1988, Sanson and Clanton, 1989, Vanzant et al., 1990, Pordomingo et al., 1991, Grigsby et al., 1993, Freeman et al., 1993, Galloway et al., 1993a,b and Olson et al., 1999). In contrast, supplementation has increased total VFA levels in many reported studies and had varied effects on individual VFA profiles, which is in agreement with studies without an effect on total VFA concentration (Martin and Hibberd, 1990, Sunvold et al., 1991, Hannah et al., 1991, Barton et al., 1992, Carey et al., 1993, Köster et al., 1996 and Olson et al., 1999).

Ruminal ammonia concentrations. In a review of ruminant starch utilization, Huntington (1997) suggested that the relatively rapid rate of NH_3 absorption would limit the utilization of NH_3 in the absence of ruminally fermentable energy. Starch fermentation increases microbial capture of NH_3 , amino acids and peptides, increasing ruminal outflow of microbial protein (Spicer et al., 1986) and consequently increasing the requirements for NH_3 , amino acids and peptides. However, on low CP forages (< 6-8% CP), responses to protein supplements are often believed to be from the correction of a ruminal NH_3 deficiency (McCollum and Horn, 1990), as it is a vital nutrient for ruminal cellulolytic microorganisms. Fibrolytic bacteria utilize only NH_3 as a source of N for microbial CP synthesis, while amylolytic species can use peptides, amino acids or NH_3 (Russell et al. 1992). However, interpretation of ruminal NH_3 levels is difficult since low values actually indicate an imbalance of DIP and fermentable OM rather than describing the causative agent. Horn and McCollum (1987) also suggested that ruminal ammonia concentrations are more indicative of the balance between ruminally-degraded protein and energy. Russell et al. (1992) stated that carbohydrate availability can determine the

fate of peptides between microbial incorporation by amylolytic bacteria or deamination and conversion to NH_3 . This balance may become increasingly complicated as differences in physiological status, site of digestion and metabolic modifiers are considered. These can act as nitrogen sinks, decreasing systemic or circulating nitrogen levels and increasing DIP requirements via reduced recycling of urea to ruminal or large intestinal NH_3 (McCullum and Horn, 1990, Owens et al., 1991).

Horn and McCullum (1987) suggested that the addition of readily fermentable carbohydrates (grain) could result in deficiencies of ruminal NH_3 . Addition of grain supplements to low-quality forage diets would be expected to depress ruminal NH_3 and may prevent adequate fiber fermentation. However, nitrogenous compounds other than NH_3 , such as protein, peptides, or individual amino acids, may also be involved (Horn and McCullum, 1987). Owens et al. (1991) suggested that ruminal ammonia is a useful indicator of N available for ruminal fermentation while still being a function of supply (DIP) and demand (microbial CP synthesis). Russell et al. (1992) suggested that non-structural carbohydrate fermenting bacteria reduce ruminal NH_3 levels during rapid growth. Owens et al. (1991) suggested that ionophores can alter NH_3 by increasing ruminal concentrations of peptides and AA's. This may be important since amylolytic bacteria prefer NAN for growth and some cellulolytic microbes require branched-chain VFA from fermentation of branched-chain amino acids for growth. Also, Owens et al. (1991) mentioned that microbial growth rate is limited by cell turnover (outflow and death), while total microbial numbers are limited by nutrient supply. With low-quality forages, NH_3 is the first limiting nutrient, while energy is first limiting on most other diets. However, green forages, silages and high-quality hays may have increased DIP

requirements due to reduced salivary flow, resulting in decreased urea recycling compared to dry, low-quality forages. This reduction in salivary flow may be due to stem to leaf ratios, decreased chewing, rumination or an osmotic effect (McLeod et al., 1990). Owens et al. (1991) also suggested the greatest benefit of DIP for fiber digestion was from forage protein since it is located near the site of cellulolytic microbial incorporation.

Satter and Slyter (1974) found a range of between 2 and 5 mg/dl to limit microbial protein production in continuous culture with no increase in microbial CP as NH_3 was increased 16-fold. It would appear that above 5 mg/dl, ammonia was no longer first limiting for microbial growth. Possibly, branched-chain amino acids or energy limited microbial growth, not NH_3 concentration. They also indicated that maximum microbial production or efficiency might not be synonymous with maximum fiber digestion. Satter and Slyter (1974) also reported that ruminally digested OM controlled ammonia accumulation, pointing to the balance between ruminally degradable nitrogen and energy. El-Shazly et al. (1961) reported increasing urea increased *in vitro* and *in vivo* cellulose digestibility when forage to concentrate ratios were greater than 1:1. In addition, they reported that the addition of autoclaved ruminal fluid supernatant also increased cellulose digestibility. This may well have been an effect of NH_3 , amino acids, or buffering capacity of the rumen fluid. Martin and Hibberd (1990), suggested the low NH_3 values observed in their trial were due to increased OM fermentation as SBH increased, resulting in greater incorporation of NH_3N by microbes rather than a NH_3 deficiency. Sanson and Clanton (1989) suggested that the low levels of NH_3 in their steers could have depressed the digestibility of all diets. Matejovsky and Sanson (1995) suggested that increases in DMD of low-quality forages supplemented with DIP were due to correcting a NH_3

deficiency in the rumen. Hoover (1986) suggested that competition for ruminal NH_3 between fibrolytic and amylolytic microorganisms could also play a part in reductions of fiber degradation when readily fermentable carbohydrates were fed with forage.

Results of research involving ruminal concentrations of NH_3N have shown varied effects due to supplementation. Some researchers (Chase and Hibberd, 1987 and Olson et al., 1999) have noted that ruminal NH_3N levels decreased as starch increased while Carey et al. (1993) found beet pulp, corn or barley to decrease NH_3 vs a soybean meal treatment. Other workers (Jones et al., 1988, Vanzant et al., 1990, Galloway et al., 1993b and Forster et al., 1993) found no effect on ruminal NH_3 levels due to supplementation. The greatest number of reports have indicated that supplementation has increased NH_3N over an unsupplemented control, with greater increases in NH_3 coming from increased supplemental CP, which is reflective of increased supplemental DIP (Arelovich, 1983, McCollum and Galyean, 1985, Guthrie and Wagner, 1988, Fleck et al., 1988, Sanson and Clanton, 1989, Sanson et al., 1990, Martin and Hibberd, 1990, Hannah et al., 1991, Sunvold et al., 1991, Chan, 1992, Barton et al., 1992, Galloway et al., 1993a, Grigsby et al., 1993, Freeman et al., 1993, Köster et al., 1996 and Olson et al., 1999). Brake et al. (1989) and Feng et al. (1995) found greater NH_3 levels from barley than corn supplements.

Fractional dilution rates. Many investigations into the interactions between intake and digestion have pointed to the importance of rate of passage from the rumen, as well as total tract, having a major impact on digestion, both rate and extent, and intake. Chase and Hibberd (1987) caused a slight linear decline in particulate passage rate (K_{pp}) (3.9, 4.04, 3.72 and 3.68%/h) due to corn supplementation that may have been related to

decreased DIP, especially in relation to DOM. McCollum and Horn (1990) reported that many studies have shown protein supplementation to increase rate of passage. Passage rates for particulate matter have been reported to be increased by either energy or protein supplementation by a wide variety of researchers (McCollum and Galyean, 1985, Guthrie and Wagner, 1988, Brake et al., 1989, Martin and Hibberd, 1990, Sunvold et al., 1991, Hannah et al., 1991, Chan, 1992, Beaty et al., 1994 and Olson et al., 1999) as have fluid passage rates (McCollum and Galyean, 1985, Brake et al., 1989, Hannah et al., 1991, Chan, 1992, Freeman et al., 1993, Galloway et al., 1993b, Beaty et al., 1994, Köster et al., 1996 and Olson et al., 1999). However, other workers have found no effect of supplementation on particulate passage rates (Fleck et al., 1988, Vanzant et al., 1990, Pordomingo et al., 1991, Freeman et al., 1993, Carey et al., 1993, Galloway et al., 1993a and Barton et al., 1992) or on fluid passage rates (Fleck et al., 1988, Vanzant et al., 1990, Sunvold et al., 1991, Pordomingo et al., 1991, Carey et al., 1993 and Galloway et al., 1993a). Jones et al. (1988) and Grigsby et al. (1993) reported no effect on fluid or particulate passage, but this was expected since these cattle were limit-fed. Feng et al. (1995) found no differences between corn or barley supplements on particulate passage rate.

Ruminal capacity. Varying results of supplementation and the restrictions of particle size for ruminal outflow have led some to suggest that ruminal fill controls intake, both for low-quality high-roughage diets and for animals at a high rate of production. Owens et al. (1991) suggested that some factor other than ruminal fill must limit intake since rumen fill was not constant. McCollum and Horn (1990) indicated that ruminal fill is often increased by protein supplementation. DelCurto et al. (1990a,b,c) and Hannah et al.

(1991) found increases in fill from milo-soybean meal supplements with increasing CP concentrations, dehydrated alfalfa pellets or alfalfa hay, while Sunvold et al. (1991) found increased ruminal fill that was greatest for high-levels of wheat middlings. Vanzant et al. (1990) reported no change in fill due to amount of supplemental milo. Olson et al. (1999) found little change in ruminal fill across four levels of DIP and three levels of starch that were infused into the rumen, or an unsupplemented control. Since intake and passage increased and ruminal fill was similar, ruminal digestion rates may not have increased, or possibly infusion may have altered expected responses. Köster et al. (1996) found decreases in ruminal fill as DIP increased. Carey et al. (1993) reported that compared to SBM supplementation, ruminal fill was decreased by beet pulp and barley, but not corn supplementation while Chan (1992) found NDF fill decreased for corn and increased for soy-hull supplements. McCollum and Galyean (1985) and Barton et al. (1992) found no difference between unsupplemented and protein supplemented cattle in estimated total gastrointestinal tract fill.

Ruminal fermentation. Ruminant animals are alone in their ability to maintain a population of microbes in the forestomach that have the enzymatic capacity to degrade the β , 1-4 linkages of cellulose; a feature that non-ruminants lack to a great extent. However, this creates unique challenges in the study of ruminant nutrition, since the microbial population has the first opportunity to degrade feedstuffs. These bacteria also have certain requirements for nutrients that must be met in order for optimum ruminal function to occur. The NRC (1996) uses the MP system to express animal requirements for absorbed protein in the small intestine. The MP system was incorporated since all

feed proteins do not have equal ruminal degradation (NRC, 1996). Absorbed protein can be supplied from microbial protein (protein degraded in the rumen) or intact feed protein reaching the small intestine. Microbial protein reaching the small intestine is calculated from bacterial CP synthesis based on carbohydrate digestion in the rumen and can supply anywhere from half to all of the MP requirements depending on UIP and level of production (NRC, 1996). Release of NH_3 , peptides and amino acids, as well as the type of carbohydrate, dilution rate and pH can all influence efficiency of bacterial CP synthesis (NRC, 1996). Russell et al. (1992) suggested that efficiency of microbial protein can be reduced by imbalances between N and fermentable energy. DIP requirements are considered to be equal to microbial synthesis of CP, as NH_3 loss from the rumen is assumed to be equal to urea recycling back into the rumen (NRC, 1996, Burroughs et al., 1975). It has been suggested that increased utilization of DIP would occur if degradation rates were similar for both protein and energy (NRC, 1996). The relationship between ruminal degradation of protein and energy is opposite for forages and grains. The TDN of forage is slowly degraded, while DIP is released quickly and vice versa for grains (NRC, 1996). However, little advantage in production has resulted from synchrony, due to recycling and intake occurring more than once per day. The NRC (1996) suggests that MP deficiencies are difficult to produce as cattle get heavier, possibly due to greater duodenal flow from increased intake on an absolute basis.

Effects of supplementation on post-ruminal nutrient flow

An increased understanding of the mechanisms by which supplementation affects animal performance would allow nutritionists to supply nutrients to target specific mechanisms (McCullum and Horn, 1990). These mechanisms include ruminal nitrogen

deficiencies, small intestinal non-ammonia nitrogen stimulus, specific amino acid deficiencies, increased total amino acid supply (for greater deposition), increased supply of glucogenic precursors, or greater recycled nitrogen. All of these factors may stimulate forage intake, ME efficiency, or both. Enhanced intestinal flow of nitrogen and other nutrients may be of greater importance in regulating intake response and consequent animal performance than rate or extent of digestion. However, these are difficult to separate since increases in post-ruminal nitrogen flow often either cause or are the result of increased digestion (Egan and Moir, 1965). While several researchers have shown a response from supplementing individual amino acids, practical application may be limited due to rapid degradation of these amino acids in the rumen (McCollum and Horn, 1990, Owens et al., 1991). Possibly, determining a more energetically efficient amino acid profile for microbial use would allow lower levels of CP to be fed, resulting in an increase in efficiency and production. While total supply of amino acids to the small intestine may be more important than quality, it is possible that increased flow of amino acids to the small intestine is simply alleviating a single amino acid deficiency. Studies have shown an increase in intake when ruminal factors (passage rate, extent of digestion or fill) have not changed, indicating the presence of a non-ruminal effect. This may stem from an increased supply of protein to the small intestine from increased microbial output or additional feed protein that has escaped ruminal degradation. However, amino acid flow to the duodenum alone does not explain increased feed and water intake, decreased ruminal capacity with no change in passage rates or digestibility when post-ruminal infusions of urea-glucose or casein were compared (Garza et al., 1991). Owens et al. (1991) suggested that TDN has a greater impact on duodenal CP flow than bypass

protein. There are very few instances (young rapidly growing cattle, high levels of lactation) where cattle could benefit from bypass protein supplementation (Owens et al., 1991). Russell et al. (1992) stated that bacterial yield and consequent duodenal flow are dependent on fermentable carbohydrate supply. Owens et al. (1991) also indicated that supply of amino acid or glucogenic substrates to the small intestine is an important controller of intake. In addition to altering efficiency of nutrient utilization, increasing post-ruminal supply of nutrients also can alter the hormonal (insulin, glucagon and somatostatin) status or profile (IGF-I and GH) of animals (Houseknecht et al., 1988). Huntington (1997) indicated that capacity for glucose transport can double in two to four days in the small intestine as cattle adapt to starch. Increased energy intake or duodenal protein flow results in increased pancreatic enzyme secretion, leading to an increase in starch digestibility in the duodenum (Huntington, 1997). Increasing ruminal fermentation could effect digestibility in the small intestine (Huntington, 1997) due to increased energy supply or duodenal protein flow from microbes and UTP supplied by the feed. Karges et al. (1992) found greater microbial protein production from energy supplements (cornstarch/molasses) than from degradable protein supplements (corn steep liquor). Krysl et al. (1989) reported no effect on duodenal flow of microbial or bypass protein or on microbial efficiency for low-level energy supplementation of blue grama range. Grigsby et al. (1993) showed total N flow to the small intestine was greatest when three pounds of corn was fed, was greater for all energy supplements than a hay/protein control and accounted for 72-88% of total small intestinal N flow for hay diets fed with energy supplements.

Alternative factors

Owens et al. (1991) suggested the possibility that fecal output limited intake. Garza et al. (1991) also suggested that constant fecal output indicated that ruminal fill was not the major limitation of intake. Mieres (1992) found slight increases in fecal output with increasing corn. Elizalde et al. (1999) found a linear decrease in fecal output with increasing corn supplementation and lower fecal output for steers fed a restricted intake of unsupplemented fresh alfalfa than supplemented animals. Although the large range of fecal output values and variation between species and forage types indicates less promise for this theory, within animal and forage type, this theory does show promise. Owens et al. (1991) found dietary ADF to be a more useful predictor of DMI than NDF and suggested that ADF excretion could be playing a role in the regulation of intake. Owens et al. (1991) suggested that intake and fecal output increased as supply of N to the large intestine and cecum increased. They also suggested that since BUN, blood osmotic pressure and salivary flow are linked, intake could be influenced by BUN indirectly increasing salivary flow via osmotic pressure (Owens et al., 1991). Protein supplementation of low-quality forages has typically increased NH_3 , resulting in greater fermentation. Greater fermentation causes increased total VFA concentrations, which increase digesta osmotic pressure, resulting in an influx of water and saliva into the GI tract. Since low-quality forage diets typically have dry digesta, increases in salivary flow would be expected to increase fluid passage rates, resulting in greater particulate passage. Increased passage allows greater intake. Jones et al. (1988) and Brake et al. (1989) found lower fecal pH for grain supplementation. Fecal pH could be assumed to be indicative of

greater cecal fermentation, possibly the result of changes in site of digestion and(or) greater urea recycling.

Effects of supplementation on livestock production

Performance of livestock is measured in many ways. Typically, supplementation has been practiced in order to maintain or improve animal performance. Energy may be the primary limitation on animal performance in many grazing situations. However, protein intake seems to exert primary control over energy intake and utilization (McCullum and Horn, 1990). Caton and Dhuyvetter (1997) indicated animal production would typically be unaffected or improved by energy supplementation. This is critically important since performance is directly related to intake and protein is one of the factors controlling intake (Owens et al., 1991). This suggests a much greater positive impact on performance from protein supplementation than energy supplements. Animals in negative energy or protein balance will increase catabolism of tissue protein and increase circulating levels of N, thereby increasing salivary concentration of N and increasing recycling to the rumen (Owens et al., 1991). This alters DIP requirements and can be effected by dietary nutrient concentrations (fat, ionophore, protein, grain) that alter metabolic status of the animal or other growth stimulants (implants) by modifying tissue protein catabolism. Poppi and McLennan, (1995) suggested that additional protein can increase live weight gain of cattle grazing low-quality forages. Sanson and Clanton (1989) found no effect of corn supplementation on cow weight gain, BCS or conception rate. This may have been due to animal requirements being met by feeding hay alone. Hollingsworth-Jenkins et al. (1996) found low-levels of added DIP did not maintain gestating cow BCS during late winter that may have been a result of supplement type.

Greater maintenance of weight and BCS has been found in either gestating or lactating cows when energy was fed with protein and little or no effect on subsequent reproductive performance, calf ADG, or weaning weight (Lusby and Wettemann, 1988, Sanson et al., 1990, Beaty et al., 1994 and Heldt et al., 1998). Increased ADG in grazing steers (Karges et al., 1992, Forster et al., 1993, Galloway et al., 1993a, Owensby et al., 1995 and Garcés-Yépez et al., 1997) or hay-fed lightweight calves (Purvis et al., 1996) has been reported as a result of energy supplementation.

Conclusions

Many of the researchers who have found varying effects of supplementation have attempted to offer reasonable explanations for their results. Some of these reasons have included level of feeding, basal forage type, the results of supplements on ruminal fermentation, animal requirements and(or) physiological status, or the effects of the supplement of flow of nutrients through the GI tract.

Chase and Hibberd (1987) increased corn and decreased cottonseed meal, which reduced supplemental DIP from 153 to 84 g/hd/d while increasing supplemental TDN from 458 to 2362 g/hd/d. They stated that formulating grain-based supplements for CP requirements may decrease forage utilization when fed at high levels (2-3 kg/hd daily) and may actually decrease energy intake and was a direct result of grain-based supplements formulated with DIP deficiencies. The deficiency of ruminal NH_3N decreased the rate of ruminal digestion and may have decreased microbial growth, resulting in reduced hay digestion. This could be corrected by balancing for DIP to increase NH_3N to overcome negative associative effects on intake, digestibility and DOM. Chase and Hibberd (1987) suggested that feeding ruminally degradable protein

with grain would overcome ruminal deficiencies of NH_3N and increase forage utilization. Martin and Hibberd (1990) found soybean hulls to be an effective supplement for low-quality hay that did not appear to decrease cellulolytic activity and increased energy supply of the cattle. These two studies had large differences in the concentration, as well as the absolute amount of DIP fed daily. Guthrie and Wagner (1988) found positive associative effects of supplementation from protein supplements and no negative associative effects on hay DMI or DMD due to energy supplementation from grain when greater supplemental CP (and DIP) was fed. They suggested the importance of protein to DE ratios. Olson et al. (1999) reported improved DOM as DIP increased regardless of the level of starch inclusion, pointing to the importance of DIP in supplementation of low-quality forages, particularly when increasing fermentable organic matter. Carey et al. (1993) suggested that energy supplementation can effect rate of forage protein fraction digestion possibly through changes in forage digestion. Poppi and McLennan (1995) suggested that differences in form or type (starch, sugars or fiber) of energy supply would alter animal performance response due to both the supply of additional energy and from the increase in protein flow to the duodenum. They also suggested that differences within energy supplements (starch, sugar, fiber) could alter SI flow of protein based on the efficiency with which microbes use these energy sources to convert ruminally degraded protein to microbial protein (Poppi and McLennan, 1995).

In conclusion, while many unknowns remain about how supplementation effects animal response, it seems clear that several major issues should be consistently addressed. First, DIP and the energy needs of the microbes must be met. Next, any opportunity to measure ruminal fill, passage rates and fecal output must be taken advantage of. Third,

some measure of the physiological effect must be made. Simply reporting fecal N or pH may allow us to infer effects on large intestinal fermentation. Lastly, when practical and possible, duodenal flows and protein concentration should be determined. While these factors may not explain all the effects of supplementation, it may allow us to gain a greater understanding of the entire system and how individual mechanisms function.

Literature Cited

- Adams, D. C. 1985. Effect of time of supplementation on performance, forage intake and grazing behavior of yearling beef steers grazing Russian wild ryegrass in the fall. *J. Anim. Sci.* 61:1037-1042.
- Agricultural and Food Research Council. 1992. Technical committee on responses to nutrients, report no. 9. Nutritive requirements of ruminant animals: Protein. *Nutrition Abstracts and Reviews, Series B: Livestock Feeds and Feeding.* 62:787-835.
- Allen, M. S. 1996. Physical constraints on voluntary intake of forages by ruminants. *J. Anim. Sci.* 74:3063-3075.
- Arelovich, H. M. 1983. Effect of protein and starch supplementation on the utilization of low-quality roughages. M. S. Thesis. Oklahoma State University.
- Arelovich, H. M. D. G. Wagner and C. A. Hibberd. 1983. Effect of supplementation program on intake and utilization of low-quality prairie hay by steers. *Oklahoma Agric. Exp. Sta. MP-114.* pp. 72-75. Stillwater.
- Barton, R. K. L. J. Krysl, M. B. Judkins, D. W. Holcombe, J. T. Broesder, S. A. Gunter, and S. W. Beam. 1992. Time of daily supplementation for steers grazing dormant intermediate wheatgrass pasture. *J. Anim. Sci.* 70:547-558.
- Beaty, J. L. R. C. Cochran, B. A. Lintzenich, E. S. Vanzant, J. L. Morrill, R. T. Brandt, Jr. and D. E. Johnson. 1994. Effect of frequency of supplementation and protein concentration in supplements on performance and digestion characteristics of beef cattle consuming low-quality forages. *J. Anim. Sci.* 72:2475-2486.
- Brake, A. C. A. L. Goetsch, L. A. Forster, Jr. and K. M. Landis. 1989. Feed intake, digestion and digesta characteristics of cattle fed bermudagrass or orchardgrass alone or with ground barley or corn. *J. Anim. Sci.* 67:3425-3436.
- Burroughs, W. K. A. H. Trenkle, and R. L. Vetter. 1974. A system of protein evaluation for cattle and sheep involving metabolizable protein (amino acids) and urea fermentation potential of feedstuffs. *Vet. Med. Small Anim. Clin.* 69:7113.
- Burroughs, W. K. D. K. Nelson and D. R. Mertens. 1975. Cow: The metabolizable protein feeding standard. *J. Anim. Sci.* 41:933-944.
- Carey, D. A. J. S. Caton and M. Biondini. 1993. Influence of energy source on forage intake, digestibility *in situ* forage degradation and ruminal fermentation in beef steers fed medium quality brome hay. *J. Anim. Sci.* 71:2260-2269.

- Catlett, G. E. 1991. Effect of supplemental corn processing on utilization and intake of low quality native grass hay and performance of beef cows. M. S. Thesis. Okla. State Univ. Stillwater.
- Caton, J. S. and D. V. Dhuyvetter. 1997. Influence of energy supplementation on grazing ruminants: Requirements and responses. *J. Anim. Sci.* 75:533-542.
- Chan, W. W. 1992. Corn versus soybean hull supplements for beef cows fed low-quality native grass hay. M. S. Thesis. Okla. State Univ. Stillwater.
- Chase, C. C. Jr. and C. A. Hibberd. 1987. Utilization of low-quality native grass hay by beef cows fed increasing quantities of corn grain. *J. Anim. Sci.* 65:557-566.
- Cochran, R. C. H. H. Köster, K. C. Olson, J. S. Heldt, C. P. Mathis and B. C. Woods. 1998. Supplemental protein sources for grazing beef cattle. Proc. 9th Annual Florida Ruminant Nutrition Symposium. pp. 123-136.
- Crabtree, J. R. and G. L. Williams. 1971a. The voluntary intake and utilization of roughage-concentrate diets by sheep. 1. Concentrate supplements for hay and straw. *Anim. Prod.* 13:71-82.
- Crabtree, J. R. and G. L. Williams. 1971b. The voluntary intake and utilization of roughage-concentrate diets by sheep. 2. Barley and soya bean meal supplementation of hay diets. *Anim. Prod.* 13:83-92.
- DelCurto, T. R. C. Cochran, D. L. Harmon, A. A. Beharka, K. A. Jacques, G. Towne, and E. S. Vanzant. 1990a. Supplementation of dormant tallgrass-prairie forage: I. Influence of varying supplemental protein and(or) energy levels on forage utilization characteristics of beef steers in confinement. *J. Anim. Sci.* 68:515-531.
- DelCurto, T. R. C. Cochran, D. L. Harmon, A. A. Beharka, K. A. Jacques, G. Towne, and E. S. Vanzant. 1990b. Supplementation of dormant tallgrass-prairie forage: II. Performance and forage utilization characteristics in grazing beef cattle receiving supplements of different protein concentrations. *J. Anim. Sci.* 68:531-542.
- DelCurto, T. R. C. Cochran, T. G. Nagaraja, L. R. Corah, A. A. Beharka and E. S. Vanzant. 1990c. Comparison of soybean meal/sorghum grain, alfalfa hay and dehydrated alfalfa pellets as supplemental protein sources for beef cattle consuming dormant tallgrass-prairie forage. *J. Anim. Sci.* 68:2901-2915.
- Egan, A. R. and R. J. Moir. 1965. Nutritional status and intake regulation in sheep. I. Effect of duodenally infused single doses of casein, urea, and propionate upon voluntary intake of low-protein roughage by sheep. *Aust. J. Agric. Res.* 16:437-449.
- El-Shazly, K. B. A. Dehority and R. R. Johnson. 1961. Effect of starch on the digestion of cellulose *in vitro* and *in vivo* by rumen microorganisms. *J. Anim. Sci.* 20:268-273.

- Elizalde, J. C. N. R. Merchen and D. B. Faulkner. 1999. Supplemental cracked corn for steers fed fresh alfalfa: I. Effects on digestion of organic matter, fiber and starch. *J. Anim. Sci.* 77:457-466.
- Feng, P. C. W. Hunt, G. Pritchard and S. M. Parrish. 1995. Effect of barley variety and dietary barley content on digestive function in beef steers fed grass hay-based diets. *J. Anim. Sci.* 73:3476-3484.
- Fick, K. R. C. B. Ammerman, C. H. McGowan, P. E. Loggins, and J. A. Cornell. 1973. Influence of supplemental energy and biuret nitrogen on the utilization of low quality roughage by sheep. *J. Anim. Sci.* 36:137-143.
- Fisher, D. S. 1996. Modeling ruminant feed intake with protein, chemostatic, and distension feedbacks. *J. Anim. Sci.* 74:3076-3081.
- Fleck, A. T. K. S. Lusby, F. N. Owens and F. T. McCollum. 1988. Effects of corn gluten feed on forage intake, digestibility and ruminal parameters of cattle fed native grass hay. *J. Anim. Sci.* 66:750-757.
- Fontenot, J. P. W. D. Gallup and A. B. Nelson. 1955. Effect of added carbohydrate on the utilization by steers of nitrogen in wintering rations. *J. Anim. Sci.* 14:807-817.
- Forbes, J. M. 1996. Integration of regulatory signals controlling forage intake in ruminants. *J. Anim. Sci.* 74:3029-3035.
- Forster, L. A. Jr. A. L. Goetsch, D. L. Galloway, Sr. and Z. B. Johnson. 1993. Feed intake, digestibility, and live weight gain by cattle consuming forage supplemented with rice bran and(or) corn. *J. Anim. Sci.* 71:3105-3114.
- Freeman, A. S. M. L. Galyean, and J. S. Caton. 1993. Effects of supplemental protein percentage and feeding level on intake, ruminal fermentation, and digesta passage in beef steers fed prairie hay. *J. Anim. Sci.* 70:1562-1572.
- Galloway, D. L. Sr. A. L. Goetsch, L. A. Forster, Jr. A. C. Brake and Z. B. Johnson. 1993a. Digestion, feed intake, and live weight gain by cattle consuming bermudagrass and supplemented with different grains. *J. Anim. Sci.* 71:1288-1297.
- Galloway, D. L. Sr. A. L. Goetsch, L. A. Forster, Jr. A. R. Patil, W. Sun, and Z. B. Johnson. 1993b. Feed intake and digestibility by cattle consuming bermudagrass or orchardgrass hay supplemented with soybean hulls and(or) corn. *J. Anim. Sci.* 71:3087-3095.
- Galloway, D. L. Sr. A. L. Goetsch, L. A. Forster, Jr. W. Sun, and Z. B. Johnson. 1991. Feed intake and digestibility by holstein steers fed warm or cool season grass hays with corn, dried molasses, or wheat midds. *J. Dairy Sci.* 74:1038-1046.

- Garcés-Yépez, P. W. E. Kunkle, D. B. Bates, J. E. Moore, W. W. Thatcher, and L. E. Sollenberger. 1997. Effects of supplemental energy source and amount on forage intake and performance by steers and intake and diet digestibility by sheep. *J. Anim. Sci.* 75:1918-1925.
- Garza, J. D. F. N. Owens and S. Welty. 1991. Effect of post-ruminal protein infusions on feed intake and utilization of low quality hay by beef steers. *Oklahoma Agric. Exp. Sta. MP-134.* pp. 106-113. Stillwater.
- Grigsby, K. N. M. S. Kerley, J. A. Paterson and J. C. Weigel. 1993. Combinations of starch and digestible fiber in supplements for steers consuming a low-quality bromegrass hay diet. *J. Anim. Sci.* 71:1057-1064.
- Guthrie, M. J. and D. G. Wagner. 1988. Influence of protein or grain supplementation and increasing levels of soybean meal on intake, utilization and passage rate of prairie hay in beef steers and heifers. *J. Anim. Sci.* 66:1529-1537.
- Hannah, S. M. R. C. Cochran, E. S. Vanzant and D. L. Harmon. 1991. Influence of protein supplementation on site and extent of digestion, forage intake, and nutrient flow characteristics in steers consuming dormant bluestem-range forage. *J. Anim. Sci.* 69:2624-2633.
- Heldt, J. S. R. J. Pruitt, C. P. Birkelo, P. S. Johnson and Z. W. Wicks III. 1998. Evaluation of wheat middlings as a supplement for beef cows grazing native winter range with differing forage availabilities. *J. Anim. Sci.* 76:378-387.
- Hess, B. W. L. J. Krysl, M. B. Judkins, D. W. Holcombe, J. D. Hess, D. R. Hanks and S. A. Huber. 1996. Supplemental cracked corn or wheat bran for steers grazing endophyte-free fescue pasture: Effects on live weight gain, nutrient quality, forage intake, particulate and fluid kinetics, ruminal fermentation and digestion. *J. Anim. Sci.* 74:1116-1125.
- Hollingsworth-Jenkins, K. J. T. J. Klopfenstein, D. C. Adams, and J. B. Lamb. 1996. Ruminally degradable protein requirements of gestating beef cows grazing native winter sandhills range. *J. Anim. Sci.* 74:1343-1348.
- Hoover, W. H. 1986. Chemical factors involved in ruminal fiber digestion. *J. Dairy Sci.* 69:2755-2766.
- Horn, G. W. and F. T. McCollum III. 1987. Energy supplementation of grazing ruminants. *Proc. Grazing Livestock Nutrition Conference.* July 23-24, Jackson WY. Univ. of WY. pp. 125-136.
- Houseknecht, K. L. D. L. Boggs, D. R. Champion, J. L. Sartin, T. E. Kiser, G. B. Rampacek and H. E. Amos. 1988. Effect of dietary energy source and level on

- serum growth hormone, insulin-like growth factor I, growth and body composition in beef heifers. *J. Anim. Sci.* 66:2916-2923.
- Huntington, G. B. 1997. Starch utilization by ruminants: From basics to the bunk. *J. Anim. Sci.* 75:852-867.
- Illius, A. W. and N. S. Jessop. 1996. Metabolic constraints on voluntary intake in ruminants. *J. Anim. Sci.* 74:3052-3062.
- Jones, A. L. A. L. Goetsch, S. R. Stokes and M. Colberg. 1988. Intake and digestion in cattle fed warm- or cool-season grass hay with or without supplemental grain. *J. Anim. Sci.* 66:194-203.
- Karges, K. K. 1990. Effects of rumen degradable and escape protein on cattle response to supplemental protein on native range. M. S. Thesis. Univ. of Neb. Lincoln.
- Karges, K. K. T. J. Klopfenstein, V. A. Wilkerson and D. C. Clanton. 1992. Effects of ruminally degradable and escape protein supplements on steers grazing summer native range. *J. Anim. Sci.* 70:1957-1964.
- Ketelaars, J. M. H. and B. J. Tolcamp. 1996. Oxygen efficiency and the control of energy flow in animals and humans. *J. Anim. Sci.* 74:3036-3051.
- Köster, H. H. R. C. Cochran, E. C. Titgemeyer, E. S. Vanzant, I. Abdelgadir, and G. St-Jean. 1996. Effect of increasing degradable intake protein on intake and digestion of low-quality tall-grass-prairie forage by beef cows. *J. Anim. Sci.* 74:2473-2481.
- Krysl, L. J. M. E. Branine, A. U. Cheema, M. A. Funk and M. L. Galyean. 1989. Influence of soybean meal and sorghum grain supplementation on intake, digesta kinetics, ruminal fermentation, site and extent of digestion and microbial protein synthesis in beef steers grazing blue grama rangeland. *J. Anim. Sci.* 67:3040-3051.
- Krysl, L. J. and B. W. Hess. 1993. Influence of supplementation on behavior of grazing cattle. *J. Anim. Sci.* 71:2546-2555.
- Lake, R. P. R. L. Hildebrand, D. C. Clanton and L. E. Jones. 1974. Limited energy supplementation of yearling steers grazing irrigated pasture and subsequent feedlot performance. *J. Anim. Sci.* 39:827-833.
- Lusby, K. S. and R. P. Wettemann. 1988. Wheat middlings vs soybean meal and corn/soybean meal supplements at two protein levels for wintering spring calving beef cows. *Oklahoma Agric. Exp. Sta. Stillwater. MP-125. pp. 72-75.*
- Marston, T. T. and K. S. Lusby. 1995. Effects of energy or protein supplements and stage of production on intake and digestibility of hay by beef cows. *J. Anim. Sci.* 73:651-656.

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- Martin, S. K. and C. A. Hibberd. 1990. Intake and digestibility of low-quality native grass hay by beef cows supplemented with graded levels of soybean hulls. *J. Anim. Sci.* 68:4319-4325.
- Matejovsky, K. M. and D. W. Sanson. 1995. Intake and digestion of low-, medium, and high-quality grass hays by lambs receiving increasing levels of corn supplementation. *J. Anim. Sci.* 73:2156-2163.
- McCollum, F. T. and M. L. Galyean. 1985. Influence of cottonseed meal supplementation on voluntary intake, rumen fermentation and rate of passage of prairie hay in beef steers. *J. Anim. Sci.* 60:570-577.
- McCollum, F. T. III and G. W. Horn. 1990. Protein supplementation of grazing livestock: A review. *The Professional Animal Scientist.* 6(2):1-16.
- McLeod, M. N. P. M. Kennedy and D. J. Minson. 1990. Resistance of leaf and stem fractions of tropical forage to chewing and passage in cattle. *Br. J. Nutr.* 63:105-119.
- Mieres, J. M. 1992. Forage intake and digestibility by growing beef cattle supplemented with corn while grazing native range. M. S. Thesis. Okla. State Univ. Stillwater.
- Minson, D. J. 1990. Forage in ruminant nutrition. Academic Press. San Diego, CA.
- Mertens, D. R. 1977. Dietary fiber components: Relationship to the extent and rate of ruminal digestion. *Fed. Proc.* 36(2):187-192.
- Mertens, D. R. and J. R. Loften. 1980. The effect of starch on forage fiber digestion kinetics *in vitro*. *J. Dairy Sci.* 63:1437-1446.
- NRC. 1984. Nutrient requirements of beef cattle. (6th Ed.). National Academy Press. Washington D. C.
- NRC. 1985. Ruminant nitrogen usage. National Academy Press. Washington D. C.
- NRC. 1987. Predicting feed intake of food-producing animals. National Academy Press. Washington D. C.
- NRC. 1996. Nutrient requirements of beef cattle. (7th Ed.). National Academy Press. Washington D. C.
- Olson, K. C. R. C. Cochran, T. J. Jones, E. S. Vanzant, E. C. Titgemeyer and D. E. Johnson. 1999. Effects of ruminal administration of supplemental degradable intake protein and starch on utilization of low-quality warm-season grass hay by beef steers. *J. Anim. Sci.* 77:1016-1025.

- Owens, F. N. J. Garza and P. Dubeski. 1991. Advances in amino acid and N nutrition in grazing ruminants. Proc. Grazing Livestock Nutrition Conference. Aug. 2-3, Steamboat Colo. 109-137.
- Owensby, C. E. R. C. Cochran, R. T. Brandt Jr. E. S. Vanzant, L. M. Auen and E. M. Clary. 1995. Grain supplementation on bluestem range for intensive-early stocked steers. J. Range Mgmt. 48:246-250.
- Poppi, D. P. and S. R. McLennan. 1995. Protein and energy utilization by ruminants at pasture. J. Anim. Sci. 73:278-290.
- Pordomingo, A. J. J. D. Wallace, A. S. Freeman and M. L. Galyean. 1991. Supplemental corn grain for steers grazing native rangeland during summer. J. Anim. Sci. 69:1678-1687.
- Purvis, H. T. II, C. R. Floyd and K. S. Lusby. 1996. Supplements for light-weight calves received and grown on forages. The Professional Animal Scientist. 12:91-98.
- Rittenhouse, L. R. D. C. Clanton and C. L. Streeter. 1970. Intake and digestibility of winter-range forage by cattle with and without supplements. J. Anim. Sci. 31:1215-1221.
- Russell, J. B. J. D. O'Connor, D. G. Fox, P. J. Van Soest, and C. J. Sniffen. 1992. A net carbohydrate and protein system for evaluating cattle diets: I. Ruminal fermentation. J. Anim. Sci. 70:3551-3561.
- Sanson, D. W. and D. C. Clanton. 1989. Intake and digestibility of low-quality meadow hay by cattle receiving various levels of whole shelled corn. J. Anim. Sci. 67:2854-2862.
- Sanson, D. W. D. C. Clanton and I. G. Rush. 1990. Intake and digestion of low-quality meadow hay by steers and performance of cows on native range when fed protein supplements containing various levels of corn. J. Anim. Sci. 68:595-603.
- Satter, L. D. and L. L. Slyter. 1974. Effect of ammonia concentration on rumen microbial protein production *in vitro*. Br. J. Nutr. 32:199-208.
- Spicer, L. A. C. B. Theurer, J. Sowe and T. H. Noon. 1986. Ruminal and post-ruminal utilization of nitrogen and starch from sorghum grain-, corn- and barley-based diets by beef steers. J. Dairy Sci. 62:521-530.
- Stern, M. D. H. Hoover, C. J. Sniffen, B. A. Crooker and P. H. Knowlton. 1978. Effects of nonstructural carbohydrate, urea and soluble protein levels on microbial protein synthesis in continuous culture of rumen contents. J. Anim. Sci. 47:944-956.

Owensby, C. E. R. C. Cochran, R. T. Brandt Jr. E. S. Vanzant, L. M. Auen and E. M. Clary

- Sunvold, G. D. R. C. Cochran and E. S. Vanzant. 1991. Evaluation of wheat middlings as a supplement for beef cattle consuming dormant bluestem-range forage. J. Anim. Sci. 69:3044-3054.
- Vanzant, E. S. R. C. Cochran, K. A. Jacques, A. A. Beharka, T. DelCurto and T. B. Avery. 1990. Influence of level of supplementation and type of grain in supplements on intake and utilization of harvested, early-growing season, bluestem-range forage by beef steers. J. Anim. Sci. 68:1457-1468.

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CHAPTER III

EFFECT OF SUPPLEMENT TYPE ON FORAGE INTAKE, DIGESTION, RUMINAL PARAMETERS AND ANIMAL PERFORMANCE OF GROWING BEEF CATTLE^{1,2}

T. N. Bodine, H. T. Purvis II³, and D. L. Lalman
Oklahoma State University, Department of Animal Science, Stillwater

Abstract

Two experiments were conducted to determine the effects of supplement type on intake, apparent total tract digestibility, ruminal fermentation and kinetics, *in situ* DM disappearance and ruminal forage protein degradation by steers fed prairie hay and rate of gain by heifers grazing bermudagrass pasture. In Exp. 1, 4 ruminally cannulated steers (311 ± 22 kg) had ad libitum access to low-quality (4.1% DIP, 47% TDN, 73% NDF, 40% ADF) prairie hay and were individually fed monensin-containing (200 mg/(steer*d)) treatments consisting of: 1) Mineral mix + corn, MINCR, (.11 kg mineral with .45 kg (as-fed, AF) cracked corn as a carrier, 19 g degradable intake protein (DIP) and .41 kg TDN); 2) Mid-protein pelleted supplement, MP, (1.41 kg DM, 335 g DIP and 1.05 kg TDN); 3) High-fiber, HF, or 4) High-grain, HG, (2.85 kg DM, 340 and 360 g DIP, respectively, and 2.11 kg TDN) pelleted energy supplements in a 4 x 4 Latin square with 14 d adaptation and 6 d sampling periods. Experiment 2 utilized 45 heifers (284 ± 24 kg) grazing late summer bermudagrass pasture for 91 d to determine the effects on animal performance of no supplement (CON) or individually fed MINCS (.09 kg of mineral mix and .23 kg cottonseed hulls as a carrier/heifer daily), MP (1.13 kg/heifer daily), or HF and HG (2.27 kg/heifer daily). In Exp. 1, steers fed MP consumed more ($P < .05$) hay OM than energy or MINCR supplemented cattle. Total OMI was greater ($P < .01$) for animals

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³To whom correspondence should be addressed

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consuming HF, HG or MP than MINCR-fed cattle. Apparent forage OM digestibility (OMD) was not affected ($P > .19$) by supplementation. Total diet OMD was greater ($P < .01$) for energy supplementation than MINCR or MP-fed animals. *In situ* DM disappearance (ISDMD) of prairie hay was not altered ($P > .92$) by treatment while rate was greater ($P < .01$) for energy and protein vs MINCR treatments. In Exp. 2, energy and protein supplemented heifers had greater ($P < .01$) rates of gain than CON treatments, and energy supplemented cattle had greater ($P < .11$) ADG than MP-fed heifers. Results from these studies indicate that feeding milo vs fiber-based energy supplements formulated to provide adequate DIP did not alter forage intake, ISDMD, forage OMD and increased ADG of cattle consuming low-quality forages. Energy supplements balanced for total diet DIP as a percentage of TDN increased total OMI and animal performance over non-supplemented animals regardless of energy source. Adequate DIP:TDN balance reduced or eliminated many negative associative effects noted when high-starch supplements are fed to cattle consuming low-quality hay.

Keywords: Protein, Fiber, Grain, Intake, Beef cattle, Prairie hay

Introduction

Many producers supplement cattle grazing or fed harvested forages with feeds providing predominately either energy or protein in order to achieve acceptable levels of production. Livestock responses to protein supplementation of low-quality forages are well-documented (McCollum and Horn, 1990, Owens et al., 1991), as are the responses typically seen with supplementation from either grain or fiber-based energy sources (Horn and McCollum, 1987, Caton and Dhuyvetter, 1997). Previous research has lead to the common belief (Nickel, 1999) that feeding grain-based supplements tend to decrease

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forage intake and utilization. However, feeding low to moderate amounts of protein supplements will increase forage intake and utilization. In previous research of a variety of supplement sources conducted using the CP system, response of cattle consuming low-quality forage has typically been variable. This variation may be a result of differences in the ruminally degradable portion of both protein and energy contained in the feeds. Recent research (Cochran et al., 1998) has focused on meeting the requirements for degradable intake protein (DIP) based on the amount of total digestible nutrients (TDN) in the diet. This research was undertaken to determine the effects of feeding different types of supplements on forage utilization when the supplements were formulated to provide adequate total diet DIP in relation to the TDN concentration of the total diet.

Materials and Methods

Experiment 1

Animals. Four ruminally cannulated (i.d. 10 cm) steers (Angus and Angus x Hereford; 311 ± 22 kg) were used in a 4 x 4 Latin square design (Steel et al., 1997). Steers were weighed at the initiation and completion of each period, assigned to treatment at the beginning of each period and housed in individual indoor 3 x 4 m pens with ad libitum access to fresh water. Protocols for both experiments were approved by the university animal care and use committee.

Diets. Predominate vegetation species of the prairie hay was composed of big bluestem (*Andropogon gerardii*), little bluestem (*Schizachyrium scoparium*), indiagrass (*Sorghastrum nutans*) and switchgrass (*Panicum virgatum*). Steers were offered ad libitum access to prairie hay by feeding 2.27 kg as-fed (AF) more than the amount of hay intake recorded the previous day. Nutrient and ingredient composition of treatments and

hay are shown in Table 1. Treatments were: 1) MINCR, mineral/vitamin mix fed with cracked corn; 2) MP, mid-protein supplement (cottonseed meal/byproduct-based); or 3) HF, high-fiber (wheat midds/soybean hulls-based) or 4) HG, high-grain (sorghum grain-based) energy supplement. All treatments were formulated to meet or exceed mineral and vitamin requirements (NRC, 1996) and provided 200 mg/(steer*day) monensin. Supplements were pelleted and provided similar calculated DIP levels (1.1 g DIP/ kg BW) for MP, HF and HG. Treatments (Table 1) were fed at 0800 prior to hay at the daily rate of 110 g (AF) mineral mix with 454 g (AF) cracked corn as a carrier for MINCR, .5% BW (DM) for MP or 1% of BW (DM) for HF or HG.

Sampling Procedures. Feeds were weighed daily, supplement refusals were dosed via ruminal cannula and subsamples of hay and supplements were collected d 15 through d 20. Supplement samples were composited across days for each animal within each period and hay samples were composited across days and animals within period. All feed samples were ground through a 2-mm screen in a Wiley mill. Chromic oxide (Cr_2O_3 , 5 g) was dosed intraruminally twice daily d 10 through 18 of each period at 0800 and 2000 in gelatin capsules as an indigestible marker to quantify fecal excretion. At 0 h of d 18 of each period Co-EDTA (200 ml; .41 g Co; Uden et al., 1980) and ytterbium (Yb) -labeled prairie hay (100 g DM, .53 mg Yb, Teeter et al., 1984) were dosed via ruminal cannula for determination of fluid (FPR) and particulate passage rate (PPR). During each period fecal grab samples were collected d 15 through 18 at 0800 and 2000 and stored frozen (-10°C). Fecal samples were thawed, oven-dried (50°C, 96 h), ground (2 mm screen) and composited by steer within day and period for determination of Cr and acid-detergent

insoluble ash (ADIA) concentration. Starting on d 16 of each period, ruminal fluid samples were collected from the center of the ruminal mat and strained through 8 layers of cheesecloth at 0, 2, 4, 8, 12, 16 and 24 h post supplementation. A portable, combination electrode pH meter (Corning 870, Corning, NY) was used to determine pH immediately. Strained samples were acidified with 7.2 N H₂SO₄ at the rate of 1 ml of acid/100 ml of strained rumen fluid and stored frozen (-10°C). Ruminal contents were subsampled and strained through 8 layers of cheesecloth at 0, 2, 4, 8, 12, 16, 24, 30 and 36 h post-feeding on d 18 and 19 from 3 locations in the rumen (caudal ventral, medial ventral and cranial ventral) for determination of marker concentration. Fluid portions were immediately acidified and frozen for determination of Co concentration while particulate matter was reserved and frozen for determination of Yb concentration.

In Situ Procedures. Dacron bags (10 cm X 20 cm, 53 ± 15 µm pore size, Ankom, Fairport, NY) with heat sealed edges were used to determine *in situ* DMD. Five grams (AF) of ground (2-mm screen) prairie hay were incubated in dacron bags in the rumen for 0, 2, 4, 8, 12, 16, 24, 36, 48 or 72 h. Two bags containing hay and 1 blank (empty) bag for each incubation time were placed in the rumen at 0700 on d 16 after soaking for 20 minutes in 39°C water except for 0 and 72 h bags. The 72 h bags were placed in the rumen at 0700 on d 17 and removed at 0700 on d 20 to avoid bag removal and content loss during the sampling period (d 18 and 19) for passage rate markers. Zero hour bags were soaked for 20 minutes and not incubated in the rumen. Bags were placed under the ruminal mat in nylon mesh bags (36 cm X 42 cm). Upon removal, bags were rinsed with 39°C water to remove particles adhering to the outside of bags and stored frozen (-10°C).

In situ forage protein fraction degradation (NDIN, Mass et al., 1999) was determined using similar dacron bag techniques as described for ISDMD with two bags containing hay incubated for 2, 8, 16 or 96 h placed in the rumen at 0700 on d 16. At the completion of the trial, all bags were thawed and washed in a washing machine on delicate setting 10 times for 1-minute rinse and 2-minute spin cycles with a maximum load of 100 bags. Bags were oven-dried (50°C, 72 h) and weighed to determine ISDMD.

Laboratory Analyses. Fecal samples, supplements and forage were analyzed for dry matter by oven drying either at 100°C for 48 h or at 50°C for 96 h. Ash levels of fecal samples, supplements and forage were determined by ashing at 500°C for 6 h in a muffle furnace. Crude protein content of forage and supplements was determined by combustion method (Leco NS2000, St. Joseph, MI) in accordance with AOAC (1990). Forage and supplement CP was classified as UIP or DIP by the enzymatic procedure of Roe et al. (1990). Concentrations of NDF, ADF and ADIA in supplements, hay and fecal samples were determined by methods in accordance with Van Soest et al. (1991). Fiber-bound protein (NDIN) of the residue from dacron bags was determined by NDIN analysis (Mass et al., 1999). Starch levels of supplements were determined enzymatically from α -linked glucose using o-Toluidine (Sigma Chemical, St. Louis, MO) colorimetrically (Galyean, 1997). Ruminal VFA concentrations were determined by deproteinizing 5 ml of rumen fluid with 1 ml of 25% metaphosphoric acid (Erwin et al., 1961) and centrifuging at 20,000 X g for 15 min. Individual VFA were separated by gas chromatography (Perkin Elmer Autosystem, 9000 series, Norwalk, CT) with 8 ml/min flow rate of ultra-high purity helium as a carrier gas and 2-ethylbutyric acid as an internal standard. Ruminal

ammonia nitrogen ($\text{NH}_3\text{-N}$) concentration was determined colorimetrically by enzymatic procedure (Sigma, 1995). Ruminant fluid samples were centrifuged (15,000 X g for 5 min.) and analyzed for Co concentration by atomic absorption spectroscopy (Perkin Elmer Model 4000, Norwalk, CT) with an air plus acetylene flame (Hart and Polan, 1984). Ruminant particulate samples were thawed, dried (50°C, 96 h), ground (2-mm screen) and analyzed for Yb concentration by atomic absorption spectroscopy (Hart and Polan, 1984). Chromium concentrations of fecal composites were quantified by atomic absorption spectroscopy with an air plus acetylene flame (Williams et al., 1962).

Calculations. Fecal OM output was estimated as the average of the ratios of Cr intake to Cr concentrations in feces and ADIA intake and ADIA concentrations in feces. Forage digestibility was calculated by subtracting indigestible OM from supplement from total fecal output. Supplement indigestible OM was assumed to be 100-TDN (Table 1). Rate of ISDMD and NDIN were calculated by regressing the natural logarithm of the percentage of potentially digestible DM or NDIN remaining on time of incubation (4 to 48 or 2 to 16 h, respectively). *In situ* DMD after 72 h of ruminal incubation was considered to represent extent of ISDMD. *In situ* forage protein (NDIN) remaining after 96 h of ruminal incubation was considered to be completely ruminally undegradable. Passage rates of fluid (FPR) and particulate (PPR) matter from the rumen were calculated from the regression of the natural logarithm of Co or Yb concentration, respectively, on sampling time. Fluid volume was calculated by extrapolating the log curve to time zero, taking the inverse natural log and dividing by initial Co dose, fluid flow rate (L/h) was

calculated by multiplying FPR by volume and ruminal fluid turnover time was calculated as one divided by FPR (Galyean, 1997).

Statistical Analyses. Data for intake, digestion, UIP and DIP values, fluid volume, fluid flow rate (L/h) and ruminal fluid turnover time were analyzed using the GLM procedure of SAS (1992). Rates of ISDMD and NDIN degradation, PPR and FPR were determined by using sampling time as a covariate in the GLM procedure of SAS (1992). Effects included in the model were steer, treatment and period. Response of pH, ISDMD, $\text{NH}_3\text{-N}$ and VFA concentrations were determined as a split-plot using the MIXED procedure of SAS (1996). Fixed effects included steer, treatment and period in the whole plot, while time and treatment x time were the sub plot effects. Steer x period x treatment was a random effect and represented the error term used to test whole plot effects. Residual error was used to test subplot effects. Treatment means were calculated using the LSMEANS option. Treatment comparisons were made using preplanned contrasts of MINCR vs MP, MINCR vs (HF + HG)/2, MINCR vs (HF + HG + MP)/3, HF vs HG and (HF + HG)/2 vs MP.

Experiment 2

Animals, Study Site and Sample Collection. Forty-five 1996 fall-born Angus and Angus x Hereford heifers (284 ± 24 kg) grazed a 9 ha common bermudagrass (*Cynodon dactylon*) pasture from July 1 to September 30, 1997 (91 d). This resulted in stocking densities of 1282 and 1560 kg of liveweight per ha on July 1 and September 30, respectively. Pasture management included application of picloram + 2, 4-D (Grazon® P+D, Dow AgroSciences, Indianapolis, IN) and 112 kg N/ha on May 15, 1997. Rainfall

was 693 mm from April 1 to September 30, with 355 mm falling during the experimental grazing period. In order to ensure adequate forage allowance during the trial, grazing of the experimental pasture was minimal between May 15 and July 1, 1997. Forage availability was 3885 and 2994 kg DM/ha on July 1 and September 30, 1997, respectively, as determined from 10 clipped .19 m² quadrats. Diet quality (Table 1) was determined from forage masticate samples collected by four esophageally cannulated heifers (442 kg). Masticate samples were collected mid-July, mid-August and mid-September. Heifers were restricted from feed for three hours prior to sampling and allowed to graze freely for 60 min. Values for July and August were averaged and values for August and September were averaged to reflect changes in forage quality between early (July 1 to August 18) and late (August 18 to September 30) periods. Heifers were treated for internal and external parasites, (Eprinex™, Merial Limited, London, UK), weighed and randomly assigned to one of five treatments on June 24, 1997. Cattle were re-weighed on July 1 (initiation of supplementation), August 18 (interim, d 49) and September 30 (completion, d 91) following a 15 h removal of access to feed and water.

Diets and Feeding Procedures. Treatments consisted of no supplement (CON), mineral mix with cottonseed hulls (MINCS), or one of the three supplements fed in Exp. 1. Cattle were gathered daily at 0700 and fed six times weekly with feedings pro-rated to achieve as-fed intakes of 85 g/(heifer*day) of mineral mix with 227 g CSH as a carrier (MINCS), 1.13 kg/(heifer*day) of MP, or 2.27 kg/(heifer*day) of HF or HG and provide similar monensin (150 mg/(heifer*day)). Treatments were fed in individual stalls in a

feeding barn adjacent to the pasture with supplements offered to animals in the barn for one hour daily.

Statistical Analyses. Variables of initial BW and rate of gain for the first 49 d, second 42 d and entire period were analyzed as a completely randomized design using the GLM procedure of SAS (1992) and comparisons were made using preplanned contrasts of CON vs MINCS, CON vs (HF + HG + MP)/3, HF vs HG and (HF + HG)/2 vs MP. Effects included in the model were heifer and supplement treatment. Individual feeding in a common pasture allowed heifer to be used as the experimental unit (Steel et al., 1997).

Results and Discussion

Experiment 1

DIP levels (Table 1) as determined by enzymatic degradation (Roe et al., 1990) for all supplements varied (± 20 g DIP) from values calculated based on NRC (1996) tabular data. Variation in DIP values points to the need for a quick, easy and accurate assay for DIP in order to effectively utilize the MP system.

Ruminal NH₃-N. There was a diet x time ($P < .01$) interaction for rumen NH₃-N (Figure 1) that was a result of different increases in NH₃-N due to treatment over time. Since this interaction was expected, diet averages are reported in Table 4. Average concentration of NH₃-N post-supplementation was lower ($P < .02$) for MINCR than the supplements and tended ($P = .10$) to be greater for HF vs HG. Average ruminal NH₃-N values were low (< 2 mg/dl) and may have been kept at these low levels by the action of monensin on obligate amino acid fermenting bacteria (Russell, 1996) or by the low protein concentration of the hay. Chase and Hibberd (1987), reported similar ruminal

NH₃ concentrations to those found in the current study and suggested they were characteristic of a deficiency of DIP. Rumen NH₃ levels are indicative of the balance between fermentable organic matter and DIP (Horn and McCollum, 1987, Owens et al., 1991). Addition of rapidly fermentable carbohydrate feeds that are low in DIP would exacerbate a deficiency of ruminal NH₃. If grain supplementation decreased ruminal forage digestion, ruminal forage protein degradation may also be reduced, and decreasing forage DIP content would further compound the protein deficiency. Sanson and Clanton (1989) and Sanson et al. (1990) found no effect on ruminal NH₃ concentration due to additional corn, as would be expected, since already low levels of ruminal NH₃ would not be increased by the addition of greater energy in relation to DIP. Prairie hay has a DIP concentration of about 8 g/100 g of TDN, not much greater than corn (5 g/100 g TDN), explaining one possible mechanism for the negative effect corn supplementation has on ruminal NH₃ and forage digestion in low-quality forages. Ruminal NH₃ values between 2-5 mg/dl are believed to be adequate for microbial growth and fiber digestion (Satter and Slyter, 1974). Our findings reflected average ruminal NH₃-N values below that range. Additionally, time above 2 mg/dl was minimal. Total diet DIP for supplemented steers was adequate in the current study under NRC (1996) Level 1 model guidelines assuming 11% microbial CP yield from TDN. Studies that have found large negative associative effects (Chase and Hibberd, 1987) in relation to ruminal digestibility of low-quality forages have usually been grossly DIP deficient (225 g DIP). El-Shazly et al. (1961) suggested that the addition of increasing DIP *in vivo* and *in vitro* improved the digestion of cellulose when starch levels increased in the diet. This points to the importance of DIP in the utilization of ruminally fermentable OM. Cochran et al. (1998) suggested that total

diet DIP be at least 10% of DOM in order to achieve optimum forage utilization. It would appear that if total diet DIP requirements were met, large negative associative effects may not be observed when supplementing with high levels of starch.

Ruminal pH. A diet x time interaction ($P < .11$) was observed in ruminal pH (Figure 2) that was a result of greater decreases in ruminal pH due to treatment over time. Since this was the expected result, only diet averages are shown in Table 4. Ruminal pH was greater ($P < .01$) for MINCR-fed steers than for energy or protein supplemented cattle and tended ($P > .13$) to be greater for MINCR than MP. Energy supplements exhibited similar ($P > .90$) ruminal pH between fiber and grain sources, while MP tended ($P < .06$) to be greater than energy supplemented cattle. Caton and Dhuyvetter (1997) suggested that reductions in ruminal pH alone could not explain all reductions in forage utilization. Similar values as observed in this study have been found when low-quality prairie hay has been fed with energy supplements (Chase and Hibberd, 1987, Martin and Hibberd, 1990 and Sunvold et al., 1991). It is thought that when rumen pH drops below 6.0, growth and maintenance of fiber degrading bacteria is hindered (Hoover, 1986). It is possible that monensin limited the growth of both hydrogen and lactate producing bacteria (Russell, 1996) which allowed pH to be maintained above 6.0. Additionally, since forage and NDF intakes (Table 2) were not decreased by supplementation, effective NDF (eNDF) intakes would be expected to result in similar or greater buffering capacity for all diets due to rumination (NRC, 1996). In past studies, declines in forage intake would have decreased eNDF intake, decreasing the buffering capacity either directly or indirectly from forage.

VFA Concentration. Total concentration of VFA (Figure 3) and molar proportions of acetate and butyrate (not shown, but similar to total VFA) exhibited a diet x time interaction ($P < .08$). The interaction was expected and a result of different increases in VFA over time due to treatment, so only diet averages are reported in Table 4. No diet effects ($P > .63$) were observed for branched-chain VFA (Table 4). Energy supplementation resulted in greater ($P < .07$) total VFA and acetate than MINCR or MP-fed steers. Butyrate concentrations were greater ($P < .06$) for energy vs MP supplemented animals, while all supplements had increased ($P < .01$) butyrate over MINCR treatments. Propionate was greater ($P < .01$) for energy fed cattle than MP or MINCR and was greater ($P < .08$) for HG than HF. Acetate to propionate ratios were greater ($P < .01$) for MP and MINCR than energy supplemented cattle and tended ($P < .19$) to be greater for HF than HG treatments. Previous research (Grigsby et al., 1993, Carey et al., 1993) has shown energy supplementation to change VFA profiles, Olson et al. (1999) reported increased branched-chain and total VFA concentration as DIP supplementation increased and feeding by-product based supplements increased total VFA's (Martin and Hibberd, 1990, Sunvold et al., 1991). However, Chase and Hibberd (1987), Sanson and Clanton (1989) and Fleck et al. (1988) found no increase in total VFA concentration. Ruminal concentrations of VFA increased with greater amounts of fermentable OM, with different responses for protein and energy supplements. This would be expected to improve energy status of supplemented cattle over that of unsupplemented animals.

Ruminal Kinetics. Fluid passage rates (Table 3) were greater for HF and HG vs MP ($P > .07$) or MINCR ($P < .01$) treatments. Cattle supplemented with HF had greater ($P <$

.08) FPR than HG treatments. Carey et al. (1993), Krysl et al. (1989) and Fleck et al. (1988) all reported no effect on FPR, while Olson et al. (1999) noted increased ruminal fluid kinetics with supplements varying in combinations of starch and DIP. Differences due to treatment were not found ($P > .32$) for ruminal fluid turnover time (Table 3). Supplemented steers had greater ($P < .02$) ruminal fluid volumes (Table 3) than MINCR treatments, while rumen volume of HF cattle tended ($P < .13$) to be greater than HG fed steers. Fluid flow rates (L/h) were greater ($P < .01$) for supplemented steers than MINCR treatments and were greater ($P > .12$) for HF than HG-fed animals. Particulate passage rates (Table 3) were not different ($P = .42$) between treatments and were similar to values seen previously (Chase and Hibberd, 1987, Guthrie and Wagner, 1988). While no differences were seen in the current research, protein supplementation is thought to increase particulate passage (McCollum and Horn, 1990, Guthrie and Wagner, 1988). However, energy supplementation has not always resulted in a consistent change in PPR. Chase and Hibberd (1987) found a decrease, Martin and Hibberd (1990) and Sunvold et al. (1991) noted an increase and Carey et al. (1993) reported no effect.

In Situ DMD. No diet x time interaction ($P > .54$) was found for *in situ* DMD (Table 3) nor were any diet effects ($P > .92$) noted, similar to Chase and Hibberd (1987), Sanson and Clanton (1989) and Arelovich (1983). However, rate of ISDMD (Table 3) was greater ($P < .01$) for supplemented cattle vs MINCR. Chase and Hibberd (1987) reported decreased rate of ISDMD as corn supplementation increased, while Carey et al. (1993) found no difference in rate of digestion (ISDMD). Differences in rate of digestion may be more indicative of greater microbial fermentation of *in situ* forage.

Forage Protein Fractions. Rate of NDIN degradation (Table 5) was not affected ($P = .48$) by diet. Prairie hay UIP and DIP fractions showed no differences ($P > .29$) due to supplementation, possibly due to low forage CP concentration. This differs from Carey et al. (1993) who used moderate quality cool season grass hay and found large differences in rate of forage CP disappearance from *in situ* bags. Protein and beet pulp-supplemented steers had *in situ* CP disappearance rates 3-4 fold greater than barley or corn-fed steers resulting in a reduction of forage DIP due to grain supplementation (Carey et al., 1993).

Intuitively, different supplementation strategies would alter ruminal fermentation of forage, affecting the degradation of fiber-bound protein. If energy supplementation alters rates of passage, rumen retention time would be altered, possibly resulting in differences in specific ruminally available fractions of forage protein. This would agree with the DIP values reported by AFRC (1992) where ruminal outflow rate alters DIP values with increasing flow rates resulting in decreased availability of DIP.

In the current study, DIP from hay was highest for MINCR (79.5% of CP) and lowest for HG (69.5% of CP), possibly due to extent of ruminal fiber digestion. Differences in extent of ruminal fiber degradation may be explained by differences in ruminal retention time and rate of ruminal digestion. Increased rate of ISDMD can infer increased rate of ruminal DM digestion. Changes in digestion rates would also be reflected in rate of NDIN digestion and may be related to changes in lag time, substrate preference or specific components of plants (Hoover, 1986). A 13% decrease in DIP from forage (HG vs MINCR, assuming numeric differences in forage intake did not exist and intakes were equal) resulted in a 70 g increase (approximately .25 lb soybean meal) in DIP required from supplement. It appears that type and amount of supplement offered

affects DIP realized from forage and indicates a need for additional research in forage DIP availability before the metabolizable protein system can be effectively used in forage-feed beef cattle ration formulation.

Digestion. Apparent digestibility (Table 3) of hay OM was not altered by treatment ($P > .19$). This agrees with Sanson and Clanton (1989), Sanson et al. (1990) and Heldt et al. (1998) who found no effect of corn supplementation on hay digestion. Feeding corn with added CP (from soybean meal) may have resulted in similar hay digestion in the trials of Sanson et al. (1990) and Heldt et al. (1998). Chan et al. (1991) reported increased hay OMD from corn or soybean hull supplementation when compared to an unsupplemented control. Fleck et al. (1988) and Marston and Lusby (1995) reported increased hay DMD while Heldt et al. (1998) found no change from by-product supplementation. Typically by-products have had greater DIP than grains and created a different ruminal fermentation profile resulting in less competition between microbes for ruminal NH_3 . Chase and Hibberd (1987) reported decreased hay DMD when corn was fed with decreasing DIP from cottonseed meal and compared with a cottonseed meal control with 153 g of DIP. Supplying DIP in the current study circumvented the negative effects associated with energy supplementation of low-quality forages. Total diet apparent OMD (Table 3) was greater ($P = .01$) for steers fed HF or HG treatments than MINCR or MP fed animals, while TOMD was not different ($P > .30$) due to energy source. Total OMD was affected by the large amount of supplement fed. Chase and Hibberd (1987) reported total diet DMD decreases with high levels of corn that do not agree with the findings of the current research, mainly as a result of their decreased fiber digestion. However, many others have found similar or increased total diet digestion

from supplementation (Guthrie and Wagner, 1988, Carey et al., 1993 and Olson et al., 1999). While balancing total diet DIP, DIP as a percentage of TDN may explain the observed increased TOMD, the same effect may be a result of increasing the amount of highly digestible supplement fed while maintaining forage digestion.

Intake. Initial consumption of mineral mix was low, so 454 g cracked corn was used as a carrier. Hay OMI (Table 2) was greater ($P < .05$) for steers fed MP than energy or MINCR treatments and similar ($P > .55$) for HF and HG as well as similar ($P > .21$) for energy and MINCR treatments. Intake of NDF and total OM (Table 2) were greater ($P < .01$) for energy and protein supplemented than MINCR-fed steers. No negative associative effects were observed for forage intake (substitution) even though supplement (HF or HG) intake ($42 \text{ g/kg BW}^{.75}$) was greater than amounts previously suggested ($30 \text{ g/kg BW}^{.75}$) to alter forage intake (Horn and McCollum, 1987). Forage intake did not decrease when DIP sufficient for fermentation of hay and supplemental energy was provided with high-level energy supplementation (HF and HG). In agreement with our results, Chan et al. (1991) found increased hay and total diet intake for either of two levels (1.5 or 3 kg) of corn or soybean hulls over a negative control with no difference between energy source or level. Starch intake (.42% BW) for HG-fed cattle was equivalent to levels (.38% BW) previously found to decrease forage intake (Chase and Hibberd, 1987, Sanson et al., 1990). This may be reflective of differences in degradability of the protein sources fed between studies. In many studies where isonitrogenous supplements were fed, increases in corn resulted in decreases of oil-seed meal in the supplement formulation. This resulted in greater TDN and decreased DIP, causing ruminally degradable protein to energy ratios of low-quality forage diets to

become considerably more imbalanced (lower). Using casein as a DIP source, Olson et al. (1999) observed no decrease in forage intake response when low-quality tallgrass prairie hay was fed with up to .3% BW starch and compared to an unsupplemented control.

Experiment 2.

Animal Performance. Initial consumption of mineral mix was low so 227 g of cottonseed hulls (CSH) was used as a carrier. Intake of mineral then averaged 43 g per heifer daily (75 mg/(heifer*d) monensin) and was likely due to the short period of time MINCS was offered for consumption each day. Intake of the same mineral mix was greater (62 g/(steer*d)) for steers grazing both tall and mid-grass pastures with ad libitum access to mineral (Bodine et al., 1998). The MP, HF and HG supplements were generally consumed within 30 minutes.

Heifers fed either CON or MINCS did not differ in ADG for either the first ($P > .28$) or second ($P > .26$) period or the entire grazing season ($P > .76$; Table 6). Average rate of gain was increased ($P < .01$) by supplementation (MP, HF, HG) vs CON heifers over the entire grazing period by .26 kg/d with a greater increase coming in the second period due to decreases in forage quality (Table 1). No difference was found between HF and HG supplemented cattle in rate of gain for either period ($P > .63$) or the entire trial ($P > .92$). No difference ($P > .78$) was found for energy (HF, HG) vs MP supplemented heifers during the early grazing period (July 1 to August 18). However, energy supplementation tended ($P < .15$) to increase ADG of heifers from August 18 to September 30 (late period) and the total grazing season ($P = .10$) vs MP supplemented heifers. Steers grazing mid-grass prairie in western Oklahoma and fed the same

treatments during the same time period had similar ADG (Bodine and Purvis, unpublished data) to heifers grazing bermudagrass pasture. This agrees with results in increased gain found by Garcés-Yépez et al. (1997) for steers fed bermudagrass hay and corn, soybean hulls or wheat midds.

Supplement Conversions. Since no differences were found between CON and MINCS, comparisons for supplement conversion were made using the average rate of gain for the entire trial of heifers on those two treatments as a baseline. Additional kilograms of supplement per kg of added gain above the baseline of non-supplemented cattle were 5.43, 8.10 and 8.10 for MP, HF and HG, respectively. These values are similar to those suggested by McCollum and Horn (1990) for energy supplementation, but not as great as would be expected from small amounts of high-protein supplements.

Implications

When evaluating supplements for low-quality forages one needs to consider total diet degradable intake protein requirements. Feeding high-starch pelleted supplements at relatively large amounts (1% BW) did not cause negative associative effects on forage utilization when total diet degradable intake protein requirements were met. Additionally, it would appear that supplement type and level of supplement fed may impact the amount of degradable intake protein available from low-quality forages.

Literature Cited

- Agricultural and Food Research Council. 1992. Technical committee on responses to nutrients, report no. 9. Nutritive requirements of ruminant animals: Protein. Nutrition Abstracts and Reviews, Series B: Livestock Feeds and Feeding. 62:787-835.
- AOAC. 1990. Official Methods of Analysis. (15th Ed.). Association of Official Analytical Chemists, Arlington, VA.
- Arelovich, H. M. 1983. Effect of protein and starch supplementation on the utilization of low-quality roughages. M. S. Thesis. Oklahoma State University. Stillwater.
- Bodine, T. N., H. T. Purvis, M. T. Van Koeving, E. E. Thomas, D. L. Lalman, C. Worthington, B. R. Karges and J. R. Weir. 1998. Effects of supplementation on late summer weight gain of stocker cattle grazing tallgrass and midgrass prairie. Oklahoma Agric. Exp. Sta. Res. Rept. P-965. pp. 162-167. Stillwater.
- Carey, D. A., J. S. Caton and M. Biondini. 1993. Influence of energy source on forage intake, digestibility *in situ* forage degradation and ruminal fermentation in beef steers fed medium quality brome hay. J. Anim. Sci. 71:2260-2269.
- Caton, J. S. and D. V. Dhuyvetter. 1997. Influence of energy supplementation on grazing ruminants: Requirements and responses. J. Anim. Sci. 75:533-542.
- Chan, W. W., C. A. Hibberd, R. R. Scott and K. Swenson. 1991. Corn vs soybean hull supplements for beef cows fed low quality native grass hay. Oklahoma Agric. Exp. Sta. MP-134. pp. 172-178. Stillwater.
- Chase, C. C. Jr. and C. A. Hibberd. 1987. Utilization of low-quality native grass hay by beef cows fed increasing quantities of corn grain. J. Anim. Sci. 65:557-566.
- Cochran, R. C., H. H. Köster, K. C. Olson, J. S. Heldt, C. P. Mathis and B. C. Woods. 1998. Supplemental protein sources for grazing beef cattle. Proc. 9th Annual Florida Ruminant Nutrition Symposium. pp. 123-136.
- El-Shazly, K., B. A. Dehority and R. R. Johnson. 1961. Effect of starch on the digestion of cellulose *in vitro* and *in vivo* by rumen microorganisms. J. Anim. Sci. 20:268-273.
- Erwin, E. S., D. J. Marco and E. M. Emery. 1961. Volatile Fatty Acid Analysis of Blood and Rumen Fluid by Gas Chromatography. J. Dairy Sci. 44:1768-1771.
- Fleck, A. T., K. S. Lusby, F. N. Owens and F. T. McCollum. 1988. Effects of corn gluten feed on forage intake, digestibility and ruminal parameters of cattle fed native grass hay. J. Anim. Sci. 66:750-757.

- Galyean, M. L. 1997. Laboratory Procedures in Animal Nutrition Research. Available at: http://www.asft.ttu.edu/home/mgalyean/lab_man.pdf.
- Garcés-Yépez, P., W. E. Kunkle, D. B. Bates, J. E. Moore, W. W. Thatcher, and L. E. Sollenberger. 1997. Effects of energy source and amount on forage intake and performance by steers and intake and diet digestibility by sheep. *J. Anim. Sci.* 75:1918-1925.
- Grigsby, K. N., M. S. Kerley, J. A. Paterson and J. C. Weigel. 1993. Combinations of starch and digestible fiber in supplements for steers consuming a low-quality bromegrass hay diet. *J. Anim. Sci.* 71:1057-1064.
- Guthrie, M. J. and D. G. Wagner. 1988. Influence of protein or grain supplementation and increasing levels of soybean meal on intake, utilization and passage rate of prairie hay in beef steers and heifers. *J. Anim. Sci.* 66:1529-1537.
- Hart, S. P. and C. E. Polan. 1984. Simultaneous extraction and determination of ytterbium and cobalt ethylenediamine-tetraacetate complex in feces. *J. Dairy Sci.* 67:888-892.
- Heldt, J. S., R. J. Pruitt, C. P. Birkelo, P. S. Johnson and Z. W. Wicks III. 1998. Evaluation of wheat middlings as a supplement for beef cows grazing native winter range with differing forage availabilities. *J. Anim. Sci.* 76:378-387.
- Hoover, W. H. 1986. Chemical factors involved in ruminal fiber digestion. *J. Dairy Sci.* 69:2755-2766.
- Horn, G. W. and F. T. McCollum III. 1987. Energy supplementation of grazing ruminants. Proc. Grazing Livestock Nutrition Conference. July 23-24, Jackson WY. pp. 125-136.
- Krysl, L. J., M. E. Branine, A. U. Cheema, M. A. Funk and M. L. Galyean. 1989. Influence of soybean meal and sorghum grain supplementation on intake, digesta kinetics, ruminal fermentation, site and extent of digestion and microbial protein synthesis in beef steers grazing blue grama rangeland. *J. Anim. Sci.* 67:3040-3051.
- Marston, T. T. and K. S. Lusby. 1995. Effects of energy or protein supplements and stage of production on intake and digestibility of hay by beef cows. *J. Anim. Sci.* 73:651-656.
- Martin, S. K. and C. A. Hibberd. 1990. Intake and digestibility of low-quality native grass hay by beef cows supplemented with graded levels of soybean hulls. *J. Anim. Sci.* 68:4319-4325.

- Mass, R. A., G. P. Lardy, R. J. Grant and T. J. Klopfenstein. 1999. *In situ* neutral detergent insoluble nitrogen as a method for measuring forage protein degradability. *J. Anim. Sci.* 77:1565-1571.
- McCullum, F. T. III and G. W. Horn. 1990. Protein supplementation of grazing livestock: A review. *The Professional Animal Scientist.* 6(2):1-16.
- Nickel, R. 1999. Management: Help or hindrance. Supplementing with grain can be an inefficient way to add energy to a winter ration. *Beef Today.* Jan. pp. 25.
- NRC. 1996. *Nutrient Requirements of Beef Cattle.* (7th Ed.). National Academy Press. Washington, DC.
- Olson, K. C., R. C. Cochran, T. J. Jones, E. S. Vanzant, E. C. Titgemeyer and D. E. Johnson. 1999. Effects of ruminal administration of supplemental degradable intake protein and starch on utilization of low-quality warm-season grass hay by beef steers. *J. Anim. Sci.* 77:1016-1025.
- Owens, F. N., J. Garza and P. Dubeski. 1991. Advances in amino acid and N nutrition in grazing ruminants. *Proc. Grazing Livestock Nutrition Conference.* Aug. 2-3, Steamboat Colo. pp. 109-137.
- Roe, M. B., L. E. Chase and C. J. Sniffen. 1990. Techniques for measuring protein fractions in feedstuffs. In: *Proc. Cornell Nutr. Conf. Feed Manuf.,* Syracuse, NY. p 81. Cornell Univ., Ithaca, NY.
- Russell, J. B. 1996. Mechanisms of ionophore action in ruminal bacteria. *Scientific Update on Rumensin/Tylan/Micotil for the Professional Feedlot Consultant.* Elanco Animal Health. pp. E-1-E-19.
- Sanson, D. W. and D. C. Clanton. 1989. Intake and digestibility of low-quality meadow hay by cattle receiving various levels of whole shelled corn. *J. Anim. Sci.* 67:2854-2862.
- Sanson, D. W., D. C. Clanton and I. G. Rush. 1990. Intake and digestion of low-quality meadow hay by steers and performance of cows on native range when fed protein supplements containing various levels of corn. *J. Anim. Sci.* 68:595-603.
- SAS. 1992. *SAS/STAT[®] Users Guide* (Release 6.08). SAS Inst. Inc., Cary, NC.
- SAS. 1996. *SAS[®] System for MIXED Models.* SAS Inst. Inc., Cary, NC.
- Satter, L. D. and L. L. Slyter. 1974. Effect of ammonia concentration on rumen microbial protein production *in vitro.* *Br. J. Nutr.* 32:199-208.
- Sigma. 1995. Ammonia. Quantitative, enzymatic determination of ammonia in plasma at 340 nm. *Procedure 171-UV* (Rev. Ed.). Sigma Diagnostics, St. Louis, MO.

- Steel, R. G. D., J. H. Torrie and D. A. Dickey. 1997. Principles and Procedures of Statistics: A Biometrical Approach (3rd Ed.) McGraw-Hill Publishing Co. New York.
- Sunvold, G. D., R. C. Cochran and E. S. Vanzant. 1991. Evaluation of wheat middlings as a supplement for beef cattle consuming dormant bluestem-range forage. *J. Anim. Sci.* 69:3044-3054.
- Teeter, R. G., F. N. Owens and T. L. Mader. 1984. Ytterbium chloride as a marker for particulate matter in the rumen. *J. Anim. Sci.* 58:465-473.
- Uden, P., P. E. Colucci and P. J. Van Soest. 1980. Investigation of chromium, cerium and cobalt as markers in digesta. Rate of passage studies. *J. Sci. Food Agric.* 31:625-631.
- Van Soest, P. J., J. B. Robertson and B. A. Lewis. 1991. Symposium: Carbohydrate Methodology, Metabolism and Nutritional Implications in Dairy Cattle. *J. Dairy Sci.* 74:3583 - 3597.
- Williams, C. H., D. J. David and O. Iismaa. 1962. The determination of chromic oxide in faeces samples by atomic absorption spectrophotometry. *J. Agric. Sci. (Camb.)* 59:381-385.

Table 1. Nutrient and ingredient composition (% of DM) of monensin-containing treatments (MINCR = mineral mix + corn, MP = mid-protein, HF = fiber-based, HG = grain-based) and prairie hay fed to steers (Exp. 1) or to heifers grazing bermudagrass¹ (Exp. 2).

Item	Prairie Hay	Bermuda Per. 1	Bermuda Per. 2	Treatment			
				MINCR	MP	HF	HG
<i>Nutrient</i>							
DM	89.79	--	--	88.38	89.91	89.78	89.61
Ash	7.44	13.81	10.95	11.76	10.10	8.57	7.83
CP	5.49	15.69	10.25	7.89	29.07	16.24	18.56
NDF	72.60	60.15	64.52	10.12	37.64	46.96	44.21
ADF	39.97	57.05	56.41	4.02	20.12	21.10	9.31
Starch	.74	--	--	61.79	12.74	18.19	41.13
DIP	4.12	--	--	3.37	23.78	11.93	12.64
TDN	46.62	--	--	72.45	74.12	74.12	74.12
DIP(g/100gTDN)	8.84	--	--	4.65	26.98	16.10	17.05
<i>Ingredient (% DM)</i>							
Corn				81	0	0	0
Milo				0	0	0	50
Soybean hulls				0	20	40	0
Wheat midds				0	10	50	28
Peanut hulls				0	0	3	5
Distillers grain				0	15	2	0
Cottonseed meal				0	46	0	9
Soybean meal				0	2	0	0
Premix ²				19	7	5	8

¹Grazed bermudagrass forage masticate diet quality samples; Period 1 = July 1 – August 18; Period 2 = August 18 – September 30.

²Premix contained mineral-vitamin mix, salt, dicalcium phosphate, limestone, urea, molasses and monensin.

Table 2. Average NH₃-N, pH and VFA values of steers fed low-quality prairie hay and monensin-containing treatments (MINCR = mineral mix + corn, MP = mid-protein, HF = fiber-based, HG = grain-based; Exp. 1).

Ruminal Parameter	Treatment				Contrast ¹	SE
	MINCR	MP	HF	HG		
NH ₃ -N (mg/dl)	.27	1.31	1.86	1.25	1 ^{**}	.22
pH	6.81	6.64	6.44	6.42	1 ^{**} , 2 [†]	.10
Total VFA (mmol/L)	71.06	74.47	84.25	86.01	2 [†] , 5 [*]	3.88
Individual VFA (mol/100 mol)						
Acetate	71.34	71.06	67.62	66.57	2 ^{**} , 5 ^{**}	.59
Propionate	19.19	18.73	21.31	22.60	2 ^{**} , 3 [†] , 5 ^{**}	.43
Butyrate	7.04	7.81	8.70	8.26	1 ^{**} , 2 [*]	.23
Isobutyrate	.00	.02	.03	.04	NS	.02
Isovalerate	1.29	1.33	1.36	1.53	NS	.13
Valerate	1.14	1.05	0.99	1.01	NS	.08
Acetate:Propionate	3.82	3.83	3.30	3.01	2 ^{**} , 5 ^{**}	.14

¹Contrasts; 1 = MINCR vs (MP + HF + HG)/3; 2 = MP vs (HF + HG)/2; 3 = HF vs HG; 4 = MINCR vs MP; 5 = MINCR vs (HF + HG)/2; ** = $P < .01$; * = $P < .05$; † = $P < .10$; NS = not significant ($P > .10$).

Table 3. Average daily intake (g/kg BW) by steers fed low-quality prairie hay and monensin-containing treatments (MINCR = mineral mix + corn, MP = mid-protein, HF = fiber-based, HG = grain-based; Exp. 1).

Item	Treatment				Contrast ¹	SE
	MINCR	MP	HF	HG		
Hay OMI	12.62	18.07	14.04	15.05	1 [†] , 2 [*] , 4 [*]	1.13
Supplement OMI	1.41	4.12	8.44	8.38	ND	.15
NDF	10.06	15.63	15.76	16.19	1 ^{**}	.92
Starch	1.11	2.08	1.95	4.20	ND	.04
Total OMI	14.05	22.64	23.28	24.20	1 ^{**}	.92
Supplement DIP	.06	1.08	1.09	1.16	ND	--
Total diet DIP	.58	1.82	1.67	1.78	ND	--
Total DIP (g/100 g TDN)	9.33	18.96	13.78	14.79	ND	--

¹Contrasts; 1 = MINCR vs (MP + HF + HG)/3; 2 = MP vs (HF + HG)/2; 3 = HF vs HG; 4 = MINCR vs MP; 5 = MINCR vs (HF + HG)/2; ** = $P < .01$; * = $P < .05$; † = $P < .10$; ND = not determined.

Table 4. Ruminal fermentation, kinetics and apparent OM digestibility of hay and total diet by steers fed low-quality prairie hay and monensin-containing treatments (MINCR = mineral mix + corn, MP = mid-protein, HF = fiber-based, HG = grain-based; Exp. 1).

Item	Treatment				Contrast ¹	SE
	MINCR	MP	HF	HG		
Fluid passage rate (%/h)	6.69	7.05	8.04	7.33	2 [†] , 3 [†] , 5 ^{**}	.32
Fluid volume (L)	34.20	45.58	47.39	40.47	1 [*]	2.78
Fluid flow rate (L/h)	2.29	3.20	3.82	2.97	1 ^{**} , 3 [*]	.24
Fluid turnover time (h)	15.31	14.27	12.46	14.77	NS	1.04
Particulate passage rate	2.72	2.71	3.97	3.88	NS	.72
ISDMD, (average)	41.17	41.76	40.91	39.46	NS	2.49
ISDMD, extent (72 h)	68.05	67.79	66.72	63.83	NS	4.59
ISDMD, Kd ² (%/h)	1.92	3.30	3.35	2.88	1 ^{**}	.32
Forage OMD (%)	41.23	35.85	40.53	37.13	NS	1.78
Total diet OMD (%)	44.23	42.40	52.05	49.73	2 ^{**} , 5 ^{**}	1.64

¹Contrasts; 1 = MINCR vs (MP + HF + HG)/3; 2 = MP vs (HF + HG)/2; 3 = HF vs HG; 4 = MINCR vs MP; 5 = MINCR vs (HF + HG)/2; ** = $P < .01$; * = $P < .05$; † = $P < .10$; NS = not significant ($P > .10$).

²Rate of ISDMD of ground prairie hay from dacron bags.

Table 5. Forage protein fractions¹ of low-quality prairie hay fed to steers with monensin-containing treatments (MINCR = mineral mix + corn, MP = mid-protein, HF = fiber-based, HG = grain-based; Exp. 1).

Item	Treatment				Contrast ²	SE
	MINCR	MP	HF	HG		
NDIN, K _d (%/h) ³	1.54	1.06	.45	.38	NS	.59
UIP, (% DM)	1.21	1.36	1.69	1.81	NS	.22
DIP, (% DM)	4.73	4.58	4.25	4.13	NS	.22
UIP, (% CP)	20.44	22.96	28.42	30.46	NS	3.76
DIP, (% CP)	79.57	77.04	71.58	69.54	NS	3.76

¹UIP = undegradable intake protein, DIP = degradable intake protein.

²Contrasts; 1 = MINCR vs (MP + HF + HG)/3; 2 = MP vs (HF + HG)/2; 3 = HF vs HG; 4 = MINCR vs MP; 5 = MINCR vs (HF + HG)/2; NS = not significant ($P > .10$).

³NDIN, K_d = rate of ruminal degradation of forage protein.

Table 6. Rate of gain, total gain and supplement conversions for heifers grazing late summer bermudagrass pasture and fed monensin-containing treatments (CON = no supplement, MINCS = mineral mix + cottonseed hulls, MP = mid-protein, HF = fiber-based, HG = grain-based; Exp. 2).

Item	Treatment					Contrast ¹	SE
	CON	MINCS	MP	HF	HG		
Heifers (n)	9	9	9	9	9		
Initial weight, kg	254	254	263	256	255	NS	6.29
Final weight, kg	296	295	324	323	322	2**	8.41
Total gain, kg	43	41	61	67	67	2**, 4 [†]	4.84
Period 1 ADG (49 d)	.59	.48	.70	.74	.71	NS	.08
Period 2 ADG (42 d)	.32	.42	.62	.72	.77	2**	.07
Overall ADG (91 d)	.47	.45	.67	.74	.74	2**, 4 [†]	.04
Supplement conversion ²	--	--	5.43	8.10	8.10		--

¹Contrasts; 1 = CON vs MINCS; 2 = CON vs (MP+HF+HG)/3; 3 = HF vs HG; 4 = MP vs (HF+HG)/2; ** = $P < .01$; [†] = $P < .10$; NS = not significant ($P > .10$).

²Conversion in kg of supplement fed (AF) per kg of added gain above baseline of CON/MINCS.

Figure 1. Ruminal $\text{NH}_3\text{-N}$ for 24 h post-supplementation of steers fed prairie hay with one of four monensin-containing treatments in Exp. 1 (MINCR = mineral mix + corn, MP = mid-protein, HF = fiber-based, HG = grain-based).

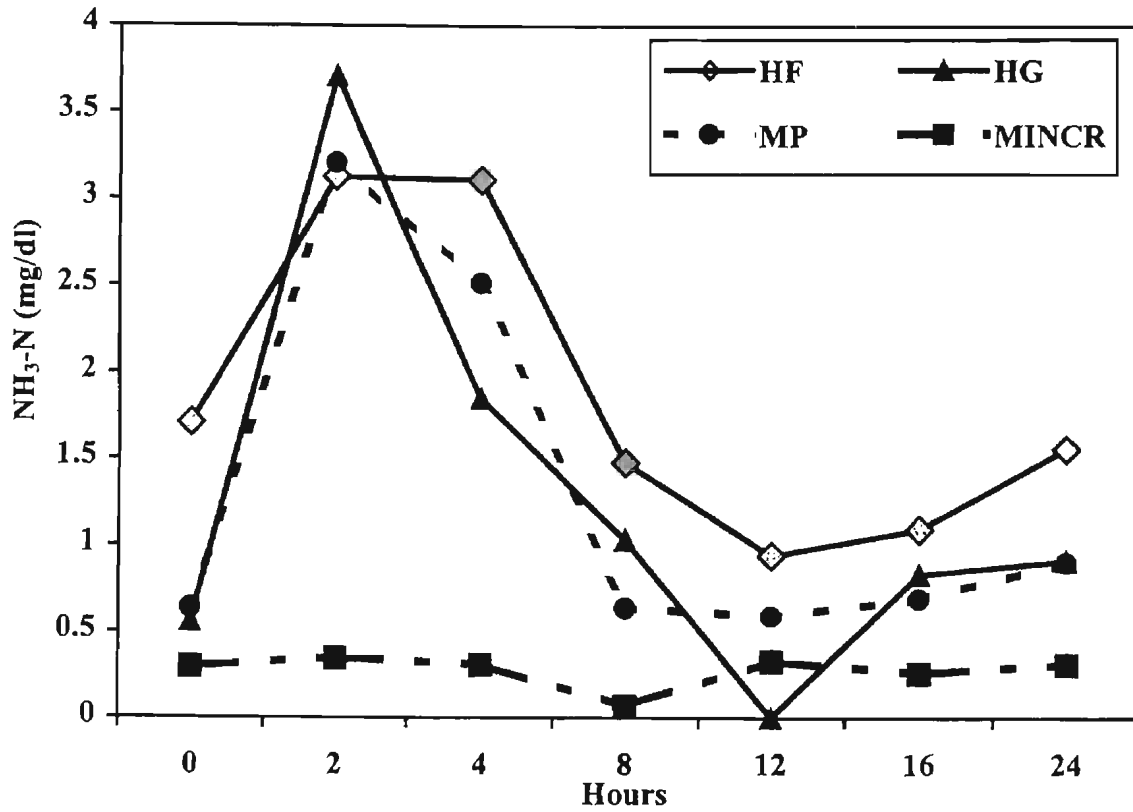


Figure 2. Ruminal pH for 24 h post-supplementation of steers fed prairie hay with one of four monensin-containing treatments in Exp. 1 (MINCR = mineral mix + corn, MP = mid-protein, HF = fiber-based, HG = grain-based).

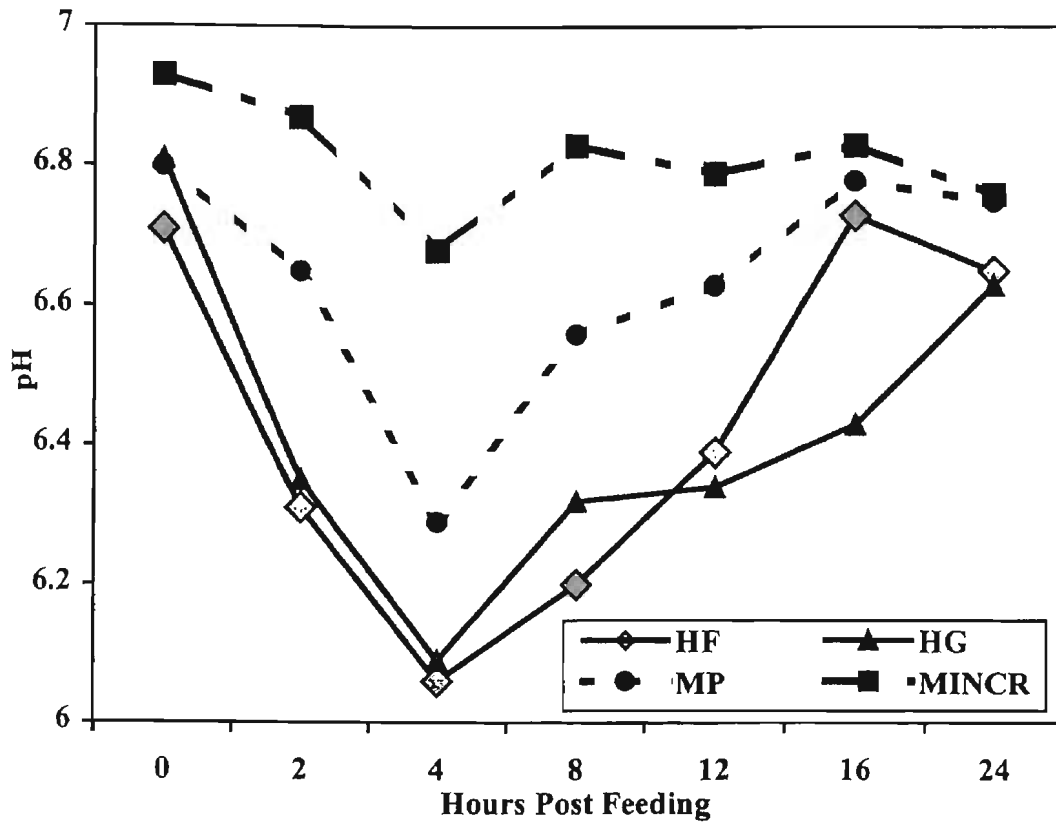
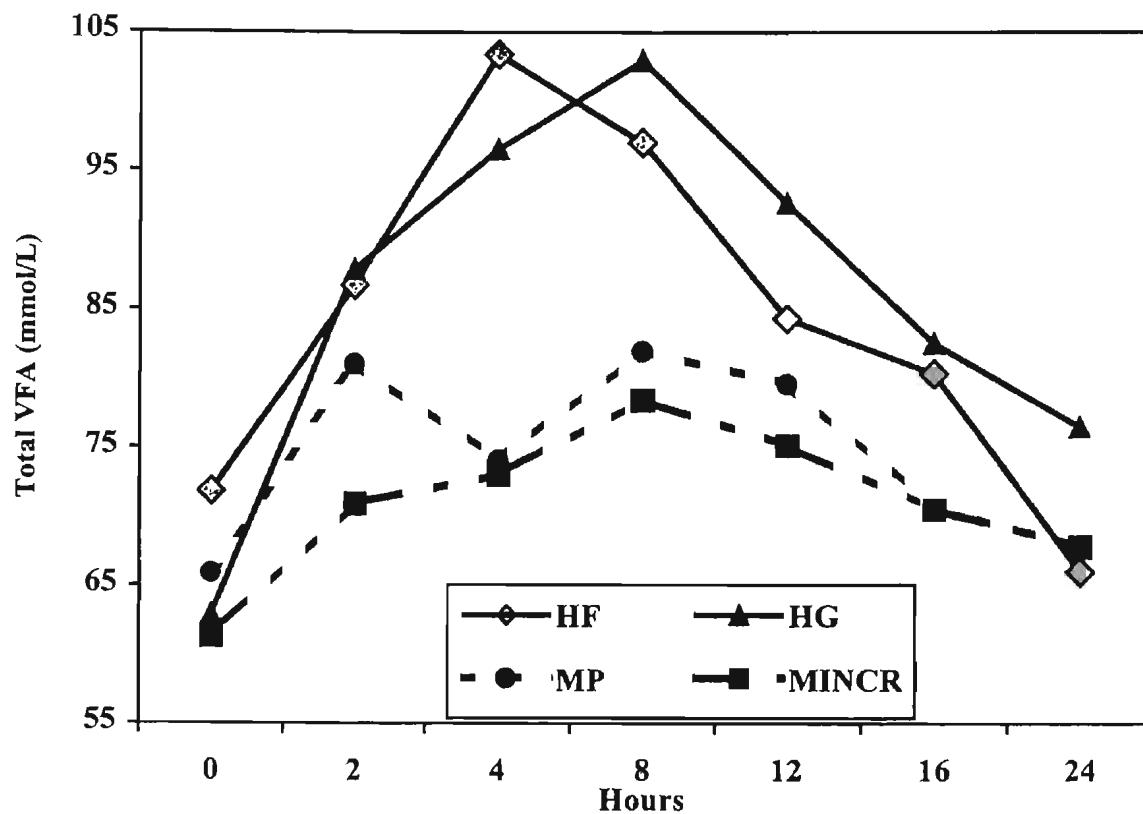


Figure 3. Total VFA concentration for 24 h post-supplementation of steers fed prairie hay with one of four monensin-containing treatments in Exp. 1 (MINCR = mineral mix + corn, MP = mid-protein, HF = fiber-based, HG = grain-based).



CHAPTER IV

EFFECTS OF SUPPLEMENTATION OF PRAIRIE HAY WITH TWO LEVELS OF CORN AND FOUR LEVELS OF SOYBEAN MEAL ON INTAKE, UTILIZATION AND RUMINAL PARAMETERS IN BEEF STEERS^{1,2}

T. N. Bodine*, H. T. Purvis II³, C. J. Ackerman⁴ and C. L. Goad⁵

*Dept. of Animal Science, Oklahoma State University, Stillwater

ABSTRACT

Prairie hay and supplements with various levels of corn and degradable intake protein (DIP) were fed to steers in two experiments to determine the effects on intake, digestion, ruminal fermentation and kinetics. In Exp. 1, ten steers had ad libitum access to chopped prairie hay and were fed .75% BW dry-rolled corn and .25% BW soybean meal and hay intake and digestion were determined for use in Exp. 2. In Exp. 2, eight ruminally cannulated beef steers (317 ± 25 kg) were allotted to an 8 x 8 Latin square experiment with a 2 x 4 factorial arrangement of treatments and fed prairie hay and supplements consisting of dry-rolled corn fed at either 0% (NC) or .75% (CR) of BW (DM) and one of four levels of DIP (from soybean meal; 0, 33, 66, and 100% of NRC (1996) requirements; 0.4, 0.7, 1.0, 1.3 g DIP/kg BW, respectively). Steers were fed for 10 d adaptation and 4 d collection periods. Diets were formulated by balancing total diet DIP to TDN for the CR100 diet and multiplying supplemental DIP for remaining CR treatments by 0, 33 or 66%. Supplements without corn contained equal g DIP/kg BW as respective CR supplements. Supplement DM intake was equalized within NC (1.22 kg/(steer*day)) and CR (3.27 kg/(steer*day)) treatments with cottonseed hulls. In Exp. 1, hay intake was 1.85% BW and hay digestibility was 48%. During Exp. 2, intake of hay

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²The authors would like to acknowledge Steve Welty for his assistance in this experiment.

³To whom correspondence should be addressed.

⁴Present address, Dept. of Anim. Sci., Oregon State Univ., Corvallis.

⁵Department of Statistics, Oklahoma State University.

OM responded with different ($P < .01$) quadratic increases ($P < .06$) as supplemental DIP increased for CR and NC-supplemented cattle. Separate ($P < .01$) quadratic increases ($P < .05$) were noted in total OM intake (TOMI) for CR vs NC treatments as level of DIP increased. A quadratic ($P = .06$) improvement in hay OM digestibility (HOMD) for CR supplements due to increasing DIP was observed. Total OM digestibility had linear increases ($P < .04$) in response to increased DIP that were different ($P < .02$) for treatments with or without corn. Intake of digestible OM (DOM) was greater ($P < .01$) for CR vs NC treatments, while increasing DIP resulted in increased ($P < .01$) DOM regardless of level of corn. Inadequate ruminally degradable protein in grain-based supplements decreased forage intake, digestibility and energy intake of cattle fed low quality prairie hay. Balancing total diet DIP to TDN appeared to overcome negative associative effects typically found when low-quality forages are supplemented with large quantities of low-protein, high-starch feeds.

Keywords: Degradable Intake Protein, Grain, Intake, Digestion, Passage, Beef cattle

Introduction

Supplementation of growing beef cattle to improve rate of gain, efficiency of gain and(or) cost of gain affects a major portion of the beef cattle industry. Cattle producers commonly provide protein or energy supplementation when forage alone does not support a desired level of production. While grains can be the most inexpensive source of energy available, current supplementation strategies indicate an aversion towards inclusion of grains in energy supplements (Nickel, 1999). Previous research (Chase and Hibberd, 1987) has indicated the presence of negative associative effects possibly caused by a deficiency of ruminal ammonia. Current research (Cochran et al., 1998) is

addressing the impact that supplementation has on meeting the DIP requirements of forage-fed cattle as determined from energy intake. This trial was undertaken to determine if negative associative effects of starch supplementation of low-quality forages still occur when total diet degradable intake protein requirements, based on TDN intake, are met.

Materials and Methods

Experimental Design. In Exp. 1, ten ruminally cannulated (i.d. 10 cm) steers (284 ± 9 kg) were fed prairie and a corn and soybean meal supplement to determine forage intake and digestibility to be used in Exp. 2. In Exp. 2, eight ruminally cannulated (i.d. 10 cm) steers (Angus and Angus x Hereford; 317 ± 25 kg) were used in an eight period crossover design (Kuehl, 1994). The order that supplements were fed to each animal during each period was determined using a randomized, unbalanced 8 x 8 Latin square, since adaptation periods were such that carryover effects were assumed to be negligible. Animals were weighed at the initiation and completion of each 14-d period with treatments assigned at the beginning of each period. Individual steers were housed in individual indoor 3 x 4 m pens with ad libitum access to fresh water and trace mineral salt for a 7-d adaptation period. On d 8 animals were moved to individual metabolism stalls for an additional 3-d adaptation period to minimize effects of animal stress followed by a 4-d collection period. All experimental protocols were approved by the university animal care and use committee.

Diets. Prairie hay was from a single source for both Exp. 1 and 2, and had predominate vegetation species composition of big bluestem (*Andropogon gerardii*), little bluestem (*Schizachyrium scoparium*), indiangrass (*Sorghastrum nutans*) and switchgrass

(*Panicum virgatum*). Steers were offered ad libitum access to prairie hay by feeding 2.27 kg as-fed (AF) more than the amount of hay intake recorded the previous day. Nutrient composition of forage and supplements is shown in Table 1. Intake of hay and supplements was recorded daily and hay orts were weighed back. In Exp. 1, all supplements were generally consumed within an hour and consisted of dry-rolled corn fed at .75% initial BW (DM) with a soybean meal-based supplement (93% soybean meal (SBM), plus molasses, dicalcium phosphate, trace mineral salt and vitamin A premix) added to meet DIP requirements (NRC, 1996). Diets were balanced (NRC, 1996 software, Level 1) for DIP requirements (100%) by using analyzed DM, ash, CP and NDF values, known corn intake (based on initial steer BW taken each period), initial BW, NRC (1996) tabular TDN values in Exp. 1, while apparent total tract OM digestibility of hay from Exp. 1 was used as TDN for prairie hay in Exp. 2, 11% microbial CP yield from TDN in Exp. 1 and 11.5% in Exp. 2 and forage DIP (70% CP) and intake (1.75% BW) from a previous grain-based energy-supplemented prairie hay trial (Bodine et al., 1999) during Exp. 1 and during Exp. 2, intake (1.85% BW) from Exp. 1 was used. A soybean meal supplement (Exp. 1) or soybean meal (Exp. 2) was then added to the CR supplement to meet DIP requirements (100%). For Exp. 2, remaining corn (CR) supplements were formulated by multiplying supplemental DIP (g/kg BW) of the CR100 diet by 0, 33 or 66% and balancing SBM, corn and cottonseed hulls (CSH) to equal that amount. Equal supplemental DIP (g/kg BW, Table 1) was fed in respective diets that did not contain corn (NC). Supplement DMI was equalized within CR (3.27 kg/(steer*day)) or NC treatments (1.22 kg/(steer*day)) with CSH. In Exp. 2, all supplement refusals were dosed via ruminal cannula.

Sample Collection and Preparation. Feeds were weighed daily and samples of hay and supplements were collected d 11 through 14. Supplement samples were composited across days for each animal within period and hay samples were composited across days and animals within period for determination of DM, OM, NDF, ADF, acid detergent insoluble ash (ADIA), starch, CP and DIP. All feed samples were ground through a 2-mm screen in a Wiley mill and composited across periods at the completion of the trial since all feeds were from a single source. During each period total fecal collection was performed on d 11 through 14. Fecal samples were weighed, oven-dried (50°C, 96 h, to determine DM) and ground (2-mm screen). At 0 h on d 12 of each period Co-EDTA (200 ml; 1.2 g of Co; Uden et al., 1980) was dosed via ruminal cannula for determination of fluid passage rate (K_{pt}) and ruminal fluid samples were collected from 3 locations in the rumen (caudal-ventral, medial-ventral and cranial-ventral) by straining through 8 layers of cheesecloth at 0, 2, 4, 8, 12, 16 and 24 h post-supplementation. A portable, combination electrode pH meter (Corning 314i pH/mV/temperature portable pH meter with an ISFET (ion selective field effect transistor) electrode, Corning, NY) was used to determine pH. Strained rumen fluid samples (100 ml) were each acidified with 1-ml 7.2 N H₂SO₄ and stored frozen (-10°C). On d 14 ruminal contents were removed, weighed, and volume determined at 0 and 4 h post-supplementation. Contents were then subsampled and placed back into the rumen. Concentration of ADIA in the subsample was used as an indigestible internal marker for particulate passage rate (K_{pp})

Laboratory Analyses. Dry matter was determined by oven-drying at 50°C for 96 h. Ash levels of fecal and ruminal samples, supplements and forage were determined by

ashing at 500°C for 6 h in a muffle furnace. Nitrogen content of forage and supplements was determined by combustion method (Leco NS2000, St. Joseph, MI) in accordance with AOAC (1996). Supplement and hay NDF, ADF and ADIA concentrations were determined by methods in accordance with Van Soest, et al. (1991). Starch content of feeds was determined enzymatically (Megazyme, Wicklow, Ireland) with α -amylase, amyloglucosidase and a colorimetric glucose determination reagent (GOPOD; high purity) in accordance with AOAC (1996). Ruminal fluid samples were thawed, centrifuged (10,000 X g; 10 min.) and subsampled for Co-EDTA, ammonia nitrogen ($\text{NH}_3\text{-N}$) and VFA determination. Subsamples for $\text{NH}_3\text{-N}$ and VFA analysis were composited across time within steer and period. Concentration of Co was determined by atomic absorption spectroscopy (Perkin Elmer Model 4000, Norwalk, CT) with an air plus acetylene flame (Hart and Polan, 1984). Ruminal $\text{NH}_3\text{-N}$ concentration was determined colorimetrically by enzymatic procedure (Sigma, 1995). Concentrations of VFA's were determined by deproteinizing 5 ml of rumen fluid with 1 ml of 25% metaphosphoric acid (Erwin et al., 1961) and centrifuging at 20,000 X g for 15 min. Individual VFA's were separated by gas chromatography (Perkin Elmer Autosystem, 9000 series, Norwalk, CT) with 8 ml/min flow rate of ultra-high purity helium as a carrier gas and 2-ethylbutyric acid as an internal standard. Ruminal particulate samples were thawed, dried (50°C, 96 h), ground (2-mm screen) and analyzed for ADIA concentration (Van Soest et. al., 1991).

Calculations. Apparent total tract forage OMD was calculated by assuming a constant indigestibility of supplement using 100 - tabular TDN values (NRC, 1996) and

subtracting that amount from total fecal output. Fluid dilution rate was calculated from the regression of the natural logarithm of Co concentration on sampling time (Galyean, 1997). Particulate passage rate from the rumen was determined by dividing daily ADIA intake by ADIA fill at each evacuation time (Waldo et al., 1972).

Statistical Analyses. Effects of intake, digestion, average ruminal volume, ruminal OM and ADF fill, ruminal particulate passage rate (K_{pp}), ruminal disappearance rates (K_r), $\text{NH}_3\text{-N}$ and VFA concentrations were analyzed as a 2 x 4 factorial (2 levels of corn and 4 levels of DIP). Ruminal fluid passage rate (K_{pf}) was determined by the addition of sampling time as a covariate to the model. All appropriate interactions were included in the model. To determine the effects of pH, a repeated measures in time analysis was conducted with period, steer, level of corn, level of DIP and sampling time included as fixed effects. Steer by period combinations defined the subjects on which the repeated measures were taken. Correlation structures among the repeated measures of ruminal pH were examined due to heterogeneous variances between periods and between measurements taken at different sampling times. Heterogeneity of variances among periods and a first order autoregressive (AR(1)) correlation structure among repeated measures were adopted. Results were supported by the model fitting criteria calculated by the MIXED procedure. Modeling the covariance structure is necessary since standard errors, and consequently observed significance levels, for all fixed effects comparisons depend on this structure. All analyses were done using SAS/MIXED (SAS Institute, 1996). Satterthwaite degrees of freedom approximation techniques were used. Means were calculated using the LSMEANS option. Linear orthogonal contrasts for level of

corn and linear and quadratic orthogonal contrasts for level of corn and level of corn by level of DIP interactions were used to determine differences.

Results and Discussion

Experiment 1. Hay OM intake averaged across all ten steers was 1.85% BW and apparent total tract OM digestion was 48% when low-quality prairie hay was supplemented with 1% BW of a corn-soybean meal supplement. These values were used in Exp. 2 and all remaining results refer to that experiment.

Ruminal NH₃-N. An interaction ($P < .01$) between levels of corn and DIP was found for ruminal NH₃-N (Figure 1). Ammonia N concentration in the rumen quadratically increased ($P < .01$) for CR treatments as DIP increased while NC-fed steers had a linear ($P < .01$) increase in NH₃-N with increasing DIP. El-Shazly et al. (1961) found large improvements in fiber utilization with urea addition *in vitro* and *in vivo*, that would be expected to increase concentration of NH₃-N in ruminal fluid. Steers fed either NC0 or CR0 supplements had similar ($P > .45$) ruminal concentrations of NH₃-N, while NC100 and CR100-fed cattle also had similar ($P > .35$) ammonia N concentrations. However, NC33 or NC66 treatments were greater ($P < .04$) than CR33 or CR66 treatments. Ruminal NH₃-N values indicate the balance between carbohydrate fermentation and ruminally degradable protein conversion to NH₃. Consequently, low ruminal ammonia may be a result of low DIP or high carbohydrate fermentation. Average ruminal NH₃-N values in the current study were low, and increased as DIP increased, suggesting that increased DIP provided N for microbial needs to allow fermentation of both starch and fiber. Supplemental DIP levels near requirements set by NRC (1996) were necessary to achieve ruminal NH₃-N values that fell within the 2-5 mg/dl range suggested by Satter

and Slyter (1974). Other studies have shown low levels of ruminal ammonia for corn supplementation of low-quality native grass hay (Chase and Hibberd, 1987, Sanson and Clanton, 1989 and Sanson et al., 1990). Fick et al. (1973) did not report ruminal ammonia, but did show a large increase in BUN, which may have been indicative of improved N status and subsequent recycling of N for sheep fed graded levels of an energy supplement with 0 or 10 g of biuret.

Ruminal pH. Interactions ($P < .01$) between level of corn and time (Figure 1) as well as between levels of corn and DIP (Figure 2) occurred for ruminal pH. Averaged across all levels of DIP, ruminal pH was lower ($P < .02$) for CR diets than for NC diets at all times (0 - 24 h). Increasing ruminally fermentable OM should increase microbial growth and increase fermentative end-products that serve to reduce ruminal pH. Other workers have noted addition of starch to decrease ruminal pH (Chase and Hibberd, 1987, Vanzant et al., 1990, Carey et al., 1993). Treatments without additional DIP from SBM (CR0 and NC0) had similar ($P > .35$) average pH values (Table 3), while CR treatments depressed ($P < .01$) pH when compared with NC-fed steers at all three levels (33, 66, 100) of supplemental DIP. Ruminal pH responded to increased DIP with different ($P < .04$) quadratic trends for treatments with ($P < .01$) or without ($P < .01$) corn. While added protein will not increase fermentable OM to the same extent as grains, it should increase forage degradation and acid load by increasing total VFA concentration. This is supported by data from Guthrie and Wagner (1988) and Köster et al. (1996) who found decreased pH as protein supplementation increased.

Ruminal Fermentation. Microbial fermentation of feeds in the rumen results in many end-products. This fermentation is affected by a variety of factors, including many

of those responses measured in this study. Feed intake, rate of passage as well as supply and balance of substrates all can alter the amount and profile of VFA's produced. Total VFA concentration (Table 2) did not exhibit a level of corn by level of DIP interaction ($P > .20$). Diets with CR had greater ($P < .01$) total VFA than NC diets, while increasing DIP resulted in a linear increase ($P < .01$) in total VFA concentration. Acetate, propionate and acetate:propionate ratios (Table 2) exhibited an interaction ($P < .01$) between levels of corn and DIP with quadratic ($P < .01$) responses for CR diets and no effect ($P > .33$) for NC diets. Acetate and acetate:propionate were greater ($P < .01$) while propionate was reduced ($P < .01$) for NC control (0 supplemental DIP from SBM) supplemented diets vs CR0 treatments. Butyrate, isobutyrate and isovalerate (Table 2) were greater ($P < .03$) for CR than NC treatments, while valerate (Table 2) was not affected ($P > .16$) by treatment. Many researchers have found little increase in, or effect on total VFA (Chase and Hibberd, 1987, Sanson and Clanton, 1989 and Freeman et al., 1993) with acetate:propionate ratios most often reduced and butyrate increased by supplemental starch (Hannah et al., 1991, and Olson et al., 1999). It is unclear if this is a result of increased fermentation of the supplement, reductions in the microbial degradation of the basal forage or due to changes in ruminal absorption of specific VFA.

Ruminal Kinetics and Capacity. Ruminal particulate passage rate (K_{pp}), as measured by using ADIA as an indigestible internal marker (Table 3), did not exhibit an interaction ($P > .11$) between levels of corn and DIP. Treatments with corn had greater ($P < .03$) K_{pp} than diets without corn. Grain supplementation has been shown to increase K_{pp} as well as DMI of forage in previous research (Hannah et al., 1991). However, Freeman et al. (1993) and Vanzant et al. (1990) found no difference in K_{pp} with increasing levels of

corn/cottonseed meal or milo grain, respectively, vs an unsupplemented control and also found no effect on forage intake. Results from the present trial indicated that passage of particulate matter from the rumen exhibited a quadratic increase ($P < .06$) as DIP was added. Increases in rate of passage should be linked to increases in intake (Guthrie and Wagner, 1988), since ruminal outflow is required for intake. McCollum and Galyean (1985), Hannah et al. (1991) and Köster et al. (1996) reported increasing levels of protein supplementation increased fluid passage rate (K_{pr}) from the rumen as well as increasing forage DMI. In the current study, K_{pr} exhibited an interaction ($P < .01$) between levels of corn and DIP (Table 3). Level of DIP did not effect ($P > .15$) fluid passage rate of CR treatments while K_{pr} of NC treatments had a quadratic ($P < .10$) increase as level of DIP increased. This may be a result of rapid fluid passage on CR diets while fluid passage on the NC0 treatment was much slower, offering an opportunity for a greater response. This is supported by Olson et al. (1999), who also suggested that increasing levels of starch appeared to reduce the increases in ruminal passage due to DIP supplementation. Increases in rate of passage with no change in total tract digestion for NC treatments appears to indicate greater rate of ruminal digestion or a shift to increased lower tract digestion as a result of protein supplementation. Increased rate of ruminal digestion is supported by increased total VFA and acetate concentrations, indicating more rapid ruminal digestion of the hay-based diet. Rate of ruminal OM disappearance (K_{OM} , Table 3), as a result of passage and digestion, was greater ($P < .01$) for CR than NC treatments with a quadratic ($P < .02$) increase as DIP increased across either CR or NC supplemented diets. Rate of ruminal ADF disappearance (K_{ADF} , Table 3) exhibited an interaction ($P < .09$) between levels of DIP and corn. Steers fed CR supplements had a

linear increase ($P < .01$) in K_r ADF while NC treatments resulted in a quadratic increase ($P < .01$) as DIP increased. Rate of ADF removal from the rumen was greater ($P < .04$) for NC diets at the 33 and 66 level of DIP but did not differ ($P > .52$) from respective CR diets at 0 or 100 supplemental DIP.

Ruminal OM fill (Table 3) did not differ ($P > .98$) between NC and CR treatments but did linearly decrease ($P < .01$) as DIP increased, regardless of level of corn. This is in agreement with Vanzant et al. (1990), who reported little effect of increasing grain on ruminal fill. However, most reports indicate that ruminal fill increases with increasing protein (McCullum and Horn, 1990 and Hannah et al., 1991). Yet, in agreement with our findings, Köster et al. (1996) found increasing DIP to result in decreasing ruminal fill with a low-quality prairie hay similar to the one used in the current study. Reductions in ruminal fill indicate that intake did not increase to as great an extent as did disappearance, resulting in decreased fill and indicating that ruminal fill or physical factors did not control intake. However, ADF fill (Table 3) was greater ($P < .01$) for NC than CR-supplemented steers and decreased quadratically ($P < .06$) as DIP increased. This also suggests that the slowly degraded fiber portion (ADF) of the diet did not limit intake since ADF leaving the rumen (via digestion or passage) increased to a greater extent than did fiber intake. Ruminal volume (Table 3) exhibited a similar trend as ADF fill, with NC treatments having greater ($P < .01$) ruminal volume than CR-supplemented animals, and a quadratic decrease ($P < .08$) in ruminal volume occurring as DIP increased.

Digestion. Apparent total tract hay OM digestibility (HOMD, Figure 4) was not affected ($P > .43$) by level of DIP for NC treatments while a quadratic increase ($P < .03$) in HOMD was noted as supplemental DIP increased. This increase in intake and constant

digestibility can be explained by increased passage rates. McCollum and Horn (1990), Owens et al. (1991) and Caton and Dhuyvetter (1997) suggested that protein supplementation can increase digestion. One mechanism to explain increased forage digestion is that DIP reduces competition between ruminal microbes providing NH_3 for fibrolytic microbial use, thereby increasing forage digestion (El-Shazly et al., 1961, Russell et al., 1992). Total diet OM digestibility (TOMD, Table 4) exhibited an interaction ($P = .01$) between levels of corn and DIP. Treatments with or without corn responded to dietary DIP addition from supplement with unique ($P = .03$) linear increases ($P < .01$) in TOMD. Increasing the amount of highly digestible supplement, while either not decreasing or improving forage digestion resulted in greater diet digestibility for grain diets. This is in agreement with Fick et al. (1973) and Sanson et al. (1990) who found increasing levels of grain fed with a source of ruminally available N increased total diet OMD.

Intake. Hay OM intake (HOMI, Figure 5) exhibited an interaction ($P < .01$) between level of corn and level of DIP. Hay OMI responded to increasing DIP supplementation with different ($P = .06$) quadratic increases for treatments with or without corn. Steers fed CR supplements increased hay OMI quadratically ($P < .01$) as DIP increased with the numerically greatest OMI of forage occurring for steers fed the CR66 supplement. Previous research has documented the increase in low-quality forage intake due to supplementation (McCollum and Galyean, 1985, Guthrie and Wagner, 1988). However, few researchers have compared the effects of increasing protein with or without starch. The current study differs from many previous studies in several ways. The first is the use of a negative control with zero additional DIP from SBM. The second is maintaining a

constant supplement DMI within level of corn supplementation. Additionally, one last difference is the use of the NRC (1996) framework to ensure adequate DIP for the high level of grain supplementation. Increasing fermentable OM from supplementation has often decreased forage intake in previous work when DIP supply was not sufficient (Chase and Hibberd, 1987). Our results suggest that forage intake can be increased over a true negative control with high levels of energy supplementation by the addition of DIP. Steers receiving supplements without SBM had similar ($P > .96$) HOMI with or without corn, which does not agree with Chase and Hibberd (1987), who reported corn decreased hay intake. Forage intake was greater ($P < .01$) for each NC diet with supplemental DIP (33, 66, 100) than its respective CR diet (33, 66, 100). Since supplemental DMI was maintained constant within level of corn, total diet OMI (Table 4) had the same pattern of responses, with CR diets having greater ($P < .04$) total OMI at all levels of DIP than NC treatments due to the increased supplement fed. Intake of digestible OM (DOM) did not exhibit an interaction ($P > .41$) between levels of corn and DIP as shown in Figure 6. In addition, individual treatment means are reported in Table 4. Across levels of corn, greater supplemental DIP increased ($P < .01$) intake of DOM quadratically. Steers fed NC vs CR supplements had reduced ($P < .01$) DOM. In the current study, steers fed low-quality forage and individually supplemented with corn and DIP from SBM increased forage intake over control animals to a lesser extent than steers fed only SBM. However, grain supplementation increased total intake and total digestible intake.

Implications

Cattle consuming low-quality warm season grass hay can efficiently utilize a 20% CP supplement made from 2/3 corn - 1/3 soybean meal without negative associative

effects on forage intake or utilization. When the supplemental protein source provided adequate ruminally degradable protein to ferment organic matter from grain and hay protein, feeding a grain-based supplement increased intake, utilization and ruminal fermentation of animals fed low-quality hay, which improved energy intake. This would be expected to result in greater animal performance and indicates that added ruminally degraded protein balanced for total diet TDN supply will alleviate negative associative effects of high levels of supplemental grain when fed with low-quality grass hay. While many other researchers have found differing results in regards to grain supplementation of low-quality results, previous research conducted with added ruminally degradable protein sources supports the current findings.

Literature Cited

- AOAC. 1996. Official Methods of Analysis. (16th Ed.). Association of Official Analytical Chemists, Arlington, VA.
- Bodine, T. N., H. T. Purvis, M. T. Van Koeveering and E. E. Thomas. 1999. Effects of supplement source on intake, digestion and ruminal kinetics of steers fed prairie hay. Oklahoma Agric. Exp. Sta. Res. Rept. In press. Stillwater.
- Carey, D. A., J. S. Caton, and M. Biondini. 1993. Influence of energy source on forage intake, digestibility, *in situ* forage degradation, and ruminal fermentation in beef steers fed medium quality brome hay. J. Anim. Sci. 71:2260-2269.
- Caton, J. S. and D. V. Dhuyvetter. 1997. Influence of energy supplementation on grazing ruminants: Requirements and responses. J. Anim. Sci. 75:533-542.
- Chase, C. C. Jr. and C. A. Hibberd. 1987. Utilization of low-quality native grass hay by beef cows fed increasing quantities of corn grain. J. Anim. Sci. 65:557-566.
- Cochran, R. C., H. H. Köster, K. C. Olson, J. S. Heldt, C. P. Mathis and B. C. Woods. 1998. Supplemental protein sources for grazing beef cattle. Proc. 9th Annual Florida Ruminant Nutrition Symposium. pp. 123-136.
- El-Shazly, K., B. A. Dehority and R. R. Johnson. 1961. Effect of starch on the digestion of cellulose *in vitro* and *in vivo* by rumen microorganisms. J. Anim. Sci. 20:268-273.
- Erwin, E. S., D. J. Marco and E. M. Emery. 1961. Volatile Fatty Acid Analysis of Blood and Rumen Fluid by Gas Chromatography. J. Dairy Science. 44:1768-1771.
- Fick, K. P., C. B. Ammerman, C. H. McGowan, P. E. Loggins and J. A. Cornell. 1973. Influence of supplemental energy and biuret nitrogen on the utilization of low quality roughage by sheep. J. Anim. Sci. 36:137-143.
- Freeman, A. S., M. L. Galyean, and J. S. Caton. 1993. Effects of supplemental protein percentage and feeding level on intake, ruminal fermentation, and digesta passage in beef steers fed prairie hay. J. Anim. Sci. 70:1562-5172.
- Galyean, M. L. 1997. Laboratory Procedures in Animal Nutrition Research. Available at: http://www.asft.ttu.edu/home/mgalyean/lab_man.pdf.
- Guthrie, M. J. and D. G. Wagner. 1988. Influence of protein or grain supplementation and increasing levels of soybean meal on intake, utilization and passage rate of prairie hay in beef steers and heifers. J. Anim. Sci. 66:1529-1537.
- Hannah, S. M., R. C. Cochran, E. S. Vanzant and D. L. Harmon. 1991. Influence of protein supplementation on site and extent of digestion, forage intake, and nutrient

- flow characteristics in steers consuming dormant bluestem-range forage. *J. Anim. Sci.* 69:2624-2633.
- Hart, S. P. and C. E. Polan. 1984. Simultaneous extraction and determination of ytterbium and cobalt ethylenediamine-tetraacetate complex in feces. *J. Dairy Sci.* 67:888-892.
- Köster, H. H., R. C. Cochran, E. C. Titgemeyer, E. S. Vanzant, I Abdelgadir, and G. St-Jean. 1996. Effect of increasing degradable intake protein on intake and digestion of low-quality tallgrass-prairie forage by beef cows. *J. Anim. Sci.* 74:2473-2481.
- Kuehl, R. O. 1994. *Statistical principles of research design and analysis.* Wadsworth Publishing Co. Belmont, CA.
- McCollum, F. T. and M. L. Galyean. 1985. Influence of cottonseed meal supplementation on voluntary intake, rumen fermentation and rate of passage of prairie hay in beef steers. *J. Anim. Sci.* 60:570-577.
- McCollum, F. T. III and G. W. Horn. 1990. Protein supplementation of grazing livestock: A review. *The Professional Animal Scientist.* 6(2):1-16.
- Nickel, R. 1999. Management: Help or hindrance. Supplementing with grain can be an inefficient way to add energy to a winter ration. *Beef Today.* Jan. pp. 25.
- NRC. 1996. *Nutrient Requirements of Beef Cattle.* (7th Ed.). National Academy Press. Washington, DC.
- Olson, K. C., R. C. Cochran, T. J. Jones, E. S. Vanzant, E. C. Titgemeyer and D. E. Johnson. 1999. Effects of ruminal administration of supplemental degradable intake protein and starch on utilization of low-quality warm-season grass hay by beef steers. *J. Anim. Sci.* 77:1016-1025.
- Owens, F. N., J. Garza and P. Dubeski. 1991. Advances in amino acid and N nutrition in grazing ruminants. *Proc. Grazing Livestock Nutrition Conference.* Aug. 2-3, Steamboat Colo. 109-137.
- Russell, J. B., J. D. O'Connor, D. G. Fox, P. J. Van Soest, and C. J. Sniffen. 1992. A net carbohydrate and protein system for evaluating cattle diets: I. Ruminal fermentation. *J. Anim. Sci.* 70:3551-3561.
- Sanson, D. W. and D. C. Clanton. 1989. Intake and digestibility of low-quality meadow hay by cattle receiving various levels of whole shelled corn. *J. Anim. Sci.* 67:2854-2862.
- Sanson, D. W., D. C. Clanton and I. G. Rush. 1990. Intake and digestion of low-quality meadow hay by steers and performance of cows on native range when fed protein supplements containing various levels of corn. *J. Anim. Sci.* 68:595-603.

- SAS Institute. 1996. SAS® System for MIXED Models. SAS Inst. Inc., Cary, NC.
- Satter, L. D. and L. L. Slyter. 1974. Effect of ammonia concentration on rumen microbial protein production *in vitro*. Br. J. Nutr. 32:199-208.
- Sigma. 1995. Ammonia. Quantitative, enzymatic determination of ammonia in plasma at 340 nm. Procedure 171-UV (Rev. Ed.). Sigma Diagnostics, St. Louis, MO.
- Uden, P., P. E. Colucci and P. J. Van Soest. 1980. Investigation of chromium, cerium and cobalt as markers in digesta. Rate of passage studies. J. Sci. Food Agric. 31:625-631.
- Van Soest, P. J., J. B. Robertson and B. A. Lewis. 1991. Symposium: Carbohydrate Methodology, Metabolism and Nutritional Implications in Dairy Cattle. J. Dairy Sci. 74:3583 – 3597.
- Vanzant, E. S., R. C. Cochran, K. A. Jacques, A. A. Beharka, T. DelCurto and T. B. Avery. 1990. Influence of level of supplementation and type of grain in supplements on intake and utilization of harvested, early-growing-season, bluestem-range forage by beef steers. J. Anim. Sci. 68:1457-1468.
- Waldo, D. R., L. W. Smith, and E. L. Cox. 1972. Model of cellulose disappearance from the rumen. J. Dairy Sci. 55:125-129.

Table 1. Ingredient (kg of DM) and nutrient (% of DM) composition of supplements with two levels of corn (NC or CR; 0 or .75% BW) and four levels of degradable intake protein (DIP; as a percentage of NRC (1996) requirements) and prairie hay.

Item	Corn Level DIP Level	NC (0% BW)				CR (.75% BW)			
		0	33	66	100	0	33	66	100
Intake (kg DM)									
Corn		0	0	0	0	2.32	2.32	2.32	2.32
Soybean meal		0	.58	.86	1.16	0	.29	.58	.87
Cottonseed hulls		1.15	.58	.29	0	.86	.57	.29	0
Mineral mix		.07	.06	.07	.06	.07	.07	.06	.06
Supplement		1.22	1.22	1.22	1.22	3.25	3.25	3.25	3.25
Intake (g/kg BW)									
Supplement		3.84	3.84	3.84	3.84	10.23	10.23	10.23	10.23
DIP		.08	.67	.95	1.28	.36	.65	.95	1.28
TDN ^a		1.51	2.36	2.74	3.24	7.65	8.09	8.50	9.16
Starch		.01	.04	.05	.07	5.18	5.19	5.20	5.34
Nutrient (% DM)									
	Prairie hay								
DM	92.01	90.16	89.74	89.55	89.33	88.20	88.13	88.06	87.98
OM	93.69	92.26	90.13	88.98	87.99	96.16	95.72	95.30	94.91
NDF	74.63	85.29	48.83	30.85	12.54	38.51	31.81	25.08	18.20
ADF	44.66	65.34	36.07	21.64	6.94	22.67	17.31	11.92	6.39
Starch	.78	.35	1.06	1.40	1.76	51.10	51.08	51.11	51.17
CP	6.07	4.01	27.26	38.64	50.36	7.76	12.10	16.43	20.87
DIP	4.13	2.01	17.42	24.97	32.74	3.52	6.41	9.28	12.22
TDN ^a	48.00	40.01	61.61	72.13	83.05	75.69	79.63	83.58	87.66
DIP (g/100g TDN)	8.61	5.06	28.30	34.63	39.45	4.73	8.05	11.11	13.95

^aCalculated from actual intake and tabular (NRC, 1996) TDN values for corn (90), soybean meal (87) and cottonseed hulls (42).

Table 2. Average ruminal VFA concentrations of beef steers fed prairie hay and supplemented with two levels of corn (NC or CR; 0 or .75% BW) and four levels of degradable intake protein (DIP; as a % of NRC (1996) requirements).

Item	Corn Level DIP Level	NC (0% BW)				Contrast ¹	CR (.75% BW)				SE	
		0	33	66	100		0	33	66	100		
Total VFA, mmol/L		76.52	95.70	102.25	113.74	DIP, L	88.52	98.11	110.72	115.81	DIP, L	2.86
Individual VFA, mol/100 mol												
Acetate		74.07	73.80	72.89	73.10	NS	67.11	71.47	73.02	70.65	Q	.55
Propionate		15.84	16.01	16.65	16.54	NS	19.85	15.61	14.64	16.36	Q	.56
Butyrate		7.97	7.95	8.23	8.30	a	10.24	10.01	9.58	10.41	b	.31
Valerate		1.26	.93	1.17	.93	NS	1.39	1.10	1.25	1.11	NS	.17
Isovalerate		.67	.94	.90	.77	a	1.13	1.07	.95	1.11	b	.11
Isobutyrate		.19	.37	.17	.36	a	.28	.75	.57	.37	b	.14
Acetate:Propionate		4.72	4.72	4.39	4.45	NS	3.41	4.62	5.01	4.38	Q	.18

¹DIP = no level of corn by level of DIP interaction, average values for main effect differ by level of DIP across both levels of corn as determined by: L = linear, Q = quadratic, D = different ($P < .01$) response across levels of DIP between levels of corn; l = linear, q = quadratic, d = different ($P < .11$) responses across levels of DIP between levels of corn; NS = no significant trend ($P > .12$) across levels of DIP.

^{a,b}No level of corn by level of DIP interaction, average values for main effect of level of corn within ruminal measure without common superscripts differ ($P < .03$).

Table 3. Average ruminal kinetics and capacity of beef steers fed prairie hay and supplemented with two levels of corn (NC or CR; 0 or .75% BW) and four levels of degradable intake protein (DIP; as a % of NRC (1996) requirements).

Item	Corn Level DIP Level	NC (0% BW)					CR (.75% BW)					SE
		0	33	66	100	Contrast ¹	0	33	66	100	Contrast ¹	
K _{pp} , (ADIA), %/h		1.93	3.01	3.30	3.52	DIP, q, b	2.58	3.08	3.25	3.79	DIP, q, a	.16
K _{pf} , (Co-EDTA), %/h		6.12	8.80	8.53	9.81	q	9.87	9.78	10.37	10.97	L	.43
K _t , (OM), %/h		2.20	3.65	4.03	4.32	DIP, q, b	3.00	3.87	4.29	5.15	DIP, q, a	.17
K _t , (ADF), %/h		2.14	3.65	4.05	4.27	Q	2.29	3.17	3.56	4.34	L	.17
OM Fill, kg		9.47	8.69	8.14	7.78	DIP, L	9.76	8.92	8.33	7.05	DIP, L	.33
ADF Fill, kg		5.15	4.02	3.60	3.31	DIP, q, b	4.72	3.95	3.48	2.48	DIP, q, a	.16
Volume, L		65.46	66.55	63.28	60.91	DIP, q, b	63.90	63.97	60.63	56.66	DIP, q, a	1.55

¹DIP = no level of corn by level of DIP interaction, average values for main effect differ by level of DIP across both levels of corn as determined by: L = linear, Q = quadratic ($P < .01$); q = quadratic ($P < .06$) across levels of DIP.

^{a,b}No level of corn by level of DIP interaction, average values for main effect of level of corn within ruminal measure without common superscripts differ ($P < .04$).

Table 4. Average digestibility (% of OM intake) and daily intake (g/kg BW) of hay, diet and nutrients by beef steers fed prairie hay and supplemented with two levels of corn (NC or CR; 0 or .75% BW) and four levels of degradable intake protein (DIP; as a % of NRC (1996) requirements).

Item	Corn Level DIP Level	NC (0% BW)				Contrast ¹	CR (.75% BW)				Contrast ¹	SE
		0	33	66	100		0	33	66	100		
Total OMD, %		54.13	56.79	59.75	60.72	L, d	51.99	60.3	58.89	65.59	L, d	1.25
Total OMI, g/kg BW		15.58	23.20	23.99	24.71	Q, d	21.90	25.5	26.43	26.46	Q, d	.61
Total Starch, g/kg BW		.07	.21	.23	.25		5.29	5.3	5.34	5.48		
Total DIP, g/kg BW		.61	1.55	1.87	2.23		.89	1.3	1.69	2.01		
Total DIP, g/100 g DOM		7.09	11.78	12.97	14.92		7.78	8.7	10.95	11.61		
Total DOM, g/kg BW		8.42	13.09	14.33	14.99	DIP, Q, a	11.34	15.4	15.52	17.35	DIP, Q, b	.55

¹DIP = no level of corn by level of DIP interaction, average values for main effect differ by level of DIP across both levels of corn as determined by: L = linear, Q = quadratic ($P < .01$); q = quadratic, d = different trends ($P < .06$); NS = no significant trend ($P > .43$) across levels of DIP.

^{a,b}No level of corn by level of DIP interaction, average values for main effect of level of corn within intake or digestion measure without common superscripts differ ($P < .01$).

Figure 1. Average ruminal NH₃-N concentration (mg/dl) of steers fed prairie hay with supplements containing 0 or .75% BW dry-rolled corn (NC or CR) and four increasing levels of DIP (as a percentage of NRC (1996) requirements). Cattle fed CR supplements exhibited quadratic increases ($P < .01$) in NH₃-N while NC-fed steers exhibited linear increases ($P < .01$) in NH₃-N with increasing DIP.

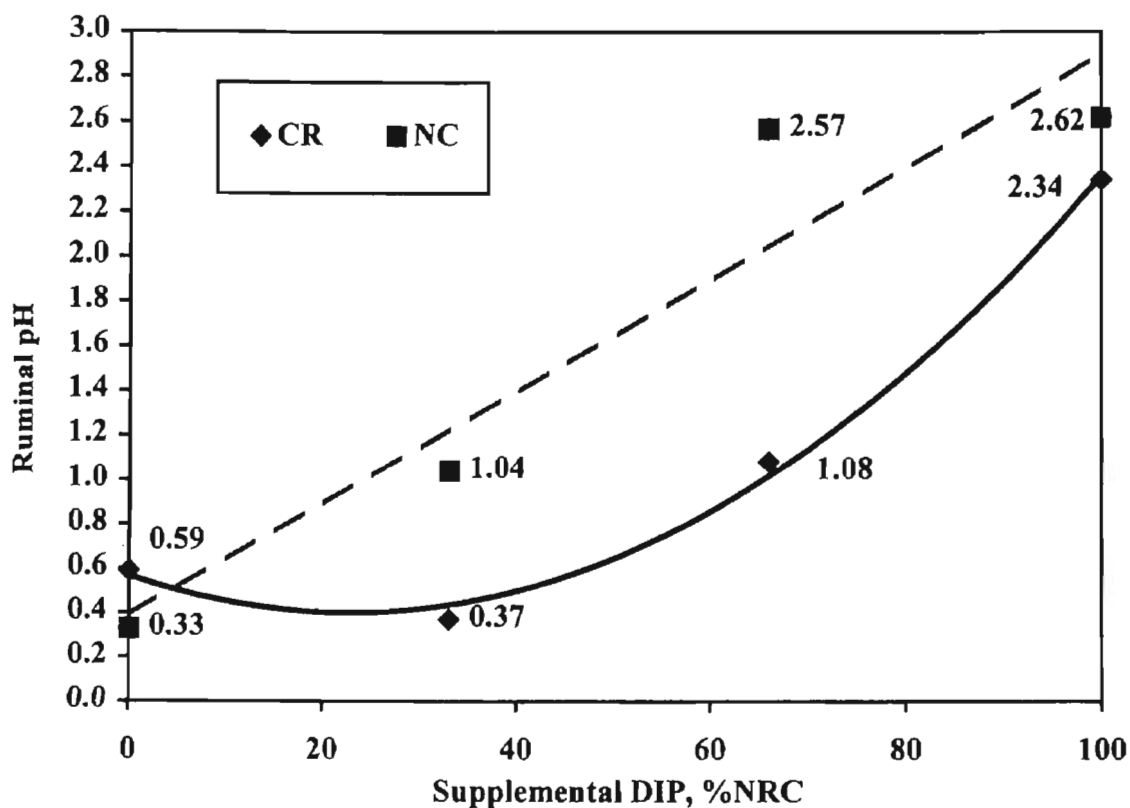


Figure 2. Ruminal pH for 24 h post-supplementation of steers fed prairie hay with supplements containing 0 or .75% BW dry-rolled corn (NC or CR). Means differ ($P < .05$) between level of corn in supplement at each time point.

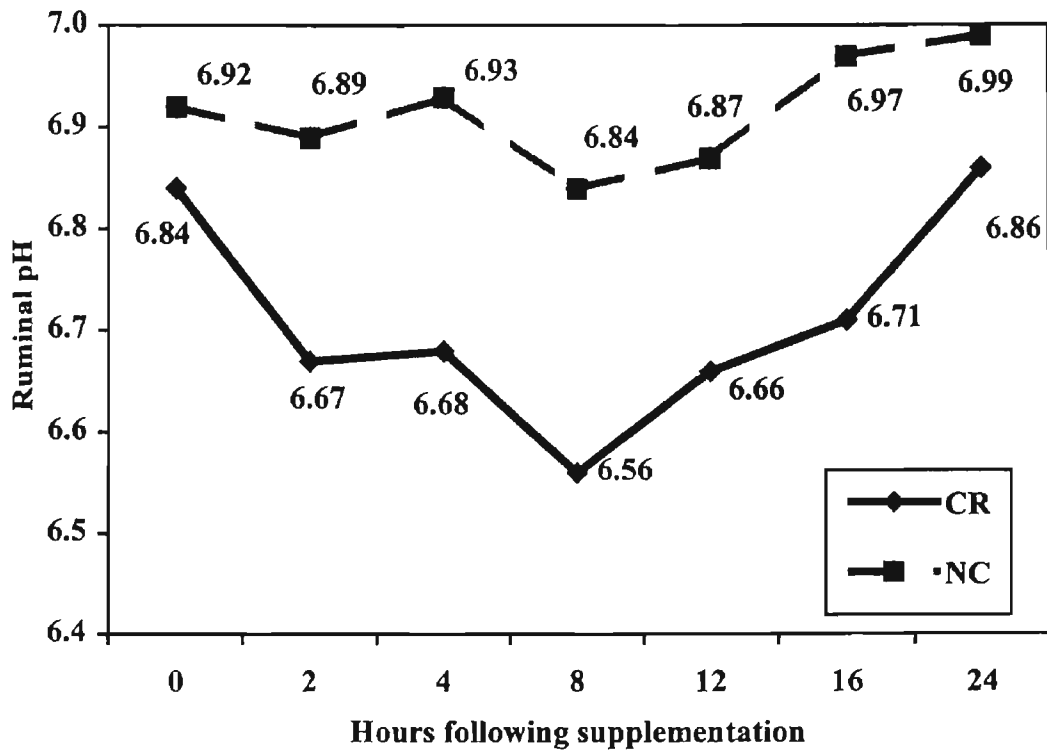


Figure 3. Average ruminal pH of steers fed prairie hay with supplements containing 0 or .75% BW dry-rolled corn (NC or CR) and four increasing levels of DIP (as a percentage of NRC (1996) requirements). Quadratic decreases ($P < .01$) in pH with increasing DIP for NC- and CR-supplemented cattle that are different ($P < .06$) between levels of corn.

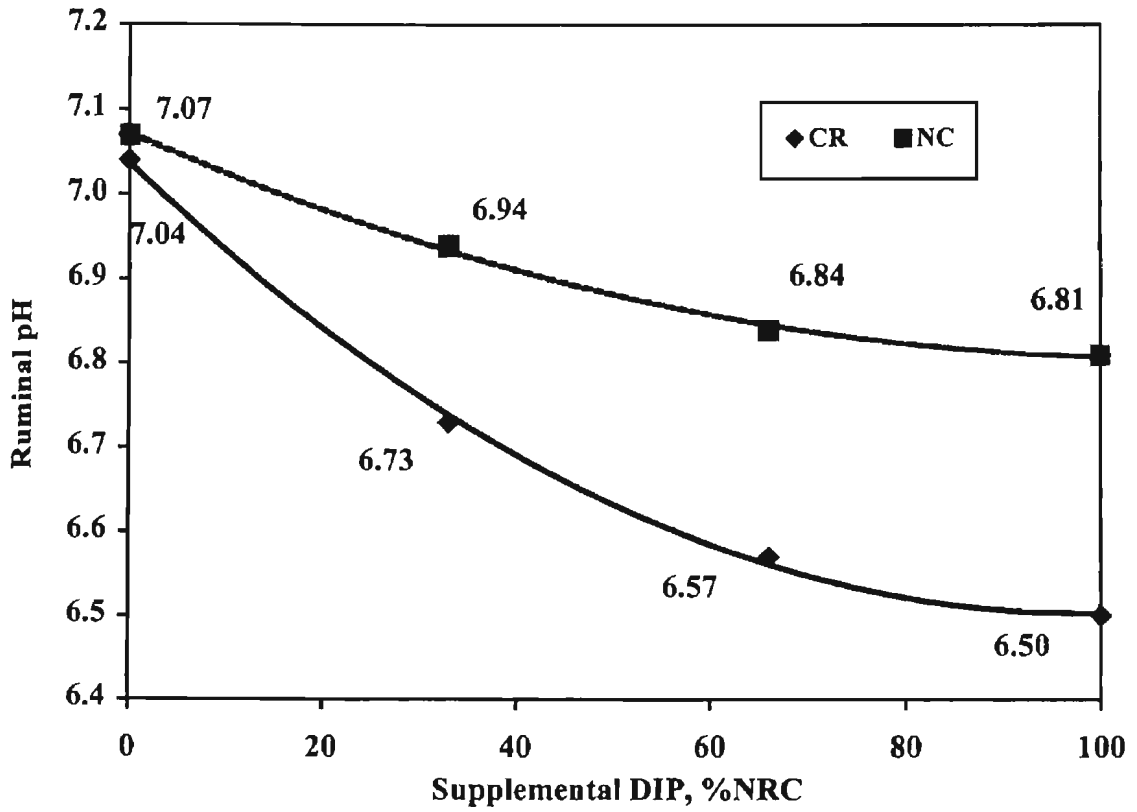


Figure 4. Organic matter digestibility of prairie hay (% of OM intake) fed with supplements containing 0 or .75% BW dry-rolled corn (NC or CR) and four increasing levels of DIP (as a percentage of NRC (1996) requirements). Quadratic increases ($P < .03$) in digestibility with increasing DIP for CR-supplemented cattle, while NC-fed steers exhibited no effect ($P > .43$).

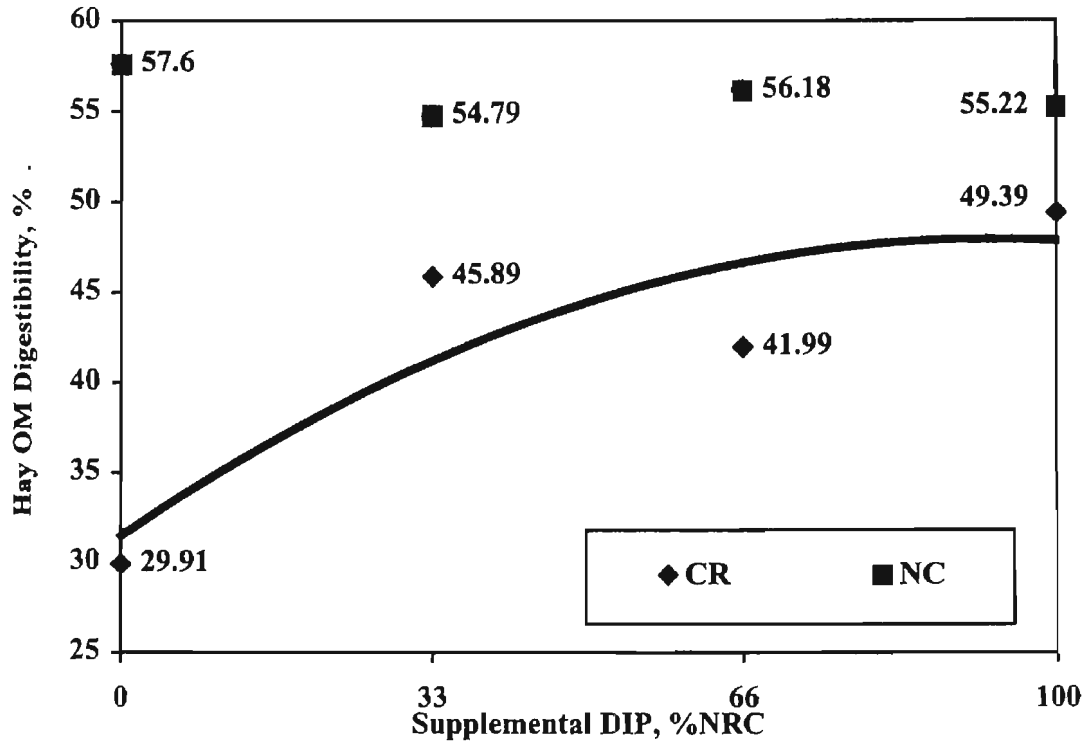


Figure 5. Organic matter intake of prairie hay (g/kg BW) fed with supplements containing 0 or .75% BW dry-rolled corn and four increasing levels of DIP (as a percentage of NRC (1996) requirements). Quadratic increases ($P < .01$) in intake with increasing DIP for NC- and CR-supplemented cattle that are different ($P < .06$) between levels of corn.

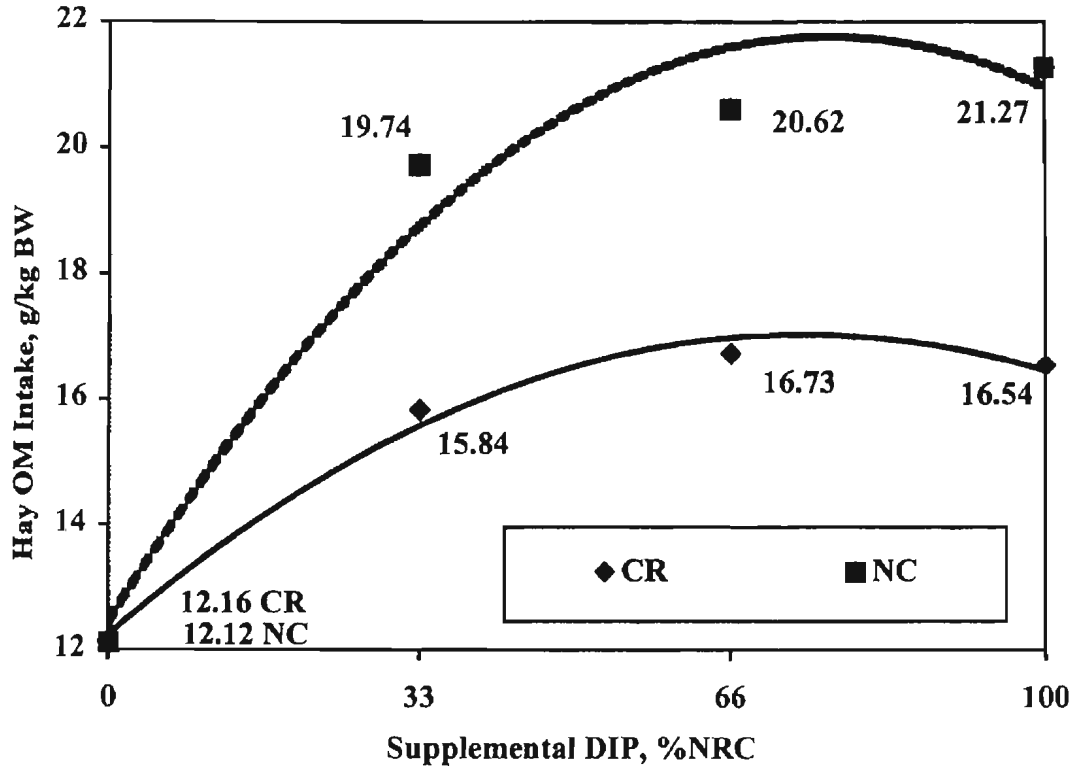
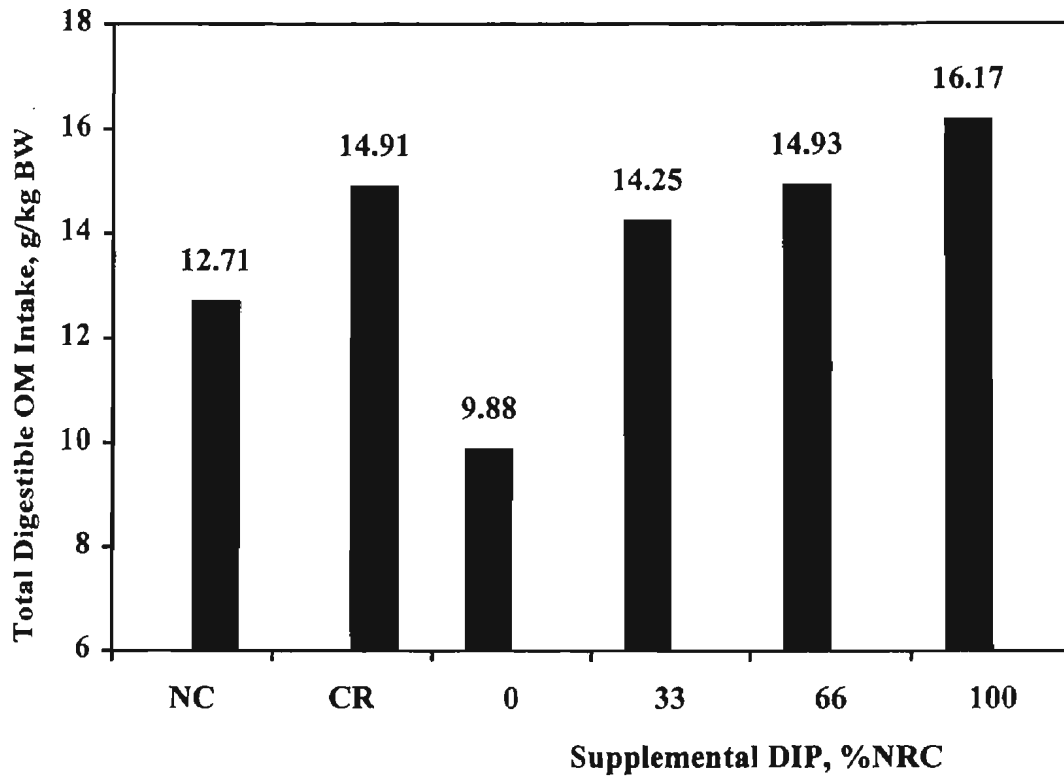


Figure 6. Total digestible organic matter intake (g/kg BW) of prairie hay and supplements containing 0 or .75% BW dry-rolled corn and four increasing levels of DIP. No level of corn by level of DIP interaction ($P > .41$). Cattle fed CR supplements had greater ($P < .01$) intake than NC-fed cattle. Quadratic increases ($P < .01$) in intake with increasing DIP.



Chapter V

Summary and Conclusion

Supplementing cattle consuming low-quality forages is a necessity for many livestock producers. Rates of gain of stocker cattle, body condition of cows and generation of a yearly income from beef cattle production based on a forage resource may require the addition of limiting nutrients via supplementation to maintain economical viability. A greater understanding of the interactions between dietary components, livestock behavior and mechanisms of digestion and metabolism will improve the ability of nutritionists to aid producers in achieving desired production goals in the most efficient manner.

Two experiments were undertaken to evaluate accepted beliefs from previously conducted research regarding supplementation of the two most commonly limiting nutrients in beef production, energy and protein. Common thought would indicate that grain-based supplements can not be used for cattle consuming low-quality forages without running the risk of reducing forage intake and(or) utilization, resulting in a negative associative effect. It also seems to dictate that fibrous by-products will not reduce forage intake or utilization, indicating simple additive effects. Another commonly accepted idea is that protein supplementation will increase forage intake and consequently improve energy and protein status of the animal more than can be explained by the simple additive effects of the individual feedstuffs, resulting in a positive associative effect.

The first experiment was designed to use commercial pelleted supplements similar to those available to livestock producers. The pelleted supplements contained monensin and

were compared to a monensin-containing mineral/vitamin mixture. Ruminally cannulated steers with ad libitum access to low-quality prairie hay were utilized. No differences were noted in forage intake for the mineral treatment or energy supplements formulated from either fibrous by-products or grain sources. Protein supplemented cattle had greater intake of low-quality prairie hay. Total intake of DM and OM were greater for supplemented cattle and not different between supplement sources. Forage digestibility was not different due to treatments, while total diet digestion was greater for the energy supplements.

The second experiment was designed to evaluate the effects of increasing supplemental DIP from soybean meal on low-quality prairie hay diets fed with or without corn to ruminally cannulated steers. The addition of large amount of ruminally fermentable OM to a diet that is deficient in ruminally degradable protein would be expected to exacerbate ruminal ammonia deficiencies and decrease forage fiber digestion, rates of passage and consequently intake and overall energetic status of the animal. In order to investigate this problem, graded levels of supplemental DIP from soybean meal were added to these diets to determine intake, utilization and ruminal parameters of beef steers fed supplements consisting of two levels of corn and four levels of DIP from soybean meal. Interactions between levels of corn and DIP were noted for many variables. However, these interactions were a result of unique responses to added DIP for diets with or without. In general, DIP increased intake and utilization of prairie hay by improving ruminal fermentation and passage. Added grain did not decrease forage intake or utilization when compared to the control without added DIP. Forage intake had a greater response due to supplementation of SBM and no corn, while total diet intake and

intake of digestible OM was greater due to corn supplementation, especially when DIP was adequate.

The results from these intensive digestion and metabolism studies indicate that supply of DIP in relation to total diet digestible nutrients is of much more importance in improving animal response than energy source. It remains to be seen if the observed intake and digestion effects will translate into production improvements in practice. However, it does suggest that the livestock feeding industry should consider DIP levels when formulating supplements for low-quality forage supplements and that the present aversion to grain inclusion is unjustified and based on artifacts of a few research trials where DIP was inadequate for ruminal fermentation of supplement and forage. Feeding grain-based energy supplements should be considered, when economically justified, with the provision of adequate DIP inclusion and the acceptance of greater management requirements.

APPENDIX
ACCESSORY DATA

Table 1. Weight gain, total gain and rate of gain for stocker cattle (233 head) grazed at the Cross Timbers Research Range (Tallgrass) from June 20, 1997 to September 27, 1997 and fed no supplement (CON), a mineral mix (MIN) or 1.13 kg/(steer*day) prorated for 3 feedings per week of a protein supplement (MP), or stocker cattle (164 head) grazed at the Marvin Klemme Range Research Station (Mixed-grass) from June 24, 1997 to October 7, 1997 and fed a mineral mix (MIN), 1.13 kg/(steer*day) prorated for 3 feedings per week of a protein supplement (MP), or a fiber (HF)- or grain (HG)-based energy supplement at 2.26 kg/(steer*day) prorated for 6 feedings per week.

Weights (kg)	Diets				
	CON	MIN	MP	HF	HG
Tallgrass					
<i>In weight (6/20)</i>	316 ^a	288 ^b	290 ^b	---	---
<i>Out weight (9/27)</i>	360 ^a	381 ^b	400 ^c	---	---
<i>Total gain</i>	44 ^a	93 ^b	110 ^c	---	---
<i>Total ADG</i>	.73 ^a	.97 ^b	1.14 ^c	---	---
Mixed-grass					
<i>In Weight (6/24)</i>	---	352	342	340	335
<i>Out weight (10/7)</i>	---	414	410	411	414
<i>Total gain</i>	---	62	68	72	79
<i>Total ADG</i>	---	.62	.67	.71	.78

^{a,b,c}Values within row without like superscripts differ ($P < .05$) between treatments.

Table 2. Pearson correlation coefficients between daily intake of forage DM, daily fecal DM output and apparent total tract digestibility of forage DM for steers fed low-quality prairie hay and supplemented with two levels (0 or .75% BW) of dry-rolled corn and four levels of degradable intake protein (DIP; as a percentage of NRC (1996) requirements).

	Hay DMI	Fecal DM Output	Hay OM Digestion
Hay DMI	---	.5648 <i>P</i> < .0001	.3287 <i>P</i> < .0091
Fecal DM Output	.5648 <i>P</i> < .0001	---	-.5379 <i>P</i> < .0001
Hay OM Digestion	.3287 <i>P</i> < .0091	-.5379 <i>P</i> < .0001	---

Table 3. Pearson correlation coefficients between daily intake of forage DM, weight of ruminal DM contents, weight of ruminal ADF contents, rate of ruminal particulate passage, rate of ruminal OM disappearance and rate of ruminal ADF disappearance for steers fed low-quality prairie hay and supplemented with two levels (0 or .75% BW) of dry-rolled corn and four levels of degradable intake protein (DIP; as a percentage of NRC (1996) requirements).

	Hay DMI	Ruminal DM fill	Ruminal ADF fill	Ruminal Kpp	Ruminal Kt (OM)	Ruminal Kt (ADF)
Hay DMI	---	-.0054 <i>P</i> > .96	-.2080 <i>P</i> < .11	.5439 <i>P</i> < .0001	.5135 <i>P</i> < .0001	.6928 <i>P</i> < .0001
Ruminal DM fill	-.0054 <i>P</i> > .96	---	.9220 <i>P</i> < .0001	-.6723 <i>P</i> < .0001	-.6736 <i>P</i> < .0001	-.6619 <i>P</i> < .0001
Ruminal ADF fill	-.2080 <i>P</i> < .11	.9220 <i>P</i> < .0001	---	-.7788 <i>P</i> < .0001	-.8314 <i>P</i> < .0001	-.7943 <i>P</i> < .0001
Ruminal Kpp	.5439 <i>P</i> < .0001	-.6723 <i>P</i> < .0001	-.7788 <i>P</i> < .0001	---	.9486 <i>P</i> < .0001	.9038 <i>P</i> < .0001
Ruminal Kt (OM)	.5135 <i>P</i> < .0001	-.6736 <i>P</i> < .0001	-.8314 <i>P</i> < .0001	.9486 <i>P</i> < .0001	---	.9230 <i>P</i> < .0001
Ruminal Kt (ADF)	.6928 <i>P</i> < .0001	-.6619 <i>P</i> < .0001	-.7943 <i>P</i> < .0001	.9038 <i>P</i> < .0001	.9230 <i>P</i> < .0001	---

Figure 1. Total diet organic matter intake (g/kg BW) of prairie hay and supplements containing 0 or .75% BW dry-rolled corn (NC or CR) and four increasing levels of DIP (as a percentage of NRC (1996) requirements). Quadratic increases ($P < .01$) in intake with increasing DIP for NC- and CR-supplemented cattle that are different ($P < .06$) between levels of corn.

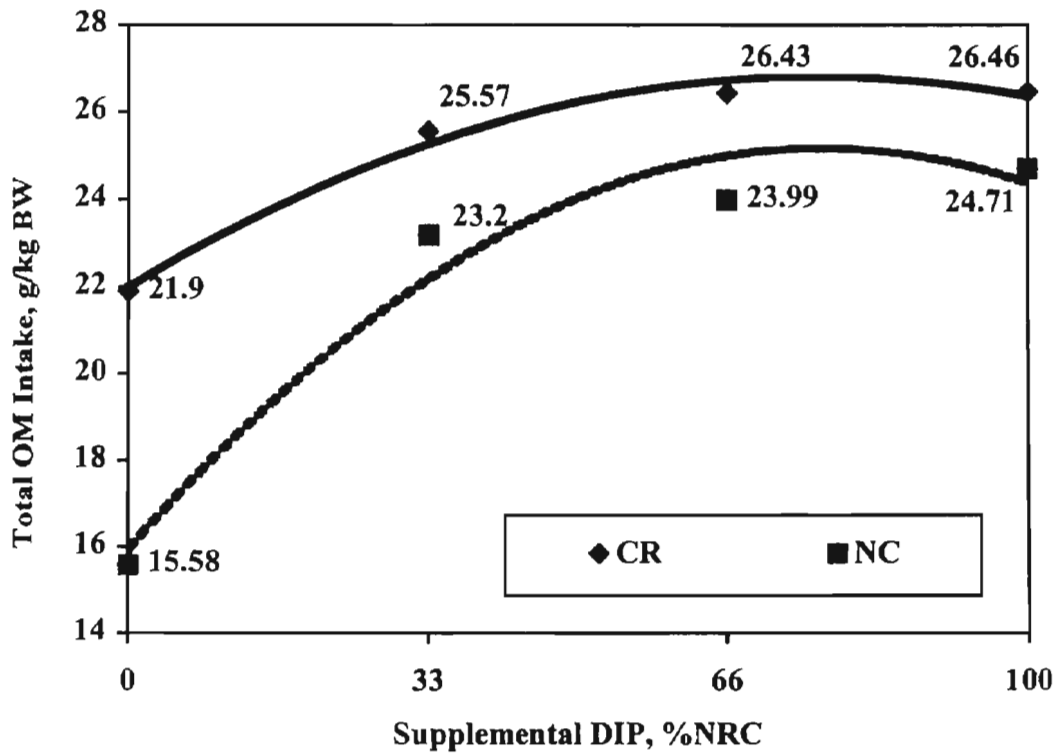
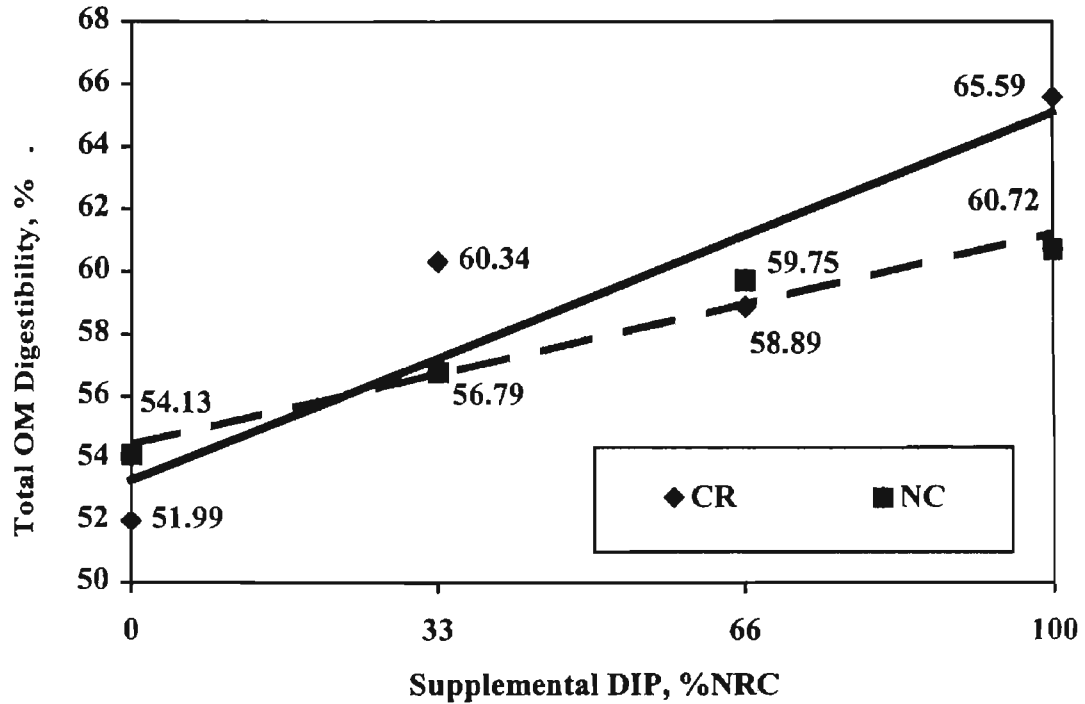


Figure 2. Total diet organic matter digestibility (% of OM intake) of prairie hay and supplements containing 0 or .75% BW dry-rolled corn (NC or CR) and four increasing levels of DIP (as a percentage of NRC (1996) requirements). Linear increases ($P < .01$) in digestibility with increasing DIP for NC- and CR-supplemented cattle that are different ($P < .06$) between levels of corn.



VITA

Timothy Noel Bodine

Candidate for the Degree of

Master of Science

Thesis: SUPPLEMENTATION OF CATTLE FED LOW-QUALITY PRAIRIE
HAY WITH FIBER, STARCH OR PROTEIN

Major field: Animal Science

Biographical:

Personal Data: Born in San Francisco, California, on November 13, 1968, the son of David and Roberta Bodine.

Education: Graduated from Grangeville High School, Grangeville, Idaho in May, 1987; attended Boise State University August, 1988 to May, 1991; received Bachelor of Science degree in Animal Science and Agribusiness with a minor in Agricultural Economics in August, 1997 from the University of Idaho. Completed the requirements for the Master of Science degree with a major in Animal Science at Oklahoma State University in July, 1999.

Professional Experience: Raised on a farm and ranch near Grangeville, Idaho, employed as farm and ranch laborer; auto parts shipping and receiving; custom chemical applicator; ASBSU student body senator; heavy equipment operator; telecommunications installer; landscaper; road construction laborer; logging laborer; agricultural chemical and fertilizer technician; feedmill employee; feed truck driver; graduate research assistant at Oklahoma State University; lab technologist II at Oklahoma State University.

Professional Memberships: Alpha Zeta, American Society of Animal Science, Gamma Sigma Delta, Mortar Board, Phi Kappa Phi, Society for Experimental Biology and Medicine