

THE EFFECT OF MONENSIN, EXTRUDED UREA-GRAIN,  
AND SLOW RELEASE LIQUID SUPPLEMENTS  
ON RANGE BEEF CATTLE

By

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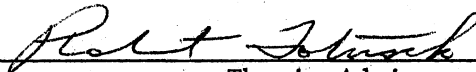
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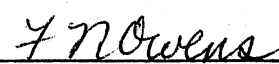
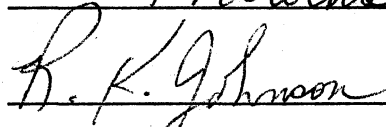
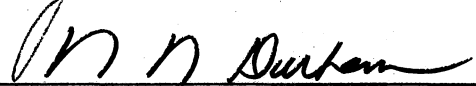
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## CHAPTER I

### INTRODUCTION

In the past several years there has been an increasing demand by livestock producers for an economical way to supplement cattle on dry winter range. This problem has grown more acute recently with the decline in cattle prices and the rise in cost of supplemental protein.

The ruminant possesses a unique digestive tract that enables it to utilize energy and nitrogen sources that are not readily available to non-ruminants. The reticulo-rumen in the ruminant digestive tract supports extensive microbial fermentation and allows the ruminant to utilize energy from cellulose and other plant polysaccharides which are poorly digested by non-ruminants. These plant energy sources are degraded to short chain volatile fatty acids (VFA's) which are absorbed and utilized by ruminant tissues. Monensin, a mycelial product of a yeast, appears to shift the ratio of VFA's produced in the rumen to a more energetically efficient pattern.

The microorganisms in the reticulo-rumen can also utilize dietary non-protein nitrogen (NPN) compounds to synthesize high quality microbial protein. This protein is subsequently digested post-rationally and can be utilized for tissue protein synthesis. Non-protein nitrogen products have been used with variable degrees of success. The amount of NPN which can be utilized by ruminants depends largely on the



availability of fermentable carbohydrate, presence of certain minerals and the level of NPN in the ration.

The purposes of this study were: 1) to compare extruded urea-grain mixtures containing high levels of alfalfa with natural protein mixtures for lactating range cows; 2) to evaluate monensin addition to extruded urea-grain and natural protein supplements for pregnant and lactating cows; and 3) to compare three protein sources -- a slow release liquid urea, natural protein and an extruded urea-grain mixture -- for weaned heifer calves grazing dry winter range grass.

## CHAPTER II

### REVIEW OF LITERATURE

#### Introduction

Nutritionists generally agree that protein is most commonly the limiting nutrient for animal production worldwide. This is especially true of ruminants, primarily because legumes are not grown extensively in many cattle-producing areas. Urea and other non-protein nitrogen (NPN) compounds are available in these areas at relatively low cost and can be used to supplement the diets of ruminants.

Ruminants have evolved an ability to regulate their ruminal environment, which fosters bacteria in the proximal part of the gut called the reticulo-rumen. Pre-gastric fermentation here allows ruminants to obtain energy from ligno-cellulose complexes which are poorly utilized by non-ruminants. In addition, the ruminal microorganisms can utilize NPN compounds to synthesize microbial protein which, in turn, can be converted by the animal to animal protein such as in meat and milk.

In 1879, Weiske et al. discovered that ruminants could convert NPN to protein. During the next 60 years, this subject received considerable attention by German researchers and others. American work on this subject began in 1939 with Hart et al. reporting that either urea or ammonium carbonate was utilized by growing heifers. They also found that soluble dietary carbohydrate was necessary for NPN utilization. This

was the forerunner of a series of experiments having as a common goal the study of the metabolic aspects of NPN utilization by ruminants.

Since that time an extensive amount of research has been conducted with urea and other NPN compounds. The practical value of urea in many beef cattle rations is well documented in experiments reviewed by Reid (1953), McLaren (1964), Briggs (1967), Chalupa (1968), Loosli and McDonald (1968), Smith (1969) and Helmer and Bartley (1971). These reviews also point out that urea occasionally is not a satisfactory supplementary source of nitrogen in beef cattle feeding practices, despite use of all feeding recommendations for successful urea utilization.

#### Urea Utilization

Urea is well utilized in ruminant rations containing high levels of grain. However, researchers have not been as successful in developing protein supplements containing urea for cattle fed poor-quality forages under winter range conditions. Nelson and Waller (1962), Williams et al. (1969), Rush (1974), Wright (1974) all found that urea-containing supplements were inferior to isonitrogenous natural supplements under winter range conditions in Oklahoma.

Urea is used by the ruminant only after microbial protein, synthesized from urea, is degraded in the abomasum and intestinal tract and the amino acids absorbed into the blood are carried to tissues for deposition in protein. The importance of microbial protein to protein nutrition in the ruminant was discussed by Weller et al. (1968, 1962) who found that about 80% of the nitrogen passing into the omasum of sheep was microbial nitrogen.

The amino acid composition of rumen bacteria and protozoa have been studied by Weller (1957), Bergen et al. (1967a) and Meyer et al. (1967). Purser and Buechler (1966) found striking similarity between mixed bacteria and protozoa populations in amino acid composition considering the environmental, experimental, feed and species variables involved. However, Bergen et al. (1967b) found a considerable variation among individual bacterial strains when studying protein quality of individual rumen bacteria using an in vitro enzymatic digestion system. Furthermore, the proportion of essential amino acids released during the digestion of different bacterial strains varied markedly, which suggests that modification of the bacterial population may be an important factor in the nitrogen status of an animal and the animal's response to dietary change. Loosli et al. (1949) discovered the rumen microorganisms were capable of synthesizing all ten essential amino acids in large amounts. Compared to animal needs for amino acids, however, Chalupa (1968), Jacobson et al. (1970) and Oltjen (1969) suggested that one or more amino acids may limit productivity of sheep and cattle. These conclusions are a result of studies of alterations in plasma amino acid profiles associated with urea feeding and of production responses from post ruminal administration of amino acids and protein (Broderick et al., 1970; Hatfield, 1970; Hogan and Weston, 1970; Nimrick et al., 1970a, 1970b; Oltjen et al. 1970; Schelling, 1970; Schelling and Hatfield, 1968).

In most rations, ammonia is an important intermediate in the conversion of food nitrogen to microbial nitrogen. The ammonia production is excessive if large amount of urea are eaten and hydrolyzed rapidly. If the rate of production of ammonia exceeds the rate at which the bacteria can utilize it, the concentration of ammonia in the rumen

rises. This is most evident in rations deficient in readily fermentable carbohydrates such as low starch rations; cellulose energy is released too slowly to match ammonia release rate from urea (Lewis and McDonald, 1958; Lewis, 1962; Hogan, 1964; Christian and Williams, 1966; Oltjen and Putnam, 1966; Purser and Moir, 1966a; Davis and Stallcup, 1967; Deif et al., 1968). Ammonia accumulation is also influenced by the composition of the microbial population. When the protozoal populations are suppressed, ammonia concentrations are reduced (Abou Akkada and el-Shazly, 1964; Christiansen et al., 1965; Luther et al., 1966; Purser and Moir, 1966b; Chalmers et al., 1968). This is probably associated with the concomitant increase in the bacterial population (Eadie and Hobson, 1962) and more efficient utilization of ammonia.

Assimilation of ammonia by rumen bacteria requires the presence or construction of short carbon chains. Intermediates produced during carbohydrate fermentation and fermentation end products plus carbon dioxide and volatile fatty acids are primary sources of carbon for amino acid biosynthesis (Hoover et al., 1963; Allison, 1969; Chalupa, 1968; Tillman and Sidhu, 1969). The greatest efficiency in the utilization of urea nitrogen for the synthesis of microbial protein would be with simultaneous appearance in rumen fluid of ammonia, from the hydrolysis of urea, and carbon skeletons, from the hydrolysis of dietary carbohydrates together with sufficient energy for rapid microbial growth.

The rapid ruminal hydrolysis of urea to ammonia is the primary reason for interest in other NPN compounds. Urea hydrolysis normally occurs at a faster rate than uptake of the liberated ammonia. Ammonia release rates which parallel ruminal VFA production should result in more

efficient utilization of urea nitrogen and maximize microbial protein synthesis.

Starch appears to be the most available native carbohydrate for microbial growth and, thereby, use of ammonia for protein synthesis. The availability of starch can be further increased by gelatinization or cooking (McNaught, 1951; Karr et al., 1965; Meyer et al., 1967; Helmer et al., 1970; Stiles et al., 1970). Cooking starch makes it more susceptible to microbial breakdown (Husted et al., 1968; Osman et al., 1966). Since the rate of energy release from cooked starch more nearly parallels the rate of ammonia release from readily hydrolyzable compounds such as urea, rumen microorganisms should utilize the ammonia more efficiently.

#### Extruded Urea-Grain Mixture

Urea in ruminant rations tends to reduce feed consumption (Huber and Sandy, 1965; Huber and Cook, 1969) and efficiency of nitrogen utilization (Harris and Mitchell, 1941; Harris et al., 1943; Grainger et al., 1960). This is most apparent with rations high in roughage and low in readily available carbohydrates. Deyoe et al. (1968) and Bartley et al. (1968) attempted to overcome these problems by reacting urea and grain under proper heat, moisture and temperature conditions to produce a product they called Starea. This process gelatinizes the starch. Ruminal ammonia levels from Starea are lower than obtained from urea and unprocessed grain fed in equivalent quantities (Stiles et al., 1970). Starea has been reported to be equivalent to soybean meal as a protein supplement for dairy cows (Helmer et al., 1970), feedlot steers (Thompson et al., 1972) and sheep (Shieh-zadeh and Harbers, 1974). However, Tucker and Harbers (1972) and Wright (1974) reported that weight loss of mature

cows was intermediate for Starea-supplemented cattle when compared to an equivalent amount of natural protein and control of an unprocessed milo-urea mixture or a lower protein level.

#### Molasses and Slow Release Urea

Utilization of NPN sources as a protein supplement for cattle grazing dry winter range is relatively poor due to the low energy availability of dry winter range grass. Bohman et al. (1954) conducted growth studies with dairy heifers to determine whether supplemental molasses improved urea use in low quality hay rations. Nitrogen balance data indicated that molasses did not improve the utilization of urea under these conditions. Several studies, however, have shown favorable animal response to various combinations of molasses and urea (Tillman et al., 1951; Evans et al., 1963; and Hussaini et al., 1968) but none of these trials used a control urea ration to permit meaningful comparisons. Related research (Kropp and Johnson, 1974) indicates that utilization of urea can be equivalent to natural protein if it is fed hourly. Since this is an impractical management procedure, the search for a slow release urea product has been intensified.

Slow release of urea in the rumen may aid in prevention of the subacute ammonia toxicity problems (Chalupa et al., 1970) besides stimulating animal performance through improved intraruminal utilization of the urea. Huston (1971) noted that slow release urea in the diet may increase the amount of nitrogen passing into the abomasum as compared with feeding untreated urea. In contrast, Males and Johnson (1974) observed in some of the slow release liquid supplements tested, urea was so tightly bound that it was not hydrolyzed at all.

### Effect of Minerals

The presence of urea does not appear to change the requirements for any mineral, but substitution of urea for intact protein sharply changes the mineral supply for the ruminal bacteria and the host animal. Sulfur is especially important for microbial synthesis of sulfur-containing amino acids and other compounds. Several researchers suggest that the optimum nitrogen:sulfur ratio is approximately 12-15:1 for cattle.

The addition of certain minerals, such as sulfur, to an NPN supplement has been found to be advantageous in vitamin formation, cellulose digestion and nitrogen utilization (Hunt et al., 1954; Barton et al., 1971; Chalupa et al., 1973; and Gil et al., 1973). However, Leibholz (1972) found no sulfur addition was necessary for young calves in Australia.

### Effect of Alfalfa

Researchers have suggested the presence of some factor(s) in feedstuffs aids in the utilization of urea nitrogen by ruminant animals. Horn and Beeson (1969) reported that added dehydrated alfalfa meal enhanced urea utilization by beef steers. Matrone et al. (1964) observed invigorating influence of alfalfa meal on rumen microflora and Lowrey and McCormick (1969) stated that feed consumption and gain were increased by the addition of 5% alfalfa meal to high urea diets. Alfalfa ash has been shown to stimulate cellulose digestion on poor quality roughage and Ellis et al. (1958) suggested that at least part of the stimulatory effect may be due to its content of molybdenum.



### Monensin

Converting plant energy sources to VFA's results in a sizable energy loss in the form of methane, hydrogen and heat as a result of microbial fermentation. Of the three principal VFA's produced in the rumen, propionate can be used by the animal with the highest energetic efficiency.

The predominant VFA normally produced in the rumen is acetate. As the level of concentrate in the ration increases, the percent propionate increases. The digestible energy of a high concentrate ration is utilized more efficiently by the ruminant animal for maintenance and meat production than the digestible energy of a high roughage ration. This difference can partially be explained by the higher percentage of propionate produced in the rumen with a high concentrate ration and the fact that rumen fermentation energy losses are reduced.

Altering the ruminal fermentation so that more propionic acid and less acetic acid are produced by the microorganisms should increase feed efficiency. A feed efficiency increase would be expected since it has been reported that: 1) the propionic acid fermentation is energetically more efficient (Hungate, 1966), 2) propionic acid is utilized by the host animal more efficiently (Smith, 1971), and 3) propionic acid may have a protein sparing effect since propionate is a precursor of glucose (Leng et al., 1967) and normally ruminants obtain some of their glucose from amino acids (Reilly and Ford, 1971).

Monensin is a biologically active compound produced by a strain of Streptomyces cinnamomensis (Haney and Hoehn, 1967). It prevents coccidiosis in poultry and has a moderate in vitro activity against gram-positive organisms. Monensin increases the molar proportion of rumen propionic acid in vitro and in vivo with high grain rations (Raun et al.,

1974b) and increases feed efficiency of cattle fed finishing rations (Raun et al., 1974a).

In vitro experiments have also shown an increase in propionic acid production of 45% when monensin was added at 1.0 ppm (Richardson et al., 1974). This response was consistent with ruminal fluid from either grain-fed cattle or sheep incubated with a high concentrate substrate. Monensin produced a similar increase in molar percentage of propionate when added to rumen fluid from pasture cattle incubated with a high roughage substrate. In vivo experiments carried out by Richardson et al. (1974) and Potter et al. (1974) showed that 200 mg/head/day appeared to be optimal for cattle on a high roughage diet.

## CHAPTER III

### THE EFFECT OF MONENSIN, EXTRUDED UREA-GRAIN,

### AND SLOW RELEASE LIQUID SUPPLEMENTS

### FOR RANGE BEEF CATTLE

#### Summary

Two trials were conducted to evaluate the supplemental value of monensin with Starea and natural (30 and 15%) supplements for beef cows grazing low quality dry winter range grass. A third trial was conducted to evaluate the supplemental value of Starea and slow release liquid supplements for weaned heifer calves.

Cow weight change on monensin supplements averaged over nitrogen levels was not significantly altered by monensin addition ( $P > .05$ ). Cows wintered on 30% natural supplements lost less weight than cows receiving Starea of 15% natural supplements ( $P < .05$ ). Condition loss of cattle followed the same trend as weight loss, with cattle losing the most weight also losing the most condition. Monensin supplemented cows did not differ in ruminal total nitrogen, ammonia, non-ammonia nitrogen or sodium from cows receiving control supplements. However, addition of monensin to supplements decreased by ruminal molar percent of acetate ( $P < .005$ ) and butyrate ( $P < .05$ ), and increased ruminal propionate ( $P < .005$ ) and potassium ( $P < .005$ ).

Heifer weight loss during a 56 day wintering trial was lowest for heifers supplemented with Starea ( $P < .05$ ) and greater for heifers supplemented with the slow release liquid supplement.

### Introduction

Altering ruminal fermentation so that more propionic acid and less acetic acid is produced by the microorganisms should increase feed efficiency. A feed efficiency increase would be expected since it is reported that: 1) propionic acid fermentation is energetically more efficient (Hungate, 1966), 2) propionic acid is utilized by the host animal more efficiently (Smith, 1971), and 2) propionic acid may have a protein sparing effect as a precursor of glucose (Leng et al., 1967).

Monensin has been shown to increase the molar proportion of rumen propionic acid in vitro and in vivo with a high grain rations (Raun et al., 1974a) and increase feed efficiency of cattle fed finishing rations in the feedlot (Raun et al., 1974b). With cattle on pasture, as well, Potter et al. (1974) observed increased molar proportions of propionate and average daily gains of cattle on pasture. In vivo experiments conducted by Richardson et al. (1974) and Potter et al. (1974) suggest that 200mg/head/day is optimal for cattle fed high roughage diets.

The purpose of this study was: 1) to evaluate the addition of monensin to extruded urea-grain and natural protein supplements for pregnant and lactating cows; 2) to compare the utilization of an extruded urea-grain containing alfalfa with natural protein supplements for cows, and 3) to compare a slow release liquid supplement containing urea with natural protein and extruded urea-grain mixtures for weaned heifer

calves. All animals were grazing dry winter range grass during the experiments.

### Experimental Procedure

Three winter trials were conducted in Central Oklahoma on native tall-grass range with climax vegetation of little bluestem (Andropogon scoparius), big bluestem (Andropogon gerardi), Indian grass (Sorghastrum nutans) and switch grass (Panicum virgatum). Ingredient makeup of experimental supplements fed in the trials are shown in Table I. The nitrogen:sulfur ratio for all supplements was approximately 12:1. Initial and final weights and condition scores appraised visually were obtained after a 12 hour shrink. A condition score of 1 to 9 was placed on each individual cow with 1 being the thinnest and 9 the fattest rating.

#### Trial 1

Seventy-eight mature Angus and Hereford cows were randomly allotted, after blocking by breed and breeding date, to six treatments for a 107 day wintering trial. The six supplemental treatments are 1 through 6 in Table I. Treatments were: 30% natural crude protein supplements with and without monensin; and 30% crude protein supplements (with one-half the protein equivalent coming from Starea 44) with and without monensin. Supplement was fed at a rate of 1.14kg/head/day six days per week initially and increased to 1.48kg/head/day for the remainder of the trial. Monensin was fed at a calculated level of 200mg/head/day. The analyzed amounts of monensin provided per day were 177.8, 197.7 and 181.0 for 30%, 15% and Starea supplements, respectively.

TABLE I  
INGREDIENT MAKEUP OF PROTEIN SUPPLEMENTS  
(PERCENT)

Item	International Reference Number	1 Natural, 0	2 30% 200	3 Natural, 0	4 15% 200	5 Starea, 0	6 30% 200	7 Natural, 0	8 30% 200
Crude protein, % <sup>a</sup>		30.69	32.07	15.21	18.24	31.32	32.66	29.59	29.64
Dry matter, %		87.82	88.47	88.05	88.36	85.96	88.26	91.79	92.01
Corn, dent, grain gr 2 US mn 54 wt., (4)	4-02-915	22.77	27.77	68.75	68.75	---	---	27.77	27.77
Soybean, seed, solv-extd. grnd, mx 7 fibr., (5)	5-04-604	58.25	58.25	17.25	17.25	12.40	12.40	58.25	58.25
Alfalfa, hay S-C grnd, stemmy, (1)	1-99-118	5.00	5.00	5.00	5.00	32.80	32.80	5.00	5.00
Sugarcane molasses, mn 48 invert sugar mn 79.5 degrees brix, (4)	4-04-696	5.00	5.00	5.00	5.00	5.00	5.00	5.00	5.00
Sodium phosphate, monobasic NaH <sub>2</sub> PO <sub>4</sub> H <sub>2</sub> O, cp, (6)	6-04-287	2.50	2.50	2.75	2.75	4.35	4.35	2.50	2.50
Calcium phosphate, dibasic commercial, (6)	6-01-080	0.75	0.75	1.20	1.20	---	---	0.75	0.75

TABLE I (Continued)

Item	International Reference Number	1 Natural, 0	2 30% 200	3 Natural, 0	4 15% 200	5 Starea, 0	6 30% 200	7 Natural, 0	8 30% 200
Sodium sulfate Na <sub>2</sub> SO <sub>4</sub> 10 H <sub>2</sub> O, cp, (6) <sup>b</sup>	6-04-292	0.68	0.68	---	---	1.40	1.40	0.68	0.68
Trace mineral mix		0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05
Vitamin A palmitate, comm, (7) <sup>c</sup>	7-05-143	+	+	+	+	+	+	+	+
Starea 44 <sup>d</sup>		---	---	---	---	44.00	44.00	---	---

<sup>a</sup>Crude protein as determined by Kjeldahl procedure on dry matter basis.

<sup>b</sup>Formulated to supply 12:1 nitrogen:sulfur ratio.

<sup>c</sup>22,000 IU per kg of supplement.

<sup>d</sup>Starea 44, 4.5% moisture, 46.1% protein, 12.5% urea, 97% starch damage determined by alpha-amylase procedure.

The 30% and 15% natural crude protein supplements were the positive and negative controls, respectively. All cows were allowed to graze in a common pasture and individual fed their respective supplement in individual stalls six mornings per week. Feed refusals were recorded daily and minor intake adjustments were made by periodically providing an extra feeding on the seventh day to equalize supplement intake across all treatments. Cows calved from September 5th to February 1st, with a mean calving date of October 26th. Calving commenced before the trial began and was completed before the trial ended. Because the number of cows calved before the trial was not equal across all treatments, initial weight of the cows that had calved before the start of the trial were adjusted to a non-pregnant weight basis. This was done by using a regression equation derived from data obtained from trials in which cows were accurately weighed prior to and after calving and the calves were weighed at birth (Ewing et al., 1966, unpublished data). This equation was used to adjust the initial winter trial weights of the cows which had not calved to a calved basis.<sup>2</sup>

$$\text{Adjusted initial weight} = \text{actual initial weight} - \sqrt{(\text{calf birth weight} \times 1.9697) - 19.07}$$

*non-pregnant wt basis.*

Rumen samples were obtained from five randomly selected cows per treatment on day 84 of the trial for volatile fatty acid determinations. Rumen fluid was sampled a second time from eight randomly selected cows per treatment on day 98 of the trial for determination of certain mineral and nitrogen parameters. Samples were taken by rumen tube with a screen developed by Raun and Burroughs (1962). Rumen samples on day 84 were taken an average of 4½ hours post-supplement feeding for VFA analysis and microbial action was stopped by adding 5gm phosphoric acid-meta



analytical reagent per 50ml rumen fluid. Samples on day 98 were taken after cows were fed supplement and allowed to graze  $3\frac{1}{2}$  hours before sampling. Microbial action in this case was stopped by adding 2ml of saturated mercuric chloride per 60ml of rumen fluid. Volatile fatty acids were determined by the procedure of Erwin et al. (1961), rumen ammonia by Kjeldahl distillation over magnesium oxide (A.O.A.C., 1960) and total nitrogen by the Kjeldahl procedure. Sodium and potassium were determined by Flame Spectrophotometry.

Cows' weight and condition changes were analyzed as a randomized block with a 3 x 2 factorial arrangement of treatments; all other parameters were analyzed as a completely randomized design.

### Trial 2

Seventy-six mature Hereford cows were used in a 140 day trial. Cows were blocked by weight and expected calving date and randomly allotted to two treatment groups with two replications. The cows were placed on four pastures and rotated among pastures at 14 day intervals to minimize pasture and location effects. The 2 treatments consisted of supplements 7 and 8 in Table I. Cows were fed 1.36kg/head/day of range cubes six days per week consisting of 30% natural crude protein supplement with or without added monensin at a calculated level of 200mg/head/day. Subsequent analysis indicated that the amount provided was 210.4mg/head/day.

Cows calved from February 2nd to May 19th, with a mean calving date of March 23rd. Because the number of cows which had not calved by the end of the trial was not equal among treatments, the final weight of the cows that had not calved were adjusted to a calved basis by using the equation of Ewing et al. (1966).

Rumen samples were taken on day 84 and 97 of the trial to determine the effect of monensin on proportions of acetate, propionate and butyrate. Rumen samples on day 84 were taken an average of 4½ hours post-supplement feeding and on day 97 immediately preceding daily supplementation. Preservation and analytical procedures were the same as in Trial 1.

### Trial 3

Twenty-four yearling heifers were used in a 56 day trial. Heifers were blocked by weight and allotted to four treatments. Four supplemental protein sources were compared; 30% natural crude protein, 15% natural crude protein, 30% crude protein with one-half the protein equivalent coming from Starea 44 and a 30% crude protein from a slow release liquid supplement. The composition of the supplements is shown in Table I. The supplements were self-fed with consumption regulated by including salt in the meal supplements and by tying the wheel on the lick tank for the liquid supplement as necessary to limit intake.

Heifer weight and condition changes were analyzed as a randomized block experiment.

## Results and Discussion

### Trial 1

Cow performance results are shown in Table II. Average daily supplement intakes were approximately equal on all treatments for the trial. Palatability of both the Starea and Starea + monensin supplements were lower than the natural protein supplements. There were no apparent

TABLE II  
 PERFORMANCE OF COWS DURING WINTER SUPPLEMENTATION  
 IN TRIAL 1 (107 DAYS)

Item	Protein source and monensin level, mg/head/day						S.E. <sup>b</sup>
	Natural, 30%		Natural, 15%		Starea, 30%		
	0	200	0	200	0	200	
Cows, number	13	13	13	13	13	13	
Ave. Daily supplement, kg	1.05	1.05	1.05	1.05	1.02	1.03	
Daily C.P. intake, kg <sup>c</sup>	0.32	0.33	0.16	0.19	0.32	0.34	
Ave. calving date	Oct. 27	Oct. 28	Oct. 23	Oct. 27	Oct. 20	Oct. 28	
Initial cow wt., kg	540.4	450.7	450.5	450.5	450.5	450.7	
Adjusted cow wt. loss, kg	97.9 <sup>d</sup>	105.6 <sup>d</sup>	147.4 <sup>f</sup>	135.1 <sup>ef</sup>	117.1 <sup>e</sup>	131.3 <sup>e</sup>	4.96
Initial cow condition score <sup>i</sup>	5.69	5.62	6.08	5.62	5.69	5.85	
Condition score change <sup>j</sup>	-2.08 <sup>d</sup>	-1.92 <sup>d</sup>	-3.69 <sup>h</sup>	-2.92 <sup>f</sup>	-2.38 <sup>e</sup>	-3.31 <sup>g</sup>	0.25

<sup>a</sup>Starea to furnish 50% of total crude protein equivalent.

<sup>b</sup>Standard error of means.

<sup>c</sup>Dry matter basis.

<sup>d,e,f,g,h</sup>Means with different superscripts are significantly different (P < .05).

<sup>i</sup>Based on a scale of 1 to 9, 1 the thinnest and 9 the fattest.

<sup>j</sup>Difference in initial and final condition.

palatability problems with the natural protein supplements with or without monensin.

Cows consuming the 30% natural supplements, with and without monensin, lost about 18% less weight than cows receiving Starea, with or without monensin, and about 28% less weight than cows fed the 15% natural protein supplements, with and without monensin ( $P < .05$ ). Cow weight losses were about 3% greater with monensin supplementation averaged over nitrogen levels ( $P > .05$ ). Cows on Starea supplements were intermediate between cows on 30% natural and 15% natural supplements when averaged over monensin ( $P < .05$ ) suggesting that the nitrogen from Starea was about 65% as well utilized as the 30% natural protein supplement. This is in agreement with Rush (1974) and Wright (1974). Response to the 30% protein supplements indicates a need for a higher level of available supplemental protein than provided by either the Starea or 15% natural supplements.

Cows fed 30% natural protein supplements lost less condition than cows on the other supplements ( $P < .05$ ). Condition loss then followed an order of Starea, 15% natural + monensin, Starea + monensin and 15% natural supplements with differences significant statistically ( $P < .05$ ). These results indicate that monensin decreased condition loss on the 15% natural supplement but increased condition loss on the Starea supplement ( $P < .05$ ). In general, condition loss paralleled weight losses.

Total and molar percentages of volatile fatty acids are shown in Table III. Averaged over supplements, monensin decreased acetate ( $P < .005$ ) and butyrate ( $P < .05$ ) and increased propionate ( $P < .005$ ). Total molar concentration was not different across all treatments ( $P > .10$ )

TABLE III

TOTAL MOLAR PERCENTAGES OF VOLATILE FATTY ACIDS  
IN RUMEN FLUID OF COWS IN TRIAL 1

Item	Protein source and monensin level, mg/head/day					
	Natural, 30%		Natural, 15%		Starea, 30%	
	0	200	0	200	0	200
Acetate, <sup>f</sup> molar %	72.48 ± 1.74 <sup>bc</sup>	70.14 ± 1.56 <sup>cd</sup>	73.04 ± 1.56 <sup>bc</sup>	67.69 ± 1.56 <sup>d</sup>	75.19 ± 1.56 <sup>b</sup>	66.17 ± 1.74 <sup>d</sup>
Propionate, <sup>f</sup> molar %	20.12 ± 1.67 <sup>cd</sup>	23.89 ± 1.50 <sup>bc</sup>	19.76 ± 1.50 <sup>cd</sup>	25.45 ± 1.50 <sup>b</sup>	18.48 ± 1.50 <sup>d</sup>	28.38 ± 1.67 <sup>b</sup>
Butyrate, <sup>e</sup> molar %	7.41 ± 0.53 <sup>b</sup>	5.97 ± 0.47 <sup>bc</sup>	7.20 ± 0.47 <sup>b</sup>	6.85 ± 0.47 <sup>bc</sup>	6.34 ± 0.47 <sup>bc</sup>	5.45 ± 0.53 <sup>c</sup>
Total conc., mM/l	35.14 ± 6.31	49.59 ± 5.64	32.52 ± 5.64	28.56 ± 5.64	46.09 ± 5.64	43.83 ± 6.31

<sup>a</sup>Values are least square means ± standard deviation.

<sup>b,c,d</sup>Means with different superscripts are significantly different (P < .05).

<sup>e</sup>Main effect of monensin statistically significant (P < .05).

<sup>f</sup>Main effect of monensin statistically significant (P < .005).

but were not influenced by protein source. These results agree with those of Potter et al. (1974) and Raun et al. (1974a).

Ruminal sodium, potassium and nitrogen parameters are shown in Table IV. Dry matter of ruminal contents was higher ( $P < .01$ ) for monensin supplemented cows than for control cows.

Total nitrogen content of rumen fluid did not differ consistently across protein sources or with monensin addition. Rumen ammonia concentrations were higher for cows fed Starea and 30% natural supplements than for cows fed 15% natural supplements. This again indicates the need for a positive and negative control to evaluate the effect of NPN supplements. The results reported here with the natural supplements do not concur with the results of experiments conducted by Eli Lilly and Company (L. H. Carroll, personal communication) in which decreased rumen ammonia was observed when monensin was fed. Differences between cows fed different supplements did not influence non-ammonia nitrogen and sodium in rumen fluid ( $P > .05$ ). The concentrations of sodium reported here are in agreement with those found in experiments conducted by Bailey (1961).

Potassium in rumen contents did not differ ( $P > .05$ ) between protein supplement sources. However, averaged over nitrogen sources, monensin was associated with about a 10% increase in ruminal potassium ( $P < .005$ ). Potassium represents an important fraction of the cation content of the rumen fluid essential in maintaining a desirable medium for bacterial fermentation. Hubbert et al. (1958) have shown that potassium is essential for cellulose digestion in an in vitro system. Maintenance of osmolarity with plasma is important in maintaining a desirable moisture content of the rumen fluid (Balch and Johnson, 1950; and Micholson et al., 1960). Therefore, it is possible that cows receiving monensin may have

TABLE IV  
RUMINAL MINERAL AND NITROGEN PARAMETERS OF COWS  
IN TRIAL 1

Item	Protein source and monensin level, mg/head/day						S.E. <sup>e</sup>
	Natural, 30%		Natural, 15%		Starea, 30%		
	0	200	0	200	0	200	
Dry matter, %	1.96 <sup>c</sup>	2.42 <sup>a</sup>	2.02 <sup>bc</sup>	2.29 <sup>ab</sup>	2.18 <sup>abc</sup>	2.40 <sup>ab</sup>	0.14
Total nitrogen, mg N/100ml	58.6 <sup>abc</sup>	64.8 <sup>a</sup>	52.2 <sup>bc</sup>	49.6 <sup>c</sup>	58.4 <sup>abc</sup>	61.7 <sup>ab</sup>	4.34
Rumen ammonia mg NH <sub>3</sub> /100ml	6.3 <sup>bc</sup>	8.1 <sup>ab</sup>	3.2 <sup>c</sup>	3.8 <sup>c</sup>	11.2 <sup>a</sup>	8.0 <sup>ab</sup>	1.19
Non-ammonia nitrogen, mg NAN/100ml	52.3	56.7	49.0	45.8	47.2	53.7	4.21
Sodium, ppm	3000	2843	2878	2640	2547.38	3318	458.00
Potassium, ppm	464 <sup>bcd</sup>	595 <sup>a</sup>	444 <sup>bcd</sup>	502 <sup>bc</sup>	413 <sup>d</sup>	508 <sup>b</sup>	26.94

a,b,c,d Means with different superscripts are significantly different (P < .05).

<sup>e</sup>Standard error of means.

an increased bacterial fermentation efficiency as shown by the decreased molar percent of acetate and butyrate and increased molar percent propionate. The higher dry matter content of rumen fluid from cows supplemented with monensin could also be due to increased fermentation and slower ruminal turnover rate. Ruminal mineral and nitrogen parameter correlations for all animals on all treatments are shown in Table V. The results show that total ruminal nitrogen and ruminal potassium are significantly correlated ( $P < .005$ ) suggesting again that bacterial fermentation increases as potassium increases. The VFA, potassium, total nitrogen and dry matter of rumen fluid reported here support the theory of Eli Lilly and Company that monensin does cause a shift in microbial populations.

### Trial 2

Response of cows on Trial 2 were similar to those of Trial 1 (Table VI). Changes in weight or condition of cows receiving the 30% natural supplement were not significantly different from those of cows fed the 30% natural + monensin supplement ( $P > .05$ ).

VFA samples collected on cows prior to supplement feeding (Table VII) indicate there were no differences in acetate, propionate or butyrate concentrations due to protein supplement. Cows fed monensin sampled  $4\frac{1}{2}$  hours post-feeding were lower in acetate and butyrate ( $P < .05$ ) and higher in propionate ( $P < .05$ ) on the monensin supplement.

### Trial 3

Results of Trial 3 are shown in Table VIII. Daily intakes were different on the four supplements. Heifers on the natural supplements



TABLE V  
CORRELATIONS OF RUMINAL MINERAL AND  
NITROGEN PARAMETER FOR COWS  
IN TRIAL 1

Item	Rumen ammonia, mg NH <sub>3</sub> -N/100ml	Non-ammonia nitrogen mg NAN/100ml	Na, ppm	K ppm
Total nitrogen, mg N/100ml	0.2478*	0.6164**	0.0464	0.5016**
Rumen ammonia, mg NH <sub>3</sub> -N/100 ml		-0.1247	-0.0308	-0.0088
Non-ammonia nitrogen, mg NAN/100ml			0.0180	0.0327
Na, ppm				0.1819

<sup>a</sup>Correlations are based on 48 observations.

\*Approaching significance (P < .10).

\*\*Significant (P < .005).

TABLE VI  
 PERFORMANCE OF COWS DURING WINTER  
 SUPPLEMENTATION IN TRIAL 2  
 (140 DAYS)

Item	Monensin, mg/head/day		S.E. <sup>a</sup>
	0	200	
Cows, number	38	38	
Ave. daily supplement, kg <sup>b</sup>	1.25	1.25	
Daily crude protein intake, kg <sup>b</sup>	0.37	0.37	
Ave. calving date	Mar. 25	Mar. 22	
Initial cow wt., kg	444.92	447.79	
Adjusted cow wt. loss, kg	88.64	89.19	4.2
Initial cow condition score <sup>c</sup>	5.66	5.45	
Condition score changed <sup>d</sup>	-1.76	-1.90	0.18

<sup>a</sup>Standard error of means.

<sup>b</sup>Dry matter basis.

<sup>c</sup>Based on scale of 1 to 9, 1 the thinnest and 9 the fattest.

<sup>d</sup>Difference in initial and final condition.

TABLE VII  
 TOTAL MOLAR PERCENTAGES OF VOLATILE FATTY  
 ACIDS IN RUMEN FLUID OF COWS IN  
 TRIAL 2

Item	Monensin, mg/head/day		S.E. <sup>c</sup>
	0	200	
Sampled 22 hr. post-supplement feeding			
Acetate, molar %	73.44	73.38	2.66
Propionate, molar %	20.40	21.09	2.64
Butyrate, molar %	6.16	5.53	0.60
Total conc., mM/l	42.28	39.92	6.31
Samples 4½ hr. post-supplement feeding			
Acetate, molar %	76.26 <sup>a</sup>	68.46 <sup>b</sup>	0.43
Propionate, molar %	16.08 <sup>b</sup>	25.68 <sup>a</sup>	0.31
Butyrate, molar %	7.65 <sup>a</sup>	5.86 <sup>b</sup>	0.20
Total conc., mM/l	38.66	36.04	3.81

<sup>a, b</sup>Means with different superscripts are significantly different ( $P < .05$ ).

<sup>c</sup>Standard error of means.

TABLE VIII  
 PERFORMANCE OF HEIFERS DURING WINTER  
 SUPPLEMENTATION IN TRIAL 3  
 (56 DAYS)

Item	Protein supplements				S.E. <sup>c</sup>
	30% Natural	15% Natural	Starea <sup>a</sup>	Liquid <sup>b</sup>	
Heifers, number	8	8	8	8	
Daily non-salt supplement intake, kg	0.49	0.49	0.44	0.38	
Daily supp. crude protein intake, kg	0.15	0.07	0.13	0.11	
Initial wt., kg	199.4	199.4	198.9	199.2	
Body wt. loss, kg	32.7 <sup>e</sup>	36.4 <sup>ef</sup>	26.1 <sup>d</sup>	39.2 <sup>f</sup>	2.14
Initial heifer condition score <sup>g</sup>	4.9	4.9	4.8	5.0	
Condition score change <sup>h</sup>	-2.1 <sup>f</sup>	-2.6 <sup>ef</sup>	-3.0 <sup>de</sup>	-3.5 <sup>d</sup>	0.25

<sup>a</sup>Starea to furnish 50% of total crude protein equivalent.

<sup>b</sup>Cargill's slow release liquid supplement containing 30% protein equivalent.

<sup>c</sup>Standard error of means.

<sup>d,e,f</sup>Means with different superscripts are significantly different ( $P < .05$ ).

<sup>g</sup>Based on a scale of 1 to 9, 1 the thinnest and 9 the fattest.

<sup>h</sup>Differences in initial and final condition.

had the highest daily supplement intake (.49kg) and heifers consuming the liquid supplement (.38kg) the lowest daily intake, with Starea supplemented (.44kg) heifers intermediate. Palatability problems appeared in heifers fed the Starea and liquid supplements as the trial progressed. This is in agreement with results found in experiments conducted by Rush (1974) and Wright (1974).

Starea supplemented heifers lost the least amount of weight during the 56 days ( $P < .05$ ). Heifers fed the natural supplements were not different ( $P > .05$ ) from each other but 30% natural supplemented heifers lost less weight than liquid supplemented heifers ( $P < .05$ ). Bohman et al. (1954) observed that molasses was a poor carbohydrate for supplementing urea when cattle were fed little or no starch. In vitro experiments conducted by Males and Johnson (1974) suggested that urea in some slow release supplements was tightly bound and poorly utilized. The results reported here suggest that either the carbohydrate from molasses is not adequate, or the NPN was not hydrolyzed by the rumen microbial population well enough to synthesize amino acids.

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**APPENDIX**

**TABLES**

TABLE IX  
ANALYSIS OF VARIANCE FOR COW WEIGHT LOSS  
(TRIAL 1)

Source of variation	df	Mean square	F value
Block	12	422.6032	1.3251
Treatment	5	3190.3602	10.0032*
Nitrogen	2	7658.4631	24.0127*
Monensin	1	6.6222	0.0208
Nitrogen x monensin	2	314.1264	0.9849
Block x Treatment	60	318.9335	---

\*P < .005

TABLE X  
ANALYSIS OF VARIANCE FOR COW CONDITION LOSS  
(TRIAL 1)

Source of variation	df	Mean square	F value
Block	12	1.8611	2.2345
Treatment	5	6.6051	7.9302***
Nitrogen	2	11.7820	14.1458***
Monensin	1	0.0513	0.0616
Nitrogen x monensin	2	4.7051	5.6490*
Block x Treatment	60	0.8329	---

\*P < .025  
\*\*P < .01  
\*\*\*P < .005

TABLE XI  
ANALYSIS OF VARIANCE FOR ACETATE (TRIAL 1)

Source of variation	df	Mean square	F value
Nitrogen	2	2.1392	0.0177
Monensin	1	214.4023	17.7002*
Nitrogen x monensin	2	24.9121	2.0566
Error	22	12.1130	---

\*P < .005

TABLE XII  
ANALYSIS OF VARIANCE FOR PROPIONATE (TRIAL 1)

Source of variation	df	Mean square	F value
Nitrogen	2	4.5602	0.4065
Monensin	1	288.4640	25.7137*
Nitrogen x monensin	2	22.0100	1.9697
Error	22	11.2183	---

\*P < .005



TABLE XIII  
ANALYSIS OF VARIANCE FOR BUTYRATE (TRIAL 1)

Source of variation	df	Mean square	F value
Nitrogen	2	3.1332	2.7968*
Monensin	1	5.4920	4.9022**
Nitrogen x monensin	2	0.7066	0.6307
Error	22	1.1203	---

\*P < .10  
\*\*P < .05

TABLE XIV  
ANALYSIS OF VARIANCE FOR TOTAL VFA  
CONCENTRATION (TRIAL 1)

Source of variation	df	Mean square	F value
Nitrogen	2	565.8862	3.5556*
Monensin	1	51.9102	0.3262
Nitrogen x monensin	2	236.1857	1.4840
Error	22	159.1549	---

\*P < .10

TABLE XV  
ANALYSIS OF VARIANCE FOR DRY MATTER IN RUMEN  
CONTENTS (TRIAL 1)

Source of variation	df	Mean square	F value
Nitrogen	2	0.0746	0.4947
Monensin	1	1.2352	8.1910*
Nitrogen x monensin	2	0.6600	0.4377
Error	42	0.1508	---

\*P < .01

TABLE XVI  
ANALYSIS OF VARIANCE FOR TOTAL NITROGEN IN  
RUMEN CONTENTS (TRIAL 1)

Source of variation	df	Mean square	F value
Nitrogen	2	548.0000	3.6291*
Monensin	1	64.0000	0.4238
Nitrogen x monensin	2	82.0000	0.5430
Error	42	151.0000	---

\*P < .05

TABLE XVII  
ANALYSIS OF VARIANCE FOR RUMEN AMMONIA (TRIAL 1)

Source of variation	df	Mean square	F value
Nitrogen	2	150.9877	13.3450*
Monensin	1	0.7752	0.0685
Nitrogen x monensin	2	27.1940	2.4035
Error	42	11.3142	---

\*P < .005

TABLE XVIII  
ANALYSIS OF VARIANCE FOR RUMEN NON  
AMMONIA-NITROGEN (TRIAL 1)

Source of variation	df	Mean square	F value
Nitrogen	2	205.3698	1.4509
Monensin	1	78.8738	0.5572
Nitrogen x monensin	2	105.4813	0.7452
Error	42	141.5476	---

TABLE XIX  
ANALYSIS OF VARIANCE FOR RUMEN SODIUM (TRIAL 1)

Source of variation	df	Mean square	F value
Nitrogen	2	151881.06	0.0905
Monensin	1	189003.00	0.1126
Nitrogen x monensin	2	1257157.56	0.7491
Error	42	1678134.60	---

TABLE XX  
ANALYSIS OF VARIANCE FOR RUMEN POTASSIUM  
(TRIAL 1)

Source of variation	df	Mean square	F value
Nitrogen	2	21594.271	3.7181*
Monensin	1	108300.000	18.6472**
Nitrogen x monensin	2	5329.188	0.9176
Error	42	5807.833	---

\*P < .05  
\*\*P < .005

TABLE XXI  
ANALYSIS OF VARIANCE FOR COW WEIGHT LOSS  
(TRIAL 2)

Source of variation	df	Mean square	F value
Block	19	4714.6199	1.4441
Treatment	1	28.2395	0.0086
Block x treatment	19	3114.9544	0.9315
Error	36	3343.8848	---
Pooled error	55	3264.7998	---

TABLE XXII  
ANALYSIS OF VARIANCE FOR COW CONDITION LOSS  
(TRIAL 2)

Source of variation	df	Mean square	F value
Block	19	2.1018	1.8028*
Treatment	1	0.3810	1.3268
Block x treatment	19	1.2168	1.0684
Error	36	1.1389	---
Pooled error	55	1.1658	---

\*P < .05

TABLE XXIII

ANALYSIS OF VARIANCE FOR ACETATE 22 HOURS  
POST-SUPPLEMENT FEEDING (TRIAL 2)

Source of variation	df	Mean square	F value
Treatment	1	0.0102	0.0002
Pasture w/i treatment	2	42.4891	6.4763*
Animal w/i pasture	8	6.5607	---

TABLE XXIV

ANALYSIS OF VARIANCE FOR PROPIONATE 22 HOURS  
POST-SUPPLEMENT FEEDING (TRIAL 2)

Source of variation	df	Mean square	F value
Treatment	1	1.4283	0.0342
Pasture w/i treatment	2	41.7161	7.9736*
Animal w/i pasture	8	5.2318	---

\*P < .025

TABLE XXV

ANALYSIS OF VARIANCE FOR BUTYRATE 22 HOURS  
POST-SUPPLEMENT FEEDING (TRIAL 2)

Source of variation	df	Mean square	F value
Treatment	1	1.1970	0.5588
Pasture w/i treatment	2	2.1419	11.6788*
Animal w/i pasture	8	0.1834	---

\*P < .005

TABLE XXVI

ANALYSIS OF VARIANCE FOR TOTAL VFA CONCENTRATION  
22 HOURS POST-SUPPLEMENT FEEDING (TRIAL 2)

Source of variation	df	Mean square	F value
Treatment	1	16.7437	0.0700
Pasture w/i treatment	2	239.2313	3.1818*
Animal w/i pasture	8	75.1879	---

\*P < .10

TABLE XXVII

ANALYSIS OF VARIANCE FOR ACETATE 4½ HOURS  
POST-SUPPLEMENT FEEDING (TRIAL 2)

Source of variation	df	Mean square	F value
Treatment	1	304.7462	165.7941*
Pasture w/i treatment	2	0.9972	0.5425
Animal w/i pasture	16	1.9432	---
Pooled error	18	1.8381	---

\*P &lt; .005

TABLE XXVIII

ANALYSIS OF VARIANCE FOR PROPIONATE 4½ HOURS  
POST-SUPPLEMENT FEEDING (TRIAL 2)

Source of variation	df	Mean square	F value
Treatment	1	460.0323	466.9904*
Pasture w/i treatment	2	0.0132	0.0134
Animal w/i pasture	16	1.1066	---
Pooled error	18	0.9851	---

\*P &lt; .005



TABLE XXIX

ANALYSIS OF VARIANCE FOR BUTYRATE 4½ HOURS  
POST-SUPPLEMENT FEEDING (TRIAL 2)

Source of variation	df	Mean square	F value
Treatment	1	15.9490	38.7300*
Pasture w/i treatment	2	0.8216	1.9951
Animal w/i pasture	16	0.3606	---
Pooled error	18	0.4118	---

\*P < .005

TABLE XXX

ANALYSIS OF VARIANCE FOR TOTAL VFA CONCENTRATION  
4½ HOURS POST-SUPPLEMENT FEEDING (TRIAL 2)

Source of variation	df	Mean square	F value
Treatment	1	34.3833	0.2364
Pasture w/i treatment	2	198.3109	1.3636
Animal w/i pasture	16	138.8164	---
Pooled error	18	145.4269	---

TABLE XXXI

ANALYSIS OF VARIANCE FOR HEIFER WEIGHT LOSS  
(TRIAL 3)

Source of variation	df	Mean square	F value
Block	7	62.5138	1.7097
Treatment	3	254.9824	6.9735*
Block x treatment	21	36.5644	---

\*P &lt; .005

TABLE XXXII

ANALYSIS OF VARIANCE FOR HEIFER CONDITION LOSS  
(TRIAL 3)

Source of variation	df	Mean square	F value
Block	7	0.3393	0.6868
Treatment	3	2.7083	5.4824*
Block x treatment	21	0.4940	---

\*P &lt; .10

VITA<sup>N</sup>

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