Effect of Different Levels of Rumensin in Diet on Rumen Fermentation, Nutrient Digestibility and Methane Production in Cattle

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ABSTRACT : Twelve rumen fistulated cross-bred calves were divided into three groups and fed wheat straw and concentrate mixture according to their maintenance requirement. Animals in group II and III were fed 50 and 100mg rumensin per day, in addition to basal feed. Supplementation of rumensin in the diet decreased the dry matter intake significantly ($p<0.05$) along with a significant decrease in the straw intake. Digestibility coefficients of all the nutrients were not affected significantly except that of CF digestibility which was lower ($p<0.05$) in groups II and III as compared to group I. Among N-parameters in the rumen fluid, mean NH_3-N was significantly lower in groups II and III (19.13 and 18.63 mg N/100 ml respectively) than in group I (22.68); total-N and TCA-ppt-N did not differ among the three groups. Total VFA concentration did also not differ among the three groups, however, propionate increased from 24.33 molar % to 32.73 while acetate and butyrate decreased respectively from 65.85 to 58.81% and 9.79 to 8.46%. Total VFA, bacteria and protozoa production rates were not affected significantly due to rumensin in diet. Methane production per kg DDM as well as % of methane in total gas were reduced at both the levels of rumensin on different concentrate ratios with wheat straw as roughage. Similar trend was also observed with rice straw and concentrate mixture as substrate with rumensin addition. (*Asian-Aus. J. Anim. Sci. 1999. Vol. 12, No. 8 : 1215-1221*)

Key Words : Cattle, Rumensin, Rumen Fermentation, Methane

INTRODUCTION

Scarcity of good quality feeds has led to either improvement of poor quality feeds or manipulation of fermentation pattern in the rumen. Manipulation of rumen fermentation is possible either by supplementation of nutrients or by feed additives like ionophores. Originally used as anticoccidial feed additives for poultry, ionophores are produced by various strains of *Streptomyces* (Bergen and Bates, 1984) and include monensin, lasalocid, salinomycin, narasin and rumensin. Addition of ionophores in high concentrate feed has resulted in increased propionic acid (Thornton et al., 1976), depression in methanogenesis (Thornton and Owens, 1981) decreased proteolysis and deamination of dietary proteins (Chalupa, 1980) and improvement in feed conversion efficiency. Therefore, experiments were conducted to study the effect of two levels of rumensin in a straw based diet on rumen fermentation, nutrient utilization and methanogenesis.

MATERIALS AND METHODS

Experiment I

Twelve rumen fistulated cross-bred calves (273-311 kg) were divided into three groups of four in a randomised block design. The animals were housed in clean, dry and airy byres in such a way that they should not have access to another animal's diet. The byres and the animals were washed and cleaned daily to remove feaces and dirt to maintain hygienic conditions. The animals were fed once a day in the morning, first the concentrate mixture and then wheat straw. Clean drinking water was provided twice a day. Vitablend (Vitamin A supplement of Glaxo India Ltd.) was given weekly in the drinking water to meet vitamin A requirement.

The animals were fed wheat straw as basal roughage and concentrate mixture (groundnut cake 27.5, maize 30, wheat bran 40, mineral mixture 2 and salt 0.5 parts) to meet the maintenance requirement (NRC, 1984). In group II and III the animals were given rumensin 50 and 100 mg/d, respectively.

After preliminary feeding of 27 d, 7 d metabolic trial was conducted. Daily samples of feed offered, residue, urine and faeces were collected for the estimation of proximate principles (AOAC, 1984). Rumen liquor (RL) samples were collected with the help of stainless steel probes having holes covered with nylon cloth and placed at different parts in the rumen to obtain composite samples at different time intervals during the metabolic trial.

Total volatile fatty acids

Single dose isotope dilution technique was followed (Chaturvedi et al., 1973). 1,2-^{14}C sodium acetate was infused into the rumen through a cannula and samples of RL were taken at 2, 2.5, 3, 3.5, 4, 5 and 7 h post infusion. Total VFA concentration was estimated and radio activity was counted with a scintillation counter. Production rate of TVFA was calculated from the dose infused and the rate of decline in radio activity assuming first order kinetics. Volatile fatty acid

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Received November 11, 1998; Accepted March 24, 1999

fractions were estimated by gas liquid chromatography using a column filled with chromosorb 101 (Erwin et al., 1961).

Bacterial production rate

The method described by Singh et al. (1974) was used to estimate bacterial production rates. Bacteria were labelled with ^{35}S sodium sulphate and were infused into the rumen. After 2 h of infusion RL samples were collected at 120, 150, 180, 210, 240, 300, 360 and 420 min. Bacteria were separated and dissolved in toluene. The radio activity in the samples was counted and production rates were calculated.

For N-parameters, RL samples were collected during two hourly feeding to get the steady state condition in the rumen. Seven samples were collected starting from the morning at 09:00 at one hour intervals. Total N, NPN, TCA-ppt-N (AOAC, 1984) and NH_3-N (Conway, 1962) were estimated in these samples.

Protozoa production rate

Protozoa production rate in the animals was determined as per the method of Leng (1982) using ^{14}C-methyl choline after about 45 days of TVFA production estimation. Rumen liquor was incubated with ^{14}C-Choline for 90 min to label protozoa. Labelled protozoa were then separated and infused into the rumen of experimental animals. After 2h of infusion, hourly samples of rumen liquor were collected up to 9h. The collected samples were processed immediately for separation of protozoa. Half of this sample was kept for measuring radio activity and half was used for N determination. From the decline in radio activity with time, pool size and protozoa production rates were calculated as :

$$\text{Pool size} = \frac{\text{Dose infused}}{\text{Radio activity at zero time}}$$

$$\text{Production rate} = \text{Pool} \times m$$

Where, m = the rate of decline in radio activity.

Experiment II

The cross-bred calves used in experiment I (12 animals) were divided into two groups of 6 each. Each group was further divided into 3 subgroups of 2 each. All the animals were fed for maintenance as in experiment I.

One group of animals was fed wheat straw (1) and the other group was fed rice straw (2) as basal roughage. In the subgroups, concentrate mixture and straw were fed in the ratio of 25:75 (1A, 2A) 50:50 (1B, 2B) and 75:25 (1C, 2C). Feeding was done for 20 days and then rumen liquor was taken for methane production estimation.

After preliminary feeding, RL was collected to study *in vitro* methane production from the animals of each subgroup. Half gram quantities of substrate (table 1), being the feeds given to the animals, were incubated with 10 ml rumen liquor and 40 ml McDougal buffer in 100 ml air tight syringe for 24 h. After incubation, total gas in the syringes was measured. Saturated sodium hydroxide was sucked into the syringe which absorbed CO_2 and the amount of gas left inside represented the methane. Due care was taken to subtract trace gases. Thus methane produced with all feed combinations was estimated and methane produced per kg DDM was calculated by estimating DMD of the sample. Data were analysed statistically (Snedecor and Cochran, 1968) and expressed as means with standard error (SE).

Table 1. Substrates used in experiment II

Treatment	Concentrate	Straw
	Wheat Straw	
1	0	100
1A	25	75
1B	50	50
1C	75	25
Concentrate	100	0
	Rice Straw	
2	0	100
2A	25	75
2B	50	50
2C	75	25

1= Wheat straw. 2= Rice straw.
A, B and C=Ratios of concentrate and straw.

RESULTS

Experiment I

Chemical composition of the feeds is presented in table 2 and feed intake is given in table 3. It was seen that total DMI was depressed in groups II and III though it was not significant statistically ($p>0.05$). This depression was significant ($p<0.05$) when DMI was expressed as kg per 100 kg BW, however no significant difference was observed between groups II and III. Wheat straw intake was also significantly lower in rumensin fed groups as compared to control. Digestibility coefficients are also shown in table 3. No significant differences were observed for DM, OM, CP, EE and NFE digestibilities; that of CF was significantly ($p<0.05$) higher in control group as compared to treatment groups.

Total N (mg/100ml SRL) as well as TCA-ppt-N

Table 3. Dry matter intakes and digestibility coefficients of various nutrients for animals given different levels of rumensin

	Groups		
	I	II	III
Body weight (kg)	291±15.72	295±9.86	298±15.18
Feed intake			
Concentrate (kg)	1.98	2.02	2.02
Wheat straw* (kg)	4.48±0.90[a]	3.82±0.89[b]	3.79±0.95[b]
Total (kg/d)	6.46±0.87	5.84±1.00	5.81±0.60
Total *(kg/100 kg BW)	2.22±0.08[a]	1.98±0.18[b]	1.95±0.35[b]
Digestibility coefficient			
DM	55.13±1.29	54.79±2.17	54.12±0.88
OM	58.65±0.97	58.10±2.13	57.88±1.02
CP	61.48±2.46	60.45±3.17	60.78±3.48
FE	48.76±2.58	49.45±1.78	48.88±2.78
CF*	55.21±4.08[a]	49.77±1.25[b]	48.98±3.18[b]
NFE	68.34±4.87	69.28±3.82	68.57±5.92

Figures bearing different superscripts in a row differ significantly. * $p<0.05$.
Values are expressed as means with standard error.
Group I = Control; Group II = Control + 50 mg rumensin; Group III = Control+ 100 mg rumensin.

did not differ significantly among the groups (table 4). The values ranged from 126.81 to 130.23, and 76.12 to 79.44. Ammonia-N values were significantly lower in groups II (19.13) and III (18.63) as compared to group I (22.68). However, blood urea did not vary significantly among the three groups and the values were in the physiological limits.

TVFA concentrations and production rates

There was no significant difference in TVFA concentration in the RL among all the groups (table 5). While acetate percentage decreased ($p<0.05$), the propionate percentage increased significantly ($p<0.01$) in experimental groups as compared to control group. However, TVFA production rate (mol/d) did not differ significantly among the three groups (table 6). The values were 12.11 (gp I), 11.98 (gp II) and 12.08 (gp III).

Table 2. Chemical composition of feeds

	Concentrate	Wheat straw
DM	98.50	97.44
OM	91.58	87.86
CP	18.08	3.62
CF	4.58	35.92
EE	4.47	1.25
NFE	65.95	49.63
Total Ash	6.92	9.58

Table 4. Effect of feeding rumensin on the nitrogenous constituents (mg/100ml) in rumen fluid

	Groups		
	I	II	III
Total-N	126.81±4.6	130.23±5.89	128.53±5.96
Ammonia-N	22.68±1.11[a]	19.13±0.70[b]	18.63±0.43[b]
TCA-ppt-N	78.32±6.68	79.44±3.88	76.12±5.36
NPN	48.49±4.18	50.79±2.80	52.41±7.53
Blood urea (m/100ml)	14.18±1.03	15.31±0.89	14.10±0.46

Figure bearing different superscripts in a row differ significantly. * $p<0.05$.
TCA-ppt-N = Trichloroacetic acid precipitatable nitrogen.
Values are expreesed as means with standard error.
Group I = Control; Group II = Control + 50mg rumensin; Group III = Control + 100 mg rumensin.

Table 5. TVFA concentration and molar percentage of individual VFAs

Groups	TVFA (mg/100ml RL)	Molar %		
		Acetate*	Propionate**	Butyrate
I	11.16±0.63	65.85±1.32[a]	24.33±0.99[a]	9.79±0.86
II	12.01±0.48	58.81±1.05[b]	32.73±1.81[b]	8.46±0.30
III	11.52±0.78	58.86±0.27[b]	32.29±0.58[b]	8.85±0.58

Figures bearing different superscripts in a row differ significantly. * $p<0.05$. ** $p<0.01$.
Values are expressed as means with standard error.
Group I = Control, Group II = Control + 50mg rumensin, Group III = Control + 100mg rumensin.

Table 6. Effect of feeding rumensin on VFA, and bacterial and protozoal production rates

	Groups		
	I	II	III
VFA production rate			
VFA pool (Mol)	4.08± 0.04	4.17±0.12	3.98±0.08
VFA produciton rate (Mol/d)	12.11± 1.12	11.98±0.88	12.08±1.17
Bacterial production rate			
Bacterial pool (g)	101.48± 6.84	99.86±9.53	100.32±3.86
Bacterial prodction rate (g/d)	248.63±11.43	250.10±8.87	249.44±7.03
Protozoa production rate			
Protozoal pool (N)	8.44± 0.46	9.08±0.57	8.68±0.69
Protozoal production rate (g N/d)	10.17± 0.78	11.23±1.12	10.88±1.70

Values are expressed as means with standard error.
Group I = Control; Group II = Control + 50 mg rumensin; Group III = Control + 100 mg rumensin.

Table 7. Gas production with various additions of rumensin to wheat straw and concentrate mixture

Rumensin (mg/d)	Total gas (L/kg DDM)	CH_4 (L/kg DDM)	% CH_4 in Total gas
Wheat Straw			
0	108.0±2.08	40.1±0.72	38.8±0.66
50	100.9±1.50	32.2±0.51	31.9±1.01
100	98.9±1.39	30.5±0.35	31.0±0.29
Conc. 25:WS75			
0	114.1±2.44	41.1±0.64	33.2±0.32
50	124.1±0.70	35.4±0.30	28.5±0.42
100	122.1±1.89	31.8±0.55	26.1±0.46
Conc. 50:WS50			
0	110.5±0.32	33.8±0.40	30.6±0.47
50	110.5±0.49	21.8±0.86	21.5±0.30
100	117.9±0.26	21.7±0.32	18.5±0.25
Conc. 75:WS25			
0	108.7±1.20	28.7±0.58	26.5±0.40
50	107.2±0.35	22.3±0.23	22.5±0.21
100	105.1±0.72	19.7±0.26	18.7±0.35
Conc. 100:WS0			
0	110.7±0.55	28.6±0.47	25.6±0.35
50	111.4±0.57	18.9±0.21	17.1±0.11
100	125.6±0.44	16.6±0.35	13.2±0.23

Conc. = Concentrate, WS = Wheat Straw.

Table 8. Gas production with various additions rumensin to rice straw and concentrate mixture

Rumensin (mg/d)	Total gas (L/kg DDM)	CH_4 (L/kg DDM)	% CH_4 in Total gas
Rice Straw			
0	109.7±0.55	42.1±0.55	38.4±0.61
50	109.7±0.68	35.8±0.32	32.6±0.26
100	111.2±0.79	32.7±0.35	29.4±0.75
Conc. 25:RS75			
0	111.3±0.90	33.6±0.47	34.6±0.59
50	108.6±0.44	33.2±0.72	30.5±0.53
100	107.3±0.45	30.7±0.61	28.6±1.00
Conc. 50:RS50			
0	112.3±0.46	33.6±0.26	29.9±0.38
50	111.5±0.44	25.9±0.57	23.2±0.68
100	107.3±0.50	22.9±0.86	21.3±0.80
Conc. 75:RS25			
0	108.7±0.45	27.5±0.41	25.3±0.76
50	108.1±0.61	23.7±0.58	21.9±0.15
100	110.3±0.70	20.6±0.25	18.6±0.45

Conc. = Concentrate, RS = Rice Straw.

Bacterial and protozoal production rates

Production rates of bacteria (in liquid phase) and protozoa are given in table 6. Bacterial pool size (g) in the rumen was 101.48, 99.86 and 100.32 in groups I, II and III, respectively and production rates (g/d) were 248.63, 250.10 and 249.44 in the three groups, respectively. These values did not differ significantly among the groups. Protozoal production values as well as pool size did also not differ significantly among the three groups. Thus feeding of 50 and 100 mg rumensin per day per animal did not affect the VFA, bacteria and protozoa production rates in rumen fed maintenance diet of concentrate and wheat straw.

Experiment II

Methane production from wheat straw and from rice straw with different proportions of concentrate mixture are depicted in table 7 and 8. Total gas (L/kg DDM) from wheat straw and concentrate combinations ranged from 98.90 (only straw) to 125.60 (concentrate). However, no significant ($p<0.01$) difference was observed either with increase in the concentrate level or with addition of rumensin. Methane (L/kg DDM) decreased from 40.10 (wheat straw) to 28.60 (concentrate); methane % in total gas also reduced consistently with the increase in the concentrate level. Rumensin also had a significant decreasing effect on methane production at all the levels of concentrate

mixture. Similarly, it also affected methane production when rice straw was used as substrate. Percent reduction in methane increased with increase in level of rumensin supplementation with wheat straw (18-40%) while with rice straw rumensin had a similar effect at all levels of concentrate (28-29%). The methane produced ranged from 42.10 (only straw) to 20.60 (with 75:25, concentrate: straw and 100 mg rumensin). Only concentrate supplementation decreased methane production from 42.10 to 27.50 L/kg DDM.

DISCUSSION

DMI and digestibilities of nutrients

No significant difference was observed in the total DMI by the animals of different groups in this study though there was a decrease in DMI per unit BW due to rumensin feeding which significantly decreased by 9.6 to 10% the wheat straw intake. Several workers in had also reported a decrease in feed intake on feeding monensin or lasalocid (Davis and Erhat, 1976; Raun et al., 1976; Faulkner et al., 1985 and Stock et al., 1995). The reduction in feed intake could be related to increased concentration of ruminal propionate (Theurer et al., 1974) which increases the availability of energy from the same amount of feed. It varied with the type of feed as diets containing considerable linked carbohydrates depicted less prominent depression. In one study Goodrich et al. (1984) observed rather contradictory results in which there was increase in forage intake by 13%. There was no effect on nutrient digestibilities except that of CF which decreased on rumensin feeding in this experiment because rumensin affects cellulolytic bacteria. In most of the earlier studies, no effect on DM and OM digestibility was observed (Dinius et al., 1976 and Beever et al., 1987). However a decrease in DM and ADF digestibilities was reported by Lemenager et al. (1978), and some studies have shown increased digestibility of DM and decreased protein degradation in the rumen (Thornton and Owens, 1981; Whetstone et al., 1981 and Goodrich et al., 1984). All this could be due to the changes in fermentation pattern at microbial level in rumen.

Rumen fermentation

While studying rumen nitrogen parameters, no significant change was observed in total-N as well as in TCA-ppt-N except a significant decrease in NH_3-N due to feeding of rumensin. This might be because of less protease and deaminase activity in experimental groups. Decrease in NH_3-N concentration was also reported by several other workers (Dinius et al., 1978 and Ricke et al., 1984). Poos et al. (1979) reported a lower decrease with urea supplemented diet (38.4 to 26.6 mg/100ml RL) as compared to a grain supplemented diet (18.7 to 5.8 mg/100 ml) showing the effect of monensin not only on deamination but on protease activity also. However, Bogart et al. (1991) and Rogers et al. (1991) reported up to 53% less NH_3-N in monensin treated cows.

Total VFA concentration did not vary among the groups, while there was significant increase in propionate proportion and decrease in the proportion of acetate and butyrate in the rumen of animals fed rumensin. Effect of level of rumensin in the diet was not apparent. In all the studies conducted so far, increase in propionate proportion was evident with no change in total VFA concentration. A shift towards more propionate led to decrease in both acetate and butyrate concentrations (Raun et al., 1976; Whetstone et al., 1981; Goodrich et al., 1984; Ricke et al., 1984 etc). Increased propionate production could lead to better feed utilization and improved feed conversion efficiency. Production rates of VFA did not differ significantly as a result of rumensin in the diet.

There are several studies which reported a change in microbial population and type of microbes which led to decrease in microbial growth and microbial protein synthesis (Dennis et al., 1981; Faulkner et al., 1985 and Haimoud et al., 1995). However, no significant change occured in this study in bacterial production rates due to rumensin feeding. Similarly, protozoal production also did not vary, though a slight decrease in protozoal number was reported in some studies (Poos et al., 1979)

Experiment II

In vitro, total gas as well as methane produced with wheat straw and concentrate mixture (in various proportions) is depicted in table 7. Effect of rumensin on all the combinations showed a significant ($p>0.01$) effect on methane production which did not increase with the increase in the level of rumensin. Total gas produced was not affected significantly with the increase of level of concentrate mixture. Supplementation with rumensin decreased total gas from straw only because of its antibacterial (cellulolytic) action on already available less number of microbes. It had a positive effect when concentrate mixture was also present in the feed because of its effect on some selected bacteria like *Ruminococcus albus, R. flavifaciens* and *Butyrivibrio fibrisolvens*. (Chen and Wolin, 1979). Methane percentage decreased up to 30% on 75% concentrate mixture diet as compared to 100% wheat straw diet. This decrease was further enhanced on rumensin addition from 18% with only wheat straw to 40% when wheat straw and concentrate mixture were in the ratio of 50:50. The opposite trend was observed by Thornton and Owens (1981) who showed that monensin decreased methane production by 16% at a lower roughage level and by 24% at a

higher roughage level.

Depending upon the *in vitro* methane production values with and without rumensin and on different ratios of straw and concentrate, actual production by the animals was calculated. In *in vivo* experiment the animals consumed the ration in the ratio of 30 : 70 in group I and 35 : 65 in groups II and III. Total gas (L/kg DDM) produced on these ratios was not significantly different among the three groups (110.0, 111.0 and 100.5) while methane decreased significantly (37.8, 29.0 and 26.9 L/kg DDM) in group II and III. The decrease was 23% in group II and 28% in groups III. Group II and III did not differ significantly from each other.

In table 8, total gas and methane values are shown with rice straw as roughage and concentrate mixture in different ratios. Total gas produced was not affected significantly on addition of concentrate mixture or rumensin in the straw diet, however methane was reduced up to 34% on concentrate mixture supplementation up to 75% in the diet. Rumensin addition caused further a decrease up to 28-29% with straw only as well as with various proportions of concentrate and rice straw. The decrease in methane production observed may be because of the inhibition of the organisms that produce formic acid and hydrogen as reported by Chen and Wolin (1979) and Dennis et al. (1981). However, Delfino et al. (1988) did not observe any difference in methane production between a lasalocid fed group and control.

Hence, this study showed an improvement in ruminal fermentation by way of increased propionate production and reduced acetic acid and methane production. However a decrease in fibre digestibility was due to shift in microbial type in rumen with ultimately led to better utilisation of the feed.

REFERENCES

AOAC. 1984. Official methods of analysis (14th Ed.) Association of Official Agricultural Chemists, Washington, DC.

Beever, D. E., H. R. Losada, D. L. Gala, M. C. Spooner and M. S. Dhanoa. 1987. The use of monensin to control the digestion of the nitrogenous constituents of perennial rye grass and white clover in the rumen of cattle. Br. J. Nutr. 57:57-67.

Bergen, W. G. and D. B. Bates. 1984. Ionophores: Their effect on production efficiency and mode of action. J. Anim. Sci., 58:1465-1483.

Bogart, C., L. Gomez and J. P. Jouany. 1991. Effect of lasalocid and cationomycin on the digestion of plant cell walls in sheep. Can. J. Anim. Sci. 71:379-388.

Chalupa, W. 1980. Chemical control of rumen microbial metabolism. In Digestive Physiology and Metabolism in Ruminants. p. 325. AVI Publishing Company Inc. Wastport, CT.

Chaturvedi, M. L., U. B. Singh and S. K. Ranjhan. 1973. Effect of alkali treatment of wheat straw on feed consumption, digestibility and VFA production in cattle and buffaloe calves. Indian J. Anim. Sci. 43:677-682.

Chen, M. and M. J. Wolin. 1979. Effect of monensin and lasalocid-sodium on the growth of methanogenic and rumen saccarobytic bacteria. Appl. Environ. Microbiol. 38:72-77.

Conway, E. J. 1962. Micro diffusion analysis and volumetric error (5th Ed.). Crossby Lockwood and Sons Ltd., London.

Davis, G. V. and A. B. Erhat. 1976. Effect of monensin and urea in finishing steer rations. J. Anim. Sci., 43 : 1-8.

Delfino, J., G. W. Mathison and M. W. Smith. 1988. Effect of lasalocid on feedlot performance and energy partioning in cattle. J. Anim. Sci. 66:136-150.

Dennis, S. M., T. G. Nagaraja and E. E. Bartley. 1981. Effect of lasalocid or monensin on lactate producing or using rumen bacteria. J. Anim. Sci. 52:418-426.

Dinius, D. A., H. K. Goering, R. R. Oltjen and H. R. Cross. 1978. Finishing beef steers on forage diets with additives and supplemental lipids. J. Anim. Sci. 46:761-768.

Dinius, D. A., M. E. Simpson and P. B. Marsh. 1976. Effect of monensin fed with forage on digestion and the ruminal ecosystem of steers. J. Anim. Sci. 42:229-234.

Erwin, E. S., G. J. Macro and E. M. Energy. 1961. Volatile fatty acid analysis of blood of rumen fluid by gas liquid chromatography. J. Dairy Sci. 44:1768-1771.

Faulkner, D. B., T. J. Klopfenstein, T. N. Trotter and R. A. Britton. 1985. Monensin effects on digestibility, ruminal protein escape and microbial protein synthesis on high fiber diets. J. Anim. Sci. 61:654-660.

Goodrich, R. D., J. E. Garrett, D. R. Gast, M. A. Krick and J. C. Meiske. 1984. Influence of monensin on the performance of cattle. J. Anim. Sci. 58:1484-1498.

Haimoud, D. Ali, M. Vernay, C. Bayourthe and R. Moncoulon. 1995. Avoparcin and monensin effects on the digestion of nutrients in dairy cows fed a mixed diet. Can. J. Anim. Sci. 75:379-385.

Lemenager, R. P., F. N. Owens, K. S. Lusby and T. Robert. 1978. Monensin, forage intake and location of range beef cows. J. Anim. Sci. 47:247-254.

Leng, R. A. 1982. Dynamics of protozoa in the rumen of sheep. Br. J. Nutr. 48 : 399-416.

NRC. 1984. Nutrient requirements of domestic animals. National Research Council, Washington, DC.

Poos, M. I., T. L. Hanson and T. J. Klopfenstein. 1979. Monensin effects on diet digestibility, ruminal protein by-pass and microbial protein synthesis. J. Anim. Sci. 48:1516-1523.

Raun, A. P., C. O. Cooley, E. L. Potter, R. P. Rathmacher and L. F. Richardson. 1976. Effect of monensin and feed efficiency of feedlot cattle. J. Anim. Sci. 43:670- 677.

Ricke, S. C., L. L. Berger, P. J. Van der Aar and G. C. Fahey, Jr. 1984. Effects of lasalocid and monensin on nutrient digestion, metabolism and rumen characteristics of sheep. J. Anim. Sci. 58:194-202.

Rogers, M., J. P. Jouany, P. Thivend and J. P. Fontenot. 1991. Comparative effects of feeding and duodenal infusion of monensin in digestion in sheep. Can. J. Sci. 71:1125-1133.

Singh, U. B., D. N. Verma, A. Verma and S. K. Ranjhan. 1974. Measurement of rate of the production of bacteria in the rumen of buffalo calves. J. Agric. Sci. Camb. 83: 13-18.

Snedecor, G. W. and W. G. Cochran. 1968. Statistical Methods, Oxford and IBH Publishing Company, Calcutta, India.

Stock, R. A., S. B. Lauder, W. W. Stroup, E. M. Larson, J. C. Paraoty and R. A. Britton. 1995. Effect of monensin and monensin and tylarin cambination on feed intake variation of feedlot steers. J. Anim. Sci. 73:39-44.

Theurer, B., H. Bristol and W. H. Hale. 1974. Dietary regimen and post-parandial plasma VFA levels in cattle. J. Anim. Sci. 39:255(Aabstr.).

Thornton, J. H. and F. N. Owens. 1981. Monensin supplementation and *in vivo* methane produciton by steers. J. Anim. Sci. 52:628-634.

Thornton, J. H., F. N. Owens, R. P. Lemenagar and S. Totusek. 1976. Monensin and ruminal methane production. J. Anim. Sci. 43:336(Abstr.).

Whetstone, H. D., C. L. Davis and M. P. Bryant. 1981. Effect of monensin on breakdown of protein by ruminal microorganisms *in vitro*. J. Anim. Sci. 53:803-809.