Supplementation effect of ammonium sulphate and sulphur coated urea on rumen metabolites in crossbred cow

ANSHU RAHAL

Departmentof Animal Nutrition, College of Veterinary and Animal Science, G. B. Pant University of Agriculture and Technology, Pantnagar-263145 (U. S. Nagar, Uttarakhand)

ABSTRACT : In order to study effect of supplementation of ammonium sulphate and sulphur coated urea on rumen metabolites, fifteen lactating crossbred cows were selected from Instructional dairy farm. Thirty gram ammonium sulphate and sulphur coated urea was given to group II and group III respectively in two equal doses at the time of milking while group I was taken as control. Rumen liquor was collected in mid of feeding trial at every two hour interval (0, 2, 4, 6 & 8 hours after feeding). The rumen liquor pH of supplemented groups decreased and varied significantly from the control. The lowest values were found at 2 hours after feeding .Total nitrogen in group III varied significantly from other two groups .Supplemented groups had lower ammonical nitrogen than group I and varied significantly($P<0.01$). Highest TCA-precipitable nitrogen was noted in group III which varied significantly. Total volatile fatty acids concentration also was high in supplemented groups and varied significantly ($P<0.01$). Supplementation creates favourable environment for more microbial protein production.

Key words: Ammonium sulphate, crossbreds, rumen metabolites, sulphur coated urea

In India 70% population is dependent for their livelihood on agriculture. The scarcity of feed resources has reduced production of livestock. Various technical interventions, strategies and policy decisions can enhance the availability of fodder in country without competing with food crops. Making available the appropriate feed having suitable consistency of required nutrients and fodder for the animal based on its biology and genetics along with suitable farm management practices will improve milk productivity of the animals. Common NPN source, urea has been used in one or the other form for increasing utilization of fibrous feed residues. Ruminal pH has great impact on rumen fermentation efficiency (Rahal, 1996; Wanapat, 2003).Urea toxicity due to careless handling always pose animal at risk. Slow release of ammonia has been tried using oil, carbohydrate, formaldehyde etc (Cherdthong and Wanapat, 2010). The fermentation pattern is severely affected by sulphur and ammonia concentration of the rumen. In this trial supplementation of ammonium sulphate and sulphur coated urea was supplemented to study the impact on rumen fermentation characteristics in lactating dairy cow.

MATERIALS AND METHODS

Feeding trial of 90 days was conducted at Instructional dairy Farm, Nagla, Govind Ballabh Pant University of Agriculture & Technology, Pantnagar, Uttarakhand. Fifteen Lactating crossbred cows were selected from the herd and divided into three groups consisting of five animals each .Group I was taken as control while in group II, 30g ammonium sulphate and in group III 30g sulphur coated urea was added in concentrate mixture of animals in two doses (15g, each dose). During adaptation period, all the cows were maintained on mixed green fodder (green oat, green berseem, green mustard approximately in 1:1:1 ratio), wheat straw and concentrate was provided as per their requirement (NRC, 2001).The concentrate mixture was offered at the time of milking i.e.2.30AM and 2.30PM. All the animals were housed in well ventilated shed having concrete floor with individual feeding arrangement and tied with iron chain having tail to tail arrangement during adaptation & experimental feeding period. The cows were tied at such a distance that they had freedom for free movement and preventing them from getting access to the manger of other cows. The animals were let loose to open enclosure between 8.00AM to 9.00 AM daily to have exercise and access to fresh drinking water. Water was offered again in evening by buckets at 4.00 PM. The byre was washed and cleaned daily. All the animals were cleaned regularly by splashing with water and then groomed. Proper hygienic conditions and

healthy surroundings were maintained in shed throughout experimental period. Feed analysis was done using methods detailed in AOAC (1995).

Rumen liquor was collected in mid of feeding trial at every two hour interval (0, 2, 4, 6 & 8 hours after feeding) by rumenocentesis using 16 gauge 6 inch needle. Samples were centrifuged at 2000 rpm for 5 minutes and clear supernatant was obtained. Two drops of saturated mercuric chloride were added to the supernatant of rumen liquor as preservative. The pH of rumen liquor was measured immediately after each collection by digital pH meter. The total nitrogen was determined by Kjeldahl method. Ammonical nitrogen was analysed by microkjeldahl method. For the estimation of TCA precipitable N, rumen liquor was taken in centrifuge tube mixed with equal volume of 20% Trichloroacetic acid and tubes were left overnight. Next day tubes were centrifuged at 2000 rpm for 10 minutes. The whole precipitated content was transferred to kjeldahl flask with repeated washings of distilled water and then, it was processed for digestion, distillation and titration as for total nitrogen estimation. Non proteinous Nitrogen was calculated by subtracting sum of ammonical nitrogen and TCA-precipitable nitrogen from total nitrogen. Total volatile fatty acid was determined by method of Barnett and Reid (1957).

The data recorded for various parameters were subjected to statistical analysis as per Snedecor and Cochran (1994) using one-way, Two-way and three-way anova (STPR-43) to find out significance of differences between the treatment groups.

RESULTS AND DISCUSSION

The dry matter content of mixed green fodder, dry fodder, mixed fodder and concentrate was found to be 20.93, 88.43, 27.06 and 87% respectively while on dry matter basis, the organic matter content was 91.7, 92.8, 91.8 and 93.4% respectively. Mixed green fodder contained considerable variation in dry matter ranging from 19 to 35% during the experimental period. Mixed fodder contained crude protein 13.66%, crude fibre 27.3%, nitrogen free extract 49.4%, total ash 8.2%, Acid Insoluble ash 4.62%, neutral detergent fibre 60.95% and Acid detergent fibre 43.64%. Mixed green fodder contained green berseem, green oat and green mustard

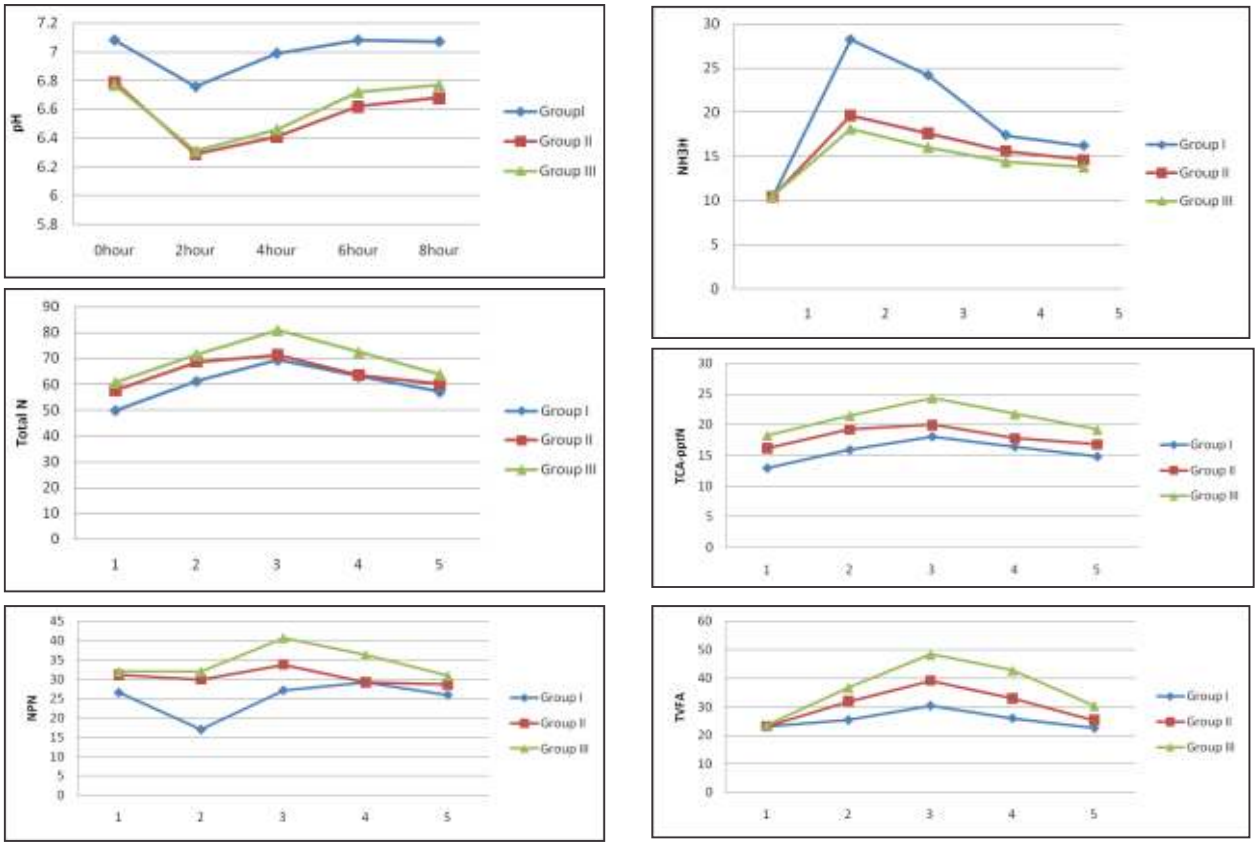

Fig. 1: Effect of supplementation of ammonium sulphate and sulphur coated urea on rumen metabolites in lactating crossbred cows

approximately in 1:1:1 ratio. Mixed green fodder contained 14.7% crude protein, 26% crude fibre, 49.5% nitrogen free extract, 8.3% total ash, 4.6% Acid insoluble ash, 59% neutral detergent fibre and 43% Acid Detergent Fibre. Wheat Straw contained 3.32% crude protein, 40.5% crude fibre, 48.48% nitrogen free extract, 7.2% total ash, 4.8% Acid Insoluble ash, 80.45% neutral detergent fibre and 50% Acid detergent fibre. The composition of wheat straw remained almost identical during the feeding period. The concentrate offered to the animals contained 19.95% crude protein, 9.5% crude fibre, 60.95% nitrogen free extract 6.6% Total ash, 1.4% Acid insoluble ash, 39.8% neutral detergent fibre and 12.25% Acid detergent fibre.

The supplementation of ammonium sulphate and sulphur coated urea resulted in drastic changes in rumen parameters of crossbred lactating cows which has been depicted in Table No.1.

pH : Highly significant difference (P<0.01) was found in group II and Group III compared to control (Group I). Supplementation decreased the rumen pH of animal. The decrease in pH more was noted in group II. Highly significant difference (P<0.01) was also found within the group with respect to time interval. The pH decreased significantly after two hours of supplementation in all groups and then increased slowly. The interaction between time interval and treatment was also found to be significantly different (P<0.05). The pH of group II and group III differed non-significantly. The pH was within the range considered optimal for microbial digestion of fibre and protein i.e. 6.0 to 7.0 (Hoover, 1986; Rahal,1997; Wanapat and Cherdthong, 2009)

Table 1: Rumen Liquor metabolites in Lactating crossbred cow

Group	0 hour	2 hour	4 hour	6 hour	8 hour	mean
			pH			
Group I	7.08^{ax}±0.084	6.76^{bx}±0.089	6.99^{ax}±0.137	7.08^{ax}±0.105	7.07^{ax}±0.092	7.00^{x}
Group II	6.79^{ay}±0.038	6.29^{dy}±0.038	6.41^{cy}±0.055	6.62^{bz}±0.164	6.68^{bz}±0.107	6.56^{y}
Group III	6.77^{ay}±0.055	6.31^{cy}±0.119	6.46^{by}±0.109	6.72^{ay}±0.078	6.77^{ay}±0.042	6.61^{y}
Mean**	6.88a	6.45^{c}	6.62^{b}	6.81^{a}	6.84^{a}	HxT*
			Total Nitrogen(mg/dl)			
Group I	49.88^{cy}±4.68	61.2^{by}±4.26	69.4^{ay}±3.84	63.0^{by}±3.24	57.0^{by}±3.6	60.0^{9y}
Group II	57.68^{bx}±5.45	68.8^{ax}±7.46	71.4^{ay}±6.42	63.6^{by}±8.73	$60.0b^{x}$±6.28	64.2^{9y}
Group III	60.80^{cx}±9.25	71.6^{bx}±3.13	81.08^{ax}±11.54	72.5^{bx}±5.17	63.94^{cx}±6.74	69.9^{8x}
Mean**	56.12^{c}	67.2^{b}	73.96^{a}	66.36^{b}	60.31^{c}	HxT^{ns}
			NH3-N(mg/dl)			
Group I	10.36^{dx}±0.27	28.26^{ax}±1.24	24.2^{bx}±1.48	17.4^{cx}±2.19	16.2^{cx}±1.48	19.2^{8x}
Group II	10.44^{dx}±0.41	19.6^{ay}±2.30	17.6^{by}±1.67	15.6^{cy}±1.81	14.6^{cy}±1.94	15.5^{6y}
Group III	10.5^{dx}±0.60	18.08^{az}±0.91	16.0^{bz}±1.58	14.4^{cz}±1.14	13.8^{cy}±1.48	14.5^{5y}
Mean**	10.43^{d}	21.98^{a}	19.26^{b}	15.8^{c}	14.86^{c}	HxT**
			TCA- Precipitable N (mg/dl)			
Group I	12.96^{cz}±1.21	15.91^{bz}±1.10	18.04^{az}±1.00	16.38^{az}±0.84	14.82^{bz}±0.93	15.62^{z}
Group II	16.15^{by}±1.52	19.26^{ay}±2.09	19.99^{ay}±1.79	17.80^{by}±2.44	16.80^{by}±1.76	18.00^{y}
Group III	18.24^{cx}±2.77	21.48^{bx}±0.93	24.32^{ax}±3.46	21.75^{bx}±1.55	19.18^{cx}±2.02	20.99^{x}
Mean**	15.78^{c}	18.88^{b}	20.78^{a}	18.64^{b}	16.93	HxT^{ns}
			NPN(mg/dl)			
Group I	26.55^{ay}±3.33	17.02^{by}±3.39	27.15^{az}±4.04	29.22^{ay}±3.96	25.98^{ay}±2.86	25.18^{z}
Group II	31.09^{ax}±4.00	29.93^{ax}±7.10	33.80^{ay}±5.92	29.19^{ay}±7.76	28.60^{bx}±4.50	30.52^{y}
Group III	32.06^{bx}±6.85	32.04^{bx}±1.95	40.75^{ax}±7.85	36.35^{ax}±3.73	30.96^{bx}±4.67	34.43^{x}
Mean**	29.90^{a}	26.33^{b}	33.90^{a}	31.58^{a}	28.51^{b}	HxT^{ns}
			TVFA(mM/dl)			
Group I	23.0^{bx}±2.23	25.4^{bz}±2.19	30.40^{az}±2.70	26.0^{bz}±1.58	22.6^{by}±3.43	25.48^{z}
Group II	23.2^{cx}±5.76	32.0^{0by}±4.69	39.2^{ay}±3.56	33.0^{by}±5.56	25.4^{cy}±5.98	30.56^{y}
Group III	23.6^{ex}±4.98	36.8^{cx}±4.76	48.4^{ax}±4.39	42.8^{bx}±3.34	30.40^{dx}±2.30	36.40^{x}
Mean**	23.26^{c}	31.4^{b}	39.33^{a}	33.93^{b}	26.13	HxT**

H-Time interval, T-treatment, ns-nonsignificant
Means bearing superscripts x, y, z within a column differ significantly from each other,**P<0.01
Means bearing superscripts a, b, c, d within a row differ significantly from each other,**P<0.01

Total Nitrogen: The total nitrogen in group III (69.98) was significantly different (P<0.01) compared to group I (60.09) and II (64.29). Group I and Group II differed non-significantly. Highly significant difference (P<0.01) also existed within the group with respect to time interval. The highest value for total nitrogen was found at 4 hours after feeding and thereafter there was a gradual fall in the values. The interaction between time interval and treatment was found to be non-significant. More nitrogen was available for ruminal protein synthesis and relatively less ammonia for urea formation in the liver (Verma *et al*, 1981).

Ammonical Nitrogen: The supplemented groups had lower ammonical nitrogen compared to control group and differed significantly (P<0.01). Non-significant difference was found among the supplemented groups (group II and Group III). Highly significant difference (P<0.01) was found within the group with respect to time interval. Values at two hours differed significantly. Higher values were noted after two hours of supplementation. Least value was found of ammonical nitrogen in group III compared to group II and group I and values were highly significant. Non-significant differences existed between 6th and 8th hour after feeding. The interaction between time interval and Treatment was also highly significantly different(P<0.01).Controlled rate of nitrogen degradation in rumen led to slow rate of ammonical -N release. Similar results have been reported by Chanjula *et al* (2003), Chanjula *et al*, 2004), Cherdthong *et.al* (2010), Pinos-Rodriguez *et al* (2010), Taylor-Edwards *et al*. (2009) ,Wanapat *et al* (2008), Tan and Murphy (2004) and Akinlade and Osoanya (2016).

TCA-precipitable N: Highly significant difference (P<0.01) was found among the three groups. Group III had highest TCA-precipitable Nitrogen (20.99) followed by group II (18) and group I (15.62). Significant difference (P<0.01) also existed within the group with respect to time interval. Highest TCA-precipitable N was noted at 4 hour after feeding followed by gradual decrease. Non-significant difference was found in the interaction between treatment and time interval. Similar results have been reported by Rahal *et al* (1997).

NPN: Highly significant difference (P<0.01) existed between the groups. Highest values were noted for group III (34.43) followed by group II (30.52) and group I (25.18). Each group differed significantly from each other. Significant difference (P<0.01) was also noted within the group with respect to time. Highest values were noted at 4 hour after feeding. The interaction between time interval and treatment were non-significant.

TVFA: Highly significant difference (P<0.01) existed among the groups. Highest values were noted in group III (36.4) followed by group II (30.56) and group I (25.48) Each group differed significantly from each other. Significant difference (P<0.01) was also noted within the group with respect to time interval. Highest values were noted in group III and peak of Total volatile fatty acids in all groups reached at about 4 hours after feeding. The interaction between time interval and treatment was found to be highly significant (P<0.01).Increased TVFA in group II was due to increased dry matter intake which provide more substrate for synthesis of *de-novo* fatty acids. Klusmeyer *et al* (1987) and Umunna and Woods (1975) also reported that ammonium sulphate and elemental sulphur are most effective in promoting synthesis of rumen microbial protein and volatile fatty acids.

CONCLUSION

The supplementation of ammonium sulphate and sulphur coated urea help in creating favorable conditions for rumen fermentation yielding more of microbial protein to meet protein demand of lactating animal.

ACKNOWLEDGEMENTS

The author acknowledges the help rendered by Dr Ripusudan Kumar, Assistant Professor, KVK, Majhera in carrying out this research work. Financial support from Director Research, Pantnagar is thankfully acknowledged to complete this project.

REFERENCES

Akinlade A.T. and Osoanya T.O. (2016). Effect of ammonium sulphate fortification on growth performance, Nutrient Digestibility and Nitrogen Balance of West African Dwarf Rams. *Journal of Animal Production Advances*, 6(5):943-949.

AOAC(1995).*Official Methods of Analysis*. 16thedn. Association of Official Analytical Chemists. Arlington,VA,USA.

Barnett, A.J.G. and Reid, R.L., (1957). Studies on the production of volatile fatty acids from grass by rumen liquor in an artificial rumen. 1. The volatile acid production from fresh grass. *Journal of Agricultural Science*, *48*:. 315-321.

Chanjula, P., Wanapat, M., Wachirapakorn, C., Uriyapongson, S. and Rowlinson, P., (2003). Ruminal degradability of tropical feeds and their potential use in ruminant diets. *Asian Australasian Journal of Animal Sciences, 16*(2): 211-216.

Chanjula, P., Wanapat, M., Wachirapakorn, C. and Rowlinson, P. (2004). Effect of synchronizing starch sources and protein (NPN) in the rumen on feed intake, rumen microbial fermentation, nutrient utilization and performance of lactating dairy cows.*Asian Australasian Journal of Animal Sciences*, *17*(10):.1400-1410.

Cherdthong A and Wanapat,M.(2010). Development of Urea Products as Rumen Slow-Release Feed for Ruminant Production: A Review. *Australian Journal of Basic and Applied Sciences*, 4(8): 2232-2241.

Cherdthong., A, Wanapat, M, Wachirapakom, C., and Van Amburgh, M.E., (2010). Evaluation of urea-calcium mixtures (UCM) as slow release: Fermentation characteristics using in vitro gas technique. Proceeedings of the Agriculture Conference 11 January 25-26, 2010. Kawee Jutikul Auditorium, Khon Kaen, Thailand,pp 138-141.

Hoover,W.H. (1986).Chemical factors involved in ruminal fiber digestion. *J. Dairy Sci.*, 69(10): 2755-2766.

Klusmeyer, T.H., Clark, J.H., Vicini, J.L., Murphy, M.R. and Fahey, G.C., (1987). Effects of Feeding or Infusing ammonium salts of Volatile Fatty Acids on Ruminal Fermentation, Plasma Characteristics, and Milk Production of Cows1. *J of Dairy Science*, *70*(1): 50-63.

NRC (2001).Nutrient Requirements of Dairy Cattle. 6th edn. National Research Council. National Academy of sciences,Washington,D.C.USA.

Pinos-Rodriguez, J.M, Pena, L Y., Gonzalez-Munoz, S.S., Barcena, R., Barcena, R, Salem, A (2010). Effects of a slow- release coated urea product on growth performance and ruminal fermentation in beef steers. *Italian Journal of Animal Science*, 9:1, e4.

Rahal, A., Singh, A. and Singh, M., (1997). Effect of urea treatment and diet composition on, and prediction of nutritive value of rice straw of different cultivars. *Animal Feed Science and Technology*, 68 (1-2):165-182.

Rahal, A., (1996). A Rapid, Simple and Economic Technique to Predict the Nutritive Value of Rice Straw of Different Cultivars. *Indian Journal of Animal Nutrition*, 13(3): 125-132.

Rahal, A., (1997). Rumen Fermentation Pattern in Crossbred Cattle as Influenced By Different Diets. *Indian Journal of Animal Nutrition*, 14(3):186-188.

Snedecor, G.W and Cocharan, W.G. (1994). Statistical Methods. The Iowa State University Press, Iowa, USA.

Tan, Z., & Murphy, M. R. (2004). Ammonia production, ammonia absorption, and urea recycling in ruminants. A review. *Journal of Animal and Feed Sciences*, *13*(3): 389-404.

Taylor-Edwards, C.C., Elam, N.A., Kitts, S.E., McLeod, K.R., Axe, D.E., Vanzant, E.S., Kristensen, N.B. and Harmon, D.L., (2009). Influence of slow-release urea on nitrogen balance and portal-drained visceral nutrient flux in beef steers. *J.Anim.Sci.*, 87(1):209-221.

Umunna, N. N., & Woods, W. R. (1975). Nitrogen utilisation by lambs fed a high roughage ration and supplemented with compacted starch-urea or sulphur-coated urea. *Journal of the Science of Food and Agriculture*, 26(4): 413-419.

Verma, D. N., Dass, R. S., Mazumdar, A., Singh, U. B., & Srivastava, U. S. (1981). Effect of feeding urea and ammonium sulphate as source of nitrogen on rumen metabolism in buffalo. *Indian Veterinary Medical Journal*, 5(1): 19-21.

Wanapat, M. (2003). Manipulation of cassava cultivation and utilization to improve protein to energy biomass for livestock feeding in the tropics.Asian-Aust. *J.Anim.Sci*., 16(3): 463-472.

Wanapat, M., Cherdthong, A., Pakdee, P., Wanapat, S., (2008).Manipulation of rumen ecology by dietary lemongrass (Cymbopogon citrates Stapf.) powder supplementation. *J.Anim.Sci*., 86(12):3497-3503.

Wanapat, M. and Cherdthong, A.(2009) Use of real-time PCR technique in studying rumen cellulolytic bacteria population as affected by level of roughage in swamp buffalo.*Curr.Microbio*.,58(4):294-299.

Received: May 3, 2017
Accepted: July 7, 2017