Development and Evaluation of Protected Fat in Wheat Straw Based Total Mixed Ration

S. K. Sirohi*, Raman Malik and T. K.Walli

Dairy Cattle Nutrition Division, National Dairy Research Institute, Karnal-132 001, India

ABSTRACT : Ca salt of soybean oil (PSO) and that of mustard oil plus mahua oil (PMOMO) (50:50) were prepared using double decomposition method, and further tested for their fatty acid composition and degree of saponification. Furthermore, the different levels of protected fat of PSO and PMOMO were evaluated in wheat straw based total mixed ration (TMR) *in vitro*. Results indicated that capric, lauric, myristic, palmitic, steric, oleic, linoleic, leinolenic acids were traces, traces, traces, 10.00, 2.00, 25.00, 58.50, 5.0% in PSO while the corresponding values in PMOMO were 1.08, 0.28, 0.45, 16.9, 12.95, 44.38, 17.46 and 6.50%, respectively. The degree of saponification of both protected fat supplements was more than 80%. Six treatment combinations were tested i.e., blank without feed and fat supplement (T1); control diet with out fat supplement (T2); control diet plus bypass fat supplement (PSO) so that diet contain 5% fat (T3); control diet plus bypass fat supplement (PSO) so that diet contain 7.5% fat (T4); two more diets viz. T5 and T6 were formulated using bypass fat supplement from PMOMO containing 5 and 7.5% fat respectively. TMR was prepared using 50% concentrate mixture and 50% wheat straw. Result indicated that TVFA, NH_3-N,TCA-N, total-N and total gas production were increased in treatment diets at 7.5% level of supplementation, however, fermentation pattern remain similar at 5.0% level of supplementation with respect to control diet. Nevertheless, IVDMD and IVOMD values remained unchanged, rather non-significant at both fat levels and with the both fat sources. On the basis of results it was concluded that Ca-salt of Soybean oil or Mustard plus Mahua oil did not show any negative effect either on digestibility or on microbial protein synthesis in rumen, hence the dietary fat upto 7.5% level in total mixed ration based on wheat straw, could be safely used without any adverse effect on rumen fermentation. ***(Asian-Aust. J. Anim. Sci. 2001. Vol 14, No. 10 : 1405-1408)***

Key Words : Total Mixed Ration, By-Pass Fat, Rumen Fermentation, Digestibility, Unconventional Oil

INTRODUCTION

Ruminants are generally fed a dietary fat ranging from 3-4% in total diet. Increase in fat percentage in diet results in reduction of microbial activity in rumen and depresses the digestion of cellulose (Czerkawski et al., 1966 and Henderson 1973). Moreover, a constant increase in the prices of oil cakes and other fat supplements limits the use of such feed ingredients in the diet of ruminants in developing countries. Consequently, energy density in diet of productive animals is low, which in turn affect productivity. Calcium salts of fatty acids (CSFA) have been used successfully in dairy animals to increase the energy density of the diet without any adverse effect of these lipids on rumen environment (Jenkins and Palmquist 1984; Coppock and Wilks 1991). CSFA can also serve as method of manipulating the fatty acid composition of milk fat (Fearon et al., 1994). The present study was carried out to develop a method for the preparation of CSFA, using conventional and unconventional oils and to test the effectiveness of fat protection, using some lab techniques.

MATERIALS AND METHODS

Ca-soaps of soybean oil (PSO) and mustard oil plus mahua oil (PMOMO) (50:50) were prepared in laboratory using double decomposition method. PSO and PMOMO were first saponified with sodium hydroxide and then Ca-soaps were precipitated with saturated solution of calcium chloride, which were washed with water, dried and crushed with grinder. Fatty acids were analyzed as per the method given by Sukuhija and Palmquist (1988). Degree of saponification was calculated after estimating total fatty matter and unsaponified fat portion, as described by Garg (1997). The protected fat supplements prepared were tested in six treatment combinations under *in vitro* environment. The following six treatment combinations were formulated viz. Blank without feed and fat supplement (T1); control diet without fat supplement (T2); control diet plus bypass fat supplement (PSO) so that diet contain 5% fat (T3); control diet plus bypass fat supplement (PSO) with diet containing 7.5% fat (T4); and the diets were formulated with bypass fat supplement from PMOMO containing 5 and 7.5% fat in T5 and T6 respectively. The above six treatments with six replicates in each treatment were evaluated for analysis of variance in randomised block design. In control group, total mixed ration (TMR) was prepared using 50 parts concentrate mixture and 50 parts roughage (wheat straw) and ground in hammer mill to 1 mm and used for *in vitro* incubations. The concentrate mixture consisted of maize 40, deoiled ground nut cake 30, wheat bran 27, mineral mixture 2 and salt 1 parts. All treatment were evaluated under *in vitro* conditions by

* Corresponding Author: S. K. Sirohi. Tel: +91-0184-259064, E-mail: ssirohi@ndri.hry.nic.in.

Received December 1, 2000; Accepted May 31, 2001

incubating the 0.5 g of total mixed feed sample in 40 ml McDougall's (1948) buffer and 10 ml rumen liquor anaerobically for 24 h in 100 ml bottles as per the Van Soest et al. (1966). After 24 h *in vitro* incubations, dry matter and organic matter digestibilities were estimated and filtrate were analyzed for total volatile fatty acids (TVFA), Ammonia-N, TCA precipitable nitrogen and total nitrogen as per standard procedures. Total gas production was estimated as per the method described by Theordorou et al (1994) using transducer. Methane was analyzed by absorbing carbon dioxide in 1N sodium hydroxide and making necessary correction of some other gases present in ruminal gas pool. Data obtained were analyzed statistically according to Snedecor and Cochran (1968).

RESULTS AND DISCUSSION

Chemical composition of TMR was presented in table 1. Fatty acid composition and their degree of saponification of calcium soaps prepared from soyabean oil and mustard oil plus mahua oil (50:50) was given in table 2. Result indicated that capric, lauric and myristic acids percentage were very low in both PSO and PMOMO. However, oleic acid was higher (44.38%) in PMOMO but linoleic acid was higher (58.0%) in PSO. In both of the protected fat supplements, unsaturated fatty acid content was higher than saturated fatty acid content. Degree of saponification in both calcium soaps were about 80%.

Results of TVFA, individual VFAs and their ratio (A: P and A: B) are presented in table 2. Total volatile fatty acids concentration was significantly higher ($p<0.01$) in different treatment combinations in comparison to control. However, the variation among the treatments was insignificant. In both PSO and PMOMO supplemented groups TVFA's concentration was higher at 5.0% level as compared to 7.5% level, however, the TVFA concentration was similar at different levels of fat in PSO and PMOMO supplemented groups. Similar results were obtained in individual concentrations of VFAs. Individual VFAs was slightly higher in fat supplemented group in comparison to control group. Acetate (A) to propionate (P) ratios during fermentation remained unchanged but acetate (A) to butyrate (B) ratio showed significant effect, which could be due to little increase in butyrate concentration in different treatment groups compared with control group. Thus, the A:B ratio was higher in control group than in treatment (T3, T4, T5 and T6) respectively, indicating that microbial activity was not hampered due to protected fat supplementation. These results are in agreement with those who only reported a little or no effect of supplemental fat in protected form on rumen VFA concentration or acetate: propionate ratios (Grummer et al., 1993 and Abdullah et al., 2000). Results of total gas production, methane production and nitrogen fractions on various treatments are mentioned in table 3. Total gas production (ml/0.5 g substrate/ 24 h) also showed a similar trend as TVFAs, the values were higher in treatments groups than in control group. Gas production was higher at 7.5% fat level than at 5.0% fat level in PMOMO supplemented groups, however, no such variation was found in PSO supplemented groups. Methane production showed a decreasing trend, especially at 5.0% fat level in both supplemented groups, however, at 7.5 % fat level, the values were remained somewhat similar in both the supplemented groups. Methane production was reduced to 20% and 4% respectively in PSO and PMOMO supplemented groups at 5% fat level.

Results (table 4) of nitrogen fractions like NH_3-N, TCA-N and Total- N indicated that different fractions were increased during fermentation as compared to blank group. However, differences between control and treatment groups supplemented with fat, remained unchanged statistically, indicating that microbial protein synthesis was not affected in any way by addition of protected fat in the diet. Results of DM and OM digestibility (table 5) showed that values remained unchanged and the difference between treatment groups and control group were nonsignificant. These results

Table 1. Chemical composition of TMR on DM basis

Chemical composition	TMR(%)
OM	88.5
EE	3.3
CP	12.0
NDF	52.0
ADF	30.5
Total ash	15.5

TMR is prepared by using wheat straw and concentrate mixture in 1:1 ratio

Table 2. Fatty acid composition (g/100 g) and degree of saponification of calcium soaps of soyabean oil and Mustard oil plus Mahua oil

Particulars of Ca soap	$C_{10:0}$	$C_{12:0}$	$C_{14:0}$	$C_{16:0}$	$C_{18:0}$	$C_{18:1}$	$C_{18:2}$	$C_{18:3}$	$C_{20:0}$	Deg.of saponification (%)
	Percentage									
Soyabean oil	Tr.[1]	Tr.	Tr.	10.0	2.00	25.00	58.00	5.00	Tr.	82
Mustard oil +Mahua oil (50:50)	1.08	0.28	0.45	16.90	12.95	44.38	17.46	6.50	-	80

Tr.[1]= trace

Table 3. Effect of type and level of Bypass fats supplementation on individual VFAs and total VFA's (m mole/100ml) production in TMR.

Treatments	Acetic	Propionic	Butyric	TVFA's	A/P	A/B
T1 (Blank)	1.60	0.63	0.43	2.66	2.539	3.72
T2 (Control)	3.51	1.42	0.74	5.66	2.471	4.74
T3	4.00	1.60	1.06	6.66	2.500	3.77
T4	3.90	1.43	1.00	6.33	2.720	3.90
T5	4.10	1.56	1.00	6.66	2.628	4.10
T6	3.99	1.53	1.16	6.66	2.607	3.43
LSD	1.51**	0.70**	0.31**	0.38**	NS	0.65**

** Result are significant at 1% level ($p<0.01$), LSD Least significant difference.

Table 4. *In vitro* total gas production and nitrogen fractions as effected by protected fat supplementation in TMR

Treatments	Total gas	Methane	NH_3-N	TCA-N	Total -N
T1 (Blank)	21.00	2.06	20.07	10.92	57.77
T2 (Control)	69.00	6.90	25.86	12.88	97.53
T3	72.00	5.52	23.07	15.35	99.40
T4	72.66	8.13	25.13	14.84	98.00
T5	70.00	6.67	27.53	14.84	99.87
T6	73.33	7.00	24.73	15.12	100.80
LSD	3.16**	0.92**	3.75*	2.09**	4.02**

1 Total gas and methane in ml/0.5 g substrate/24 h, 2 NH_3-N, TCA-N and total-N in mg/100 ml, 3 LSD- Least significant difference.
** Result are significant at $p<0.01$

Table 5. Effect on digestibility of TMR as affected by protected fat supplementation

Treatments	DMD(%)	OMD(%)
T1 (Blank)	-	-
T2 (Control)	40.00	40.77
T3	42.26	43.17
T4	39.67	40.57
T5	40.17	40.97
T6	41.30	41.87
Average	40.68	41.47
Significance ($p<0.05$)	N.S.	N.S.

once again confirmed that the supplementation of protected fat in TMR does not affect the rumen fermentation, as also reported by previous workers : Elliot et al. (1994), Palmquist and Conrad (1980), Schauff and Clark (1989), West and Hill (1990), Garg (1997) and Abdullah et al. (2000). Devendra and Lewis (1974) postulated four theories, to explain the decrease in fibre digestibility as a result of fat supplementation in ruminant diet i.e. (a) physical coating of fibre to prevent microbial attack (b) modification of the rumen microbial population from possible toxic effects of fat on certain microorganisms (c) inhibition of microbial activity from surface active effects of fatty acids on cell membranes (d) reduced cation availability from formation of insoluble complexes with long chain fatty acids. Ca salts of fatty acids seems to be more or less inert at normal rumen pH, as seen in the present *in vitro* studies. Hence, the theories as suggested by above workers with regard to the effects rumen fermentation due to fat supplementation may not hold good in case of Ca saponified salts (Jenkins and Palmiquist, 1984 and Chalupa et al., 1984). On the basis of above results, it may be said that the supplementation of ruminant diet with protected fat did not effect the rumen environment or its functions and the digestibility of OM and DM. Ca saponified salts of fatty acids supplemented even at 7.5% level in total mixed ration appeared to remain inert under a *in vitro* rumen conditions.

CONCLUSION

Supplemental protected fat (as calcium saponified-salts) had no effect on molar concentration of TVFA's and nitrogen fractions in the rumen *in vitro* system. No differences were also observed in even the proportion of volatile fatty acids, and digestibility of OM and DM in TMR remained unchanged due to fat supplementation. Total gas production values increased slightly in comparison to control, however, the methane production was reduced as a result of fat supplementation. It appears from the results that protected fat could be used in total mixed ration efficiently upto 7.5% level in diet, without any adverse effect on rumen environment and its fermentation pattern.

REFERENCES

Abdullah, M., J. W. Young, H. D. Tyler and G. Mohiuddin. 2000. Effect of feeding high forage diets with supplemental fat on blood metabolites, rumen fermentation and dry matter

digestibility in dairy cows. Asian-Aus. J. Anim. Sci. 13:451-456.

Cappack, C. E. and D. L. Wilks. 1991 Supplemental fat in high energy ration for lactating cows: Effects on intake, digestion milk yield and composition. Anim. Sci. 69:3829.

Chalupa, W., D. Rickabaugh, D. S. Kronfield and D. Sklan. 1984. Ruminal fermentation *in vitro* as influenced by long chain fatty acids J. Dairy Sci. 67:1439.

Czerkawski, J. W., K. L. Baxter and F. W. Wainman. 1966. The metabolism of oleic and lenoleic, linoleneic acids by sheep with reference to their effects on methane production. Br. J. Nutr. 20:354-362.

Devendra, C and D. Lewis. 1974. The interaction between dietary lipids and fibre in the sheep 2. Digestibility studies. Anim. Prod. 19:67-74.

Elliott, J. P., T. R. Overton and J. K. Dreckley. 1994. Digestibility and effects of three forms of mostly saturated fatty acids. J. Dairy Sci. 77:789.

Fearon, A. M., C. T. Charlton and D. J. Kilpalrick. 1994. A further in investgation on the influence of dietary protected lipid supplements on the characteristics of cows milk fat. J. Sci. Food Agric. 66:247-256.

Grag, M. R. 1997. Bypass fat production using acid oil, its effect on *in vitro* rumen fermentation and effect of its feeding in *in sacco* DM disappearance in sheep. Asian- Aus. J. Anim. Sci. 10:571-574

Grummer, R. R. 1993. Etiology of lipid related metabolic disorders in periparturient dairy cows. J. Dairy Sci. 76:3882.

Henderson, C. 1973. The effects of fatty acids on pure culture of rumen bacteria. J. Agric. Sci. 81:107-112.

Jenkins, T. C. and D. L. Palmquist. 1984. Effect of fatty acid or calcium soaps on rumen and total nutrient digestibility of dairy rations. J. Dairy Sci. 67:978.

McDougall E. I. 1948. The composition and output of sheep's saliva. Biochem. J. 43:99-104.

Palmquist, D. L. and H. R. Concard. 1980. High fat rations for dairy cows. Comparison of tallow and hydrolyzed, blended animal vegetable fat at two levels of intake. J. Dairy Sci. 63:391-395.

Schauff D. J. and J. H. Clark. 1989. Effects of prilled fatty acids and calcium salts of fatty acids on rumen fermentation, nutrient digestibility, milk production and milk composition. J. Dairy Sci. 72:917-927.

Snedecor, G. W. and W. G. Cochran. 1968. Statistical Methods. 6th Ed. Iowa State University Press, Ames, Iowa, USA.

Theordorou, M. K., A. W. Barbar, S. D. Mewa, A. B. McAllan and J. France. 1994. A simple gas production method using pressure transducer to determine the fermentation kinetics of ruminant feeds. Anim. Feed Sci. Technol. 48:185-197.

Van Soest, P. J., R. H. Wine and L. A. Moore. 1966. Proc. 10th Int. Grassland. Congr. Helsinki, Finland: 438-441.

West, J. W. and G. M. Thill. 1990. Effect of protected fat product on productivity of lactating Holstein and Jersey cows. J. Dairy Sci. 73:3200.