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Effect of pre partum supplementation of rumen protected fat and protein on the performance of Murrah buffaloes

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ABSTRACT

The present experiment was conducted to study the effect of pre partum rumen protected fat and protein supplementation on the performance of Murrah buffaloes. Eighteen Murrah buffaloes (2 nd - 4th lactation) were divided into 2 groups (9 each) on the basis of most probable production ability (MPPA). Buffaloes in group 1 (control group; MPPA 2204.17 kg) were fed chaffed wheat straw, chopped green maize fodder and concentrate mixture as per requirements; buffaloes in group 2 (treatment group; MPPA 2210.64 kg) were fed same ration as control group plus 2.5% rumen protected fat (on DM intake basis) and concentrate mixture containing formaldehyde treated mustard and groundnut oil cake (1.2 g HCHO/100 g CP) in place of normal mustard and groundnut oil cakes. Group 2 buffaloes were supplemented rumen protected fat and protein 60 days pre partum. Average DM intake was 11.13 and 11.69 (kg/d) in groups 1 and 2, respectively, which was significantly higher in group 2. The average CP and TDN intakes were higher in group 2 than that of group 1. During last fortnight, group 2 buffaloes showed higher body weight gain than that of group 1. Average birth weights of the calves were higher by 10.8% in group 2 (35.38 kg) than that of group 1 (31.94 kg). The calving percentage was 100% in both groups. There was no effect on plasma glucose, NEFA, triglycerides and cholesterol concentrations among 2 groups, whereas BUN concentration was lower in group 2 during. Incidence of retention of foetal membranes, still births and premature births were reduced in group 2 buffaloes. It may be concluded that rumen protected fat and protein supplementation during pre partum period to advanced pregnant buffaloes increased the calf weight, decreased the incidences of retention of foetal membranes and premature birth in high yielding buffaloes.

Key words: Buffalo, Calving performance, Pre partum, Protected fat, Protected protein

Generally, the feeding of non-lactating animals is neglected (Tyagi *et al.* 2010), which may be responsible for lower growth of foetus, an increased incidence of peri-parturient health disorders and lower productive as well as reproductive performance. Dairy animals of good genetic potential usually experience negative energy balance prior to calving due to reductions in feed intake and modest increases in energy requirements during late gestation for growth of foetus (Grummer 2007).

Supplementing ration of cows and buffaloes with protected fat enhanced the energy intake without compromising rumen cellulolytic bacterial activity (Tyagi *et al.* 2009a, Thakur and Shelke 2010). However, feeding bypass protein supplies more energy to the energy deficient animals, through the same feed, as the extra amino acids supplied through bypass protein feeding are converted to glucose in liver (Chaturvedi and Walli 2000, Walli and Sirohi 2004). Keeping these points in view, a feeding trial was conducted to study the effect of pre-partum rumen protected fat and protein supplementation on parturition related parameters in buffaloes.

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MATERIALS AND METHODS

Animals, feeding and management: Pregnant Murrah buffaloes (18), selected from the herd at NDRI, Karnal, were divided into 2 groups (9 each) on the basis of most probable production ability (MPPA) and lactation number (second to fourth lactation; 5 to 7 years old). Buffaloes in group 1 (control group, MPPA 2204.17 kg) were fed chaffed wheat straw (particle size, 1.5 to 2.0 cm), chopped green maize fodder (particle size, 2.0 to 2.5 cm) and concentrate mixture as per requirements (Kearl 1982). However, buffaloes in group 2 (treatment group, MPPA 2210.64 kg) were fed the same ration as control group plus 2.5% rumen protected fat (on DM intake basis) and concentrate mixture containing formaldehyde treated mustard (MC) and groundnut oil (GNC) cake (1.2 g HCHO/100 g CP) in place of untreated cakes as a rumen protected protein source.

Composition (%) of the concentrate mixture was: maize 33, groundnut cake 21, mustard cake 12, wheat bran 20, deoiled rice bran 11, mineral mixture 2 and common salt 1. Green maize forage was fed separately whereas wheat straw and concentrate mixture was mixed prior to feeding and fed as per weekly calculated requirements (Kearl 1982) of each buffalo. The concentrate mixture was offered twice a day in equal parts at 05.00 and 18.00 h. Bypass fat was fed through concentrate mixture at one time i.e. 5.00 h. Fresh green maize forage were fed at 10.00 and 19.00 h in addition to wheat straw, which was offered at 5.00 h. DM content of forage and left over was determined to calculate the daily DM intake. The buffaloes were housed in a well ventilated paddock having individual feeding mangers and space for separate tying of individual animal and provided with fresh and clean tap water 3 times daily at 8.00 h, 12.00 h and 18.30 h. After adaptation period of 10 days, the buffaloes of group 2 were supplemented rumen protected fat and protein 60 days pre partum. Separate calving pen were provided to each animal and feeding and management of all animals was same as described earlier. Daily nutrient intake, fortnightly changes in body weights (on two consecutive days) and calving performance of pre parturient buffaloes were recorded.

Formaldehyde treatment of cakes: Rumen protected fat (Ca salts of fatty acids) was procured commercially. Individual GNC and MC were crushed in feed mill and treated with formaline (40% formaldehyde) at the level of 1.2 g formaldehyde/100 g CP of cake, in a horizontal mixer of feed mill. After treatment, the cakes were mixed thoroughly and finally stored in tightly sealed plastic bags for at least 4 to 5 days, as per Chatterjee and Walli (2003). The RDP (rumen degradable protein) and UDP (undegradable protein) values of cakes were estimated by *in sacco* nylon bag technique (Mehrez and Orskov 1977). These formaldehyde treated cakes were used for preparation of compounded concentrate mixture for feeding of buffaloes in treatment group.

Analytical techniques: The degree of protection of rumen protected fat was judged by estimating the degree of saponification of the Ca soaps (Tyagi *et al.* 2010). The dried samples of concentrate mixture, wheat straw and green maize forage were ground to pass through 1 mm sieve; pooled samples were analyzed for proximates (AOAC 2005) and cell wall constituents (Goering and Van Soest 1970). The fatty acid analysis of feed samples (green maize forage, wheat straw, concentrate mixture of control and treatment group) and rumen protected fat was done using saponification method (Gulati and Ashes 2000). The analysis was carried out on GLC fitted with flame ionization detector and 50 m length of capillary column. Initial temperature of the column was 140ºC. The RAMP rate was 2 ºC/min. Identification of peaks was made through retention time of the reference standards.

Blood samples were collected at start of experiment and after fortnightly intervals up to parturition. Blood glucose (GOD-POD), triglycerides (GPO-PAP), cholesterol (CHOD-PAP) and blood urea nitrogen (NED-dye) were estimated by using liquid gold kit. The non-esterified fatty acids (NEFA) concentration in the plasma samples was standardized using the extraction method of Itaya and Ui (1965) and the colour development procedure of Duncombe (1963).

Statistical analysis: Statistical analysis of the data were carried out by Students't' test as per Snedecor and Cochran (1986).

RESULTS AND DISCUSSION

Chemical composition and fatty acid profile of feeds and fodders: The chemical composition of feed ingredients is presented in Table 1. Total fat content in the supplemented bypass fat was 76.23% and the protection level from rumen hydrolysis (saponifiable portion) was 57.35%. The predominant fatty acids (% of total fatty acids) in green fodder, wheat straw, bypass fat and concentrate feed of control and treatment group are presented in Table 2. Formaldehyde treatment of MC increased its UDP content from 4.97 to 28.55%. Similarly, in case of GNC, formaldehyde treatment increased its UDP content from 15.86 to 32.66%.

Changes in body weight: Fortnightly changes in body weights of pre parturient buffaloes are presented in Table 3. In third fortnight (day 30 to 15 days pre parturient), group 2 gained higher ($P<0.05$) body weight (12.73 kg) than that of group 1 (10.45 kg). During transition period, feed intake decreases at a time when the energy requirements are increasing due to rapid growth of the conceptus (Bellows *et al*. 2001). So, rumen protected fat and protein supplementation to the treatment group, increased the energy density of the consumed ration. Higher ($P<0.05$) loss in weight was observed in group 2 (83.65 kg) than that of group 1 (76.23 kg), due to parturition and release of the fetal membranes.

Nutrients intake and plane of nutrition: Average dry matter intake (DMI) was 11.13 and 11.69 (kg/d) in group 1 and 2, respectively (Table 4), which was significantly ($P<0.05$) higher in group 2 than that of group 1 due to higher body

Table 1. Chemical composition of feed ingredients offered (% DM basis)

Parameter	Concentrate (control)	Concentrate (treatment)	Green maize	Wheat straw
DM	89.14	89.23	12.98	90.07
OM	91.17	91.25	88.94	91.56
CP	20.24	20.65	8.74	3.12
CF	8.07	9.13	25.06	39.9
EE	3.89	3.91	2.01	1.32
NFE	58.97	57.56	53.13	47.22
NDF	37.11	38.24	52.47	77.78
ADF	13.48	14.40	32.8	50.43

Table 2. Fatty acid profile (% of total fatty acids) of feed ingredients offered

Fatty acids	Feed ingredients				
	Green fodder	Wheat straw	Concentrate (control)	Concentrate (treatment)	Bypass fat
Caprylic acid (C8:0)	0.59	ND	0.56	0.3	0.23
Capric acid (C10:0)	0.71	0.51	0.78	0.43	0.30
Lauric acid (C12:0)	3.31	8.67	1.89	2.05	0.65
Myristic acid (C14:0)	2.4	2.03	1.56	1.13	0.35
Myristoleic acid (C14:1)	0.67	ND	ND	0	0.54
Palmitic acid (C16:0)	21.77	21.56	14.37	15.83	13.09
Palmitoleic acid (C16:1)	1.02	0.32	1.48	1.23	0.53
Margaric acid (C17:0)	ND	3.12	ND	0.25	ND
Stearic acid (C18:0)	3.02	39.50	32.98	30.08	11.83
Elaidic acid (C18:1t9)	2.06	ND	2.65	3.11	8.32
Oleic acid (C18:1c9)	4.68	14.89	21.06	23.03	26.45
Linoleic acid (C18:2)	17.12	ND	2.73	1.98	32.67
Linolenic acid (C18:3)	38.87	ND	15.41	16.26	2.01
Arachidic acid (C20:0)	0.79	4.34	0.33	0.78	0.46
Total	97.01	94.94	95.80	96.46	97.43

ND, Not detected.

Table 3. Fortnightly changes in body weights (kg) of buffaloes fed experimental diets

Day	Group 1	Group 2
-60	670.33±21.24	713.88±10.98
-45	678.83±21.58	722.83±10.44
-30	688.25±21.44	733.19±10.48
-15	698.70±21.29	745.92±10.53
0*	622.47±19.89	662.27±10.54
Change in body weight		
-45	8.50±0.83	8.95±1.13
-30	9.42±0.36	10.36±1.06
-15	10.45^{a}±0.83	12.73^{b}±0.59
0*	-76.23^{a}±1.89	-83.65^{b}±2.56

*Day of parturition; $^{a, b}$Means having different superscripts in the same row differ significantly (P<0.05)

Table 4. Average nutrients intake and plane of nutrition in buffaloes fed experimental diets

Parameters		Group 1	Group 2
DMI (kg/day)		11.13^{c}±0.13	11.69^{d}±0.08
CPI (g/day)		915.45^{a}±4.81	954.88^{b}±5.50
TDNI (kg/day)		5.52^{a}±0.02	6.19^{b}±0.02
DMI (kg/100 kg BW)		1.63±0.02	1.60±0.02
CPI (kg/100 kg BW)		133.85±0.95	131.01±0.95
TDNI (kg/100 kg BW)		0.81^{a}±0.00	0.85^{b}±0.00
Plane of nutrition			
DM	Requirement * (kg/d)	11.70	12.20
	Deficit in DMI (%)	4.88	4.19
CP	Requirement * (g/d)	995.66	1040.89
	Deficit in CPI (%)	8.05	8.26
TDN	Requirement * (kg/d)	5.90	6.09
	Deficit in TDNI (%)	6.44	+ 1.64

*Requirements as per Kearl (1982); $^{c, d}$ means having different superscripts in the same row differ significantly (P<0.05); $^{a, b}$ means having different superscripts in the same row differ significantly (P<0.01).

weight. It was observed that prior to calving, feed intake was slightly lower in both groups. DMI/100 kg body weight was 1.63 and 1.60 kg/d in group 1 and 2, respectively. The results of present study indicated that there was no adverse effect of rumen protected fat and 1.60±0.02 protein supplementation on DMI of buffaloes. Crude protein intake (CPI) was 915.45 and 954.88 (g/d) in group 1 and 2, respectively, which was significantly higher (P<0.01) in group 2 than that of group 1 due to slightly higher DMI observed in this group. However, there was no difference among 2 groups in CPI/100 kg body weight. Total digestible nutrients intake (TDNI) was 5.52 and 6.19 (kg/d) in groups 1 and 2, respectively, which was significantly higher (P<0.01) in group 2 than that of group 1 due to supplementation of rumen protected fat. Similarly, higher TDNI/100 kg body weight was observed in group 2 than that of group 1. These results are in agreement with those of Tyagi *et al.* (2009b) who reported there was no adverse effect of pre partum bypass fat supplementation to crossbred cows. Walli and Sirohi (2004) also reported no adverse effect of feeding formaldehyde protected mustard cake on nutrient intake to crossbred cows.

The DMI was 4.88 and 4.19% and CPI was 8.05 and 8.26% lower than recommended (Kearl 1982) in group 1 and 2, respectively (Table 4), due to decrease in DMI before calving. However, TDN intake was undersupplied by 6.64% in group 1, whereas, in group 2 the intake was as per requirements due to supplementation of rumen protected fat. Though the DMI and CPI was lower than requirements in all the animals, the deficit in TDN intake was partially met in group 2 due to increased energy density of ration on supplementation of rumen protected fat in this group. The results of present study are in agreement with those reported

Table 5. Blood parameters and calving performance of buffaloes fed experimental diets

Parameters	Group 1	Group 2
Blood glucose (mg/dl)	58.79±0.67	57.39±1.23
NEFA (mg/l)	114.59±2.38	125.37±4.39
Plasma triglyceride (mg/dl)	21.93±1.13	24.09±1.47
Plasma cholesterol (mg/dl)	213.23±9.37	232.63±14.18
BUN (mg/dl)	22.49^{a}±0.72	17.29^{b}±1.15
Calving performance		
Calf weight (kg)	31.94^{c}±1.15	35.38^{d}±0.94
Calving %	100	100
Premature birth	1	-
Retention of foetal membranes	3	1

[c, d] Means having different superscripts in the same row differ significantly (P<0.05); [a, b] means having different superscripts in the same row differ significantly (P<0.01).

by Tyagi *et al.* (2009b).

Blood parameters: Blood glucose concentration (Table 5) was 58.79 and 57.39 mg/dl in group 1 and 2, respectively. The blood glucose concentration remained within the normal range (Fahey *et al.* 2002, Tyagi *et al.* 2009a). The reason may be a high metabolic rate of utilization of glucose and homeostatic mechanism of animal body that does not allow appreciable change in glucose level. Blood urea nitrogen (BUN) concentration (mg/dl) was lower (P<0.01) in group 2 (17.29) than that of group 1 (22.49), due to supplementation of rumen protected protein in group 2 buffaloes. The higher value of blood urea level in group 1 is indicative of less efficient utilization of dietary nitrogen for microbial protein synthesis due to higher ammonia level. Feeding of rumen protected protein not only results in more supply of amino acids, but also saves energy wasted in urea synthesis. Similar results were reported by Tiwari and Yadava (1994) on feeding formaldehyde treated mustard cake to buffalo calves and Sahoo and Walli (2005) to lactating goats.

Plasma NEFA concentration was 114.59 and 125.37 mg/litre in group 1 and 2, respectively. The overall turnover of the adipose tissues for deposition and mobilization of fat increases on bypass fat supplementation (Fahey *et al.* 2002), which might be the reason behind higher NEFA concentration observed in group 2. Plasma triglycerides concentration was 21.93 and 24.09 mg/dl in group 1 and 2, respectively. Plasma triglycerides were on higher side in group 2 on feeding of protected fat and protein, which is expected because of enhanced uptake of dietary fatty acids (Sklan and Tinsky 1993). Similar results were reported by Fahey *et al.* (2002) and Delbecchi *et al.* (2001). Plasma cholesterol concentration was 213.23 and 232.63 mg/dl in group 1 and group 2, respectively. Higher cholesterol concentration is associated with better reproductive performance in high yielding dairy cows, as it acts as a precursor of steroid hormones (Son *et al.* 1996). Fat supplementation is generally associated with higher cholesterol concentration (Grummer and Carroll 1988) which could not be observed in the present experiment. The results of present study are similar to that of Delbecchi *et al.* (2001) and Tyagi *et al.* (2009a).

Parturition related parameters: The effect of feeding rumen protected fat and protein on the calving performance of animals is given in Table 5. Average body weight of calves at the time of birth was 31.94 kg in group 1 and was higher (P<0.05) by 10.8% in group 2 than that of group 1, which may be due to one premature calving recorded in group 1. Higher birth weight of the calves observed in group 2 may be due to the higher energy intake as the TDNI was 0.37 kg/d more in group 2 than that of group 1(Table 4). Higher plasma progesterone concentration was reported on fat supplementation (Spicer *et al.* 1993, Son *et al.* 1996). The higher progesterone level may be responsible for better nourishment of foetus in the final stages of pregnancy. There was no case of premature birth observed in group 2, which may be due to higher concentration of linoleic acid (32.67%) in the supplemental bypass fat fed group 2 (Table 2). Linoleic acid can inhibit $PGF_2\alpha$ synthesis during pregnancy by competitive inhibition of enzymes D6-desaturase and cyclooxygenase responsible for desaturation and elongation of linoleic acid to form arachidonic acid (Staples *et al.* 1998). Due to check on the synthesis of $PGF_2\alpha$, there are lesser chances of contraction of smooth muscles of uterus and that might reduce the incidence of premature calvings or abortions. Our results were in agreement with those of Lammoglia *et al.* (1999) and Tyagi *et al.* (2009b). There was no effect of supplementation of rumen protected fat and protein on calving% between 2 groups. There was 1 case of premature birth in group 1 and no such case was observed in group 2. Three cases of retention of fetal membranes were observed in group 1 while 1 case was observed in group 2. The major problem associated with such calving related abnormalities is delay in reaching the peak yield and also lower peak milk yield which in turn reduces the full lactation milk production and also decreases post calving reproductive performance of buffaloes.

Our results indicated that rumen protected fat (@ 2.5% of DMI) and protein (formaldehyde treated cakes) supplementation 60 days pre partum, increased the calf birth weight and decreased the incidence of retention of foetal membranes and premature births due to higher TDN intake.

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