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Effect of Formaldehyde Treated Mustard Cake on Nutrient Utilization and Milk Production Performance in Crossbred Cows Fed Wheat Straw Based Diet

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ABSTRACT

An experiment was conducted to study the supplementation effect of formaldehyde treated mustard cake on its chemical composition, nitrogen fractions, in situ degradability, nutrient utilization and milk production performance in crossbred (Karan- Fries) dairy cows. Twelve lactating crossbred cattle were randomly divided into two groups on the basis of milk yield (10.12 kg/day) and days of calving (90 days) in a randomized block design. They were fed green berseem fodder, wheat straw and concentrate mixture supplemented with 0.9 kg untreated (raw) mustard cake as fed basis (8 per cent of total diet) in the control group, whereas untreated mustard cake was replaced by formaldehyde treated mustard cake in the treatment group to make both diets isonitrogenous and isocaloric. The roughage to concentrate ratio was 55:45 in both the diets. The cows were housed in individual byres and fed experimental diet for 90 days. The total dry matter intake apparently higher in the treatment group but values remained statistically was similar in both groups. The digestibility of dry matter, organic matter, crude protein, ether extract and neutral detergent fiber was not significantly different in both groups. However, average milk production(10.05kg/d) and fat corrected milk yield per day(11.03 kg) was significantly($P<0.05$) higher in treatment group as compared control group(8.99 and 10.18kg/d). Milk composition almost remained unaffected and was similar in both groups. Thus, rumen bypassing of protein fractions in present experimental study showed beneficial effect on milk production in medium producing crossbred cows.

Keywords Rumen bypass protein, Mustard cake, Crossbred cow, Milk production.

INTRODUCTION

Mustard cake (MC) is one of commonly used feed ingredient in ruminant diets and rich in many essential amino acids (*e.g.*, methionine and lysine), but high rumen degradability of its CP in the rumen reduces its nutritive value (Chatterjee andWalli, 2003). Formaldehyde (FA) treatment is an effective method to reduce the rumen degradability of proteins (Wulf and Sudekum, 2005)..Rapid and extensive degradation of valuable proteins in the rumen lead research to develop the concept of protein protection from ruminal degradation with the principal objective of enhancing the supply of essential amino acids to the productive animal and reduction of nitrogen losses as urea in the urine (Walli 2005 ; 2008).

MATERIALS AND METHODS

Whole mustard cake in raw and formaldehyde treated supplied by NDDB (National Dairy Development Board) Gujrat, India and stored in well ventilated store before use. Mustard cake was ground to pass a 1.0 mm sieve (Chatterjee and Walli 2003). The mustard cake was treated with 1.2 g of formaldehyde per 100 g CP of mustard cake followed by thorough mixing and stored in tightly sealed plastic bags for 7 days. During this period, formation of complexes between amide and aldehyde groups, occurred which can resist proteolysis attack in the rumen (Ashes 1995). Representative uniform samples (500 g) of treated and untreated mustard cake samples were retained for an in sacco nylon bag study and protein quality evaluation.Samples of raw and treated mustard cake were subjected to protein quality evaluation by in situ nylon bag technique and the laboratory method of N-fractionation.

Three crossbred rumen canulated male calves of 200-250 kg body weight were fed wheat straw and concentrate (maize 50%, groundnut cake 30%, wheat bran 17%, mineral mixture 2% and common salt 1%)

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along with 5 kg green, maintaining the roughage concentrate ratio of around 65:35.

Preweighed nylons bags (9 x 15 cm with 40 μ size) containing 5 g of milled raw and treated mustard cake (ground to pass 2.5 mm sieve) were tied by nylon threads and fastened tightly to three iron chains and placed in the rumen of three fistulated crossbred adult calves. The bags were taken out at intervals of 4, 8, 12, 16, 24, 36 and 40 hours for each feed. After removal, the bags were thoroughly washed in a water trough, till clear water emerged. The washed bags were then dried at 60°C for 24 h followed by 90° C for 24 h. The DM loss was measured from the residue left in the bag. The dried residue was further subjected to N estimation by kjeldhal's method, for estimating protein degradability. From the degradability data at different hours with respect to DM and CP degradability in rumen, the constants a, b, c from the expression p = a + bc/c+k (Orskov and Mc Donald 1979). were obtained by analysis, where 'p' is effective degradability, 'a' is the intercept on y axis, 'b' is the potentially degradable fraction, 'c' the rate constant and 'k' is the rumen out flow rate, taken as 0.04% h-1. The mustard cake samples (untreated and treated) were ground to pass 1 mm sieve. The partitioning dietary protein into different fractions was done according to Chalupa and Sniffen 1996. The values were expressed as A+B1, B2, B3 and C fractions which represented values of different protein fractions mentioned below :

1. A+B1: corresponds to non protein nitrogen, all globulins and some albumins. This N fraction is soluble in phosphate buffer and completely degradable in rumen.
2. B2: degradable in rumen and rest are digestible in lower tract which represents rest of albumin and all gluetins partially.
3. B3: mostly the prolamin, extension proteins and denatured proteins, hardly degradable in rumen, but partially digestible in the lower tract.
4. C: corresponds to Millard products and N bound to lignin. This fraction is neither degraded in rumen nor digestible in the lower tract.

Twelve crossbred (*Bos indicus* × *Bos taurus*) cows, weighing 361.87±15.55 kg, mostly in their third month of lactation and yielding around 10 liters of milk, were randomly divided into two groups of six animal each untreated and treated mustard fed groups. The animals were made free from ecto and endoparasites before start of the trial. The experiment was started in the summer month of June and ended in early winter. A 90 day lactation trial was conducted on the 12 cows after adapting them to a untreated mustard cake based diet for 2 weeks. The animals were kept in well ventilated byres with ad lib access to fresh water and having separate mangers for fodder and concentrate in addition to provision of ad lib access to fresh water. The animals were milked thrice a day (0500, 1200 and 1800 hours). Milk yield and dry matter intake (DMI) through different feeds was recorded for each animal separately. Milk samples from each animal in both groups were collected at weekly intervals in proportion to their milk yield (one over 100th). Milk of evening, morning, and noon were sampled, kept at 4°C until mixing was done after noon milking (bringing them all to room temperature) and analyzed for milk composition. Milk samples were analyzed for fat, protein, lactose, and solids not fat (SNF), using a pre-calibrated milk analyzer (Lacto Star, FUNKE GERBER, Article No. 3510, Berlin). Fat-corrected milk at 4% (4% fat-corrected milk, FCM) was calculated as per (Tyrell and Reid 1965). Body weights were recorded on 2 consecutive days at the start and end of the experimental period and fortnightly once throughout the experiment.

Both the groups of animals were fed concentrate having maize 50, groundnut cake 30, wheat bran 17, mineral mixture 2 and common salt 1 parts and green Berseem fodder ad lib to meet or exceed their requirements (NRC 2001). In addition to that, control group and experimental group were given 0.91 kg raw mustard cake and 0.91 kg formaldehyde treated mustard cake through concentrate.The concentrate was fed at the time of milking and berseem fodder at 11:00 and 16:00 h with recording of orts at 08:00 h daily. Feed offered was weighed at each feeding and calculated to result in about 10% orts, which were weighed and recorded daily. A conventional digestion trial of 7 days duration was conducted on both groups of animals in the mid-experiment period, keeping the 24-h record of intake of

feeds, feces voided out and orts if any. Aliquots of feces were taken daily separately and pooled for CP and DM estimation.

Representative pooled samples of berseem fodder, concentrate, untreated mustard cake, formaldehyde treated mustard cake, wheat straw, orts and faeces were taken and grounded to a particle size of 1 mm by a hammer mill after oven drying. Then representative samples of offered feeds and faces were analyzed for proximate composition (AOAC 2005). Fibre fractions were analyzed according to Van Soest et al. 1991 without sodium sulphite and heat stable amylase and expressed inclusive of residual ash.

Data obtained was analyzed by paired t test by (SYSTAT 7.0 1997) software and statistical significance was expressed at $P<0.05$.

RESULTS AND DISCUSSION

The chemical composition of feeds and fodders is given in Table 1 and effective protein degradability fractions of both untreated and formaldehyde treated mustard cake are mentioned in Table 2.The initial CP content of raw mustard cake was approximately equal to formaldehyde treated mustard cake (Table 1). By subjecting the mustard cake to formaldehyde treatment, the effective protein degradability and rumen degradable protein (RDP) content was reduced with obvious increase in the undegraded dietary protein (UDP) content.

The highly rumen degradable fraction (A+B1 fraction) was higher in the raw mustard cake as compared to formaldehyde treated mustard cake, showing that formaldehyde treatment had significant effect in the reduction of this fraction (Table 2). Slowly degradable RDP fraction was higher ($P<0.05$) in raw mustard cake as compared to formaldehyde treated mustard cake where as UDP was also significantly higher ($P<0.05$) in formaldehyde treated mustard cake than raw mustard cake. These results clearly showed that there was increase in the bypass ability of protein in mustard cake after formaldehyde treatment of raw mustard cake which imitates the effective protein degradability.

Treatment of proteins with formaldehyde is the most widely used process at the present time and it has been exploited commercially. Treatment of high quality proteins result in the formation of cross-links with amino group and makes the protein less susceptible to microbial attack (Czerkawski 1986).

Such treatments of protein rich feedstuffs has been shown to increase the protein digested in the intestine and nitrogen retention. The concentration of amino acids in the plasma is generally increased depending on tissue demands and the balance of amino acids supplied (Ferguson 1975). Formaldehyde treatment of mustard cake resulted in increasing the bypass value of its protein as reflected by reduction in effective protein degradability and a corresponding increase in UDP value of the formaldehyde treated mustard cake. The UDP content was also increased significantly in formaldehyde treated mustard cake over untreated mustard cake. The RDP content was significantly lower in formaldehyde treated mustard cake in comparison to untreated mustard cake.Similar results were reported by different workers during formaldehyde treatment of cakes (Yadav and

Table 1. Nutrient composition of feeds (gram per kilogram DM)

Feed	Berseem	Concentrate	Wheat straw	Untreated mustard cake	Treated mustard cake
Organic matter	834	871	885	898	914
Crude protein	210	233	36	348	350
Ether extract	35	37	43	107	78
Neutral detergent fiber	442	424	785	364	400
Acid detergent fiber	225	130	493	180	178
Hemicellulose	221	294	292	184	222
Total ash	165	128	115	101	85

Table 2. Protein fractions degradability of untreated and formaldehyde treated mustard cake (on DM Basis)

Particulars	Treated MC	Untreated MC
Crude protein(%)	35.00	34.82
Effective CP Degradability %	53.30	71.60
UDP(%)	16.35	9.88
RDP(%)	18.65	24.93

RDP : Rumen Degraded Protein UDP : Undegraded Protein

Chaudhary 2004; Misra et al. 2006; Bugallia and Chaudhary 2010).

The chemical composition of the treatment feeds is given in Table 3. The formaldehyde treatment did not have any major influence on the chemical composition, including the fiber fractions of the mustard cake. The DMI was not affected significantly due to feeding of formaldehyde treatment of mustard cake over untreated cake(Table 4).The roughage to concentrate ratio of the diets in the two groups was also remained similar i.e. 47.52:52.28 and 47.20:52.79 in untreated and formaldehyde treated mustard cake-fed groups, respectively. Digestibility coefficients of different nutrients were also did not vary significantly between the groups.

In the present experiment, feeding of formaldehyde treated mustard cake neither had any effect on the total DM intake of the animals, nor affected the total tract apparent digestibility of nutrients, including fiber fractions (Table 4). Sahoo and Walli 2008 on feeding raw and formaldehyde treated mustard cake in case of kids also found similar results and did not observe any significant difference in digestibility of nutrients except in digestibility of ether extract.

The weekly milk production in the experimental period versus control group is presented in Fig 1, whereas the average values are presented in Table 5.Average Milk yield (per day) and FCM yield were significantly($P<0.05$) different in both groups and found higher in formaldehyde supplementedgroups. However, the average percentage of milk composition i.e fat, protein, and SNF showed a non-significant variation ($P>0.05$) between the two groups fed either untreated mustard cake or formaldehyde treated mustard cake (Table 5).

The milk production remained higher in formaldehyde treated mustard supplemented cows than control animals received raw mustard cake. The milk production of crossbred cows after feeding formaldehyde treated mustard cake was higher (15 percent) when compared with their initial levels which confirmed the more efficient utilization of protein and amino acids for milk production. Similar trend was observed in case of fat corrected milk yield which also shown significant increased in comparison to control group cows. Similar results were reported by various workers by feeding bypass protein supplements in lactating animals which

Table 3. Different nitrogen fractions in untreated and formaldehyde treated mustard cake

Particulars	Untreated mustard cake		Treated mustard cake	
	% of DM	% of CP	% of DM	% of CP
A + B1[a]	19.29	55.39	17.12	48.91
B2[b]	13.72	39.41	17.88	51.09
B3[c]	1.71	4.91	1.89	5.41
C[d]	0.10	0.28	0.08	0.22

[a] N fraction soluble in phosphate buffer and completely degradable in rumen; [b] Degradable in the rumen and the rest are digestible in the lower tract; [c] N fraction hardly degradable in the rumen, but partially digestible in the lower tract; [d] Millard products and N bound to lignin

Table 4. Intake and nutrient digestibility in lactating crossbred cows supplemented with raw and formaldehyde treated mustard cake

Particulars	Treated MC Group	Untreated MC Group	P Value
Average BW (Kg)	356.81	366.93	-
Dry matter intake (g/day/animal)			
Berseem fodder (g/day)	3090	3060	-
Straw (g/day)	2230	2170	-
Concentrate (g/day)	5040	4820	-
Mustard cake (g/day)	910	910	-
Total (g/day)	11270	10960	0.299
Total(g/kg$^{0.75}$/day)	138.11	130.57	0.298
Total (Kg/100kg/day)	3.15	2.99	0.192
Roughage to concentrate ratio	47.20 : 52.79	47.72 : 52.28	-
Apparent digestibility			
Dry matter	0.627	0.649	0.368
Organic matter	0.696	0.720	0.199
Crude protein	0.697	0.677	0.570
Ether extract	0.744	0.726	0.566
Neutral detergent fiber	0.511	0.522	0.738
Acid detergent fiber	0.272	0.270	0.948
Hemicellulose	0.720	0.742	0.582

may be due to increased pool of amino acid at tissue level for utilization (Kaim et al. 1987; Hamilton et al. 1992; Gulati et al. 2002; Misra et al. 2006; Bugallia and Chaudhary 2010). Feeding of formaldehyde protected protein at higher levels in the ration of crossbred cattle (Sampath et al. 1997; Shelke and Thakur 2011) and buffaloes (Chatterjee and Walli 1998) improved the milk yield by 16-20 per cent. The increase in milk production reported on feeding protected protein in the present study could also be due to more availability of protein for digestion in the intestine, thereby increasing supply of precursors of milk production (Forster et al. 1983).

Increase in milk yield with a little depression of milk protein has been reported by Moore et al. 2004 on

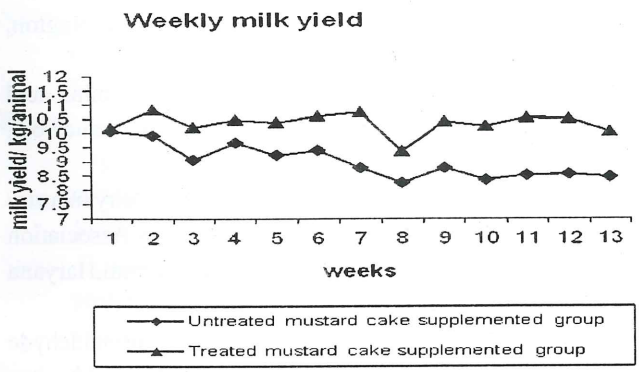

Figure 1. Milk yeild (kg/animal/day) in different treatment groups supplemented with mustard cake in crossbred cows

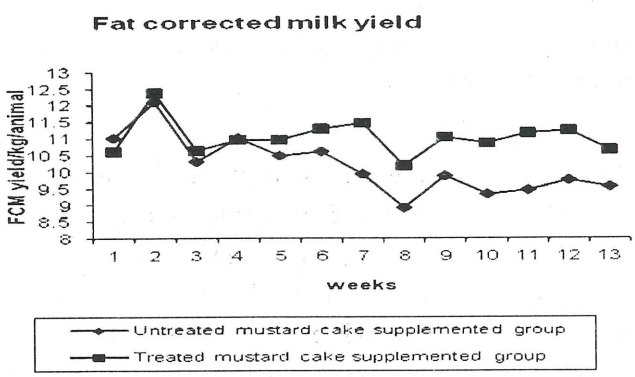

Figure 2. 4% Fat corrected milk yeild (kg/animal/ day) in different groups supplemented with mustard cake in crossbred cows

Table 5. Effect of mustard cake based rations on production performance in crossbred cows

Particulars	Treated MC Group	Untreated MC Group	P Value
Milk composition (g/Kg)			
Protein	25.30	26.40	0.123
Fat	48.80	44.50	0.364
Solids not fat	84.60	84.40	0.211
Total Solids	12.91	13.32	0.345
Milk yield (Kg/d)			
Initial milk yield	10.17	10.07	0.223
Final milk yield	10.05[a]	8.43[b]	0.004
Average milk yield	10.35[a]	8.99[b]	0.004
4% Fat corrected milk	11.03[a]	10.18[a]	0.005
Feed Conversion Efficiency			
Total DMI (Kg/day)	11.27	10.96	0.677
DMI /Kg Milk produced	1.08[a]	1.22[b]	0.003
Gross Energetic Efficiency	28.85	26.96	0.357

Different superscript in rows differ significantly at $P<0.05$

feeding rumen protected soybean based diet than control which was in accordance to the result of present experiment with rumen protected mustard cake based diet. However, total milk protein produced per animal remained the same in most of the reports as milk yield balances the depression. Santos et al.1998 after thorough review reported varying trend with milk fat also. Positive effect was reported by Voss et al. 1988, whereas others workers had reported no effect on milk fat percentage (Mohamed et al. 1988; Misra et al. 2006; Fathi Nasri et al. 2007; Bugallia and Chaudhary 2010). Milk lactose and SNF content also have not been affected on feeding different levels of rumen un-degradable protein or fat (Santos et al. 1998; Fathi Nasri et al. 2007).

CONCLUSION

On the basis of results of the present study, it was concluded from present study that protection of mustard cake protein increased the UDP content and supplementation of such cake was beneficial in terms of improved milk production in cows which clearly reflects better utilization of amino acids at tissue level.

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