

Influence of dietary cation-anionic difference on hemato-biochemical profile, mineral metabolism, post-partum reproductive and productive performance of Haryana cows

Bokan Abhay Mohanrao, Vinod Kumar*, Debashis Roy, Muneendra Kumar,
Mukesh Srivastava and Vivek Prasad Gupta

College of Veterinary Sciences & Animal Husbandry, U.P. Pt Deen Dayal Upadhyaya Pashu Chikitsa Vigyan Vishwavidyalaya,
Evam Go-Anusandhan Sansthan, Mathura- 281 001(U.P), India.

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ABSTRACT

Eighteen multiparous periparturient Haryana cows (*Bos indicus*) (3-4 parity) in last month of their pregnancy were blocked into three groups ($n=6$) and fed for 30 days pre-partum to 30 days post-partum. The objective was to determine the effects of manipulating dietary cations and anions on hemato-biochemical profile, mineral metabolism and performance of cows. Nutrient requirements of cows were met by feeding concentrate and roughage. Varying levels of salt supplementation were used to formulate mineral mixture having DCAD of +11, +21 and +31 meq (Na+K - Cl+S) / 100 g of feed. Feeding DCAD of +11, +21 and +31 meq/100 g of DM did not have any effect ($P>0.05$) on feed intake and body weight change during periparturient period. Blood calcium level was higher for the lowest DCAD (+11 meq/100 g of DM), suggesting that low DCAD helps in mobilization of stored Ca in periparturient cows. The pH of blood and urine increased curvilinearly with increasing DCAD and found maximum in +31 meq/100 g of DM DCAD fed group. Feeding of three different DCAD diets did not ($P>0.05$) have any adverse effect on reproductive performance but milk yield was significantly higher ($P<0.05$) in +21 meq/100 g of DM fed cows. Feeding of +21 meq/100 g of DM DCAD diets provided sufficient buffer and mineral balance during experimental period. Results suggest that feeding of low DCAD diet during advanced pregnancy helps to maintain blood Ca homeostasis and prevent chances of milk fever. However, feeding of high DCAD diet increase blood and urinary pH and leads to metabolic alkalosis during periparturient period.

Key words: Blood metabolite, Dietary cation anion difference, Mineral, Periparturient cows.

INTRODUCTION

In developing countries, non productive animals are fed on crop residues with little or no concentrate supplementation (Lanyasunya *et al.*, 2005). These crop residues are poor in energy, protein and mineral contents which greatly affects the nutrient availability, reproductive and productive performance of the animals (Prasad *et al.*, 1995). In the periparturient period from advanced pregnancy to early lactation, the clearance of Ca to the placenta ceases, but the lactational Ca demand increases rapidly (Ramberg *et al.*, 1984). The reduction of body fluids pH by addition of low dietary cation-anion difference (DCAD) diet can help in Ca mobilization (Takagi and Block, 1988; Bushinsky *et al.*, 1993; Schonewille *et al.*, 1994). The direct implementation of the findings obtained from the studies executed on exotic dairy cows to the indigenous breed that differs in physiological aspects and thrive on dry fodder dominated feeding system would not be a scientific approach. Therefore, this study was conducted to assess the effect of altering DCAD on hemato-

biochemical profile, mineral metabolism, and performance of Haryana cows.

MATERIALS AND METHODS

This experiment was conducted in the Instructional Livestock Farm Complex (ILFC), College of Veterinary Science & Animal Husbandry, Mathura, India. All procedures related to animal care were conducted with the approval of the Institutional Animal Ethics Committee (IAEC) constituted as per the article number 13 of the CPCSEA-rules, laid down by Government of India.

Animals, feeding and management: A total of 18 periparturient Haryana cows were selected from herd of ILFC and allocated to three dietary treatments having six cows in each group on the basis of their expected producing ability (EPA) (Lush, 1945). The experimental cows were housed in well ventilated shed and individually tied by rope with a provision of feeding and watering trough. The experimental cows were free from anatomical, physiological and infectious

*Corresponding author's e-mail: vinodsidhu@rediffmail.com.

disorders. All the cows were maintained for a period of 60 days from 30 days pre-partum to 30 days post-partum. The nutrient requirements of animals were met by feeding concentrate mixture, maize fodder and wheat straw (NRC, 2001). Ingredient and chemical composition of basal diet fed to the experimental cows is presented in Table 1.

Dietary cation-anion difference: The DCAD was calculated by equation of Tucker *et al.* (1992) using the quantities of the salts administered and the measured mineral concentrations and dry matter intake of all feeds.

$$\text{DCAD (meq/100 g DM)} = (\text{Na} + \text{K}) - (\text{Cl} + \text{S}).$$

The DCAD offered before and after calving was manipulated by adding salts of either Na, Cl, or S such that the three treatments received a planned DCAD of +11, +21 and +31 meq/100 g DM. MgSO_4 , MgCl_2 and CaCl_2 were used to reduce the DCAD, and NaHCO_3 was used to increase it. The cows of three respective groups received 83, 50 and 109 g DCAD manipulated mineral mixture per cow per day by mixing in concentrate mixture.

Chemical composition and biochemical analysis: The experimental cows were monitored daily for DMI and milk yield. Cows were weighed and blood samples were collected at 30, 15 days pre-partum and at 7 day post-partum. Blood samples were collected in sterile heparinized vacutainer (Becton Drive, Franklin Lakes, NJ, USA) from jugular vein puncture, posing minimum disturbance to the animal. Collected blood samples were used for the analysis of hemato-biochemical and mineral profile.

The proximate composition of experimental diets and residue left was analysed in triplicate (AOAC, 2000) and crude fibre by acid digestion (1.25%) followed by alkali digestion (1.25%). NDF and ADF were analyzed according to the methods described by Van Soest *et al.* (1991).

Blood samples were used for determination of pH, glucose, hemoglobin (Hb), packed cell volume (PCV), total leukocyte count (TLC), differential leukocyte count (DLC), creatinine, Ca, P, Na and K. Remaining blood samples were centrifuged at $1200 \times g$ at 4°C for 20 min to separate the plasma for the analysis of minerals and creatinine. The pH of whole blood was determined by using an electronic pH meter (Riviera), however, blood Hb, PCV, TLC, and DLC were estimated as per method described by Jain (1986). Glucose level in blood was estimated by method described by Hultmann (1959). Na, K and Cl were estimated on flame photometer after digestion. Plasma was used for estimation of minerals by using atomic absorption spectrophotometer (Model -AA 400, Perkin Elmer Atomic absorption Spectrophotometer, Perkin Elmer Corporation, USA). Creatinine level in plasma was analyzed by Span diagnostics kits using modified Jaffe's Reaction (Jaffe, 1886).

A sample of urine, from midstream, was collected in a 30-ml container. Within 30 min of collection, pH was measured, and 15 ml of sample was frozen for mineral and creatinine analysis. Urinary Ca, P, Na, K and Cl were expressed as ratios to creatinine concentration to overcome variations in urine volume among animals (Roche *et al.*, 2002). Creatinine in urine was analyzed by Span diagnostics kits using modified Jaffe's Reaction (Jaffe, 1886) however mineral content in urine was analyzed by using atomic absorption spectrophotometer.

Reproductive and productive parameters: All the experimental cows were kept under close observation during parturition. One week before expected date of calving, the animals were kept individually in calving pens and the period up to the expulsion of the fetal membranes was recorded. Reproductive parameters studied include birth weight of

Table 1: Ingredients and nutrient composition of Experimental diets (% DM basis) offered to periparturient cows

Attribute	Formulated DCAD (mEq/100 g of DM)		
	+11	+21	+31
Ingredient (% on DM basis)			
Wheat straw	52.02	51.35	51.51
Maize fodder	28.44	29.05	28.78
Barley grain	9.77	9.80	9.86
Wheat bran	3.91	3.92	3.94
Mustard cake	4.88	4.90	4.93
Gram chunni	0.98	0.98	0.99
Dicalcium phosphate	0.39	0.24	0.51
CaCl_2	0.29	-	-
CaCO_3	-	0.18	0.38
NaCl	0.21	0.13	-
NaHCO_3	-	-	0.28
MgCl_2	0.02	-	-
MgSO_4	-	0.01	0.03
Trace mineral mixture ¹	0.06	0.03	0.08
Composition ² (%)			
CP	8.31	8.34	8.35
TDN	57.57	57.67	57.66
Ash	9.22	9.23	9.25
Ca	0.42	0.39	0.40
P	0.29	0.30	0.30
Na	0.12	0.12	0.14
K	0.21	0.21	0.22
Cl	0.24	0.22	0.21
S	1.00	0.08	0.08
DCAD ³ mEq per 100 g DM	+11	+21	+31

¹Trace mineral mixture contained FeSO_4 (41.60 %), CuSO_4 (34.94%), ZnSO_4 (12.48%), CoCl_2 (0.83%), MnCl_2 (9.98%) and KI (0.17%). ²Calculated values ³DCAD = (Na + K) - (Cl + S) mEq/100 g of dietary DM (Tucker *et al.*, 1992) Abbreviation: DCAD, Dietary cation-anion difference; TDN: Total digestible nutrients

calves, total time required in the act of parturition, assistance during parturition and mortality of calves if any among 3 groups. In order to study the uterine involution period, rectal palpation was started one week postpartum and performed twice a week until completion of involution. Different locations of the uterine horns (abdominal, half pelvic and pelvic) were palpated. Pelvic location of the uterine horns was considered to be the indication of completion of uterine involution (Chaudhry, 1985). Erect and turgid condition of the uterine horns was also considered to be the indication of completion of uterine involution.

Milk yield record of post calving 30 days was averaged and the 30th day milk was analysed with Automatic Funke-gerbe measuring range 0.00% to 44.00%), protein (measuring range 0.00% to 10.00%), lactose (measuring range 0.00% to 10.00%) and SNF (measuring range 0.00% to 15.00%).

Statistical analysis: Data were analyzed using the MIXED procedure of statistical software package SPSS version 19 (SPSS for windows, V19.0; SPSS Inc., Chicago, IL, USA). Repeated measures on DMI, body weight, haemato-biochemical and fractional mineral excretion were analyzed using mixed model analyses. The statistical model was used to estimate sampling day effect, treatment group (DCAD), and their interaction:

$$Y_{ijk} = \mu + T_i + D_j + (T \times D)_{ij} + e_{ijk}$$

Where, Y_{ijk} = dependent variable; μ = overall mean of the population; T_i = mean effect of the treatment group

(DCAD) D_j = mean effect of day of sampling ($j = -30, -15$ and 7 days of parturition) with day as a repeated factor; $(T \times D)_{ij}$ = effect of the interaction between effect of DCAD and day of sampling; e_{ijk} = unexplained residual element assumed to be independent and normally distributed.

RESULTS AND DISCUSSION

Nutrient intake and body weight changes: The nutrient intake and body weight changes in cows of three respective groups are summarized in Table 2. The intake of DM, CP, and TDN was not affected ($P > 0.05$) by feeding of three different DCAD manipulated diets. However intake of DM, CP, and TDN was increased ($P > 0.05$) towards calving and increase was found maximum at 15 days before calving in all three respective groups. Present findings were similar to findings of Hersom *et al.* (2010) and Shahzad *et al.* (2011) who also found no effect of feeding similar DCAD diets. In contrast to present findings, other researchers (Tucker *et al.*, 1988a; Hu and Murphy 2004; Hu *et al.*, 2007) reported increased DMI with increasing DCAD level. This may be due to lower DCAD levels than those used in the present study. The absence of an effect of DCAD on DMI in the present study might be less difference in treatment groups. In present findings, the dietary Ca, P, Na and Cl concentrations were enough to meet NRC (2001) recommendations (0.44, 0.26, 0.14 and 0.20%, respectively). The intake of CP, TDN, Ca and P were higher to those recommended by Ranjhan (1998) for Indian cows in last two month of pregnancy.

Table 2: Nutrient intake and body weight changes in periparturient cows fed DCAD modified diets

Parameter	Formulated DCAD (mEq/100 g of DM)			SEM	P-value		
	+11	+21	+31		DCAD	Time	DCAD×Time
BW changes (kg)							
-30 days	356.4	356.18	357.77	8.34	0.69	0.03	0.98
-15 days	360.13	375.17	374.65	8.17			
+7 days	332.55	341.4	343.84	7.93			
DMI (kg/day)							
-30 days	7.67	7.62	7.51	0.07	0.22	<0.001	0.93
-15 days	8.73	8.75	8.68	0.01			
+7 days	8.55	8.57	8.49	0.01			
DMI (% of BW)							
-30 days	2.17	2.15	2.10	0.05	0.35	<0.001	0.99
-15 days	2.45	2.34	2.31	0.06			
+7 days	2.52	2.52	2.47	0.06			
CP intake (g/day)							
-30 days	637.28	635.80	627.18	6.05	0.37	<0.001	0.92
-15 days	725.23	730.25	724.52	1.08			
+7 days	709.78	714.73	709.03	1.08			
TDN intake (kg/day)							
-30 days	4.26	4.40	4.33	0.04	<0.001	<0.001	0.83
-15 days	4.85	5.05	5.01	0.03			
+7 days	4.74	4.94	4.89	0.02			

Abbreviation: SEM, Standard error of mean (n=6)

No differences ($P>0.05$) in body weight changes were observed in three respective groups (Table 2). The increased body weight towards calving was due to rapid development of fetus. Similarly, reduced postpartum body weight of cows is due to calving and expulsion of fetal membrane.

Blood and urinary metabolites: Blood plasma concentration of Ca, P, Na, K and Cl are presented in Table 3. Blood plasma concentration of Ca increased linearly with decreased DCAD level and found maximum in + 11 mEq/100 g of DM fed cows. The results of present study are in line with finding of Tucker *et al.* (1992) and Jackson *et al.* (1992). Increased plasma Ca level in + 11 mEq/100 g of DM fed diet might be due to higher dietary Ca level. High plasma Ca level may also be attributed to increased Ca absorption from the alimentary tract (Lomba *et al.*, 1978) and Ca mobilization from bones (Bushinsky *et al.*, 1993) due to mild metabolic acidosis, induced by feeding low DCAD diet. Feeding a low DCAD increased the flow of Ca through the readily exchangeable Ca pool (Takagi and Block, 1988) and enhanced the concentration of ionized Ca in the blood (Oetzel *et al.*, 1988).

Increased plasma P concentration was positively correlated with the period of feeding but its concentration was similar ($P<0.05$) in all three group showing lack of marked effect of DCAD on plasma P level. These results are in concordance with findings of earlier reports showing no effect of feeding different DCAD diets on plasma P concentration (Delaquis and Block, 1995; Shahzad *et al.*,

2011). Increased plasma P concentration towards calving, within period is due to increased DMI.

Increased plasma Na^+ concentration was positively correlated with increase in the dietary DCAD levels and plasma Na^+ level was found maximum in +31 mEq/100 g of DM DCAD fed cows. The present findings are in agreement with Tucker *et al.* (1988b) who also reported increased plasma Na^+ at high DCAD levels. Increased plasma Na^+ concentration in +31 mEq/100 g of DM DCAD fed periparturient cows is due to higher intake of Na^+ . +31 mEq/100 g of DM DCAD manipulated diet has 0.14% higher Na content as compared to +21 mEq/100 g of DM DCAD diet. Plasma K^+ concentration in periparturient cows remained unaffected ($P<0.05$) by feeding different DCAD manipulated diets. These results are in consistent with previous reports in lactating cows and calves (Tucker *et al.* (1988b) and buffaloes (Shahzad *et al.*, 2011).

A linear increase in plasma Cl^- concentration was observed with decrease in the DCAD levels and plasma Cl^- concentration was found maximum in + 11 mEq/100 g of DM fed cows. Similarly, Tucker *et al.* (1988b) also reported a linear increase in plasma Cl^- concentration with decrease in the DCAD levels (+20 to -10 mEq/kg of DM). Increased plasma Cl^- concentration in low DCAD fed cows might be due to high intake of CaCl_2 used for reduction of dietary DCAD level as reported by Tucker *et al.* (1991). The plasma Na, K and Cl level in present finding is closely associated with increased blood pH in cows fed on high DCAD diet.

Table 3: Plasma mineral profile of periparturient cows fed DCAD modified diets

Parameter	Formulated DCAD (mEq/100 g of DM)				P-value		
	+11	+21	+31	SEM	DCAD	Time	DCAD×Time
Plasma Ca (mg/dl)							
-30 days	9.50	9.40	9.52	0.15	0.006	0.09	0.21
-15 days	10.40	9.56	9.24	0.25			
+7 days	10.46	9.89	9.46	0.24			
Plasma P (mg/dl)							
-30 days	5.75	5.93	6.07	0.11	0.62	0.015	0.78
-15 days	6.14	6.10	6.09	0.13			
+7 days	6.35	6.23	6.43	0.08			
Plasma Na (mEq/L)							
-30 days	142.16	145.77	146.03	0.77	<0.001	0.014	0.005
-15 days	143.16	144.60	151.73	1.46			
+7 days	142.36	146.63	151.33	1.40			
Plasma K (mEq/L)							
-30 days	4.33	4.21	4.36	0.06	0.45	0.98	0.58
-15 days	4.67	4.20	4.31	0.06			
+7 days	4.32	4.33	4.29	0.05			
Plasma Cl (mEq/L)							
-30 days	99.23	104.00	98.20	1.49	0.002	0.17	0.004
-15 days	103.20	98.40	98.73	0.88			
+7 days	102.30	97.90	95.50	1.08			

Table 4: Haemato-biochemical profile of periparturient cows fed DCAD modified diets

Parameter	Formulated DCAD (mEq/100 g of DM)			SEM	P-value		
	+11	+21	+31		DCAD	Time	DCAD × Time
Glucose (g/dl)	46.05	47.35	48.05	0.15	0.44	0.75	0.13
Blood pH	6.93	7.21	7.28	0.23	<0.001	0.16	0.005
Blood Hb (%)	11.17	11.78	10.31	0.16	0.001	0.32	<0.001
PCV (%)	37.33	38.66	36.67	0.75	0.45	0.32	0.53
TLC ($\times 10^6/\text{mm}^3$)	3.78	3.71	4.01	0.06	0.053	0.47	0.65
Lymphocyte (%)	60.89	60.00	60.56	0.55	0.67	0.95	0.95
Neutrophils (%)	33.33	34.00	33.11	0.35	0.27	0.54	0.54
Eosinophils (%)	3.53	3.60	3.57	0.07	0.82	0.15	0.09
Monocytes (%)	5.30	4.75	4.73	0.07	0.90	0.64	0.64

Haemato-biochemical profile: Haemato-biochemical profile of periparturient cows fed DCAD modified diets is presented in Table 4. In present study, blood glucose level was found similar ($P>0.05$) and within normal range in all three groups. Glucose plays a fundamental role of fetal development, especially during advanced pregnancy (Bell, 1995). The decrease in blood pH with decreasing DCAD approached significance ($P < 0.5$), and cows receiving the lowest DCAD (+ 11 mEq/100 g of DM) had a significantly lower blood pH than the other two treatments (Table 4). This supports the theory of Stewart (1981, 1983), that an increased dietary concentration of Cl and S would reduce the pH of body fluids. Results of present findings are also in agreement with earlier report (Shahzad *et al.*, 2011). Low blood pH in + 11 mEq/100 g of DM DCAD fed cows might be due to increased supplemental level of Cl by dietary addition of CaCl_2 and MgCl_2 .

In contrast to pattern of blood pH, blood Hb level decreased as the supplemental level of DCAD increased ($P<0.05\%$) and it was found minimum in +31 mEq/100 g DM of DCAD fed group (Table 4). Increased dietary supplemental level of DCAD is responsible for increased blood base. Increased blood HCO_3^- concentration in high DCAD fed diet is mainly due to increased intake of NaHCO_3 (Block, 1994). PCV, neutrophil, eosinophil and monocyte count was found similar in all three groups. The PCV values in present study were similar to earlier reports of Sivaraman *et al.* (2002) and Mirzadeh *et al.* (2010) in advanced pregnant

cows. The values of neutrophil, eosinophil and monocyte in present study were similar to findings of earlier report (Parrish *et al.*, 2010) in advanced pregnant cows during summer season.

Urinary parameters: Urinary pH and mineral excretion of periparturient cows fed on three levels of DCAD based diet is presented in Table 5. Similar to blood pH, urinary pH was also found lower ($P<0.05$) in +11 meq/100 g of DM fed cows. Reduced urinary pH in +11 meq/100 g of DM fed group is due to feeding of high acidogenic salt. The results of present study are consistent with Tucker *et al.* (1988b). This increased urine pH at high DCAD levels might be due to decreased concentrations of H^+ and increased HCO_3^- concentration (Fredeen *et al.*, 1988; Tucker *et al.*, 1988b). Increased urine pH at high DCAD diets may be associated with increased blood HCO_3^- concentration and reduced serum Cl⁻ concentration. Similar results were also observed by Tucker *et al.* (1988b). Low DCAD diets resulted in increased Cl⁻ concentration in the diet and increased urinary excretion of Cl⁻. Changes in urine pH might be due to the animals attempt to balance any change in the blood by excreting acidic or alkaline urine.

In present study, excretion of Ca was significantly higher in low DCAD fed groups (+ 11 meq/100g of DM). The present findings of increased urinary Ca excretion are in agreement with other researchers (Fredeen *et al.*, 1988; Shahzad *et al.*, 2011).

Table 5: Average urine mineral excretion and pH of periparturient cows fed on three levels of DCAD based diet

Parameter	Formulated DCAD (mEq/100 g of DM)			SEM	P-Value		
	+11	+21	+31		DCAD	Time	DCAD × Time
Urine pH	6.04	7.03	7.74	0.88	<0.001	<0.001	<0.001
Fractional mineral excretion ¹							
Ca/creat	2.77	2.13	1.87	0.14	<0.001	<0.001	<0.001
P/creat	0.038	0.039	0.037	0.001	0.659	0.07	0.09
Na/creat	6.49	7.81	8.48	0.30	<0.001	<0.001	<0.002
K/creat	118.13	110.93	103.56	2.37	0.010	0.051	0.02
Cl/creat	82.49	70.94	66.20	2.65	0.005	<0.001	<0.001

¹Fractional excretion = ([urinary mineral] / [plasma mineral]) × ([plasma creatinine] / [urinary creatinine]) × 100; (Vander, 1991).

Table 6: Effect of feeding DCAD manipulated diet during periparturient period on reproductive parameters in cows

Attribute	Formulated DCAD (mEq/100 g of DM)			SEM	P-value	Normal value
	+11	+21	+31			
Parturition time (h) ¹	4.3	4.6	4.5	0.20	0.82	2.5-7
Placenta expulsion period (h)	4h 27m	4h 21m	5h 25m	0.27	0.25	3-8
Birth weight of calves (kg)	20.27	20.53	20.48	0.17	0.63	18-28
Uterine involution period (days)	34.3	33.0	36.6	1.01	0.84	28-45
Assistance during parturition	Nil	Nil	Nil	-	-	-
Mortality of calf	Nil	Nil	Nil	-	-	-

¹Parturition time (Dilatation of cervix and expulsion of fetus) (stage 1 & 2)

Table 7: Milk yield and milk composition of post parturient cows fed three different DCAD diets

Parameter	Formulated DCAD (mEq/100 g of DM)			SEM	P-value*
	+11	+21	+31		
Milk yield, kg per day	3.67 ^b	4.89 ^a	4.31 ^{ab}	0.43	0.04
Milk composition, %					
Fat	4.15	4.06	4.13	0.03	0.06
Protein	2.98	2.93	2.92	0.05	0.07
Lactose	4.34	4.34	4.36	0.04	0.34
SNF	7.21 ^a	7.75 ^b	8.01 ^b	0.25	0.03
Specific gravity	1.03	1.03	1.03	0.004	0.62
Acidity	0.137	0.133	0.140	0.015	0.41

*Means bearing different superscript in row (a, b) differs significantly (P<0.05)

In present findings, urinary excretion of P was not affected (P>0.05) by feeding three different DCAD manipulated diet. Urinary P excretion in present findings are similar to the earlier reports in lactating animals (Delaquis and Block, 1995; Shahzad *et al.*, 2011). In contrast to present finding, West *et al.* (1991) has shown a cubic response in urinary P to reductions in DCAD (from + 31 to 11 mEq/100 g DM) in lactating cows. Increased urinary excretion of Na in high DCAD fed diet is agreement with Tucker *et al.* (1988b). However, urinary excretion of K⁺ and Cl⁻ was found significantly (P<0.05) lower in high DCAD fed groups. The higher K⁺ excretion at high DCAD may be due to higher Na⁺ excretion in this group. Higher Cl⁻ excretion noticed in present study in low DCAD group was in agreement with earlier reports (Tucker *et al.*, 1988b; Hu *et al.*, 2007) who reported decreased Cl⁻ excretion at high DCAD diets.

Reproductive traits and productive parameters: The differences in reproductive parameters, *i.e.*, placental expulsion period, uterine involution period, birth weight of calves, assistance during parturition and calf mortality among three groups were similar (P>0.05; Table 6). Normal time required for foetus expulsion (stage 1 and 2) ranged from 2.5 to 7 hours. In the present study, placenta expulsion time of cows in three respective groups was within reported ranges *i.e.* 4.3, 4.6 and 4.5 h. Similarly, time taken for involution of uterus was also within reported time. Birth weight of calf born was around 20 kg which was also found similar in all

three groups. There was no any calf mortality and assisted birth in the present study.

In present study average milk yield were 3.67, 4.89 and 4.31 kg/day in +11, 21 and 31 meq/100 g of DM DCAD diet group, respectively (Table 7). Milk yield was found significantly higher (P<0.05) in +21 meq/100 g of DM DCAD fed group. There was no significant (P>0.05) variation in milk composition due to feeding different DCAD diets except in SNF which was significantly (P<0.05) lower in +11 mEq/100 g of DM DCAD fed cows. The results of present findings are in agreement with other workers (Tucker *et al.*, 1988a; Delaquis and Block, 1995; Hu *et al.*, 2007; Sharif *et al.*, 2008) who also found no alteration in milk composition in different DCAD fed groups. This might be due to less difference in dietary DCAD ranges. In contrast to present findings, other researchers reported great variation in milk composition of cows fed on different DCAD diets having much wider dietary DCAD ranges (Roche *et al.*, 2003; Wildman *et al.*, 2007).

CONCLUSION

Supplementation of low cations anions manipulated diet to periparturient cow may be beneficial in maintaining blood Ca homeostasis and preventing chances of milk fever. However, feeding of high DCAD diet increase blood and urinary pH and leads to metabolic alkalosis during periparturient period. Feeding of medium DCAD (21 mEq/100g DM) diet helps to improve milk production without altering milk composition.

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REFERENCES

- AOAC (2000). Official Methods of Analysis, 16th Ed. Association of Official Analytical Chemists, Washington, DC, USA.
- Bell, A.W. (1995). Regulation of organic nutrient metabolism during transition from late pregnancy to early lactation. *J. Anim. Sci.*, **73**: 2804-2819.
- Block, E. (1994). Manipulation of dietary cation-anion difference on nutritionally related production diseases, productivity, and metabolic responses of dairy cows. *J. Dairy Sci.*, **77**: 1437-1450.
- Bell, A.W. (1995). Regulation of organic nutrient metabolism during transition from late pregnancy to early lactation. *J. Anim. Sci.*, **73**: 2804-2819.
- Bushinsky, D.A., Wolbach, W., Sessler, N., Mogilevsky, R. and Levi-Setti, R. (1993). Physicochemical effects of acidosis on bone calcium flux and surface ion composition. *J. Bone Min. Res.*, **8**: 93-102.
- Chaudhry, M.A. (1985). Studies on postpartum buffaloes: changes in reproductive organs and onset of estrus after parturition. *MSc (Hons) Thesis*, College of Veterinary Sciences, Lahore, Pakistan.
- Delaquis, A.M. and Block, E. (1995). Dietary cation-anion difference, acid-base status, mineral metabolism, renal function, and milk production of lactating cows. *J. Dairy Sci.*, **78**: 2259-2284.
- Fredeen, A.H., DePeters, E.J. and Baldwin, R.L. (1988). Effects of acid-base disturbances caused by differences in dietary fixed ion balance on kinetics of calcium metabolism in ruminants with high calcium demand. *J. Anim. Sci.*, **66**: 174-184.
- Hersom, M.J., Hansen, G.R. and Arthigton, J.D. (2010). Effect of dietary cation-anion difference on measures of acid-base physiology and performance in beef cattle. *J. Dairy Sci.*, **88**(1): 374-82.
- Hu, W. and Murphy, M.R. (2004). Dietary cation-anion difference effects on performance and acid-base status of lactating dairy cows: A meta-analysis. *J. Dairy Sci.*, **87**: 2222-2229.
- Hu, W., Murphy, M.R., Constable, P.D. and Block E. (2007). Dietary cation-anion difference effects on performance and acid-base status of dairy cows postpartum. *J. Dairy Sci.*, **90**: 3367-3375.
- Hultmann, E. (1959). Rapid specific method for determination of aldohexoses (aldosaccharides) in body fluids. *Nature* **103**: 108-109.
- Jackson, J.A., Hopkins, D.M., Xin, Z. and Hemken, R.W. (1992). Influence of cation anion balance on feed intake, body weight gain, humoral response of dairy calves. *J. Dairy Sci.*, **75**: 1281-1286.
- Jaffe, M. (1886). Über den Niederschlag, welchen Pikrinsäure in normalem Harn erzeugt und über eine neue Reaction des Kreatinins. *Zeitschrift für physiologische chemie* **10**(5): 391-400.
- Jain, C.N. (1986). Schalm's Veterinary Hematology. 4th edn., Lea and Febiger, Philadelphia.
- Lanyasunya, T.P., Musa, H.H., Yang, Z.P., Mekki, D.M. and Mukisira, E.A. (2005). Effects of poor nutrition on reproduction of dairy stock on smallholder farms in the tropics. *Pak. J. Nutr.*, **4**(2): 117-122.
- Lomba, F., Chauvaux, G., Teller, E., Lengele, L. and Bienfet, V. (1978). Calcium digestibility in sows as influenced by the excess of alkaline ions over stable acid ions in their diets. *Br. J. Nutr.*, **39**: 425-429.
- Lush, J. (1945). Animal Breeding Plans, Iowa State University Press, Ames, Iowa, USA.
- Mirzadeh, K., Tabatabaei, S., Bojarpour, M. and Mamoei, M. (2010). Comparative study of hematological parameters according strain, age, sex, physiological status and season in Iranian cattle. *J. Anim. Vet. Adv.* **9**(16): 2123-2127.
- National Research Council. (2001). Nutrient Requirements of Dairy Cattle. 7th rev. edn. National Academy Press, Washington, DC.
- Oetzel, G.R., Olson, J.D., Curtis, C.R. and Fettman, M.J. (1988). Ammonium chloride and ammonium sulfate for prevention of parturient paresis in dairy cows. *J. Dairy Sci.*, **71**: 3302-3309.
- Parrah, J., Moulvi, B.A., Hussain, S.S., Athar, H., Malik, H.U., Mir, M.S. and Darzi, M.M. (2010). Peritoneal fluid cytology in clinical cases of bovine obstructive urolithiasis. *Iranian J. Vet. Sci. Tech.*, **2**(1): 17-24.
- Prasad, S., Kaushish, S.K. and Mittal, J.P. (1995). Grazing behaviour of *Rathi* cows under hot arid conditions. *Indian J. Anim. Prod. Mngmt.* **11**(1): 49-53.
- Ramberg, C.F., Mayer, G.P., Kronfeld, D.S., Phang, J.M. and Berman, M. (1984). Calcium homeostasis in cows, with special reference to parturient hypocalcaemia. *Am. J. Physiol.* **246**: 698-704.
- Ranjhan, S.K. (1998). Nutrient Requirement of Livestock and Poultry. Directorate of Information and Publication of Agriculture, Krishi Anusandhan Bhawan, Indian Council of Agricultural Research, New Delhi.

- Roche, J.R., Dalley, D.E., Moate, P., Grainge, C., Rath, M. and O'Mara, F. (2003). Dietary cation-anion difference and the health and production of pasture-fed dairy cows. 1. Dairy cows in early lactation. *J. Dairy Sci.*, **86**: 970–978.
- Roche, J.R., Morton, J. and Kolver, E.S. (2002). Sulfur and chlorine play a non-acid base role in periparturient calcium homeostasis. *J. Dairy Sci.*, **85**: 3444–3453.
- Schonewille, J.T., Van't Klooster, A.T., Dirkswager, A. and Bayen, A. (1994). Stimulatory effect of an anion (chloride)-rich ration on apparent calcium absorption in dairy cows. *Livest. Prod. Sci.*, **40**: 233–240.
- Shahzad, M.A., Sharif, M., Nisa, M., Sarwar, M., Khalid, M.F. and Saddiqi, H.A. (2011). Changing certain dietary cationic and anionic minerals: Impact on blood chemistry, milk fever and udder edema in buffaloes during winter. *Afr. J. Biotech.*, **10**(62): 13651–13663.
- Sharif, M., Mahr-un-Nisa, Sarwar, M. and Shahzad, M.A. (2008). Influence of varying levels of dietary cation anion difference on ruminal characteristics, acid base status and milk yield of early lactating animals (a review). *Pak. J. Agric. Sci.*, **45**(2): 288–296.
- Sivaraman, T., Shanmugaseundaram, S., Arunachalam, S., Sivakumar, T. (2002). Blood profile of Jersey crossbred cows under different physiological stages. *Indian J. Anim. Res.*, **36**(2): 114–117.
- Stewart, P.A. (1981). How to understand acid-base-A quantitative acid-base primer for biology and medicine. Edward Arnold, London.
- Stewart, P.A. (1983). Modern quantitative acid-base chemistry. *Can. J. Physiol. Pharmacol.*, **61**: 1444–1461.
- Takagi, H. and Block, E. (1988). Effect of manipulating dietary cation-anion balance on calcium kinetics in normocalcemic and EGTA infused sheep. *J. Dairy Sci.*, **71**(Suppl 1): 153 (abstr.).
- Tucker, W.B., Harrison, G.A. and Hemken, R.W. (1988a). Influence of dietary cation-anion balance on milk, blood, urine, and rumen fluid in lactating dairy cattle. *J. Dairy Sci.*, **71**: 346–354.
- Tucker, W.B., Hogue, J.F., Waterman, D.E., Swenson, T.S., Xin, Z., Hemken, R.W., Jackson, J.A., Adams, G.D. and Spicer, L.J. (1991). Role of sulfur and chloride in the dietary cation-anion balance equation for lactating dairy-cattle. *J. Anim. Sci.*, **69**: 1205–1213.
- Tucker, W.B., Hogue, J.F., Waterman, D.F., Swenson, T.S., Xin, Z., Hemken, R.W., Jackson, J.A., Adams, G.D. and Spicer, L.J. (1992). Sulfur should be included when calculating the dietary cation-anion balance of diets for lactating dairy cows. In Animal Science Research Report, Pp. 141–150.
- Tucker, W.B., Xin, Z. and Hemken, R.W. (1988b). Influence of dietary calcium chloride on adaptive changes in acid-base status and mineral metabolism in lactating dairy cows fed a diet high in sodium bicarbonate. *J. Dairy Sci.*, **71**: 1587–1597.
- Van Soest, P.J., Robertson, J.B. and Lewis, B.A. (1991). Method for dietary fibre, neutral detergent fibre and non starch polysaccharides in relation to animal nutrition. *J. Dairy Sci.*, **74**: 3583–3597.
- Vander, A.J. (1991). Renal Physiology. 4th ed. McGraw-Hill Inc., NY.
- West, J.W., Mullinix, B.G. and Sandifer, T.G. (1991). Changing dietary electrolyte balance for dairy cows in cool and hot environments. *J. Dairy Sci.*, **74**: 1662–1674.
- Wildman, C.D., West, J.W., Bernard, J.K. (2007). Effect of dietary cation-anion difference and dietary crude protein on performance of lactating dairy cows during hot weather. *J. Dairy Sci.*, **90**: 1842–1850.