

SULFUR SHOULD BE INCLUDED WHEN CALCULATING THE DIETARY CATION-ANION BALANCE OF DIETS FOR LACTATING DAIRY COWS

W.B. Tucker¹, J.F. Hogue², D.F. Waterman³, T.S. Swenson³,
Z. Xin³, R.W. Hemken³, J.A. Jackson³,
G.D. Adams⁴ and L.J. Spicer¹

Story in Brief

Ten Holstein cows averaging 120 d in lactation were arranged in replicated 5 x 5 Latin squares with 3 wk periods to evaluate the role of sulfur (S) on dietary cation-anion balance. Diets were based upon corn silage in Experiment 1 and sorghum silage in Experiment 2. Supplemental S and chloride (Cl) from Dynamate^R and CaCl₂ were utilized to manipulate the dietary cation-anion balance from 0 to +30 meq when expressed as $\text{meq}((\text{Na} + \text{K}) - (\text{Cl} + \text{S}))/100$ g diet DM, and from +19 to +49 meq when expressed as $\text{meq}((\text{Na} + \text{K}) - \text{Cl})/100$ g diet DM. Blood pH was not affected by cation-anion balance, although both S and Cl supplementation tended to lower pH. Blood HCO₃⁻ and urine pH decreased, whereas plasma calcium and urinary calcium excretion increased as anion was added to the diet. Milk fat production tended to be increased by the low S supplementation. Dietary Cl and S had similar effects on acid-base status. Therefore, we suggest that S should be included with Cl when calculating the dietary cation-anion balance equation for lactating dairy cows as follows: $\text{meq}((\text{Na} + \text{K}) - (\text{Cl} + \text{S}))/100$ g diet DM. Although response of acid-base status to S and Cl was similar, as more data comparing the acidogenicity of S vs. Cl becomes available, it may be necessary to include a modifying coefficient for S in the equation to adjust for differences between S and Cl in acid-generating potential. This coefficient may be further dependent upon source of dietary S.

(Key Words: Dairy Cows, Sulfur, Ion Balance, Acid Base Equilibrium.)

¹Assistant Professor ²Graduate Student ³University of Kentucky ⁴Instructor

Introduction

Despite the apparent importance of dietary fixed ions to numerous physiological responses, objective determination of which ions need to be included in the dietary cation-anion balance equation for ruminants is nonexistent. The most common equation used for poultry (Mongin, 1980) is $\text{meq (Na+K)-Cl}/100 \text{ g diet DM (DCAB)}$; this completely disregards potential effects of S, which could be accounted for as $\text{meq (Na+K)-(Cl+S)}/100 \text{ g diet DM (DCAB:S)}$. The objective of this trial was to evaluate the relative influence of dietary S vs Cl on systemic acid-base status, milk yield, milk composition and mineral metabolism in lactating dairy cows.

Materials and Methods

Experiment 1 was conducted at the University of Kentucky. Five primiparous lactating Holstein cows averaging $123 \pm 13 \text{ d (X} \pm \text{SD)}$ postpartum and producing $33 \pm 4.5 \text{ kg milk/day}$ were arranged in a 5×5 Latin square with experimental periods of 3 weeks. A similar design was utilized to conduct Experiment 2 at Oklahoma State University, with the exception that experimental animals consisted of one primiparous and four pluriparous Holstein cows, averaging $116 \pm 25 \text{ d postpartum}$ and producing $30 \pm 5.7 \text{ kg milk/d}$.

In Experiment 1, the treatments were 5 diets (Table 1) containing corn silage and concentrate in a 48:52 DM ratio with 0 to +30 DCAB:S and +19 to +49 DCAB. Decreasing the cation-anion balance of the diet by equal increments with both S and Cl allowed direct comparison of the effects of these anions. In addition, calculation of DCAB for C/0, C/+15 and Ctrl/+30 yielded values of +19, +33.9 and +48.9. Because these 3 diets all contained .3% S, this allowed us to test the response of dairy cattle to DCAB higher than +19.

A similar approach was used for the diets in Experiment 2 (Table 2) except that sorghum silage replaced corn silage. Other exceptions included lower dietary Ca, P and Cl, and higher Mg, K and S concentrations than for Experiment 1. Both DCAB and DCAB:S were formulated to be identical to Experiment 1, but DCAB:S was approximately 7 meq lower in Experiment 2 due to higher than expected dietary S concentrations. The experiments were designed to allow 2 principal evaluations: 1) the physiological effects of lowering dietary cation-anion balance, disregarding the anion utilized to lower it, and 2) a direct comparison of the effects of S to the effects of Cl.

Blood and urine samples were collected at 4 h postfeeding on the last day of each period. In both experiments, feed intake was recorded daily and daily

Table 1. Calculated ingredient and nutrient composition of experimental diets^a for experiment 1.

Ingredients:	Diet				
	Ctrl/+30	C/+15	S/+15	C/0	S/0
Corn silage	47.71	47.71	47.71	47.71	47.71
Ground corn	26.58	26.58	26.58	26.58	26.58
Soybean meal (44%)	19.50	19.50	19.50	19.50	19.50
CaCO ₃	1.497	.730	1.513	1.528
Dicalcium phosphate	.512	.512	.512	.512	.512
MgO	.444	.446	.223	.446
NaHCO ₃	.960	1.009	.964	.990	.964
KHCO ₃	1.409	1.382	.901	1.355	.390
Salt ^b	.251	.207	.228	.207	.207
CaCl ₂881	1.719
Dynamate ^c	.354	.354	1.407	.354	2.462
Selenium premix ^d	.100	.100	.100	.100	.100
Vitamin ADE premix ^e	.050	.050	.050	.050	.050
SiO ₂	.636	.542	.315	.480
Crude protein	16.39	16.39	16.39	16.39	16.39
NE _L (Mcal/kg) ^f	1.58	1.58	1.58	1.58	1.58
ADF	17.25	17.25	17.25	17.25	17.25
NDF	34.29	34.29	34.29	34.29	34.29
Ca	1.09	1.09	1.09	1.09	1.09
P	.47	.47	.47	.47	.47
Mg	.47	.47	.47	.47	.47
Na	.43	.43	.43	.43	.43
K	1.51	1.51	1.51	1.51	1.51
Cl	.30	.83	.30	1.36	.30
S	.30	.30	.54	.30	.78
meq/100 g diet DM:					
(Na+K)-(Cl+S)	+30.2	+15.2	+15.2	+ .2	+ .2
(Na+K)-Cl	+48.9	+33.9	+48.9	+19.0	+48.9

^a Ingredients and nutrients listed as a percentage of diet DM.

^b Trace mineralized salt contained 3500 ppm Zn, 3400 ppm Fe, 2000 ppm Mn, 330 ppm Cu, 70 ppm I and 50 ppm Co.

^c Double sulfate of K and Mg.

^d Contained 200 ppm Se.

^e Contained 10,008,818 IU Vitamin A/kg, 2,204,586 IU Vitamin D/kg, and 1,102 IU Vitamin E/kg.

^f Calculated from ADF; all other nutrients are from diet analysis.

Table 2. Calculated ingredient and nutrient composition of experimental diets^a for experiment 2.

Ingredients:	Diet				
	Ctrl/+30	C/+15	S/+15	C/0	S/0
Sorghum silage	38.70	38.70	38.70	38.70	38.70
Ground corn	13.45	13.45	13.45	13.45	13.45
Soybean meal (44%)	10.20	10.20	10.20	10.20	10.20
Dried corn					
distillers grains	32.14	32.14	32.14	32.14	32.14
CaCO ₃	1.115	.459	1.133	.039	1.151
Dicalcium phosphate	.372	.373	.372	.374	.372
MgO	.457	.480	.233	.495	.010
NaHCO ₃	.971	.664	.973	.007	.975
KHCO ₃	1.586	1.565	1.075	1.552	.564
Salt ^b	.298	.500	.276	.946	.254
CaCl ₂648	1.062
Dynamate ^c	.001	.002	1.057	.003	2.114
SiO ₂	.707	.815	.387	1.029	.066
Nutrients:					
Crude protein	17.7	17.1	16.7	17.5	17.5
NEI (Mcal/kg) ^d	1.52	1.52	1.52	1.52	1.52
ADF	25.3	25.2	25.6	24.8	25.3
NDF	32.8	35.0	39.0	32.6	35.1
Ca	.71	.71	.74	.74	.81
P	.45	.47	.47	.47	.48
Mg	.55	.64	.66	.63	.60
Na	.41	.45	.45	.41	.43
K	1.68	1.81	1.78	1.73	1.71
Cl	.43	.93	.51	1.43	.46
S	.43	.49	.66	.46	.91
meq/100 g diet DM:					
(Na+K)-(Cl+S)	+21.8	+9.1	+9.5	-7.0	-7.3
(Na+K)-Cl	+48.7	+39.6	+50.7	+21.7	+49.5

^a Ingredients and nutrients listed as a percentage of diet DM.

^b Trace mineralized salt contained 3500 ppm Zn, 3400 ppm Fe, 2000 ppm Mn, 330 ppm Cu, 70 ppm I and 50 ppm Co.

^c Double sulfate of K and Mg.

^d Calculated from ADF; all other nutrients are from diet analysis.

dry matter intake was averaged by week. Milk samples were collected one day per week during consecutive p.m. and a.m. milkings.

Results and Discussion

Blood and Urine Acid-Base Status

Blood $[H^+]$ was not affected ($P > .10$) by cation-anion balance, although it was higher numerically for both S and Cl supplementation (Table 3). Blood pH values for all diets were similar to previously published values (Tucker et al., 1988). Blood HCO_3^- was reduced ($P = .018$) in our study by anion supplementation, whereas blood pCO_2 was unaffected ($P > .10$). Tucker et al. (1988) reported that reducing DCAB from +20 to -10 reduced blood pH from 7.427 to 7.369, and reduced blood HCO_3^- from 23.2 to 19.3 meq/liter. Whiting and Cole (1986) supplemented the diets of rats with Cl or S to evaluate their effects on urinary net acid excretion. Their results indicated that if differences in apparent absorption of Cl and S were considered, the two anions produced similar increases in urinary net acid excretion and would be expected to have similar effects on systemic acid-base status. Chloride vs. S contrasts (Table 3) were nonsignificant for all indicators of acid-base status in our study; therefore, our data supports the concept of similar acidogenicity for Cl and S. Reducing DCAB tended to increase urine $[H^+]$ in both experiments, but the effect of the highest concentrations of Cl and S were much more pronounced in Experiment 2.

Chloride and S generate acid systemically as they are absorbed from the gastrointestinal tract. However, apparent absorption of Cl by ruminants may exceed 95% (Church and Fontenot, 1979), whereas apparent absorption of S in Hereford steers has ranged from 51.8 to 60.8% (Spears et al., 1985). The impact of S on systemic acid-base status might be less than that of Cl because less S is absorbed. Moreover, the variety of organic and inorganic forms in which S may be absorbed and utilized by the body adds to the variability of its effect on acid-base status.

Plasma and Urine Minerals

Plasma Na and K concentrations (Table 4) were not significantly affected ($P > .10$) by cation-anion balance of the diets, but differences in dietary concentrations of these elements were slight. Plasma Ca was elevated by Cl and S supplementation in Experiment 1 but not in Experiment 2. Nevertheless, plasma Ca was numerically higher for the C/+15 diet than for the control in Experiment 2. In the merged data analysis, plasma Ca was increased ($P = .003$) by anion supplementation (Table 4). Plasma Mg and P

Table 3. Least square means and orthogonal contrasts for blood and urine acid-base status.

	Ctrl/ +30	C/ +15	S/ +15	C/ 0	S/ 0	SE	P values ^a			
							Ctrl/ +30 vs others	C/ vs S diets	C/+15 vs C/0	S/+15 vs S/+0
Blood [H ⁺], neq/L	36.6	38.1	36.8	38.0	38.3	.73	NS	NS	NS	NS
Blood pH	7.437	7.419	7.434	7.420	7.417					
Blood pCO ₂ , (mm Hg)	43.9	43.7	41.2	42.3	42.2	1.27	NS	NS	NS	NS
Blood HCO ₃ ⁻ , (meq/L)	29.2	28.1	28.2	27.1	26.9	.57	.018	NS	NS	NS
Urine [H ⁺], neq/L	6.7	11.4	8.2	149.1	40.9	36.8 ^b	NS	NS	.014	NS
pH	8.17	7.94	8.09	6.83	7.39					

^aP values for Type I error; NS = nonsignificant (P > .10).

^bSquare by treatment interaction (P = .057).

Table 4. Least square means and orthogonal contrasts for plasma and urine mineral responses to dietary cation-anion balance.

	Ctrl/ +30	C/ +15	S/ +15	C/ 0	S/ 0	SE	P values ^a			
							Ctrl/+30 vs others	CI vs S diets	C/+15 vs C/0	S/+15 vs S/+0
Plasma: (meq/L)										
Na	142.0	140.7	138.9	141.4	141.0	1.01	NS	NS	NS	NS
K	5.48	5.61	5.59	5.66	5.55	.120	NS	NS	NS	NS
Cl	98.2	102.3	99.8	103.7	101.2	.87	.001	.008	NS	NS
Ca	4.69	4.81	4.78	4.78	4.74	.025 ^b	.003	NS	NS	NS
Mg	1.92	1.91	1.97	1.95	1.95	.034	NS	NS	NS	NS
P (mg/L)	65.9	65.4	66.7	65.6	69.0	2.18	NS	NS	NS	NS
Cation-anion balance ^c	49.3	44.1	44.7	43.3	45.3	1.34	.003	NS	NS	NS
Urine excretion (mg mineral/mg creatinine):										
Na	3.94	6.74	5.42	3.99	3.16	.994	NS	NS	.061	NS
K	13.7	15.4	13.8	13.8	11.9	.95	NS	.072	NS	NS
Cl	1.42	7.87	3.04	9.16	1.96	1.166	.004	.001	NS	NS
Ca	.567	.871	.772	.735	.719	.0980	.069	NS	NS	NS
Mg	.449	.529	.498	.490	.445	.0610	NS	NS	NS	NS
P	.392	.578	.373	.393	.289	.0921	NS	NS	NS	NS
Cation-anion ^d balance	.482	.466	.502	.268	.386	.0354 ^e	.065	.038	.001	.029

^aP values for Type I error; NS = nonsignificant ($P > .10$).

^bSquare by treatment interaction ($P = .020$).

^cExpressed as meq((Na + K)-Cl)/liter.

^dExpressed as meq((Na + K)-Cl)/mg urine creatinine.

^eSquare by treatment interaction ($P = .012$).

concentrations were not affected ($P > .10$) by Cl or S supplementation (Table 4). Factors involved in Mg homeostasis have not been clearly identified, although extracellular Mg concentration is regulated by renal excretion (Guyton, 1986). If lowering the dietary cation-anion balance resulted in an increase in parathyroid hormone release, P would be mobilized from bone but the threshold for reabsorption of phosphate in the renal tubules also would be reduced (Guyton, 1986) so that more P would be lost in the urine; hence, plasma P concentrations may not be affected.

Plasma cation-anion balance (Table 4) was reduced ($P = .003$) by adding dietary Cl or S. This is in agreement with Tucker et al. (1988) who reported that reducing DCAB from +20 to -10 resulted in a reduction ($P < .05$) of serum cation-anion balance from 6.8 to 5.8 meq/100 ml.

Urinary Na excretion (Table 4) tended to decrease ($P = .061$) as Cl supplementation increased, whereas K excretion tended to be higher ($P = .072$) for supplementation of Cl vs S. Urinary Cl excretion increased with dietary Cl supplementation. Urinary Ca excretion tended to be higher ($P = .069$) with Cl and S supplementation and paralleled plasma Ca response. Urinary Mg and P excretion were not affected ($P > .10$) by Cl or S supplementation.

As noted by Tucker et al. (1988), urinary cation-anion balance (Table 4) tended to decrease ($P = .065$) with decreasing dietary cation-anion balance in the merged data analysis. This reduction was more extensive ($P = .038$) for Cl than S supplementation but continued as DCAB:S decreased from +15 to 0 meq. The larger response to the Cl diets likely can be explained by the exclusion of S when calculating urinary cation-anion balance. Urinary S concentration was not determined.

Feed Intake, Milk Yield and Composition

Daily milk yield (Table 5) was lowered by reducing DCAB:S from +15 to +0 with either Cl or S. Milk fat percentage, milk fat yield and 4% fat-corrected milk production tended to be highest for cows fed the S/+15 diet. In addition, milk protein yield was higher ($P = .017$), but protein percentage was not different for S/+15 vs. S/0. The tendency for elevated 4% FCM and milk fat and protein production for the S/+15 diet in our study may be related to improved nutrient digestibility in the rumen, although S concentrations for all diets were above the requirement of .25% listed by the NRC (1988).

Dry matter intake was numerically highest for cows receiving the C/+15 diet, and was higher ($P = .019$) for C/+15 than for C/0. Tucker et al. (1988) reported that daily DM intake decreased from 18.6 kg to 16.8 kg as DCAB was decreased from +20 to -10. However, in that study, the sharpest reduction in intake occurred for the -10 diet; therefore, the lack of intake depression in our study might be due to the higher DCAB.

Table 5. Least square means and orthogonal contrasts for milk yield and composition, and dry matter intake.

	Ctrl/ +30	C/ +15	S/ +15	C/ 0	S/ 0	SE	P values ^a			
							Ctrl/+30 vs others	C/ vs S diets	C/+15 vs C/0	S/+15 vs S/+0
Daily milk yield, kg	23.2	23.4	23.5	21.6	21.0	.56	NS	NS	.029	.004
4% FCM, kg	21.6	21.2	24.3	20.0	20.1	.84	NS	.062	NS	.001
Milk fat, (%)	3.54	3.37	4.34	3.46	3.65	.268	NS	.040	NS	.080
Milk fat, kg	.82	.79	1.00	.75	.78	.051	NS	.032	NS	.005
Milk protein, (%)	3.31	3.26	3.32	3.34	3.29	.042	NS	NS	NS	NS
Milk protein, kg	.76	.76	.77	.72	.69	.023	NS	NS	NS	.017
DM intake, kg	19.3	20.1	19.7	19.0	19.0	.32	NS	NS	.019	NS

^aP values for Type I error; NS = nonsignificant ($P > .10$).

The ratio of fixed cation to anion in the diet is an important determinant of dietary impact on systemic acid-base status. Dairy farmers should use this effect to control acid-base status-responsive disorders such as milk fever. In addition, manipulation of this dietary balance may prove useful in attenuating dietary acid or base challenges for lactating dairy cows effected by specific feeding regimens; e.g., the high-concentrate diets offered to high-producing dairy cows. However, the importance of specific fixed ions to this response must be determined before its widespread application will be practical. The results of our study indicate that in the range of 0 to +30 meq((Na+K)-(Cl+S))/100 g diet DM, the effect of S on systemic acid-base status is similar to that of Cl. Therefore, the dietary cation-anion balance equation for lactating dairy cows should include S and should be calculated as: meq((Na+K)-(Cl+S))/100 g diet DM. After further evaluation of different forms and sources of dietary S, a modifying coefficient for S may need to be developed which will account for the acidogenicity of S relative to Cl.

Literature Cited

- Church, D.C. and J.P. Fontenot. 1979. The macro (major) minerals. In: D.C. Church (ed.) Digestive Physiology and Nutrition of Ruminants. pp 82-83. O&B Books, Inc., Corvallis, OR.
- Guyton, A.C. 1986. Textbook of Medical Physiology. 7th ed. W.B. Saunders Co., Philadelphia, PA.
- Mongin, P. 1980. Electrolytes in nutrition: review of basic principles and practical application in poultry and swine. pp 1-15 in Proc. Third Annu. Int. Miner. Confer., Orlando, FL.
- National Research Council. 1988. Nutrient requirements of dairy cattle. 6th ed., Natl. Acad. Sci., Washington, DC.
- Spears, J.W. et al. 1985. Sulfur fertilization of cool season grasses and effect on utilization of minerals, nitrogen, and fiber by steers. *J. Dairy Sci.* 68:347.
- Tucker, W.B. et al. 1988. Influence of dietary cation-anion balance on milk, blood, urine and rumen fluid in lactating dairy cattle. *J. Dairy Sci.* 71:346.
- Whiting, S.J. and D.E.C. Cole. 1986. Effect of dietary anion composition on acid-induced hypercalciuria in the adult rat. *J. Nutr.* 116:388.