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## The effects of a cation-anion balanced diet on calcium and phosphorus metabolism in growing lambs

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### ABSTRACT

During the past decade, there has been renewed interest and research in the role of the dietary cation-anion balance (DCAB) in controlling Ca and P homeostasis. Two diets differing in DCAB (either high DCAB, 250 meq/kg DM, or low DCAB, -100 meq/kg DM) were fed to 10 Romney lambs. Feeding low DCAB decreased urine pH by 2.0 pH units; from 8.2 (SE=0.45) to 6.2 (SE=0.50), indicative of the anticipated changes in the ionic balance associated with alterations in the dietary CAB. Plasma concentrations of Ca and P, and whole-body Ca and P balance (intake-excretion) were similar for both groups of sheep. Reducing the dietary CAB caused a 7-fold increase in the urinary excretion of Ca (low DCAB 0.75 g/day vs high DCAB 0.11 g/day;  $P=0.001$ ). The apparent absorption of Ca in the low DCAB and high DCAB groups were 1.6 and 1.3 g/day, respectively, although there was no significant difference between the two groups. These data indicate that lowering the DCAB increases urinary excretion of Ca of growing lambs without affecting Ca balance due to an alteration in the movement of Ca to and from bone. Similar changes in Ca absorption and utilisation have been observed in dairy cows, suggesting that the growing lamb is a suitable model for future studies in the regulation of Ca homeostasis in dairy cattle.

Keywords: cation-anion balance; calcium metabolism; phosphorous metabolism; milk fever, ruminant nutrition.

### INTRODUCTION

Calcium (Ca) and phosphorus (P) are involved in a number of important physiological and biochemical functions. Together they are part of the complex salt which forms the inorganic matrix of bone and teeth. Besides its structural function, the bone also acts as a store for Ca and P which is available when demand for Ca increases and exceeds the dietary supply, as is often the case during lactation. Ca homeostasis is complex and the movements of Ca between the skeleton, intestine and kidney are affected by the hormones parathyroid hormone and calcitonin as well as vitamin D (Grace, 1983).

A changing physiological status, such as growth (Chrisp *et al.*, 1989a) or lactation (Chrisp *et al.*, 1989b) alters the demand and metabolism of Ca. Milk fever or parturient paresis is the most serious problem involving Ca deficiencies in ruminants. Milk fever in dairy cows has been successfully prevented with a negative cation-anion balance in the diet (Block, 1984). The cation-anion balance was defined as the summation in milliequivalents (meq kg<sup>-1</sup>) of the cations, sodium (Na) and potassium (K), minus the anion, chloride (Cl). A trial was undertaken to examine the effects of dietary cation-anion balance on Ca and P homeostasis and to assess how well young sheep serve as models for dairy cows.

### MATERIALS AND METHODS

#### Experimental Procedures

Ten 10-month old lambs (30-35 kg) were individually housed in metabolism crates. Lambs were allocated into two groups (5 lambs per group) and fed either a diet of high

DCAB (control) or a diet with a low DCAB (treatment) for 19 days. A period of 12 days was used to allow for adaptation to the diet and the environment. Experimental rations were fed at 1.3 x maintenance energy requirements from day 0 of the trial. The control diet was 250 g barley pellets containing 13 g Na<sub>2</sub>CO<sub>3</sub> per kg mix + 1 kg lucerne pellets. The treatment diet was 250 g barley pellets containing 10% NH<sub>4</sub>Cl (100 g NH<sub>4</sub>Cl per kg mix) + 1 kg of lucerne pellets. The diets were also balanced for Na. The composition of the two diets is given in Table 1. Feed was offered via overhead feeders at hourly intervals.

Each lamb fed the low DCAB was pair-fed with a lamb, closely matched by liveweight, that was fed the high DCAB. Fresh water was provided *ad libitum*. Two three-day collections of total urine, faeces and feed refusals were conducted over days 12-14 and days 17-19. Two days before starting the first collection period, we found that the urine of two animals with the low DCAB were contaminated with dust from the diet. This was immediately recti-

**TABLE 1:** The concentrations of calcium (Ca), phosphorus (P), and sodium (Na), and the metabolisable energy (ME) for sheep fed either the low- or high-DCAB rations, and the calculated DCAB of both rations.

	High DCAB	Low DCAB
Ca (g/kg DM)	11.73	11.18
P (g/kg DM)	2.22	2.12
Na (g/kg DM)	1.21	1.15
ME (MJ/kg DM) <sup>1</sup>	10.6	10.6
Calculated DCAB (meq/kg DM)	250	-100

<sup>1</sup> Calculated energy content based on the Farm Technical Manual (1991).

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fied to ensure urine was collected free of contamination. A 10% sub-sample of, feed, faeces and urine were taken for later analyses and calculation of calcium and phosphorus balance.

**Ca and P Analysis**

Following acid digestion, samples of feed, faeces and urine were analysed by inductively-coupled plasma emission spectrometry (Lee, 1983) for Ca, Na and P concentrations.

**Statistical Analysis**

Significance of differences between treatment means and between balance period means for data relating to Ca and P were tested by ANOVA using the SAS computer programme (SAS, 1985). The SAS programme was also used to test differences in plasma Ca and P concentrations and urinary pH.

**RESULTS**

**Health of the Lambs**

No health problems were encountered with lambs in either group. The mean live weights for the high DCAB group was 32.5 ± 0.77 kg and 33.9 ± 0.77 kg for the low DCAB group. These weights remained constant throughout the experimental period.

**Mineral Content of Plasma and Urine pH**

No significant differences in Ca<sup>2+</sup> or PO<sub>4</sub><sup>3-</sup> concentrations in plasma collected over three days during the balance periods were detected (Ca<sup>2+</sup> 2.5 mmol L<sup>-1</sup>; PO<sub>4</sub><sup>3-</sup> 2.5 mmol L<sup>-1</sup>).

During the first balance period (days 12-14), the lambs fed the high DCAB had urinary pH values that were 1.3 pH units higher than those fed the low DCAB (Table 2; P=0.05). During the second balance period (days 17-19), the difference in urinary pH had increased to 2.0 pH units (P=0.01).

**TABLE 2:** Urinary pH of lambs fed high- or low-DCAB rations during two balance periods.

Diet	Balance period <sup>1</sup>	pH
High DCAB	1	8.9 ± 0.45 <sup>a</sup>
High DCAB	2	8.2 ± 0.45 <sup>ab</sup>
Low DCAB	1	7.6 ± 0.41 <sup>b</sup>
Low DCAB	2	6.2 ± 0.50 <sup>c</sup>

<sup>1</sup> Balance period 1; days 12-14, and balance period 2; days 17-19.  
<sup>abc</sup> Means within columns having different superscripts are significantly different (P < 0.05).

**Ca and P Balance**

Since there were no statistically significant period effects detected in Ca and P balance, the data were pooled and are summarised in Table 3.

The intake of Ca and P was similar in both groups of lambs, as was the daily faecal excretion of both Ca and P. As a result, the apparent absorption of Ca and P were not different between the high DCAB and low DCAB groups.

**TABLE 3:** The intake, excretion, apparent absorption and retention of calcium (Ca) and phosphorous (P) in lambs fed high- or low-DCAB rations

	Treatment		SEM	
	High DCAB (g/day)	Low DCAB (g/day)		
Ca intake	13.4	13.1	0.59	n.s.
Ca loss				
- Urine	0.1	0.7	0.16	*
- Faeces	12.1	11.5	0.71	n.s.
Apparent Ca absorption	1.3	1.6	0.31	n.s.
Ca retention	1.2	0.9	0.43	n.s.
P intake	2.6	2.3	0.11	n.s.
P loss				
- Urine	0.01	0.01	0.002	n.s.
- Faeces	2.5	2.2	0.26	n.s.
Apparent P absorption	0.1	0.1	0.17	n.s.
P retention	0.1	0.1	0.17	n.s.

Mean values for five lambs per treatment. Probability levels : \* P < 0.05; n.s., not significant.

There was a 7-fold greater loss of Ca in the urine of lambs on the low DCAB diet (low DCAB 0.7 g/day vs high DCAB 0.1 g/day; P=0.02; Table 3), there was no difference between groups in the urinary excretion of phosphorus (P=0.66). The lambs fed the low DCAB had a net retention of 0.9 g Ca/day compared with the retention of 1.20 g Ca/day for the lambs fed high DCAB although there was no statistical difference between the two groups.

**DISCUSSION**

The differences in the pH of the urine between the lambs on the two diets (Table 2) reflects the effects of the DCAB on the acid-base balance of the animals (Tucker *et al.*, 1988; Erdman, 1993) and indicates a significant effect of the diet in this trial. On diets with a high DCAB, such as normally consumed by ruminants, the excess cations are excreted in association with bicarbonate and the pH of the urine is usually alkaline. Lowering the DCAB until there is an overall excess of fixed anions results in their excretion in association with H<sup>+</sup> and an acidic urine.

The Ca concentration in the plasma of the lambs fed the low DCAB were similar to that of the group fed the high DCAB indicating that Ca homeostasis was being satisfactorily maintained in both groups. This is similar to the response with growing lambs (Abu Damir *et al.*, 1991) but differs from the response observed in cows (Block, 1984), in which plasma Ca concentrations were increased by lowering the DCAB. Nevertheless, the lack of difference in plasma Ca concentrations does not preclude an effect of diet on the metabolism of Ca.

Ramberg *et al.* (1984) outlined three major controlling mechanisms for counterbalancing disturbances in Ca metabolism and maintaining Ca homeostasis; intestinal Ca absorption, bone Ca resorption, and renal Ca reabsorption. The two diets used in the present study had similar concentrations of Ca and P in each and, with pair-feeding,

the intake of the minerals in both treatment groups was similar. Neither the apparent absorption of Ca nor that of P were significantly different between the groups (Table 3). Low DCAB has been reported to either increase calcium absorption or have no effect in the dairy cow (Freedeen *et al.*, 1988; Leclerc and Block, 1989) and Schonewille *et al.* (1994) consider the issue is still unresolved. There was, however, a 7-fold greater excretion of Ca in the urine by lambs on the low DCAB (Table 3). A similar response has also occurred in cows fed low DCAB, and has been attributed to an increase in the resorption of Ca from the bone (Block, 1984). The higher urinary excretion of Ca in lambs fed low DCAB was associated with a non-significant increase in apparent absorption of Ca (1.6 vs 1.3 g Ca/day; equivalent to Ca digestibility values of 15% and 10%, respectively). Analysis of blood or urine samples for markers of bone metabolism such as hydroxyproline or deoxypyridinoline (Hodsman *et al.*, 1993) would be needed to confirm whether the increase in urinary Ca excretion was associated with an increase in Ca resorption from bone.

A lower DCAB reduced urine pH and increased in Ca excreted in the urine, but did not alter Ca retention. Similarly, no changes in P retention were found. It is suspected that the mode of action of the reduced dietary cation-anion balance is through the increased bone resorption or reduced bone accretion of Ca in the growing lambs. If so, the lamb may be a suitable model for future studies in the regulation of Ca homeostasis in dairy cows since Ca movement from bone also appears to be the main site of regulation for Ca metabolism in lactating cows.

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