

# The effects of calcium and sodium loading on organic matter digestibility and mineral absorption in sheep

## 1. Digestion in the forestomachs and small intestine

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### ABSTRACT

Experiments were conducted on sheep cannulated to the rumen, duodenum and ileum to study the effects of Ca and Na loading (5 and 10 times the ARC requirements) on digestion in the rumen and intestines. The introduction of calcium lactate (19.6%) or sodium chloride (9.6%) into extruded feed mixtures had no significant effect on degradation in the rumen (apparent digestibility of organic matter in the forestomachs averaged 40%, ileal digestibility 61%) or on the pH of ruminal (5.73), duodenal (3.37) or ileal (7.63) digesta. Excess sodium in the diet reduced ( $P < 0.05$ ) microbial protein synthesis in the rumen (7.69g N-MP/d) in comparison with control sheep and those loaded with Ca (10.46 and 10.74 g N-MP/g) as well as the apparent digestibility of total nitrogen in the small intestine.

**KEY WORDS:** Ca, Na, sheep, digestion, protein synthesis

### INTRODUCTION

Fibrous feeds are often treated with calcium or sodium alkali in order to improve their digestibility in ruminants, while rations containing large amounts of starch are fed along with calcium-containing buffers. However, these processes also lead to loading with Na and Ca (Łukowski, 1980). A high calcium and sodium content in the diet of ruminants can reduce feed intake and utilization (Wilson and Dudziński, 1973; Goodrich et al., 1985) and the digestibility of organic matter (Hemsley, 1975; Jacobson et al., 1975; Gralak et al., 1994). Changes in the concentration of mineral components in the digestive

tract, including Ca and Na, can change digesta pH (Rogers et al., 1982), cellulolytic activity (van Soest, 1982; Harris et al., 1989) and microbial protein synthesis in the rumen (Durland and Kawashima, 1980; Leontowicz et al., 1994).

The objective of this study was to assess the effect of rations containing excess calcium and sodium (5 and 10 times more, respectively, than ARC requirements) on ruminal and intestinal digestion dynamics and ruminal microbial protein synthesis in sheep.

## MATERIAL AND METHODS

### *Animals*

The study was carried out on 9 Merino wethers with an average initial body weight of  $24 \pm 1.5$  kg. The sheep were divided into 3 groups: control (I), calcium-loaded (II; 20% calcium lactate) and sodium-loaded (III; 8.5% NaCl in the ration). The calcium content in the diet fed group II was 5 times, while that of sodium in group III, 10 times higher than given in the ARC requirements (1980). The sheep received extruded concentrate mixtures (Table 1) and chopped meadow hay at a proportion of 1:1.

All of the sheep were cannulated with a Jarett cannula to the rumen, a straight T cannula (PVC) to the duodenum (2-3 cm after the pancreatic-bile duct orifice) and to the ileum (7 cm before the appendix).

### *Experimental design*

The experiment has started with a 3-week adaptation period and 2-week period of digesta collection. The sheep were fed 3 times daily; extruded mixtures (0.4 kg/animal/day) at 9h, followed an hour later by 1/2 of the hay ration and the remainder at 16h. The animals were watered twice daily at 8 and 15 h. Feed and water intake were controlled. Chromium in the form of chromic oxide impregnated paper was given twice daily through the cannula to the rumen 4 days before collection and throughout the collection period at a dose of 1.64 g  $\text{Cr}_2\text{O}_3/\text{d}$ .

### *Sample collection and chemical analysis*

Duodenal and ileal digesta were collected on days 1 and 2 of the fourth and fifth week of the experiment for the first 48h in 6-h intervals. The pH was measured in each digesta sample, after which they were mixed and the unfiltered samples were pooled to obtain daily samples for dry matter (DM), total nitrogen and ammonia determination. The remaining part of the digesta sampled from the

TABLE I

Composition and nutritional value of concentrates for sheep, g/kg

Ingredients, g/kg	Concentrates		
	I control	II Ca-loading	III Na-loading
Barley	680	554	600
Soyabean meal	219	195	215
Fodder yeast	25	20	24
Polfamix D	25	25	25
Calcium lactate	50	196	50
Urea	—	9	—
Sulphur <sup>1</sup>	1	1	1
NaCl	—	—	85
Per kg feed			
dry matter, g	875.3	888.4	878.5
metabolic energy, <sup>2</sup> MJ	10.7	10.5	9.7
RDP, g <sup>3</sup>	92.0	95.0	83.5
UDP, g	68.1	52.6	62.2

<sup>1</sup> sulphur was supplied as a 1:1 proportion of Na<sub>2</sub>SO<sub>4</sub> and elemental sulphur<sup>2</sup> the metabolic energy content of the ration was calculated using the regression coefficient for sheep according to van Es (1978)<sup>3</sup> the RDP and UDP contents in the feed rations were calculated according to the ARC (1980) based on the determined crude protein content in the individual components of concentrates and hay

duodenum and ileum was frozen and pooled into daily samples after freeze-drying. DM was determined at 105°C, ash, chromium according to Schürch et al. (1950) and total nitrogen was determined by the Kjeldahl (Kjeltec-Tecator) method in these samples. Nucleic acids were determined in duodenal digesta (Zinn and Owens, 1982).

On the third day of weeks four and five of the experiment the rumen content was sampled before and 1, 2, 3, 4, 5 and 6 h after morning feeding. The pH was determined in all rumen samples, and the following in selected samples (0, 1 and 3 hours): ammonia by the phenol reagent method, volatile fatty acids (VFA) by gas chromatography (Ziolecki and Kwiatkowska, 1973) and lactic acid (+) (Boehringer test kit).

### Calculations

The apparent digestibility of DM and organic matter (OM) in the rumen and small intestine and their duodenal and ileal flows were calculated on the basis of the chromium content of the digesta from these two sites, using the formula given by Ruszczyc (1983). Microbial protein synthesis (MP) in the rumen was determined according to Czerkawski (1976) on the basis of duodenal nucleic acid flow.

The results analyzed statistically by two-way variance analysis using Statgrafics software, while the difference between means was evaluated by the Tukey test.

## RESULTS

The chemical composition of extruded feed mixtures and hay is given in Table 2. The sheep consumed all of the offered concentrates, while leaving some of the hay (expressed in % of DM): 27.0, 6.3 and 12.9 in groups I, II and III, respectively. The animals fed the Ca ration consumed more DM ( $P < 0.05$ ) than controls (Table 3). Water intake in groups I-III equalled 2577, 2765 and 3455 ml/d ( $P < 0.05$ ).

Chemical composition of feeds, g/kg DM

TABLE 2

Nutrients	Extruded concentrate			Meadow hay
	I control	II Ca-loading	III Na-loading	
Crude protein	210	203	199	106
Crude fibre	40	35	33	330
Fat	21	24	19	40
Ash	54	149	170	82
N-free extractives	675	589	579	442
Calcium	8.0	45.3	14.5	6.7
Sodium	1.1	3.3	30.09	1.2

Digestibility of dry matter and organic matter in the forestomachs and small intestine of sheep ( $x \pm SD$ ;  $n = 12 = 3$  sheep  $\times$  2 samples  $\times$  2 collections)

TABLE 3

Indices	Group		
	I control	II Ca-loading	III Na-loading
DM intake, g/d	651 $\pm$ 63 <sup>a</sup>	725 $\pm$ 26 <sup>b</sup>	704 $\pm$ 70 <sup>ab</sup>
	Apparent DM digestibility, %		
forestomachs <sup>1</sup>	29.8 $\pm$ 11.4	29.1 $\pm$ 11.0	39.8 $\pm$ 8.3
small intestine <sup>2</sup>	27.7 $\pm$ 9.5	23.7 $\pm$ 11.2	22.2 $\pm$ 5.5
forestomachs + small intestine <sup>3</sup>	57.5 $\pm$ 12.7	52.8 $\pm$ 7.80	60.2 $\pm$ 7.6
OM intake, g/d	608 $\pm$ 58	635 $\pm$ 24	617 $\pm$ 64
	Apparent OM digestibility, %		
forestomachs	38.4 $\pm$ 10.2	35.7 $\pm$ 10.6	45.4 $\pm$ 7.6
small intestine	23.3 $\pm$ 9.1	22.4 $\pm$ 9.6	18.4 $\pm$ 6.5
forestomachs + small intestine	61.7 $\pm$ 11.7	58.1 $\pm$ 7.1	63.8 $\pm$ 7.4

a, b -  $P < 0.05$

<sup>1</sup> forestomachs = intake minus duodenum

<sup>2</sup> small intestine = duodenum minus ileum

<sup>3</sup> forestomachs + small intestine = intake minus ileum

The highest pH value of rumen digesta (6.90) was found before feeding and differed ( $P < 0.05$ ) from the remaining values; they were lowest between 2 and 3 h (5.29-5.33). No significant differences were noted, however, in pH values among groups (Table 4). The ammonia content in the rumen of animals receiving 20% calcium lactate in their feed was higher ( $P < 0.05$ ) than in the control and sodium groups. The intensity of fermentation, expressed as the concentration of VFA and lactic acid, was similar in the rumen of all of the sheep, and the molar ratios of  $C_2:C_3:C_4$  did not differ significantly among groups (Table 4).

The pH of the duodenal content ranged from 3.3 (I) to 3.43 (II), while that of the ileum from 7.61 to 7.65 (Table 4). The timing of sampling did not influence the pH of the duodenal content. The pH of the ileum was significantly higher before feeding in all groups ( $P < 0.05$ ; 7.84) than in the successive samples during the day (7.58, 7.64, 7.48).

Table 3 gives the apparent digestibility coefficients for DM and OM in the studied parts of the digestive tract, i.e. in the forestomachs (intake minus duodenal flow), the small intestine (duodenal minus ileal) and overall in the forestomach plus small intestine (intake minus ileal flow). DM apparent digestibility coefficients did not differ among groups and averaged 33, 25 and 57% in the forestomachs, small intestine and in the forestomachs + small intestine, respectively. The apparent digestibility coefficients of OM averaged 40% in the forestomachs, 21% in the small intestine and 61% in the forestomachs + small intestine.

TABLE 4

The pH of ruminal and intestinal digesta, ammonia, VFA and lactic acid concentrations in ruminal fluid in sheep ( $x \pm SD$ ;  $n = 18 = 3 \text{ sheep} \times 2 \text{ days collection} \times 3 \text{ samples}$ )

Indices	Group		
	I control	II Ca-loading	III Na-loading
pH	5.59 ± 0.63	5.85 ± 0.68	5.74 ± 0.61
Ammonia, mmol/l	1.2 ± 0.9 <sup>a</sup>	9.4 ± 6.2 <sup>b</sup>	1.2 ± 0.7 <sup>a</sup>
LKT, mmol/l	114.6 ± 14.0	118.8 ± 13.0	116.4 ± 12.9
Acetic acid, mmol/l	59.7 ± 17.6	66.5 ± 18.1	62.3 ± 16.8
Propionic acid, mmol/l	37.2 ± 5.2	34.7 ± 7.0	35.8 ± 4.4
Butyric acid, mmol/l	13.5 ± 2.9	13.9 ± 3.7	14.6 ± 4.1
Other VFA, mmol/l	4.2 ± 1.7	3.8 ± 1.9	3.7 ± 1.3
Lactic acid mmol/l	4.2 ± 2.8	3.9 ± 3.0	3.2 ± 2.9
Duodenal pH <sup>1</sup>	3.33 ± 0.37	3.34 ± 0.39	3.35 ± 0.41
Ileal pH <sup>2</sup>	7.65 ± 0.25	7.62 ± 0.29	7.64 ± 0.23

a, b -  $P < 0.05$

<sup>1</sup>  $n = 36 = 3 \text{ sheep} \times 6 \text{ samples} \times 2 \text{ collections}$

<sup>2</sup>  $n = 24 = 3 \text{ sheep} \times 4 \text{ samples} \times 2 \text{ collections}$

Total nitrogen intake was similar in all groups, while its flow through the duodenum and ileum (expressed in per cent of intake) did not differ among groups, although in sheep loaded with Ca and Na it was somewhat lower than in the control group (Table 5).

NaCl lowered ( $P < 0.05$ ) duodenal nucleic acid-N flow; ruminal microbial protein synthesis calculated on the basis of this flow was lower in group III loaded with Na (7.7 g N-MP/d) than in groups I and II (10.46 and 10.74 g N-MP/d, respectively) (Table 5). Excess Na in the diet reduced the digestibility of total N in the small intestine ( $P < 0.05$ ). Loading sheep with Ca and Na did not significantly affect absorption of total nitrogen in the small intestine, calculated in relation to the amount of nitrogen reaching the duodenum, which equaled 67 (II) to 69% (I) (Table 5).

## DISCUSSION

Sheep receiving calcium lactate consumed more DM ( $P < 0.05$ ) (Table 3), which could be related to the reduced energy concentration in a diet containing large amounts of Ca (19 g/d) (Table 1). A similar relationship, although statistically insignificant, was observed in sheep loaded with sodium chloride (12 g NaCl/d). Ruminants loaded with Na usually consume more water, as this is necessary to maintain electrolyte balance when the rumen is hypertonic and to support increased salivation in order to buffer the ruminal contents (Łukomski, 1980; Reffett and Boiling, 1985; Chiy and Phillips, 1993). In our study, water intake by sheep loaded with Na was 34% higher than in controls. Rogers et al.

TABLE 5  
Nitrogen and nucleic acid flow in the duodenum, total-N digestibility and absorption in the small intestine of sheep ( $x \pm SD$ ;  $n = 12 = 3$  sheep  $\times$  2 samples  $\times$  2 collections)

Indices	Group		
	I control	II Ca-loading	III Na-loading
N intake, g/d	17.4 $\pm$ 1.06	18.2 $\pm$ 0.4	17.7 $\pm$ 1.1
N flow, % of intake			
duodenum	139.7 $\pm$ 24.4	119.6 $\pm$ 21.3	119.0 $\pm$ 16.1
ileum	43.5 $\pm$ 13.4	38.8 $\pm$ 6.5	39.5 $\pm$ 8.3
N-NA, g/d	2.14 $\pm$ 0.43 <sup>a</sup>	2.20 $\pm$ 0.30 <sup>a</sup>	1.58 $\pm$ 0.33 <sup>b</sup>
N-MP, g/d	10.6 $\pm$ 2.10 <sup>a</sup>	10.74 $\pm$ 2.60 <sup>a</sup>	7.69 $\pm$ 1.62 <sup>b</sup>
Small intestine			
N-digestibility, % intake	96.2 $\pm$ 15.5 <sup>a</sup>	80.8 $\pm$ 19.2 <sup>ab</sup>	79.5 $\pm$ 10.8 <sup>b</sup>
N-absorption, % inflow	69.3 $\pm$ 7.5	67.0 $\pm$ 5.6	66.9 $\pm$ 4.7

a,b -  $P < 0.05$

(1979) observed increased water consumption and dilution rate occurring proportionately to changes in the pH and increasing rate of VFA evacuation from the rumen.

The pH of the rumen content was low in all of the sheep and averaged 5.59 in the control group, 5.85 in the calcium and 5.73 in the sodium group (Table 4). A rise in rumen pH and dilution rate after feeding diets loaded with Na was observed in bulls by Rogers et al. (1982) and Estell and Gaylean (1985), and in sheep by Thomson et al. (1978).

Calcium lactate as the calcium source in the diet is, in contrast to alkali and calcium buffers, a natural metabolite that is rapidly converted in the rumen to VFA. Loading sheep with Ca and Na did not cause any significant changes in the lactic acid and VFA contents, which averaged 3.74 and 117 mmol/l and in the proportions of C<sub>2</sub>:C<sub>3</sub>:C<sub>4</sub> equalling 56:32:12, respectively (Table 4), typical for rations containing a large proportion of concentrates. Rogers et al. (1982), Wiedemeier et al. (1987) and Erdman (1988) feeding cows with forage found increased C<sub>2</sub>:C<sub>3</sub> proportion accompanied with higher cellulolytic bacteria activity in the rumen liquor after supplementing diet with NaHCO<sub>3</sub>. However, increased cellulolytic activity in the rumen liquor is usually met at higher than determined in our experiment pH value.

Calcium lactate was used in all groups of sheep, in groups I and III for Ca supplementation. In previous studies (Leontowicz et al., 1994) were found negative retention of Ca in a control group of sheep fed a diet balanced according to ARC requirements (1980), that is why the amount of this element, so important for growing animals, was increased in the current experiment to 5 g Ca/d in the control group.

Despite the loading with Ca and Na, DM duodenal flow was similar in all sheep, and its apparent digestibility in the forestomachs (expressed in absolute terms) was higher in the group loaded with Na ( $P < 0.05$ ) than in the control (280 vs 192 g/d) (Table 3). The higher digestibility of DM in the forestomachs of sheep loaded with Na may have been not only the result of greater Na and Ca absorption (Leontowicz et al., 1995), but also the somewhat higher digestibility of OM in the forestomachs (Table 3). The apparent digestibility coefficients of DM did not differ significantly among groups and ranged from 29 (I) to 40% (III) in the forestomachs, while from 22 (II, III) to 28% (I) in the small intestine. Phillipson (1983), Mess et al. (1985) and Leontowicz et al. (1993) did not find any effect of Na and/or Ca (Leontowicz et al., 1994) on the apparent digestibility of DM or OM.

The sheep received feeds containing similar amounts of nitrogen (17-18 g/day) (Table 5), and the concentration of NH<sub>3</sub> in the rumen liquid was low (Table 4). It was higher only in sheep loaded with Ca as the result of introducing urea (9 g/kg mixture) into the Ca mixture to compensate the additional energy introduced by

calcium lactate. The low  $\text{NH}_3$  concentration may have been related to the slower rate of degradation of extruded feeds in the rumen and their efficient utilization by microorganisms, accompanied by only slight absorption from the forestomachs under conditions of lowered pH (Table 3).

The amount of total N entering the duodenum, expressed in per cent of intake, did not differ significantly among groups ( $P > 0.05$ ), although in sheep loaded with Ca and Na this value was lower than in controls (119 vs 139%) (Table 5). Increased duodenal N flow was found by Wanderly et al. (1987) in bulls and by Leontowicz (1991) in sheep fed feeds slowly fermenting in the rumen in comparison with animals receiving a diet with a high proportion of concentrates.

Loading sheep with Ca did not affect nucleic acid-N flow through the duodenum, while loading with Na significantly ( $P < 0.05$ ) lowered the amount entering the duodenum (Table 5) and the ruminal microbial protein synthesis calculated on this basis; in sheep receiving sodium chloride it equaled 7.69 g N-MP/d and was significantly ( $P < 0.05$ ) lower than in the remaining groups (10.46 (I) and 10.74 (II) g N-MP/d). These results did not differ from figures reported by other authors (Czerkawski, 1974; Evans et al., 1975; Leontowicz, 1991). In searching for the cause of reduced protein synthesis in sheep loaded with Na, it can be supposed that a higher dilution rate reduces the extent of ruminal proteolysis and deamination (Reffet and Boiling, 1985), which could lead to a rise in the amount of feed protein digested in the small intestine (Hemsley, 1975).

The depressing effect of Ca and Na on apparent digestibility of crude protein in the small intestine is evident: 96, 81 and 80% in groups I, II and III, respectively (Table 5). In contrast, the absorption coefficients of crude protein in the small intestine did not show such differences among groups loaded with Ca or Na (67%) and the control group (69%), similarly as duodenal and ileal pH, which in sheep fed extruded mixtures equaled 3.37 and 7.63, respectively.

## CONCLUSIONS

Introducing calcium lactate and sodium chloride into the diets of sheep in amounts exceeding by 5 and 10 times the ARC requirements for Ca and Na did not have a significant effect on fermentation, apparent digestibility of OM in the part of the digestive tract from the mouth to the ileum or on absorption of crude protein in the small intestine.

Intake of excessive amounts of Na (12 g/d) in the form of NaCl lowered ruminal microbial protein synthesis and the apparent digestibility of crude protein in the small intestine.



The introduction of excess calcium lactate (19.6%) and sodium chloride (8.5%) into the composition of extruded mixtures for sheep did not significantly change the pH in the digestive tract; average pH values of ruminal, duodenal and ileal digesta equaled 5.73, 3.37 and 7.63, respectively.

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## STRESZCZENIE

**Wpływ nadmiaru wapnia lub sodu w diecie na trawienie składników organicznych i wchłanianie związków mineralnych u owiec 1. Trawienie w przedżołądkach i jelicie cienkim**

W doświadczeniu przeprowadzonym na owcach z kaniułami do żwacza, dwunastnicy i jelita biodrowego badano wpływ nadmiaru Ca i Na w dawkach (5 i 10 razy więcej niż zapotrzebowanie wg ARC) na procesy trawienne w żwaczu i w jelitach. Wprowadzenie w skład ekstrudowanych

mieszanek mleczanu wapniowego (19.6%) lub chlorku sodowego (9.6%) nie miało istotnego wpływu na przebieg procesów trawiennych w żwaczu (strawność pozorną substancji organicznej wynosiła w przedżołądkach średnio 40%, w jelitach 61% oraz wartość pH treści żwacza (5.73), dwunastnicy (3.37) i jelita biodrowego (7.63). Nadmiar Na w dawce obniżał ( $P < 0.05$ ) syntezę białka mikroorganizmów w żwaczu (7.69 g N-MP/d) w porównaniu z uzyskaną u owiec kontrolnych i obciążanych Ca (10.46 i 10.74g N-MP/g) oraz strawność pozorną N-całkowitego w jelicie cienkim.