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## THE EFFECT OF CONCENTRATE FEEDING ON CALCIUM METABOLISM AND PLASMA GASTRIN CONCENTRATION IN PARTURIENT COWS\*

By

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LUTHMAN, J., S.-O. JACOBSSON & G. NILSSON: *The effect of concentrate feeding on calcium metabolism and plasma gastrin concentration in parturient cows.* Acta vet. scand. 1979, 20, 546—554. — The effect of concentrate feeding on calcium metabolism was studied in pregnant cows. The concentrate (5 kg/day) was added to the diet about three weeks before expected calving. A control group was fed only hay during the whole dry period. It was earlier observed in sheep that concentrate feeding was followed by a disturbance in calcium homeostasis, but no such disturbances were observed in the cows. The concentrate fed cows consumed 50 % more calcium than the controls and were found to mobilize less calcium from the skeleton immediately after calving. There was no evidence for the theory that the gastro-intestinal hormone gastrin is involved in calcium homeostasis in the parturient cow.

concentrate; calcium; gastrin; parturient  
paresis.

The addition of concentrate to the diet of hay-fed sheep and heifers is followed by a disturbance in calcium metabolism (Luthman & Persson 1977). In some animals serum calcium fall to levels which is often associated with paresis. The composition and mineral content of the concentrate seem to be of no importance, since the same changes can be induced by crushed oats as well as by a well balanced mineralized concentrate mixture. Recent studies did not support the theory that the disturbance in calcium homeostasis was caused by a reduced

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gastro-intestinal uptake of calcium (Jones & Luthman 1978). The primary aim of the present investigation was to study if the same changes can be induced in the parturient dairy cow.

The endocrinological changes which occur in cows at the time around partus have been intensely studied, special attention has been given to the calcium regulating hormones, parathyroid hormone and calcitonin.

It has been suggested that some gastro-intestinal hormones e.g. gastrin play a role in calcium homeostasis at least in non-ruminant animals. Gastrin is mainly considered as a calcitonin secretagogue (Cooper *et al.* 1972), but Schulak & Kaplan (1974) showed that gastrin can induce hypocalcaemia independent of the thyroid gland, and Krishnamra & Limlongwongse (1978) presented evidence for the theory that gastrin-induced hypocalcaemia may in part be explained by an increased excretion of calcium into the gastro-intestinal lumen.

Although the role of gastrin in calcium homeostasis is far from established, it was considered to be of interest to study changes in plasma gastrin concentration in parturient cows fed different diets.

## MATERIAL AND METHODS

Sixteen cows were used in the experiment, 14 were of the Swedish Red Breed (SRB) and two were Swedish Friesians. The animals were divided into two groups, one group comprising nine, and the other seven cows. The experiment started at the beginning of the dry period. The dry period varied from six to eight weeks.

The animals were fed according to the following scheme:

Group 1. Only hay (10 kg/day).

Group 2. Hay as in Group 1 plus concentrate (5 kg/day). The concentrate was introduced within the course of three days, 2 kg was given on the first day, 3 kg on the second day and from the third day the full amount. Concentrate feeding began about three weeks before expected calving.

All animals were fed twice daily. The concentrate mixture was of the following composition: oats 40 %, barley 40 %, soy bean meal 8 %, rape seed meal 5 %, molass 2 %, mineral and vitamins 2 %.

Table 1. Calculated daily intake of energy (MJ), digestible protein (g) and minerals (g).

	Group 1	Group 2
Energy	69.7	127.1
Digestible protein	92	782
Ca	41	63.5
P	24	51.5
Mg	12	20

Feed analysis was performed at the State Agrichemical Laboratory, Uppsala, according to the methods officially used in Sweden. The daily intake of nutrients was calculated from the feed analysis. The results are shown in Table 1.

The following blood parameters were studied: Glucose, non-esterified fatty acids (NEFA), calcium, inorganic phosphorus, magnesium, aspartate amino transferase (ASAT), alkaline phosphatase (AP) and gastrin. Glucose was determined on whole blood and plasma was used for analysis of NEFA and gastrin. All other analysis were performed on serum.

Basal plasma gastrin concentration was determined on three occasions, four—six weeks ante partum, one—three days ante partum, and one—three days post partum. The gastrin concentration was calculated from the analysis of three samples taken with 15 min interval before the morning feeding.

Protamine is a drug with calcitonin-like activity, e.g. it inhibits bone resorption, and can thus be used to study the role of bone resorption in various physiological conditions. The intravenous injection of protamine is followed by a fall in serum calcium which is a direct reflection of the outflow of calcium from the skeleton. This technique has been used earlier in both sheep and cattle (*Persson & Luthman 1975, Luthman et al. 1977*).

Protamine tests were performed one—three days post partum. The cows were given an intravenous injection of protamine sulphate at a dose of 10 mg/kg, and blood was sampled for calcium determination after 1, 2, 4 and 6 h.

The following analytical methods were used: NEFA (*Dole 1956*), serum calcium (*Luthman & Persson 1975b*), inorganic phosphorus (*Fiske & Subbarow 1925*), serum magnesium by atomic absorption according to instructions given by the manu-

facturer of the instrument (Pye Unicam), ASAT and AP were determined kinetically in a Kemion enzyme analyzer (reagents from AB Kabi, Stockholm). Gastrin was analyzed by the radio-immunological method described by Nilsson (1975).

Conventional statistical methods (Student's t-test for paired and unpaired data) were used for comparison of the results.

## RESULTS AND DISCUSSION

It was observed in earlier studies that addition of concentrate to the diet was followed by changes in serum calcium after a few days in non-pregnant sheep, while in pregnant animals the greatest changes occurred after 15—30 days (Luthman & Persson 1977). In the present study no changes in serum calcium was observed immediately after the change of the diet.

The blood chemical data obtained in the two groups of cows are summarized in Table 2.

Plasma NEFA increased significantly at the time of calving in both groups. In Group 1 the rise in NEFA began already one week before parturition and was thereafter significantly higher than in Group 2. Increased lipid mobilization has been described previously in parturient cows (Luthman & Persson 1975a, Horst *et al.* 1976). Decreased insulin secretion rate as a result of hypocalcaemia and increased production of catecholamines and cortisol are generally considered as the causes of the increased lipid mobilization, but the present study showed that also the nutritional status of the cow is of importance.

According to Swedish feeding standards the energy requirement of SRB cows in late pregnancy is 72.6 MJ and the requirement of digestible protein is 650 g. As seen from Table 1 the protein intake of the cows in Group 1 was far below the requirement, while the demand for energy was almost met. Clinical ketosis occurred about one week post partum in two cows in Group 1 and in one cow in Group 2. These three cows also showed the highest NEFA level at parturition.

There were no significant changes in blood glucose and inorganic phosphorus between the groups.

The intake of calcium was about 50 % higher in Group 2 than in Group 1, but there were no significant changes in serum calcium between the groups. Serum calcium decreased, however, in both groups at partus. Compared with the value obtained

Table 2. Blood glucose, non-esterified fatty acids (NEFA), minerals (mmol/l) and serum enzymes ( $\mu$ kat/l) in parturient cows fed two different diets. Mean  $\pm$  s.e.m.

	Days ante partum			Partus	Days post partum	
	30-20	7	3		1	2
	NEFA					
Group 1	0.62 $\pm$ 0.06	0.84 $\pm$ 0.09 <sup>1</sup>	1.24 $\pm$ 0.14 <sup>3</sup>	1.29 $\pm$ 0.14 <sup>1</sup>	1.34 $\pm$ 0.15 <sup>1</sup>	1.17 $\pm$ 0.11
Group 2	0.51 $\pm$ 0.06	0.53 $\pm$ 0.06	0.57 $\pm$ 0.07	0.87 $\pm$ 0.16	0.79 $\pm$ 0.15	0.85 $\pm$ 0.32
	Glucose					
Group 1	2.02 $\pm$ 0.20	2.09 $\pm$ 0.19	2.10 $\pm$ 0.18	2.86 $\pm$ 0.28	2.27 $\pm$ 0.29	1.88 $\pm$ 0.30
Group 2	2.16 $\pm$ 0.13	1.98 $\pm$ 0.08	1.96 $\pm$ 0.11	3.47 $\pm$ 0.28	2.20 $\pm$ 0.13	2.11 $\pm$ 0.31
	Calcium					
Group 1	2.39 $\pm$ 0.04	2.33 $\pm$ 0.05	2.32 $\pm$ 0.14	2.11 $\pm$ 0.05	2.17 $\pm$ 0.05	2.20 $\pm$ 0.06
Group 2	2.54 $\pm$ 0.07	2.44 $\pm$ 0.08	2.37 $\pm$ 0.13	2.02 $\pm$ 0.11	2.02 $\pm$ 0.09	1.93 $\pm$ 0.19
	Inorganic phosphorus					
Group 1	1.63 $\pm$ 0.07	1.62 $\pm$ 0.05	1.52 $\pm$ 0.09	1.19 $\pm$ 0.17	1.23 $\pm$ 0.19	1.21 $\pm$ 0.18
Group 2	1.66 $\pm$ 0.25	1.77 $\pm$ 0.26	1.79 $\pm$ 0.37	1.29 $\pm$ 0.18	1.26 $\pm$ 0.11	1.21 $\pm$ 0.19
	Magnesium					
Group 1	0.90 $\pm$ 0.03	0.81 $\pm$ 0.02 <sup>3</sup>	0.87 $\pm$ 0.03 <sup>1</sup>	0.93 $\pm$ 0.02 <sup>2</sup>	0.93 $\pm$ 0.03 <sup>1</sup>	0.92 $\pm$ 0.05 <sup>2</sup>
Group 2	0.96 $\pm$ 0.03	1.05 $\pm$ 0.05	1.01 $\pm$ 0.04	1.14 $\pm$ 0.05	1.13 $\pm$ 0.08	1.24 $\pm$ 0.07
	S-ASAT					
Group 1	0.85 $\pm$ 0.08	0.96 $\pm$ 0.12	1.09 $\pm$ 0.19	1.26 $\pm$ 0.27	1.36 $\pm$ 0.24	1.62 $\pm$ 0.45
Group 2	0.93 $\pm$ 0.04	0.98 $\pm$ 0.03	1.03 $\pm$ 0.06	1.32 $\pm$ 0.19	1.39 $\pm$ 0.09	1.35 $\pm$ 0.11
	S-AP					
Group 1	0.88 $\pm$ 0.13	1.40 $\pm$ 0.58	1.93 $\pm$ 0.78	1.49 $\pm$ 0.42	1.62 $\pm$ 0.37	1.85 $\pm$ 0.67
Group 2	0.73 $\pm$ 0.12	1.02 $\pm$ 0.20	1.22 $\pm$ 0.32	1.40 $\pm$ 0.27	1.29 $\pm$ 0.28	1.25 $\pm$ 0.32

<sup>1</sup> 0.01 < P < 0.05

<sup>2</sup> 0.001 < P < 0.01

<sup>3</sup> P < 0.001

seven days prepartum the value at partus was significantly lower in Group 1 (0.001 < P < 0.01,  $t = 4.463$ ) and almost significantly lower in Group 2 (0.1 < P < 0.05,  $t = 3.209$ ). One cow in Group 2 died from hypocalcaemia in association with partus. The serum values obtained from this cow until death are included in the results given in Table 2.

The daily intake of magnesium was lower in Group 1, and the serum level was also significantly lower in this group from seven days prepartum and then during the whole observation period.

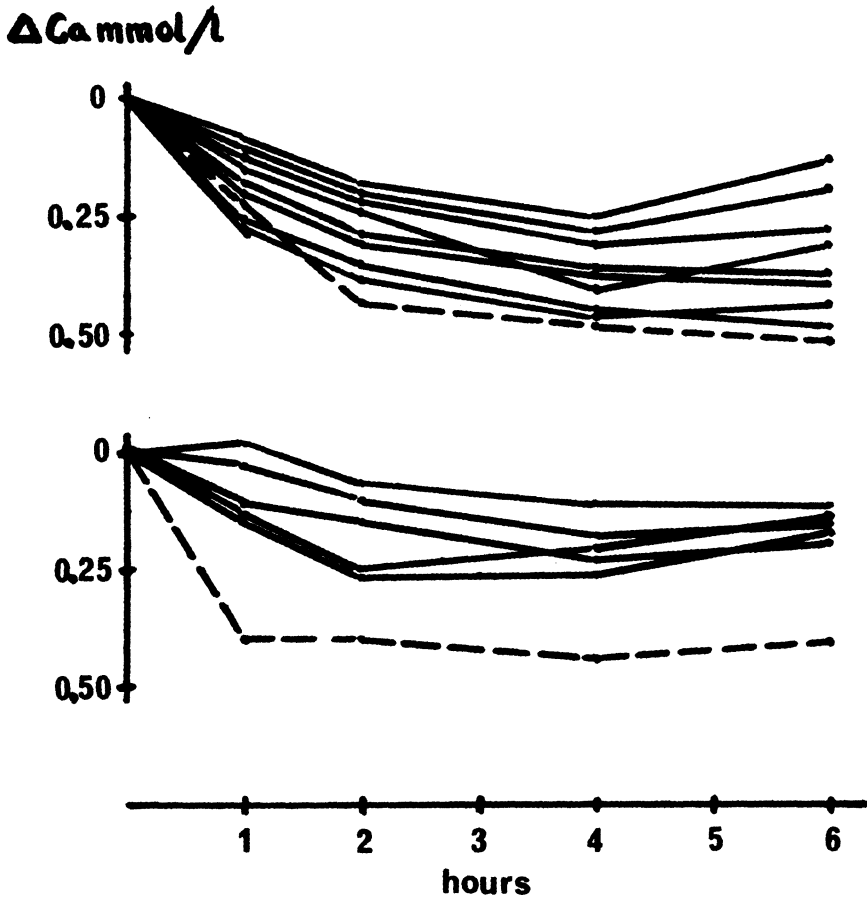


Figure 1. The hypocalcaemic response to protamine in parturient cows fed only hay (top) and hay concentrate, 5 kg/day (bottom).

— = Swedish Red cows.  
 - - - - = Swedish Friesian cows.

The results of the protamine tests are shown in Fig. 1. As shown in the figure the Friesian cows showed the greatest hypocalcaemic response. The SRB breed is more susceptible to parturient paresis than Friesians (*Ekesbo 1966*) and the responsiveness to vitamin D is different between the two breeds (*Jönsson 1978*). It may therefore be justifiable to assume that the effect of different diets on calcium metabolism also differs. The difference in protamine response between the two groups is shown in Table 3. The hypocalcaemic response was slightly

Table 3. Changes in serum calcium after intravenous injection of protamine. Mean  $\pm$  s.e.m.

	Hours			
	1	2	3	4
	$\Delta$ Ca (mmol/l)			
	All cows			
Group 1	-0.19 $\pm$ 0.02	-0.29 $\pm$ 0.02	-0.39 $\pm$ 0.03	-0.35 $\pm$ 0.05
Group 2	-0.15 $\pm$ 0.06	-0.21 $\pm$ 0.06	-0.25 $\pm$ 0.05 <sup>1</sup>	-0.22 $\pm$ 0.04
	Only SRB-cows			
Group 1	-0.19 $\pm$ 0.02	-0.28 $\pm$ 0.02	-0.38 $\pm$ 0.03	-0.32 $\pm$ 0.04
Group 2	-0.09 $\pm$ 0.03 <sup>1</sup>	-0.17 $\pm$ 0.05 <sup>1</sup>	0.21 $\pm$ 0.03 <sup>2</sup>	-0.18 $\pm$ 0.01 <sup>1</sup>

<sup>1</sup> 0.01 < P < 0.05<sup>2</sup> 0.001 < P < :0.01

greater in Group 1 after 4 h (0.05 < P < 0.01, t = 2.823). If the Friesian cows are excluded, differences occurred already after 1 h, and the difference was significant after 4 h (0.01 < P < 0.001, t = 3.999). It seems logical that Group 2 which consumed 50 % more calcium than Group 1 was less dependent on calcium mobilization from the skeleton.

As discussed previously gastrin is known as a calcitonin secretagogue, but there is evidence that gastrin may induce alterations in calcium homeostasis also by other mechanisms. Table 4 shows the plasma gastrin levels in the cows. There were no significant changes between the groups, and plasma gastrin did not change at partus. It was earlier observed that pregnant ewes fed a calcium rich diet showed higher plasma gastrin levels than ewes fed a normal diet (Luthman et al. 1977). The difference in gastrin response to diet obtained in these investigations may be explained by the fact that the sheep were fed the experimental diet for six months and the cows only for three weeks.

Table 4. Plasma gastrin (pg/ml) in parturient cows fed two different diets. Mean  $\pm$  s.e.m.

	Gastrin (pg/ml)		
	4-6 weeks ante partum	1-3 days ante partum	1-3 days post partum
Group 1	68 $\pm$ 27	78 $\pm$ 18	74 $\pm$ 26
Group 2	55 $\pm$ 8	74 $\pm$ 13	58 $\pm$ 3

*Swaminathan et al.* (1973) suggested the existence of a gastro-intestinal thyroid C cell system as an integral part of post-prandial calcium homeostasis in non-ruminant animals. The role of gastrin in ruminants is uncertain, as feeding is not followed by a rise in plasma gastrin (*Nilsson et al.* unpublished).

The influence of feeding on the incidence of parturient paresis has long been a matter for discussion. Excellent reviews on this topic were recently published by *Jorgensen* (1974) and *Jönsson*. The results obtained in the present study did not support the idea that rapid addition of concentrate to the diet increases the susceptibility to parturient hypocalcaemia, and there were no evidence for a role of gastrin in the calcium homeostasis of the parturient cow.

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#### SAMMANFATTNING

*Effekten kraftfoderutfodring på kalciummetabolismen och plasma-gastrinkoncentrationen hos kor vid kalvningen.*

Det har tidigare observerats att snabbt insättande av kraftfoder medför rubbningar i kalciumhomeostasen hos får. I en grupp högdräktiga kor insattes kraftfoder omkring tre veckor före beräknad kalvning. En kontrollgrupp utfodrades med enbart hö under hela sinperioden. Försöksgruppen konsumerade 50 % mera kalcium än kontrollgruppen. Resultaten visade att försöksgruppen mobiliserade mindre kalcium från skelettet än kontrollgruppen omedelbart efter kalvningen. Resultaten visade även att ett snabbt insättande av kraftfoder inte tycks öka risken för paresis puerperalis. Plasmakoncentrationen av gastrin ändrades inte vid kalvningen.

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