J. H. Westerhuis

Institute for Animal Feeding and Nutrition Research 'Hoorn', Hoorn

Parturient hypocalcaemia prevention in parturient cows prone to milk fever by dietary measures

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Abstract

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Over 5 years (1967-1972) at Hoorn feeding measures in dry period and over the calving period intended to prevent low concentrations post partum of calcium were tested with about 170 parturient cows prone to milk fever. The trials examined influences of changes in Ca and P contents of the diet, a day of fasting, feeding below recommended requirements for energy and protein, infusions of ethylenediaminetetraacetate intravenously or intramuscularly and their effects on postpartal concentrations of Ca, P and Mg in plasma and on packed cell volume. From the evaluation of results, dietary measures were designed that prevented low plasma levels of Ca in a trial with 45 cows prone to milk fever. These cows were fed to a Ca-poor diet (33.1-43.9 g/day) pre partum and a Ca-rich diet (148.3-196.8 g/day) post partum and adequate in energy, protein, P, Mg and vitamin D. No milk fever occured and in only 4 of the cows did plasma Ca decline below 7.5 mg/100 ml.

Recommended measures are as follows: (1) Provide a prepartal diet as low as possible in Ca ($\leqslant$ 0.50% Ca in dry matter); P and vitamin D intakes must be sufficient. (2) Just after calving, increase Ca intake by an oral dose of 250 g $CaCO_3$ (100 g Ca) by bottle and increase Ca content of the diet ($>$ 1.0% Ca in dry matter); P and vitamin D intakes must be sufficient. (3) In the weeks pre and post partum, provide an extra Mg of 30 g daily to prevent tetanic symptoms and low concentrations of Mg in plasma.

Contents

List of abbreviations and symbols

Ca	=	calcium
conc.	=	concentrate
contr.	=	control
dcp	=	digestible crude protein
dm	=	dry matter
EDTA	=	ethylenediaminetetraacetic acid
exp.	=	experimental
iu	=	international unit
Mg	=	magnesium
Na	=	sodium
nm	=	nanometer
P	=	(inorganic)phosphate
part.	=	parturition
p.c.v.	=	packed cell volume (haematocrit)
PTH	=	parathyroid hormone
rev/min	=	revolutions per minute
SE	=	starch equivalent
TCT	=	thyrocalcitonin

1 Introduction

In the Netherlands in 1971, the total number of milking cows was 1.9 million (Central Office of Statistics & the Institute for Agricultural Economics, 1972). Of these, 1.1 million (60%) were cows that had calved at least twice, among which about 45000 were susceptible to milk fever (4% according to Kemperman, 1971) and would show symptoms of milk fever at parturition time. Because a loss of milk production of about 35 litres at 0.40 Dutch guilders and at least one injection of calcium ions per cow by the veterinary surgeon at *f* 32, a direct loss can be calculated of about 2.1 million Dutch guilders, about £ 300000. An indirect loss caused by extra labour, by mastitis, death or slaughter of the cows and stillborn calves cannot be calculated.

In the last decennium in the Netherlands, a lot of work has been done on vitamin D injection which has been shown to have a preventive action.

Kemperman (1971) showed that high-yielding older cows that developed milk fever at delivery had a 25% chance of milk fever in the following year. All these considerations explain my interest in basic information about the decline of calcium in blood plasma around parturition.

The purpose of my research was to investigate the value of some feeding measures that might prevent the phenomenon of parturient hypocalcaemia. Already many research workers have studied this aspect of parturient hypocalcaemia with more or less success. In the period 1967 – 1972, 'Hoorn' carried out a number of experiments[1] with cows prone to milk fever and which were fed on diets with different amounts of minerals.

In other countries too, there has been some change in milk fever research. In 1970, three years after I started the study, the proceedings of a conference on milk fever, held in November 1968 at the University of Illinois, USA, appeared in print (Anderson, 1970). Some of the lectures at this conference supported my conviction that feeding measures can provide a means of preventing milk fever in dairy cows. Since no practical method of preventing milk fever was given at that conference, I continued the work.

My study paid no attention to the therapy of the disease or to dietary administration of vitamin D, nor to dietary measures which may support the therapy.

1. This study was initiated by the late Dr. J. van der Grift, who after a year had to stop his work in 1968, because of serious illness.

2 Literature

Shortage of minerals will occur when the output exceeds input. For calcium, the adult cow has a large reserve. This reserve can be drawn on, for example, by dairy cows early in lactation when calcium output is much greater than input.

In contrast to calcium, no reserve of magnesium is readily available when output of magnesium from blood exceeds input. In spring when the dairy cows are grazing on pasture rich in protein and potassium, the total input of magnesium may be depressed (Kemp, 1958; Kemp & Geurink, 1966) and grass tetany may occur almost immediately.

Hypocalcaemia occurs when there is a sudden increase in calcium output. Usually this happens soon after delivery with the onset of milk production.

2.1 Effect of hormones

For regulation of input and output of minerals from body fluids, the working mechanism of certain hormones must be known.

2.1.1 Parathyroid hormone

Parathyroid hormone stimulates absorption of calcium from the intestines (Cramer, 1963; Toverud, 1964) and the resorption of calcium from the bone (Mayer et al., 1967). It was thought that hypocalcaemia at delivery might be due to a parathyroid gland insufficiency (Dryerre & Greig, 1928). Investigations of the ultrastructure of parathyroid gland (Capen & Young, 1967) and the parathyroid hormone concentration of plasma (Mayer, 1970) indicate that parturient hypocalcaemia is not due to an insufficiency of the parathyroids.

Furthermore the secretory rate of parathyroid hormone is controlled by the calcium concentration in plasma: the lower the plasma calcium the greater the secretory rate (Ramberg et al., 1967; Sherwood et al., 1968). Mayer (1970) observed in general the highest plasma parathyroid hormone concentrations in severely hypocalcaemic cows with paresis. In general, parathyroid hormone stimulates calcium input, but the target organs need some time to react to changes in plasma concentration of parathyroid hormone. With certain rations bone as a target organ needs a time of 10 to 20 days to increase calcium mobilization, after a sudden increase in plasma concentration of parathyroid hormone around parturition (Ramberg et al., 1970b). The reaction time

of intestinal calcium absorption is much shorter and is pinpointed at about 3 days post partum (Ramberg et al., 1970b).

In summary, hypocalcaemia may occur within the first three days post partum despite of high concentrations in plasma of parathyroid hormone and active parathyroid glands because of the low rate of adaptation in calcium homeostasis.

2.1.2 Thyrocalcitonin

Thyrocalcitonin restores the calcium content of the bone and lowers the calcium and phosphorus content of plasma (Friedman & Raisz, 1965; Wallach et al., 1967; Barlet, 1973). High levels of plasma calcium stimulate release of calcitonin by the thyroid gland (Care et al., 1967). By intravenous infusion of exogenous thyrocalcitonin in 4 lactating cows 30 months of age, Barlet (1967) induced a decline in concentrations of calcium and inorganic phosphate in plasma of 20 and 30%, respectively. In recent histological studies, Capen & Young (1967) showed a calcitonin depletion of the thyroid gland at delivery. They concluded that an abrupt release of a significant amount of thyrocalcitonin near parturition might contribute to the development of hypocalcaemia. Also plasma thyrocalcitonin levels were found to be higher in paretic than in control cows (Barlet, 1969; Littledike et al., 1971).

However, Mayer (1972) found that during the decline in plasma calcium after calving concentration of thyrocalcitonin in plasma was not higher than pre partum in cows developing paresis. Mayer (1970) continuously infused thyrocalcitonin by vein for 4 h 2 to 5 days post partum in 4 cows 6 – 10 years old that had recovered from parturient paresis. A decline in calcium concentration in plasma was not observed. An abrupt intravenous injection of thyrocalcitonin 3 weeks post partum, even in a cow 2 years old with a good adaptability of calcium metabolism, did not cause a notable decline in plasma calcium. So Mayer could not prove that abrupt release of thyrocalcitonin, as occurs at delivery, contributes to the development of hypocalcaemia in parturient cows 2 – 5 days post partum.

On one hand, since thyrocalcitonin seems to lower concentration of calcium in blood by reducing resorption of calcium from bone (Friedman & Raisz, 1965; Milhaud et al., 1965; Wallach et al., 1967), a low bone metabolism in older cows may have been responsible for the apparent lack of response to thyrocalcitonin.

On the other hand, a better explanation of the poor results from thyrocalcitonin infusions carried out by Mayer (1970) would be that the cows he used already had an activated intestinal calcium absorption 2 – 5 days post partum (Ramberg et al., 1970b): the cows at that moment would already be under the influence of parathyroid hormone.

Continual exposure of the bone to the restoring action of thyrocalcitonin over several weeks pre partum (induced by a diet rich in calcium, for example) may induce a low turn-over rate of bone calcium and a low adaptability of the resorption mechanism to high levels of parathyroid hormone at the onset of lactation (Mayer, 1972). An abrupt release of thyrocalcitonin may have a short-lasting working action since the

half-life of thyrocalcitonin in the circulation is only 5 – 15 min (Deftos et al., 1968). Hypercalcaemia, as mentioned above, has been reported as the stimulus for thyrocalcitonin release in other species than the cow (Care et al., 1967); this mechanism may work in the cow too. Despite frequent sampling, no hypercalcaemia was observed just before the onset of hypocalcaemia (Mayer, 1970). The concentration of calcium in plasma of cows which did develop paresis usually began with a progressive decline 24 to 48 hours before parturition (Mayer et al., 1969).

On the usual calcium-rich diets for cows in calf bone resorption at parturition did not increase rapidly, despite the increased concentration of parathyroid hormone in plasma (Kronfeld, 1968). This may be due to a low turn-over rate in bone induced by a continuously high level of calcitonin in plasma before calving. The level of dietary intake of calcium pre partum seems to have caused the differences in results from thyrocalcitonin studies (Mayer, 1972; Barlet, 1973).

2.1.3 Oestrogen

The ratio of oestrogen to progesterone in blood is important during pregnancy. An excess of progesterone is then necessary to maintain the lack of sensitivity of the uterus to influences of oxytocin hormone.

In dairy cows (Short, 1958), ewes (Edgar & Ronaldson, 1958) and in rabbit (Mikhail et al., 1961) but not in woman (Short & Eaton, 1959), the progesterone level in plasma goes down a few days before parturition and the influences of oestrogen become more important. An increase in concentration of oestrogen in plasma just before parturition was mentioned by Saba (1964) in dairy cows and by Raeside (1963) in sows.

Oestrogen plays a role in intermediary metabolism too. After an injection of oestradiol valerate into goats, Anderson et al. (1973) diagnosed a decline in appetite and in phosphate concentration in blood plasma. But direct influence of oestrogen on calcium concentration has not been proved. Also at oestrus concentration of plasma oestrogen is increased and appetite lowered.

When appetite is lowered input of calcium and phosphate will be small. Thus, oestrogen will have some indirect influence on mineral concentrations in blood around parturition. According to the latest literature, the phosphate decline and the calcium decline at parturition are not connected. Two separate mechanisms are held responsible for the phosphate and calcium decline (Anderson et al., 1970).

A persistent influence on the mineral and carbohydrate metabolism from high oestrogen levels in plasma after parturition can hardly be present because oestrogen in blood is quickly broken down. Although very little is known about the influence of oestrogen on the partus syndrome, high oestrogen levels together with other disturbances of hormonal homeostasis may initiate a decline in appetite and probably parturient hypocalcaemia.

2.1.4 Adrenal corticosteroids

Littledike et al. (1968) found high concentrations of corticoid (hydrocortisone) and of glucose in plasma of paretic cows. Osinga (1959) made differential counts of the white blood cells in paretic cows and found symptoms identical with those during stress.

Blood cell pictures found by Garm (1950), Merill & Smith (1954) and Osinga (1959) are in agreement with the three stages of the differential counts of the white blood cells in the stress syndrome described by Selye (1950):

- battle stage (neutrophilia, eosinopenia and lymphopenia),
- victory stage (increasing amounts of monocytes),
- recovery stage (lymphocytaemia and return of eosinophils).

Since the concentration of corticoid in plasma is high (Littledike et al., 1968), the cortex of the adrenal glands is highly activated. In normal circumstances, the adrenal glands can be activated by an increased activity of the adenohypophysis. Osinga (1971) and Drost (1972) induced parturition nearly at term by injection of corticosteroid. They based their theory and treatment on the increase in production of foetal corticoids at the end of pregnancy (Drost et al., 1968). The question is whether the activity of the maternal or foetal adrenal gland is responsible for the high corticoid level in maternal blood plasma at parturition.

Perhaps in the first stage, these foetal corticoids will induce the parturition. Still it is not completely understood whether a high concentration of corticoid in plasma is important in inducing low calcium levels, although Littledike et al. (1968) proved a decreasing effect on calcium absorption from corticoid hormone. The possible direct relation between corticoid concentration in plasma and low calcium levels in plasma needs more investigation.

The high levels of parathyroid hormone (Mayer, 1970) and corticosteroid hormone (Littledike et al., 1970) in plasma and the differential counts of white blood cells seen under 'stress' conditions (Osinga, 1959) make it likely that a general derangement of hormonal regulation rather than a one single factor contributes to parturient hypocalcaemia.

A causal relationship of corticoid hormone and hyperglycaemia in normal cows would be expected, because of the known actions of hydrocortisone on carbohydrate metabolism and as an antagonist of insulin. In cows with parturient paresis, there is a higher positive correlation between the incidence of elevated plasma concentration of hydrocortisone and hyperglycaemia than in normal dry cows and in healthy (non-paretic) parturient cows (Littledike et al., 1970).

In normal cows the concentration of glucose in plasma starts to rise about 18 h pre partum; 3 h post partum a maximum is reached and 18 h post partum plasma glucose levels are again on prepartal levels (Osinga, 1959). Cows with parturient paresis have a significant higher glucose concentration in plasma than normally calving non-paretic cows (Osinga, 1959; Littledike et al., 1970).

2.1.5 Insulin

Insulin does increase the uptake and oxidation by peripheral tissues and the conversion of glucose into glycogen in muscles and in liver. A normal concentration of insulin in plasma is necessary to maintain the metabolism of peripheral tissues, in particular muscles.

Besides this effect of insulin in cell metabolism, insulin is necessary to keep glucose between normal levels in blood plasma. In parturient hypocalcaemic cows, there is a significantly lower level of insulin, and higher levels of glucose, corticoids and lactic acid in blood plasma (Littledike et al., 1970). Studies in vitro on rat tissues (Curry et al., 1968) and experiments with parturient hypocalcaemic cows (Littledike et al., 1970; Blum et al., 1973) suggested that calcium ions are necessary for a proper release of insulin from cells of the pancreatic gland. Higher levels of corticoid and lower levels of insulin in plasma do enhance the elevated levels of glucose found in parturient cows. This hyperglycaemia is associated with an increase in the plasma concentration of lactic acid (Littledike et al., 1970; Singh et al., 1972) and with a decrease in the number of movements of the rumen and in the amplitude of contractions (Singh et al., 1972). Thus parturient hyperglycaemia seemed to be caused by temporary deficiency of insulin released from the pancreas, which in turn is caused by shortage of calcium ions in plasma. The hyperglycaemia may induce rumino-intestinal stasis, which aggravates the shortage of calcium ions.

2.1.6 Discussion

In normal circumstances, hormonal system is adaptable enough to react to changed living conditions in dairy cows. The stress of parturition can be so intense that abnormal hormonal changes occur in plasma. The influence of oestrogens just before parturition may cause poor appetite and a smaller intake of calcium. Calcium-rich diets pre partum stimulate the production of thyrocalcitonin, which inhibits calcium mobilization from bone. They also enhance a decline in parathyroid hormone production, therefore intestinal calcium absorption remains low. In parturient normal and paretic cows, there is an increase in plasma concentration of parathyroid hormone, corticosteroid hormone, oestrogen and, perhaps for a short period, of thyrocalcitonin. Because the colostral output increases and the input decreases (poor appetite and an inhibited mobilization from bone), calcium in plasma tends to decline.

Whether the cow becomes paretic or not depends largely on the adaptability of the animal, i.e. on activation of intestinal absorption and mobilization of calcium from bone.

The literature suggests that it is not advisable to seek a single hormonal regulator to prevent parturient hypocalcaemia. Enlarging the adaptability of the cows to the sudden output of calcium with the colostral milk may have more effect.

2.2 Input and output of calcium

To know why mineral absorption and resorption is deranged at parturition time in certain dairy cows, it is necessary to balance against one another the input and output of calcium from the immediately available pool.

In pregnant dry cows, the calcium metabolism is in balance: that is input from intestine and bone equals output into bone, faeces, urine and foetus. At delivery, the exchange into the foetus becomes zero but the output to colostrum is above output to the foetus in the dry period and may cause parturient hypocalcaemia (Kronfeld & Ramberg, 1970; Ramberg et al., 1970b).

Ramberg et al. (1970b) made a diagram (Fig. 1) by a compartment building method with the digital computer program Simulation Analysis and Modeling (SAAM program) developed by Berman et al. (1962). At delivery and during the two weeks post partum, the role of calcium mobilization from bone is crucial. During the first three days post partum an important input of calcium from bone cannot be expected and an increased output of calcium may cause hypocalcaemia (Payne, 1964). To compensate for the big demand for calcium, the intestinal absorption will increase after delivery but requires a few days to reach an optimum. In the dry period in the older cows, the calcium input from intestines together with a small amount from the bone is more than enough for a normal life (Sansom, 1969; Ramberg et al., 1970b; Kronfeld & Ramberg, 1970).

Thus, to give the parturient cow more resistance against hypocalcaemia the following measures appear suitable:

- enlargement of the immediately available calcium pool,
- slow down the calcium output from the blood,
- increase the input into blood.

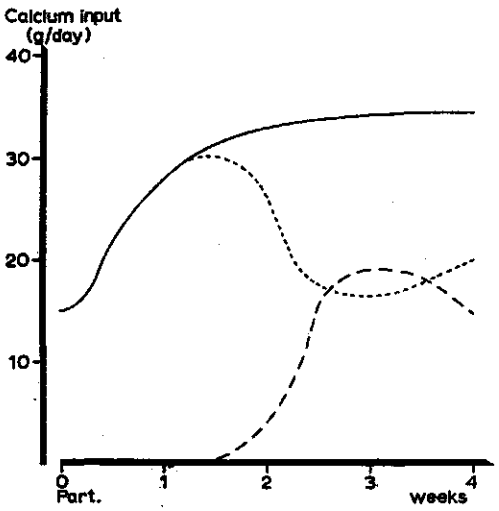

Fig. 1. Changes in calcium input in response to the onset of lactation (from Ramberg et al., 1970b). ——— = total input; — = gastrointestinal absorption; – – – = bone resorption.

Enlarging the calcium pool. A daily addition of 320000 iu vitamin D to the ration of a dairy cow can enlarge the immediately available calcium pool (Muir et al., 1968).

It is worth remembering that protection against milk fever reached a peak after 3 days on a vitamin D supplement and that protection declined drastically if parturition occurred more than 1 day after vitamin D feeding was terminated (Hibbs & Conrad, 1960).

On the basis of some trials by Hibbs et al. (1970), feeding of 20 million iu of vitamin D daily for at least 3 days and not more than 7 days before parturition offers a safe and effective method of preventing parturient hypocalcaemia.

Slowing down output from blood. In parturient cows, it is difficult to influence output of colostral calcium, which is the most important output fraction. The daily output of calcium in urine is small. A complete stop in this output can only compensate for the calcium loss of 1 litre milk. The cessation of the output to the foetus compensates for the calcium lost in about 5 litre milk. In cows with severe hypocalcaemia, endogenous faecal calcium decreases temporarily. The average decrease in endogenous faecal calcium might be the same as in 1–2 litre milk per day (Ramberg, 1972).

In severely hypocalcaemic cows a temporary suppression of the milk calcium outflow is established. This will happen by means of a decreased colostral milk production during the first days post partum (Ramberg, 1972).

Increasing the input. This may be another way of enlarging the immediately available pool. This can be achieved by an increased intestinal absorption and an increased bone resorption. It is difficult to increase bone resorption in a few hours (Ramberg et al., 1970b; Ramberg, 1972) as is necessary just after calving. But by feeding a low-calcium diet during the dry period, calcium resorption from bone will be stimulated (Ramberg, 1972).

Increasing intestinal absorption takes a few days (Hibbs & Conrad, 1960; Manston, 1967; Wasserman & Taylor, 1969). Feeding with vitamin D or a single massive injection of vitamin D will increase intestinal absorption after a few days. This is due to the fact that vitamin D is transformed into 1,25-dihydroxycholecalciferol partly in the liver and the kidneys (Lawson, 1971). This 1,25-dihydroxycholecalciferol stimulates the synthesis of a calcium-binding protein in domestic fowl (Wasserman et al., 1968). This protein seems to be necessary to transport calcium through the intestinal wall. In ruminants, it is reasonable to assume that this protein is also important in the intestinal calcium absorption mechanism, because this protein is also found in rat, monkey, dog and calf. Generally when the needs of calcium are increased, for example, by milk production of cows on a ration low in calcium or in young growing animals, the absorption of calcium can adapt itself. This occurs by a more active transport of calcium through the intestinal wall and by an enlarged surface of the small intestine able to absorb calcium (Schachter et al., 1960; Dowdle et al., 1960; Kimberg et al., 1961, Malm, 1963).

2.3 Various influences on calcium metabolism

Increasing the output or decreasing the input to the immediately available calcium pool may make calcium metabolism more adaptable. This can be done by
- removing calcium by milking,
- fasting,
- parenteral administration of a calcium-binding agent like EDTA.

2.3.1 Milking pre partum

Several people have advocated milking pre partum as a method of preventing milk fever. Research work by Smith & Blosser (1947) did not support or reject this advice. In a group of 46 cows that were milked in the dry period beginning 2 – 16 days before parturition, 9 cases of parturient paresis were diagnosed. Compared to a control group, there was no significant decline in paretic cases. Also Kendall et al. (1968, 1970) investigated the removal of colostrum pre partum. They used 8 cows as a control group and 14 cows, of which 2 had paretic histories, as an experimental group which were milked pre partum. Paresis appeared in 1 control and 7 cows milked pre partum. These 7 cows produced 2.8 kg colostrum during the last 24 h pre partum and the other 7 experimental cows which were also milked pre partum but did not develop paresis produced 8.5 kg colostrum. They concluded that cows milked pre partum which initiated colostrum secretion earlier and produced more (8.5 against 2.8 kg colostral milk) on the day before parturition did not develop the paresis syndrome but had concentrations of calcium and inorganic phosphate in blood plasma that were about normal and were comparable to those of the control cows not milked pre partum. In contrast to these cows, the cows milked pre partum that produced only small amounts of colostrum developed parturient paresis.

Through milking pre partum the loss of calcium from the body increases and when the output of calcium is bigger than the input hypocalcaemia occurs after some while when the immediately available calcium pool is exhausted. The adaptability of calcium metabolism reacts to the hypocalcaemia with a release of parathyroid hormone which in turn stimulates intestinal absorption and removal from bone of calcium ions. The secretory rate of this hormone seemed to be controlled by plasma concentration of calcium: the lower the plasma calcium the greater the secretory rate of parathyroid hormone (Ramberg et al., 1967; Sherwood et al., 1968).

Thus milking pre partum can have a positive effect upon the activation of calcium metabolism, so that milking pre partum cannot be rejected. But milking pre partum is not only far from practical, it is also abnormal physiologically.

2.3.2 Fasting in pregnant dry cows

Although few exact measurements have been made, it is a common opinion that at parturition the cow will to some extent refuse food. Some authors have reported the

effect of fasting on the mineral content of blood (Gowen & Tobey, 1936; Allcroft & Godden, 1934; Aylward & Blackwood, 1936; Smith & Dastur, 1938; Seekles, 1939; Halse, 1948, 1958a, 1958b).

Halse (1960) compared results of mineral disturbances in blood of milking cows, subjected to a short period of fasting with the decline in plasma calcium of normally calving non-paretic cows. After 2 days of fasting, he found signs of restlessness, muscular tremor and unsteadiness while serum calcium was between 5.8 and 6.5 mg/100 ml. Some cows fell in a 'mineral crisis' and injections of calcium-magnesium solution had to be used to save their lives. As a rule, serious signs of illness appeared quite suddenly about 36 – 60 h after the start of fasting. Before serious signs, he noted a decline in plasma calcium and phosphorus and an increasing magnesium content of plasma.

In summary, we may conclude that hypocalcaemia will occur even in milking cows after a few days of fasting. This will be followed by an increase in production of parathyroid hormone (Ramberg et al., 1967), resulting in a higher absorption from intestine and a higher mobilization from bone. Further investigations are necessary to prove whether fasting for some while has a preventive value in parturient hypocalcaemia.

2.3.3 Infusion or injection of ethylenediaminetetraacetate sodium

Blood calcium levels can also be lowered by intravenous injections of calcium-binding agents such as EDTA. About 95 – 98% of a calcium-EDTA complex is excreted in urine in about 6 h after injection of EDTA (Foremann et al., 1953), which means that the artificial removal of calcium from the blood is very rapid. Many research workers have used EDTA in measuring the exchangeable calcium pool (Payne, 1964; Muir et al., 1968; Hibbs et al., 1970) and to diagnose parathyroid insufficiency (Kaiser & Ponsold, 1959). In man, Kaiser & Ponsold lowered the plasma calcium from 9.5 to 7.5 mg/100 ml with an intravenous infusion of EDTA (70 mg/kg bodyweight in 2 h). About 10 h after starting infusion in patients with a good working parathyroid gland, the plasma calcium returned to a normal level again. In patients with parathyroid insufficiency, the plasma calcium was still below starting values 12 h after starting infusion.

Muir et al. (1968) measured the immediately available calcium pool with a challenge technique for blood calcium with EDTA as described by Payne (1964) but with certain modifications. In lowering plasma calcium from 10.0 to 4.5 mg/100 ml, he used an infusion of 97.9 mg EDTA per kg bodyweight in 4 h. From the recovery time of plasma calcium after the start of the infusion, the immediately available calcium pool was calculated to be 10.3 g for a cow. In the plasma, 5.2 g of calcium had to be bound and excreted in order to lower plasma calcium from 10.0 to 4.5 mg/100 ml.

Because of the low levels of plasma calcium, more parathyroid hormone may be released, causing a shorter recovery time. Therefore the value of this method of depressing plasma calcium is limited to a measurement of the immediately exchangeable

calcium pool over a short period of time.

Thus this technique can only be used in a steady state of calcium metabolism. There must be some days of rest before the immediately available pool can next be measured because this technique will lead to a non-steady state of calcium metabolism.

Later I will discuss work on EDTA infusions in stimulating calcium metabolism (Sections 3.5 and 3.6).

2.3.4 Infusions of calcium ions

Intravenous infusion of calcium ions has a hypercalcaemic effect and parathyroid hormone production will decrease (Ramberg et al., 1967). A short infusion time (about 5 min) resulted in hypercalcaemia, which continued a short time in comparison, for instance, to a continuous infusion of calcium for 1 week. A short infusion will therefore give a small depression in parathyroid gland activity.

During a long-term infusion of calcium ions, levels of plasma calcium return to normal after a hypercalcaemic period and a steady state will be achieved (Ramberg, 1972). As soon as the infusion is stopped, severe hypocalcaemia occurs. The return to normal levels is more rapid in cows fed on a diet low in calcium than in cows fed on a calcium-rich diet. Ramberg concluded that a long-term infusion of calcium pre partum causes a decline in absorption of calcium from the diet and a decline in removal of calcium from bone, in other words mechanisms that prevent hypercalcaemia were stimulated.

2.3.5 Injection of vitamin D

Vitamin D should now be viewed as a provitamin or prohormone. The liver transforms vitamin D into 25-hydroxycholecalciferol, which in turn is transformed into 1,25-dihydroxycholecalciferol by the kidneys. This 1,25-dihydroxycholecalciferol is biologically the most active metabolite of vitamin D. The physiological effect of vitamin D or its metabolites, especially 1,25-dihydroxycholecalciferol, on the biosynthesis of a calcium-binding protein is definitely established (Schachter & Kowarski, 1965). Addition of vitamin D results in an increasing absorption of calcium from the intestine, which indirectly tends to cause hypercalcaemia and an increased level of inorganic phosphate in human blood (Brickman et al., 1972) and in dairy cows (Manston & Payne, 1964).

1,25-Dihydroxycholecalciferol also stimulates some calcium removal from bone in anephric rats (Holick et al., 1972). The transformation of vitamin D into 1,25-dihydroxycholecalciferol and the production of calcium-binding protein is a process that takes some time. A massive dose of vitamin D results in a higher level of plasma calcium about 2 days after injection (Seekles & Hendriks, 1966). The concentration of plasma calcium reached a maximum about 4 days after injection of 10 million iu vitamin D_3. Therefore this injection must be administered to cows prone to milk fever not less than 2 days before calving.

Although the input of calcium seems to be higher after administration of vitamin D_3, the output of calcium also increases, since a higher excretion in urine and a higher production of urine per day could be detected (Seekles & Hendriks, 1966; Hendriks & Seekles, 1966). It is remarkable that the urinary excretion of calcium reached a maximum two days after the massive dose of vitamin D_3.

In summary, injections of massive doses of vitamin D given to cows prone to milk fever cannot be seen as an ideal means of preventing milk fever, since protection is far from complete.

Seekles et al. (1958) claimed 80 – 90% protection of cows prone to milk fever. That means that 80 – 90% of the injected cows prone to parturient hypocalcaemia did not develop milk fever symptoms. A recent study in Canada by Cote et al. (1970) on 2 Holstein-Friesian and on 2 Jersey herds showed that parturient paresis did not develop in 55% of cows with a history of milk fever and susceptible cows 5 or more years of age, which were also given 10 million iu vitamin D_3 by intramuscular injection. For the cows with a history of milk fever protection was only 48%.

Physiologically, higher concentrations of plasma calcium will depress the activity of the parathyroid glands. Lower concentrations of parathyroid hormone in plasma may result in a smaller absorption surface of the intestine and may slow down the production of calcium-binding protein (Ramberg & Olsson, 1970).

2.4 Effect of calcium and phosphate in the ration on calcium metabolism

Dryerre & Greig (1928), Sjollema (1928) and many others related the occurrence of parturient hypocalcaemia to the calcium content of the diet during the dry period and advanced the hypothesis that parturient hypocalcaemia may be due to parathyroid insufficiency. Although nowadays this hypothesis has been discarded, Boda & Cole (1954) altered the diet of Jersey cows from one high in calcium and low to normal in phosphate to one low in calcium and high in phosphate. The Ca:P ratio was changed from 6:1 to 1:3.

The number of cows studied was small, feeding levels were low, concentrates not very palatable and few analyses were made of blood. However Boda & Cole concluded that a diet low in calcium and high in phosphate stimulated the activity of the parathyroid glands and so stimulated the release of parathyroid hormone. Also Boda & Cole (1955) had good results with this type of diet because in a group of 23 Jersey cows (11 with a history of milk fever) only one got parturient hypocalcaemia with paretic symptoms. Because no control group was available some difficulties arise in interpreting their results.

In a field study reported by Stott (1965), a Jersey herd showed a decline in the incidence of paresis after 6 months of feeding with a supplement of 5% monosodium phosphate added to the concentrate. The total daily intake changed from 137 g Ca and 85 g P to 150 g Ca and 162 g P. The incidence of parturient paresis declined from 74% before to 16% after the dietary change. Stott did not verify the diagnosis of milk fever by testing blood for calcium. Still 16% of the herd went down with milk fever

symptoms. Thus more information is needed before one can state that diets rich in calcium, which predispose the cows to milk fever, can be completely corrected with a supplement of monosodium phosphate in order to bring the Ca:P ratio to 1:1.

The proposal of Boda & Cole (1954) to lower the daily intake of calcium and to lower the Ca:P ratio to 1:1 or 1:3 is more drastic than that of Stott (1965) only to lower the Ca:P ratio to 1:1 or 1:3.

In agreement with Boda & Cole (1954), Ender et al. (1962) stated that diets rich in calcium before calving must undoubtedly be characterized as a dominant causal factor in parturient paresis. They also still believed in insufficient parathyroid activity. But Jönsson (1960), Capen et al. (1965), Capen & Young (1967), Ramberg et al. (1967), Sherwood et al. (1968) and Mayer et al. (1969) concluded that parathyroid activity was the same in normal calving and in parturient hypocalcaemic cows. Other factors than parathyroid hormone production seem to play a predominant role in determining the degree of hypocalcaemia in parturient cows because Mayer (1967) reported cows with low blood calcium (about 6 mg/100 ml plasma) coupled with high blood levels of parathyroid hormone in parturient hypocalcaemic cows in contrast to normally calving cows.

On the basis of the results and advice of Boda & Cole, recent investigations have been published about diets low in calcium pre partum preventing milk fever. Data from Boda & Cole (1954, 1955) provide evidence of the efficiency of diets low in calcium pre partum in the control of parturient hypocalcaemia. Diets low in calcium pre partum stimulate bone resorption (Ramberg et al., 1970b). The decrease in incidence of parturient hypocalcaemia is related to calcium intake and not to phosphate intake or to Ca:P ratio of the diet (Niedermeier et al., 1949; Robertson et al., 1956; Mayer, 1970, 1972).

On the basis of available knowledge, Mayer (1970) stated the hypothesis, given in Figure 2. On a diet rich in calcium, the input of calcium into the plasma pool comes mainly from absorption from the intestine. There is also a net accretion of calcium in bone, but with a slow turnover rate of calcium in bone. So parathyroid production is low and the production of thyrocalcitonin is higher. Input of calcium from bone into the plasma pool is not important in this situation to maintain the concentration of plasma calcium at normal levels. Since appetite is usually low and rumino-intestinal stasis will occur at parturition calcium absorption will be diminished and hypocalcaemia will follow.

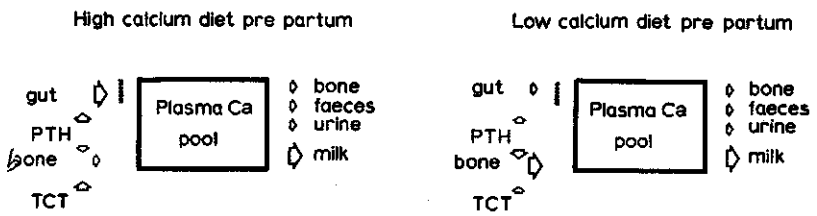

Fig. 2. Calcium homeostasis at parturition (from Mayer, 1970).

Low calcium diets (Fig. 2) will stimulate removal of calcium from bone in order to maintain normal levels of plasma calcium. At parturition, the mobilization of calcium from bone will already be activated and therefore more important for calcium homeostasis than intestinal absorption.

This hypothesis is supported by Ramberg (1972). He concluded that it is not usually possible to alter the plasma concentration of calcium by changing the calcium content of the diet because of the inverse relationship between absorption from intestine and calcium mobilization from bone. When absorption of calcium increases, the rate of calcium removal from bone decreases by an equal amount so that total input of calcium into the blood remains constant.

In ruminants the absorption of calcium from the intestine is continuous because of the gradual release of food from the forestomachs. In contrast to non-ruminants, ruminants are not geared to a sudden big demand for calcium as usually happens just after parturition. It takes a few days to increase the intestinal calcium absorption and usually many days to increase the calcium mobilization from bone (Ramberg et al., 1967; Ramberg et al., 1970b).

An increase in calcium input is necessary to cover the higher output soon after parturition with the onset of lactation. Ramberg et al. (1970b) and Ramberg (1972) reported on calcium removal from bone in two groups of parturient cows fed on a diet low in calcium or high in calcium pre partum (Fig. 3). These results support the hypothesis of Mayer (1970) that a diet low in calcium pre partum stimulates input of calcium by higher mobilization of calcium from bone.

Ramberg (1972) concludes that cows conditioned to a diet low in calcium in the dry period can rely on calcium absorption from intestine to a smaller extent at parturition and, because of the previous exercise of the mechanism for resorption of cal-

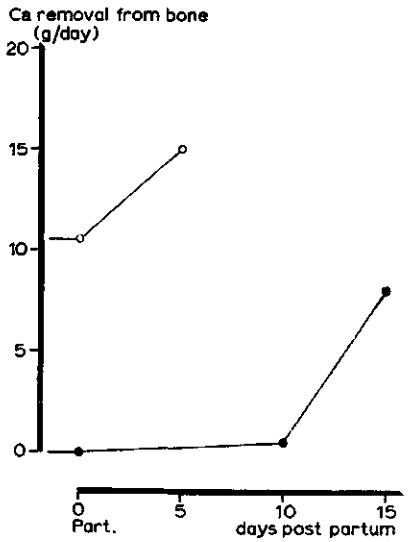

Fig. 3. Start of an increased removal of calcium from bone after delivery in cows fed on a prepartal high (●) or low (○) calcium diet (from Ramberg, 1972).

cium from bone, permits more rapid and sufficient adaptation to the onset of the calcium drain in lactation.

2.5 Influences caused by the alkali-alkalinity of the ration on calcium metabolism

2.5.1 Mineral acid silage

Although the calcium or phosphate content of the fodder seems to be important, the alkali-alkalinity can play a role in preventing parturient hypocalcaemia (Ender et al., 1962). In feeding experiments over the years 1952 – 1961 Ender et al. reported that diets rich in calcium (intakes of 140 – 170 g Ca daily) with normal to low phosphate and with a normal to high alkali-alkalinity resulted in manifest symptoms of milk fever. Few of the cows were entirely normal during the puerperium. The Ca:P ratio varied from 5:1 to 10:1.

However, symptoms of milk fever did not appear at delivery in cows fed on a large quantity of mineral acid silage (25 – 35 kg silage daily) so the diet has a low alkali-alkalinity and was rich in calcium and normal to low in phosphate.

Ender & Dishington (1970) give the following explanation: 'Acidity of the intestine gives rise to increased ionization of dietary calcium, by which calcium is more easily absorbed.'

It is a pity that no estimates were made of available bone calcium. Perhaps high excretion of calcium into the urine would cause a high output of calcium induced by the acid fodder. So the demand for calcium from the available bone calcium would increase. Although Ender et al. (1962) and Ender & Dishington (1970) had some success in preventing milk fever by feeding cows on large amounts of mineral acid silage around parturition, no complete explanation is given how and why this results was obtained.

2.5.2 Concentrates with ammonium chloride

In England, Payne (1967) has used ammonium chloride in a 'steaming-up' ration pre partum. He used a dose of 25 g up to 100 g daily (about 25 g/kg concentrate). He mentioned that in trials in Norway acid diets were highly effective in preventing milk fever and he hoped that similar beneficial results would be obtained by adding ammonium chloride in the United Kingdom.

Payne reported that his research suggested two further methods of preventing milk fever (Payne, 1970). The former involved increasing the 'acidity' of the diet by addition of ammonium chloride to the 'steaming up' ration. This method was undergoing a large-scale field trial, but trial was not then finished.

Vagg & Payne (1970) investigated the addition of ammonium chloride to the diet of non-pregnant dry Ayrshire cows with the radioisotopes ^{85}Sr and ^{45}Ca. Because of the small number of animals and the variability of the results, there was no significant increase in the size of the exchangeable calcium pool as a result of the treatment with ammonium chloride, although some cows did show an increase in that calcium pool.

Neither was there a significant increase in calcium absorption from the intestine. But an increase in urinary calcium excretion seemed to be significant. Similar experiments on cows prone to milk fever would be of interest.

The latter method involved the injection of a new calcium binding polymer, but this method needs more pharmacological development before field trials begin, according to Payne.

In the Netherlands, Unilever investigated the addition of ammonium chloride to concentrates in a similar way to Payne (unpublished results, H. van der Ploeg, Unilever Research, Maarssen). They used the following scheme:

In the dry period

- 7 th and 6 th week pre partum 1 kg 'special' cake, daily intake 25 g NH_4 Cl
- 5 th week pre partum 2 kg 'special' cake, daily intake 50 g NH_4Cl
- 4 th and 3 th week pre partum 3 kg 'special' cake, daily intake 75 g NH_4Cl
- last 2 nd week pre partum 4 kg 'special' cake, daily intake 100 g NH_4Cl

On 16 dairy farms, 82 cows (more or less prone to milk fever) were fed on these NH_4Cl concentrates and 69 cows on the same farms were controls. There was a 20% better result in the experimental group in preventing milk fever than in the control group. One has to remember that this trial was a small field trial lasting one year without support of biochemical analysis.

As mentioned by Payne, Unilever too would need more basic information about influences of NH_4Cl on the calcium metabolism before they could introduce this cake for dairy cows.

2.6 Effect of magnesium on calcium absorption and the working mechanism of parathyroid hormone

Cramer & Dueck (1962) investigated the absorption mechanism of calcium in the dog's small intestine. With calcium solutions infused into prepared loops of small intestine, they found that the absorption mechanism of calcium can be saturated. A certain amount of calcium is absorbed. This amount of absorbed calcium can be diminished by adding magnesium ions to the infused liquid.

The competition between calcium and magnesium ions and the saturation of the absorption mechanism suggested a carrier mechanism in the wall of the intestine. This competition is found not only in dogs but also in rats (Alcock & MacIntyre, 1960; Hendrix et al., 1963).

In ruminants too, there is a competition between magnesium and calcium absorption, but it is found only in the middle part of the small intestine and not in the first part (Care & van 't Klooster, 1965). Because in ruminants the main absorption of calcium takes place in the proximal part and magnesium in the distal part of the small intestine (Care & van 't Klooster, 1965), the negative influence on absorption of calcium by magnesium may be not so disastrous as suggested by the Mineral Research Committee (1973) in the Netherlands. Because the usual diets for ruminants are rich in calcium and normal or poor in magnesium there might be a more important

influence upon the intestinal magnesium absorption by an excess of calcium intake. When the needs for calcium are low, for example in the dry period, a small amount of calcium will be absorbed and the large amount of calcium left in the gut can interfere with magnesium absorption. Further research is necessary because no literature gives exact results about this problem.

Magnesium seems to be important for a good working of the parathyroid hormone as well as being essential for many enzymic processes in the cell. Estep et al. (1969) and Muldowney et al. (1970) saw that an injection of exogenous parathyroid hormone in human patients with hypocalcaemia, hypomagnesaemia and tetany did not elevate the plasma calcium but did to some extent increase excretion of phosphate into the urine. After a magnesium infusion, the action of exogenous parathyroid became normal again and that injection resulted in normal calcium plasma. It is therefore reasonable to believe that normal magnesium levels in plasma are necessary to quarantee a good working of parathyroid hormone.

2.7 Effect of Solanum malacoxylon on calcium metabolism

A disease of cattle in the lowland areas of Buenos Aires, Argentina, known as 'Enteque Seco' killed about 300000 cattle a year (Camberos et al., 1970); in Brazil too, this disease was recognized and called 'Espichamento'. The symptoms are loss of appetite, increase in blood calcium and inorganic phosphate, and stiffness with a characteristic calcification of the cardiovascular system. This disease seems to be caused by eating leaves of a plant *Solanum malacoxylon* (Carillo & Worker, 1967; Worker & Carillo, 1967).

With an aqueous extract of the leaves of *S. malacoxylon* with the active agent, which may be a glucoside, the same symptoms could be provoked in cattle, sheep and guineapigs as mentioned by Camberos et al. (1970). Balance studies with sheep and guineapigs (Camberos et al., 1970) and balance studies combined with administration of the isotope ^{47}Ca in cows (Sansom et al., 1971) showed that the unknown active agent of *S. malacoxylon* leaves induced an increased absorption of calcium, magnesium and inorganic phosphate from the diet by the gut. The absorption coefficient of calcium from the diet in cows was increased from 10% in control cows to between 40 and 50% in cows eating some leaves of *S. malacoxylon*.

In spite of a slight increase in endofaecal losses, the retention of calcium measured with a whole body counter was 5 – 7 times as great as in the control cows (Sansom et al., 1971).

Although little is known about this active agent in these leaves, it might be interesting to do some studies with pregnant dry cows prone to milk fever with an addition of leaves of *S. malacoxylon* to their diet.

3 Own trials

3.1 Introduction

During the years 1967 to 1972 cows treated for milk fever in previous years (cows prone to milk fever) were borrowed from dairy farmers in the northern part of the Province of North-Holland, situated north of the City of Amsterdam. The average dairy cow in this part of the Netherlands has a milk production which is some 500 kg higher than in the rest of the Netherlands. Most of the borrowed cows came from herds with an average annual milk production of about 6000 kg with at least 4% fat. The cows were collected and housed at a farm near Hoorn in tied stalls and cared for by the same farmer during these years. Some trials in the same period were with older dairy cows, differing in susceptibility to milk fever on the Institute farm.

Pregnant dry cows, borrowed from the farmers, arrived at the farm 4 – 6 weeks pre partum and returned to the owner 10 – 14 days after delivery. Blood was analysed at regular intervals and around delivery. Intake of energy, protein and minerals was estimated; water supply was freely available. More details of each trial are given later. The 10 trials will be treated in logical rather than chronological order.

3.2 Methods

Methods of analysis of blood and plasma, urine and feedstuffs used in the diet were as follows.

Blood The blood was taken from the jugular vein and collected in 100-ml wide-necked polythene bottles and was treated with an anticoagulant (2 drops 5% sodium heparin solution/100 ml blood). The blood was centrifuged (3000 rev/min) for 15 min with plastic granules (Styron 664). These were used in order to get an adequate separation between plasma and blood cells.

Packed cell volume (p.c.v.) The method of the International Equipment Company, Boston, Mass. was used to estimate packed cell volume (haematocrit) as percentage of whole blood. The blood was sucked into a glass capillary, 10 cm long, which was then sealed at one end and centrifuged for 3 – 4 min at a minimum speed of 11 500 rev/min. After that, the percentage of p.c.v. could easily be read from a special disk.

Calcium and magnesium in plasma Concentration of calcium and magnesium in plasma was estimated on the Techtron Atomic Absorption Spectrophotometer (Model AA-100). The plasma was diluted with a 1% solution of Na_2-EDTA (Hendriks & Klazinga, 1965; Hendriks et al., 1965) and then the readings were taken.

Phosphate in plasma Protein was precipitated from plasma with trichloracetic acid. Concentration of phosphate (as inorganic phosphorus) was estimated by the sodium molybdate method of Bell & Doisy and Benedict & Theis with a Beckman B spectrophotometer at 625 nm.

Phosphate in urine Concentration of inorganic phosphate (as phosphorus) in urine was estimated by the same molybdate method as used for plasma.

Calcium and magnesium in urine Calcium and magnesium concentrations were estimated in urine, which was diluted with a 1% lanthanium chloride solution and measured with the Techtron Atomic Absorption Spectrophotometer (Model AA-100).

Calcium, magnesium and phosphorus in roughage and in concentrates About 2 g material was ashed at 450°C. The ash was added to 5 ml aqua regia and then steamed dry. The residue was absorbed in 10 ml of nitric acid 2.5 mol/litre. This solution was then filtered and made up to 50 ml with distilled water (Solution A).

Calcium was estimated in Solution A, which was further diluted with a 1% lanthanium chloride solution until the calcium concentration was in the range 0 – 10 mg/litre.

Magnesium was estimated in Solution A, which was diluted with distilled water in such a way that the magnesium concentration was about 0 – 0.4 mg/litre. For both the calcium and magnesium concentrations, measurements were with the Techtron Atomic Absorption Spectrophotometer (Model AA-100).

Content of total phosphorus was estimated by a method which was developed by G. Misson (1908) and described later in Chemiker Zeitung, Volume 32, p. 633. A modification of the original method allowed colorimetric estimation of phosphorus in a 1:10 dilution of Solution A with a nitro vanadium molybdate reagent. The final measurements were with a Beckman DU-2 Spectrophotometer at 430 nm.

3.3 Cows prone to milk fever on a diet with a Ca:P ratio of 1:1 and a day fasting each week

3.3.1 Introduction

From December 1967 till May 1968 30 cows prone to milk fever arrived at the farm about 4 weeks before parturition. They were allotted at random to the control or experimental group. Ten days post partum they were returned to their owners.

This trial combined the method of Stott (1965) – raising the phosphorus content of

the diet until a Ca:P ratio of 1:1 was reached – and the method of one-day fasting to stimulate the adaptability of the calcium-regulating mechanism. As a consequence of fasting lower plasma calcium occurs (Halse, 1960) and the absorption of calcium from the diet is stimulated by an increased production of parathyroid hormone (Ramberg et al., 1967). Data on the control and experimental group are listed in Table 1.

The cows of the control group ate 10 kg grass hay, artificially dried with cold air, and 4 kg concentrates daily. The cows of the experimental group were fed on the same hay and a concentrate with another mineral content (Table 2). Also a concentrate without any addition of minerals was used to feed the cows of the experimental group according to energy and protein requirements. The content of dry matter, energy, protein and minerals of the ingredients of the diet are listed in Table 2.

A supply for 7 days was given to the experimental group in six days, so every week on Sunday they had a day fasting (Table 3). After delivery, the quantity of concentrates was daily increased by 0.5 kg so that on the 6th day post partum 7 kg concentrates were supplied containing the nutrients as given in Table 4.

Table 1. Data on the control and experimental group.

	Control	Experimental
Number of cows	14	16
Average age (years)	8.8	8.2
Average past frequency of milk fever	2.7	1.7
Bodyweight (kg)	608	673

Table 2. Contents of the ingredients of the ration (g/kg product).

	dm	SE	dcp	Ca	P	Mg	Ca:P
Grass hay	843	385	83	4.9	2.6	1.7	1.9
Control concentrate	900	656	130	7.9	5.5	2.9	1.4
Experimental concentrate	900	656	130	3.5	18.9	2.9	0.2
Concentrate without minerals	900	656	130	4.6	5.6	2.8	0.8

Table 3. Weekly intake (g) of nutrients per cow.

	dm	SE	dcp	Ca	P	Mg	Ca:P
Contr.	83 720	45 318	9 450	564.2	336.0	200.2	1.7
Exp.	83 720	45 318	9 450	464.1	429.8	198.2	1.1

Table 4. Daily intake (g) of nutrients on the 6th day post partum per cow.

	dm	SE	dcp	Ca	P	Mg	Ca:P
Contr.	15 560	8 442	1 740	104.3	64.5	43.3	1.6
Exp.	15 560	8 442	1 740	80.1	78.5	36.7	1.0

3.3.2 Results and discussion

Blood samples were taken and analysed at arrival, at 24 h and 7 days post partum (Table 5).

Not only are cases of milk fever important: more important to this work are the numbers or percentages of cows with a marked decline in plasma calcium after delivery. The results of this trial are listed in Table 6.

Although there were few cows with milk fever, 57% of the control group (8 of 14 cows) had a calcium concentration in blood plasma below 7.5 mg/100 ml, for the experimental group this percentage was 44 (7 of 16 cows). Also in the cows which did

Table 5. Average blood plasma content of calcium, phosphorus and magnesium (mg/100 ml) and packed cell volume (%) of the blood.

	Group	Number of cows	p.c.v.	Ca	P	Mg
day after arrival	contr.	14	32.8	10.5	6.2	2.1
(about 4 w pre partum)	exp.	16	32.0	10.6	5.3	2.2
24 h after	contr.	14	33.4	7.7	3.8	2.2
delivery	exp.	16	33.6	8.1	4.4	2.0
7 days after	contr.	14	32.0	10.0	5.1	1.9
delivery	exp.	16	32.1	9.9	5.7	2.0
just before milk	contr.	2	36.2	5.5	2.6	2.4
fever treatment	exp.	4	35.5	5.8	3.0	1.8

Table 6. Number and percentage of cows with milk fever and with a plasma decline to below 7.5. mg Ca/100 ml plasma.

Group	Number of cows			Number with	Percentage with
	normal calving	paretic	total	Ca $\leqslant$ 7.5 mg/100 ml	Ca $\leqslant$ 7.5 mg/100 ml
Contr.	12	2	14	8	57
Exp.	12	4	16	7	44

not develop milk fever the average plasma calcium of the control group was lower than that of the experimental group at 24 h after delivery, being 7.7 mg/100 ml and 8.1 mg/100 ml, respectively.

The inorganic phosphorus plasma level was lower 24 h post partum in those cows of the control group which did not develop milk fever than in similar animals of the experimental group. Also 7 days post partum, the plasma phosphorus level was somewhat higher in the experimental group than in the control group. The feeding of extra phosphate may be the cause.

A remarkable experience with one cow in the experimental group (Truus 29) was that it showed milk fever symptoms in the dry period after a day fasting and had to be infused with calcium magnesium borogluconate (Table 7). Truus 29, a Dutch Friesian 9 years old was highly prone to milk fever, because during the last 4 weeks before parturition she went down 4 times with hypocalcaemic paresis directly after a day fasting. Also after delivery, she had to be treated twice to raise her plasma calcium and save her life. Because this highly susceptible cow was not representative, we decided also to follow a fasting pregnant cow of average susceptibility in the dry period: Emma 10, Dutch Friesian 9 years old (Fig. 4). The figure shows that in Emma 10 a 48-h fasting period results in a greater decline in blood calcium (3.4 mg/100 ml) than after parturition (1.1 mg/100 ml). Emma 10 had a good appetite after calving, which is important for the absorption of calcium from the diet.

In the fasting period and after delivery, there was a decline in plasma calcium and an increase in plasma magnesium. Emma 10 received a mixed ration of hay and concentrates and had a daily intake of magnesium of more than 30 g.

The dimension of the decline in plasma calcium post partum of both cows suggest a better adaptability of the calcium absorption from intestine and calcium mobilization from bone for Emma 10, who was kept on a calcium-poor and magnesium-rich diet in the dry period instead of Truus 29 who was fed on a calcium-rich diet with sufficient but less magnesium. It also suggests that calcium absorption from the intestine immediately after delivery is more important than calcium mobilization from bone in view of Truus' reduced appetite after delivery and the big decline in plasma calcium.

This hypothesis was supported by the same Truus 29 in the next year. The cow, which was pregnant and dry at the time, was fed on a diet rich in calcium, normal in

Table 7. Concentration of Ca, P and Mg in blood plasma (mg/100 ml) from Truus 29 before and after a 24-h fast (just before treatment).

	Ca	P	Mg
Before fast	9.5	5.3	2.3
After fast	7.3	6.9	1.9

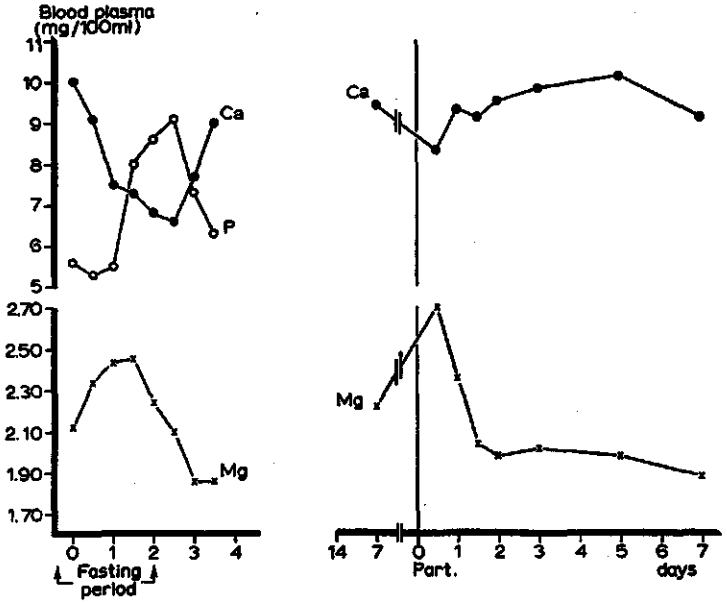

Fig. 4. Plasma concentration of calcium, phosphorus and magnesium during a fasting period of 2 days of a pregnant dairy cow prone to milk fever and plasma calcium and magnesium around parturition.

phosphorus and rich in magnesium for about 7 days. Nutrient intake was adequate, also on one day when the diet was changed from high to low in calcium, normal in phosphorus and high in magnesium. However, there were changes in concentration of plasma minerals within 48 h and the cow developed milk fever pre partum (Table 8). This agrees with my earlier opinion that calcium absorption from the intestine is more important than mobilization from bone just after delivery to prevent a serious decline in plasma calcium.

In conclusion, a Ca:P ratio 1:1 in the ration combined with a day's fast has hardly any preventive effect. Although in the control group 57% (8 of the 14 cows) and in the experimental group 44% (7 of the 16 cows) had a plasma calcium below 7.5 mg/100 ml

Table 8. Concentration of Ca, P and Mg in blood plasma (mg/100 ml) from Truus 29 before and 48 h after a dietary change from Ca-rich to Ca-poor when symptoms of milk fever appeared.

	Plasma content		
	Ca	P	Mg
Ca-rich diet	8.8	8.3	2.0
After 48 h on Ca-poor diet	4.7	4.1	2.0

a few days after delivery, the differences were too small for a definite conclusion. The reaction of the two cows to a short fast indicates the importance of the calcium absorption from intestine.

3.4 Cows prone to milk fever which were fed 20% below energy and protein requirements with a diet with a Ca:P ratio of about 1:1

3.4.1 Introduction

Because of the danger of milk fever (Truus 29: Section 3.3) after a short period of fasting and the presumable negative influence of over-feeding of pregnant dry cows during a steaming-up period on appetite, we decided to replace the fast by feeding the cows 20% below their requirements for energy during the last 4 weeks of the dry period.

The control group (16 cows prone to milk fever) was fed according to standards with barn-dried grass hay and a concentrate mixture. The experimental group (17 cows prone to milk fever) received concentrates without a mineral mixture but with a supplement of phosphorus (75 g monosodium phosphate daily) according to the theory of Stott (1965). Over 4 weeks pre partum blood samples were taken once a week and 24, 72 h after delivery and on Days 7, 10 and 14 post partum at 08 h 00 in order to assess the physiological decline in plasma calcium post partum. Within 24 h after delivery, the cows were milked completely in order to increase the output of calcium, which might also raise the incidence of milk fever. After delivery, the cows received the same concentrates as before parturition. The daily amount of concentrates increased in the same way as during the trial described in Section 3.3.

Some data are given in Table 9.

Table 9. Data on the control and experimental group and daily intake of nutrients (g).

	Control	Experimental
Number of cows	16	17
Average age (years)	7.4	7.6
Average past frequency of milk fever	2.2	2.0
Daily intake of nutrients		
dm	12000	10200
SE	5524	4212
dcp	1290	1030
Ca	82.6	53.1
P	51.0	53.5
Mg	27.6	21.7
Ca:P ratio	1.6	1.0

3.4.2 Results and discussion

Due to the ration manipulations, the experimental group received less energy, protein, dry matter, calcium and magnesium than the control group. In the experimental group 53% (9 of the 17 cows) suffered from milk fever against only 31% (5 of the 16 cows) in the control group.

Data on blood plasma at different times around parturition are given in Table 10. The data of Table 10 are plotted against time in Fig. 5.

Partitioning the data according to the lowest calcium level after delivery being above or below 7.5 mg/100 ml plasma gave the results, given in Table 11. In the experimental group, plasma calcium declined below 7.5 mg/100 ml in 13 cows (77%) and in the control group in 8 cows (50%). The average lowest plasma calcium of the cows which calved normally was 7.4 mg/100 ml at 24 h in the experimental and 8.0 mg/100 ml in the control group.

Table 10. Average packed cell volume of blood (%) and concentration in plasma of Ca, P and Mg (mg/100 ml).

	Group	p.c.v.	Ca	P	Mg
Normally calving cows					
average of once a week	contr.	31.8	10.4	5.8	2.0
sampling during 4 weeks pre partum	exp.	33.0	10.1	6.2	2.1
24 h post partum	contr.	32.0	8.0	4.2	2.1
	exp.	33.3	7.4	4.6	1.9
3 days post partum	contr.	32.0	9.2	5.1	1.9
	exp.	32.2	8.7	5.6	1.9
7 days post partum	contr.	30.2	9.5	5.5	2.0
	exp.	32.2	9.7	5.2	2.0
10 days post partum	contr.	29.5	9.7	4.7	2.1
	exp.	32.1	9.5	5.2	2.1
14 days post partum	contr.	29.1	9.9	5.1	2.1
	exp.	31.1	9.4	5.3	2.2
Cows with milk fever					
average of once a week	contr.	33.0	10.2	6.4	2.0
sampling during 4 weeks pre partum	exp.	31.8	9.8	7.0	1.9
24 h post partum	contr.	34.6	6.5	4.3	1.8
	exp.	33.4	6.7	4.3	1.5
3 days post partum	contr.	32.2	9.0	6.3	1.8
	exp.	32.3	7.8	4.0	1.9
7 days post partum	contr.	31.1	9.4	5.9	1.8
	exp.	–	–	–	–
10 days post partum	contr.	29.7	9.1	5.7	1.9
	exp.	29.4	9.2	5.5	1.9
14 days post partum	contr.	29.4	9.8	5.1	2.0
	exp.	29.0	9.4	5.4	2.1

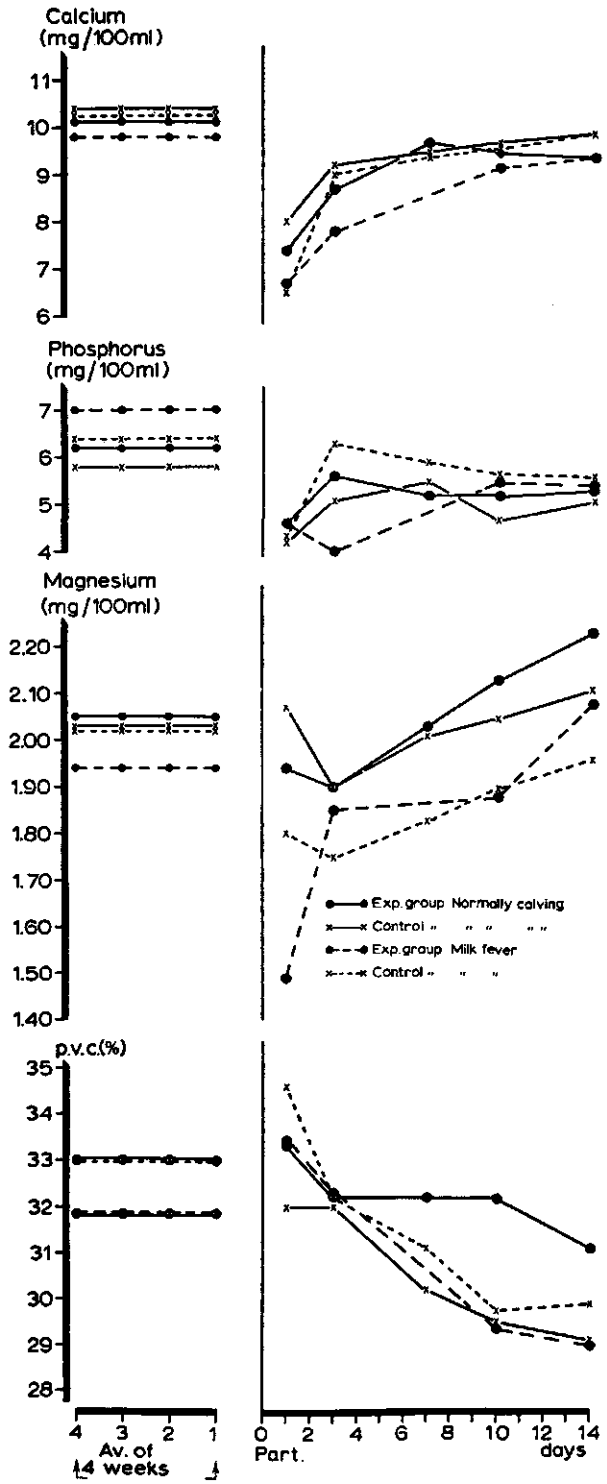

Fig. 5. Average plasma concentration of calcium, phosphorus and magnesium and packed cell volume of the blood of 4 groups of cows prone to milk fever around parturition.

Table 11. Incidence of different calcium levels and of milk fever.

Group	Plasma Ca > 7.5 mg/100 ml	Plasma Ca ⩽ 7.5 mg/100 ml	Cows with milk fever
Contr.	8	8 (= 50%)	5 (= 31%)
Exp.	4	13 (= 77%)	9 (= 53%)

Feeding cows prone to milk fever below energy and protein standards seems to have no preventive action on parturient hypocalcaemia. Figures of Table 11 suggested that this way of feeding can induce milk fever and low plasma concentration of calcium after calving.

The daily intake of minerals (Ca, P and Mg) is more important for the adaptability of calcium metabolism (Table 7 and 8). Also a sudden decrease in mineral intake was more effective than a continuously lower mineral intake in activating calcium metabolism (Fig. 4).

3.5 Experimentally induced calcium decline in plasma with Na_2-EDTA in dairy cows with no milk fever history and in calves

3.5.1 Introduction

A fall in blood calcium can be induced not only by fasting (Section 3.3) but also by an infusion of a calcium-binding agent. In human as well as in ruminant medicine, EDTA has been used as an agent for lowering calcium in blood plasma. Soon after EDTA infusion, the calcium EDTA complex is excreted in urine.

Kaiser & Ponsold (1959) infused EDTA in a quantity of 70 mg EDTA per kg bodyweight in man for 2 h. During treatment, blood calcium fell from 9.6 to 7.3 mg/100 ml.

In our experiments the phenomenon of lowering plasma calcium is used as a method of stimulating calcium metabolism by increasing the absorption of calcium from the intestine and the resorption from bone.

3.5.2 Results and discussion

Eight pregnant dry Dutch Friesian cows and three young calves were given an infusion of an EDTA solution once or twice within a certain period.

The results are given in Figures 6 – 10 and Table 12. Calcium in blood plasma of Cow 403450 and Cow 17 infused intravenously with 66 and 57 mg EDTA/kg bodyweight, respectively, decreased markedly. Because of this danger of very low calcium in blood the amount of EDTA was subsequently decreased to 50 mg/kg bodyweight (Cow 12A). The duration of infusion does not seem to influence the time taken to reach the lowest plasma calcium. In Cows 43 and 95, the lowest plasma calcium was reached 4 and 3 h, respectively, after the start of infusion but the decline is different in

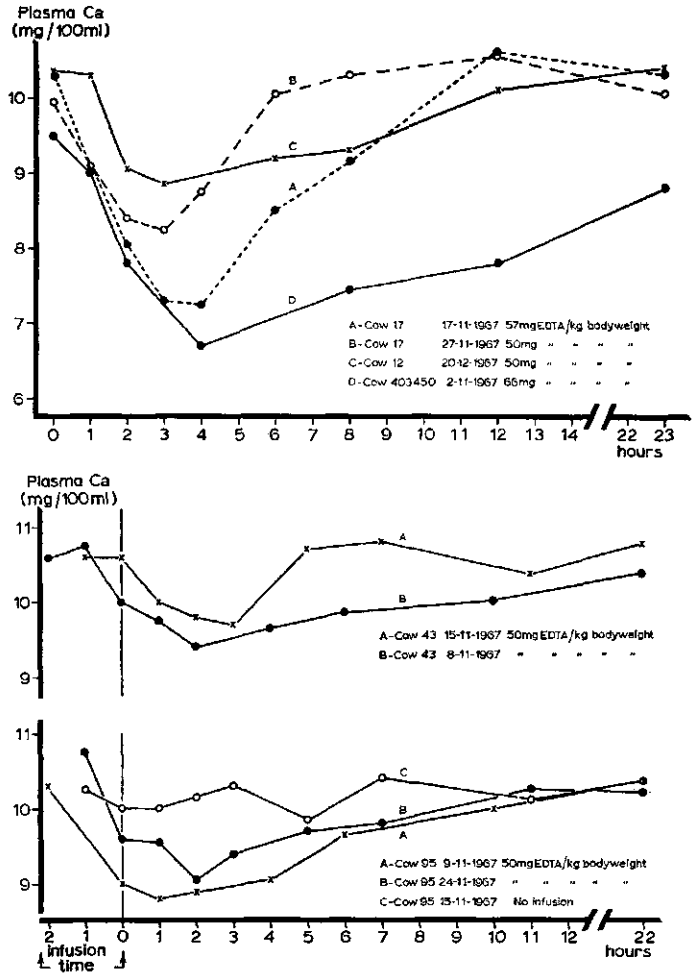

Fig. 6. Plasma calcium after EDTA infusions in 5 cows.

each cow and seems to depend upon the time interval between the first and second infusion (Cows 17, 43, 95, 10 and 68). Cow 17 (Fig. 6) was infused again 10 days later with EDTA (50 mg/kg bodyweight). As hoped, the reaction of the cow in stabilizing her plasma calcium content was better than at the first infusion. Also the recovery was faster (10 mg/100 ml plasma calcium was reached after 6 h in the second test against 12 h in the first).

Also Cow 43 (Fig. 6) showed an activated stabilizing mechanism. A sharp recovery in plasma calcium was seen within 4 – 6 h after infusion in the second test 7 days later.

With Cow 95, three tests were performed. In the first and the third test, 50 mg EDTA was infused per kg bodyweight, either in 1 or in 2 h and in the second test serving as a control no EDTA was infused. Between the first and the third test, there was an interval

Table 12. Data and results of intravenous infusion of EDTA in 8 dairy cows and 3 calves.

Cow No	Infusion on Day	Infusion time (h)	EDTA in bodyweight (mg/kg)	Decline in plasma Ca (mg/100 ml)	Time for plasma Ca to reach min. (h)
403450	0	2	66	9.5 − 6.7 = 2.8	4
17	0	1	57	10.3 − 7.3 = 3.0	4
17	10	1	50	9.9 − 8.3 = 1.6	3
12A	0	1	50	10.4 − 8.9 = 1.5	3
43	0	2	50	10.6 − 9.4 = 1.2	4
43	7	1	50	10.6 − 9.7 = 0.9	4
95	0	2	50	10.3 − 8.8 = 1.5	3
95	6	No infusion	0	10.3 − 9.9 = 0.4	6
95	9	1	50	10.8 − 9.1 = 1.7	3
12B	0	½	50	10.0 − 8.0 = 2.0	2
12B	7	(parturition)	0	10.7 − 10.3 = 0.4	12 (post partum)
Calf I	0	¼	27.7[1]	9.5 − 8.6 = 0.9	½
II	0	¼	50[1]	10.3 − 10.0 = 0.3	½
III	0	¼	70[2]	10.0 − 9.7 = 0.3	½
Cow 10	0	¼	50[1]	11.1 − 7.5 = 3.6	24
10	18	¼	44[1]	10.0 − 6.4 = 3.6	24
10	26	(parturition)	0	10.5 − 8.2 = 2.3	24 (post partum)
68	0	¼	50[2]	9.5 − 6.4 = 3.1	7
68	65	¼	50[2]	10.8 − 7.7 = 3.1	8
68	71	(parturition)	0	9.5 − 9.1 = 0.4	24 (post partum)

1. Intraperitoneal injection.
2. Intramuscular injection.

of 15 days. Figure 6 shows the changes in plasma calcium. There was no spectacular stabilizing mechanism of plasma calcium during the third test, although calcium levels fell not so far as in the first test. Probably because of a better adaptation of calcium metabolism in the cows, a reaction in plasma is seen only for a period of 7 to 10 days after experimentally induced hypocalcaemia (Cow 17, 43 and 95).

In Cow 12B (Fig. 7). I injected intravenously 50 mg EDTA/kg bodyweight in half an hour. The recovery of plasma calcium during the day of infusion was slow, but 24 h after the start of the infusion normal plasma calcium values were found again (Fig. 7). Seven days later the cow calved and there was no marked decline in plasma calcium within 36 h post partum.

The three young calves weighing 50, 50 and 70 kg, respectively, were injected intraperitoneally or intramuscularly (Table 12; Fig. 8). Calves I and II received intraperitoneally 27.7 and 50 mg EDTA/kg bodyweight, respectively, within a few minutes. Calf III received intramuscularly 70 mg EDTA/kg bodyweight also within a few minutes. All three solutions were neutralized to about pH 7.0.

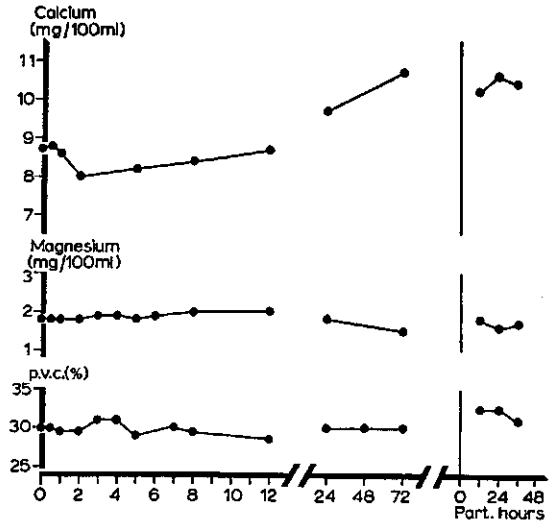

Fig. 7. Plasma calcium and magnesium and packed cell volume in blood of Cow 12B after intravenous infusion of 50 mg EDTA/kg bodyweight in 30 min and after calving.

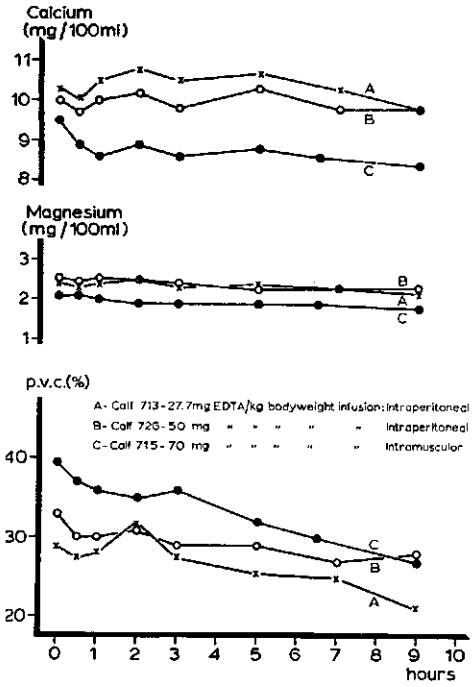

Fig. 8. Plasma calcium and magnesium and packed cell volume in 3 calves infused with EDTA intraperitoneally and intramuscularly.

After 30 days, the same infusions were repeated. Thirteen days later, the calves were slaughtered and examined. No local lesions were seen. In these calves with an active calcium metabolism, there was no decline in plasma calcium after an EDTA infusion. In these calves, a sudden loss of calcium from the blood was obviously restored by a rapid increase in calcium input; the calcium required for it seemed to be readily available.

These experiments with calves were done in order to study whether the infusion time could be shortened. Rapid intravenous infusions of EDTA (50 mg/kg bodyweight) are said to be dangerous (Goodman & Gilman, 1956). Because no lesions were seen after an intraperitoneal or intramuscular injection and because of the supposition that absorption through the peritoneum is usually slower than absorption in muscle, intraperitoneal injection was preferred. These experiments were done also because a long-term increased output of calcium might be a better stimulus to the adaptative system of calcium metabolism instead of a briefer more severe decline in plasma calcium as occurs after an intravenous injection. I therefore infused Cow 10 (Fig. 9) intraperitoneally. It showed a slow but marked decline in plasma calcium 24 h later (from 11.1 to 7.5 mg Ca/100 ml plasma). Eighteen days later during a second infusion test, there was again a fall in plasma calcium after 24 h (from 10.0 to 6.4 mg/100 ml). There is no clear indication of a more efficient adaptation of calcium metabolism due to the first infusion after the interval of 18 days. At parturition, 7 days after the second test, a physiological decline in plasma calcium was hardly seen. This agrees with my earlier observations(Table 12).

Cow 68 was twice injected intramuscularly with 50 mg EDTA/kg bodyweight. Figure 10 and Table 13 give details. In cow 68 too, the previous infusion seemed to have little effect on the lowest level of plasma Ca reached after a further infusion given about 2 months later. But at delivery, which occurred 6 days after the last infusion

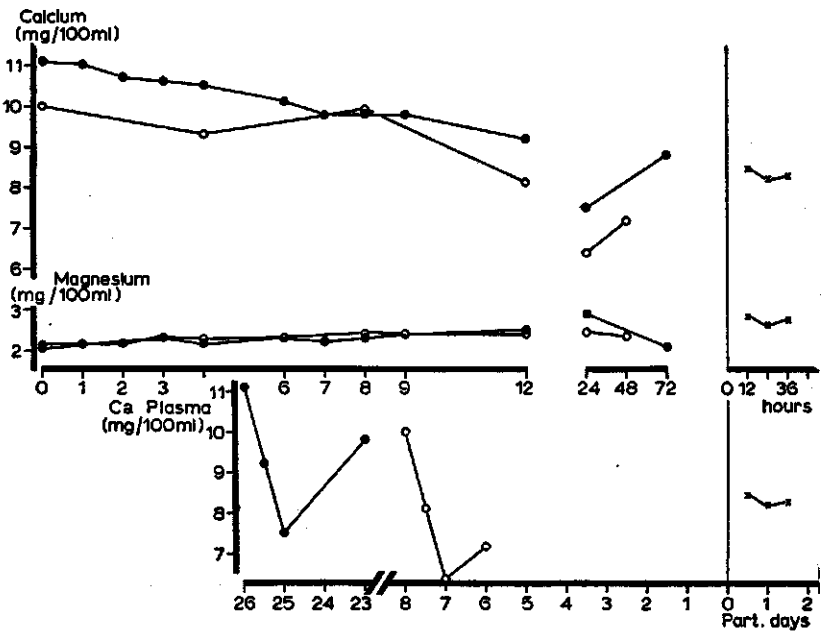

Fig. 9. Plasma calcium and magnesium in Cow 10 infused twice with EDTA intraperitoneally (for calcium only, below). ● = 8 Oct., 50 mg/kg bodyweight; ○ = 26 Oct., 44 mg/kg bodyweight; x = post partum.

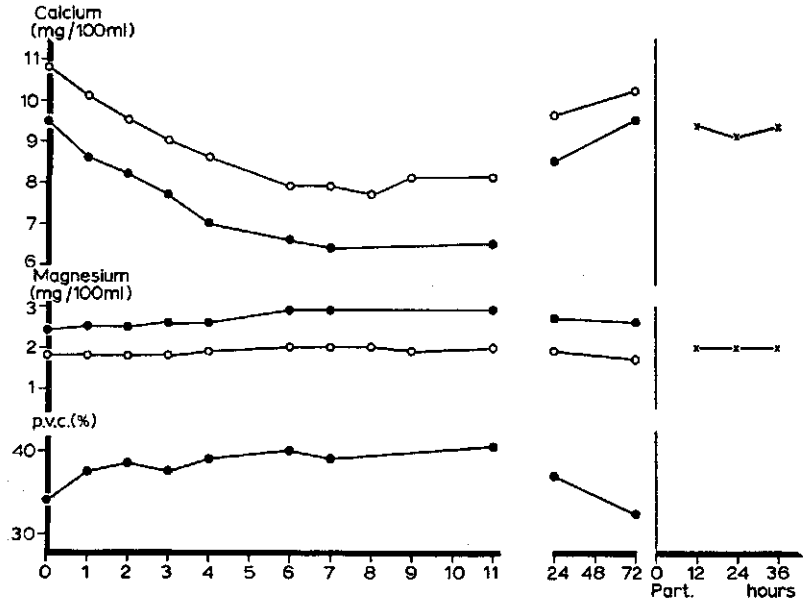

Fig. 10. Plasma calcium and magnesium and packed cell volume in blood in Cow 68 infused twice intramuscularly with 50 mg EDTA/kg bodyweight on 4 Aug. (●) and 8 Oct. (○). Delivery on 14 Oct. (x).

Table 13. Intramuscular injection of EDTA pre partum and plasma calcium of Cow 68.

Day of injection	Dose of EDTA (mg/kg bodyweight)	Plasma Ca (mg/100 ml)			Time for plasma Ca to reach minimum (h)
		before injection or parturition	lowest level after injection or parturition	difference	
0	50	9.5	6.4	3.1	7
65	50	10.8	7.7	3.1	8
71	parturition	9.6	9.0	0.6	24

test, hardly any physiological decline in plasma calcium was seen.

From the various results of the experiments in Section 3.5, the conclusion can be drawn that the decline in plasma calcium induced by an injection of EDTA will be less if the cow had received a calcium-lowering injection of EDTA a short time before. This effect was not seen when the time-lag between the two injections was greater than 10 days.

In two cows, hardly any decline in plasma calcium was seen after parturition when the cow received an injection of EDTA less than a week pre partum.

3.6 Influence of EDTA infusions on incidence of milk fever in cows prone to milk fever

3.6.1 Introduction

The literature and experiments described in Section 3.5 show that EDTA infusion causes a decrease in blood calcium. A physiological or an (experimentally) induced decline in calcium will activate the calcium-regulating mechanism. This will lead to a higher absorption from intestines or a higher mobilization of calcium from bone.

Because of the slower adaptation of bone mobilization, intestinal absorption will be particularly stimulated. This intestinal absorption will be influenced by the daily intake of calcium with food. I therefore infused a solution of EDTA into 3 cows fed on different amounts of calcium (Table 14).

3.6.2 Results and discussion

The changes in blood minerals are given in Figs. 11, 12, 13 and Table 15. EDTA can lower plasma calcium in cows prone to milk fever (Figs. 11, 12, 13). As plasma calcium decreased plasma magnesium increased. This increase in magnesium was only possible when sufficient magnesium was given daily in the diet. With a subnormal supply of magnesium as often happens also in the winter period in the Netherlands both low calcium and low magnesium are seen in the blood of parturient cows.

Table 14. Daily intake of the minerals Ca, Mg and P (g) and theoretical absorption coefficient when cow had no bone resorption. The daily requirement for these cows is 30 g Ca (ARC, 1965; NRC, 1966).

Cow	Concentrate	Intake (g)			Absorption coefficient (%)
		Ca	Mg	P	
Corry	Ca-poor, P-rich	61.4	44.2	188.0	50
Bontje 76	commercial mixture	103.0	38.2	65.0	29
Nelly 34	Ca-rich, P-normal	192.6	46.6	66.0	16

Table 15. Post partal decline in plasma calcium in 3 cows after infusion of EDTA some days before calving.

Cow	Time pre partum of last infusion (days)	Ca intake (g/day)	Decline in plasma Ca (mg/100 ml)
Corrie	5	61.4	8.7 − 8.6 = 0.1
Bontje 76	14	103.0	10.2 − 6.9 = 3.3
Nelly 34	3	192.6	10.2 − 3.4 = 6.8

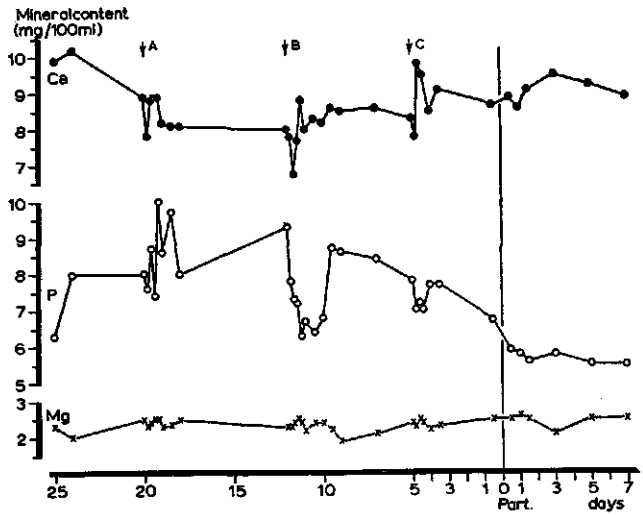

Fig. 11. Concentration of calcium, phosphorus and magnesium in plasma of Cow Corrie. With EDTA intravenously (↓) A: 50 mg/kg bodyweight in ½ h; B: 80 mg/kg bodyweight in 1 h; C: 50 mg/kg bodyweight in ½ h. Daily intake (g) in dry period Ca 61.4, P 188.0, Mg 44.2.

The speed with which the blood calcium recovered was better in Corrie than in Bontje 76 and Nelly 34. From results with Nelly 34, we may conclude that infusions of EDTA even 3 days pre partum gave no residual activation of calcium metabolism post partum; this can be ascribed to the strong inactivation of metabolism by a high-calcium diet. With Bontje 76 too, infusion of EDTA 14 days pre partum had no residual effect post partum. On a low-calcium diet (Corrie), the frequently induced experimental hypocalcaemia also activated calcium metabolism. No inactivation of calcium metabolism was seen when the cow calved 5 days after the last infusion. Although we have results from only 3 cows prone to milk fever, their results and those in Table 12 suggest that the activation or inactivation of calcium absorption, which is the main input for older dry cows, can occur in a minimum of 3 days.

These findings are supported by a field trial carried out in the winter 1970/1971 with cows prone to milk fever infused twice with EDTA within 14 days in the second half of the dry period. The cows prone to milk fever received 50 mg EDTA/kg bodyweight by intramuscular injection for the first time (about 4 weeks before parturition). Two weeks before the expected date of calving an intravenous injection was given of 50 mg EDTA/kg bodyweight, infused over 15 min. If there was milk fever at delivery the farmer informed the local veterinary surgeon and asked for milk fever treatment. Although no exact analysis of the diets were made, it can be taken that the daily calcium intake was over 100 g. In this field trial, I observed 28 cows prone to milk fever, EDTA was injected into 15 cows and 13 cows were not treated (Table 16).

A quarter of the total number of cows got milk fever, a normal proportion in cows

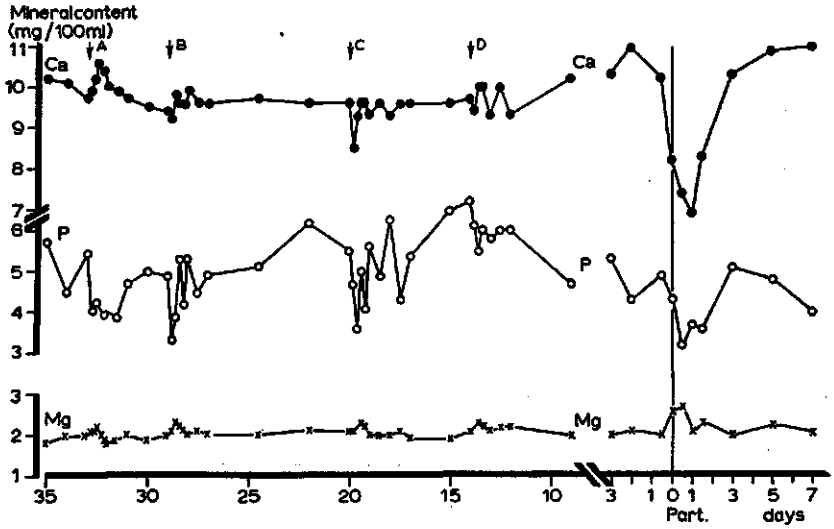

Fig. 12. Concentration of calcium, phosphorus and magnesium in plasma of Cow Bontje 76. With EDTA intravenously (↓) A & D: 50 mg/kg bodyweight in ¼ h; B & C: 80 mg/kg bodyweight in ¼ h. Daily intake (g) in dry period Ca 103.0, P 65.0, Mg 38.2.

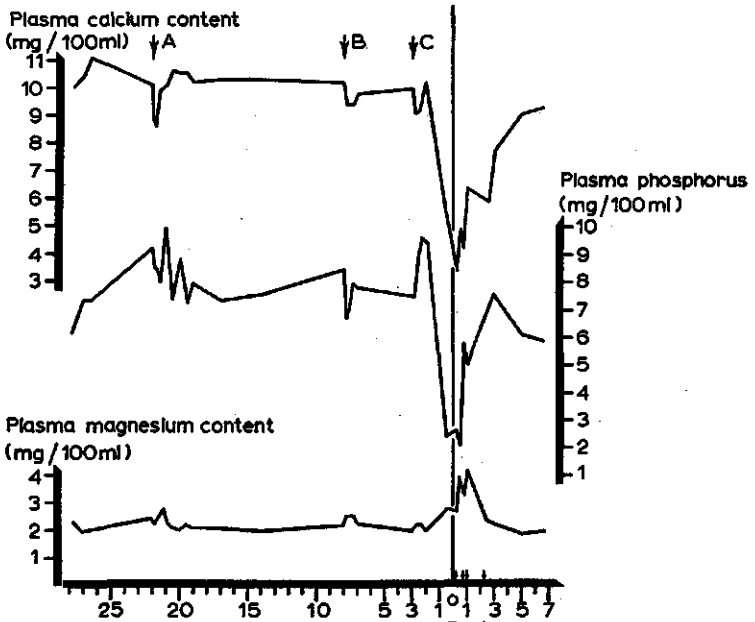

Fig. 13. Concentration of calcium, phosphorus and magnesium in plasma of Cow Nelly 34. With EDTA intravenously (↓) A, B & C: 50 mg/kg bodyweight in ¼ h. Daily intake (g) in dry period Ca 192.6, P 66.0, Mg 46.6. After calving, milk fever appeared (↑), blood samples just before treatment.

Table 16. Effect of two infusions of EDTA pre partum on incidence of milk fever in a field trial.

	Controls	EDTA injected	Total
Total number	13	15	28
Milk fever	1	6 (= 40%)	7
No milk fever	12	9 (= 60%)	21

prone to milk fever (Kemperman, 1971).

The 2 injections of EDTA, intramuscularly or intravenously, 4 weeks and 2 weeks pre partum did not prevent parturient hypocalcaemia in local farm conditions.

Further development of this method was not continued because of the constant influence of the dietary calcium, which always interferes with prevention of parturient hypocalcaemia by EDTA infusion.

3.7 Effect on calcium metabolism of changes in amounts of calcium and phosphorus in the diet of pregnant dry cows

3.7.1 Introduction

In older cows, the input of calcium into the immediately available pool comes mainly from intestinal absorption (Ramberg et al., 1967). Calcium metabolism can be activated by increasing the output (EDTA infusions) or by diminishing the input of calcium (fasting). Complete fasting is a rather drastic measure. 'Fasting' was therefore restricted to calcium only. Pregnant dry cows were fed on a calcium-rich and normal-phosphorus or a calcium-poor and phosphorus-rich diet changing weekly. Four cows were given rations alternately rich and poor in calcium for weekly intervals. Samples from blood, faeces and urine were taken twice a week. The diet, blood plasma, urine and faeces were analysed for calcium, phosphorus and magnesium contents (Tables 17, 18, 19, 20; Fig. 14).

Just after calving, all the 4 cows were fed on the diet that is usual on the Hoorn Experimental Farm. Cow 60 calved during a phosphorus-rich period; the other three in a calcium-rich period. Analyses of blood plasma are given in Table 21.

Table 17. Data on the cows.

Cow No	Bodyweight (kg)	Age (years)	Expected calving date	Real calving date
60	700	8	5–10	28–9
59	552	11	30– 9	21–9
39	547	5	30– 9	20–9
93	570	6	30– 9	23–9

Table 18. Composition of the dietary components.

	Dry matter (%)	% in the dry matter	
		Ca	P
Grass hay	83.2	0.54	0.31
Ca-rich concentrate	87.0	0.87	0.60
P-rich concentrate	85.5	0.35	2.06

Table 19. Average daily intakes of dry matter, calcium and phosphorus.

	Dry matter (kg)	Ca (g)	P (g)	Ca:P
P-rich period				
14 kg hay	11.6	62.9	36.1	
2 kg concentrates	1.7	6.0	35.2	
Total	13.3	68.9	71.3	0.97
Ca-rich period				
14 kg hay	11.6	62.9	36.1	
2 kg concentrates	1.7	15.1	10.4	
240 g $CaCO_3$	0.2	97.4	–	
Total	13.5	175.4	46.5	3.77

Table 20. Average packed cell volume (%), concentration of calcium, phosphorus and magnesium in blood plasma, urine and faeces of the four dairy cows.

Trial day	Diet	p.c.v.	In blood plasma (mg/100 ml)			In urine		Content in dry matter of faeces (%)	
			Ca	P	Mg	Ca (mg/100 ml)	P (g/litre)	Ca	P
0	normal	32.9	10.4	5.5	2.3				
2	Ca-rich	32.1	10.8	8.8	2.1	12.6	0.30	1.86	0.86
4	Ca-rich	31.6	10.6	8.0	2.0	20.4	0.14	–	–
7	P-rich	32.0	10.3	8.3	2.1	23.1	0.50	1.49	1.71
11	P-rich	32.6	9.8	7.3	2.1	16.0	0.59	–	–
14	Ca-rich	31.9	10.4	6.4	1.9	24.4	0.07	3.65	0.79
18	Ca-rich	31.9	10.1	6.9	2.0	17.0	0.17	–	–
21	P-rich	32.9	9.5	6.7	2.2	6.2	1.35	1.49	1.49
25	P-rich	32.6	9.8	7.3	2.0	7.6	0.99	–	–
28	Ca-rich	31.3	10.5	6.6	1.9	28.6	0.10	4.06	0.76

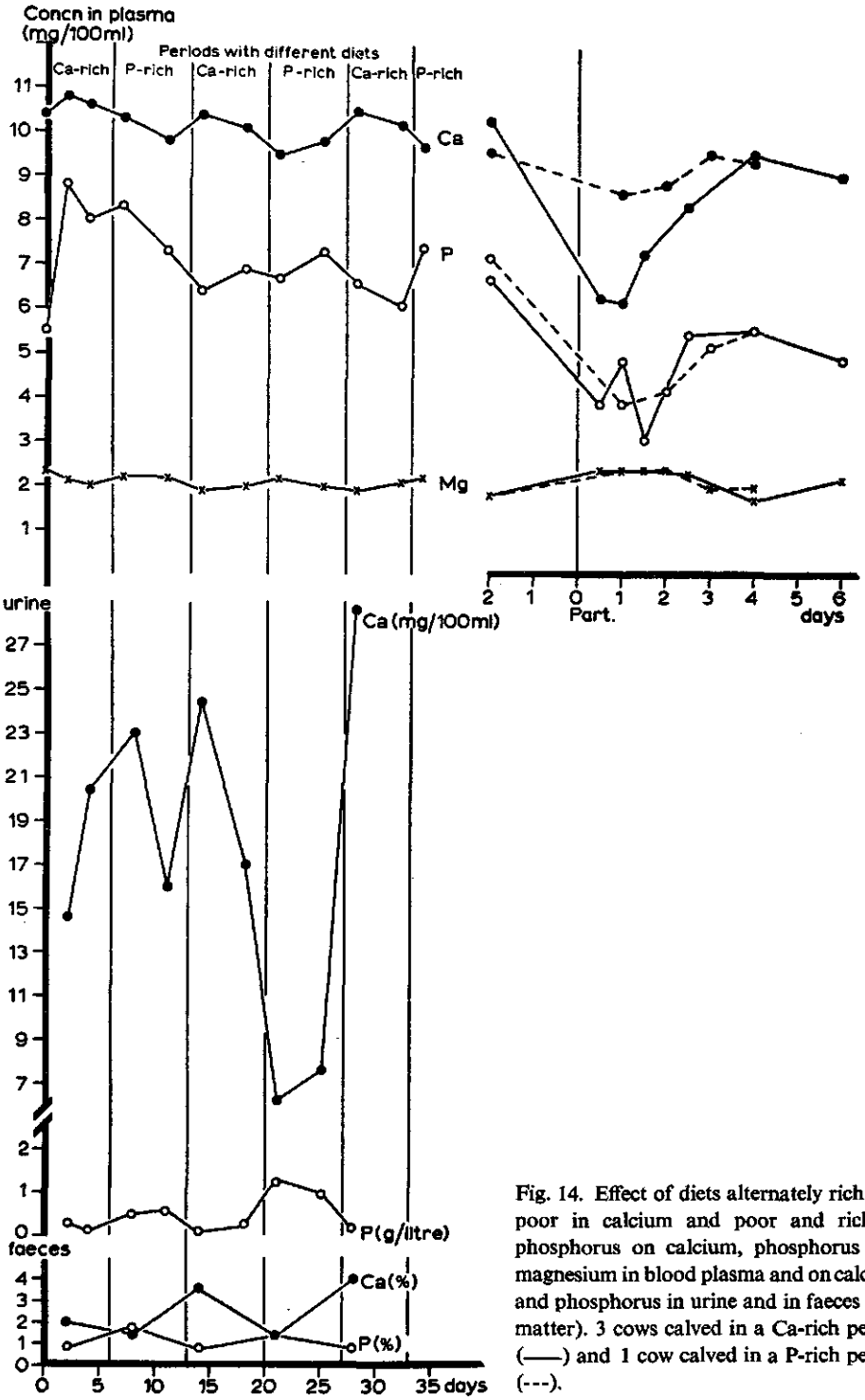

Fig. 14. Effect of diets alternately rich and poor in calcium and poor and rich in phosphorus on calcium, phosphorus and magnesium in blood plasma and on calcium and phosphorus in urine and in faeces (dry matter). 3 cows calved in a Ca-rich period (——) and 1 cow calved in a P-rich period (---).

Table 21. Packed cell volume and concentration of calcium, phosphorus and magnesium in blood plasma. Results are grouped for Cow 60 calving in a phosphorus-rich period and for the others calving in a calcium-rich period.

Day	p.c.v. (%)	In plasma (mg/100 ml)		
		Ca	P	Mg
Cow 60				
1–2 pre partum	29.1	9.7	7.2	1.8
1 post partum	32.6	8.7	3.9	2.3
2 post partum	30.2	8.9	4.1	2.3
3 post partum	30.0	9.6	5.2	2.0
4 post partum	28.7	9.4	5.8	1.9
Average of Cows 39, 93 and 59				
1–2 pre partum	31.7	10.3	6.7	1.8
½ post partum	30.4	6.3	3.9	2.4
1 post partum	28.8	6.2	4.9	2.3
1½ post partum	33.5	7.3	4.1	2.3
2½ post partum	34.7	8.4	5.5	2.2
4 post partum	34.7	9.6	5.6	1.7
5 post partum	36.0	9.1	4.9	2.2

3.7.2 Results and discussion

A slightly higher plasma content of calcium was seen during the period with a diet rich in calcium and normal in phosphorus and a slightly lower during the period with the other diet. Plasma phosphorus was high during all 4 – 5 weeks of this experiment, although some variation was present. Three of the four cows calved during or immediately after the calcium-rich diet and calcium in plasma was markedly low but no paresis occurred: plasma calcium declined from 10.3 to 6.3 (12 h post partum) and 10.3 to 6.2 mg/100 ml (24 h post partum). Values fully recovered 3 or 4 days post partum. One cow calved after a feeding period of 5 days on the calcium-poor diet. The post partal plasma calcium decline from 9.7 to 8.7 mg/100 ml (24 h post partum) was negligible in comparison with the three cows mentioned earlier. The decline in postpartal plasma phosphorus was the same in all four cows.

Although Cow 93 calved one day after a change from the Ca-rich to the Ca-poor diet, her postpartal decline in plasma calcium was the same as that of Cows 59 and 39 which calved during the calcium-rich period. This may be taken as an indication that the adaptation of the calcium metabolism takes more than a single day.

This conclusion is in agreement with the theory that the production of calcium-binding protein in the intestinal wall and the increase of the intestinal absorption of calcium (Ramberg, 1972) takes some time. This protein seems to be responsible for the transport of calcium through that wall and its production is induced by parathyroid hormone (Wasserman & Taylor, 1969).

The excretion of calcium and phosphorus into the urine is an interesting facet. In the last complete period (Days 21 – 27) when a phosphorus-rich diet was given, the excretion of phosphorus was much higher than in the second period (Days 7 – 13). But excretion of calcium in the last period was less than in the second. Perhaps this phenomenon is a result of a greater production of parathyroid hormone and a higher adaptivity of the calcium homeostasis.

There is some indication of a negative correlation between plasma calcium and magnesium in Figure 14.

3.8 Cows prone to milk fever and fed on a diet poor in calcium in the dry period but rich in calcium around parturition

3.8.1 Introduction

A diet rich in phosphorus and poor in calcium seems to stimulate calcium input either by higher absorption from intestine or higher mobilization from bone (Section 3.7).

Attempts were made to activate the adaptability of the calcium homeostasis in cows prone to milk fever (which were again borrowed from local farmers) by feeding them on a phosphorus-rich and calcium-poor diet for 4 weeks of the dry period. Then the last week before delivery, the cows of the experimental group were fed in the same way as cows of the control group in order to replenish the possibly exhausted pool of immediately available calcium and to take advantage of the possibly activated calcium absorption from the intestine.

The cows arrived at the experimental farm from November till May and were divided into two equal groups according to their history of milk fever and age (Table 22). Data on the diets used during the first 4 weeks (Period I) of the experiment and during the last week (Period II) of the dry period are given in Table 23. The change in feed composition after the 4 weeks was accentuated by a single oral dose of 250 g $CaCO_3$ mixed with water by bottle. The experimental cows were fed like the control cows during the last week before parturition. The amount of concentrates was steamed up from 6 to 8 – 10 kg per day in the last days before calving. Thus at parturition, the daily intakes of calcium and phosphorus were, respectively, 119.6 – 137.0 and 71.0 –

Table 22. Data on the cows prone to milk fever.

	Experimental	Control
Number of cows	14	13
Average bodyweight (kg)	648	658
Average age (years)	9.1	8.7
Average past frequence of milk fever	2.2	2.5

Table 23. Average daily intakes of feedstuffs and nutrients.

	Four weeks of the dry period		One week before parturition
	control	experimental	control and experimental group
Grass hay (kg)	10	10	10
Concentrates (kg)	4	4	6 (8 – 10)
dm (kg)	12.4	12.4	14.2
SE (g)	5014	5014	7326
dcp (g)	1410	1410	1670
Ca (g)	84.8	66.8	102.2 (119.6 – 137.0)
P (g)	49.0	97.8	60.0 (71.0 – 82.0)
Mg (g)	30.6	29.4	37.4
Ca:P	1.7	0.7	1.7

82.0 g for both groups. Within 12 h of delivery, the cows were milked in order to stimulate calcium output.

3.8.2 Results and discussion

Results of this experiment are given in Table 24. Several blood samples were taken and analysed; average values per group are given in Table 25.

During Period I, higher levels of plasma phosphorus were found in the experimental group. This might be due to the higher daily intake of phosphorus. Although large amounts of phosphorus were supplied, plasma phosphorus tended to decrease during Period I. An adaptation to a high phosphorus intake may play a role. That adaptation (lower absorption or higher excretion in urine) seems to be slow and may take 4–5 weeks.

Plasma magnesium levels were normal with the high-phosphorus diet (Period I), just before parturition (Period II) and after parturition. No decline or increase in plasma magnesium was seen near parturition: a sufficient supply of magnesium was guaranteed by the diet.

When the diet of the experimental group was changed about a week before delivery, there was an unexpected increase in plasma calcium, probably from the single oral

Table 24. Results of both groups.

	Experimental	Control
Number of cows	14	13
Incidence of milk fever	7	7
Number of cows with plasma Ca $\leqslant$ 7.5 mg/100 ml after parturition	13	12

Table 25. Average packed cell volume (%) and plasma concentration of calcium, phosphorus and magnesium (mg/100 ml).

		p.c.v.		Ca		P		Mg	
		contr.	exp.	contr.	exp.	contr.	exp.	contr.	exp.
Preliminary period	start	31.5	32.5	10.0	10.2	5.8	6.3	2.2	2.2
Period I	5th week	29.9	32.0	10.2	10.8	6.4	7.5	2.2	1.9
	4th week	30.9	31.4	10.2	10.1	5.7	8.0	2.2	2.1
	3rd week	31.5	31.6	10.1	9.9	6.2	7.6	2.1	2.1
	2nd week	31.3	32.0	10.1	9.8	5.8	7.0	2.1	2.1
Period II	1st week	31.7	32.6	10.2	11.0	5.9	6.3	2.0	2.0
	0 – 2 days	31.8	34.4	9.2	9.2	5.4	5.2	2.0	2.2
Post partum	12 h	33.7	35.1	6.9	7.6	4.3	3.9	2.2	2.2
	24 h	32.0	33.9	6.5	7.1	3.8	3.4	2.1	2.2
	36 h	31.7	33.1	6.9	6.5	3.6	3.0	2.2	2.2
	3–4 days	31.0	31.8	8.4	8.2	4.9	4.0	1.9	2.2
	5–7 days	30.8	31.6	9.4	8.8	5.5	5.3	1.9	2.0

dose of $CaCO_3$. A blood sample was taken at 16 h 00. Immediately after this sampling, 250 g $CaCO_3$, dissolved in water, was given by bottle. Another blood sample was taken at 8 h 00 next morning. The results are given in Table 26.

Eight cows were given that excessive amount of calcium; four cows were not.

The analysis of variance (Tables 27, 28) gives grounds for concluding that within an unreliability of 5%:

- An oral administration of $CaCO_3$ gives a higher average increase in plasma calcium than without $CaCO_3$.
- The upper limit of the one-sided 95% confidence interval for the increase in plasma calcium due to an addition of $CaCO_3$ in the cows without milk fever is 1.465. In effect, in the circumstances of the experiment and with cows like ours (prone to milk fever) and with the experimental treatment described above, followed by an oral addition of $CaCO_3$ the chance is 95% or more that a cow with an increase of at least 1.5 mg Ca/100 ml plasma gets milk fever. Also it is evident that a cow with an increase in plasma calcium less than 1.5 mg Ca/100 ml after the $CaCO_3$ treatment is very unlikely to get paretic parturient hypocalcaemia.

Because of the adaptability of calcium homeostasis, the time from the change in diet and oral administration of $CaCO_3$ to delivery might be important for the postpartal decline in plasma calcium (Table 29). Within 3 days of the dietary change, 3 of the 14 cows had a decline in plasma calcium to 7.5 mg/100 ml or less and 2 cows had a decline to a minimum above 7.5 mg/100 ml. There is obviously an indication that the time between the dietary change and the delivery influences the decline in plasma calcium post partum.

The probably activated adaptability of the cows' calcium metabolism by a diet

Table 26. Changes of calcium in blood plasma of cows prone to milk fever around the switch from a calcium-poor phosphorus-rich diet to one rich in calcium and normal in phosphorus with or without oral administration of $CaCO_3$ at the time of the switch a week pre partum.

	Ca in plasma (mg/100 ml)		
	before switch	after switch	difference
With 250 g $CaCO_3$			
No milk fever			
Ant 90	10.00	10.28	+ 0.28
Thade 12	11.08	11.10	+ 0.02
Dirkje 83	9.25	9.60	+ 0.35
Marie 40	9.05	11.30	+ 2.24
Average			+ 0.73
With milk fever			
Elly 20	9.10	12.15	+ 3.05
Jonker 31	10.75	12.45	+ 1.70
Sijtje 13	9.60	12.02	+ 2.42
Schilder 41	7.20	10.70	+ 3.50
Average			+ 2.67
Without 250 g $CaCO_3$			
No milk fever			
Juliana 66	9.86	9.80	− 0.06
Tini 6	9.81	10.11	+ 0.30
Average			+ 0.12
With milk fever			
Kuiper 78	9.52	10.25	+ 0.73
Schilder 51	9.92	10.62	+ 0.70
Average			+ 0.72

Table 27. Analysis of variance on data from Table 26.

Cause of variation	Degrees of freedom	Sum of squares	Mean square	F and significance	
$CaCO_3$ or no $CaCO_3$	1	4.3606		6.89*	
Milk fever or no milkfever	1	6.6901	4.0871	10.57*	6.46*
Interaction $CaCO_3$ x milk fever	1	1.2105		1.91	
Residual variance	8	5.0632	0.6329		

* Significant at $P \leqslant 0.05$.

Table 28. Average increase in plasma calcium (mg/100 ml) of the twelve experimental cows after the moment of dietary change 1 week pre partum.

Cows at parturition	With $CaCO_3$	No $CaCO_3$	Average	
With milk fever	2.668	0.715	2.017	difference 1.494
No milk fever	0.725	0.120	0.523	
Average	1.696	0.418	1.270	

Table 29. Association of different times (days) from dietary switch to calving with severe or slight decline in plasma calcium post partum.

	Days										
	0	½	1½	2	3	4	9	10	11	15	17
11 cows with plasma Ca $\leqslant$ 7.5 (mg/100 ml)				1	2	2	2		2	1	1
3 cows with plasma Ca $>$ 7.5 (mg/100 ml)		1			1		1				

poor to normal in calcium and rich in phosphorus can be disturbed by feeding for about a week on a diet rich in calcium and normal in phosphorus. This results in a deep decline in plasma calcium after parturition as happened in the control group (Table 24).

3.9 Diets rich or poor in calcium pre partum to cows not prone to milk fever

3.9.1 Introduction

Sections 3.3, 3.4, 3.7 and 3.8 suggest a relation of calcium intake some weeks before parturition with the incidence of milk fever as well as with the severity of the decline in plasma calcium after parturition (Table 30).

Figure 15 shows that an increasing daily intake of calcium is correlated with an increasing percentage of cases with a decline in plasma calcium to below 7.5 mg/100 ml. To prove the relationship older dairy cows with no history of milk fever were fed on two diets. Barley straw was given as roughage together with a concentrate with cottonseed, barley and milocorn and without a mineral mixture. Each cow received 50 g MgO (30 g Mg) daily so that magnesium supply was sufficient.

This trial was started in May 1970 when the cows were in the last third of their lactation. All cows, from May onwards, were fed according to standards, except for those of Group I for calcium. The calcium standards in the Netherlands are 0.40 – 0.45% and 0.60% in the dry matter of the ration for cows with an annual yield of up to, and above 6000 kg milk, respectively.

Group I (5 dairy cows with no history of milk fever) received 0.17% calcium in the

Table 30. Daily intake of calcium and phosphorus, incidence of milk fever, and decline in plasma calcium post partum in the control and experimental groups in trials described in Sections 3.3, 3.4, 3.7 and 3.8.

Section		Number of cows	Daily intake (g)		Ca:P in diet	Cows with milk fever		Cows with a plasma Ca decline ⩽ 7.5 mg/100 ml	
			Ca	P		absolute	relative (%)	absolute	relative (%)
3.2	contr.	14	80.6	48.0	1.7	2	15	8	57
	exp.	16	77.3	71.6	1.1	4	25	7	44
3.4	contr.	16	82.6	51.0	1.6	5	31	8	50
	exp.	17	53.1	53.5	1.0	9	53	13	77
3.7	contr.	3	175.4	46.5	3.8	0	0	3	100
	exp.	1	68.9	71.3	1.0	0	0	0	0
3.8	contr.	13	102.2	60.0	1.7	7	53	12	92
	exp., fed like control one week pre partum	14	102.2	60.0	1.7	7	50	13	93

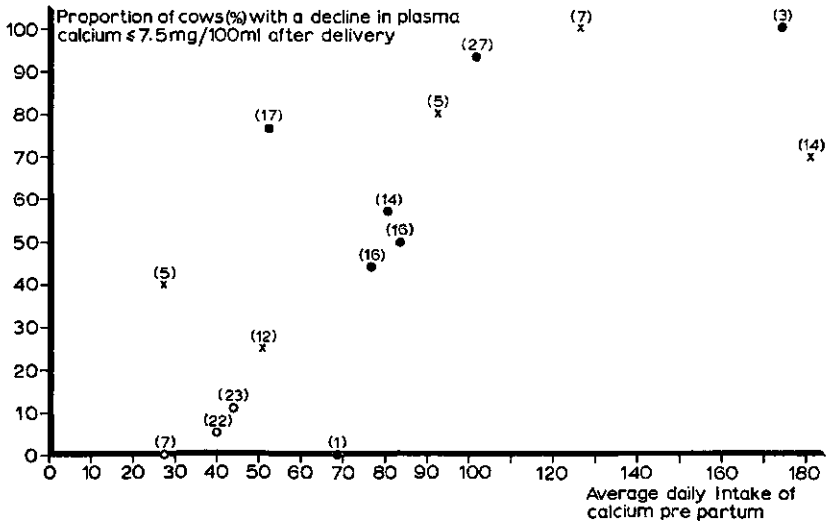

Fig. 15. The relation of prepartal intake of calcium (g/day) and percentage of cows prone to milk fever with a plasma calcium decline $\leqslant$ 7.5 mg/100 ml after calving. In round brackets are numbers of cows in the groups. The results of trials still to be described have been added. Results from Sections 3.3, 3.4, 3.7 and 3.8 (●), from 3.9 and 3.10 (x), and from 3.11 and 3.12 (○).

Table 31. Intakes of dietary components and of some minerals.

	Week 1 + 2		Week 3, 4, 5, 6		Week 7, 8 and the first week post partum	
	group I	group II	group I	group II	group I	group II
Straw (kg)	7	7	7	7	7	7
Concentrates (kg)	4	4	7	7	9	9
$CaCO_3$ (g)	–	250	–	250	–	250
Ca (g)	19.6	119.6	23.8	123.8	26.6	126.6
P (g)	16.0	16.0	39.6	39.6	42.4	42.4

dry matter of the ration during 3 months of lactation, 2 months dry period and the first week post partum.

Group II (7 dairy cows with no history of milk fever) received 0.80% calcium in the dry matter of the ration and again during the last 3 months of lactation, 2 months dry period and the first week post partum. The diet during the first week post partum was the same as during the last two weeks pre partum. The diet during the 8 weeks of the dry period and the daily intake of minerals are stated in Table 31.

Our cows with a normal milk production (up to 6000 kg milk a year) needed 0.35 – 0.40% phosphorus in the dry matter of the ration (standards as given by the Committee on Mineral Nutrition, 1973). An intake of 16.0 g phosphorus means

0.17% phosphorus in the dry matter of the ration. Only for the first 14 days of the dry period did the cows receive this phosphorus-poor diet. For the last 2 weeks pre partum the cows received 42.4 g phosphorus (0.30% P in dry matter).

3.9.2 Results and discussion

The results of this trial and of blood analysis around parturition are given in Table 32 and 33.

It is striking that all of the cows in Group II (0.80% Ca in dry matter of the diet) compared with 2 cows in Group I (0.17% Ca in dry matter of the diet) had a decline in plasma calcium to below 7.5 mg/100 ml. In the cows of Group II, which were not prone to milk fever, a considerable decline in plasma calcium after parturition can obviously be induced by giving an excess of calcium pre partum. Moreover, the decline in Group II is more severe and of longer duration than in Group I, although the daily intake of calcium pre and post partum was greater in Group II (Table 34; Fig. 16).

The recovery from hypocalcaemia started sooner in Group I (at 36 h post partum) than in Group II. We may conclude that the adaptability of the calcium homeostasis is on average better in the cows of Group I than of Group II. In the lactation period from May till September, neither milk fever symptoms occurred nor broken legs were seen and that milk production remained at a good level in spite of low daily intakes of calcium.

At the time of the decline in plasma calcium after delivery, an increase in plasma magnesium was seen, equal in the two groups. It should be remembered that the daily intake of magnesium was more than enough. Although the supply of phosphorus was

Table 32. Plasma minerals (mg/100 ml) of Groups I and II.

	Group I			Group II		
	Ca	P	Mg	Ca	P	Mg
1–2 weeks pre partum	10.0	6.4	2.34	9.9	7.1	2.48
parturition	–	–	–	–	–	–
12 h post partum	8.9	6.3	3.19	8.6	5.0	3.10
24 h post partum	8.3	5.6	3.29	7.5	4.4	3.35
36 h post partum	8.4	5.7	3.28	7.3	4.1	3.37

Table 33. Incidence of milk fever and hypocalcaemia.

	Group I	Group II
Number of cows	5	7
Milk fever	1	0
Cows with plasma Ca $\leqslant$ 7.5 mg/100 ml	2	7

Table 34. Average change from normal values 1–2 weeks pre partum to the values post partum of calcium, phosphorus and magnesium in blood plasma (mg/100 ml) in Group I and II.

	Group I			Group II		
	Ca	P	Mg	Ca	P	Mg
12 h post partum	− 1.1	− 0.1	+ 0.85	− 1.3	− 2.1	+ 0.62
24 h post partum	− 1.7	− 0.8	+ 0.95	− 2.4	− 2.7	+ 0.87
36 h post partum	− 1.6	− 0.7	+ 0.94	− 2.6	− 3.0	+ 0.89

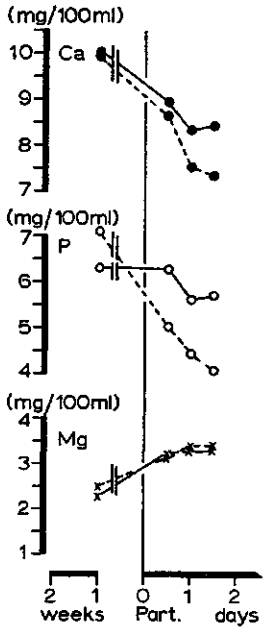

Fig. 16. Average concentration of minerals in plasma of Group I fed on a calcium-poor diet (——) and Group II fed on a calcium-rich diet (----).

marginal, the decline in plasma phosphorus in Group II was more severe than in Group I. This suggests a more adequate adaptation mechanism for calcium as well as for phosphorus in Group I.

3.10 Diets poor in calcium and rich in phosphorus during the last month of pregnancy and the first week of lactation in cows prone to milk fever

3.10.1 Introduction

Because hopeful results are seen in Section 3.9 with the cows not prone to milk fever fed with 0.17% calcium and 0.30% phosphorus in dry matter, it is necessary to investigate also a calcium-poor and phosphorus-rich diet or a calcium-poor and phos-

phorus-normal diet for cows prone to milk fever. The calcium content of the diet seemed more important in preventing paretic hypocalcaemia than the phosphorus content or the Ca:P ratio of the diet. To prove this theory, cows prone to milk fever were fed on a diet poor in calcium and rich in phosphorus and with calcium and phosphorus equal or even more phosphorus than calcium, as Boda & Cole (1955) did. Observations were not so much on incidence of milk fever as on the decline in plasma calcium after parturition.

The cows in this trial were older than 6 years with a high annual milk production and with a history of milk fever (Table 35). The control group (14 cows) received a commonly used diet (hay, straw and concentrates). Experimental Group I (12 cows) received the same dietary ingredients but the diet was poor in calcium and rich in phosphorus. Experimental Group II (5 cows) received the same diet as Experimental

Table 35. Summary of the data and the results from the trial with different amounts of calcium and phosphorus in diets for cows prone to milk fever.

	Control	Exp. I	Exp. II
Number of cows	14	12	5
Average bodyweight (kg)	648	668	588
Average past frequency of milk fever	2.0	2.1	1.8
Average age (years)	8.0	8.5	8.0
Average milk prod. 7 days post partum (kg/day)	28.4	29.8	27.0
Incidence of milk fever	4	1	1
Cows with plasma Ca $\leqslant$ 7.5 mg/100 ml	10 (71 %)	3 (25 %)	4 (80 %)

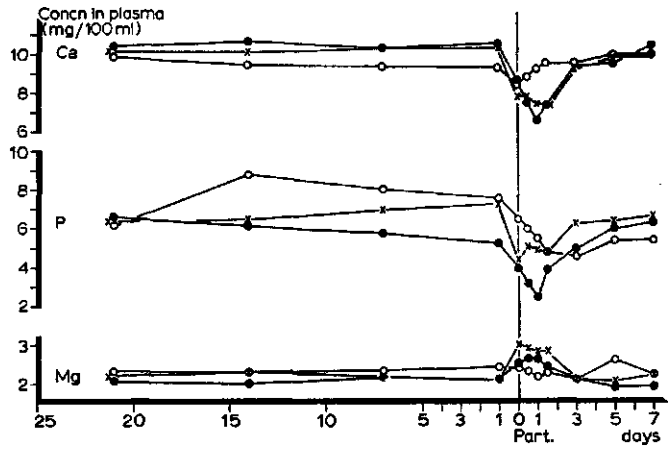

Fig. 17. Average concentration of minerals in plasma before and after parturition in cows with different amounts of calcium and phosphorus in the diet. Control group (x). Experimental Group I (○) and Experimental Group II (●).

Table 36. Average daily intakes of minerals (g/day) and average concentrations in blood plasma (mg/100 ml).

	Daily intake (g)			Concn in plasma (mg/100 ml)			
	Ca	P	Mg	Ca	P	Mg	
Start							
contr.	84.2	51.8	30.9	10.1	6.3	2.2	
exp. I	84.2	51.9	31.2	9.9	6.2	2.3	
exp. II	84.4	51.6	30.6	10.4	6.5	2.1	
14 d pre partum							
contr.	165.4	51.3	42.2		10.0	6.4	2.3
exp. I	43.8	137.5	35.9	9.4	8.9	2.3	
exp. II	84.4	51.6	30.6	10.6	6.3	2.0	
7 d pre partum							
contr.	181.2	58.2	43.4	10.2	6.9	2.2	
exp. I	51.2	153.3	39.1	9.3	8.0	2.3	
exp. II	93.7	58.3	34.4	10.2	5.7	2.2	
2 d pre partum							
contr.				10.2	7.2	2.1	
exp. I				9.2	7.5	2.4	
exp. II				10.3	5.2	2.1	
0 d[1]							
contr.	93.0	22.2	28.8	7.7	4.2	3.0	
exp. I	25.4	76.4	19.6	8.3	6.3	2.4	
exp. II	46.9	29.2	17.2	8.4	2.9	2.5	
½ d post partum							
contr.				7.6	5.0	2.9	
exp. I				8.7	5.9	2.3	
exp. II				7.4	3.1	2.6	
1 d post partum							
contr.	185.4	42.3	58.5	7.3	4.8	2.8	
exp. I	176.2	58.2	42.3	9.1	5.4	2.2	
exp. II	93.7	58.3	34.4	6.5	2.3	2.6	
1½ d post partum							
contr.				7.1	4.6	2.8	
exp. I				9.4	4.7	2.3	
exp. II				7.2	3.8	2.4	
3 d post partum							
contr.	247.0	78.1	55.8	9.1	6.2	2.1	
exp. I	247.5	84.6	56.2	9.3	4.5	2.1	
exp. II	103.0	65.0	38.2	9.2	4.9	2.1	
5 d post partum							
contr.				9.7	6.3	2.0	
exp. I				9.8	5.3	2.6	
exp. II				9.4	5.9	1.9	
7 d post partum							
contr.	276.7	87.5	68.2	9.7	6.5	2.2	
exp. I	282.3	90.4	66.8	9.8	5.3	2.2	
exp. II	112.3	71.7	42.0	10.3	6.2	1.9	

1. Assumption: intake half a diet.

Group I but with somewhat more calcium and less (normal) phosphorus. The three diets were given for about 4 weeks before parturition. One week before parturition, the cows received 1 kg extra concentrates of their particular type (extra) to 'steam' them up. After delivery, Group I was fed exactly like the control group and received thus more calcium. Group II received the same diet as before parturition. Daily intake of minerals and mineral content in plasma at different times are given in Table 36 and Fig. 17.

3.10.2 Results and discussion

The daily calcium intake of Group I was greater after than before parturition (Table 36). In sensitive cows with a low dietary intake of calcium, blood calcium will increase within 12 – 16 h after a single oral dose of $CaCO_3$. After a feeding period that activates calcium metabolism (low Ca intake), the just calved cows were therefore fed on a calcium-rich diet. Cows with good appetite after parturition showed hardly any 'physiological' calcium decline. Of the cows prone to milk fever, of experimental group I a quarter (25%) had bad appetite and did show a marked decline in plasma calcium. In the control group given excess of calcium pre and post partum, 71% of the cows showed a decline in plasma calcium. Group II fed with somewhat less calcium than the control group reacted like the controls. Table 35 gives the data and the final results of the groups. The χ square test of the difference between the control group and Group I in the proportion of cows with plasma Ca $\leqslant$ 7.5 mg/100 ml is just significant at $P = 0.05$.

In Table 37, a theoretical absorption coefficient is given, for which it was assumed

Table 37. Theoretical absorption coefficient for a daily calcium requirement of 30 g for a 600-kg dry pregnant dairy cow.

		Ca intake (g/day)	Absorption coeff. (%)	Input to Ca pool (g/day)
start	control	84.2	35.6	30
	Group I	84.2	35.6	30
	Group II	84.4	35.6	30
14 d pre partum	control	165.4	18.1	30
	Group I	43.8	67.1	30
	Group II	84.4	35.6	30
7 d pre partum	control	181.2	16.6	30
	Group I	51.2	58.6	30
	Group II	93.7	32.0	30
1 d post partum	control	185.4	16.6	30
	Group I	176.2	58.6	103
	Group II	93.7	32.0	30

that all the input of the immediately available calcium pool comes by absorption from the intestine and is sufficient to keep plasma calcium in the normal range. A 600-kg dry pregnant dairy cow needs 30 g Ca/day according to ARC (1965) and NRC (1966).

As we have seen in the rapid diet change in Section 3.7 and 3.8 and in experiments of Ramberg et al. (1970b) and Ramberg (1972) an adaptation of the absorption coefficient or the absorption ability in the intestinal wall took some time (2 – 3 days). Perhaps the hormonally induced production of calcium-binding protein, which takes some time, may play an important role. It is reasonable to believe that just before parturition the absorption coefficient is the same as 7 days pre partum because the diet is not changed and the total need is hardly changed. The input of calcium into the immediately available pool on Day 1 post partum must therefore be 30, 103 and 30 g for the cows of the control group, Groups I and II, respectively, because only the intake of calcium in Group I is increased from 51.2 to 176.2 g daily.

Our good results (Table 35) in preventing a decline in plasma calcium post partum might be explained by a sudden increase in total calcium absorption due to a quick increase in calcium intake and an absorption coefficient which is fixed at about 60% for 2 – 3 days.

3.11 Diets poor in calcium in the dry period and rich in calcium just after delivery in older pregnant dairy cows with no history of milk fever

3.11.1 Introduction

It is necessary to pay attention to two questions:

1. Is it necessary during the dry period and after parturition that phosphorus intake be high as well as calcium intake low in order to prevent milk fever as suggested by Boda & Cole (1954)?
2. Can intestinal calcium absorption give a better contribution to the big demand for calcium at the onset of lactation than Mayer (1970) and Ramberg (1972) indicate?

During the summer of 1971, 8 Dutch Friesian pregnant milking cows without a history of milk fever and on the average 7 years old were fed indoors on a winter ration. The diet was poor in calcium and normal in phosphorus during the last 3 months of lactation (Table 38) and the last 2 months of pregnancy (dry period) (Table 39). Immediately after parturition, the cows received a single oral dose of 250 g $CaCO_3$ and calcium-rich and phosphorus-normal concentrates. Only one cow did not receive the dose of $CaCO_3$ immediately after delivery. This cow will not contribute to the average data given in Table 40 and Fig. 18, but its data are given separately.

3.11.2 Results and discussion

On average the cows gave 16.1 and 18.3 kg colostrum on Days 1 and 2 post partum, respectively. None of the eight cows had a plasma calcium decline below 7.5 mg/100

Table 38. Data on cows with no history of milk fever, diet and daily intake during the last part of the lactation period.

	15/4	4/5	18/5	1/6	15/6	29/6	14/7	28/7	11/8	25/8
Number of cows in milk	8	8	8	8	8	8	7	7	7	2
Number of cows dry	0	0	0	0	0	0	1	1	1	6
Average bodyweight (kg)	530	534	534	544	554	565	579	586	591	596
Average daily milk (kg)	18.1	15.9	14.8	14.2	13.5	12.2	12.7	11.6	10.9	11.6
Daily ration (kg)										
straw (wheat (W), barley (B))	5 (B)	6 (B)	6 (B)	6 (B)	6 (B)	6 (B)	6 (B)	6 (B)	6 (B)	6 (W)
potatoes	10	10	10	10	10	10	10	10	10	10
concentrates	6	7.1	7.7	8.4	6.1	6.7	7.1	7.3	7.6	7.6
Daily intake										
dm (kg)	11.3	13.6	14.0	15.1	13.3	13.8	14.2	13.7	14.7	15.0
SE (g)	6666	7997	8431	8937	7274	7708	8597	8142	8359	7035
dcp (g)	1002	1195	1277	1373	1058	1140	1195	1222	1263	1233
Ca (g)	42.2	50.6	51.8	51.2	45.4	46.5	48.6	51.0	49.6	25.6
P (g)	30.7	36.7	39.0	41.8	35.8	38.5	38.5	39.4	41.7	41.7
Mg (g)	22.3	26.5	28.4	30.7	23.3	25.2	28.2	27.9	29.9	29.9
Ca:P	1.3	1.3	1.3	1.2	1.3	1.2	1.3	1.3	1.2	0.6
Content in dry matter (%)										
Ca	0.38	0.37	0.37	0.34	0.34	0.34	0.34	0.37	0.34	0.17
P	0.27	0.27	0.28	0.28	0.27	0.28	0.27	0.29	0.29	0.28
Mg	0.20	0.19	0.20	0.20	0.18	0.18	0.20	0.20	0.20	0.20

Table 39. Average daily intake and diet composition of 8 pregnant dry dairy cows, with no history of milk fever, during the dry period and after delivery.

	Weeks of dry period			Days post partum		
	1 – 4	5 – 6	7 – 8	0	1 – 3	4 – 7
Average bodyweight (kg)	590	602	605		–	–
Daily ration (kg)						
straw wheat	6	6	6		6	6
potatoes	10	10	10		10	10
concentrates	2	4.5	7		7	9
Daily intake						
dm (kg)	9.1	11.3	13.5		10.7	12.6
SE (g)	3986	5794	7601		7601	9047
dcp (g)	466	809	1151		1151	1425
Ca (g)	14.2	26.9	28.5	100[1]	142.9	179.1
P (g)	17.6	28.0	35.9		34.0	41.6
Mg (g)	11.4	20.9	28.1		34.5	43.1
Ca:P	0.8	1.0	0.8		4.2	4.3
Content of minerals in dry matter (%)						
Ca	0.16	0.24	0.21		1.34	1.42
P	0.19	0.25	0.27		0.32	0.33
Mg	0.13	0.18	0.21		0.32	0.34

1. Given as an oral dose of $CaCO_3$.

ml and no milk fever occurred.

Cows with a milk production around 4000 to 4500 kg a year need 0.40 – 0.45% calcium and 0.35 – 0.40% phosphorus in dry matter. In this trial the cows received about 0.34% calcium and 0.28% phosphorus in dry matter from May till the end of August; the average daily milk production was 18.1 and 11.6 kg in May and in August, respectively. From May till parturition in October or November, the cows were far below the recommended standards for calcium and phosphorus. Plasma magnesium was on a normal level so intake seems to have been sufficient. This means that the cow's calcium and phosphorus metabolism had to work hard because no cow had low plasma calcium or phosphorus. For calcium, this would result in a higher intestinal absorption and a higher removal from bone.

To avoid any effect from bad appetite after delivery, the cows were given orally $CaCO_3$ with a bottle just after delivery to take advantage of their activated calcium metabolism (intestinal absorption). In addition, they were given a diet with 1.34 and 1.42% calcium in dry matter 1 – 3 and 4 – 7 days post partum, respectively, instead of 0.20% Ca pre partum.

Remarkable is the difference of average post partal plasma calcium between the 7 cows and the cow which did not receive $CaCO_3$ immediately after delivery and which had a bad appetite for some reason. This phenomenon probably means that the in-

testinal absorption of calcium is important during the first 2 – 3 days post partum in preventing parturient hypocalcaemia.

In this group of 7 parturient cows, the lowest plasma calcium was observed in one cow 12 h post partum and was estimated as 8.5 mg/100 ml.

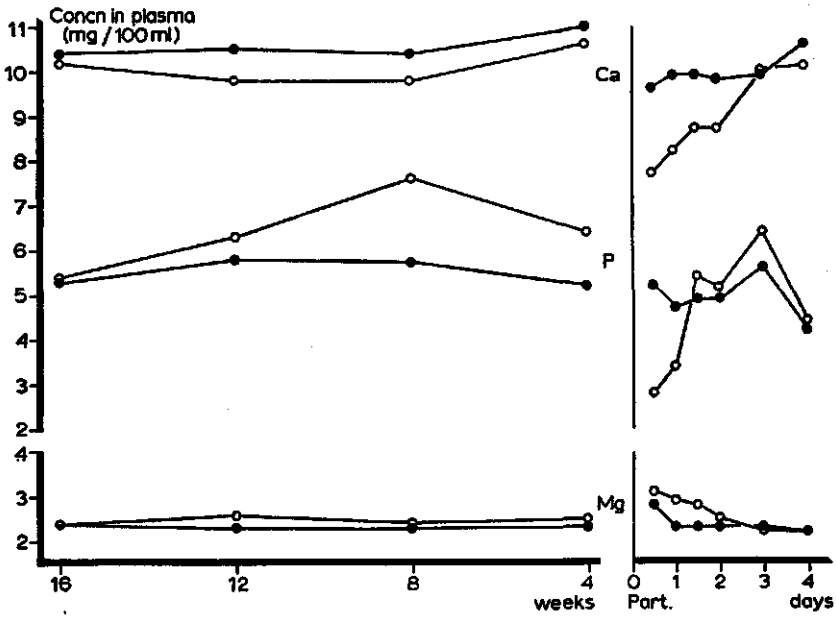

Fig. 18. Average concentration of plasma minerals of cows on a calcium-poor diet pre partum and a calcium-rich diet post partum. Just after calving, 7 cows received an oral dose of $CaCO_3$ (●) and 1 cow did not (○).

Table 40. Mineral content in blood plasma (mg/100 ml) averaged for 7 cows given a dose of 250 g $CaCO_3$ at delivery and of 1 cow without the dose.

	Weeks pre partum				Days post partum					
	16	12	8	4	½	1	1½	2	3	5
7 cows										
Ca	10.4	10.5	10.4	11.0	9.6	9.9	9.9	9.8	9.9	10.6
P	5.3	5.8	5.8	5.2	5.2	4.7	4.9	4.9	5.6	4.2
Mg	2.37	2.33	2.30	2.27	2.80	2.31	2.26	2.26	2.30	2.15
1 cow										
Ca	10.1	9.8	9.8	10.6	7.7	8.2	8.7	8.7	10.0	10.1
P	5.4	6.3	7.6	6.4	2.8	3.4	5.9	5.2	6.4	4.4
Mg	2.38	2.58	2.36	2.48	3.09	2.93	2.84	2.45	2.16	2.15

3.12 Cows prone to milk fever on a diet preventing milk fever and with or without a magnesium supplement

3.12.1 Introduction

The trials described in Sections 3.7, 3.8, 3.9, 3.10 and 3.11 suggest that parturient hypocalcaemia can be prevented by two dietary measures:

– In the dry period, the diet has to be as poor as possible in calcium. The daily requirement of calcium of a pregnant dry cow (550 kg bodyweight) is about 30 g (ARC, 1965; NRC, 1966). A 'steaming-up' ration in the last days of pregnancy has no effect on the induction of parturient hypocalcaemia provided that daily intake of calcium is kept as low as possible. This feeding measure has to be taken because a calcium-poor diet pre partum (a daily intake of calcium of about 30 instead of 100 – 120 g) enlarges the absorptive capacity of the intestinal wall as well as stimulating the bone turnover rate or resorption (Ramberg, 1972). With the onset of lactation, this enlarged absorptive capacity must be given a chance to absorb the required amount of calcium.

– Therefore immediately after delivery, the diet has to be changed from calcium-poor to calcium-rich. Because the reaction time for adaptation in the absorptive system of the intestines is about 2 – 3 days, a high dietary intake of calcium immediately after delivery will result in an increased input of calcium into the immediately available pool; in the first 3 days post partum plasma calcium will not fluctuate more than in the dry period, whereas usually there is a severe 'physiological' decline.

After delivery when plasma calcium declines, plasma magnesium increases under certain dietary conditions (Section 3.6, Fig. 11, 12, 13; Section 3.7, Fig. 14; Section 3.9, Fig. 16; Section 3.10, Table 36). Perhaps within certain limits of plasma concentration, magnesium ions can take over the action of calcium ions. However for a good action of a numerous enzymes and of the parathyroid hormone a normal plasma magnesium is required too. Because cows prone to milk fever are the most sensitive cows to parturient hypocalcaemia, further studies with these cows were made on the dietary measures used previously but with or without a supplement of magnesium.

Two groups of cows prone to milk fever were used to study magnesium supplementation to the diet. The cows of Group I received no dietary supplement and the cows of Group II received about 22 g magnesium daily extra.

Table 41. Data of cows of Group I and Group II.

	Group I	Group II
Number of cows	22	23
Average age (years)	9.2	9.0
Average bodyweight (kg)	651	643
Average past frequency of milk fever	2.0	2.0

Table 42. Content of nutrients in hay and concentrates (g/kg).

	dm	SE	dcp	Ca	P	Mg
Hay	780	249	57	3.1	1.9	1.3
Concentrate						
Ca-poor	872	723	137	1.9	3.5	2.1
Ca-rich	872	723	137	17.2	3.4	3.1
Mg-rich	872	723	137	4.6	3.6	22.3

This trial lasted 5 weeks for each cow (4 weeks pre partum and 1 week post partum). The cows were borrowed as usual from local dairy farmers and were rather prone to milk fever. Data on the cows in the groups are given in Table 41.

Except for the difference in magnesium intake, the cows received the same prepartal diet, which seemed to prevent parturient hypocalcaemia. This prepartal diet was calcium-poor. During the last week of pregnancy, cows were 'steamed up' with an extra 2 kg hay and 1 kg of concentrate daily. Immediately after parturition was complete, the cow was checked for deglutitions (swallowing movements) before being given a single oral dose of 250 g $CaCO_3$. The check avoided the cow swallowing the wrong way. This oral dose given by bottle was used because some cows after delivery had poor appetite. But as soon as the cow had a normal appetite, a calcium-rich diet could be taken, the diet being changed from calcium-poor to calcium-rich immediately after delivery.

Except for calcium, the ingredients of the ration and of the concentrate were kept the same. To make sure the calcium output was as large as possible, the parturient cows were completely milked soon after delivery.

Nutrients in the hay and the three types of concentrate used are given in Table 42. The prepartal and postpartal diet and the daily intake of nutrients and minerals are given in Table 43.

3.12.2 Results and discussion

Table 44 shows the absence of milk fever and the number of cows with a decline in plasma calcium more than 7.5 mg/100 ml or to 7.5 mg/100 ml or less.

In this trial, the feeding measures gave good results in preventing severe hypocalcaemia post partum. Average blood values for the groups are contained in Table 49. There seems to be a relation between the decline in plasma calcium and the increase in plasma magnesium, as suggested in the introduction to this section (3.12.1). Since any value between 9 and 12 mg/100 ml could be due to normal fluctuation, values below 9 were taken as a criterion of a true decline in plasma calcium in seeking a relation with an increase in magnesium (Table 45).

In Group II, the decline in plasma calcium was more severe than in Group I.

Table 43. Average daily intakes of dietary components, nutrients and minerals by cows of Group I and II.

	Weeks pre partum				Days post partum			
	4 – 2		1		0 – 3		5 – 7	
	I	II	I	II	I	II	I	II
Hay (kg)	7	7	9	9	9	9	9	9
Concentrates (kg)[1]	6 (A)	5 (A) 1 (C)	7 (A)	6 (A) 1 (C)	7 (B)	6 (B) 1 (C)	10 (B)	9 (B) 1 (C)
dm (kg)	10.7	10.7	13.1	13.1	13.1	13.1	15.0	15.0
SE (g)	6081	6081	7302	7302	7302	7302	9222	9222
dcp (g)	1221	1221	1472	1472	1472	1472	1826	1826
Ca (g)	33.1	35.8	41.2	43.9	148.3	135.7	196.8	184.2
P (g)	34.3	34.4	41.6	41.7	40.9	41.1	49.2	50.4
Mg (g)	21.7	41.9	26.4	46.6	33.4	52.6	41.4	60.6

1. Concentrate A was calcium-poor; Concentrate B was calcium-rich; C was magnesium-rich.

Table 44. Results of the trial with diets normal (Group I) or rich (Group II) in magnesium.

	Group I	Group II	Both groups
Number of cows	22	23	45
Cows with milk fever	0	0	0
Cows with plasma Ca > 7.5 mg/100 ml	21	20	41
Cows with plasma Ca ⩽ 7.5 mg/100 ml	1 (4.6%)	3 (11.0%)	4 (8.9%)
Daily milk production on Day 7 post partum (kg)	27.8	25.7	26.7

Table 45. Average plasma calcium and magnesium (mg/100 ml) of 10 cows of Group I (magnesium-poor diet) and 13 cows of Group II (magnesium-rich diet) with a postpartal decline in plasma calcium to below 9.0 mg/100 ml.

	Group I		Group II	
	Ca	Mg	Ca	Mg
Weeks pre partum				
5	9.9	1.87	10.2	2.08
4	9.8	2.03	10.2	2.25
3	9.9	2.09	10.1	2.26
2	9.9	2.09	9.9	2.27
1	9.6	2.17	9.8	2.16
2 Days pre partum	9.3	2.07	9.5	2.36
Days post partum				
½	9.3	2.16	8.6	2.65
1	9.0	2.12	8.3	2.62
1½	8.9	2.25	8.4	2.54
2	8.8	2.28	8.6	2.64
3	9.1	2.14	9.2	2.21
5	9.3	1.96	9.5	2.12
7	9.4	1.99	9.5	2.15

Although on Day 2 post partum plasma calcium was 8.8 mg/100 ml in group I, plasma magnesium had hardly increased but certainly had in Group II.

Dietary magnesium in calving cows can increase plasma magnesium at the moment that a decline in plasma calcium is detected just after parturition. As a support for this trial, I reexamined data from cows in previous trials (Section 3.3 – 3.11).

Cows prone to milk fever were regrouped into those on magnesium-rich diets (Table 46) and those with hardly sufficient dietary magnesium to keep plasma magnesium above the normal limit of 2.0 mg/100 ml (Table 47; Fig. 19).

In this material too, the increase in plasma magnesium was greater when the cows received a magnesium supplement (Tables 46 and 47). The χ square test for the

Table 46. Plasma calcium and magnesium (mg/100 ml) in cows prone to milk fever and fed with a supplement of magnesium. The 15 cows were not treated despite severely low plasma calcium. All recovered spontaneously.

	Cow No																Average	
	1		2		3		4		5		6		7		8		Av. of 7 other cows		Ca	Mg
	Ca	Mg	Ca	Mg	Ca	Mg	Ca	Mg	Ca	Mg	Ca	Mg	Ca	Mg	Ca	Mg	Ca	Mg		
Weeks pre partum																				
5									11.3	2.1	10.3	2.1			9.7	1.8			10.4	2.0
4									11.2	2.2	10.2	2.1			10.0	1.9			10.5	2.1
3	9.3	2.1	9.6	2.1	10.4	2.1	10.5	2.0	11.3	2.1	10.5	2.0			10.0	2.0			10.2	2.1
2	10.0	2.1	10.4	2.0	10.4	2.1	10.4	2.3	10.0	2.3	9.9	2.1	9.5	–	10.5	2.1	10.7	–	10.2	2.1
1	9.5	2.1	9.7	1.9	10.1	2.1	10.4	2.1	9.7	2.3	10.0	2.1	9.8	2.6	10.3	2.3	9.9	2.5	9.9	2.2
Days pre partum																				
2	8.8	1.9	10.4	1.3	9.8	2.3	10.8	2.2							10.2	2.1			10.0	2.0
Days post partum																				
½	8.1	2.3	6.9	2.0	8.5	2.6	6.8	3.1	6.1	3.7	5.7	2.4	7.1	3.5	6.6	2.7	8.6	3.1	7.2	2.8
1	7.4	2.1	5.9	2.3	7.0	2.2	6.6	2.7	6.8	4.1	4.6	2.3	5.5	4.0	5.7	2.6	7.5	3.4	6.3	2.9
1½	6.3	1.9	6.4	2.5	6.4	2.1	8.0	2.7	8.1	3.5	5.2	2.7	5.1	4.2	5.8	2.5	7.3	3.4	6.5	2.8
2									9.0	2.5										
3	7.5	1.5	9.2	1.9	8.5	2.6	10.8	2.0	9.5	1.3	8.3	2.2			7.7	2.4			8.8	2.0
5	9.1	1.8	9.5	1.7	9.7	2.2	10.1	2.1	9.4	1.5	9.5	1.9			11.3	1.9			9.8	1.9
7	8.8	2.1	9.0	2.1	9.3	2.2	10.6	2.1	9.7	1.8									9.5	2.1

Table 47. Plasma calcium and magnesium (mg/100 ml) in cows prone to milk fever and fed with hardly sufficient dietary magnesium. The 8 cows were not treated despite severely low plasma calcium. All recovered spontaneously.

	Cow No														Average	
	1		2		3		4		5		6		7		8		Ca	Mg
	Ca	Mg	Ca	Mg	Ca	Mg	Ca	Mg	Ca	Mg	Ca	Mg	Ca	Mg	Ca	Mg		
Weeks pre partum																		
5					9.9	2.0	10.0	2.1									10.0	2.0
4					9.3	1.9	10.3	2.1									9.8	2.0
3	8.1	2.3	9.9	2.2	9.4	1.9	9.8	2.0									9.3	2.1
2	8.7	2.2	10.5	2.2	9.6	2.0	9.3	2.1	9.8	1.9	10.5	2.0	10.1	2.3	10.2	2.1	9.8	2.1
1	8.0	2.1	10.4	2.4	10.2	2.2	10.8	1.8	9.8	1.9	10.5	2.0	10.1	2.3	9.6	2.0	9.9	2.1
Days pre partum																		
2	8.9	2.3	10.3	2.2	9.7	2.3	9.4	1.9									9.6	2.2
Days post partum																		
½	6.2	2.2	9.0	2.4	7.5	2.1	6.8	1.9							5.9	2.1	7.1	2.1
1	7.1	2.1	7.4	2.3	5.9	2.0	6.4	1.8	5.9	2.3	5.4	2.1	6.0	2.0	5.0	1.7	6.1	2.0
1½	7.7	2.2	6.6	2.3	5.4	1.9	6.0	2.0							5.5	1.6	6.2	2.0
2					6.2	1.9											6.2	1.9
3	8.1	2.2	10.0	2.1					5.5	1.7	8.3	2.1	8.8	1.8	9.3	1.8	8.3	2.0
5	8.7	2.2			8.0	2.0	9.0	1.5									8.6	1.9
7	9.3	2.3	9.3	2.0					9.2	2.0	9.7	2.0	9.3	2.1			9.4	2.1

Table 48. Average packed cell volume (%) and plasma minerals (mg/100 ml) in 71 cows prone to milk fever which formed 4 different control groups in 4 years.

	Week(s) pre partum				Days pre partum	Day(s) post partum					
	4	3	2	1	2	0	½	1	1½	3	5	7
p.c.v.	31.7	31.7	31.6	32.2	33.1	–	34.4	33.0	32.4	31.6	31.2	32.1
Ca	10.3	10.2	10.2	10.5	9.5	7.7	7.3	7.2	6.8	8.7	9.3	9.5
P	6.1	6.2	6.1	6.3	6.0	4.2	4.4	4.0	3.7	5.2	5.7	5.5
Mg	2.1	2.1	2.1	2.1	2.1	3.0	2.4	2.2	2.4	2.0	2.0	2.0

Table 49. Packed cell volume (%) and concentration of minerals in blood plasma (mg/100 ml) of Group I and Group II of Section 3.12 compared with the 'aggregate' control group (III).

	p.c.v.			Ca			P			Mg		
	I	II	III	I	II	III	I	II	III	I	II	III
5 Weeks	32.5	32.6		10.1	10.2		6.1	5.7		1.92	2.08	
4	31.2	31.8	31.7	9.9	10.3	10.4	6.3	5.6	6.1	2.13	2.17	2.1
3	32.1	31.6	31.7	10.1	10.2	10.2	6.3	6.0	6.2	2.11	2.25	2.1
2	32.4	31.4	31.6	10.2	10.1	10.2	6.5	5.7	6.1	2.19	2.25	2.1
1	32.8	30.9	32.2	10.0	10.0	10.5	6.1	5.9	6.3	2.15	2.18	2.1
2 Days	31.4	31.7	33.1	9.8	9.9	9.5	6.5	5.9	6.0	2.17	2.29	2.1
Parturition			–			7.7			4.2			3.0
½ Day(s)	34.9	33.1	34.4	9.9	9.4	7.3	4.9	4.8	4.4	2.17	2.48	2.4
1	32.7	31.8	33.0	9.5	9.0	7.2	4.4	4.5	4.0	2.04	2.38	2.2
1½	33.5	31.6	32.4	9.5	9.0	6.8	4.5	4.8	3.7	2.13	2.38	2.4
2	32.8	31.2		9.5	9.2		5.1	5.1		2.15	2.44	
3	32.7	30.8	31.6	9.6	9.5	8.7	5.3	5.2	5.2	2.03	2.14	2.0
5	31.5	30.8	31.2	9.7	9.7	9.3	4.9	4.9	5.7	1.88	2.12	2.0
7	31.0	30.0	31.2	9.8	9.7	9.5	4.8	4.8	5.5	1.94	2.10	2.0

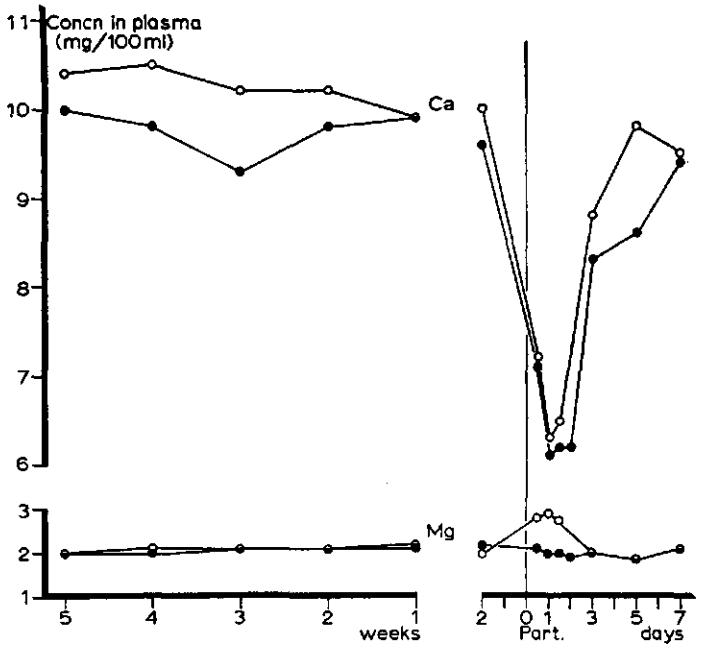

Fig. 19. Average calcium and magnesium concentration in plasma of cows prone to milk fever. One group fed on a magnesium-poor diet (●), the other on a magnesium-rich diet (○) and with no milk fever.

difference in the proportion of cows with an increase or a decline in plasma magnesium is significant at $0.01 < P < 0.025$. The average decline in plasma calcium post partum in cows listed in Tables 46 and 47 are about the same. This means that in practice when low plasma calcium and magnesium are seen together in parturient paretic cows, the diet needs a supplement of magnesium. Not only for proper working of parathyroid hormone but also for prevention of milk fever with tetanic symptoms attention must be given to the daily magnesium supply.

In the 4 winter trials (Sections 3.3, 3.4, 3.8, 3.10), 71 cows prone to milk fever were borrowed from local dairy farmers and fed on a diet commonly used in the Netherlands. These cows formed the four control groups in different years. Some cows were used in a control group more than once but not in the same year. There were no operative restrictions in using plasma contents of these cows sampled in different winter trials in constructing an aggregate control group from the 4 control groups of 14, 16, 27 and 14 cows in the years 1967/1968, 1968/1969, 1969/1970 and 1970/1971, respectively.

Average packed cell volume and plasma calcium, phosphorus and magnesium of these 71 cows are listed in Table 48.

The 'aggregate' control group can be used as a control for Groups I and II of winter trial 1971/1972 (Section 3.12). We have listed the results (plasma concentrations etc.) with the plasma content as given in Table 48 of the 'aggregate' group (Table 49).

The results of Table 44 and Table 49 showed clearly that our feeding measures can contribute to the prevention of parturient hypocalcaemia and therefore also to the prevention of parturient paresis.

4 General discussion and recommendations

4.1 General discussion

Parturient hypocalcaemia is a derangement of the calcium homeostasis of the parturient cow. So far, scientists have accepted a 'physiological' plasma calcium decline post partum as 'normal' in parturient cows. The decline appears also in the 'aggregate' control group (Section 3.12). The small fluctuation in plasma calcium in non-parturient dairy cows casts doubts whether this decline post partum is indeed a normal phenomenon (Section 3.12).

After delivery, a sudden calcium output with colostrum cannot be compensated in dairy cows by normal dairy feeding practice. Immediately after delivery, an increase in calcium input into the immediately available pool is difficult to achieve because calcium mobilization from bone starts 14 – 20 days later and the increase in absorption from the intestine needs about 3 days (Ramberg et al., 1970b). Therefore there is usually a decline in plasma calcium. But in lactating cows (7 – 8 years old) on a calcium-poor diet, up to 70% of the calcium intake seems to be absorbed (van Leeuwen, draft). Feeding cows on calcium-poor diets pre partum considerably increases bone calcium turn-over rate over those with calcium-rich diets (Ramberg, 1972). This means that the cow's ability to maintain calcium homeostasis soon after parturition can be activated by a calcium-poor diet before parturition.

Indeed, in my trials over the years 1967 to 1972, rations could be composed in such a way that the 'physiological' plasma decline did not on average occur.

After delivery, high levels of parathyroid hormone in plasma are found (Mayer, 1970). For good function, parathyroid hormone needs magnesium ions (Estep et al., 1969; Muldowney et al., 1970). Therefore our feeding measures to prevent parturient hypocalcaemia are based on three main principles:

1. A calcium-poor diet pre partum activates calcium metabolism, and therefore increases bone turn-over rate and absorption of calcium from the intestine.
2. By offering the cows sufficient calcium just after delivery when a decline in plasma calcium is imminent (during the first 3 days), calcium homeostasis has a good chance of being maintained.
3. Because magnesium is needed for good working of parathyroid hormone, plasma magnesium has to be kept at a normal level ($\geqslant$ 2.0 mg/100 ml). Thus sufficient attention should be paid to the magnesium supply of the cow near parturition.

To prevent ketosis, steaming-up rations are usually prescribed. But this kind of ra-

tion can also induce milk fever if rich in calcium (Section 3.8). A steaming-up ration low in calcium did not cause a decline in plasma calcium after delivery (Section 3.12).

In practice, cows with milk fever sometimes have tetanic symptoms as in grass tetany. With a diet balanced for magnesium, the cow does not develop hypomagnesaemia just after delivery and has the benefit of an optimum function of the parathyroid hormone to prevent parturient hypocalcaemia.

4.2 Recommendations

This research was done with the aid of many Dutch dairy farmers who lent us their most valuable dairy cows. The results of our work are also of great importance to these farmers who have for years experienced the symptoms and problems of milk fever. I will translate our results into a number of practical recommendations.

We concentrated attention to the feeding of the dry cows for 4 weeks before and one week after parturition because the speed of adaptability of the intestinal absorption of calcium is about 3 days and the adaptability of calcium mobilization from bone about 21 days.

Usually in the same 4 weeks before parturition a 'steaming-up' system of feeding is used to accustom the cows to the high feeding regime necessary for a good start of milk production.

Diet before parturition The net calcium needs of a pregnant dry cow (550 kg bodyweight) are rather low (about 17 g daily). With an apparent absorption coefficient of 70%, as found at our Institute in 8-year-old milking cows (van Leeuwen, draft), this means a daily intake with the diet of 24 g calcium.

Every gram of calcium given above the needs has a depressing effect on the absorption coefficient. Because the need for calcium is rather high after parturition, a high apparent absorption coefficient of calcium in the intestine and also an activated calcium metabolism of bone is necessary to keep the input and output of calcium in balance.

Because diets low in calcium stimulate both mechanisms, it is highly advisable to feed the pregnant dry cow on a diet as poor in calcium as possible.

In the Netherlands, roughage (grass hay and grass silage) often contains 6 – 8 g calcium per kg. so that 3–4 kg grass hay will cover the calcium needs of a pregnant dry cow. The dry matter composition of prepartal diets commonly used is 10 kg grass hay or silage and 1 – 2 kg concentrates. Hence the calcium content of the concentrates has to be as low as possible and the calcium content of the roughage may decline from 0.60 – 0.80% to about 0.30%.

The supply to grassland of magnesium-containing fertilizers has a depressing effect on the calcium content of the grass and its products. Till now, this effect has been viewed as alarming (Committee on Mineral Nutrition, 1973, p. 22). Because the cow can raise her absorptive ability as high as 70% of the total calcium intake, lowering of the calcium content of herbage is a point for discussion.

The dietary magnesium supply before and after parturition in dairy cows has to be made safe. A possible interaction with the absorption of calcium after addition of 30 g magnesium (1 kg antitetany cake) daily for 4 weeks before and 1 week after parturition does not promote the frequency of parturient hypocalcaemia (Section 3.12). Because low levels of plasma magnesium must be prevented in connection with tetanic symptoms in paretic cows and because magnesium is necessary for a good working of parathyroid hormone, a daily addition of 30 g magnesium to the rest of the diet of the pregnant dry cow and the just calved cow is necessary in my opinion.

Concentrates without any addition of minerals and minor elements except magnesium and vitamins can be used in the dry period. The composition of the concentrates has to be in such a manner that calcium-rich products like pulp are not used.

Parturition time Because a calving cow is too occupied with calving, she will not take up any food, so I make no recommendations for feeding a cow during calving.

Diet after parturition After parturition, the colostral calcium output is high and the demand for calcium upon the immediately available calcium pool is high too. The needs for maintenance and pregnancy are about 17 g/day. With a production of 15 kg colostrum, the total net needs are increased to about 47 g/day (by 175%).

To increase the input of calcium into the blood, it is necessary to increase the oral intake of calcium after parturition. With a need of 50 g calcium daily and an absorption coefficient of 50%, a sudden uptake of about 100 g calcium can supply sufficient the immediately available calcium pool.

Soon after delivery (about 12 to 24 h), a decline in plasma calcium is usual as demonstrated in the 'aggregate' control group (Section 3.12.1). It is therefore necessary to give the cow an oral dose of calcium as soon as possible after calving. We used a single dose of 250 g $CaCO_3$ suspended in water and gave it from a bottle.

A cow with paretic symptoms cannot swallow. Therefore, attention has to be paid to the swallow reflex before the treatment with 250 g $CaCO_3$. This can easily be done by letting the cow drink or by giving her a bit of hay or concentrates.

After that, the diet of the cow must be calcium-rich (1.00% Ca in the dry matter). Because Dutch concentrates for dairy cows have a calcium content of 0.80 – 1.00% in dry matter, these concentrates can be used after parturition.

Substitutes of roughage and concentrates Before parturition, only products poor in calcium can be used, for instance:

potatoes
products of potatoes
tulip bulbs
brewer's grain
milocorn
cottonseed products.

After parturition, calcium-rich products have to be used, for example:

- sugar-beet pulp
- sugar-beet leaves and heads (silage)
- products of clover and lucerne
- turnips and Brussels sprouts
- calcium salts in a mineral mixture

Housing Plasma calcium values in the 'aggregate' control group (Section 3.12) indicate that the average cow becomes normal again 3 days post partum.

To avoid injury, it is advisable to give the cow a place where she has no chance of hurting herself for at least 3 days post partum. For example, the lying bed must be soft, but the floor must be rough and the dunging passage must be covered over.

Summary

In the Netherlands, milk fever in dairy cows is still a problem. Besides financial losses, the inconvenience must not be underestimated for the dairy farmer, who has to give his attention to many cows, nowadays. Milk fever in dairy cows, mostly happening within a couple of days post partum, is preceded and accompanied by a severe decline in plasma calcium.

Prevention of a decline in plasma calcium and reduction of milk fever incidence is the subject of the research. Generally, cows prone to milk fever borrowed from local farmers were used in the study. The cows arrived about 4 weeks pre partum and were returned 1 week after parturition.

In the first trial (3.3) the cows were divided into a control and an experimental group. A ration of hay and concentrates, meeting energy and protein requirements, with a Ca:P ratio of 1:7 was fed to the cows of the control group. Cows of the experimental group were fed on the same diet, but with a Ca:P ratio of 1.1. The ration for 7 days was given in 6 days, so the experimental cows had a day's fast. The diet and the way of feeding of the cows of the experimental group did not contribute to the prevention of low plasma levels of calcium and of milk fever cases after calving (Tables 2, 3 and 4). The frequency of milk fever cases were 25 and 15% for the experimental and control group, respectively. In one cow (pregnant and dry), the same depressing effect on plasma concentration of calcium was seen, as well on a diet low in calcium, as with complete fasting.

In the next study (3.4), cows of the experimental group in contrast to the control group, both group containing cows with a milk fever history, were fed on a ration of hay and concentrate, 20% below energy and protein requirements and with a Ca:P ratio of 1.0. Milk fever incidence was 53 and 31% in the experimental and control group, respectively.

The effect of experimentally induced plasma decline of calcium on the activation of calcium metabolism was studied in cows fed on hay and concentrate ration (3.5). A significant decline in plasma calcium, induced by repeated EDTA infusions, activated calcium metabolism. After calving, the decline in plasma calcium was minimal if the last induced decline in plasma calcium was established 3 – 10 days before calving. This phenomenon was checked in a trial with 28 cows prone to milk fever under farm conditions (3.6). In the last 4 weeks of the dry period, 15 cows were infused twice with 50 mg EDTA/kg bodyweight. There were 14 days between the first and the second infusion. The other 13 cows served as controls. The infusions did not contribute to the prevention of milk fever. These results could be explained by experiments with 3

cows infused with EDTA and fed on a diet containing different amounts of calcium. The activating ability of repeated EDTA infusions interfered with the inactivating ability of rations with a high calcium content.

Because the content of calcium in the diet is an important key in preventing milk fever, diets low in calcium and high in phosphorus were alternated weekly with diets high in calcium and low in phosphorus for 4 cows in order to study the effect of alternation on the activity of calcium metabolism (3.7). After calving, a severe decline in plasma calcium was seen in 3 cows fed on a diet rich in calcium and poor in phosphorus during the last prepartal week, in contrast to no plasma calcium decline in 1 cow fed on a diet poor in calcium and rich in phosphorus.

Since a diet poor in calcium during the last weeks of pregnancy can activate the calcium metabolism, another experiment with 29 milk fever prone cows was carried out (3.8). For some weeks pre partum, a diet poor in calcium was given to 14 experimental cows, the others (controls) were fed on a normal (Ca-rich) diet. During the last prepartal week, the diet was changed to calcium-rich for the experimental cows, in order to replenish the possibly exhausted calcium pool. At the time of diet change the experimental cows received an oral dose of 250 g $CaCO_3$. Activated calcium metabolism hardly remained after calving (severe decline in plasma calcium) because of the 1 week on a calcium-rich diet. Cows with an increase of at least 1.5 mg Ca/100 ml plasma after the oral dose of $CaCO_3$ got milk fever. In this experiment, the frequency of milk fever was 50% for both the control and the experimental group. There was also no marked decline in the number of cows with a decline in plasma calcium below 7.5 mg Ca/100 ml plasma.

With 5 cows (3.9) fed on a diet containing 0.17% Ca in dry matter and with 7 cows fed on a diet containing 0.80% Ca in dry matter of the whole ration, an experiment was made during the last 3 months of lactation, the last 2 months of pregnancy and the first week post partum. The cows fed on a diet poor in calcium had the same milk production as the cows fed on a diet rich in calcium. After calving, the 5 cows (calcium-poor diet) had less decline and a quicker recovery of plasma calcium than the cows fed on a calcium-rich diet. The daily intake of phosphorus did not influence this result.

Section 3.10 describes an experiment with 26 cows prone to milk fever. During the last 4 prepartal weeks and the first postpartal week a diet poor in calcium was supplied to 12 experimental cows. In the control group (14 cows) that was fed on a normal (Ca-rich) diet, 4 cows (29%) had to be treated for milk fever and 10 cows (71%) had a decline in plasma calcium to 7.5 mg/100 ml or less. In the experimental group, 1 cow (8%) had to be treated for milk fever and 3 cows (25%) showed a decline in plasma calcium to 7.5 mg/100 ml or less (Table 35). Moreover, in an experiment on 8 cows with no milk fever history and fed on a diet poor in calcium (3.11), it appeared that the increased activity of calcium metabolism and an intestinal absorption coefficient of calcium, that remained at a high level, can be used to increase the calcium input by giving the cows a calcium-rich diet soon after calving. This enlarged intestinal calcium input was responsible for a slight decline and a rapid recovery of plasma calcium post

partum and no milk fever was seen (Table 40).

In 3.12, the effect was studied of a daily magnesium supplement for cows prone to milk fever and fed on a diet claimed to prevent low calcium levels in plasma and milk fever after calving. No milk fever was seen in the 45 cows, while a small number (9%) had a decline in plasma calcium to 7.5 mg/100 ml or less. The cows produced on average 26.7 litre milk on Day 7 post partum. From numerous experiments (Sections 3.3 – 3.11) I found indications that there was an interaction between magnesium and calcium in blood plasma. That interaction was seen only when the daily dietary magnesium supply was sufficient. Therefore two groups were formed. One received daily 20 g magnesium extra with the diet; this group had a higher concentration of plasma magnesium after calving than the control group (Table 45). In other cows too (Tables 46 and 47), this interaction was seen in plasma. I conclude that at the moment of declines in plasma calcium, usually seen in parturient cows, an increase in plasma magnesium is seen only when the daily intake of magnesium is sufficient pre and post partum.

To judge the results of the 45 cows of Section 3.12, an 'aggregate' control group was formed from 4 control groups of 4 different years (total 71 cows, prone to milk fever; Table 48). The average plasma concentration of minerals of the 71 cows contribute to an interpretation of the plasma figures of the cows justified in Section 3.12. Hardly any decline in plasma calcium was seen in contrast to those of the 'aggregate' control group. This 'aggregate' control group of 71 cows needed 3 days or more to return to normal levels of plasma calcium. There was no milk fever in the 45 cows against 15, 31, 53 and 29% incidence in the 4 control groups that formed the 'aggregate' control group. Also the number of cows with a plasma calcium decline to 7.5 mg/100 ml or less was reduced (9%) in contrast to the 4 groups of the 'aggregate' control group (57, 50, 92 and 71%).

Recommendations about the diet and the consequences for the dairy farmer, feedstuff millers and others, are given in 4.1 and 4.2.

Samenvatting

Melkziekte bij koeien komt in Nederland nog steeds veelvuldig voor. Dit betekent voor de veehouder niet alleen financiële schade, ook het ongemak moet niet onderschat worden. Melkziekte, die doorgaans voorkomt binnen enkele dagen na de partus, wordt voorafgegaan door, en gaat gepaard met een duidelijke daling van het calciumgehalte in bloedplasma. Het optreden van calciumdaling in het bloed na de partus en daarop aansluitend melkziekte is het onderwerp van het in dit proefschrift beschreven onderzoek.

Het onderzoek werd uitgevoerd met hoofdzakelijk melkziektegevoelige koeien die ons ter beschikking werden gesteld door een groot aantal veehouders uit het noordelijk deel van de provincie Noord-Holland. De koeien arriveerden gewoonlijk 4 weken voor de berekende kalfdatum op een gehuurd bedrijf en werden ongeveer 1 week na het kalven teruggebracht.

In de eerste proef (3.3) werden de koeien verdeeld over een controle- en een proefgroep. Gedurende de laatste 4 weken van de dracht werd de controlegroep een rantsoen van hooi en krachtvoer gevoerd overeenkomstig de geldende energie- en eiwitnormen en met een Ca/P verhouding van 1,7. De proefgroep ontving hetzelfde rantsoen maar met een Ca/P verhouding van 1,1 en met dien verstande dat het rantsoen van 7 dagen in 6 dagen verstrekt werd zodat de koeien een dag moesten vasten. Het rantsoen en de manier van voeren, waarvan verwacht werd dat het de calciumstofwisseling activeert, bleken geen wezenlijke bijdrage te leveren ter voorkoming van lage plasma-calcium-waarden en melkziekte na het kalven (tabellen 2, 3 en 4). Het percentage gevallen van melkziekte bedroeg 25 en 15% voor de proefgroep, respectievelijk controlegroep. Bij een droogstaande drachtige koe had het niet verstrekken van calcium, zonder dat de opname van energie en eiwit werd verlaagd, hetzelfde verlagende effect op het plasma-calcium-gehalte als een algeheel vasten.

In de volgende proef (3.4) werden de melkziektegevoelige koeien van de proefgroep, met het doel de calciumstofwisseling te activeren, in tegenstelling tot de controlegroep, 20% onder de geldende energie- en eiwitnormen gevoerd met een rantsoen (hooi en krachtvoer) dat een Ca/P verhouding had van 1,0. De percentages van gevallen van melkziekte waren 53 en 31% voor de proefgroep, respectievelijk controlegroep.

Bij een aantal koeien, dat gevoerd werd met een hooi en krachtvoer rantsoen (3.5) werd het effect van een experimenteel geïnduceerde plasma-calcium-verlaging op het activeren van de calciumstofwisseling onderzocht. Een belangrijke daling van het plasmagehalte van calcium, veroorzaakt door herhaalde EDTA-inspuitingen tijdens de

droogstand, bleek de calciumstofwisseling te activeren. Wanneer deze herhaalde plasma-calcium-daling tevens tot stand werd gebracht 3 – 10 dagen voor het afkalven, bleek het calcium-metabolisme zo geactiveerd te zijn, dat na het kalven de plasma-calcium-daling minimaal was. Daarom werden op praktijkschaal in een proef met 28 melkziekte gevoelige dieren (3.6) 15 koeien tijdens de laatste 4 weken van de dracht twee keer, met 14 dagen tussenruimte, ingespoten met 50 mg EDTA/kg lichaamsgewicht. De andere 13 dieren dienden als controle. Onder de plaatselijke voedingsomstandigheden bleken de injecties geen bijdrage te kunnen leveren ter preventie van melkziekte. Bij drie melkziekte gevoelige koeien, geïnjecteerd met EDTA, die gevoerd werden met verschillende hoeveelheden calcium in het rantsoen, bleek dat het activerend vermogen van deze EDTA-inspuitingen teniet gedaan werd door een hoog calciumgehalte van het rantsoen.

Aangezien het calciumgehalte in het voer een belangrijke sleutel is in de preventie van melkziekte, werd geprobeerd om door middel van per week wisselend veel en weinig calcium en fosfor in het voer, het calcium-metabolisme te activeren (3.7). Na het afkalven lieten 3 van de 4 koeien, in de laatste week van de dracht gevoerd met calciumrijk en fosforarm voer, een belangrijke plasma-calcium-daling zien in tegenstelling tot één koe die de laatste week voor het kalven calciumarm en fosforrijk gevoerd werd.

Aangezien een calciumarm rantsoen tijdens de laatste weken van de dracht een activerende werking op het calcium-metabolisme leek te bezitten, werd in een proef met 29 melkziekte gevoelige koeien (3.8) aan 14 proefkoeien gedurende een aantal weken voor het afkalven calciumarm voer verstrekt om hun calcium stofwisseling te activeren, terwijl de 15 controlekoeien het normale rantsoen ontvingen. De laatste week voor het kalven werden de proefkoeien echter weer gevoerd zoals de controlekoeien (Ca-rijk) teneinde een mogelijk te klein geworden mobiele 'calcium-pool' weer aan te vullen. Bovendien kreeg een aantal koeien bij de overgang van calciumarm naar calciumrijk voer een orale dosis van 250 g $CaCO_3$ toegediend. Na activatie van het calcium-metabolisme bleek het voeren van calciumrijk voer gedurende 1 week voor het kalven een zodanig effect te hebben dat na het kalven van een geactiveerde calciumstofwisseling geen sprake was. De koeien die een calciumarm rantsoen ontvingen en bij de overgang van het rantsoen van calciumarm naar calciumrijk op een orale $CaCO_3$-gift reageerden met een plasma-calcium-stijging van 1,5 mg/100 ml of meer, kregen melkziekte. Ook deze proefbehandeling leverde niet de gewenste daling in het aantal melkziektegevallen ($\pm$ 50% melkziekte in zowel de controle- als de proefgroep) en in het aantal dieren met een plasma-calcium-daling tot beneden 7,5 mg/100 ml op.

Met 5 koeien die calciumarm gevoerd werden (0,17% Ca in de droge stof van het rantsoen) en met 7 koeien calciumrijk gevoerd (0.80% Ca in de ds) werd de volgende proef uitgevoerd (3.9). Deze voeding werd verstrekt gedurende de laatste 3 maanden van de lactatie, de 2 maanden droogstand en 1 week na het kalven. De calciumarm gevoerde koeien bleken evenveel melk te produceren en na het kalven een minder sterke daling en een sneller herstel van het plasma-calcium te vertonen dan de contro-

ledieren. Een hoge fosfor- in combinatie met lage calcium-opname bleek op deze effecten geen invloed te hebben.

Vervolgens werd bestudeerd of het calcium-metabolisme van 12 melkziekte gevoelige dieren (3.10) te activeren was door gedurende de laatste 4 weken van de dracht en de eerste week na het kalven calciumarm te voeren. In de controlegroep (14 koeien), gevoerd met een normaal (Ca-rijk) rantsoen, kregen 4 koeien (29%) melkziekte, bij 10 koeien (71%) daalde het plasma-calcium-gehalte tot beneden 7,5 mg/100 ml. In de proefgroep kreeg 1 koe (8%) melkziekte, terwijl bij 3 koeien (25%) het plasma-calcium-gehalte daalde tot beneden 7,5 mg/100 ml (tabel 35). Bovendien bleek uit een proef met 8 niet voor melkziekte gevoelige koeien die gevoerd werden met een calciumarm rantsoen (3.11), dat de toegenomen activiteit van het calcium-metabolisme en een daardoor op een hoog niveau gefixeerde absorptie-coëfficiënt van calcium in de darm na het kalven uitgebuit kon worden door het verstrekken van een calciumrijk rantsoen na dit kalven, hetgeen resulteerde in een plotseling vergrote opname van calcium. Door deze vergrote calciumopname bleek de daling minder en het herstel van de daling sneller te zijn, bovendien trad geen melkziekte op (tabel 40).

Het effect van de dagelijkse voorziening van magnesium werd bestudeerd (3.12) bij melkziekte gevoelige koeien welke volgens de reeds beschreven wijze werden gevoerd, een wijze die pretendeert lage calciumwaarden en melkziekte na het kalven te voorkomen. Van 45 koeien kreeg er niet één melkziekte, in een klein aantal (9%) daalde het plasma-calcium-gehalte tot beneden 7,5 mg/100 ml. Zeven dagen na het kalven produceerden de koeien gemiddeld 26,7 liter melk per dag (tabellen 44 en 49). Uit een aantal proeven (3,3 – 3.11) werd de indruk verkregen dat er een wisselwerking tussen magnesium en calcium in bloedplasma bestaat welke alleen tot uiting komt bij een voldoend hoge dagelijkse magnesiumvoorziening. Twee groepen werden geformeerd, waarvan de één met het rantsoen 22 g magnesium extra per dag kreeg toegediend. Het bleek dat deze koeien een gemiddeld hoger magnesium-gehalte in het plasma na het kalven hadden dan de controledieren (Tabel 45). Ook andere koeien (tabellen 46 en 47) lieten duidelijk het verband zien tussen dalingen van calcium en stijgingen van magnesium in plasma. Geconcludeerd wordt dat in plasma bij de na het kalven doorgaans optredende calciumdaling het magnesium pas zal stijgen als de dagelijkse voorziening van magnesium voor en na het kalven voldoende is. Teneinde de 45 koeien te kunnen vergelijken met een controlegroep werden 4 controlegroepen van 4 verschillende proeven met melkziekte gevoelige dieren (totaal 71 koeien) samengevoegd tot één controlegroep (tabel 48). Het totaalbeeld van de plasmagehalten van deze 71 koeien levert een aanzienlijke bijdrage tot een juiste interpretatie van het in twee groepen verdeelde aantal van 45 koeien (3.12). Op de tijdstippen ½, 1, 1½ dag na de partus (tabel 49) werden voor de controlegroep calcium-gehalten gevonden variërend tussen 7,3 en 6,8 mg/100 ml tegen gehalten van de proefgroepen tussen 9,9 en 9,0 mg/100 ml plasma. Drie dagen na de partus bleken de plasma-calcium-waarden van de controlegroep zich pas te gaan herstellen. Bij de 45 koeien werd geen melkziekte (0%) gesignaleerd tegen 15, 31, 53 en 29% in de 4 controlegroepen welke in totaal 71 dieren omvatten. Ook het percentage koeien waarvan het plasma-calcium-gehalte

daalde tot beneden 7,5 mg/100 ml bleek sterk verminderd (9% voor de 45 proefkoeien) in tegenstelling tot het percentage van de 71 koeien van de 4 verschillende controlegroepen dat respectievelijk 57, 50, 92 en 71% bedroeg.

Voor aanbevelingen voor de veehouder, veevoederfabrikant en anderen, zij verwezen naar 4.1 en 4.2.

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Curriculum vitae

De auteur werd op 3 september 1941 geboren te Oosternieland (gemeente Uithuizermeeden). Na de lagere school behaalde hij in juni 1960 het diploma HBS-B aan de Rijks-HBS te Warffum. Na de zomervakantie begon hij zijn studie aan de Faculteit der Geneeskunde van de Rijksuniversiteit te Utrecht. Na het behalen van het propaedeutisch examen in 1961 veranderde hij van studierichting en liet zich inschrijven aan de Faculteit der Diergeneeskunde.

Vooruitlopend op een beslissing van de Minister van Defensie meldde hij zich in het najaar van 1961 aan voor het vervullen van de militaire dienst, welke hij, als wachtmeester der lichte luchtdoelartillerie, grotendeels vervulde in het voormalige Nederlands Nieuw-Guinea. Aan het einde van deze periode werd hem, naast het Herinneringskruis (K.B. 29 september 1962 Nr. 1), een 'Tevredenheidsbetuiging wegens buitengewone plichtsbetrachting en bijzondere toewijding gedurende de periode van paraatheid te velde' verleend.

Op 1 januari 1963 hervatte hij de studie, waarna hij als dierenarts op 1 oktober 1968 afstudeerde aan de Faculteit der Diergeneeskunde te Utrecht.

Sinds 1 november 1968 is hij als onderzoeker verbonden aan het Instituut voor Veevoedingsonderzoek 'Hoorn' en werkzaam op het gebied van de relatie voeding-gezondheid bij rundvee.

TOWNS AND VILLAGES

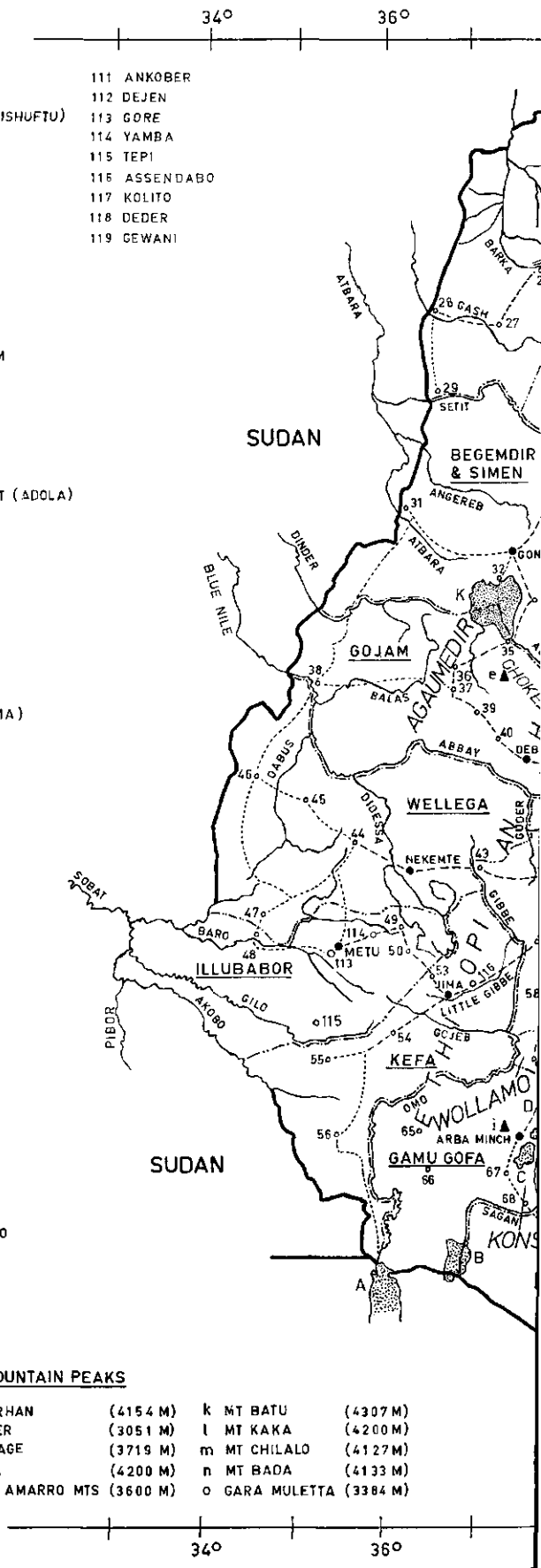
1 DEBRE BIRHAN
2 DEBRE SINA
3 KARA KORE
4 KOMBOLCHA
5 BATI
6 TENDAHO
7 SERDO
8 ASSAB
9 WOLDYA
10 KOBO
11 ALAMATA
12 LALIBELA
13 SOKOTA
14 MAICHEW
15 ENDA MEDHANE ALEM
16 ABIY ADI
17 AXUM
18 ADUA
19 ADIGRAT
20 SENAFE
21 ADI KAYEH
22 ADI UGRI
23 DEKEMHARE
24 MASSAWA
25 KEREN
26 AGORDAT
27 BARENTU
28 TESENEY
29 OM HAJER
30 DEBAREK
31 METEMA
32 GORGORA
33 ADDIS ZEMEN
34 DEBRE TABOR
35 BAHAR DAR
36 DANGLA
37 INJIBARA
38 GUBA
39 BURE
40 DEMBECHA
41 FICHE
42 AGERE HIWET (AMBO)
43 BAKO (SHOA)
44 GIMBI
45 MENDI
46 ASOSA
47 DEMBI DOLO
48 GAMBELA
49 BEDELLE
50 DEMBI
51 GHION (WOLLISO)
52 WELKITE
53 AGARO
54 BONGA
55 MIZAN TEFERI
56 MAJI
57 BUTAJIRA
58 HOSAINA
59 *DEBRE ZEIT (BISHUFTU)*
60 MOJO
61 MAKI
62 ADAMI TULU
63 SHASHAMANE
64 SODDO
65 BULKI
66 BAKO
67 GIDOLE
68 GIARSO
69 YABELO
70 BURJI
71 AGERE MARIAM
72 FISHA GENET
73 YIRGA CHAFFE
74 DILA
75 WONDO
76 YIRGA ALEM
77 AGERE SELAM
78 KEBRE MENGIST (ADOLA)
79 NEGELLI
80 MEGA
81 MOYALE
82 DOLO
83 EL KERE
84 GINIR
85 ADABA
86 DODOLA
87 BEKOJI
88 TICHO
89 NAZRET (ADAMA)
90 METAHARA
91 AWASH
92 MIESO
93 ASBE TEFERI
94 BEDESSA
95 GELEMSO
96 HIRNA
97 KOBBO
98 DIRE DAWA
99 ALEMAYA
100 FIK
101 IMI
102 JIJIGA
103 DEGEH BUR
104 AWARE
105 WERDER
106 GELADI
107 SHILALO
108 KEBRE DEHAR
109 KELAFO
110 FERFER
111 ANKOBER
112 DEJEN
113 GORE
114 YAMBA
115 TEPI
116 ASSENDABO
117 KOLITO
118 DEDER
119 GEWANI

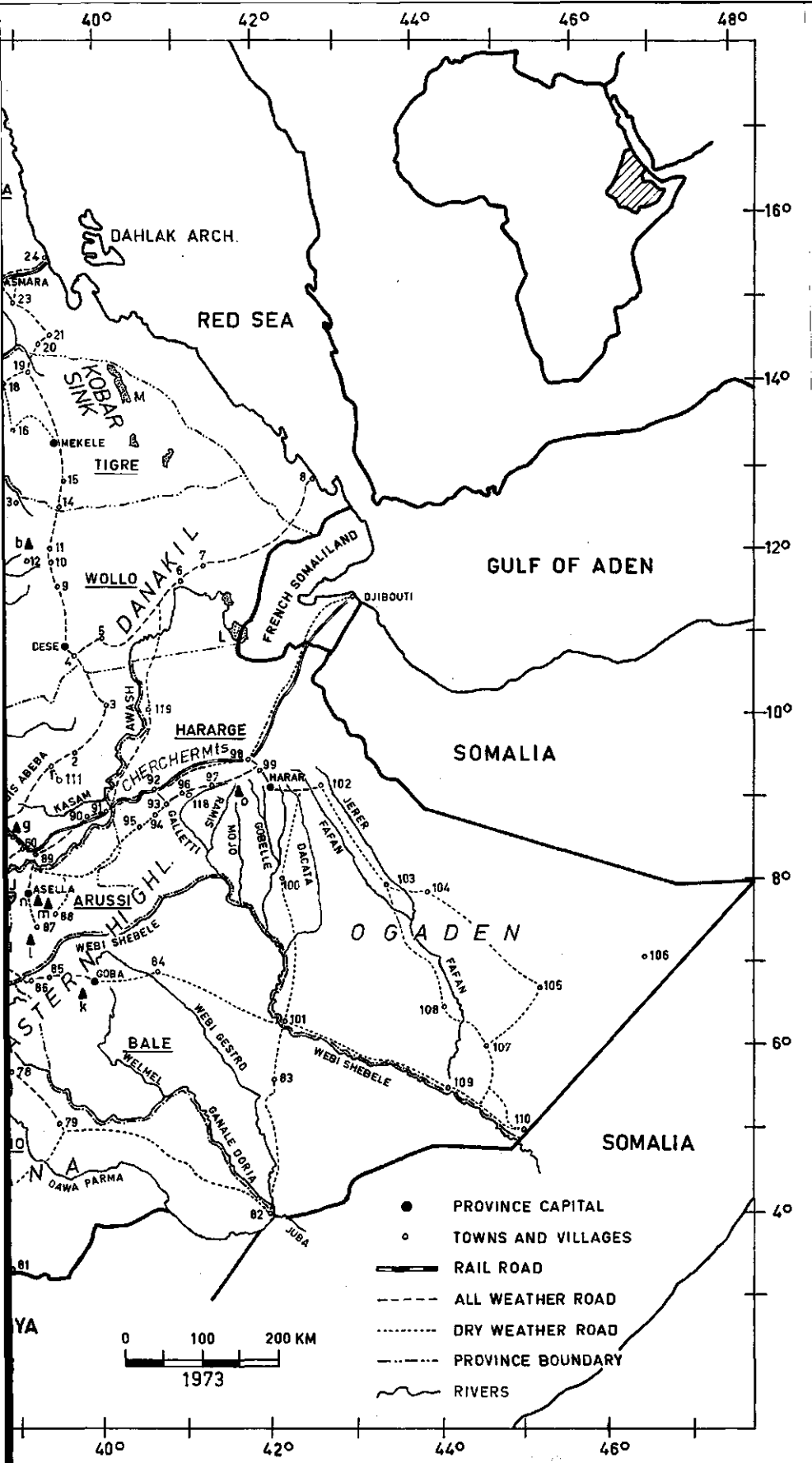
LAKES

A LAKE RUDOLF
B LAKE CHEW BAHIR
C LAKE CHAMO (RUSPOLI)
D LAKE ABAYA (MARGHERITA)
E LAKE AWASA
F LAKE SHALA
G LAKE ABAYITA
H LAKE LANGANO
J LAKE ZIWAI
K LAKE TANA
L LAKE ABBE
M LAKE ASALE

MOUNTAIN PEAKS

a RAS DASHAN (4620 M)
b MT ABUNA YOSEF (4194 M)
c MT GUNA (4261 M)
d AMBA FARIT (3978 M)
e MT AMEDAMIT (3619 M)
f RAS BIRHAN (4154 M)
g MT YERER (3051 M)
h MT GURAGE (3719 M)
i MT TOLA (4200 M)
j PEAK IN AMARRO MTS (3600 M)
k MT BATU (4307 M)
l MT KAKA (4200 M)
m MT CHILALO (4127 M)
n MT BADA (4133 M)
o GARA MULETTA (3384 M)

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DAHLAK ARCH.
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KOBAR SINK
MEKELE
TIGRE
WOLLO
DANAKIL
FRENCH SOMALILAND
DJIBOUTI
DESE
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HARARGE
CHERCHERMts
HARAR
KASAM
GALLETTI
RAMIS
GOBELLE
DACATA
JERER
FAFAN
ARUSSI
HIGHL
OGADEN
WEBI SHEBELE
GOBA
BALE
WELMEL
WEBI GESTRO
GANALE DORIA
DAWA PARMA
JUBA
PROVINCE CAPITAL
TOWNS AND VILLAGES
RAIL ROAD
ALL WEATHER ROAD
DRY WEATHER ROAD
PROVINCE BOUNDARY
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