INFLUENCE OF FERTILISER TREATMENT OF GRASSLAND ON THE INCIDENCE OF HYPOMAGNESAEMIA AND HYPOMAGNESAEMIC TETANY (GRASS TETANY) IN MILKING COWS [1]

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Summary

1. Following a statistical study (Kemp and 't Hart, 1957) grazing experiments were conducted in 1956 and 1957 in which 16 milking cows were examined throughout the grazing season in order to ascertain what effect dressing grassland with potash, phosphate and nitrogen had on the magnesium levels of the blood serum and the incidence of grass tetany.

2. It was found that a heavy potash dressing goes with lower serum magnesium levels, and that this effect is considerably greater in spring and autumn than in the summer. A heavy nitrogen dressing had a similar effect, although not so marked. The greatest differences in serum magnesium levels occurred between two groups of animals one of which grazed on plots which had received a light potash and nitrogen dressing, and the other on plots which had been given a heavy potash and nitrogen dressing. Over the entire experimental grazing season the mean differences were 0.82 mg Mg per 100 ml serum, and even more than 1.20 mg/100 ml in the spring and autumn. It was only in the autumn that a heavy phosphate dressing was accompanied by a slight drop in the serum magnesium levels, but the significance of this is doubtful.

3. In these grazing trials we succeeded in inducing grass tetany experimentally. In the spring of 1957 six animals were suffering from grass tetany, and four of these seriously. At the time of the attack the serum magnesium levels of these animals were 0.2, 0.4, 0.4, 0.4, 0.5 and 0.7 mg per 100 ml. All these cases arose on the plots with a heavy potash dressing, both with a low and a high nitrogen dressing; during the same period the serum magnesium levels of the remaining animals were also low, and occasionally very low.

4. The size of the variability of serum magnesium levels of milking cows grazing the same pasture depends on the conditions governing the size of the mean level of the entire group. The smallest variability of the single observations were found under favourable and very unfavourable conditions, and hence with low and high mean serum magnesium levels per group.

5. An other investigation carried out on 23 farms showed that the serum magnesium levels of 90 milking cows grazing the same pastures on which cases of grass tetany had occurred during the period of sampling, had been reduced in nearly every case and may sometimes be very low. Of these 90 animals, only one had a level higher than 2.0 mg/100 ml Mg, while 60% of the animals had values of less than 1.1 mg/100 ml. Under the conditions prevailing in Holland this means that a considerable proportion of milking cows suffer from hypomagnesaemia in the spring and autumn in particular.

Introduction

Sjollema (1931) found that the occurrence of grass tetany is invariably associated with a condition of hypomagnesaemia. Hence it was on the basis of this study that he developed the Ca-Mg therapy which is still being successfully applied today, Sjollema distinguished two stages in the course of this "disease", viz. the first stage in which there is a fairly unstable condition,

[1] Received for publication September 15, 1958.

the magnesium levels of the blood serum having fallen to lower or very low values (hypomagnesaemia). The second stage is characterised by the occurrence of the such well-known clinical symptoms as hyperaesthesia, convulsions and prostration. According to this research worker, the main cause of the hypomagnesaemic condition, which always precedes the clinical pattern, lies in the feed (SJOLLEMA, 1952). He believes that the composition of the herbage is of decisive importance, and attention was drawn to the relationship between the fertilisation of the grassland and the incidence of grass tetany. Potash and nitrogen fertilisation were said to be the chief factors.

Subsequent workers, BARTLETT et al. (1954), found in grazing experiments carried out on trial swards seeded with cocksfoot and clover that the application of sulphate of ammonia affected the incidence of hypomagnesaemia and grass tetany. The harmful effect of a sulphate of ammonia dressing was connected by the said workers with the suppression of clover from the sward. In an investigation carried out on a great number of farms, KEMP and 'T HART (1957) found a connection between dressing grassland with potash and nitrogen on the one hand and the incidence of grass tetany on the other. The data of this investigation also revealed a correlation between the potassium, calcium and magnesium contents of the sward and the percentage incidence of grass tetany.

But despite the above investigations, there is a very limited amount of data on studies under controlled conditions of the effect which fertilising grassland has on the incidence of hypomagnesaemia and grass tetany. It was therefore decided to lay down grazing experiments, paying particular attention to the effect which dressing the pasture with potash, nitrogen and phosphate has on the composition of the pasture and the magnesium level of the blood serum of milking cows. This article will only deal with the fertilisation of the pasture and its effect on the serum magnesium levels; a subsequent article will discuss the composition of the pasture in connection with the magnesium level of the blood serum [2]).

LAYOUT OF THE EXPERIMENTS

The experiments were carried out on permanent grassland, belonging to the "Droevendaal" Experimental Farm, Wageningen, situated on fairly low-lying sandy soil with 8% organic matter, 10% clay, and what by Dutch standards is a normal phosphate and potash condition in the 0–5 cm layer. In 1956 the experimental field was 6.09 hectares and divided into 21 plots. All these plots received an equal amount of nitrogen, but seven had a very heavy potash dressing and seven others a normal potash dressing. These fourteen plots all received the same phosphate dressing. The last seven plots, however, were dressed with a normal amount of potash, but received a very heavy phosphate dressing. The result was a grazing experiment with three treatments, according to the following plan. (The treatments are designated by symbols, a capital letter denoting a heavy dressing and a small letter a light dressing).

[2]) The entire statistical material relating to experiments conducted in 1956 and 1957 will be given in a separate stencilled report which will be sent free of charge to persons interested on application to the Institute for Biological and Chemical Research on Field Crops and Herbage, Wageningen.

1956 : 6.09 hectare experimental pasture with three treatments.

Treatment	Area of treatment in hectares	Number of plots per treatment	Number of cows	Dressing in kg per hectare		
				N	P_2O_5	K_2O
NpK	1,53	7	4	230	30	50
NPk	1,53	7	4	230	260	50
Npk	3.03	7	8	230	30	450

On the plots of the Npk and NpK treatments the nitrogenous fertiliser was applied in the form of nitro-chalk of which 80 kg per hectare were given in the spring on 21st March and the remainder after the first four grazings. On the other hand, the plots of the NPk treatment received the nitrogen in the form of ammonium phosphate, in which form the heavy phosphate dressing was also applied to these plots. This means, therefore, that on these plots the phosphate dressing was given in portions over the whole season. The plots of the Npk and NpK treatments only received 30 kg P_2O_5 per hectare in the form of superphosphate in the spring on 15th March. The potash dressing was given to all plots in the form of 60% muriate of potash; on 15th March 300 kg K_2O per hectare were applied to the NpK plot and 40 kg to the Npk and NPk plots. The remaining quantities, viz. 150 and 10 kg, K_2O per hectare were given in August.

In 1957 no further data were assembled on the effect of a heavy and light phosphate dressing; in their place a heavy and a light nitrogen dressing were included in the investigation. To this end four treatments were given with potash and nitrogen in the following combinations: nk, Nk, nK and NK. For this experiment the same plots were employed as in 1956 but with an added 1.55 hectares, so that the total area in 1957 was 7.64 hectares. This area was divided into 36 plots. In 1957 the plots of the four treatments were fertilised in accordance with the following plan, nitrogen being given in the form of nitro-chalk, potash in the form of 60% muriate of potash, and phosphate in the form of superphosphate.

1957 : 7.64 hectare experimental pasture with 4 treatments.

Treatment	Area per treatment in hectares	Number of plots per treatment	Number of cows	Dressing in kg per hectare		
				N	P_2O_5	K_2O
nk	2.54	9	4	20	30	20
Nk	1.28	9	4	210	30	20
nK	2.54	9	4	20	30	200–300
NK	1.28	9	4	210	30	200–300

The parts of the experimental field which received in 1956 a heavy potash dressing received also a heavy potash dressing in 1957; the parts of the experimental field which received in 1956 a light potash dressing received also a light potash dressing in 1957. The 1956 plots high and low potassium were divided in a ratio 1 : 2 on which the N and n plots were laid out, respectively. It should be noted that although there are the same number of cows to a treatment, owing to the difference in the amounts of nitrogen there would be great differences in the sward growth. In 1957 the Nk and nk plots were laid down on the 1956 Npk and NPk plots. In this case also the nk plots receiving a light N dressing were twice the area of the Nk plots receiving a heavy N dressing. Shifting the fencing ensured that the effect of the heavy phosphate dressing of the NPk plots in 1956 was equally marked on the Nk and nk plots in 1957. The spring nitrogen dressing was applied in two portions to the plots with a heavy nitrogen dressing, viz. 40 kg N per hectare on 22nd March and 40 kg per hectare on 9th April. As in 1956, the remainder was given after the first four grazings. The plots with a light nitrogen dressing only received 20 kg N per hectare on 22nd March. The nK and NK plots received 200 kg K_2O per hectare on the 20th March. The nK and NK plots on the part of the experimental field which was added in 1957 received an addional potash dressing of 100 kg K_2O per hectare in August, to compensate the difference resulting from the after effect of the potash dressing in 1956. The nk and Nk plots only received 20 kg K_2O per hectare on 20th March. The phosphate dressing was also given on 20th March.

Grazing

In both years the experiments lasted from the beginning to the end of the grazing period, viz. in 1956 from 4th May to 26th October, and in 1957 from 23rd April to 24th October. Both in 1956 and 1957 there were two periods in the summer and early autumn when there was no longer enough herbage available to the animals on the experimental pastures. During these periods all animals were turned out together on a pasture outside the experimental field.

In both years the number of milking cows was 16; 8 of the 16 animals used in 1956 were employed again in 1957. The ages varied from three to eight years. The cows used for the 1956 experiment all calved in the period November 1955 to April 1956, and those used for the 1957 experiment from October 1956 to April 1957. During both years the animals' milk production fluctuated around 17 to 25 kg per head per day before the experiment was begun. Of the cows used for the experiment, none had previously suffered from grass tetany, and so far as could be ascertained grass tetany had likewise never occurred on the experimental plots in previous years. As stated earlier, in 1956 three groups of cows were assembled and in 1957 four. Factors taken into account in making this division were age, date of calving, milk production and the Mg level of the blood serum on two dates before grazing began. In this way the groups assembled were as uniform as possible. Owing to the experiments being divided up into a fairly large number of plots, as well as the number of cows to a plot, it was possible to employ a rotational grazing system in which, depending on the sward growth, the animals did not graze the same plot for longer than four to five days on the average, after which they were transferred to a new plot. When the experiment was begun in 1956 the percentage of herbs was about 5% and the percentage of clover considerably lower. But in order to reduce the percentage of clover and herbs still further, all plots were given hormonal sprays in both years, as a result of which very little clover or herbs appeared in the sward during the further course of the experiments. All groups of animals always grazed closed to each other so as to ensure that as far as possible all groups were affected in the same way at the same time by differences in fertility within the entire experimental field. All 16 cows were always transferred to new plots at the same time. During the entire grazing periods the animals were given grass only, viz. no supplementary feed, and they also remained in the pasture at night. The only exception which had to be made was during the period from 10th to 16th May 1957 when owing to the many cases of grass tetany all animals were given cakes containing supplementary magnesium oxide.

Sampling

Except that samples of urine and faeces were also taken partly for other investigations, care was primarily taken to ensure a systematic and fairly frequent sampling of herbage and blood throughout the grazing season. In 1956 one herbage sample was taken from each plot where cows were grazing every two or three days. All treatments were always sampled simultaneously. In these samples the N, K, Na, Ca, Mg, Cl, S and P contents were determined. In 1957 the herbage samples were taken just before the cows had been transferred to a new plot, viz. depending on the date of transferring. In this year as well, all treatments were always sampled simultaneously, and the analysis was confined to the N, K, Na, Ca and Mg contents.

During the stall period, blood samples were taken in the spring before the cows went to pasture, and in the autumn about 10 days after stalling. The spring sampling also helped in assembling the groups, while the object of the autumn sampling was to ascertain the serum magnesium levels after the cows in the stall had been given the same feed for some time. During the 1956 grazing period, blood samples were taken twice a week from 8th May to 5th June, viz. eight animals on Tuesday, and the other eight on Friday. From 5th June to the end of the grazing period, samples were taken of the blood of eight cows a week, and samples of the other eight in the following week. In 1957 more blood samples were taken, a blood sample being taken of every animal every Thursday throughout the grazing period. In these samples, which were taken from the jugular vein, the magnesium level of the serum was determined, and also the Ca and P contents in a number of the 1956 samples.

Results

The trend of the serum magnesium levels at the various treatments is shown in Figures 1 to 8. These figures show the trend of the mean magnesium levels of the blood serum of four cows from each separate treatment throughout the growing season, and all four treatments are compared. As was stated above in the introduction, all individual levels are recorded in a separate report.

1956. With regard to the 1956 experiment, in Figs. 1 and 2 the serum magnesium levels of the cows of the NpK and NPk plots respectively are compared with the trend of these levels on the Npk plots. In both these figures it is possible to distinguish different periods, viz. in the first place the parts with horizontal hatching before the experiment began on 4th May and after it was completed on 26th October. During these periods the animals were in the stalls and all receiving the same feed. During the grazing season there are two periods with vertical hatching, viz. from 27th June to 11th July, and from 15th August to 11th September. During this time there was no longer sufficient grass available on the experimental pastures and all animals used in the experiment grazed on a pasture outside the experimental field, so that they all consumed the same grass.

Fig. 1 shows that the magnesium levels of the blood serum of the cows of the NpK plots fell very considerably immediately after going to graze, and especially during the first weeks the levels differed appreciably from those of the Npk plots. In the period from 27th June to 11th July, when all animals were grazing on the same pasture, these differences disappeared. But after they had again been put to pasture in the experimental area on 11th July there was again a steep decline in the serum magnesium levels on the NpK plots, and consequently there was once again a considerable difference between these levels and the levels of the Npk plots. From 14th August to 11th September all experimental animals were again on the same pasture, and the cows which had been grazing the NpK plots recovered, so that after some 10 days there were no further systematic differences between the groups. Of the entire grazing period, the lowest serum magnesium levels occurred in the autumn, viz. in October. Very soon after the animals had again been put to pasture on the experimental area there were great differences between these treatments; these differences in levels no longer occurred after the cows had been in the stalls for about 10 days and had consumed the same feed.

Fig. 2 shows an entirely different pattern in the trend of the serum magnesium levels. From the beginning of the grazing period until October the mean serum magnesium level of the cows from the NPk plots are admittedly somewhat lower than those of the Npk plots, but there can be no question of systematic differences when the spreads of the individual observations are also taken into account. In October, however, there was a fairly sharp decline in the serum magnesiuum levels of the NPk plots, although in view of the small number of observations to which these differences relate, it would be inadvisable to base any far-reaching conclusions on this. But the fact that the differences in level no longer occurred in the stalls at the beginning of November does suggest that the phosphate dressing had an adverse effect on the serum magnesium levels. In this connection it should be observed that the phosphatic dressing on the NPk plots was given simultaneously with the nitrogen throughout the season, so that the maximum amount had been reached by the beginning of the autumn. This phosphate effect will be discussed in greater detail in a subsequent article dealing with the composition of the sward.

In order to give an approximate idea of the size of the differences in level between the three treatments, the mean serum magnesium levels over the various periods are reported in Table 1.

Table 1 Mean serum magnesium levels of milking cows at different periods and under various conditions.

	Stall	Experimental pastures	Outside per experiment	Experimental pastures	Outside the experiment	Experimental pastures	Stall
Period → / Groups ↓		4/5-27/6	27/6-11/7	11/7-14/8	14/8-11/9	11/9-26/10	
Npk	2.51	2.18	2.18	2.20	2.25	2.24	2.28
NPk	2.50	2.16	2.38	2.07	2.33	1.80	2.30
NpK	2.53	1.86	2.22	1.71	2.19	1.12	2.13

This shows first of all that the differences in serum magnesium levels between the Npk and NpK plots increase as the season advances, and that in the last grazing period the mean level on the NpK plots is half the level on the Npk plots. These differences in level, which occurred throughout the experimental grazing season, were manifestly caused by the difference in potash dressing. The greater differences in level in the autumn compared to the spring levels are probably to be associated with the increasing effect of the potash dressing on the pasture grazed by the cattle. No clinical symptoms of grass tetany occurred during this year. The lowest serum magnesium level occurring in a single animal was measured on 26th October on the plots with a heavy potash dressing and amounted to 0.3 mg/100 ml. The mean level of all eight animals at that date was 0.96 mg/100 ml and the maximum value 1.7 mg/100 ml. On the same date the lowest level measured in the group of four animals on the Npk plots was 2.1 mg/100 ml, the average of the entire group being 2.33 mg/100 ml.

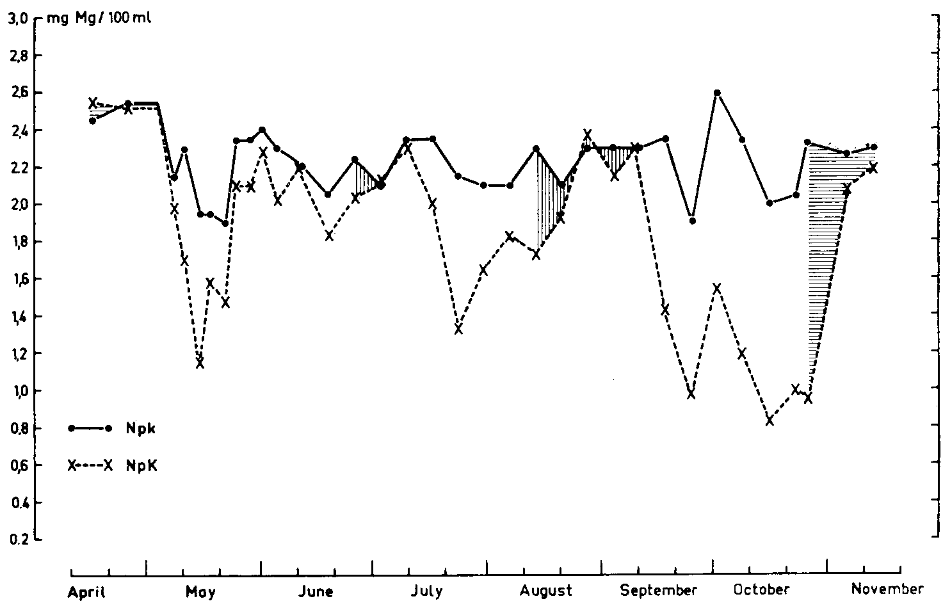
From the winter-feeding period to mid-August the serum-calcium levels were also determined. Nearly all values fluctuated between 10 and 12 mg Ca per 100 ml serum and there were no systematic differences between the treatments, but there was a noticeable rise in the serum-calcium level the first week after the animals had been turned out to grass. Subsequently, however, there was again a gradual fall to the end of May, after which the means per group continued to fluctuate around 10 and 11 mg Ca/100 ml until mid-August.

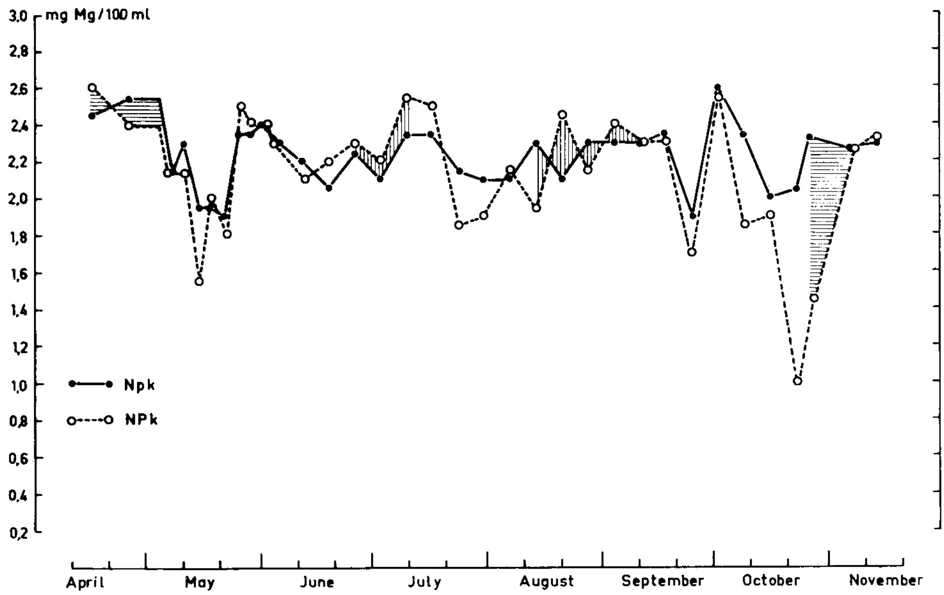
1957. As stated earlier, in this year attention was paid to the effect on the serum magnesium levels of potash and nitrogen dressings in four combinations. Hence there were four treatments, and four cows on the plots of each treatment. Unlike 1956, the same groups of animals did not remain on the same treatments throughout the experimental grazing season. Thus the group of animals which had begun grazing plots of a particular treatment (e.g. group I on the nk plots) did not always remain on the same treatment, but grazed the plots of another treatment for certain periods (e.g. group I was on the nK plots from 4th to 17th May). Table 2 shows the treatments successively grazed by the four groups of animals.

Table 2 Grazing plan of four groups of milking cows on plots with varying potash and nitrogen dressings.

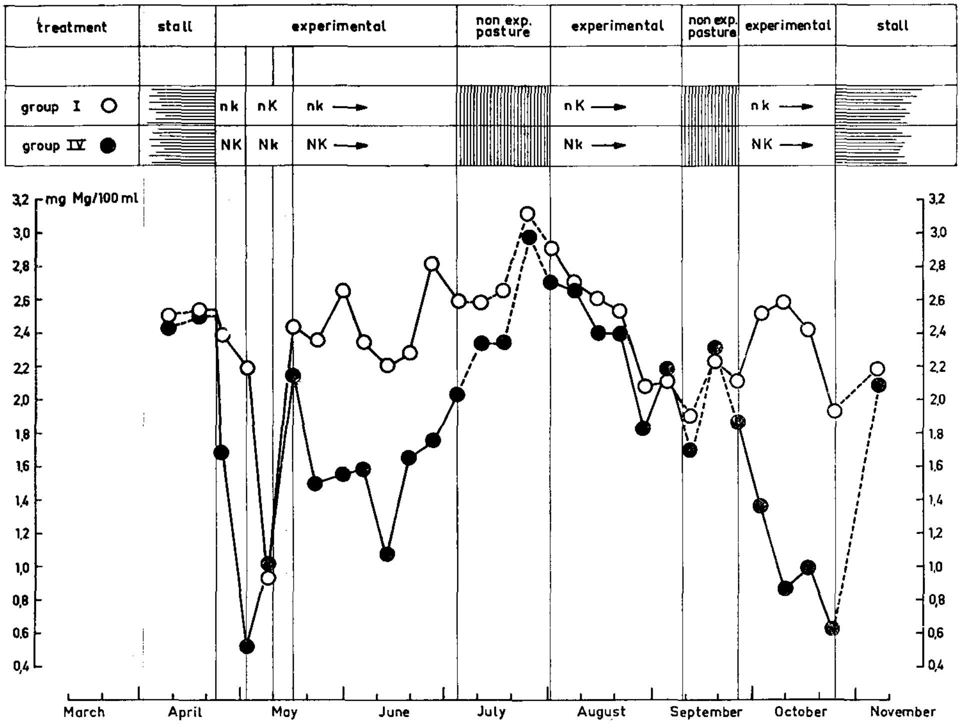
Period ➤	23/4-4/5	4/5-17/5	17/5-4/7	4/7-1/8	1/8-12/9	12/9-25/9	25/9-24/10
Group ➤	Treatment	Treatment	Treatment		Treatment		Treatment
I	nk	nK	nk	outside the experiment	nK	outside the experiment	nk
II	Nk	NK	Nk		NK		Nk
III	nK	nk	nK		nk		nK
IV	NK	Nk	NK		Nk		NK

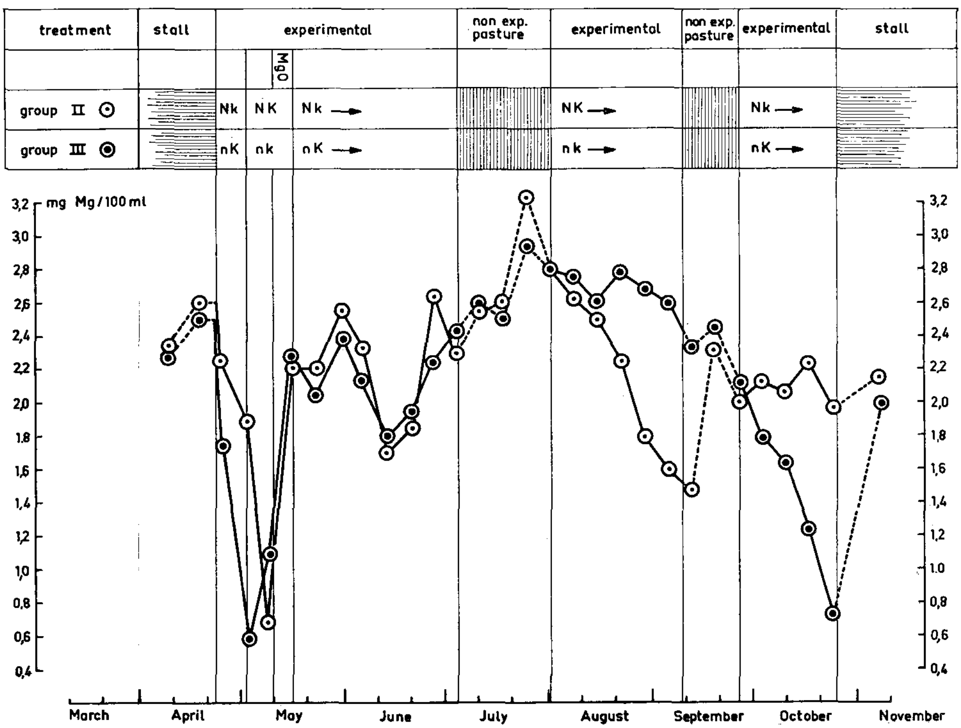
Throughout the experimental grazing season cows of groups I and III grazed on plots with light nitrogen dressings, and groups II and IV on plots with heavy nitrogen dressings. Hence the groups were only changed from plots with a heavy potash dressing to plots with a light potash dressing and vice versa. Before entering into a discussion of the overall trend of the mean



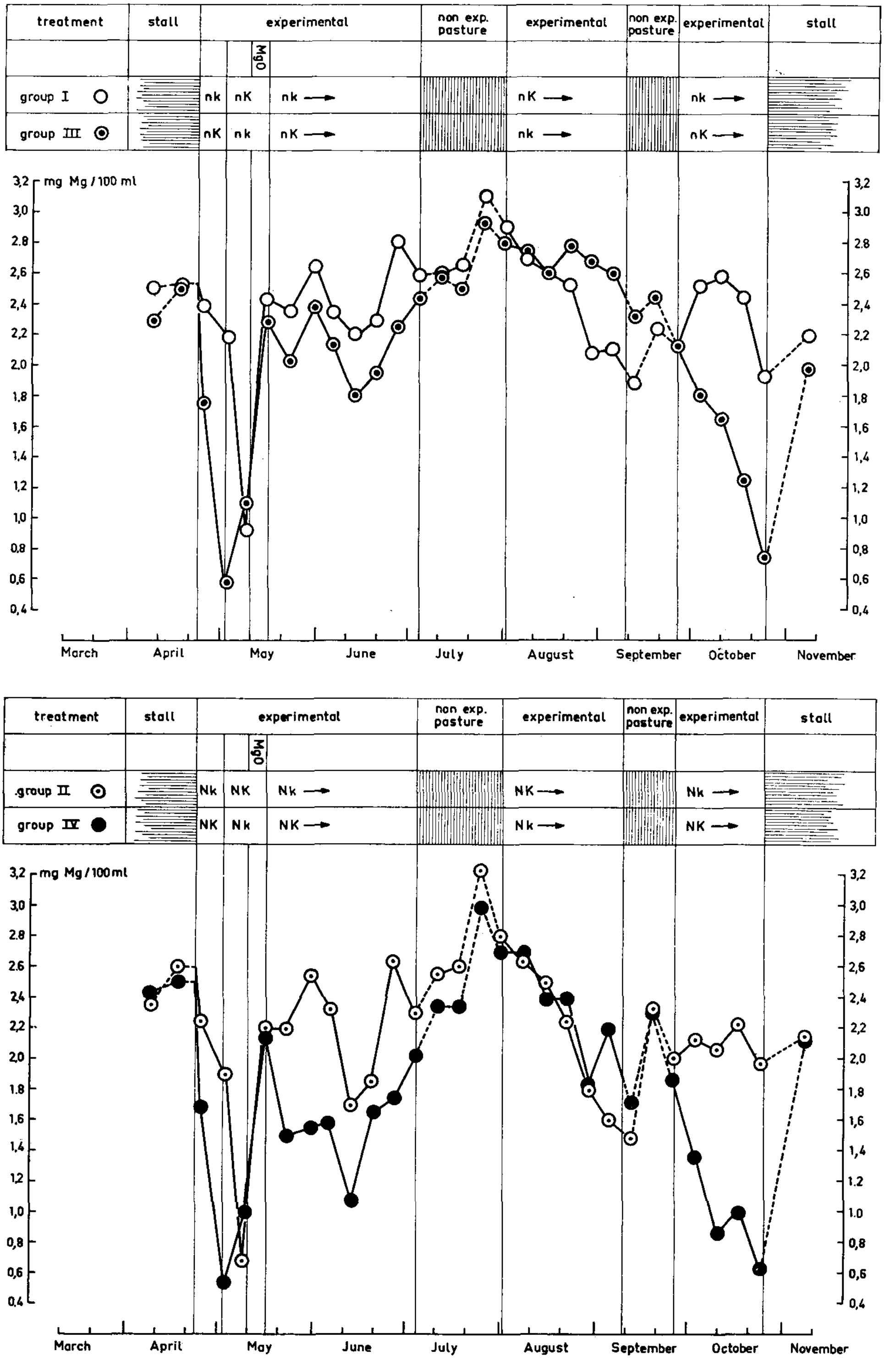


FIGS. 1 AND 2 TREND OF THE MAGNESIUM LEVELS OF THE BLOOD SERUM OF MILKING COWS ON GRASSLAND WITH A LIGHT AND HEAVY POTASH DRESSING (FIG. 1) AND WITH A LIGHT AND HEAVY PHOSPHATE DRESSING (FIG. 2).

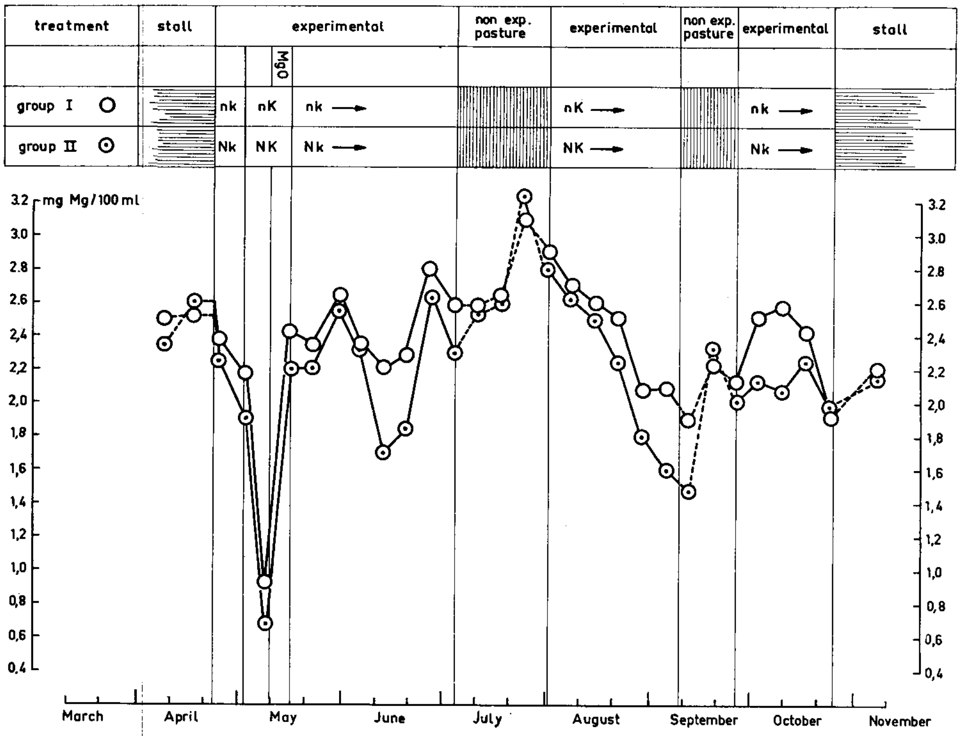


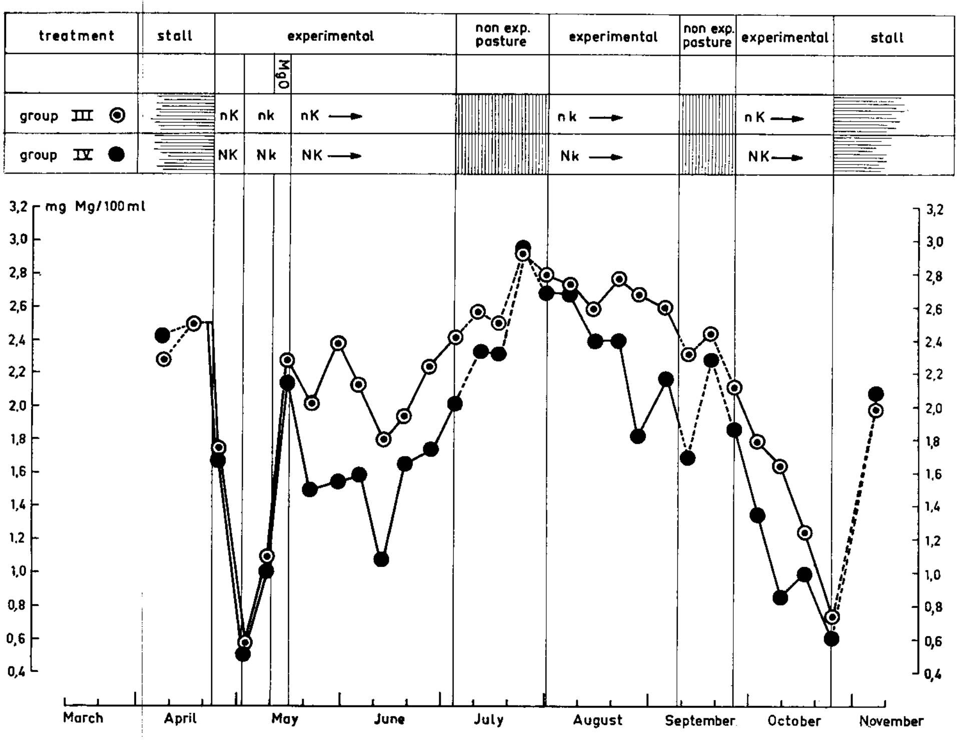


FIGS. 3 AND 4 TREND OF THE MAGNESIUM LEVELS OF THE BLOOD SERUM OF MILKING COWS ON SWARDS WITH HEAVY AND LIGHT POTASH AND NITROGEN DRESSINGS.



FIGS. 5 AND 6 TREND OF THE MAGNESIUM LEVELS OF THE BLOOD SERUM OF MILKING COWS ON SWARDS WITH HEAVY AND LIGHT POTASH AND NITROGEN DRESSINGS.





FIGS. 7 AND 8 TREND OF THE MAGNESIUM LEVELS OF THE BLOOD SERUM OF MILKING COWS ON SWARDS WITH HEAVY AND LIGHT POTASH AND NITROGEN DRESSINGS.

serum magnesium levels on the four treatments with reference to Figs. 3–8, the spring period will be dealt with separately, since during this time there were six cases of grass tetany which occurred exclusively on plots which had received a heavy potash dressing.

After all sixteen cows with a normal serum magnesium level had entered the experimental swards on 23rd April, it was found on 25th April that there was already a considerable fall in the levels of groups III and IV grazing the nK and NK plots respectively (see Table 3).

Table 3 Mean serum magnesium levels in mg Mg per 100 ml of four groups of animals grazing on plots with different fertiliser treatments.

Date →		23/4	25/4	1/5	3/5	4/5	9/5	9/5	10/5	16/5
Group ↓	Winter feed	Treatment				Treatment				
I	2.52	nk	2.38	–	2.18	nK	0.93	c.	extra MgO + lean swards	2.43
II	2.48	Nk	2.25	–	1.90	NK	0.68	c.		2.20
III	2.40	nK	1.75	c.c.c.	0.58	nk	1.10	–		2.28
IV	2.46	NK	1.68	c.	0.53	Nk	1.00	–		2.15

c = one cow with clinical symptoms of grass tetany.

This fall continued until there were very low average values on 3rd May, whereas the serum magnesium levels of groups I and II grazing plots with a light potash dressing were considerably higher. On 1st May, i.e. a week after the cattle had been turned out to grass, four cases of grass tetany occurred in groups III and IV, three of which were serious. These cows displayed such clinical symptoms as convulsions and prostration and the serum magnesium levels were 0.4, 0.4 and 0.5 mg/100 ml. The fourth animal did not suffer from convulsions, but exhibited such symptoms as hyperaesthesia and muscle tremors. The serum magnesium level of this animal was 0.7 mg/100 ml. All four animals recovered fairly rapidly after an intravenous injection of a Ca and Mg solution had been given by the local veterinary surgeon. They then remained in the stalls for a few days and were given a ration of hay, beet pulp and cakes containing supplementary magnesium oxide, after which they were again allotted to the experimental groups.

Nor could the four remaining cows of groups III and IV all be termed clinically normal. The serum magnesium levels were very low on 3rd May, the highest value being 0.8 mg/100 ml. On the other hand, the eight cows of groups I and II grazing the nk and Nk plots displayed no single anomaly during this period. None of these animals was found to have a serum magnesium level less than 1.5 mg/100 ml on 3rd May. In order to prevent any possible accident to the remaining four animals on the nK and NK plots, they were transferred on 4th May to the plots which had received a light potash dressing. At the same time the experimental animals of groups I and II were taken from the nk and Nk plots and put to pasture on the plots with a heavy potash dressing. Table 3 now shows a reversed pattern on the following sampling date, 9th May; the serum magnesium levels of groups I and II had fallen appreciably and those of groups III and IV had risen. At this date two more animals displayed marked symptoms of grass tetany, one from group II on the NK treatment to a very serious degree with a serum magnesium level of 0.2 mg/100 ml. The second, less serious case, occurred in group I on the nK treatment. This animal had a serum magnesium level of 0.4 mg. These levels, like those of the four other patients, were measured in blood samples taken immediately prior to intravenous injection. The other animals in groups I and II also had extremely reduced serum magnesium values on 9th May. The two last patients, like the first four animals, also recovered in a few days after an intravenous injection and a stall ration.

Hence six animals in all suffered from grass tetany during the spring of this year, four on the plots with a heavy potash and light nitrogen dressing and two on the plots with heavy potash and nitrogen dressings. During this spring period it was the potash dressing in particular that greatly influenced the serum magnesium levels. But Table 3 also shows the tendency that lower serum magnesium levels were measured in cows grazing the plots with a heavy nitrogen dressing than those grazing the plots with a light nitrogen dressing. This phenomenon was more in evidence as the year advanced. In this connection it should also be taken into account that the plots with a light nitrogen dressing received 20 kg N per hectare in the spring and nothing more during the remainder of the season.

Since all sixteen animals were in a fairly unstable condition on 9th May, it was decided to give them a supplementary feed of cakes in which additional MgO had been incorporated; in this way an additional 60 grams MgO were given per head per day. The cows were also transferred to the part of the experimental field less rich in potash and which in 1956 was still not included in the experiment and had therefore first received its first trial dressing in the spring of 1957. The effect of taking these steps is shown in the last column of Table 3. It will be seen that on 16th May the mean serum magnesium values of all groups were again at a normal level. No level lower than 1.9 mg/100 ml was observed.

In Figs. 3–8 the trend of the mean serum magnesium levels of the four groups of cows are compared in various combinations. The heading of each figure shows the groups concerned and which treatments of the experimental field were grazed. The horizontally hatched parts show the periods in which the cattle were in the stalls and all received the same feed. As in 1956, in this year there were also two periods in which there was no longer sufficient herbage available on the experimental swards, so that all animals were turned out on a pasture outside the experiment and consequently also consumed the same feed (vertical hatching). The serum magnesium levels during these periods are denoted by broken lines, while the trend of the mean levels on the experimental swards are shown by full lines.

As in 1956, all the figures again emphasise the fact that there were no essential differences between the serum magnesium levels of the groups of cows during the periods in which they were all grazing on the same sward outside the experiment, and also after they had received the same feed in the stalls in autumn. This strengthens the assumption that the differences in serum magnesium levels between the groups were solely duo to the effects of fertilisation. In Figs. 3 and 4 the mean serum magnesium levels are compared of groups I and IV and II and III respectively. Taking these two figures together, it is possible to ascertain the trend of the serum magnesium levels of the nk plots compared to the NK plots throughout the experimental grazing season, i.e. to compare the effects of light and heavy dressings and the trends on the nK plots and the Nk plots. Great differences in serum magnesium values occur between the nk and NK plots, the differences in the spring and autumn being considerably greater than in the summer. It can be seen from Table 4, which shows the size of these differences, that the mean difference in serum magnesium level over the entire experimental grazing season is 0.82 mg/100 ml Mg, and in spring and autumn even higher than 1.20 mg/100 ml.

The lowest levels occurred on 24th October on the NK treatment; on this date the mean level of this group was 0.63 mg/100 ml Mg and the lowest single value 0.4 mg/100 ml. Two animals were seen to have suspicious symptoms on this date, but there no clear cases of grass tetany.

The differences between the nK and Nk plots were smaller, at any rate during the summer, since it can be seen from Figs. 4 and 3 that in this period there were no systematic differences between these treatments. This is in contradistinction to the spring and autumn when the serum magnesium levels on the nK plots were at a considerably lower level than on the Nk plots, so that the heavy potash dressing had an appreciably more harmful effect than the heavy nitrogen dressing. It can be seen from Table 4 that the mean difference in serum magnesium level throughout the year between these treatments is 0.21 mg/100 ml, however, the differences were much greater during the spring and autumn.

Very low serum magnesium levels were also measured on the nK plots both in autumn and spring without the animals displaying any clinical symptoms of grass tetany.

Table 4 Mean serum magnesium levels throughout the grazing season and during four periods thereof of four groups of cows on plots with four different treatments.

Period →	23/4-4/5	17/5-4/7	1/8-12/9	25/9-24/10	Entire experimental grazing period
Treatment ↓					
nk	2.28	2.45	2.62	2.36	2.39
Nk	2.08	2.21	2.20	2.10	2.11
nK	1.16	2.14	2.32	1.36	1.90
NK	1.04	1.59	2.04	0.97	1.57

In Fig. 5 the serum magnesium levels are compared of groups I and III of which the animals alternately grazed plots with a heavy and light potash dressing, the nitrogen applicaction being low in both cases. In Fig. 6, which shows the trend of the levels of groups II and IV, it is possible to ascertain this potash effect when the nitrogen application is high. In both figures the lines denoting the serum magnesium levels on the plots with a heavy potash dressing are noticeably lower than those denoting the trend on the plots with a light potash dressing. The differences in serum magnesium levels caused by the potash dressing when the nitrogen dressing is heavy are not found to be consistently greater or smaller than when the nitrogen dressing is light. If the mean differences are calculated for the entire year, the differences as between the potash dressings with a high nitrogen application are substantially the same as with a low nitrogen level and amount to 0.54 mg/100 ml and 0.49 mg/100 ml Mg respectively (see Table 4).

Finally, Figures 7 and 8 show the serum magnesium levels on the plots with a heavy and light nitrogen dressing at high and low potash levels. These figures show that systematic differences exist between the serum magnesium levels at a high and a low nitrogen level which on balance are to the disadvantage of a high nitrogen level. It will be seen that during the entire experimental grazing season the lines denoting the serum magnesium values of the cows on the plots with a heavy nitrogen dressing are consistently and sometimes considerably below the lines denoting the levels on the plots with a light nitrogen dressing. The form of these lines also shows that in this case, the differences in summer are as great, or greater, than in the autumn (see also Table 4). This is just the reverse from what was found on the plots with high and low potassium. There is no difference in nitrogen effect whether with a heavy or light potash dressing. Table 4 shows that the differences in serum magnesium levels between high and low nitrogen with a heavy and light potash dressing are 0.33 and 0.28 mg/100 ml Mg respectively. The size of the differences mentioned above only relates, of course, to such conditions as were experienced in these two experimental years in these grazing experiments.

The results of these grazing experiments over the two years showed, therefore, that both a heavy potash dressing and a heavy nitrogen dressing of the sward is concomitant with lower serum magnesium levels in milking cows. Very large differences occurred between the NK and nk plots. It was also found that the adverse effect of a heavy potash dressing during the spring and autumn in particular was considerably greater than the effect of a heavy nitrogen dressing. A possible factor in this case is the fact that despite the small percentage of clover in the sward, the nitrogen dressing may have helped the grass/clover ratio in the wrong direction just in the summer when clover grows best, always assuming that a high clover content of the sward is accompanied by higher serum magnesium levels (Bartlett et al., 1954). In this connection it should, however, be remembered that only very small differences in clover content are involved, e.g. from 0–4 %, partly due to the fact that a hormonal spray had been applied.

Individual differences between the animals

In the literature reference is sometimes made to the considerable individual differences in serum magnesium levels between animals of the same group consuming the same feed, and also between animals grazing the same sward. In this connection it should first of all be observed that although a group of animals may consume the same feed in equal amounts, this does not necessarily imply that they are all subject to the same dietary conditions. Usually this will not be the case. It is also known that not all animals of a group grazing the same sward consume the same feed (selective grazing) or consume it in equal amounts. These alone would be sufficient reasons for expecting differences in serum magnesium levels. In fact these differences sometimes emerged fairly clearly in these experiments on the various sampling dates, although sometimes only slightly. It appears, however, that these differences in serum magnesium levels between animals of the same group grazing the same sward are associated with the mean serum magnesium level of the entire group and with the date of sampling. If the conditions to which the animals are subject are such that the mean serum magnesium level of the group is very low, then all individual levels are either low or reduced. Vice versa, when the animals are subject to very favourable conditions under which very favourable mean serum magnesium levels are found, then all individual values are on a considerably higher level. This is illustrated in table 5. Here the frequency distribution of the magnesium levels of the individual cows is given in the columns and the frequency distribution of the means of the groups in the rows.

Table 5 Frequency distribution of the individual observations with very low to very high mean serum magnesium levels per groups of milking cows.

Number of observations per class	Means per group in mg/100 ml						
	0.40-0.80	0.81-1.20	1.21-1.60	1.61-2.00	2.01-2.40	2.41-2.80	> 2.80
< 0.4	4	1	0	0	0	0	0
0.4–0.8	29	35	4	0	0	0	0
0.9–1.3	5	34	26	16	0	0	0
1.4–1.8	1	18	26	59	28	0	0
1.9–2.3	0	1	16	71	163	22	0
2.4–2.8	0	0	0	8	111	154	5
2.9–3.3	0	0	0	0	0	8	12
> 3.3	0	0	0	0	0	0	3

If a group of four milking cows had a mean serum magnesium level of 0.5 mg/100 ml on a given date, then the individual observations from which this mean was calculated were distributed over the said classes in the first column of Table 5. Thus in the first column all individual observations from groups having a mean serum magnesium level of from 0.4 to 0.8 mg/100 ml in the corresponding classes < 0.4, 0.4–0.8, etc. were counted and the total shown in the table. In this way it was found that of all groups of cows of which the mean serum magnesium level per group was higher than 2.40 mg/100 ml, there was no individual level lower than 1.9 mg/100 ml. In the same way only one level higher than 1.8 mg/100 ml occurred in the groups of which the average per group was lower than 1.20 mg/100 ml.

The greater spread of the serum magnesium levels in the middle range is associated with differences in the animals' speed of reaction to a change of circumstances. If, for instance, a group of four milking cows with normal serum magnesium levels is transferred to a sward in which these levels will fall considerably, this fall will be completed more rapidly in one animal than in another. This means than when evaluating the size of the individual differences in serum magnesium contents of a single group, one should take into account both the level and the trend of these contents over a certain period. In this connection there is little point in a single sampling of a group of animals, particularly when the mean level of the group fluctuates widely over a period of time. It was quite obvious, however, that there were individual differences between the serum magnesium contents, and some interesting information comes to light when these contents are studied over an entire year, for instance, in the case of a number of individual animals. It is then found that the serum magnesium contents of, say, two animals over an entire season rise and fall fairly simultaneously but on a different level. It would be desirable to have more information on the cause of this.

In a different experiment from the preceding one, magnesium levels of milking cows on pastures of 23 farms were determined in the spring of 1957. On these pastures one or more cows had clearly been suffering from grass tetany a day previously. This investigation was carried out on farms belonging to the ambulatory practice of the Veterinary Faculty of the University of Utrecht. In this manner 90 blood samples in which the magnesium level was determined were taken on these 23 farms. No blood samples were taken of the patients. Table 6 shows the frequency distribution of the serum magnesium levels of these animals.

Table 6 Frequency distribution of serum magnesium levels of 90 milking cows without clinical symptoms of grass tetany, grazing pastures on which one or more animals had been suffering from grass tetany shortly before.

	Magnesium content of the blood serum in mg/100 ml		
	> 2.0	2.0–1.1	< 1.1
Number of cows in each group	1	38	51

Of all these milking cows only one was found to have a normal magnesium level in the blood serum, viz. one higher than 2.0 mg/100 ml. Nearly 60% of the animals had very low levels, and the remainder had subnormal levels of between 2.0 and 1.1 mg/100 ml. A detailed observation showed that many of these were not clinically normal. This result is in complete agreement with those of the above-mentioned grazing experiments.

The conclusion to be drawn from the above investigation is that when one or more animals of a single group suffer from grass tetany, the serum magnesium levels of most of the other animals are either subnormal or very low. It may therefore be presumed that in Holland, where it is estimated that 1% or 2% of milking cows per annum suffer from grass tetany, a high percentage of the "clinically normal" animals are in a more or less unstable state of hypomagnesaemia. It is not known what harmful consequences this might have.

Acknowledgements

The author is indebted to Mr. H. J. Wentink for technical assistance and Mr. H. J. Immink for the analytical work. The investigations were performed with the financial aid of The National Council for Agricultural Research, T.N.O.

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